Croatian mayflies (Insecta, Ephemeroptera): species diversity and distribution patterns

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Abstract
Knowledge of the mayfly biodiversity in the Balkan Peninsula is still far from complete. Compared to the neighbouring countries, the mayfly fauna in Croatia is very poorly known. Situated at the crossroads of central and Mediterranean Europe and the Balkan Peninsula, Croatia is divided into two ecoregions: Dinaric western Balkan and Pannonian lowland. Mayflies were sampled between 2003 and 2013 at 171 sites, and a total of 66 species was recorded. Combined with the literature data, the Croatian mayfly fauna reached a total of 79 taxa. Of these, 29 species were recorded for the first time in Croatia while 15 species were not previously recorded in Dinaric western Balkan ecoregion. Based on the mayfly assemblage, sampling sites were first structured by ecoregion and then by habitat type. In comparison with the surrounding countries, the Croatian mayfly fauna is the most similar to the Hungarian and Bosnian fauna. Some morphologically interesting taxa such as Baetis cf. nubecularis Eaton, 1898 and Rhithrogena from the diaphana group were recorded. Ephemerida cf. parnassiana Demoulin, 1958, the species previously recorded only from Greece, was also recorded.

Keywords
Ephemeroptera, species list, biodiversity, Balkan Peninsula
Introduction

Mayflies (Ephemeroptera) have a worldwide distribution, being absent only from Arctic region, Antarctica and some remote oceanic islands (Barber-James et al. 2008). According to the literature (Bauernfeind and Soldán 2012), 369 species are recorded for Europe and North Africa. Mayflies are a merolimnic insect order (i.e. with aquatic larval stages and terrestrial adults) that plays a critical role in running and standing waters where they hold an important position in secondary production, as an important food source for diverse freshwater and terrestrial predators. In recent decades, human impacts on the distribution and abundance of many aquatic insects, including mayflies, are becoming more and more evident. During the 20th century, increasing industrialisation, population growth, overexploitation of natural resources and different types of pollutions have greatly impacted many European freshwater ecosystems, and also endangering the species inhabiting them (Brittain and Sartori 2009). Highly sensitive, confronted with habitat alteration, mayfly species are among the first to disappear. Therefore they are important indicators of freshwater health and widely used in bio-monitoring programmes over the world (Elliott et al. 1988, Sartori and Brittain 2015). The knowledge of the mayfly biodiversity in the Balkan Peninsula is still far from complete. Moreover, many taxa lack appropriate morphological descriptions for the larval and/or adult stages. The mayfly fauna in Croatia is no exception. Published data on Croatian mayflies are generally part of diverse limnological studies (e.g. Matoničkin 1959, 1987, Matoničkin and Pavletić 1961, 1967, Filipović 1976, Habdija and Primc 1987, Habdija et al. 1994, 2004) in which mayflies were investigated only as part of the overall macroinvertebrate fauna. In most studies, identification tools are generally not cited, thus the accuracy of mayfly species identification is questionable. In summary, 50 mayfly species were recorded from Croatia (Bauernfeind and Soldán 2012, Kovács and Murányi 2013, Ćuk et al. 2015). In comparison with the number of species recorded in the neighbouring countries, i.e. 68 in Slovenia, 106 in Italy, and 93 in Hungary (Bauernfeind and Soldán 2012), it can be assumed that the Croatian mayfly fauna has been underestimated to date.

Studies on distribution and biodiversity are of crucial importance in determining the conservation status of certain species and in investigating factors that influence that diversity (de Silva and Medellín 2001). Therefore, knowledge of the mayfly faunal composition, seasonal dynamics, distribution, ecology, biogeography and especially their sensitivity as bio-indicators can enable high-quality classification and protection of Croatian freshwater habitats.

Materials and methods

This research is based on recent mayfly studies conducted in the last decade (2003–2013). The results of field studies were then combined with the literature data given in Bauernfeind and Soldán (2012), Kovács and Murányi (2013) and Ćuk et al. (2015), for the purpose of obtaining a comprehensive checklist of the Croatian mayfly fauna.
Sampling and laboratory methods

Croatia is a relatively small country situated at the crossroads of Central and Mediterranean Europe and Balkan Peninsula, and is divided into two ecoregions: Dinaric western Balkan (ER5) and Pannonian lowland (ER11) (Illies 1978). Specimens were collected in lotic and lentic freshwater habitats throughout the Croatian territory (Fig. 1). Additionally, specimens housed in the collection of the Slovene National History Museum were identified.

The list of the 171 sampling site names with number codes (site ID), altitude, latitude and longitude is presented in Table 1 as well as on the map (Fig. 1). Larvae were sampled using a Surber sampler and hand net, adults using hand nets and pyramidal emergence traps.

![Map of the mayfly fauna sampling sites, Croatia (See Table 1 for codes).](image-url)
Table 1. The list of the sampling sites in Croatia. Ecoregions are taken from Illies (1978); Dinaric western Balkan (5) and Pannonian lowland (11). BS = Black Sea Basin; AS = Adriatic Sea Basin.

| Site ID | Sampling site                                      | Altitude | Longitude     | Latitude      | Ecoregion | Basin |
|---------|----------------------------------------------------|----------|---------------|---------------|-----------|-------|
| 1       | Karašica River, Valpovo                           | 85       | N45°37'44"    | E18°27'28"    | 11        | BS    |
| 2       | Vučica River, Valpovo                             | 85       | N45°38'14"    | E18°25'09"    | 11        | BS    |
| 3       | Čarna channel, Tikveš, near Bilje                  | 85       | N45°40'23"    | E18°50'46"    | 11        | BS    |
| 4       | Veličanka River, Mihaljevci                       | 155      | N45°21'36"    | E17°40'54"    | 11        | BS    |
| 5       | Sava River, Slavonski Brod                         | 85       | N45°07'35"    | E18°02'18"    | 11        | BS    |
| 6       | Sava River, Stitar                                 | 80       | N45°05'47"    | E18°37'38"    | 11        | BS    |
| 7       | Sutla River, Klanjec                               | 160      | N46°02'46"    | E15°43'49"    | 11        | BS    |
| 8*      | Drava River, Varaždin                              | 170      | N46°19'50"    | E16°20'22"    | 11        | BS    |
| 9       | Drava River, Čakovec, left drainage ditch          | 165      | N46°18'49"    | E16°27'49"    | 11        | BS    |
| 10      | Drava River, Dubrava, right drainage ditch         | 145      | N46°18'54"    | E16°42'15"    | 11        | BS    |
| 11      | Stream, Trakošćan                                  | 275      | N46°15'44"    | E15°56'30"    | 11        | BS    |
| 12      | Stiper stream, Ljubešćica, Kalnik Mountain         | 185      | N46°09'04"    | E16°22'18"    | 11        | BS    |
| 13      | Bliznec stream, Medvednica Mountain                | 380      | N45°52'38"    | E15°58'33"    | 11        | BS    |
| 14      | Veliki potok stream, Medvednica Mountain, Mikulići | 300      | N45°51'29"    | E15°56'08"    | 11        | BS    |
| 15      | Kraljevec stream, Medvednica Mountain              | 565      | N45°52'48"    | E15°56'28"    | 11        | BS    |
| 16      | Sitnik spring, Žumberak-Samoborsko Gorge Mountain  | 745      | N45°44'40"    | E15°32'39"    | 11        | BS    |
| 17      | Slapnica stream, Žumberak-Samoborsko Gorge Mountain| 290      | N45°44'12"    | E15°29'29"    | 11        | BS    |
| 18*     | Kupa River, Sisak                                  | 90       | N45°28'32"    | E16°22'37"    | 11        | BS    |
| 19      | Sava River, Rugvica                                | 100      | N45°44'01"    | E16°13'11"    | 11        | BS    |
| 20      | Sava River, Mlaka                                  | 90       | N45°14'14"    | E17°01'11"    | 11        | BS    |
| 21      | Sava River, Zagreb, bridge                         | 110      | N45°47'03"    | E16°00'10"    | 11        | BS    |
| 22      | Bregana River, Jarušje                             | 560      | N45°46'21"    | E15°34'36"    | 11        | BS    |
| 23      | Stream, Mečenčani                                  | 180      | N45°17'07"    | E16°25'53"    | 11        | BS    |
| 24      | Stream Zeleni dol, Hrastovica/ Hrvatski Čuntić     | 160      | N45°21'51"    | E16°16'15"    | 11        | BS    |
| 25      | Pond Zeleni dol, Hrastovica/ Hrvatski Čuntić       | 160      | N45°21'51"    | E16°16'18"    | 11        | BS    |
| 26      | Petrinjčica River, Prnjavor Čuntićki               | 150      | N45°21'05"    | E16°16'57"    | 11        | BS    |
| 27      | Petrinjčica River, Tješnjak, bridge                | 150      | N45°22'52"    | E16°17'11"    | 11        | BS    |
| 28      | Utinja River, Križ Hrastovački                     | 140      | N45°25'15"    | E16°14'32"    | 11        | BS    |
| 29      | Žirovnica River, Donja Ljubina                     | 135      | N45°05'39"    | E16°17'39"    | 11        | BS    |
| 30      | Moštanica stream, Moštanica                       | 155      | N45°21'55"    | E16°21'06"    | 11        | BS    |
| 31      | Sunja River, Rakovac                               | 120      | N45°18'40"    | E16°32'33"    | 11        | BS    |
| 32      | Sunja River, Donji Kukuruzari                       | 150      | N45°16'01"    | E16°29'14"    | 11        | BS    |
| 33      | Kupa River, Brest                                  | 90       | N45°26'56"    | E16°15'38"    | 11        | BS    |
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|   | Site                                      | Latitude       | Longitude       | Code | Country |
|---|------------------------------------------|----------------|-----------------|------|---------|
| 34 | Kupa River, Bubnjarci                    | N45°38'42"    | E15°21'24"     | 5    | BS      |
| 35 | Una River, Hrvatska Kostajnica            | N45°13'37"    | E16°32'22"     | 11   | BS      |
| 36 | Glina River, Marinbrod                    | N45°23'19"    | E16°08'20"     | 11   | BS      |
| 37 | Glina River, Cerjak                       | N45°21'27"    | E16°04'58"     | 11   | BS      |
| 38 | Čemernica stream, Topusko                 | N45°19'08"    | E15°57'30"     | 11   | BS      |
| 39 | Sava River oxbow, Mužilovčica             | N45°23'23"    | E16°40'37"     | 11   | BS      |
| 40  | Sava River, Martinska Ves                 | N45°35'09"    | E16°22'14"     | 11   | BS      |
| 41  | Sava River, Desno Trebarjevo              | N45°35'56"    | E16°20'43"     | 11   | BS      |
| 42  | Sava River, Krapje                        | N45°18'10"    | E16°49'23"     | 11   | BS      |
| 43  | Sava River, Lukavec Posavski              | N45°24'36"    | E16°31'03"     | 11   | BS      |
| 44  | Sava River, Drenov bok                    | N45°15'58"    | E16°50'04"     | 11   | BS      |
| 45  | Mire Plavnica, Šatornja                   | N45°19'58"    | E16°00'26"     | 11   | BS      |
| 46  | Javošnica stream, Donji Javoranj          | N45°07'14"    | E16°21'44"     | 11   | BS      |
| 47  | Odra River, Sisak                         | N45°29'54"    | E16°21'04"     | 11   | BS      |
| 48  | Zrinčica River, Zrin                      | N45°11'41"    | E16°22'13"     | 11   | BS      |
| 49  | Čatlan River, Gornja Oraovica             | N45°09'26"    | E16°25'03"     | 11   | BS      |
| 50  | Spring Izvor bijele stijene Križ, Župić   | N45°25'44"    | E16°13'52"     | 11   | BS      |
| 51  | Šanja River, Gora                         | N45°25'08"    | E16°11'42"     | 11   | BS      |
| 52  | Radonja River, Vojnić                     | N45°19'26"    | E16°41'55"     | 11   | BS      |
| 53  | Lonja River, Brežnički Hum                | N46°07'34"    | E16°17'18"     | 11   | BS      |
| 54  | Lonja River, Breznica                     | N46°04'11"    | E16°18'07"     | 11   | BS      |
| 55  | Mrežnica River, Generalski stol           | N45°22'05"    | E15°24'55"     | 5    | BS      |
| 56  | Mrežnica River, Duga Resa                 | N45°27'31"    | E15°29'38"     | 5    | BS      |
| 57  | Dretulja River, Plaški, middle reach       | N45°05'31"    | E15°20'32"     | 5    | BS      |
| 58  | Dretulja River, Plaški                    | N45°05'06"    | E15°21'56"     | 5    | BS      |
| 59  | Trupinjska rijeka River, Keserov potok    | N45°17'04"    | E15°37'28"     | 5    | BS      |
| 60  | Gojačka Dobra River, Gorinci, downstream from the waterfall | N45°21'10" | E15°20'44" | 5 | BS |
| 61  | Gojačka Dobra River, Gorinci, waterfall above the dam | N45°20'60" | E15°20'45" | 5 | BS |
| 62  | Gojačka Dobra River, Tomasići             | N45°22'33"    | E15°21'18"     | 5    | BS      |
| 63  | Bukovska Dobra River, Turkovići           | N45°16'59"    | E15°10'49"     | 5    | BS      |
| 64  | Ribnjak stream, Trošmarija               | N45°19'43"    | E15°16'25"     | 5    | BS      |
| 65  | Virunčica stream, Virunj                  | N45°17'01"    | E15°09'48"     | 5    | BS      |
| 66  | Bistrica stream, Bistrac                  | N45°16'27"    | E15°17'28"     | 5    | BS      |
| 67  | Sušik stream, Drežnica                    | N45°08'44"    | E15°04'41"     | 5    | BS      |
| 68  | Bračana stream, Škuljari                  | N45°24'57"    | E13°55'36"     | 5    | AS      |
| 69  | Rečica stream, Pengari                    | N45°23'21"    | E13°59'13"     | 5    | AS      |
| 70  | Draga River, Selca                        | N45°23'36"    | E13°59'46"     | 5    | AS      |
| 71  | Račički potok stream, Juradi              | N45°20'17"    | E13°57'20"     | 5    | AS      |
| 72  | Mirna River, Koji                         | N45°22'06"    | E14°01'         | 5    | AS      |
| 73  | Jadova River, Gornja Ploča                | N44°27'03"    | E15°38'58"     | 5    | AS      |
| 74  | Obsenica stream, near Lovinac             | N44°21'09"    | E15°40'36"     | 5    | AS      |
| 75  | Ričica stream, Ričice                    | N44°20'23"    | E15°45'08"     | 5    | AS      |
| No. | Location | Distance (m) | Latitude  | Longitude  | Season  | Code |
|-----|----------|--------------|-----------|------------|---------|------|
| 76  | Lika River, Lički Ribnik | 565 | N44°29'13" | E15°27'38" | 5 | AS |
| 77  | Gacka River, Ličko Lešće | 450 | N44°48'46" | E15°19'18" | 5 | AS |
| 78  | Gacka River, Prozor | 450 | N44°50'23" | E15°15'21" | 5 | AS |
| 79* | Bijela rijeka River, NP Plitvice Lakes, upper reach | 715 | N44°50'04" | E15°33'33" | 5 | BS |
| 80* | Bijela rijeka River, NP Plitvice Lakes, spring | 760 | N44°49'56" | E15°33'22" | 5 | BS |
| 81* | Crna rijeka River, NP Plitvice Lakes, spring | 710 | N44°49'43" | E15°36'49" | 5 | BS |
| 82* | Crna rijeka River, NP Plitvice Lakes, upper reach | 680 | N44°50'10" | E15°36'30" | 5 | BS |
| 83* | Crna rijeka River, NP Plitvice Lakes, lower reach | 670 | N44°50'22" | E15°35'59" | 5 | BS |
| 84* | Korana River, NP Plitvice Lakes | 390 | N44°55'33" | E15°37'09" | 5 | BS |
| 85* | Plitvica stream, NP Plitvice Lakes | 555 | N44°54'08" | E15°36'27" | 5 | BS |
| 86* | Tufa barrier Novakovića Brod, NP Plitvice Lakes | 510 | N44°54'07" | E15°36'38" | 5 | BS |
| 87* | Tufa barrier Labudovac, NP Plitvice Lakes | 630 | N44°52'17" | E15°35'59" | 5 | BS |
| 88* | Tufa barrier Kozjak-Milanovac, NP Plitvice Lakes | 545 | N44°53'39" | E15°36'32" | 5 | BS |
| 89* | Kozjak Lake, NP Plitvice Lakes | 555 | N44°53'18" | E15°36'38" | 5 | BS |
| 90* | Prošće Lake, NP Plitvice Lakes | 665 | N44°51'51" | E15°36'06" | 5 | BS |
| 91* | Ciginovac Lake, NP Plitvice Lakes | 640 | N44°52'22" | E15°35'51" | 5 | BS |
| 92* | Kaluđerovac Lake, NP Plitvice Lakes | 540 | N44°54'05" | E15°36'41" | 5 | BS |
| 93  | Suha Ričina stream, Jurandvor, Krk island | 20 | N44°58'38" | E14°43'52" | 5 | AS |
| 94  | Zeleni vir, Skrad | 540 | N45°25'25" | E14°53'53" | 5 | BS |
| 95  | Curak stream, Zeleni vir | 330 | N45°25'37" | E14°53'33" | 5 | BS |
| 96  | Veli potok stream, Dobrinj, Krk island | 35 | N45°08'06" | E14°35'43" | 5 | AS |
| 97  | Kupica River spring, Mala Lošnica, NP Risnjak | 270 | N45°25'48" | E14°51'07" | 5 | BS |
| 98  | Mijića vrelo stream, Mijići | 60 | N44°09'37" | E15°52'38" | 5 | AS |
| 99  | Krupa River, Krupa | 130 | N44°11'34" | E15°54'34" | 5 | AS |
| 100 | Krupa River, Kudin bridge | 90 | N44°11'16" | E15°50'44" | 5 | AS |
| 101 | Pond, Zvjerinac | 245 | N43°56'45" | E16°12'56" | 5 | BS |
| 102 | Jaruga stream, Jelavica bridge, Zmijavci | 260 | N43°24'46" | E17°15'09" | 5 | AS |
| 103 | Otuća River, Deringaj, Kijani | 615 | N44°21'02" | E15°52'34" | 5 | AS |
| 104 | Vransko Lake, main channel, Biograd | 0 | N43°56'20" | E15°30'59" | 5 | AS |
| 105 | Vransko Lake, Biograd, Drage | 5 | N43°53'44" | E15°33'07" | 5 | AS |
| 106 | Krka River, Roški slap waterfall, NP Krka | 75 | N43°54'23" | E15°58'30" | 5 | AS |
| 107 | Visovac Lake, NP Krka | 50 | N43°51'38" | E15°58'55" | 5 | AS |
| No. | Location Description                             | Latitude  | Longitude  | Accuracy | Type |
|-----|-------------------------------------------------|-----------|------------|----------|------|
| 108 | Brljan Lake, NP Krka                           | 44°00'30" | 16°02'41"  | 5        | AS   |
| 109*| Kosovčica River, upper reach, Vučenoviči       | 43°58'30" | 16°12'45"  | 5        | AS   |
| 110*| Kosovčica River, lower reach, Biskupija         | 44°00'26" | 16°12'52"  | 5        | AS   |
| 111 | Krka River, Knin                               | 44°01'56" | 16°11'26"  | 5        | AS   |
| 112 | Krka River, upstream of Kosovčica river mouth, Knin | 44°02'24" | 16°13'42"  | 5        | AS   |
| 113 | Krka River, downstream of Kosovčica river mouth, Knin | 44°01'41" | 16°12'48"  | 5        | AS   |
| 114 | Orašnica River, Knin                           | 44°01'56" | 16°12'04"  | 5        | AS   |
| 115 | Zrmanja River, Mokro polje, Prkos              | 44°05'31" | 16°02'00"  | 5        | AS   |
| 116 | Zrmanja River, Vekići                          | 44°06'06" | 15°56'41"  | 5        | AS   |
| 117 | Zrmanja River, Palanka                         | 44°08'23" | 16°04'25"  | 5        | AS   |
| 118 | Zrmanja River, Muškovci, Berberi buk           | 44°11'50" | 15°46'07"  | 5        | AS   |
| 119 | Zrmanja River, Kravlja Draga, bridge           | 44°05'50" | 16°04'30"  | 5        | AS   |
| 120 | Zrmanja River, Žegar, bridge                   | 44°09'10" | 15°53'08"  | 5        | AS   |
| 121 | Zrmanja River, Draga                           | 44°09'50" | 15°50'43"  | 5        | AS   |
| 122 | Lopuško vrelo stream, Lake                    | 44°01'11" | 16°13'21"  | 5        | AS   |
| 123 | Krčić River, Kovačić                          | 44°02'19" | 16°16'42"  | 5        | AS   |
| 124 | Krčić River, Mlinica                          | 44°01'38" | 16°19'25"  | 5        | AS   |
| 125 | Šarena jezera lake, Biskupija                  | 44°01'36" | 16°13'22"  | 5        | AS   |
| 126 | Čikola River, near Rakići                      | 44°01'13" | 16°04'25"  | 5        | AS   |
| 127 | Čikola River, Otavice                          | 44°05'36" | 16°15'25"  | 5        | AS   |
| 128 | Vrba River, Vrba                              | 44°03'21" | 16°23'58"  | 5        | AS   |
| 129 | Vrba River, Čavoglave                          | 44°07'28" | 16°18'52"  | 5        | AS   |
| 130 | Butižnica River, Knin                         | 44°02'44" | 16°11'39"  | 5        | AS   |
| 131 | Brodic stream, Markovac, Biskupija             | 44°05'03" | 16°15'00"  | 5        | AS   |
| 132 | Karakašica, Karakašica                         | 43°43'04" | 16°38'19"  | 5        | AS   |
| 133 | Boggy seepages, Brvun, Gračac                 | 44°23'15" | 15°53'08"  | 5        | AS   |
| 134 | Ričina stream, Proložac                       | 44°29'20" | 16°09'11"  | 5        | AS   |
| 135*| Cetina River, Spring Glavaš                    | 43°58'36" | 16°25'48"  | 5        | AS   |
| 136 | Grab River, Spring                             | 43°38'24" | 16°46'20"  | 5        | AS   |
| 137*| Cetina River, Preočki most bridge              | 43°57'59" | 16°25'53"  | 5        | AS   |
| 138*| Cetina River, Crveni most bridge               | 43°57'35" | 16°25'46"  | 5        | AS   |
| 139*| Cetina River, Obrovac Sinjski                  | 43°43'58" | 16°41'11"  | 5        | AS   |
| 140*| Cetina River, Trilj1                          | 43°36'54" | 16°43'42"  | 5        | AS   |
| 141*| Cetina River, Ćikotina lada                    | 43°31'58" | 16°44'42"  | 5        | AS   |
| 142*| Cetina River, Radmanove mlinice                | 43°26'19" | 16°45'06"  | 5        | AS   |
| 143*| Cetina River, Trilj2                          | 43°36'19" | 16°43'28"  | 5        | AS   |
| 144 | Cetina River, Peruča Reservoir                | 43°47'45" | 16°35'32"  | 5        | AS   |
| 145 | Cetina River, Zadvarje                        | 43°26'02" | 16°53'18"  | 5        | AS   |
| 146*| Ruda River, spring                            | 43°40'07" | 16°47'39"  | 5        | AS   |
Mayflies were sampled in every season at 34 sites, while at the remainder of sites, sampling was usually performed only once between April and September. Specimens were stored in 80% ethanol and identified in the lab using a stereomicroscope and microscope. A reference collection was made by preparing permanent slide mounts of identified species. Larvae were treated with 10% KOH and 99% acetic acid to remove all muscle parts. Mouth parts, legs, gills, thorax, abdomen, paraproct plate in Baetidae and cerci, necessary for the species identification, were fixed in Euparal and examined under a microscope. Adult specimens were mostly identified by the imaginal male genitalia. The collected material (larvae and adult specimens) was identified using Müller-Liebenau

| No. | Location Information                                                                 | Latitude   | Longitude  | Sampling Sites | Waterbody Type   |
|-----|--------------------------------------------------------------------------------------|------------|------------|----------------|------------------|
| 147 | Ruda River, upper reach                                                              | N43°40'06" | E16°47'28" | 5              | AS               |
| 148 | Cetina River tributary stream, Vukovići, Paško polje                                | N43°58'06" | E16°25'07" | 5              | AS               |
| 149 | Cetina River tributary stream, Kotluša, Paško polje                                 | N43°56'54" | E16°24'06" | 5              | AS               |
| 150 | Jadro River 1, Solin                                                                 | N43°32'23" | E16°29'45" | 5              | AS               |
| 151 | Matica River, Vrgorac                                                                | N43°12'21" | E17°23'46" | 5              | AS               |
| 152 | Matica River, Umčani                                                                 | N43°10'28" | E17°22'32" | 5              | AS               |
| 153 | Stinjevac spring, Dusina                                                             | N43°10'29" | E17°25'02" | 5              | AS               |
| 154 | Cetina River, Čitluk                                                                  | N43°44'48" | E16°39'49" | 5              | AS               |
| 155 | Vukovića vrilo spring, Bitelići, Hrvace                                             | N43°49'12" | E16°37'28" | 5              | AS               |
| 156 | Ljuta River, spring                                                                  | N42°32'20" | E18°22'46" | 5              | AS               |
| 157 | Ljuta River, upper reach, Donja Ljuta                                                 | N42°32'05" | E18°22'39" | 5              | AS               |
| 158 | Vodovada stream, Palje Brdo                                                          | N42°30'29" | E18°24'34" | 5              | AS               |
| 159 | Konavоčica River, near Karasovići                                                   | N42°30'19" | E18°24'37" | 5              | AS               |
| 160 | Stream, near Zastolje                                                                | N42°31'17" | E18°23'31" | 5              | AS               |
| 161 | Stream, near Brajkovići                                                               | N42°31'49" | E18°23'14" | 5              | AS               |
| 162 | Vljiška River, Kamenmost                                                             | N43°25'52" | E17°11'42" | 5              | AS               |
| 163 | Vljiška River, Kapuše                                                                  | N43°26'33" | E17°10'32" | 5              | AS               |
| 164 | Jarun Lake, Zagreb                                                                    | N45°46'47" | E15°55'17" | 11             | BS               |
| 165 | Stream under the village Beram                                                        | N45°15'10" | E13°54'18" | 5              | AS               |
| 166 | Spring by the church, Stajnica, Porekuli                                          | N45°02'31" | E15°14'18" | 5              | AS               |
| 167 | Danube River, Ilok                                                                      | N45°13'49" | E19°23'26" | 11             | BS               |
| 168 | Ljubica stream, Baška Oštarije, Linić, Velebit Mountain                              | N44°31'37" | E15°09'41" | 5              | AS               |
| 169 | Spring by the church, Slano                                                           | N42°47'01" | E17°53'26" | 5              | AS               |
| 170 | Spring by the sea, Dubrovnik, Mali Zaton                                                | N42°42'06" | E18°02'40" | 5              | AS               |
| 171 | Tounjčica stream, Tounj                                                               | N45°14'56" | E15°20'04" | 5              | BS               |

* Sampling sites used in calculating Shannon-Weaver and Simpson indices and in Cluster analysis.
○ Samples stored in Slovene Natural History Museum. The remaining samples are stored at the University of Zagreb, Faculty of Science, Department of Biology, Division of Zoology, Zagreb.
Croatian mayflies (Insecta, Ephemeroptera): species diversity and distribution patterns

Data analysis

All recorded specimens were included in the Croatian mayfly species list. Data for the sites with the same sampling effort were statistically analysed using the PRIMER 6 software package (Clarke and Warwick 2001). As such, only 34 sampling sites were compared out of the total 171 (Table 1). These sites were sampled in all seasons, at the available microhabitats and they represent habitats in each ecoregion and each sea basin. Species diversity, evenness, and similarity between sites with respect to the mayfly composition and abundance were determined by the Shannon-Weaver and Simpson indices. For estimation of similarity and differences in the mayfly community composition, cluster analysis was used. Similarity among sites was determined using the Bray-Curtis similarity index. SIMPER (Similarity Percentage) was used to assess which taxa are primarily responsible for the similarities between the sites of the same habitat type. The Croatian mayfly species richness was compared with the surrounding countries (Bosnia & Herzegovina, Hungary, Slovenia, Italy) by compiling species list for these countries taken from Bauernfeind and Soldán (2012) and the Sørensen Index of Similarity was calculated.

Results

Species richness

In total, 79 mayfly taxa (Table 2) were recorded for Croatia. Of the 171 sites (55 in ER11, 116 in ER5) investigated during this study (Table 1), 66 taxa were sampled, of which 29 were recorded for the first time (Table 2). The presence of 13 (16%) previously recorded species could not be confirmed (Table 2). The most diverse genera were *Baetis* Leach, 1815 and *Ecdyonurus* Eaton, 1868 both with 11 species. *Baetis rhodani* (Pictet, 1843) and *Serratella ignita* (Poda, 1761) were the most widely distributed species, present in 83 and 76 sampling sites, respectively. Fourteen species were recorded at only one sampling site: *Cloeon simile* Eaton, 1870, *Procloeon nana* (Bogoescu, 1951), *Caenis pusilla* Navàs, 1913, *Ephemera cf. parnassiana* Demoulin, 1958, *Leptophlebia vespertina* (Linnaeus, 1758), *Ecdyonurus vitoshensis* Jacob & Braasch, 1984, *Ecdyonurus zelleri* (Eaton, 1885), *Electrogena mazedonica* (Ikonomov, 1954), *Heptagenia coerulans* Rostock, 1878, *H. flava* Rostock, 1878, *H. longicauda* (Stephens, 1835), *Rhithrogena iridina* (Kolenati, 1839), *Rh. gr. diaphana* and *Rh. semicolorata* (Curtis, 1834).

Approximately half of the species (30) were present in both ecoregions. A total of 50 species was recorded as present only in the Dinaric western Balkan ecoregion (ER5) and 48 only in the Pannonian lowland ecoregion (ER11) (Table 2). Nearly half the species (32)
Table 2. Croatian mayfly fauna.

| Mayfly taxa | Ecoregion | Habitat type | Basin |
|-------------|-----------|--------------|-------|
| Ametropodidae |           |              |       |
| Ametropus fragilis Albarda, 1878 | 11 | 3 | BS |
| Ameletidae |           |              |       |
| Ameletus inopinatus Eaton, 1887 | - | - | - |
| Metreletus balcanicus (Ulmer, 1920) | - | - | - |
| Siphlonuridae |           |              |       |
| Siphlonurus armatus (Eaton, 1870) | - | - | - |
| Siphlonurus croaticus Ulmer, 1920 | 11 | 2,3,4 | AS |
| Siphlonurus lacustris (Eaton, 1870) | 5, 11 | 2,3 | BS, AS |
| Bactidae |           |              |       |
| Alainites muticus (Linnaeus, 1758) | 5 | 2,3,4 | BS, AS |
| Baetis alpinus (Pictet, 1843) | 5, 11 | 1,2,3 | BS |
| Baetis buceratus Eaton, 1870 | 11 | 3 | BS |
| Baetis fuscatus (Linnaeus, 1761) | 5, 11 | 3 | BS |
| Baetis liebenauae Keffermüller, 1974 | 5, 11 | 1,2,3 | BS, AS |
| Baetis lutheri Müller-Liebenau, 1967 | 5, 11 | 1,3 | BS, AS |
| Baetis melanonyx (Pictet, 1843) | 5 | 1,2,3 | AS |
| Baetis cf. nubecularis (Eaton, 1898) | 5 | 1,2,3,4 | BS |
| Baetis rhodani (Pictet, 1843) | 5, 11 | 1,2,3,4 | BS, AS |
| Baetis scambus Eaton, 1870 | 11 | 3 | BS |
| Baetis tricolor Tshernova, 1928 | 11 | 3 | BS |
| Baetis vernus Curtis, 1834 | 5, 11 | 3 | BS, AS |
| Baetopus tenellus (Albarda, 1878) | 5, 11 | 2,3 | BS |
| Nigrobaetis niger (Linnaeus, 1761) | 5, 11 | 2,3 | BS, AS |
| Centropilum luteolum (Müller, 1776) | 5, 11 | 2,3,4,5 | BS, AS |
| Cloeon dipterum (Linnaeus, 1761) | 5, 11 | 2,3,5 | BS, AS |
| Cloeon simile Eaton, 1870 | 5 | 5 | AS |
| Procloeon bifidum (Bengtsson, 1912) | 5, 11 | 2,3 | BS, AS |
| Procloeon nana (Bogoescu, 1951) | 5 | 2 | AS |
| Procloeon penmulatum (Eaton, 1870) | 5, 11 | 3,4 | BS, AS |
| Caenidae |           |              |       |
| Brachycercus harrisellus Curtis, 1834 | 11 | 3 | BS |
| Caenis beskidensis Sowa, 1973 | 5 | 3 | AS |
| Caenis horaria (Linnaeus, 1758) | 5, 11 | 3,4,5 | BS, AS |
| Caenis macrura Stephens, 1835 | 5, 11 | 3 | BS, AS |
| Caenis pusilla Navas, 1913 | 5 | 3 | BS |
| Caenis rivulorum Eaton, 1884 | 11 | 3 | BS |
| Caenis robusta Eaton, 1884 | 11 | 2,3,5 | BS |
| Ephemeraellidae |           |              |       |
| Ephemerella mucronata (Bengtsson, 1909) | 5, 11 | 2,3 | BS, AS |
| Serratella ignita (Poda, 1761) | 5, 11 | 1,2,3,4 | BS, AS |
| Torleya major (Klapalek, 1905) | 5, 11 | 2,3,4 | BS, AS |
| Family               | Species                                      | References | Distribution | Notes   |
|---------------------|----------------------------------------------|------------|--------------|---------|
| Ephemeridae         | Ephemera danica Müller, 1764                 | 5, 11      | 2,3,4,5      | BS, AS  |
|                     | ▲ Ephemera glutops Piclet, 1843               |            |              |         |
|                     | Ephemera lineata Eaton, 1870                 | 5          | 2,3,5        | AS      |
|                     | ● Ephemera cf. parnassiana Demoulin, 1958   | 5          | 2            | AS      |
|                     | Ephemera vulgata Linnaeus, 1758              | 5, 11      | 2,3,5        | BS, AS  |
|                     | ● Ephemera zettana Kimmins, 1937             | 5          | 2,3          | AS      |
| Palingeniidae       | ▲ Palingenia longicauda (Olivier, 1791)      |            |              |         |
| Polymitarcyidae     | ▲ Ephoron virgo (Olivier, 1791)              |            |              |         |
| Leptophlebiidae     | ▲ Choroterpes picteti (Eaton, 1871)          |            |              |         |
|                     | Habroleptoides confusa Sartori and Jacob, 1986 | 5, 11      | 2,3          | BS, AS  |
|                     | Habrophlebia fusca (Curtis, 1834)            | 5, 11      | 1,2,3        | BS, AS  |
|                     | Habrophlebia lauta Eaton, 1884               | 5, 11      | 2,3,5        | BS, AS  |
|                     | ● Leptophlebia vespertina (Linnaeus, 1758)   | 5          | 2,5          | BS, AS  |
|                     | Paraleptophlebia submarginata (Stephens, 1835) | 5, 11      | 2,3,4        | BS, AS  |
|                     | ● Paraleptophlebia werneri Ulmer, 1920       | 5          | 2,5          | BS      |
| Oligoneuriidae      | Oligoneuriella rhenana (Imhoff, 1852)        | 11         | 3            | BS      |
| Potamanthidae       | Potamanthus luteus (Linnaeus, 1767)          | 11         | 3            | BS      |
| Heptageniidae       | ▲ Ecdyonurus aurantiacus (Burmeister, 1839)  |            |              |         |
|                     | Ecdyonurus dispar (Curtis, 1834)             |            |              |         |
|                     | Ecdyonurus insignis (Eaton, 1870)            | 5          | 2,3          | BS, AS  |
|                     | ● Ecdyonurus macani Thomas & Sowa, 1970      | 5, 11      | 3            | BS, AS  |
|                     | ▲ Ecdyonurus siveci Hefti, Tomka & Zurwerra, 1986 |            |              |         |
|                     | ● Ecdyonurus starmachi Sowa, 1971            | 5, 11      | 2,3          | BS, AS  |
|                     | ● Ecdyonurus submontanus Landa, 1969          | 5          | 3            | BS      |
|                     | Ecdyonurus torrentis Kimmins, 1942            | 5          | 2,3          | BS, AS  |
|                     | Ecdyonurus venosus (Fabricius, 1775)         | 5          | 2,3          | AS      |
|                     | Ecdyonurus vitosensis Jacob & Braasch, 1984  | 11         | 2            | BS      |
|                     | ● Ecdyonurus zelleri (Eaton, 1885)           | 11         | 2            | BS      |
|                     | ● Electrogena affinis (Eaton, 1883)          | 5          | 2,3          | AS      |
|                     | Electrogena lateralis (Curtis, 1834)         | 5, 11      | 2,3,4        | BS, AS  |
|                     | ● Electrogena mazedonica (Ikonomonov, 1954)  | 5          | 3            | AS      |
|                     | ● Electrogena ujhelyii (Sowa, 1981)          | 5, 11      | 1,2          | BS, AS  |
|                     | Epeorus assimilis Eaton, 1885                | 5, 11      | 1,2,3        | BS, AS  |
|                     | Heptagenia coerulans Rostock, 1878            | 11         | 3            | BS      |
|                     | Heptagenia flavus Rostock, 1878              | 11         | 3            | BS      |
|                     | ● Heptagenia longicauda (Stephens, 1835)     | 5          | 3            | BS      |
|                     | Heptagenia sulphurea (Müller, 1776)          | 11         | 3            | BS      |
were recorded in both the Black and Adriatic Sea Basins, while 25 species were recorded only for Black Sea basin and 11 species only for Adriatic Sea basin (Table 2).

The Sørensen Index of Similarity indicated the Croatian mayfly fauna had the greatest similarity with the Hungarian assemblage (Table 3).

**Mayflies (Insecta, Ephemeroptera) of Croatia**

For the distribution data, the following format was used: “Literature data” were mainly taken from Bauernfeind and Soldán (2012), which listed the presence of each species in Croatia but without reference to their exact localities. Two and one species and localities where they were recorded were mentioned in Kovács and Murányi (2013) and Ćuk et al., respectively. “Literature data with new records” corresponds to data obtained as a part of this study but were already published. “New records” are data obtained in this study but were not yet published. For every species, the site ID is listed. All sampling sites and their ID numbers are listed in Table 1.

- New records for the Croatian mayfly fauna
- Only adults recorded
I. Ametropodidae Bengtsson, 1913
1. *Ametropus fragilis* Albarda, 1878
   **Literature data:** Drava River, Donji Miholjac (Čuk et al. 2015)

II. Ameletidae McCafferty, 1991
2. *Ameletus inopinatus* Eaton, 1887
   **Literature data:** Bauernfeind and Soldán (2012)
3. *Metreletus balcanicus* (Ulmer, 1920)
   **Literature data:** Bauernfeind and Soldán (2012)

III. Baetidae Leach, 1815
4. *Alainites muticus* (Linnaeus, 1758)
   **Literature data:** Bauernfeind and Soldán (2012)
   **Literature data with new records:** 79, 80, 82, 84, 85, 86 (Vilenica et al. 2014)
   **New records:** 68, 70, 115, 150, 158, 160, 161, 162, 163, 165, 168

5. *Baetis alpinus* (Pictet, 1843)
   **Literature data:** Bauernfeind and Soldán (2012)
   **New records:** 13, 15, 57, 63

6. *Baetis buceratus* Eaton, 1870
   **New records:** 2, 36

7. *Baetis fuscatus* (Linnaeus, 1761)
   **Literature data:** Bauernfeind and Soldán (2012)
   **New records:** 5, 7, 8, 10, 18, 19, 26, 29, 31, 32, 35, 36, 40, 56, 60, 61, 62

8. *Baetis liebenauae* Keffermüller, 1974
   **New records:** 1, 2, 9, 10, 35, 36, 37, 62, 98, 109, 110, 111, 112, 113, 122, 128, 131, 134, 139, 140, 141, 143, 151, 152, 153, 162, 171

9. *Baetis lutheri* Müller-Liebenau, 1967
   **Literature data:** Bauernfeind and Soldán (2012)
   **New records:** 7, 18, 19, 35, 61, 62, 103, 116, 141, 142, 146, 147, 150, 157

10. *Baetis melanonyx* (Pictet, 1843)
    **New records:** 115, 117, 120, 146, 147, 156, 157, 158, 159, 160, 161, 162, 163

11. *Baetis cf. nubecularis* Eaton, 1898
    **Literature data with new records:** 79, 80, 81, 82, 83, 84, 85, 86, 87 (Vilenica et al. 2014)
12. *Baetis rhodani* (Pictet, 1843)  
**Literature data:** Bauernfeind and Soldán (2012)  
**Literature data with new records:** 79, 80, 81, 82, 83, 84, 85, 87, 88 (Vilenica et al. 2014)  
**New records:** 9, 10, 13, 15, 16, 23, 24, 26, 28, 29, 30, 31, 32, 34, 35, 48, 50, 51, 53, 59, 61, 62, 63, 64, 65, 66, 68, 70, 77, 78, 98, 99, 100, 103, 109, 110, 112, 113, 114, 115, 116, 117, 118, 120, 122, 123, 124, 128, 131, 132, 134, 135, 137, 138, 139, 140, 141, 142, 146, 147, 148, 149, 153, 157, 158, 159, 160, 161, 162, 163, 166, 169, 170, 171

13. *Baetis scambus* Eaton, 1870  
**Literature data:** Bauernfeind and Soldán (2012)  
**New records:** 7, 26

14. *Baetis tricolor* Tshernova, 1928 ●  
**New records:** 20, 43, 44

15. *Baetis vernus* Curtis, 1834 ●  
**New records:** 7, 9, 10, 36, 38, 53, 54, 76

16. *Baetopus tenellus* (Albarda, 1878) ●  
**New records:** 19, 64, 94

17. *Nigrobaetis niger* (Linnaeus, 1761) ●  
**Literature data with new records:** 138  
**New records:** 15, 36, 38, 93, 103, 109, 110, 128, 131

18. *Centroptilum luteolum* (Müller, 1776)  
**Literature data:** Bauernfeind and Soldán (2012)  
**Literature data with new records:** 84, 85, 86, 87, 88, 89, 90, 91, 92 (Vilenica et al. 2014)  
**New records:** 1, 12, 23, 27, 28, 31, 32, 35, 61, 62, 69, 74, 77, 78, 103, 107, 109, 110, 121, 127, 128, 141, 142, 143, 144, 159

19. *Cloeon dipterum* (Linnaeus, 1761)  
**Literature data:** Bauernfeind and Soldán (2012)  
**New records:** 1, 5, 20, 24, 35, 37, 39, 41, 43, 44, 45, 46, 47, 60, 67, 78, 101, 103, 104, 105, 121, 125, 127, 128, 129, 152

20. *Cloeon simile* Eaton, 1870  
**Literature data:** Bauernfeind and Soldán (2012)  
**New records:** 125

21. *Procloeon bifidum* (Bengtsson, 1912)  
**Literature data:** Bauernfeind and Soldán (2012)  
**New records:** 6, 19, 20, 28, 29, 31, 32, 40, 41, 42, 44, 47, 62, 68, 69, 71, 115, 121, 141
22. *Procloeon nana* (Bogoescu, 1951) ●
**New records:** 68

23. *Procloeon pennulatum* (Eaton, 1870)
**Literature data:** Bauernfeind and Soldán (2012)
**Literature data with new records:** 84, 85, 86 (Vilenica et al. 2014)
**New records:** 26, 27, 61, 127, 129

IV. Caenidae Newman, 1853
24. *Brachycercus harrisellus* Curtis, 1834
**Literature data:** Vojlovica River at the bridge of road No. 2, Vojlovica (Kovács and Murányi 2013)

25. *Caenis beskidensis* Sowa, 1973 ●
**New records:** 139, 140, 141, 143, 142

26. *Caenis horaria* (Linnaeus, 1758)
**Literature data:** Bauernfeind and Soldán (2012)
**Literature data with new records:** 86, 87, 89, 90, 91, 92 (Vilenica et al. 2014)
**New records:** 39, 73, 78, 101, 106, 107

27. *Caenis macrura* Stephens, 1835
**Literature data:** Bauernfeind and Soldán (2012)
**New records:** 8, 9, 10, 18, 26, 27, 28, 31, 32, 35, 40, 41, 54, 61, 68, 71, 115, 140, 141, 142, 143

28. *Caenis pusilla* Navàs, 1913 ●
**New records:** 62

29. *Caenis rivulorum* Eaton, 1884 ●
**New records:** 40, 41

30. *Caenis robusta* Eaton, 1884 ●
**New records:** 1, 24, 39, 47

V. Ephemerellidae Klapálek, 1909
31. *Ephemerella mucronata* (Bengtsson, 1909) ●
**New records:** 14, 134, 139, 163

32. *Serratella ignita* (Poda, 1761)
**Literature data:** Bauernfeind and Soldán (2012)
**Literature data with new records:** 83, 84, 85, 86, 88 (Vilenica et al. 2014))
New records: 1, 7, 8, 9, 10, 12, 17, 26, 27, 28, 29, 30, 31, 32, 34, 35, 36, 37, 48, 49, 53, 56, 59, 60, 61, 62, 64, 65, 66, 68, 69, 73, 76, 98, 99, 100, 103, 108, 109, 110, 113, 114, 115, 116, 117, 118, 119, 121, 122, 129, 134, 137, 138, 139, 140, 141, 142, 143, 144, 146, 147, 148, 150, 153, 157, 158, 159, 162, 163, 171

33. Torleya major (Klapalek, 1905)
Literature data: Bauernfeind and Soldán (2012)
New records: 53, 66, 117, 139, 141

VI. Ephemeridae Latreille, 1810
34. Ephemera danica Müller, 1764
Literature data: Bauernfeind and Soldán (2012)
New records: 8, 14, 17, 23, 27, 28, 30, 33, 48, 49, 53, 59, 60, 61, 63, 64, 66, 68, 95, 100, 115, 141, 142

35. Ephemera glaucops Pictet, 1843
Literature data: Bauernfeind and Soldán (2012)

36. Ephemera lineata Eaton, 1870
Literature data: Bauernfeind and Soldán (2012)
New records: 106, 107, 108, 109, 110, 118, 119, 122, 137, 138, 139, 140, 141, 142, 143, 147

37. Ephemera cf. parnassiana Demoulin, 1958 ●
New records: 98

38. Ephemera vulgata Linnaeus, 1758
Literature data: Bauernfeind and Soldán (2012)
New records: 11, 54, 55, 59, 100, 125, 128, 154, 164

39. Ephemera zettana Kimmins, 1937 ● ■
New records: 102, 118, 134, 136, 138, 141, 142, 154, 155

VII. Heptageniidae Needham, 1901
40. Ecdyonurus aurantiacus (Burmeister, 1839)
Literature data: Bauernfeind and Soldán (2012)

41. Ecdyonurus dispar (Curtis, 1834)
Literature data: Bauernfeind and Soldán (2012)
New records: 61, 63, 66, 68, 69
42. *Ecdyonurus insignis* (Eaton, 1870)

**Literature data:** Cetina River, between Podgrade and Slime (Kovács and Murányi 2013)

**New records:** 26, 27, 32, 116, 141, 145

43. *Ecdyonurus macani* Thomas & Sowa, 1970 ●

**New records:** 7, 26, 27, 137, 138, 139, 141, 147

44. *Ecdyonurus siveci* Hefti, Tomka & Zurwerra, 1986

**Literature data:** Bauernfeind and Soldán (2012)

45. *Ecdyonurus starmachi* Sowa, 1971 ●

**New records:** 13, 14, 26, 53, 103, 120

46. *Ecdyonurus submontanus* Landa, 1969 ●

**Literature data with new records:** 82, 83 (Vilenica et al. 2014)

47. *Ecdyonurus torrentis* Kimmins, 1942

**Literature data:** Bauernfeind and Soldán (2012)

**New records:** 95, 99, 118, 119, 120

48. *Ecdyonurus venosus* (Fabricius, 1775)

**Literature data:** Bauernfeind and Soldán (2012)

**New records:** 97 ■, 99, 100, 109, 110, 112, 118, 119, 120, 137, 138, 139, 141, 148, 150, 162

49. *Ecdyonurus vitoshensis* Jacob & Braasch, 1984

**Literature data:** Bauernfeind and Soldán (2012)

**New records:** 12

50. *Ecdyonurus zelleri* (Eaton, 1885) ●

**New records:** 53

51. *Electrogena affinis* (Eaton, 1883) ●

**New records:** 68, 69, 70

52. *Electrogena lateralis* (Curtis, 1834)

**Literature data:** Bauernfeind and Soldán (2012)

**Literature data with new records:** 86 (Vilenica et al. 2014)

**New records:** 12, 27, 61, 96, 165

53. *Electrogena mazedonica* (Ikonomov, 1954) ●

**New records:** 128
54. *Electrogena ujhelyii* (Sowa, 1981) ●  
**New records:** 11, 13, 16, 24, 50, 93

55. *Epeorus assimilis* Eaton, 1885  
**Literature data:** Bauernfeind and Soldán (2012)  
**New records:** 4, 13, 94, 97, 98, 99, 115, 116, 117, 120, 135, 137, 138, 141, 142, 146, 147, 156

56. *Heptagenia coerulans* Rostock, 1878  
**Literature data:** Bauernfeind and Soldán (2012)  
**New records:** 18

57. *Heptagenia flava* Rostock, 1878  
**Literature data:** Bauernfeind and Soldán (2012)  
**New records:** 167

58. *Heptagenia longicauda* (Stephens, 1835) ●  
**New records:** 63

59. *Heptagenia sulphurea* (Müller, 1776)  
**Literature data:** Bauernfeind and Soldán (2012)  
**New records:** 7, 8, 18, 21, 40, 42

60. *Kageronia fuscogrisea* (Retzius, 1783)  
**Literature data:** Bauernfeind and Soldán (2012)

61. *Rhithrogena braaschi* Jacob, 1974 ●  
**Literature data with new records:** 79, 80, 81, 82, 83, 85 (Vilenica et al. 2014)  
**New records:** 57, 58, 109, 110, 112, 117, 120, 122, 124, 135, 137, 138, 139, 141, 142, 143, 146, 147, 162, 163

62. *Rhithrogena gr. diaphana* ●  
**New records:** 32

63. *Rhithrogena germanica* Eaton, 1885  
**Literature data:** Bauernfeind and Soldán (2012)

64. *Rhithrogena iridina* (Kolenati, 1839) ●  
**New records:** 27

65. *Rhithrogena semicolorata* (Curtis, 1834)  
**Literature data:** Bauernfeind and Soldán (2012)  
**New records:** 53
VIII. Leptophlebiidae Banks, 1900
66. Choroterpes picteti (Eaton, 1871)
Literature data: Bauernfeind and Soldán (2012)

67. Habroleptodes confusa Sartori and Jacob, 1986
Literature data: Bauernfeind and Soldán (2012)
New records: 22, 120, 158

68. Habrophlebia fusca (Curtis, 1834)
Literature data: Bauernfeind and Soldán (2012)
New records: 27, 28, 30, 35, 38, 48, 59, 69, 70, 131, 168, 169

69. Habrophlebia lauta Eaton, 1884
Literature data: Bauernfeind and Soldán (2012)
Literature data with new records: 82, 83, 85, 90 (Vilenica et al. 2014)
New records: 25, 26, 27, 29, 48, 49, 61, 65, 66, 68, 70, 109, 110

70. Leptophlebia vespertina (Linnaeus, 1758)
Literature data with new records: 90, 91 (Vilenica et al. 2014)
New records: 134

71. Paraleptophlebia submarginata (Stephens, 1835)
Literature data: Bauernfeind and Soldán (2012)
Literature data with new records: 79, 83, 84, 85, 86, 87, 88 (Vilenica et al. 2014)
New records: 8, 14, 26, 53, 60, 61, 74, 77, 98, 109, 110, 118, 119, 120, 128, 134, 137, 138, 139, 141, 142, 162

72. Paraleptophlebia werneri Ulmer, 1920
Literature data with new records: 85, 90 (Vilenica et al. 2014)

IX. Oligoneuriidae Ulmer, 1914
73. Oligoneuriella rhenana (Imhoff, 1852)
Literature data: Bauernfeind and Soldán (2012)
New records: 26, 27, 32

X. Palingeniidae Albarda, 1888
74. Palingenia longicauda (Olivier, 1791)
Literature data: Bauernfeind and Soldán (2012)

XI. Polymitarcyidae Banks, 1900
75. Ephoron virgo (Olivier, 1791)
Literature data: Bauernfeind and Soldán (2012)
XII. Potamanthidae Albarda, 1888
76. Potamanthus luteus (Linnaeus, 1767)
Literature data: Bauernfeind and Soldán (2012)
New records: 7, 8, 9, 10, 18, 35, 36, 37, 40

XIII. Siphlonuridae Ulmer, 1920 (1888)
77. Siphlonurus armatus (Eaton, 1870)
Literature data: Bauernfeind and Soldán (2012)

78. Siphlonurus croaticus Ulmer, 1920
Literature data: Bauernfeind and Soldán (2012)
Literature data with new records: 82, 83, 85, 87 (Vilenica et al. 2014)
New records: 55, 66, 111, 123, 128, 130, 135■, 137

79. Siphlonurus lacustris (Eaton, 1870)
Literature data: Bauernfeind and Soldán (2012)
New records: 26, 27, 30, 73, 76

Community composition
The majority of the Croatian mayfly species were found to be associated with rivers and streams (Table 2). Among these, larvae of ten species also occurred within the spring areas (Table 2). Eleven species recorded in lakes and/or ponds were also found to inhabit flowing-water habitats. Cluster analysis (Fig. 2) showed that based on the

Figure 2. Cluster analysis of mayfly community composition, based on Bray-Curtis Similarity (See Table 1 for codes).
mayfly assemblage, sampling sites were mainly structured first by ecoregion and then by habitat type. Species richness at the sampling sites and diversity indices are presented in Table 4. Species richness ranged from 2 and 18 species, Shannon-Weaver index between 0.21 and 1.96 and Simpson index between 0.11 and 0.82. All sampling sites with the highest species richness and diversity indices were situated in the Dinaric western Balkan ecoregion (ER5).

The SIMPER analysis between sites within the same habitat type showed an average similarity ranging from 35.1% for the Pannonian lowland rivers to 57.3% for the springs (Table 5).

### Table 4. Species richness (S), Shannon-Weaver (H’) and Simpson (1-λ) indices of diversity, calculated for 34 sites. Sites with the highest H’ and 1-λ are in bold.

| Sampling site | S   | H’   | 1-λ  |
|---------------|-----|------|------|
| 8             | 7   | 1.38 | 0.65 |
| 18            | 6   | 1.05 | 0.54 |
| 40            | 6   | 1.19 | 0.62 |
| 41            | 5   | 1.09 | 0.55 |
| 42            | 2   | 0.56 | 0.4  |
| 60            | 5   | 0.31 | 0.12 |
| 61            | 11  | 0.76 | 0.31 |
| 62            | 8   | 0.85 | 0.44 |
| 79            | 5   | 0.95 | 0.56 |
| 80            | 4   | 1.01 | 0.61 |
| 81            | 3   | 0.98 | 0.59 |
| 82            | 7   | 0.77 | 0.39 |
| 83            | 10  | 1.70 | 0.75 |
| 84            | 9   | 1.43 | 0.69 |
| 85            | 12  | 1.67 | 0.75 |
| 86            | 10  | 1.51 | 0.71 |
| 87            | 7   | 1.41 | 0.67 |
| 88            | 5   | 1.06 | 0.59 |
| 89            | 3   | 0.86 | 0.56 |
| 90            | 6   | 0.52 | 0.24 |
| 91            | 4   | 0.86 | 0.43 |
| 92            | 3   | 1.06 | 0.66 |
| 109           | 10  | 1.77 | 0.75 |
| 110           | 9   | 1.42 | 0.69 |
| 135           | 2   | 0.21 | 0.11 |
| 137           | 9   | 1.30 | 0.66 |
| 138           | 9   | 1.26 | 0.65 |
| 139           | 11  | 1.35 | 0.61 |
| 140           | 6   | 1.31 | 0.65 |
| 141           | 18  | 1.96 | 0.81 |
| 142           | 11  | 1.83 | 0.82 |
| 143           | 7   | 1.09 | 0.52 |
| 146           | 4   | 1.09 | 0.59 |
| 147           | 8   | 1.08 | 0.56 |
Discussion

Due to the paucity of systematic studies, mayfly fauna and their habitat preferences in Croatia were very poorly known, with records of only 50 species (Bauernfeind and Soldán 2012, Kovács and Murányi 2013, Ćuk et al. 2015). As expected, this study showed a higher diversity: 66 taxa were recorded, of which 29 for the first time in Croatia (Table 2). Combined with the literature, the species list consists of 79 taxa. Croatia is a relatively small Balkan country divided into two Ecoregions: Dinaric western Balkan (ER5) and Pannonian lowland (ER11) (Illies 1978) due to its position on the crossroads of Central and Mediterranean Europe, which is why its mayfly fauna shows transitive characteristics.

As a result, species with wide (e.g. *Baetis rhodani*, *Cloeon dipterum*, *Caenis horaria*, *Serratella ignita*), patchy (e.g. *Procloeon nana*, *Leptophlebia vespertina*, *Caenis beskiden-
Croatian mayflies (Insecta, Ephemeroptera): species diversity and distribution patterns

sis) central European (e.g. Baetis cf. nubecularis, Ecdyonurus zelleri, Electrogena ujhelyii) as well as southern (e.g. Ephemerida zettana) and Balkan (e.g. Electrogena mazedonica, Rhithrogena braaschi, Ephemerida cf. parnassiana) distribution were recorded in Croatia. Additionally, 15 taxa were found that were not previously recorded in the Dinaric western Balkan ecoregion: Baetis cf. nubecularis, Procloeon nana, Caenis beskidensis, Ephemerida cf. parnassiana, Ecdyonurus macani, E. submontanus, E. torrentis, Electrogena affinis, E. mazedonica, E. ujhelyii, Heptagenia longicauda, Rhithrogena braaschi, Habroleptiodes confusa, Leptophlebia vespertina and Paraleptophlebia werneri (Buffagni et al. 2007, 2009, Bauernfeind and Soldán 2012).

The new records include several morphologically interesting taxa: Rhithrogena from the diaphana group, Baetis cf. nubecularis and Ephemerida cf. parnassiana. The Rhithrogena species from the diaphana group is morphologically similar to Rhithrogena savoiensis Alba-Tercedor & Sowa, 1987. However, DNA analysis based on mitochondrial COI gene shows it to be more closely related to Rhithrogena beskidensis Alba-Tercedor & Sowa, 1987 (Vuataz unpubl. results). Thus, reliable identification cannot be distinguished at this time. Comparison with other Balkan Rhithrogena diaphana group species and further detailed studied are required. A similar case is recorded for the Baetis alpinus group (sensu Müller-Liebenau, 1969), which presents the morphological characteristics that are intermediate between Baetis alpinus and B. nubecularis. Interestingly, the species is only recorded in high numbers (Vilenica et al. 2014) in the mountain Dinaric karst streams and tufa barriers in the area of Plitvice Lakes National Park (Table 1, Fig. 1). One male imago of the genus Ephemerida Linnaeus, 1758, was caught in the Lopoško vrelo stream in southern Croatia. Its morphological features correspond to Ephemerida parnassiana, a species that has currently only been recorded from Greece; however due to the small sample size, additional specimens are necessary for accurate identification of the species.

As most sites were in running waters and often with a stony substrate, the most diverse genera were Baetis and Ecdyonurus, which are known to be very common in running waters of the Northern Hemisphere (Bauernfeind and Soldán 2012). The most widely distributed species were two eurytopic and eurythermal species: Baetis rhodani and Serratella ignita. Further study is required at new sampling sites to determine the distribution of eleven species recorded only at only a single sampling site (Cloeon simile, Procloeon nana, Caenis pusilla, Ephemerida cf. parnassiana, Leptophlebia vespertina, Ecdyonurus vitosensis, E. zelleri, Electrogena mazedonica, Heptagenia coeruleus, H. flava, H. longicauda, Rhithrogena iridina, Rh. gr. diaphana and Rh. semicolorata), as well as to determine the presence of the thirteen species listed in the literature which were not confirmed in this study (Ametropus fragilis, Ameletus inopinatus, Metreletus balcanicus, Siphlonurus armatus, Brachycercus harrisellus, Ephemerida glaucops, Palingenia longicauda, Ephoron virgo, Choroterpes picteti, Ecdyonurus aurantius, E. siveci, Kageronia fuscogrisea and Rhithrogena germanica). The rare or unconfirmed presence of most of these species is likely due to the lack of seasonal sampling. It is possible that they were present at some sampling sites included in this study, but at a very young instar or even egg stage, and as such were overlooked. Additionally, some species might have
become extinct from the Croatian rivers, such as *Palingenia longicauda*, which at present likely only inhabits the Danube River and Tisza River in Hungary, Slovakia and Ukraine (Bauernfeind and Soldán 2012).

The Black Sea basin includes 62% of Croatian rivers (Jelić et al. 2008), which likely explains the higher number of mayfly species recorded in this basin than in the Adriatic Sea basin.

The Dinaric region is considered to be a biodiversity hotspot (Bânãrescu 2004, Griffiths et al. 2004, Ivković and Plant 2015). Despite a similar number of taxa recorded in each ecoregion, the highest species diversity was recorded for the fast flowing streams and rivers in the Dinaric western Balkan ecoregion. Similar results were obtained in the study of aquatic dance flies in Croatia (Ivković et al. 2013). The lowest number of mayfly species was found in springs and lakes (Table 4). Various studies have shown that mayfly species diversity is generally low in spring areas (Berner and Pescador 1988, Bauernfeind and Moog 2000, Maiolini et al. 2011). The only spring with four species was the spring of the Ruda River (146) in southern Croatia (Fig. 1), which is largely fed with water from the Buško Blato reservoir (Štambuk-Giljanović 2001, Bonacci and Roje-Bonacci 2003) that is relatively rich in nutrients and organic matter (Štambuk-Giljanović 2001). Thus, mayfly communities in the Ruda River spring are more species diverse and have a high proportion of detritivores (Vilenica unpubl. results). Most mayfly species prefer lotic habitats with a larger array of microhabitats, and these are less diverse in spring areas and lentic habitats. The present study confirmed the results of many previous studies (Berner and Pescador 1988, Elliott et al. 1988, Bauernfeind and Humpesch 2001, Bauernfeind and Soldán 2012).

Mayfly larvae inhabit flowing and standing freshwater ecosystems where they occupy a range of microhabitats in correlation with different biotic and abiotic factors. Additionally, in running water habitats, due to the longitudinal gradient of the physico-chemical characteristics of the water, different parts of the watercourse are inhabited by different mayfly species (Elliott et al. 1988, Bauernfeind and Humpesch 2001). Cluster analysis (Fig. 2) based on mayfly assemblage generally showed that sampling sites are structured first by ecoregion and then by habitat type. For this reason, due to their morphology and water properties (Lucić et al. 2015), the large, slow Pannonian lowland rivers (Sava, Drava, Kupa) are separated from the other sampling sites situated in the Dinaric western Balkan ecoregion. SIMPER analysis (Table 5) showed that the Pannonian mayfly community consisted of species that prefer epipotamalic sections of rivers, such as *Caenis macrura*, *Proclœon bifidum*, *Heptagenia sulphurea* and *Potamanthus luteus* (Buffagni et al. 2007, 2009, Bauernfeind and Soldán 2012). Due to the two common mayfly species present in high numbers, *Baetis rhodani* and *Rhithrogena braaschi* (Vilenica et al. 2014, Vilenica unpubl. results), the investigated springs clustered together with the small mountain karst rivers. Larger karst rivers clustered together due to the presence of species with a wide ecological range as *Baetis rhodani*, *Centroptilum luteolum*, *Serratella ignita* and *Paraleptophlebia submarginata*, and species with a southern European distribution such as *Rhithrogena braaschi*. Another common species was *Baetis liebenauae*, previously recorded in smaller streams with a sandy
or stony bottom as well as in large lowland rivers, where it can be found as a habitat specialist on macrophytes (Buffagni et al. 2007, 2009, Bauernfeind and Soldán 2012). The presence of a stony bottom and submerged vegetation may be a suitable habitat combination for the species. Further research is required to determine the more specific preferences at the microhabitat scale and physico-chemical properties of the water. The mayfly species diversity is generally quite poor in lentic habitats, though certain taxa can be very abundant. The main reason why lakes clustered together and apart from other sites was due to their species composition consisting of taxa from lentic (e.g. Caenis horaria) or a wide range of habitat type preferences (e.g. Centroptilum luteolum, Ephemerida danica; Bauernfeind and Soldán 2012). Due to the presence and abundance of the species Baetis rhodani, B. cf. nubecularis, Centroptilum luteolum, Serratella ignita, Ephemerida danica and Paraleptophlebia submarginata, the lower streams in the Plitvice Lakes National Park (sites 84 and 85) grouped together with the tufa-barriers (see also in Vilenica et al. 2014).

In comparison with the neighbouring countries and with consideration of their surface areas, the Ephemeroptera diversity in Croatia could be characterised as relatively high. Together with Croatia, Bosnia and Herzegovina is also situated in Dinaric western Balkan ecoregion (ER5) (Illies 1978). However, as its mayfly fauna is currently poorly known, with only 52 species recorded, and as a large part of Croatian territory belongs to the Pannonian lowland ecoregion, to which most of the Hungarian territory also belongs, the Croatian mayfly fauna was found to be most similar to the Hungarian fauna (75%, Table 3). This is due to the presence of widely distributed species and of the species inhabiting the larger rivers. Even though the mayfly fauna of Bosnia and Herzegovina is currently poorly known, 65% of the species were similar to the Croatian fauna. Thus, it is possible that a much greater similarity between these countries can be expected in the future. Italy is divided into two completely different ecoregions than Croatia: Italy (ER3) and Alps (ER4) (Illies 1978). It had a much higher mayfly diversity and the lowest similarity with the Croatian mayfly assemblage (55%, Table 3). This is possibly due to its geographical position and large surface area that includes a great variety of geographical features and diverse habitats. For example, the Alps, which are not present in Croatia, are well-known for their mayfly diversity and endemism, especially in the genus Rhithrogena Eaton, 1881 (Vuataz et al. 2011).

Conclusions

As expected, this study revealed a higher number of mayfly taxa inhabiting Croatian freshwater habitats than known from the previous literature. As two of the most similar mayfly assemblages of the neighbouring countries have several taxa that could also inhabit Croatian habitats (e.g. Baetis vardarensis Ikonomov, 1962, Rhithrogena picteti Sowa, 1971, Leptophlebia marginata (Linnaeus, 1767), Ephemerella notata Eaton, 1887, Caenis luctuosa (Burmeister, 1839)) but were not yet recorded, due to the lack of systematic sampling in all seasons, future studies should include seasonal sampling
of a higher number of sites and habitat types. Additionally, the main focus should be on the eastern lowland part of the country, where a lower number of sites was visited during this study.

In the present study, some interesting taxa with restricted European and local distributions were recorded (e.g. *Rhithrogena* gr. *diaphana*, *Baetis* cf. *nubecularis* and *Ephemera* cf. *parnassiana*). Considering these species were recorded from a small number of sites in this study, they could be considered rare. Future studies on the taxonomic status, ecological features and detailed distribution of these species is necessary.

Additionally, as *Baetis liebenauae* was recorded on larger karstic rivers, a different habitat type than previously known, more detailed information on its preferences at the microhabitat scale and water physico-chemical properties should be investigated.

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