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Aspects regarding taxonomy, phylogeny and biogeography of the semiaquatic bugs of Romania and West-Palaearctic Region (Hemiptera: Heteroptera: Gerromorpha)

ABSTRACT

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1. Introduction

1.1. Infraorder Gerromorpha

1.1.1. General aspects regarding taxonomy and biology of the Romanian families

Gerromorpha Popov, 1971 represents an infraordin of semiaquatic bugs in the order Hemiptera, which, although it includes a relatively small number of taxa, is extremely diverse in terms of morphology, ecology or adaptations (Andersen 1982).

Semiaquatic bugs of the infraorder Gerromorpha are familiar inhabitants of water surfaces in all continents, except Antarctica (Andersen 1982; Andersen & Weir 2004b). Currently, the world fauna has more than 2000 describes taxa, classified in eight families and more than 160 genera. The Palaearctic Region has six families, 14 genera and approximately 120 species (Polhemus & Polhemus 2008).

Some specific biological characteristics and taxonomic details of the higher taxa of semiaquatic bugs occurring in Romania are presented below.

Family Gerridae

Family Gerridae, with eight subfamilies, comprise one of the largest families of the semiaquatic Heteroptera, and occupy a wide variety of habitats, from open ocean to high mountain rheocrenes. All species are predators, feeding on other arthropods that fall on the water surface. Most species overwinter as adults. Their life cycle has always five larval instars. Most species are wing-dimorphic (Andersen 1995b).

Family Gerridae has 104 genera and over 712 known species; 6 genera and 51 species are known in the Palaearctic Region (Polhemus & Polhemus 2008).

Family Hebridae

This family is cosmopolitan and they are often characterized as living on the surface of ponds, however most of the species actually live on the banks, or on steep rock surfaces. Hebrids overwinter as adults; life cycle comprises five larval instars, and many species are wing-dimorphic (Andersen 1995b).

Family Hebridae has 19 genera and over 224 species; 2 genera and 17 species are known in the Palaearctic Region (Kment *et al*. 2016; Polhemus & Polhemus 2008).

Family Hydrometridae

This family is cosmopolitan, but most prevalent in the tropics. Hydrometrids overwinter as adults; life cycle comprises five larval instars, and many species are wing-dimorphic (Andersen 1995b).

Family Hydrometridae has 14 genera and over 129 described taxa; 1 genus and 6 species are known in the Palaearctic Region (Polhemus & Polhemus 2008).

Family Mesoveliidae

The Mesoveliidae occur in a wide range of both lotic and lentic habitats. But most mesoveliid species are cryptic, living on banks, damp places, mosses, or on stones in the water. All species are predators of other arthropods. They overwinter as eggs inserted into plant tissue. Their life cycle comprises five (in some species four) larval instars. Most of the species are wing-dimorphic (Andersen 1995b).

Family Mesoveliidae has 20 genera and over 56 species; 1 genus and 3 species are known in the Palaearctic Region (Polhemus & Polhemus 2008; Jehamalar *et al*. 2019).

Family Veliidae

This is the largest family of semiaquatic Heteroptera, with six subfamilies that occupy an extremely wide variety of habitats from the intertidal zone to high mountain rheocrenes, and from secluded habitats to large streams. Some are negatively phototropic, hiding in holes under the banks during the day, emerging to feed at night (Polhemus 2009). All species are predators of small arthropods, including various stages of mosquitoes. Most species overwinter as adults. Life cycle comprises five, rarely four larval instars. Most species are wing-dimorphic too (Andersen 1995b).

Family Veliidae has 78 genera and over 903 described taxa; 3 genera and 44 species are known in the Palaearctic Region (Polhemus & Polhemus 2008).

In addition to the five families presented above, family Hermatobatidae, with one genus and 12 species described so far does also occur in the Palaearctic Region (cf. Andersen 1995b; Polhemus & Polhemus 2012). *Hermatobates* Carpenter, 1892, which is the only genus within this family, was originally placed into Gerridae. All the taxa are obligatorily marine (Andersen 1982; Polhemus & Polhemus 2012).

The families Macroveliidae and Paraphrynoveliidae does not occur in the Palaearctic Region. Macroveliidae is known from Nearctic Region (USA) and Neotropical Region (Chile), containing three monotypic genera. Paraphrynoveliidae has one genus and two described species in the Afrotropical Region (South Africa) (Andersen 1995b; Polhemus & Polhemus 2008).

1.1.2. Systematics and phylogenetic relationships

The heteropteran infraorder Gerromorpha (Štys & Kerzhner 1975) has been considered a distinct group since the time of Dufour (1833), who named the group “Amphibicorisae”, thus referring to their ability to walk on the surface of water, as distinct from the aquatic bugs, “Hydrocorisae” and the terrestrial bugs, “Geocorisae”. Andersen (1982) convincingly demonstrated that Gerromorpha was a monophyletic group, recognizing eight families: Mesoveliidae, Hebridae, Hydrometridae, Veliidae, Gerridae (prezente și în România), Paraphrynoveliidae, Macroveliidae and Hermatobatidae. Gerromorpha, with other six infraorders (e.g. Enicocephalomorpha, Dipsocoromorpha, Nepomorpha, Leptopodomorpha, Cimicomorpha și Pentatomomorpha) form the suborder Heteroptera, in order Hemiptera.

Phylogeny and higher classification of the semiaquatic bugs of infraorder Gerromorpha were addressed in the last decades by Andersen (1982), Andersen & Weir (2004b) and Damgaard (2008a).

Regarding the relationships of the eight families of Gerromorpha, Andersen (1982) proposes a cladogram in which the family Mesoveliidae represents the first branch, being a closely related group (sister group) with all other families. Hebridae is the second branch of the cladogram. In its analysis, the close relationship (sister group) between Macroveliidae and Hydrometridae is supported by the structure of the head and the dorsal position of the metathoracic spiracle. The Paraphrynoveliidae is a sister group of both families mentioned above. Veliidae and Gerridae are undoubtedly very closely related, as evidenced by several synapomorphies (e.g. the preapical position of the claws, etc.). The relationship of the Hermatobatidae is uncertain, this being probably a sister group with the last two families mentioned above. The cladogram proposed by Andersen (1982) still represents one of the most credible phylogenetic hypotheses.

Monophyly of the infraorder Gerromorpha was confirmed by Andersen (1982) and Damgaard (2008a). During the last few decades, systematics has undergone a remarkable revolution in implementing computer based analyses of DNA sequence data. A series of these studies have addressed phylogenetic relationships among the major gerromorphan lineages (e.g. Muraji & Tachikawa 2000; Andersen & Weir 2004b; Damgaard *et al*. 2005), and have been summarized by Damgaard (2008a) in an analysis of 64 morphological characters and 2.5 kb of DNA sequence data from the mitochondrial genes cytochrome c oxidase subunit I + II (COI + II) and large mitochondrial ribosomal subunit (16S rRNA) and the nuclear gene large nuclear ribosomal subunit (28S rRNA) obtained from representatives of all families and most subfamilies. This study confirmed Mesoveliidae as being the sister group to all other families of Gerromorpha and also the close relationship between the Gerridae and Veliidae, but suggested that the superfamilies Gerroidea (Hermatobatidae + Gerridae + Veliidae) and Hydrometroidea (Paraphrynoveliidae + Macroveliidae + Hydrometridae), and the family Veliidae, were paraphyletic entities.

Although various molecular studies have been published in the last decades, dealing with phylogenetic relationships within families, genera or species groups, and molecular systematics is an extremely promising discipline, it is clear that production, quality and homogenization of the molecular data, as well as in the interpretation of the results regarding semiaquatic bugs, is still at an early stage.

1.1.3. Biogeography

It is assumed that the Gerromorpha, together with other infraorders, originated in the early Mesozoic, at the end of Triassic (ca. 180 Ma) (Andersen 1982).

At present, the semiaquatic heteroptera occur on all continents, except Antarctica; they are more numerous in the tropical regions, although there are taxa adapted to the temperate or even cold areas.

The *Palaearctic Region* has 6 families and more than 120 species (Polhemus & Polhemus 2008; Kment *et al*. 2016). The most numerous are Gerridae and Veliidae (subfamiliy Veliinae).

The *Nearctic Region* is more or less equivalent in species reachness to the Palaearctic, and it has 6 families and more than 105 species (Polhemus & Polhemus 2008); one additional family (Macroveliidae) does occur in this region.

In the *Afrotropical Region*, more than 340 species in 7 families are known to occur (Polhemus & Polhemus 2008, 2012). This area has the world's highest species richness of Hebridae, with over 77 species; however, here the most numerous is family Veliidae, with more than 158 known taxa. Family Paraphrynoveliidae is endemic here.

The *Oriental Region* has the highest diversity of semiaquatic bugs, with over 600 known species in 6 families (Polhemus & Polhemus 2008, 2012); This area has the world's highest species richness of Gerridae, with over 287 described species.

The *Neotropical Region* is also very diverse, with over 515 known taxa in 7 families (Polhemus & Polhemus 2008, 2012). This area has the world's highest species richness of Veliidae (over 290 described taxa), Hydrometridae (over 37 known species) and Mesoveliidae (more than 15 species).

The *Australasian Region* has 6 families and approximately 325 species (Polhemus & Polhemus 2008, 2012), with high richness of Veliidae and Gerridae.

The *Pacific Region* has a low richness of semiaquatic bugs, with 5 families and approximately 20 known species (Polhemus & Polhemus 2008, 2012).

Gerridae, Hydrometridae, Mesoveliidae and Veliidae are cosmopolitan, with at least some representation in all the biogeographic regions except Antarctica (Polhemus & Polhemus 2008).

1.2. Scope, objectives and structure of the thesis

Clarification of the range of each species is a necessary prerequisite for future studies of their ecological niches, historical biogeography and correct interpretation of phylogenetic relationships. Nevertheless, Romania and many areas in southeastern Europe still represents a "black hole" (*sensu* Polhemus & Polhemus 2008) when referring to the semiaquatic bugs and their distributions.

Althought, systematics has undergone a remarkable revolution in implementing computer based analyses of DNA sequence data providing large amounts of data applicable that has greatly improved the possibility to build robust phylogenetic reconstructions at all taxonomic levels, there are still many groups of semiaquatic bugs that have not been addressed in this regard (e.g. Hebridae, Hydrometridae, Veliidae, etc.).

Thereby, starting from the gaps identified above, **the scope** of this study is to contribute to a better knowledge of the taxonomy, phylogeny and biogeography of some given groups of semiaquatic bugs (Insecta: Hemiptera: Heteroptera: Gerromorpha) from Romania and West-Palaearctic Region.

The thesis consists of nine chapters and the bibliography. The first chapter contains a general introduction. Chapters 2–7 contain their own results and are structured into introduction, materials and methods, results and discussions. Chapter eight is dedicated to a chorological analysis, and chapter nine is showing the final conclusions. The results were obtained between 2010 and 2018.

Objectives of the studies in chapters 2–7 are presented below:

Chapter 2 – update of the taxonomic list and distribution of the family Veliidae in Romania;

Chapter 3 – conturarea unei prime perspective detaliate asupra sistematicii şi biogeografiei speciilor Vest-Palearctice din genul *Velia*;

Chapter 4 – update of the taxonomic list and distribution of the family Gerridae in Romania, review of the distribution of *Gerris maculatus* Tamanini, 1946 and *G. gibbifer* Schummel, 1832 in southeastern Europe, and a short analysis of the allary dimorphism;

Chapter 5 – review of the taxonomic status of *H. fulvinervis* Horváth, 1929 and update of the taxonomic list and distribution of the family Hebridae in Romania;

Chapter 6 – update of the distribution of the family Hydrometridae in Romania;

Chapter 7 – update of the taxonomic list and distribution of the family Mesoveliidae in Romania and adjacent countries, and to investigate the phylogenetic relationships within and among the three species found in Europe.

***Key words*:** taxonomy, phylogeny, biogeography, heteroptera, gerridae, hebridae, hydrometridae, mesoveliidae, veliidae

2. Family Veliidae in Romania[1]

2.1. Introduction

The family Veliidae is the largest group of semiaquatic bugs (Hemiptera: Heteroptera: Gerromorpha) living in a wide variety of semiaquatic, marine, and even terrestrial habitats (Andersen 1982; Polhemus 2009; Andersen & Weir 2004a; Chen *et al*. 2005; Moreira *et al*. 2010).

The Romanian territory represents a transition zone between Mediterranean and Central European ecosystems (Doniță *et al*. 2005) and thus, it can harbor diverse fauna. Despite this, studies concerning Veliidae are scarce and they are considered rare in this country (cf. Davideanu 1999; Ilie & Davideanu 2007; Ilie 2009). So far, two genera and seven species have been recorded (*Microvelia* – 2 species, *Velia* – 5 species) (Paina 1975; Davideanu 1999; Ilie 2009).

The aim of this paper is to provide an updated checklist and distribution of the Veliidae in Romania, and to fill gaps in the biogeographic patterns of this group.

2.2. Material and methods

Unless otherwise stated in material examined, all the material was collected by the first author between 2011 and 2014. In addition, some museum or private collections were also checked. Most of the specimens are preserved in 70% ethanol, while a minor part is dry mounted. The distribution maps have been made based on own collected material, revised data from other collections and reliable bibliographic references by using ArcGIS 10.2 (ESRI 2011).

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[1] Data from this chapter were published in: **Berchi, G.M.** & Kment, P. (2015) Review of the family Veliidae in Romania (Hemiptera: Heteroptera: Gerromorpha). *Zootaxa*, 3963 (1), 74–88.

2.3. Results

Review of the species

During this study 311 samples of aquatic bugs were collected across the entire territory of Romania. Of these, 64 samples contained specimens of *Velia* and 44 samples included specimens of *Microvelia*.

Velia serbica and *Microvelia buenoi* are here recorded for the first time from our country. Occurrence of *V. mancinii mancinii* is confirmed through additional records. Occurrences of *Velia currens* and *V. rivulorum*, which have been countinously reported from our country, were not confirmed during this study, analysis of specimens preserved in various collections showing that they belong to some other taxa (e.g. *V. caprai caprai*). Both latter taxa are excluded from the Romanian fauna.

2.4. Discussions

In this study, *Microvelia* species were found at low altitudes, in similar habitats represented by the banks of slow flowing streams, pools, ponds or swamps with aquatic vegetation. Contrastingly, *Velia* species were collected from lotic habitats in mountainous regions, but several samples were taken from hilly areas as well.

In Romania, many records of both *V. currens* and *V. rivulorum* are old, dating prior to Tamanini's (1947) revision. We have checked Horváth's specimens of *V. rivulorum* collected in 1873 and 1888 from Baziaș and preserved in HNHM; all the specimens are *V. mancinii mancinii*, which is in accordance with Benedek (1969b, 1970). Furthermore, we have checked Ilie's specimens identified as *V. rivulorum* (Ilie 2005, 2009) and they turned out to be *V. caprai caprai*.

Macropterous specimens were certainly rarer in our samples of Veliinae, but we observed that they were more frequent when the population density was higher at the sampling site; the number of winged specimens also increased when they occurred in intermittent streams.

3. Phylogeny and biogeography of the West-Palaearctic *Velia*[2]

3.1. Introduction

Water crickets of the genus *Velia* Latreille, 1804 (Hemiptera: Heteroptera: Gerromorpha: Veliidae) are common semiaquatic bugs from streams and rivers throughout the temperate and subtropical parts of the Palaearctic region (Andersen 1995b).

Biogeographically, most of the diversity of the West-Palaearctic *Velia* is concentrated in the circum-Mediterranean area, with only a few species occurring outside this region. Only *Velia* (*P*.) *caprai caprai* Tamanini, 1947 and *V*. (*P*.) *saulii* Tamanini, 1947 are known from the northern temperate area of the Western Palaearctic (Andersen 1995b; Damgaard 1997), and the polytypic *V*. (*P*.) *affinis* Kolenati, 1857 has the widest distribution range, occurring from the Italian Peninsula to Pakistan and northwestern India (Tamanini 1953). The major Mediterranean islands as well as Madeira and Canaries are usually inhabited by endemic species (Tamanini 1947, 1951, 1954, 1979; Csabai *et al*. 2017). The majority of the other taxa are restricted to the Iberian, Italian and Balkan peninsulas, and to northwestern Africa (Maghreb), which are well known as biodiversity hotspots and glacial refugia for Western Palaearctic biotas (Hewitt 2000; Husemann *et al*. 2014).

The phylogeny and biogeography of *Velia* are poorly known due to a scarcity of data (both distributional and molecular) and the morphological similarity among its species, which not only hampered routine identifications (Berchi & Kment 2015; Csabai *et al*. 2017), but also limited the use of morphological characters in a cladistic context.

Therefore, this study aims to provide a first comprehensive insight into the molecular systematics and biogeography of the Western Palaearctic *Velia*.

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[2] Data from this chapter were published in: **Berchi, G.M.**, Copilaş-Ciocianu, D., Kment, P., Buzzetti, F.M., Petrusek, A., Rákosy, L., Cianferoni, F. & Damgaard, J. (2018a) Molecular phylogeny and biogeography of the West-Palaearctic *Velia* (Heteroptera: Gerromorpha: Veliidae). *Systematic Entomology*, 43 (2), 262–276;
respectively: Csabai, Z., Soós, N., **Berchi, G.M.**, Cianferoni, F., Boda, P. & Mora, A. (2017) Aquatic and semiaquatic Heteroptera (Nepomorpha and Gerromorpha) fauna of Greek holiday islands (Rhodes, Crete and Corfu) with first records of three species from Europe and Greece. *Zootaxa*, 4231 (1), 51–69.

3.2. Material and methods

Taxa covered, sampling and mapping

We obtained material from 23 morphologically identified taxa presently assigned to the West-Palaearctic *Velia* (79% of all currently known taxa) as follows: subgenus *Plesiovelia* – 20 species and two subspecies; subgenus *Velia* (s. str.) – 1 species.

Molecular protocols, sequence alignment and dataset assembly

Genomic DNA was extracted using the Genomic DNA Mini Kit for tissue (Geneaid Biotech, Taipei). We sequenced four mitochondrial markers: cytochrome oxidase subunits I and II (COI +COII), including the leucine transfer RNA (tRNA-Leu), and the large mitochondrial ribosomal subunit (16S rRNA); and one nuclear marker: the large nuclear ribosomal subunit (*28S* rRNA). Primers and PCR protocols followed Damgaard *et al*. (2012).

Phylogenetic analyses

The number of variable and parsimony informative sites for each marker was calculated with MEGA 6. Loss of phylogenetic signal in the COI, COII and 16S markers was inspected by using the substitution saturation index of Xia *et al*. (2003) implemented in DAMBE 5.3 (Xia & Xie 2003).

Molecular clock calibration

Divergence times were estimated with BEAST 1.8.0 (Drummond *et al*. 2012) using the same evolutionary models as for the MrBayes analysis. Strict versus relaxed molecular clocks were tested by using path/stepping stone sampling to estimate their marginal likelihoods (Baele *et al*. 2012, 2013) and Bayes factors (BF) were calculated in TRACER.

Ancestral range reconstruction

In order to evaluate the origin and dispersal patterns of the West-Palaearctic *Velia*, we conducted a Dispersal-Extinction-Cladogenesis analysis (DEC; Ree & Smith 2008) implemented in RASP 3.2 (Yu *et al*. 2015).

Diversification rate analyses

We examined if the diversification of the West-Palaearctic *Velia* varied over time. In order to visualize changes in diversification rates, we constructed a lineage through time plot (LTT; Nee *et al*. 1994) with TRACER using 50 000 post burn-in trees from the BEAST analysis.

Haplotype networks

Haplotype networks were built for groups of closely related taxa in order to explore their patterns of intraspecific diversity and examine their relationships in more detail. For this analysis we used a concatenated alignment consisting of COI and 16S because these markers were best represented in our dataset.

3.3. Results

Sequence data

We obtained altogether 250 new sequences (68 for COI, 65 for tRNA-Leu+COII, 89 for 16S, and 28 for 28S) from 91 specimens of *Velia*. The alignment lengths of the protein-coding COI and COII markers were 821 and 497 bp, respectively, and the tRNA-Leu fragment had a length of 54 bp. Several insertions present in the outgroup taxa were removed with GBLOCKS, resulting in alignments that had lengths of 465 and 481 bp for 16S and 28S, respectively (98 and 92% of the original positions). The final concatenated alignment had a total length of 2318 bp.

Phylogenetic analyses and distribution of clades

The number of variable and parsimony informative sites per marker was: 324/263 for COI, 227/179 for COII, 175/127 for 16S, 15/9 for tRNA-Leu and 85/52 for 28S. Uncorrected *p*-distances among ingroup taxa varied from 0.5 to 11.8% at COI, 0.6 to 12% at COII, 0 to 6.2% at 16S and 0 to 1.2% at 28S.

Both the ML and BI approaches to phylogenetic reconstruction yielded almost identical topologies with most of the ingroup clades receiving high support. Both analyses recovered four strongly supported major clades, three belonging to the subgenus *Plesiovelia* (Clades 1, 2 and

3) and one to the subgenus *Velia* (s. str.) (Clade 4). Clades 1, 3 and 4 are broadly distributed in the western part of the Western Palaearctic region, whereas Clade 2 has an eastern distribution.

Molecular dating

The topology of the chronogram was almost identical with the previous methods, differing only at poorly supported nodes. The majority of the nodes received maximum support. However, the position of Clade 3 fluctuated in initial runs and it was therefore constrained to monophyly with Clade 2, as this relationship was recovered in the ML and BI analyses, albeit with low support.

Ancestral range reconstruction

The DEC model supported a Balkan origin of *Velia* (*Plesiovelia*) with a high degree of probability (65%), however the origin of the clade that contains both *Velia* (*Plesiovelia*) and *Velia* (s. str.) remains less clear (11%, 10% și 7% probability of a Balkan–northwestern African, Balkan, and Balkan–Italian origin, respectively). Clade 1 presumably originated in northwestern Africa and the Balkans (32% probability), Clades 2 and 3 are both supported as having a Balkan origin (61% and 58% probability, respectively).

Diversification rate analyses

The LTT plot indicated that the rate of lineage accumulation through time was rather constant with no obvious shifts.

Haplotype networks

A modest amount of variation was observed within most of the analysed taxa at the concatenated COI +16S mitochondrial alignment. The majority of intraspecific haplotypes differed by one to 10 mutational steps. However, a substantial amount of divergence was observed within the *V*. (*P*.) *serbica* 2 clade, where each analysed population had distinct endemic haplotypes that differed by up to 29 mutations from other populations.

3.4. Discussions

Phylogeny and taxonomy

The analysis revealed that theWest-Palaearctic *Velia* is monophyletic and its two subgenera (*Velia* s. str. and *Plesiovelia*) were recovered as sister clades. Although the phylogenetic position of the subgenus *Cesavelia* is unknown, it is probably a sister group to the clade containing *Velia* (s. str) and *Velia* (*Plesiovelia*) due to its relatively shorter middle legs, a character state which is considered plesiomorphic (Andersen 1981, 1982).

The molecular phylogeny revealed that most analysed *Velia* taxa are phylogenetically distinct and as such, it agrees well with the morphological taxonomy of the group as previously defined. Our data indicate that *V.* (*P.*) *serbica* is paraphyletic, because *V.* (*P.*) *mancinii mancinii* is nested between its two clades, which are remarkably similar in morphology.

Biogeography of the subgenus Plesiovelia

The ancestral area reconstruction analysis suggests that although *Velia* (*Plesiovelia*) has a strongly supported southeastern European origin, considerable uncertainty exists about the origin of the *Velia-Plesiovelia* clade.

The biogeographical reconstruction and molecular dating analyses indicate that the deepest splits between extant *Velia* (*Plesiovelia*) clades occurred in southeastern Europe between ca. 32 and 15 Ma – thus between the early Oligocene (based on fossil calibrations) and the middle Miocene (based on COI rates). Although the two molecular clock calibration approaches give rather different estimations, they are consistent in that *Velia* (*Plesiovelia* diversified mainly during the Neogene. Nevertheless, we consider the results of the fossil calibration more plausible, as they are based on tangible evidence rather than assumptions, and have narrower confidence intervals.

During the Neogene, southeastern Europe was a geologically dynamic archipelago, characterized by intense uplift movements due to the collision of the African and Eurasian tectonic plates (Alpine orogeny) (Rögl 1998; Meulenkamp & Sissingh 2003; Popov *et al*. 2004). From that area, the subgenus subsequently radiated to the west (Clade 1 and 3) and east (Clade 2) along the fragmented landmasses of southern Europe and Asia Minor. Similar biogeographical patterns consisting of deep splits between western and eastern clades have been described in the aquatic beetle genera *Deronectes* Sharp, 1882 (Dytiscidae) and *Hydrochus* Leach, 1817 (Hydrochidae) (Hidalgo-Galiana & Ribera 2011; García-Vázquez *et al*. 2016).

The unusually high intraspecific genetic divergence, despite geographical proximity of localities, observed within the lineages of *V*. (*P*.) *serbica* is apparently unique among its congeners. One plausible explanation for this observation is an inefficient dispersal of V. (P.) serbica that promotes diversification; the macropterous morph seems to be absent or extremely rare in these lineages. Another explanation might be that these patterns of microendemicity possibly reflect *in situ* survival throughout the Pleistocene glaciations in microrefugia (*sensu* Rull 2009).

4. Family Gerridae in Romania with a review of the distribution of *Gerris gibbifer* and *G*. *maculatus* in southeastern Europe[3]

4.1. Introduction

The water striders (Heteroptera: Gerridae) are common and well-known inhabitants of lentic and lotic freshwater habitats (Andersen 1982; Spence & Andersen 1994), and include about 70 genera and more than 750 species worldwide (Polhemus & Polhemus 2008). The species diversity in Europe declines from south to north (Damgaard *et al*. 2014), and phylogeographical studies have shown the same trend in genetic variation within several species (Damgaard 2008c). However, whereas the fauna of Gerridae is relatively well-known in northern and central Europe, the situation is less clear in southeastern and eastern Europe, where many areas are undersampled or knowledge is based mainly on old and dubious records.

The aim of this study is to provide a new and updated record of the distribution of Gerridae in Romania by extensive sampling throughout the country and by critically revising older

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[3] Data from this chapter were published in: **Berchi, G.M.**, Cianferoni, F. Csabai, Z., Damgaard, J., Olosutean, H., Ilie, D.M., Boda, P. & Kment, P. (2018b) Water striders (Heteroptera: Gerromorpha: Gerridae) of Romania with an update on the distribution of *Gerris gibbifer* and *G*. *maculatus* in south-eastern Europe. *Zootaxa*, 4433 (3), 491–519.

records. To aid the revisionary work on the allopatric *Gerris maculatus* Tamanini, 1946 and *G. gibbifer* Schummel, 1832 in southeastern Europe, additional material originating from several museum and private collections has been revised, resulting in dozens of new distributional records and corrections of previous records.

4.2. Material and methods

This study is based on data originating from i) our own extensive field survey, ii) museum and private collections, and iii) published data. Most of the material was collected by the senior author during 2010–2015 and in 2017, unless otherwise stated. The specimens were collected by using a water net and are stored in 70% ethanol in the G. M. Berchi Collection, Cluj-Napoca, Romania (GBCR); a few bugs are dry mounted in the same collection.

4.3. Results

In Romania, 11 species of gerrids were identified at 275 sites (261 sites derived from our field survey, and 14 sites originated from museum collections) from lowlands to high mountainous areas. By far the most frequently occurring species was *Gerris lacustris* Linnaeus, 1758, which was recorded from 143 sites (52.0%). Other frequent taxa were *Aquarius paludum paludum* Fabricius, 1794 recorded from 82 sites (29.8%), *G. thoracicus* Schummel, 1832 recorded from 49 sites (17.8%), and *G. costae fieberi* Stichel, 1938 recorded from 47 sites (17.1%). The least frequent species were *G. lateralis* Schummel, 1832 and *Limnoporus rufoscutellatus* Latreille, 1807, which have been recorded from 10 (3.6%) and seven sites (2.5%), respectively.

Aquarius najas De Geer, 1773 and *Gerris asper* Fieber, 1860 were collected only in the lowlands (<500 m a.s.l.), whereas *G. lateralis* was found only at higher elevations (>700 m a.s.l.). In contrast, *A. p. paludum*, *G. argentatus* Schummel, 1832, *G. costae fieberi*, *G. lacustris*, *G. maculatus* Tamanini, 1946, *G. odontogaster* Zetterstedt, 1828, *G. thoracicus* and *L. rufoscutellatus* have a wide elevational distribution.

L. rufoscutellatus, *G. costae fieberi*, *G. maculatus* and *G. thoracicus* always occurred as the longwinged (macropterous) morph in our samples, whereas populations of *A. p. paludum*, *G. lacustris*, *G. asper* and *G. lateralis* commonly were found containing both long-winged and short-winged adults. For *A. najas*, the macropterous morph was found in only six percent of the specimens.

4.4. Discussions

Alary polymorphism and habitat preferences

Althought some taxa, such as *L. rufoscutellatus*, *G. costae fieberi*, *G. maculatus* and *G. thoracicus* are always long winged (macropterous) in their adult stage in our samples, the Gerridae are well known for their wing polymorphism, meaning that the same population can include both long-winged (macropterous) adults and adults with more (apterous or micropterous) or less (brachypterous) shortened wings (Andersen 1982). For the Romanian fauna, populations of *A. p. paludum*, *G. lacustris, G. asper* şi *G. lateralis* are commonly found in both long-winged and short-winged morphs, whereas in *A. najas*, the apterous morph is prevalent.

Gerris gibbifer and G. maculatus in southeastern Europe

Most of the taxa collected through this faunistic survey are common and widely distributed in Europe and have been documented from Romania as well, with the single exception of *Gerris maculatus* Tamanini, 1946. This species was reported as present in Romania only by Andersen (1995b), although without further details or exact localities. In contrast, *G. gibbifer* Schummel, 1832 has been recorded repeatedly from this country.

The study shows that there are no confirmed records of *G. gibbifer* in Romania, Serbia, Macedonia and Slovenia; therefore, we propose its exclusion from the faunal lists of Romania, Serbia, and Macedonia. Its occurrence in Slovenia is probable (especially in the Alps), since it is present in all the neighboring countries. Therefore, both *G. gibbifer* and *G. maculatus* occur in Croatia, Hungary, Ukraine and probably Slovenia, but mainly in different parts of the countries.

5. Family Hebridae in Romania with a redescription of *Hebrus fulvinervis*[4]

5.1. Introduction

Velvet water bugs or sphagnum bugs (Hebridae) are a small group of minute water bugs represented in the World fauna by two subfamilies, 20 genera and about 220 species, most of which are accommodated in the cosmpolitan genus *Hebrus* Curtis, 1833 with about 140 described species (Kment *et al*. 2016). These insects are predaceous, living in humid terrestrial habitats (litter, moss), marginal aquatic habitats (sphagnum bogs, hygropetric habitats, intertidal habitats in mangroves), or rarely on water surfaces covered with floating plants. The Hebridae fauna of the West-Palaearctic Region is less diverse, including only a single native genus, *Hebrus*, represented by three subgenera (*Hebrus* s. str., *Hebrusella* Poisson, 1944, *Paratimasius* Poisson, 1952) and *Merragata hebroides* White, 1877, an American species of Hebrinae which occurs in the Canary Islands (Andersen 1995b).

In Romania, family Hebridae is one of the most neglected groups of the aquatic and semiaquatic bugs. Even if it does not completely solve the problem of their distribution in Romania, the paper supports the recent efforts on the study of aquatic and semiaquatic heteropterans in our country (e.g. Berchi 2013; Berchi & Kment 2015; Berchi *et al*. 2016, 2018b), aiming to revise the taxonomic status of *H. fulvinervis* Horváth, 1929, a species not recorded since 1908 (Horváth 1929; Benedek 1970) and completing the knowledge regarding the distribution of this group.

5.2. Material and methods

Measurements were taken using a Leica MZ75 stereomicroscope provided with an ocular micrometer and subsequently standardized to absolute values. Some specimens were examined using a Hitachi S-3700N environmental scanning electron microscope at the Department of

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[4] Data from this chapter were published in: Kment, P., Jindra, Z. & **Berchi, G.M.** (2016) Review of West-Palaearctic Hebridae with description of a new species and redescription of *Hebrus fulvinervis* (Hemiptera: Heteroptera). *Zootaxa*, 4147 (3), 201–239.

Paleontology, National Museum, Praha. Habitus photographs were taken using a Canon MP-E 65 mm macro lens attached to a Canon EOS 550D camera; they were stacked from multiple layers using the Helicon Focus 5.1 Pro software. Other photographs were taken using an Olympus BX44 microscope with an attached Canon EOS 1100D camera. The morphological terminology follows mostly Andersen (1982).

5.3. Results

Review of the species

The current knowledge regarding the Hebridae in Romania, as also in the West-Palearctic region is far from satisfactory.

Although described almost a century ago, *Hebrus fulvinervis* Horváth, 1929 remains in the taxonomical list of our country through Horváth's (1929) first and only reports from: Cruce (Suceava), Râu de Mori (Hunedoara) and Sibiu (Sibiu). The species is redescribed; its taxonomic status is confirmed, as well of the occurrence of the other three species previously reported in our country.

5.4. Discussions

The subgeneric division of the cosmopolitan *Hebrus* into six subgenera (*Hebrus* s. str., *Hebrusella*, *Paratimasiellus* Poisson, 1956, *Paratimasius*, *Subhebrus* Poisson, 1956, *Timasielloides* Poisson, 1952) was established by Poisson (1944, 1952, 1956) based only on West-Palaearctic, Afrotropical and Madagascar taxa, and is hardly satisfactory for application on the global diversity of the genus. The recent discovery of the apterous *H. franzi* having antennal segment IV not subdivided (Cianferoni *et al*. 2016), as well of *Hebrus atlas* Kment, Jindra & Berchi, 2016 and *H. syriacus* resembling members of *Hebrusella*, but sharing the antennal segment IV subdivided by a membranous joint as in *Hebrus* s. str., only underscores the unsatisfactory nature of the subgeneric classification. The situation requires a deeper phylogenetic study of the entire family using both morphological and molecular characters (Kment *et al*. 2016).

6. Family Hydrometridae in Romania

6.1. Introduction

Members of the family Hydrometridae are semiaquatic insects that live predominantly on the surface of both lentic and lotic ecosystems, on all continents, except Antarctica. At present, seven genera and approximately 125 species of Hydrometridae are known worldwide (Polhemus 2009). The cosmopolitan genus *Hydrometra* Latreille, 1797 includes most of the species, including the European ones: *Hydrometra gracilenta* Horváth, 1899 and *H. stagnorum* Linnaeus, 1758 (Andersen 1995b). Both taxa are known to occur in Romania, since Fuss (1862) and Horváth (1878, 1899).

Data referring to the distribution of Hydrometridae in Romania were published in various scattered papers, generally concentrated at local level, and summarized in his work by Paina (1975).

The aim of this chapter is to provide an updated distribution of the Hydrometridae in Romania, and to fill gaps in the biogeographic patterns of this group.

6.2. Material and methods

Unless otherwise stated in the material examined, most of the biological material was collected by the author in Romania, between 2010–2015 and in 2017.

Specimens were collected and preserved as in the previous chapters.

6.3. Results

List of taxa

During this study, members of *Hydrometra* Latreille, 1797 were collected at 45 sampling sites. *Hydrometra gracilenta* Horváth, 1899 was identified in only four samples, while *H. stagnorum* Linnaeus, 1758 was present at the remaining ones, with no co-occurrences. The short-winged morph seems to be much frequent in Hydrometridae; all the specimens of *H. gracilenta* were micropterous and only 5% of specimens of *H. stagnorum* were macropterous.

6.4. Discussions

Hydrometra gracilenta was known for a long time in our country only through Horváth's (1899, 1907) papers. It has been and it still is considered one of the rarest semiaquatic heteropteran species, with a low abundance (Paina 1987), but in the last decades, its presence has been reported from several additional localities in our country (Paina 1987; Ilie 2001, 2007; Ilie & Davideanu 2002a; Ilie & Ban-Calefariu 2010; Berchi et al. 2011; Olosutean & Ilie 2013a; this paper), suggesting a low sampling effort, rather than a possible recent expansion of this taxon. The species is currently known from several localities in Transylvania, Banat, Oltenia and Dobrogea. In contrast, *H*. *stagnorum* is one of the most common semiaquatic bug in our country. It was first reported by Fuss (1862), being known today in all regions of the country, except Maramureș. Both species live in lowland and hilly areas. The highest elevation for for *H*. *gracilenta* was of 111 m a.s.l., while for H. stagnorum was 516 m a.s.l. They can be found in both lotic and lentic habitats, overwintering as imago (Paina 1986, 1987; this paper). In Romania, important and interesting data on the biology of the two species were published by Paina (1986, 1987).

The phylogenetic relationships within the family Hydrometridae have not yet been addressed, but according to preliminary data it appears that *H*. *stagnorum* and *H*. *gracilenta* are not closely related (Berchi *et al*. in prep.).

7. Mesoveliidae of Romania and adjacent countries and their phylogenetic relationships[5]

7.1. Introduction

Members of the family Mesoveliidae (Hemiptera: Heteroptera: Gerromorpha) are common inhabitants of margins of ponds and lakes, but a number of species are confined to hygropetric habitats along streams and rivers or live among mosses and leaves on the floor of tropical rain forests (Andersen 1982). The Mesoveliidae have retained many primitive features, such as a welldeveloped female ovipositor for embedding eggs in the substrate, and insertion of their coxae close to the midline of the thorax (Andersen 1982). Within Gerromorpha, Mesoveliidae is placed as a sister group to all the remaining seven currently recognized families (Gerridae, Veliidae, Hydrometridae, Hermatobatidae, Macroveliidae, Paraphrynoveliidae, and Hebridae), and is foremost diagnosed on the presence of an ejaculatory bulb and pump of the male genital tract and the lack of an embryonic egg burster (Damgaard 2008a).

7.2. Material and methods

Sampling and imaging

Unless otherwise stated in material examined, the biological material for this survey was collected by the author between 2011 and 2015 in Bulgaria, Croatia, Romania, and Serbia.

Molecular methods and analyses

In order to explore the phylogenetic position of *M. thermalis* within the genus, we sequenced three mitochondrial and one nuclear markers, namely the cytochrome oxidase subunits I and II (COI+II), the large mitochondrial ribosomal subunit (16S rRNA), and the large nuclear ribosomal subunit (28S rRNA). Total genomic DNA was extracted from ethanol-preserved specimens using the Genomic DNA Tissue Mini Kit (Geneaid Biotech) according to the

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[5] Data from this chapter were published in: **Berchi, G.M.**, Kment, P., Copilaș-Ciocianu, D., Rákosy, L. & Damgaard, J. (2016) Water treaders of Romania and adjacent countries and their phylogenetic relationships (Hemiptera: Heteroptera: Mesoveliidae). *Annales Zoologici*, 66 (2), 193–212.

manufacturer's protocol. Genes were amplified by polymerase chain reaction (PCR) using the primers and protocols of Damgaard *et al*. (2012).

The protein coding genes COI and COII were concatenated as a single locus, aligned using MUSCLE (Edgar 2004) as implemented in MEGA 6 (Tamura *et al*. 2013) and translated into amino acids in order to check for stop codons that would indicate the presence of pseudogenes. Ribosomal genes 16S and 28S were aligned using MAFFT 7 (Katoh & Standley 2013). Phylogenetic analyses were performed on the concatenated data set using maximum-likelihood (ML) and Bayesian inference (BI). Molecular dating was performed with BEAST 1.8 (Drummond *et al*. 2012) using the concatenated dataset under an uncorrelated relaxed molecular clock model with a lognormal distribution (Drummond *et al*. 2006).

7.3. Results

Review of the distribution in Romania and adjacent countries

During this study, 155 individuals of the genus *Mesovelia* were collected at 51 localities from Bulgaria, Croatia, Romania, and Serbia. *Mesovelia furcata* was identified at 22 sampling sites, *M. thermalis* at 35, while *M. vittigera* was collected from two localities on the Black Sea coast (Burgas region, southeastern Bulgaria). At 8 sampling sites, *M. furcata* and *M. thermalis* were found sympatrically. In terms of wing morphs, the macropterous/apterous ratio was: *M. furcata* 0.018 : 0.982 (total 1/55), *M. thermalis* 0.083 : 0.917 (total 7/84) and *M. vittigera* 0 : 1 (total 0/8).

Based on our data, this species is widespread in the southern and western lowlands of Romania, but its presence in Transylvania (central Romania) and eastern Romania cannot be excluded. At the European level, our new records from Bulgaria, Croatia, Romania, and Serbia considerably extend the south-western limits of its distribution, effectively doubling its range. *Mesovelia vittigera* was described from Egypt (Horváth 1895), and for a long period was known in Europe only from Elbasan in Albania (Horváth 1924). In 1980 it was reported from Montenegro (Štusák 1980). Further records came from the mainland of Greece (Zimmermann 1982), Bulgaria (Josifov 1986b) and Croatia (Dalmatia) (Kment 2001; Damgaard *et al*. 2012) mostly from the coastal areas, with a Mediterranean climate. *Mesovelia furcata* is a Euro-

Siberian element, occurring in most of the European countries (Andersen 1995b). Its distribution range overlaps with that of both *M. thermalis* and *M. vittigera*.

Molecular analyses

The concatenated supermatrix contained 45 taxa and had a length of 2396 bp. Both ML and BI analyses recovered a well supported tree, with the exception of a few basal nodes, and revealed that *M. thermalis* is a possible sister species to *M. furcata*. The molecular dating analysis indicates that the crown age of *Mesovelia* is ca. 22.8 Ma (95% HPD intervals: 17–35.9 Ma). The last common ancestor of *M. thermalis* and *M. furcata* likely lived ca. 4.7 Ma ago (95% HPD intervals: 2.23–7.98 Ma).

7.4. Discussions

Mesovelia species were collected at low altitudes, along the shores of slow flowing lowland rivers, but also from pools and ponds with aquatic vegetation (*Elodea*, *Lemna*, *Nymphaea*, *Phragmites*, *Potamogeton*, *Spirodela*, *Typha* spp.). The highest altitude where *M. furcata* was collected was 370 m a.s.l., while for *M. thermalis* the maximum was 163 m a.s.l. We have found active adult specimens of *M. thermalis* between the first half of May and the end of September, often co-habiting with *M. furcata*.

The phylogenetic relationships

The phylogenetic results are very similar to Damgaard *et al*. (2012), except that *M. amoena* Uhler, 1894 is not a sister-species to *Speovelia maritima* Esaki, 1929; however, branches in both studies are poorly-supported. As pointed out in Damgaard *et al*. (2012), the remaining species of *Mesovelia* are divided into two groups, one including *M. vittigera*, *M. stysi* Polhemus & Polhemus, 2000, *M. furcata*, *M. thermalis*, *M. hungerfordi* Hale, 1926, *M. ebbenielseni* Andersen & Weir, 2004 and *M. mulsanti* White, 1879; and one including *M. horvathi* Lundblad, 1933, *M. hackeri* Harris & Drake, 1941, and two undescribed species from Nigeria (no. 2247) and New Caledonia (nos 2276 and 2279). Furthermore, Damgaard *et al*. (2012) indicated cryptic speciation in the widespread *M. vittigera*, *M. mulsanti* and *M. horvathi*. Our phylogenetic results indicate genetic differences between European and Asian individuals of *M. thermalis*. However, we only had a 28S sequence from the Chinese individual (2652) for comparison. Additionaly, we analysed the 16S and 28S sequences from Muraji & Tachikawa (2000) (GenBank accession numbers AB026617 and AB034767) labeled as "*M. vittigera*", and found that the specimen is more likely to belong to *M. thermalis* (results not shown). Therefore, a more detailed analysis encompassing a comprehensive sampling of these species across their distribution ranges should be carried out in order to fully understand their patterns of genetic diversity.

8. Checklist of the semiaquatic bugs of Romania with a chorological analysis

Thus, this work cumulates the results of a continuous work on updating the distribution of the semiaquatic heteroptera in Romania and southeastern Europe, an action started several years ago, which has shown interesting results from faunistic, taxonomic and biogeographical point of view (e.g. Berchi 2011, 2013; Berchi *et al*. 2012, 2016, 2018b; Berchi & Kment 2015; Kment *et al*. 2016).

In these past years, taxa such as *Velia currens* (Fabricius, 1794), *V. rivulorum* (Fabricius, 1775) (both Veliidae) and *Mesovelia vittigera* Horváth, 1895 (Mesoveliidae) were excluded from the faunal list of Romania, while *Microvelia buenoi* Drake, 1920 and *Velia serbica* Tamanini, 1951 (both Veliidae) were reported as new faunistic elements (see Berchi & Kment 2015; Berchi *et al*. 2016). Occurrences of *Velia mancinii mancinii* Tamanini, 1947, *Hebrus fulvinervis* Horváth, 1929, *H. montanus* Kolenati, 1857 and *G. maculatus* were confirmed too; the neglected *H. fulvinervis* was confirmed as valid species and redescribed (Berchi & Kment 2015; Kment *et al*. 2016). Finally, *G. gibbifer* was recently added to the list of excluded taxa (Berchi *et al*. 2018b). In total, 27 taxa of semiaquatic bugs (Gerromorpha) are presently known to occur in Romania, a considerably higher number when comparing to the other countries in our region (Slovenia – 20 sp., Slovakia – 20 sp., Hungary – 21 sp., Austria – 22 sp., Croatia – 22 sp., Serbia – 23 sp., Ukraine – 24 sp.) (Boda *et al*. 2015). This fact, underlines once again the importance of the Carpathians as hotspot for biodiversity (Bálint *et al*. 2011; Theissinger *et al*. 2013).

The chorotype analysis (including codes and major groups of chorotypes) follows Vigna Taglianti *et al*. (1999). A new 4-letter code ("BALK") is used for Balkan endemic taxa.

Chorotype abbreviations: ASE = Asiatic-European; BALK = Balkan endemic; EUM = Europeo-Mediterranean; EUR = European; OLA = Holarctic; PAL = Palaearctic; SEU = South-European; SIE = Sibero-European; TEM = Turano-Europeo-Mediterranean; TUE = Turano-European; WPA = West-Palaearctic.

According to the chorotype analysis, the gerromorphan fauna of Romania shows a high percentage (85.2%) of species widespread in the Holarctic Region, whilst only 14.8% of the species are restricted to Europe. This latter group also includes two Balkan endemic species, *Hebrus fulvinervis* Horváth, 1929 and *Velia serbica* Tamanini, 1951 which, along with *G. maculatus*, represent the most interesting elements of a mainly continental fauna.

Checklist of the semiaquatic bugs (Gerromorpha) occurring in Romania (cf. Paina, 1975; Berchi & Kment 2015; Berchi *et al*. 2016, 2018a,b; Kment *et al*. 2016). For chorotype, see the abbreviations list.

Taxa	**Chorotype**
Infraorder GERROMORPHA Popov, 1971	
Family MESOVELIIDAE Douglas & Scott, 1867	
Mesovelia furcata Mulsant & Rey, 1852	SIE
Mesovelia thermalis Horváth, 1915	ASE
Family HEBRIDAE Amyot & Serville, 1843	
Hebrus (*Hebrus*) *montanus* Kolenati, 1857	TUE
Hebrus (*Hebrus*) *pusillus pusillus* (Fallén, 1807)	PAL
Hebrus (*Hebrusella*) *fulvinervis* Horváth, 1929	SEU (BALK)
Hebrus (*Hebrusella*) *ruficeps* Thomson, 1871	SIE
Family HYDROMETRIDAE Billberg, 1820	
Hydrometra gracilenta Horváth, 1899	PAL
Hydrometra stagnorum (Linnaeus, 1758)	TEM
Family VELIIDAE Brullé, 1836	
Microvelia (*Microvelia*) *buenoi* Drake, 1920	OLA
Microvelia (*Microvelia*) *reticulata* (Burmeister, 1835)	SIE
Microvelia (*Picaultia*) *pygmaea* (Dufour, 1833)	TEM
Velia (*Plesiovelia*) *affinis filippii* Tamanini, 1947	TUE
Velia (*Plesiovelia*) *caprai caprai* Tamanini, 1947	EUR
Velia (*Plesiovelia*) *mancinii mancinii* Tamanini, 1947	TUE
Velia (*Plesiovelia*) *saulii* Tamanini, 1947	EUR
Velia (*Plesiovelia*) *serbica* Tamanini, 1951	SEU (BALK)
Family GERRIDAE Leach, 1815	
Aquarius najas (De Geer, 1773)	WPA
Aquarius paludum paludum (Fabricius, 1794)	PAL
Gerris (*Gerris*) *argentatus* Schummel, 1832	SIE
Gerris (*Gerris*) *costae fieberi* Stichel, 1938	TUE
Gerris (*Gerris*) *lacustris* (Linnaeus, 1758)	PAL
Gerris (*Gerris*) *maculatus* Tamanini, 1946	TEM
Gerris (*Gerris*) *odontogaster* (Zetterstedt, 1828)	SIE
Gerris (*Gerris*) *thoracicus* Schummel, 1832	PAL
Gerris (*Gerriselloides*) *asper* (Fieber, 1860)	EUM
Gerris (*Gerriselloides*) *lateralis* Schummel, 1832	SIE
Limnoporus rufoscutellatus (Latreille, 1807)	OLA

9. Conclusions

Taking into consideration the diversity of the topics, the final conclusions are structured according to objectives, results and discussions, and they are shown on chapters:

***Chapter 2*:** Two genera and eight species of the family Veliidae (*Microvelia* – 3 species, *Velia* – 5 species) are known in Romania. *Velia serbica* and *Microvelia buenoi* are new records for Romanian fauna. Occurrence of *V. mancinii mancinii* is also confirmed. *Velia currens* and *V. rivulorum*, repeatedly reported from our country, were not collected in the present study, the analysis of the specimens preserved in other collections showing that they actually belong to other taxa (e.g. *V. caprai caprai*).

The geographical distribution patterns of the Veliidae confirm that the Romanian territory represents a transition zone from Mediterranean to Central Europe.

***Chapter 3*:** The molecular phylogeny of the West-Palaearctic *Velia* agrees well with the morphological taxonomy of the group as defined by Tamanini (1947), by recovering almost all taxa as monophyletic and distinct from the others. The only exception is the paraphyly of *V.* (*P.*) *serbica* with respect to *V.* (*P.*) *mancinii mancinii*. Furthermore, the phylogeny also indicates the existence of apparently unrecognized taxa from Cyprus and Greece. The analyses reveal a sister relationship between the subgenera *Velia* (s. str.) and *Velia* (*Plesiovelia*), the latter comprising three major lineages.

The two subgenera apparently split in the Eocene or early Miocene epoch, and the diverse *Velia* (*Plesiovelia*) originated in southeastern Europe. From there, it subsequently radiated to the west and east, along the geomorphologically dynamic archipelagos of Europe and Asia Minor during the Neogene. Sudden sea-level fluctuations seem also to have played an important role in its diversification. Northwestern Africa served as the second most important diversification centre of *Velia* (*Plesiovelia*). The low intraspecific variability in two widespread European species, *V.* (*P.*) *caprai caprai* and *V.* (*P.*) *saulii*, suggests a rapid postglacial colonization, whereas the remarkably high variability within the lineages of *V.* (*P.*) *serbica* likely indicates *in situ* survival throughout the Pleistocene glaciations in microrefugia.

Chapter 4**:** Three genera and 11 species of the family Gerridae (*Aquarius* – 2 species, *Gerris* – 8 species, *Limnoporus* – 1 species) are known in Romania. Most of the taxa collected through this faunistic survey are common and widely distributed in Europe and have been documented from Romania as well, with the single exception of *Gerris maculatus* Tamanini, 1946. This species was reported as present in Romania only by Andersen (1995b), although without further details or exact localities. On the other hand, *G. gibbifer* Schummel, 1832 which has been recorded repeatedly from this country, was not collected in this study, the analysis of the specimens preserved in museums showing that they actually belong to *G. maculatus*.

The study shows that there are no confirmed records of *G. gibbifer* in Romania, Serbia, Macedonia and Slovenia. Its occurrence in Slovenia is probable (especially in the Alps), since it is present in all the neighboring countries. Therefore, both *G. gibbifer* and *G. maculatus* occur in Croatia, Hungary, Ukraine and probably Slovenia, but mainly in different parts of the countries.

A. p. paludum, *G. lacustris*, *G. asper* and *G. lateralis* commonly were found containing both long-winged and short-winged adults, while for *A. najas* the apterous morph is prevalent. *L. rufoscutellatus*, *G. costae fieberi*, *G. maculatus* and *G. thoracicus* always occurred as the longwinged (macropterous) morph in our samples.

Chapter 5**:** The current knowledge regarding the distribution of Hebridae in Romania is far from satisfactory. However, *Hebrus fulvinervis* is confirmed as valid taxon. The occurrence of *H. montanus* in our country is also confirmed.

Chapter 6**:** The current knowledge regarding the distribution of the two taxa of Hydrometridae in Romania is somehow satisfactory. The short-winged morph seems to be more frequent in Hydrometridae; all the examined specimens of *H. gracilenta* were micropterous, while only 5% of the specimens of *H. stagnorum* were macropterous.

Chapter 7**:** One genus and two species of the family Mesoveliidae are known in Romania. *M. thermalis* is a common and widely distributed species across southeastern Europe. It appears that *M. vittigera* is confined to southern coastal areas with a Mediterranean climate and does not occur north of the Danube River.

The crown age of extant *Mesovelia* is probably Early Miocene, and *M. thermalis* is a sister species to *M. furcata*. The former exhibits a significant genetic divergence between Asian and European populations, but this should be investigated through a more detailed analysis and a broader sampling across its range.

***Chapter 8*:** In total, 27 taxa of semiaquatic bugs are presently known to occur in Romania, a considerably higher number when comparing to the other countries in our region.

From chorological point of view, the Romanian fauna includes: two Holarctic elements, five Palaearctic elements, one West-Palaearctic element, one Asiatic-European element, six Sibero-European elements, three Turano-Europeo-Mediterranean elements, four Turano-European elements, one Europeo-Mediterranean element, two European and two South-European elements; the latter group also includes two Balkan endemic species. Thereby, the diversity of the Romanian semiaquatic bugs, underlines once again the importance of the Carpathians as hotspot for biodiversity.

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