BIOGEOGRAPHIC DIFFERENTIATION OF EPIGEAN FRESHWATER AMPHIPODS (AMPHIPODA: CRUSTACEA) IN BOSNIA AND HERZEGOVINA

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Summary

The inland fauna of epigean freshwater gammarids in Bosnia and Herzegovina is diverse and abundant. This region has a favorable geographical position on the Balkan Peninsula. Excluding Ponto-Caspian species originating in brackish waters and freshwater subterranean taxa, there are 6 formally recognized epigean freshwater species recorded from this country. They belong to 2 genera, from the family Gammaridae, *Gammarus* 4 species and *Echinogammarus* 2 species. We provide new data with high-resolution distribution maps, thus improving the knowledge of the ranges of these taxa. Studied species display substantial altitudinal variability and, fragmented distribution. They occur abundantly, particularly in springs and streams, from lowlands to sub-mountainous and mountainous regions. The mosaic distribution of epigean freshwater amphipod species in B&H shows that this region is particularly suitable for biogeographical analyses of this group. Their large-scale distribution patterns remain obscure due to insufficient data, consequently limiting biogeographical interpretations.

Key words: *Bosnia and Herzegovina, biogeography, distribution, Gammarus, Echinogammarus*

INTRODUCTION

Understanding past influences that have affected current species distributions can be gained via studying distribution patterns (Brown *et al*., 1996). Because of their limited ability to disperse and the fragmented nature of freshwater environments, freshwater amphipod crustaceans are particularly well-suited for biogeographical investigations (Väinölä *et al*., 2008; Hou *et al*., 2011). Amphipods are predominantly aquatic benthic animals that do not possess free-swimming larval stages or resistant propagules, and thus are prone to genetic differentiation and isolation (Barnard & Barnard, 1983). Additionally, a lot of freshwater taxa exhibit allopatric or discontinuous distributions, which are frequently thought to be the result of geologically-related secondary events like island separation, sea level changes, and continental breakup, or that adhere to prehistoric drainage patterns (Hogg *et al*., 2006; Finston *et al*., 2007; Bauzà-Ribot *et al*., 2011, 2012).

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Freshwater amphipod species are rather numerous on the European continent, and diversity rises in the southeast (Väinölä *et al*., 2008). Due to Bosnia and Herzegovina's advantageous geographic location, which places it in one of the significant biodiverse regions in Europe - the Balkan Peninsula - the country has a relatively rich diversity of amphipod species. In terms of biodiversity and endemism, Dinarid Mountains in Bosnia and Herzegovina are likewise regarded as a hotspot location (Ivković & Plant, 2015). Furthermore, the area was a significant glacier refuge throughout the Pleistocene glaciations (Brus, 2009) and underwent active geological events in the Late Tertiary (Argnani, 2018). Because of its diverse terrain, Bosnia and Herzegovina offers a suitable environment for studying amphipod biogeographical trends at a more focused, finer scale. As a result, it can shed additional light on the mechanisms shaping the geographic distributions of freshwater benthic crustaceans.

The biogeographical differentiation of Bosnia and Herzegovina is primarily conditioned by its geographical position, climate, and altitude. The vegetation in the northern, eastern, western, and central parts of Bosnia and Herzegovina has similarities with the vegetation in Central Europe, while the vegetation in Herzegovina and the western and southwestern parts of Bosnia shows similarities with the vegetation in Mediterranean areas. Three biogeographical regions can be differentiated in the area of the Dinarides: the Mediterranean, the Continental, and the Alpine region (Đug & Škrijelj, 2009).

The Ponto-Caspian brackish/freshwater taxa and generally stygobiotic (subterranean) species are excluded from this study since it primarily focuses on epigean freshwater amphipod species, which complete their life cycle in surface freshwaters and occasionally appear in subterranean waters. There are six formally recognized native amphipod taxa that fit the aforementioned requirements and are found in Bosnia and Herzegovina's inland surface freshwaters. Those species belong to the genera *Gammarus* Fabricius, 1775 and *Echinogammarus* Stebbing, 1899 from the Gammaridae family.

Although the epigean amphipod fauna of Bosnia and Herzegovina is abundant and diversified, little is known about the distribution patterns of these species. Studies addressing these issues typically concentrated on national level (Trožić-Borovac, 2014).

We analyze pertinent literature and add new, in-depth data to update distribution maps of Bosnia and Herzegovina-inhabited epigean freshwater amphipod species.

MATERIALS AND METHODS

The material for this research was collected from 90 sample locations between 2007 and 2015 across the entire country of Bosnia and Herzegovina. A benthic hand-net with a mesh size of 250 µm was used to collect the samples, which were then preserved in either 70% or 96% ethanol or 4% formaldehyde solution. We examined every microhabitat that was accessible at each sampling location. A review of the literature was done from the pertinent studies, including the most current ones, and distribution data were collected. The distribution maps were created using data from the literature

and this study. Taxa were identified using the morphological delimitation criteria of the following authors: G. Karaman & Pinkster (1977a; 1977b; 1987), Pinkster (1993) and Stock (1968). For each investigated locality, geographic coordinates were determined using a GPS device during sampling or subsequently. Distribution maps with mapped localities were created in the ArcMap program from the ArcGIS 9.2 software package.

RESULTS AND DISCUSSION

During the research for this study, a total of four species from the genus *Gammarus* and two species from the genus *Echinogammarus* were found in watercourses in Bosnia and Herzegovina (tab. 1). Within the genus *Gammarus*, the presence of the species *Gammarus balcanicus* Schaferna, 1922, *Gammarus bosniacus* Schaferna, 1922, *Gammarus fossarum* Koch, 1836 and *Gammarus roeselii* Gervais, 1835 was recorded. Within the genus *Echinogammarus*, two species *Echinogammarus acarinatus* (S. Karaman, 1931) and *Echinogammarus thoni* (Schaferna, 1922) were recorded.

Table 1. List of epigean freshwater amphipod taxa found in Bosnia and Herzegovina, their type localities, distribution ranges and habitats.
Habitat abbreviations: Sp-springs, St-streams, R-rivers, L-lakes, Rl-reservoir lakes.

Taxa	**Type locality**	**Range**	**Habitats in B&H**
Gammarus balcanicus Schaferna, 1922	Kolašin, Monte Negro	SE Europe, Asia Minor	Sp, St, R
Gammarus bosniacus Schaferna, 1922	Sarajevo, B&H	B&H, the basin of the upper course of the Bosna River	Sp, St, R
Gammarus fossarum Koch, 1836	Regensburg, Germany	Western, Central and SE Europe, Asia Minor	Sp, St, R
Gammarus roeselii Gervais, 1835	Coulanges-sur-Yonne, France	Central and SE Europe, Asia Minor	R
Echinogammarus acarinatus (S. Karaman, 1931)	Mostar, B&H	SE Europe	St, R
Echinogammarus thoni (Schaferna, 1922)	Lake Deransko, BiH	SE Europe	L, Rl

The species *G. bosniacus* is an endemic species of crayfish for Bosnia and Herzegovina, described and up to this research found only on a narrow stretch from Vrelo Bosna to Rimski Most. Until now, it was considered that this species inhabits only the source of the Bosna River, where it was described by Schaferna in 1922, and again described by Karaman G. S. in 1975. For this species, there are data that it was present in the Bosna River as far as the mouth of the Miljacka River (Šenk, 1956). In addition to the Bosna River in its headwaters, this species was also found in the rivers that are on the Bjelašnica mountain or drain water from that terrain (Bjelašnica and Bijela). Given this distribution, it is possible to assume that the center of origin of this species is on the

mentioned mountain, and that it spread to the headwaters of the Bosna River via underground waters.

The most widespread species of gammarids in Bosnia and Herzegovina is *G. balcanicus* recorded at 70 sites. It is spread through all biogeographical regions in Bosnia and Herzegovina. This species in B&H has distribution in the Glina River basin, Una River basin, Bosna River basin, Drina River basin, Cetina River basin and in the Neretva River basin. *G. balcanicus* is a species that has a wide distribution in Southeast Europe and Asia Minor (Barnard & Barnard, 1983; G. Karaman & Pinkster, 1987; Özbek & Ustaoğlu, 2006; Özbek *et al*., 2009). This species is the most widespread gammarid in Romania as well (Petrescu, 1994). According to Copilas-Cocian et al. (2014), this species has the widest habitat altitude range among freshwater amphipods in Romania, which ranged between 16 and 1530 m, and most localities are between 300 and 600 m. It occurs most often in springs and streams, and occasionally in caves and rivers. In certain localities, it was found in coexistence with the species *G. fossarum* and *G. roeselii* (Copilas-Cocianu *et al*., 2014).

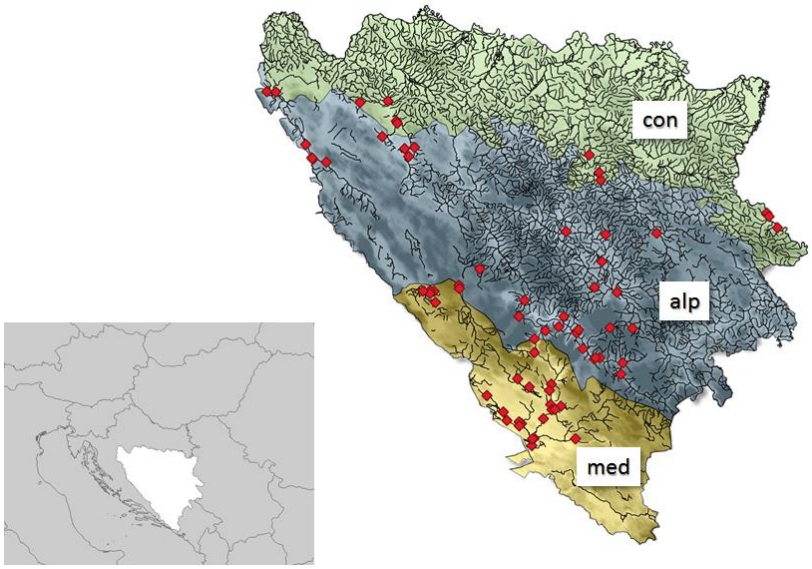

Figure 1. Distribution of *G. balcanicus*. con – continental, alp – alpine, med – mediterranean.

The species *G. fossarum* in Bosnia and Herzegovina was found only in the Black Sea basin, in the Una, Vrbas, Bosna, and Drina River basins, and in the immediate Sava River basin. *G. fossarum* has a wide range that includes western, central, and southeastern Europe and reaches northern Anatolia (G. Karaman & Pinkster, 1977a; Barnard & Barnard, 1983; Özbek & Ustaoğlu, 2006). In Romania, it occurs in the western part of the Carpathians in two isolated regions, one in the northwest and the other in the southwest. The altitude range of localities where this species has been recorded ranges from 47 to 860 m, and most often between 300 and

550 m. Populations from southwestern Romania also occur in lowland rivers, while northwestern populations are restricted to springs and streams in sub-mountainous areas. In some localities, *G. fossarum* coexists with *G. balcanicus* and *G. roeselii* (Copilas-Cocianu *et al*., 2014).

During this research, the species *G. roeselii* was found at four localities in the Una River basin. Three localities were recorded on the Una River itself (Bosanska Krupa and two localities near Bosanska Otoka), and one locality on the Sana River (Sanski Most). In Bosnia and Herzegovina, this species is found predominantly in the watercourses of the Continental region. The species *G. roeselii* is distributed in Western, Central, and South-Eastern Europe as well as South-Eastern Europe, and in the western part of Turkey (G. Karaman and Pinkster, 1977a; Barnard & Barnard, 1983; Jazdzewski & Roux, 1988; Özbek & Ustaoğlu, 2006). In Romania, it is present in several regions (Motaş *et al*., 1962; Pârvulescu 2009). It is a typical lowland taxon that occurs mainly at altitudes below 200 m. This species is the most ecologically plastic gammarid, which can be found in springs, streams, rivers, and occasionally in lakes and swamps (Motaş *et al*., 1962). It can co-occur with *G. balcanicus* and *G. fossarum*.

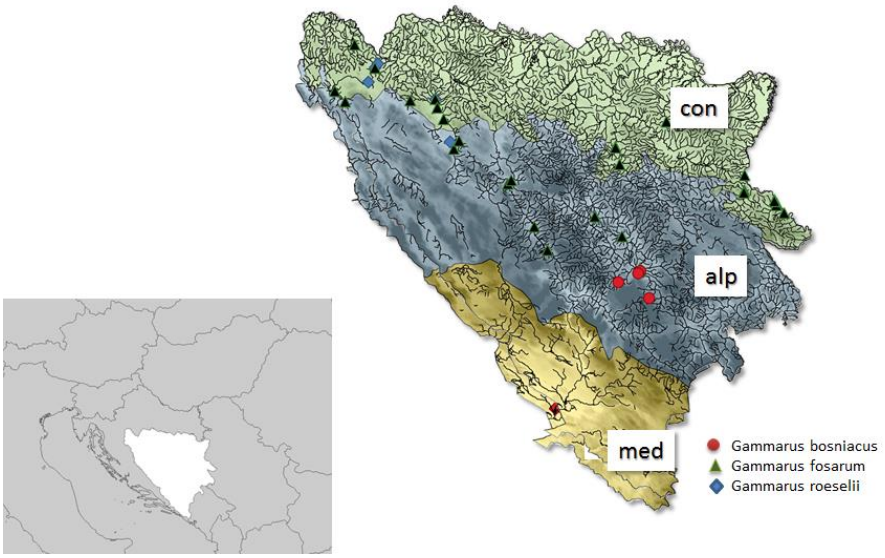

Figure 2. Distribution of *G. bosniacus*, *G. fossarum* and *G. roeselii*. con – continental, alp – alpine, med – mediterranean.

Analyzing the distribution of individual species from the genus *Gammarus*, it was determined that there are overlapping zones of certain habitats for some species. The most noticeable mixing between populations of different species was recorded between *G. balcanicus* and *G. fossarum*. For these two species, habitats overlap in the Una River basin, in the locality downstream from Bihać, and in the rivers Sana and Sanica. Common habitats for these two species were also observed in the Bosna River basin, in

its upper and middle reaches. Overlapping habitats were also noted in the area of the Drina River basin.

The genus *Echinogammarus* is the second most numerous genus within the family Gammaridae, in which 53 species have been described. Most of them inhabit freshwater and brackish waters, and only a small number are exclusively marine species. More than most of the species of this genus known so far (35 or 66%) are distributed in Southern Europe with the center of endemism in the Iberian Peninsula, where 20 endemic species have been described. The distribution and ecology of endemic species are poorly researched, and many species are known only from a few sites or from the type locality (Pinkster, 1993).

The species *E. acarinatus* was found in both major sea basins in Bosnia and Herzegovina and the Continental and Mediterranean regions. In the Black Sea basin, it was found in the Vrbas River basin in the Duboka river near Bugojno at an altitude of 715 m. In the Bosna River basin, this species was found in Plava Voda, a tributary of Lašva near Travnik, near the watershed between the Vrbas and Bosna basins. In the Adriatic Sea basin, this species is present in the Neretva River basin in two localities, the Radobolja River and the mouth of the Buna in the Neretva.

The species *E. acarinatus* was first described as a form of *G. pungens f. acarinata* (Schaferna, 1922). After ten years, Karaman, S. (1931) raised this form to species level and called it *Ostigammarus acarinatus*, while Karaman, G. (1970) included this species in the genus *Echinogammarus*. The type locality of this species is the Buna River near the mouth of the Neretva. The area of this species includes the source of the river Krka, Butižnica (a tributary of the Krka), Jadro, Lake Vrana near Biograd, salt springs near Trogir, the river Jadrtovac near Šibenik, Neretva near Metković, a small stream near Dubrovnik, a stream near Lake Vrana and the stream Stobreč in Croatia (Karaman, 1931; Karaman, 1970; Pinkster, 1993), and in the Buna and Radobolja rivers near Mostar (Karaman, 1970). The species *E.s bosnensis* S. Karaman, 1934 is synonymized with the species *E. acarinatus*. The type locality for the mentioned synonymized species is Šumeće Vrelo near Travnik. As this species is widespread in various watercourses in the areas of Croatia and Bosnia and Herzegovina, where certain abiotic factors vary greatly, it can be said that it is a eurivalent species that occur in waters where temperatures range from 10-25ºC, with electrical conductivity of 365-1007 μScm^{-1}, and inhabits saline springs and freshwater ecosystems (Karaman, 1931; Žganec, 2009). This species reaches a significantly higher number of individuals on plant substrate (Žganec, 2009). Further research is necessary to determine the range of tolerance of this species for certain abiotic factors in the aquatic environment.

The species *E. thoni* in Bosnia and Herzegovina was found only in the Neretva River basin in four localities. Earlier findings of this species in Lake Deran, Bregava, and Buna rivers were confirmed. During the research, this gammarid was also recorded in the Salakovac reservoir on the Neretva River and represents the northernmost point of distribution of this species. After the description of this species, it was found in five more localities in the Neretva delta in Croatia and Bosnia and Herzegovina, as well as in the upper course of the Jadro river near Split (Karaman, 1929, 1934; Karaman,

1969). This gammarid was also recorded in the Buna and Bregava rivers, near the confluence with the Neretva. More recently, this species has also been found in the Canj stream near Bar, and in the Orahovica river near Skadar lake in Montenegro (Grabowski & Pešić, 2005; Žganec *et al*., 2010). In more recent research, *E. thoni* was found in several new localities in Croatia and watercourses in Albania (Žganec *et al*., 2010). So far, only the species *G. balcanicus* has been found in the middle part of the Neretva River from the Amphipoda order (Škrijelj, 2002). The physical and chemical conditions recorded in this hydro accumulation show that this population of the mentioned species is well adapted to life in depths of up to 20 m, and with a relatively constant and low temperature, which is up to 10°C, which confirms its good adaptation to different living conditions in different types aquatic habitats. This species is well adapted to the conditions of high salt concentration in the water, and tolerates concentrations up to 6.9‰, and it reaches its highest abundance in summer in warm watercourses, where the temperature during this season is between 20 and 25°C. The distribution center is located in the southern part of the Neretva River delta, where it inhabits most of the lower reaches, and probably also most of its tributaries in that part (Žganec *et al.*, 2010).

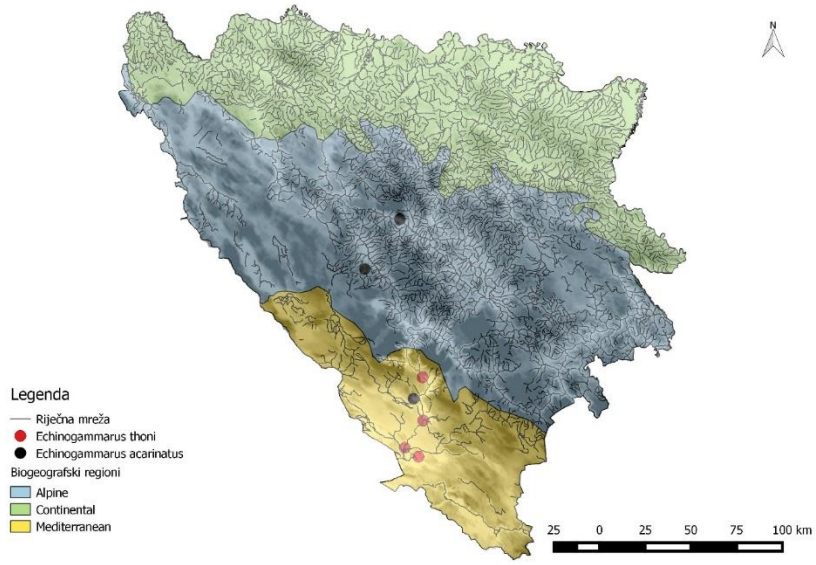

Figure 3. Distribution of *E. acarinatus* and *E. thoni*.

Biogeographical distribution of freshwater epigean amphipod species in B&H expresses different patterns. *G. bosniacus* was found only in the Alpine region and *E. thoni* only in the Mediterranean. *G. balcanicus* inhabits watercourses in all three biogeographical regions in B&H, but 60% of recorded sites are in the Alpine region, and 27% are in the Mediterranean. *G. roeselii* and G. *fossarum* were recorded in the Alpine and Continental regions. According to our data, these two species are mostly distributed in the

Continental region (fig. 4). *E. acarinatus* has equal distribution in the Mediterranean and alpine regions.

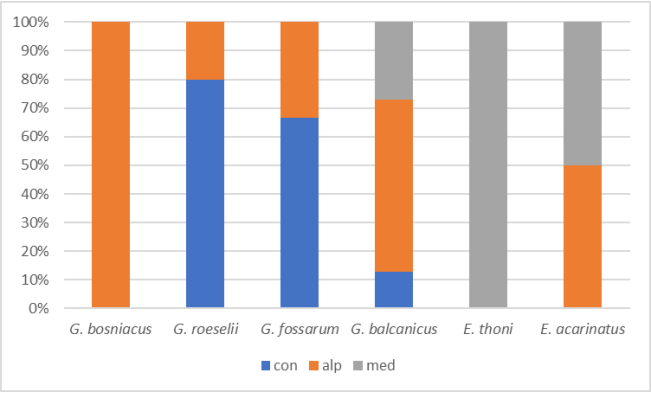

Figure 4. Biogeographical range of epigean freshwater gammarids in B&H. con – continental, alp – alpine, med – Mediterranean.

The habitat analysis of species from the genus *Gammarus* covers locations from 0 to 1200 meters above sea level. The species *G. balcanicus*, which was found in sites ranging from sea level to 1200 meters, shows the widest amplitude of variation in altitude (fig. 5).

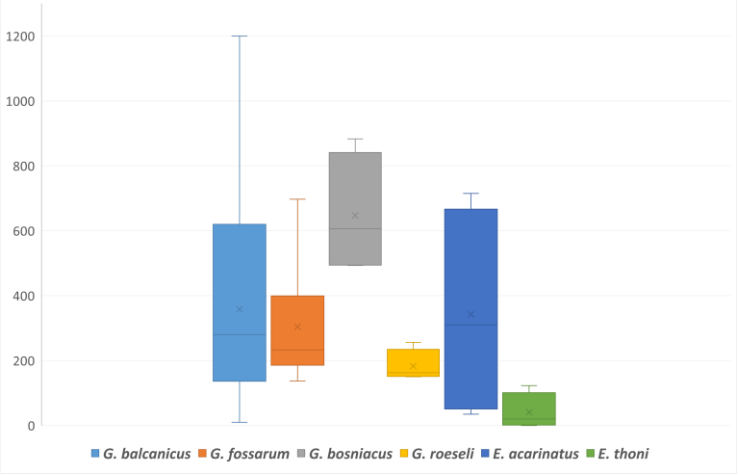

Figure 5. Boxplots representing the altitudinal ranges of studied epigean amphipod taxa in Bosnia and Herzegovina.

However, the largest number of localities where this species has been recorded ranges from 200 to 600 meters above sea level. The species *G. fossarum* was found in sites from 150 to 750 meters above sea level with most sites in the range of 400 to 600 meters. The altitude range of the habitat for the *G. bosniacus* ranged from 450 to 950 meters above sea level. The species *G. roeselii*, which was observed in sites whose altitude

ranged from 180 to 250 meters above sea level, shows the smallest range of variation in habitat height (Fig. 5). Habitats of two *Echinommarus* species ranges from 0 to 750 meters above sea level. The wider altitudinal range has *E. acarinatus*, and *E. thoni* was found only in habitats between 0 and 123 meters above sea level (fig. 5).

CONCLUSION

To fully appreciate Bosnia and Herzegovina's unique fauna of epigean freshwater amphipods, additional taxonomic research is required. The patchy and altitudinal variability of the epigean freshwater amphipod distributions in B&H is distinctive features. These make up a suitable model system for studying biogeography and phylogeography at a fine scale, with implications for further research in ecology, adaptation, and speciation of freshwater amphipods. This is due to their limited capacity for dispersal as well as the heterogeneous topography and geology of this area.

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BIOGEOGRAFIJA NADZEMNIH SLATKOVODNIH RAKUŠACA (AMPHIPODA: CRUSTACEA) U BOSNI I HERCEGOVINI

Rezime

Kopnena fauna nadzemnih slatkovodnih rakušaca u Bosni i Hercegovini je raznolika i bogata. Dinaridi imaju povoljan geografski položaj na Balkanskom poluotoku i predstavljaju jedan od centara biodiverziteta. Isključujući ponto-kaspijske vrste koje potječu iz bočatih voda i slatkovodne podzemne vrste, postoji 6 službeno priznatih nadzemnih slatkovodnih vrsta zabilježenih u Bosni i Hercegovini. Sve vrste spadaju u 2 roda, iz porodice Gammaridae, *Gammarus* 4 vrste i *Echinogammarus* 2 vrste. U ovom radu su izneseni novi podaci s kartama distribucije visoke rezolucije, čime se poboljšava poznavanje rasprostranjenosti ovih vrsta. Proučavane vrste pokazuju znatnu visinsku varijabilnost i fragmentirane obrasce distribucije. Javljaju se u većim brojevima, osobito u izvorima i potocima, od nizinskih do pretplaninskih i planinskih krajeva. Mozaička distribucija ovih vrsta u BiH pokazuje da je ovo područje posebno pogodno za biogeografske analize ove skupine. Njihovi obrasci distribucije ostaju nejasni zbog nedovoljno podataka, što posljedično ograničava biogeografska tumačenja.

Key words: *Bosna i Hercegovina, biogeografija, distribucija, Gammarus, Echinogammarus*