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Evolution, adaptation and speciation in *Anthroherpon* Reitter, a genus of highly evolved subterranean Coleoptera

Jury

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Evolution, adaptation and speciation in *Anthroherpon* Reitter, a genus of highly evolved subterranean Coleoptera

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ABSTRACT (IN FRENCH AND SERBIAN)

Cette thèse traite de l'évolution, de l'adaptation et de la spéciation en milieu souterrain, en utilisant le genre *Anthroherpon* comme modèle. Ce genre appartient à la tribu des Leptodirini (Leiodidae, Cholevinae), un groupe qui a connu une remarquable diversification dans le domaine souterrain. Toutes ses espèces ont développé des modifications troglomorphiques spectaculaires: anophthamie, aptérisme, élongation extrême des appendices, de la tête et du pronotum, et physogastrie. Pour comprendre l'histoire évolutive du groupe, ces adaptations troglomorphiques ont été replacées dans un cadre phylogénétique. La thèse est une analyse de la radiation évolutive des *Anthroherpon*, dans le cadre d'une phylogénie moléculaire datée, qui a permis de mieux comprendre les modalités de diversification du genre, de reconstruire son aire de distribution ancestrale, d'explorer la diversité des évolutions troglomorphiques en son sein et de proposer une nouvelle structure taxonomique du groupe.

* * * * * *

Doktorska disertacija predstavlja studiju evolucije, adaptacije i specijacije u podzenmim staništima troglobiontnih tvrdokrilaca roda *Anthroherpon*. Pomenuti rod pripada tribusu Leptodirini (Leiodidae, Cholevinae), grupi koja je prošla intenzivnu diverzifikaciju u uslovima podzemnih staništa. Sve vrste pomenutog roda poseduju tipične troglomorfne osobine, kao što su: anoftalmija, apterizam, ekstremno izduženi telesni nastavci, glava i pronotum, i fizogastrija. Radi razumevanja evolucione istorije grupe, troglomorfne adaptacije su studirane u filogenetskom kontekstu. U analizi evolutivne radijacije roda *Anthroherpon* korištećena je datirana molekularna filogenija kao okvir za razumevanje diverzifikacije roda, evolucije troglomorfnih karaktera i rekonstrukciju predačkog areala. U svetlu novih nalaza predložena je nova taksonomska organizacija grupe.

INTRODUCTION TO EVOLUTION IN SUBTERRANEAN ENVIRONMENT

0.1. TERRESTRIAL SUBTERRANEAN COMPARTMENT

The subterranean domain includes air- and water-filled underground habitats below the surface of the Earth, from small spaces such as tiny interstitial openings between sand grains to large spaces such as caves (Botosaneanu, 1986; Culver & Pipan, 2013). Below the ground surface two strata can be distinguished: the endogean (or euedaphic) environment that corresponds to deep soil layers and the hypogean (or subterranean in the strict sense of the word) environment below this stratum (Giachino & Vailati, 2005a). These ecological environments are closely connected and form an inseparable continuum (Racovitza, 1907; Giachino & Vailati, 2005a). Culver and Pipan (2009) categorized subterranean habitats into three main types: cave habitats, interstitial habitats, and superficial subterranean habitats (seeps, talus slopes, and small cavities in the uppermost areas of karst rocks).

Biospeleology or speleobiology (derived from Greek words "*bios*"=life and "*spelaion*"= cave) is a branch of biology dedicated to the study of organisms living in subterranean habitats. It began to develop in the first half of the 19th century, after the first scientific description of a cave-dwelling animal, the beetle *Leptodirus hohenwarti*, described by Schmidt (1832) from the Postojna cave system in Slovenia.

Subterranean habitats are present in continental regions of almost all latitudes (except the far north and far south of the planet), in all meridians and altitudes — from underwater caves to caves up to 3000 m above sea level in the mountains of the temperate zone, and up to 5000 m above sea level in the mountains of the tropical and subtropical zone (Karaman et al., 2015). According to the size, representation, and diversity of this biome, we can conclude that it is one of the most important on our planet. This is best illustrated by the fact that over 96.1% of the total fresh water on Earth, which is not in frozen state, is located in underground cavities (Shiklomanov, 1993).

Our perception of the diversity and the complexity of subterranean habitats is changing with the improvement of technical methods which enable the study of underground space inaccessible to humans. Development of biospeleology revealed that subterranean habitats are an immense and complex biome, a mosaic of diverse terrestrial and aquatic habitats, inhabited by specially adapted organisms. Due to the difficult access of these habitats, explorations require a lot of effort and our abilities to explore them are often limited to explorations of (i) caves, or (ii) "mesovoid shallow substratum" (the

"MSS", "milieu souterrain superficiel" (Juberthie et al. 1980a), accessible from the surface, while the entire networks of cracks are not directly accessible to humans. Since we are able to investigate only a small portion of the subterranean habitats, this environment is still largely unexplored. However, many observations and hypotheses drawn from cave fauna can be extrapolated to non-interstitial terrestrial subterranean habitats and fauna. In the remainder of this thesis, wherever the word ''cave'' is used, for simplicity, it should be understood as a synonym for ''subterranean habitat''. Consequently, "cave organisms" should be considered as a synonym of subterranean organisms.

0.2. KARST AND CAVE FORMATION

Karst is the term used to describe a special type of landscape containing caves and extensive underground water systems that is developed on especially soluble rocks such as limestone, marble, and gypsum (Ford & Williams, 2007). The word „karst" can be traced back to pre-Indoeuropean origins (Cvijić, 1893; Gams 1973a, 1991a, 2003; Kranjc 2001a). It derives from „karren", a proto-European term meaning „rock", found in many languages of Europe and the Middle East. This type of landscape was named after Carso (Karst), the geographic area between Italy and Slovenia. It was the first place where such landscapes have been scientifically studied, and it is the area where some of the most prominent karst phenomena are situated. Karst occupies ~12.5% of Earth's surface (Jones & White, 2012) and its thickness can reach outstanding dimensions, sometimes up to several kilometres. Karstic terrain is commonly characterized by sinking streams, caves, enclosed depressions, fluted rock outcrops, and large springs. It is formed principally by the solution of the rock, most commonly limestone. Limestone is a sedimentary rock formed on the seabed from the accumulation of calcium carbonate ($CaCO_3$) skeletons of marine animals and algal and fecal debris. Due to geotectonic movements, sedimentary rocks of marine origin are uplifted above the sea level where they are exposed to atmospheric conditions and are subject to erosive and corrosive processes. As a result, karst is formed, a landscape with a specific geomorphology, morphology, and hydrology.

0.2.1. Speleogenesis

Weathering of limestone depends on the chemical properties of its constituent minerals, almost entirely calcium carbonate, a compound with very low solubility in water. During the process of the formation and development of caves (speleogenesis), limestone is being dissolved by carbon dioxide (CO_2). Carbon dioxide is present in water due to microbial respiration in the soil. In the presence of carbon dioxide the water becomes more acidic, and dissolves the calcium carbonate, forming calcium bicarbonate, according to the following reaction:

$CaCO_3 + H_2O + CO_2 \rightarrow Ca^{++} + 2HCO_3^-$

This reaction is reversible: a disruption of equilibrium in the solution (e.g., by a change of pH, temperature or pressure) leads to a reverse reaction, i.e., CO_2 release and deposition of $CaCO_3$. As water dissolves the surface of bedrock near its cracks, or bedding planes, the bedrock continues to break down and its cracks tend to get bigger. Over geological time, cracks expand to become caves and cave systems. Speleogenesis is part of a wider geomorphological process (karst formation) of dissolving carbonate bedrock by freshwater and precipitation.

0.2.2. What are caves?

There have been lengthy debates regarding the definition of a cave, i.e. what effectively constitutes a cave and what kind of empty space in rock, carbonate or not, enters this category. The definition adopted by the International Speleological Union is that a cave is a natural underground opening in rock that is large enough for human entry (Ford & Williams, 2007). This implies a minimum size of 5 m in diameter. However, even if we accept this anthropocentric definition for practical reasons, caves should never be regarded as closed spaces, but as complex natural systems of interconnected cracks, voids, fissures of various size (Giachino & Vailati, 2010). A strictly scientific definition would be that a cave is a natural cavity in a rock which acts or has acted as a conduit for water flow between input points, such as stream sinks, and output points, such as springs or seeps (White, 1984).

Caves are formed in many rock types. Most caves are formed in limestone and related carbonate rocks, but significant number of caves are also found in lava, sandstones, gypsum, basalt, granites and ice. The great majority of natural caves are solution caves. Lava caves have been formed by volcanic processes, while other type of caves have been created principally by the dissolution of bedrock by water circulating through initial openings such as fissures and pores. The largest and most abundant solution caves are located in limestone.

There are many different ways to categorise caves but there is no single classification or a theory of genesis that has been able to encompass them all. In terms of structure, caves are sometimes classified according to their orientation relative to the gravity field into horizontal, inclined, and vertical caves, although large systems commonly have elements of various orientations. Caves can be in the form of a single cavity or an extensive underground network with multiple connections to the surface. We can also find variation in terms of size, from small caves to extraordinarily large ones like Mammoth Cave system in Kentucky, with more than 560 km of explored and mapped passages (Watson, 2005). Regarding how their spaces are filled, caves can be air-filled, water-filled, and mixed. Among aquatic caves, we find freshwater, marine, and anchialine (containing a mixture of sea water and freshwater). Caves are formed in rocks of all ages, from Cambrian, more than 500 million years old, to Holocene limestone, less than 10,000 years old (Culver & Pipan, 2013).

0.2.3. Speleology

The scientific discipline that devotes itself to the study of caves and other subterranean features is speleology. The first modern speleologist was Adolf Schmidl (1802-1863) from Czech Republic, hence considered as a „father of modern speleology“, who linked the various branches of cave study (Shaw, 1992). The earliest word for this purpose in any language was the German *Höhlenkunde*, meaning cave study, which was introduced into the scientific literature by Schmidl. His explorations took place in the mid-19th century, in Slovenia, Austria, and Hungary (Shaw, 1978). Somewhat later came Edouard A. Martel (1859-1938) whose explorations predominantly took place in France, but also in many other places in Europe as well as Russia and U.S. He was actively working on cave explorations for 26 years and his scientific contributions comprise 20 books and 780 papers. Martel himself founded the Société de Spéléologie in 1895 which was the first organisation of its kind in Europe with a significant international function. However, first cave explorations begun in China, and much earlier than in Europe– in the first half of 16th century Xu Xiake (1587-1614) explored or located more than 270 caves in Guangxi (http://www.chinaculture.org). He was a geographer of the late Ming Dynasty and a pioneer in systematic karst studies. After his work, nothing significant was published on the karst of China for more than 30 years. In the United States, organized cave exploration only began in 1941, when several caving groups formed the National Speleological Society (White & Culver, 2005).

Although organized cave explorations started more than a century ago, only a small portion of caves on Earth have been explored and documented by cavers. More than 100,000 caves are known from Europe, and nearly 50,000 are known from the US (Culver & Pipan, 2013) but this number is far from definite since new caves are constantly being discovered in many parts of the world (Testa, 2013; Grandcolas, 2014).

0.2.4. Life in caves

Although subterranean habitats are among the least explored on Earth due to difficult access, a large array of organisms is known from this environment, from unicellular organisms to vertebrates. Thousands of species from different animal groups have been described from caves and many more are yet to be discovered. The diversity of cave fauna is not proportional to the cave size. The world's longest cave, Mammoth Cave system, is inhabited by 41 cave animal species (Hobbs, 2005) while in the cave Vjeternica in Bosnia and Herzegovina, with a total length of only around 7 km, 101 cave animal species were recorded (Ozimec & Lučić, 2010), making it the cave with the richest cave fauna in the world (Ozimec & Lučić, 2010). On second place, with 84 obligate cave animal species, is the Postojna cave system in Slovenia, one of the best studied caves in the world (Hobbs, 2005).

Even though we tend to call animals inhabiting the subterranean environment "cave animals" or "cave-dwellers", it is important to emphasise that many are not confined to caves. Their habitat is far vaster, extending into the network of microfissures and crevices inaccessible to humans. This idea is more than a century old; in 1907, Racovitza wrote in the series “Biospeologica”: “… *j’incline à penser que beaucoup de cavernicoles ont leur habitat normal dans les fentes et non dans les grottes”* [... I tend to think that the natural habitat of many cave insects is in the fissures and not in caves]. However, this notice was overlooked in the following years by many “terrestrial” biospeleologists. In 1988, Vailati went back to the subject emphasizing that the fissure network is the most favourable and therefore the main habitat of subterranean fauna.

0.3. EVOLUTION IN SUBTERRANEAN ENVIRONMENTS

0.3.1. Colonisation

Since the very beginning of the development of biospeleology, questions regarding the colonisation of subterranean habitats and the origin of obligate cave species (troglobites: see the next chapter) have been raised. Various evolutionary scenarios have been proposed to explain events that have led to the colonisation of subterranean environment. Why did epigean species leave their energy-rich surface habitats and populated subterranean, energy-poor habitats? Giving an answer to this question is additionally complicated by the fact that many troglobites in the temperate zone do not have close relatives in the local epigean fauna. Generally, all hypotheses regarding the origin of the subterranean fauna can be divided into two categories: active and passive colonisation (Danielopol & Rouch, 2005). Active colonisation is understood not only as colonisation by means of the locomotory apparatuses, but more as a „voluntary“ colonisation (Stoch, 2004). On the contrary, passive colonisation was initiated either by the force of or by the change of the environmental dynamics (Danielopol & Rouch, 2005).

At the beginning of the 20th century, Neo-Lamarckians, like E.G. Racovitza and R. Jeannel in Europe and A.M. Banta in North America, considered the active colonisation of subterranean habitats the most probable scenario (Danielopol & Rouch, 2005). They assumed that active colonisation was initiated by drastic climate changes and changes in the composition of epigean communities, which increased competition and predation. French zoologist A. Vandel (1964) had a similar view and coined the idea that "old" species were not competitive enough and therefore not able to survive and follow the dynamics of the epigean environment, so they retired to subterranean habitats. These opinions were bolstered by many cases of so-called "living fossils" among troglobites (Jeannel, 1943). Jeannel considered that colonisation of the subterranean environment had been initiated by pre-adapted organisms — photophobic, humicolous or muscicolous organisms living in soil, fissures or under the stones (Vandel, 1964). The idea of preadaptation was later emphasized by many biologists (Holsinger, 2000; Langecker, 2000; Christiansen, 2005). Nevertheless, recent studies show that pre-adaptations are neither necessary, nor sufficient in subterranean environment (Romero & Paulson, 2001; Langecker & Longley, 1993).

At the end of the 19th century, E. R. Lankester proposed the passive colonisation scenario. He assumed that evolution in subterranean environment could start with organisms that accidentally colonised cave habitats (Lankester, 1893). However, these 19th century models were mostly speculations and only in the second half of the 20th century, the plausibility of these scenarios was scientifically tested and documented. Danielopol & Rouch (2005) summarised all evolutionary (not mutually exclusive) scenarios of colonisation of subterranean environment in eleven models (7 active and 4 passive). I will here briefly describe all of them.

A. Models based on active dispersal include:

1. The climatic-relict model

   This scenario surmises that the subterranean environment acts as a refugium for epigean fauna during unfavourable climatic conditions (e.g., Pleistocene glaciation and interglaciation, aridification) (Barr, 1968; Banarescu, 1975; Sbordoni, 1982; Barr & Holsinger, 1985; Peck & Finston, 1993). In periods of climatic change, many epigean species became extinct, but some of them actively colonised subterranean refugia where they managed to survive and adapt to the new environment. This hypothesis is supported by the fact that many lineages with exclusively subterranean species do not have close epigean relatives, and are considered on this basis as relicts. Many troglobites and stygobites from Europe, North America, Africa, and Australia fit this model (Danielopol & Rouch, 2005). One example is the sub-tribe Anthroherponina distributed in the Dinaric Mountains in Europe whose close epigean, geographically proximate ancestors are not known and which are considered relicts of formerly widespread epigean ancestors (Jeannel, 1943). This scenario has also been suggested for the more than 250 species of cave-adapted trechine carabid beetles in the United States (Barr & Holsinger, 1985).

2. The adaptive-shift model

   This model assumes that the colonisation of the subterranean medium is driven by the opportunity to exploit new resources (Howarth, 1980, 1987). Preadapted surface organisms (e.g. living under stones, inhabiting leaf litter, etc.) from endogean habitats are able to further colonise subsurface medium and progressively adapt to the new environment. This model presumes parapatric speciation. It is well documented by Hoch & Howarth (1993) in planthoppers (Cixiidae) in lava tubes formed in the Hawaiian Islands. These caves formed by volcanism are rich with plant roots which provide an enormous food resource for planthoppers. Howarth (1987) considered the exploitation of this food resource to be a driving force for the evolution of cave-adapted planthoppers. Another example is the study of speciation of troglobitic land snail *Georissa filiasaulae* from Malaysian Borneo (Schilthuizen at al., 2005). Molecular phylogenetic analysis shows the ongoing gene flow between cave species *G. filiasaulae* and its epigean ancestor *G. saulae* via population of intermediate morphology in the twilight zone of the cave.

Even though this model can be clearly distinguished from the climatic-relict model, in practice, it is sometimes very hard to determine which colonisation model is the most probable. It is possible that subsequently, after parapatric speciation of the subterranean population, the ancestral epigean population becomes extinct due to climate change or other distribution alterations, so that the resulting biogeographic pattern gives the (false) impression of allopatric speciation (Leys et al., 2003).

3. The Active Colonisation Model

   Assumes that colonisation events are not initiated by environmental stress. According to this scenario, proposed by Rouch and Danielopol (1987), the subterranean environment was colonised by preadapted and/or generalist organisms. This model is supported by the presence of numerous troglophiles in caves (considered as potential ancestors of future troglobites), which have a wide distribution outside subterranean habitats. One example are several crickets of the genera *Troglophilus* and *Dolicopoda* distributed in part of the Mediterranean. Even though they are widely distributed in surface habitats, they also often form large and stable populations inside caves. Therefore, they are often wrongly taken as troglobites (Karaman et al., 2011).

4. Colonisation through Marine Shallow-Water Ecotones

   Ancestors of today's troglobites inhabiting subterranean aquatic caves could penetrate to these habitats either directly through the entrance of the caves or through submarine karstic springs or porous clastic sediments along coastal areas (Danielopol & Rouch, 2005). However, colonisation from marine habitat would imply double adaptation- first to develop freshwater tolerance and then to adapt to the subterranean environment.

5. Transit of Marine Fauna through Epigean Limnic Systems

   Euryhaline marine benthic animals could colonise caves and/or interstitial habitats by adapting to surface inland waters and continuing the invasion of subterranean waters (Danielopol & Rouch, 2005). Examples are many Crustacea that have colonised Dinaric karst as well as some unique elements of Dinaric macro-stygofauna like *Congeria kusceri* (Bivalvia) and *Marifugia cavatica* (Polychaeta).

6. Colonisation from the Deep Sea

   Biological traits of many deep-sea organisms largely resemble the traits of subterranean fauna due to convergent evolution. It is hypothesized that deep-sea organisms may penetrate anchialine caves through the crevices of volcanic or karstic rocks up to the level of anchialine shallow caves (Danielopol & Rouch, 2005). This idea is supported by the occurrence of some deep-sea organisms, like sponges, Crustacea and fish, in anchialine caves.

7. Escape from Epigean Predators and/or Strong Competitive Pressure

   This model suggests that small invertebrates colonised subterranean habitats to avoid strong predation and/or competition in the epigean environment. It is assumed that many Crustacea, for example, invaded caves to escape predation by other animals, particularly insects. At the beginning of the 20th century, E. G. Racovitza gave the example of a troglobitic isopod from North Africa, *Spelaeoniscus debrugei,* suggesting that the colonisation of subterranean habitats was initiated by its incomplete capacity to protect itself from predators. Namely, this isopod rolls up into a ball when predators attack, but its antennae are left out during the rolling up of the body. Racovitza assumed that these crustaceans were therefore an easy prey for epigean invertebrates (Racovitza, 1907). This scenario is further supported by observations in the field and/or laboratory of the transition of epigean animals in subsurface habitats because of predator pressure. For example, observations of *Gammarus roeseli* exposed to fish predation showed that a fraction of the surface-dwelling animals can migrate into the sediment due to predator pressure (Danielopol & Rouch, 2005).

B. Models based on passive dispersal:

1. Passage of Epigean Animals during Massive Surface Water Infiltration into Subterranean Systems

   Epigean animals could be transported by surface streams which flow into karstic systems through sinkholes or infiltrate through macro- and micro-channels. Through the complex underground drainage system, this fauna can disperse further to the large karstic systems. Another way of passive dispersal of fauna into the subsurface can happen during floods in alluvial plains along river channels. During and after the floods, surface waters (including the fauna) infiltrate into the subsurface. As an example, Danielopol & Rouch (2005) mention periodical invasion of Cladocera (Crustacea) from the backwaters of the Danube wetlands (in Austria) into the aquifer existing below the floodplain. Also, some authors assume that this process was the origin of the highly diverse stygobitic crayfish fauna in Florida (Danielopol & Rouch, 2005).

   Moreover, studies done on the Baget, Moulis, and Dorvan karstic drainage basins have documented that a high number of animals penetrate into the subsurface during the rainy period (in Danielopol & Rouch, 2005).

2. The Regression Model

   This model assumes passive transition of marine benthic organisms to inland subterranean waters because of eustatic regressive sea level movements. The subsurface marine littoral is inhabited by a community of so-called thalassostygobionts, which are poorly vagile. J. Stock (1980), who developed this model, argued that during marine regressive phases, these animals, instead of following the regression of the marine water, remained on the coast and progressively adapted to inland non-marine subterranean water, first to brackish and later to freshwater. The idea for this scenario came from the distribution of some stygobites of marine origin: the primary habitat of amphipods of the family Ingolfiellidae is the marine interstitial and deep-sea benthos, but they are also known from subterranean freshwater of ancient marine paleo-coasts. Similar model was developed by Boutin and Coineau (1990).

3. Rafting and Resettlement of the Fauna in New Subterranean Habitats
   Animals inhabiting the marine interstitial (e.g., harpacticoids and ostracods) can be transported passively on various floating objects.

4. Erosion of Sediment and the Drift of Subsurface Fauna with the Surface Water Flow
   During storms and floods, sediment containing interstitial marine or freshwater fauna gets resuspended, so these organisms could be passively transported through the surface water column. After being transferred to other places they are able to recolonize shallow subsurface habitats.

This multitude of models, many of which are not mutually exclusive, may provide a somewhat confusing picture. However, generally, the active colonisation models emphasize the ecological benefits of cave habitats (e.g. the absence of predator pressure and the relative stability of the physical conditions), while passive colonisation models emphasize the ecological problems that incipient cave dwelling animal must solve (Kane & Richardson, 2005). Each of the models mentioned above could be supported by evidence from certain animal groups or geographic areas. However, it appears that no unique explanation can encompass all evolutionary transitions of epigean to obligate subterranean species.

0.3.2. Natural selection

Charles Darwin (1858) and Alfred Russel Wallace (1858) suggested that natural selection, leading to the change in heritable traits of populations over time, was the primary mechanism of evolution. The role of natural selection in moulding the biology of cave organisms, namely its effect on adaptation, has long been the central problem in biospeleology. The cave environment, ecologically less complex than many other types of habitats, draws the attention of evolutionary biologists because selection pressures can be more clearly recognized. Thus, caves and cave animals are ideal models for the study of evolution, particularly for the study of adaptation (Culver, 1982; Poulson & White, 1969; Culver et al., 1995; Kane & Culver, 1992).

What are the sources of selection pressures in subterranean habitats? Caves are characterized by the relative stability and consistency of the physical conditions (Christiansen, 1961; Culver, 1982). Temperature in caves is more or less constant throughout the year and usually corresponds to the average temperature in the area where the cave is located (Palmer, 2007) while relative humidity rarely falls below 80% (Culver, 1982). All subterranean habitats share several key features: permanent absence of light, absence of primary productivity (except in some rare cases where chemoautotrophy takes place), relatively constant temperature, relative humidity often near saturation, and the absence of seasonal variations of climate (Christiansen, 1961; White & Culver, 2005; Culver & Pipan, 2009). These abiotic factors are the main source of the selection pressures to which cave organisms must adapt.

Except some rare cases of bioluminescence (insect larvae in New Zealand and Australia), caves are habitats without natural source of light (Meyer-Rochow, 1990). Relying on visual senses for finding food, avoiding predators, and recognizing potential mates is, therefore, impossible. The absence of light implies the absence of photosynthesis which makes caves energy-poor in comparison to surface habitats. Finding food is extremely difficult since many environmental cues such as light/dark cycles, temperature changes, or air currents are absent (Howarth & Hoch, 2005). The absence of photosynthesis in groundwater also means that there is no oxygen production and the subterranean environment is therefore generally weakly oxygenated (Hervant & Malard, 2005). The percentage of oxygen in the groundwater depends on the oxygen transport from the surface and its consumption in the cave. Therefore, stygobites are often facing hypoxic stress and anoxia. Furthermore, high humidity makes respiration and water balance difficult to maintain and occasional flooding of voids causes additional stress (Howarth & Hoch, 2005). All these factors make caves very demanding habitats to which subterranean organisms must adapt.

On the other hand, some environmental challenges that are present in epigean habitats are lacking in caves. This refers to both abiotic (temperature and humidity) and biotic (predation and parasitism) ecological factors. Relatively constant temperature and humidity in subterranean habitats prevent desiccation and the need for strategies to avoid droughts, heat stress or cold. Lower energy input allows for fewer trophic levels, which reduces predation and parasitism in comparison to epigean habitats. All these are advantages for cave-adapted animals. Jointly, all these factors generate selection pressures which are very different in comparison with surface habitats.

Given the absence of primary producers, most subterranean habitats fully depend on external food resources (Poulson, 2005). Culver and Pipan (2009) indicated five main mechanisms of energy transport to the subterranean habitats from the surface: percolating water from the epikarst, freshwater, wind and gravity, animal activity, and plant roots. Even though caves are generally considered as oligotrophic habitats, it is important to emphasize that caves in the humid tropics are usually richer in organic matter than caves in temperate regions. There are several reasons for this difference: higher annual rainfall, relatively young age of the caves, location more close to the surface and very often having multiple entrances, and presence of large bat colonies during the whole year (which ensures large quantities of guano) (Mitchell, 1969).

0.4. EVOLUTION OF TROGLOMORPHY IN GENERAL

0.4.1. Classification of cave organisms

Efforts to classify cave organisms began to emerge during the early development of biospeleology. In the middle of the 19th century, knowledge about the ecological conditions in caves was scarce. The main environmental factor which was taken into account for making the classification was the most obvious one — the amount of light animals were exposed to. The first classification was proposed by Danish zoologist C.

Schiödte (1849, 1851) who classified animals that could be found in caves into four categories: *Skygge-Dyr* or shade-animals, *Tusmørke-Dyr* or twilight-animals, *HuleDyr* or cave-animals, and *Drypsteenshule-Dyr* or stalactite-cave-animals (Sket, 2008). A few years later, Schiner (1854) proposed a classification based on the ecological relationships between the animal and the subterranean environment. He distinguished three groups: trogloxenes, troglophiles, and troglobites. Trogloxenes are animals which appear sporadically in caves, but normally live outside subterranean habitat; troglophiles are cave-dwelling animals that may complete their life cycle in a cave, but can also survive outside caves; troglobites are obligate cave-dwellers which cannot survive outside and complete their life cycle in caves. After minor improvement of terminology by Racovitza (1907), this classification is still widely in use today, even though many other classifications have emerged (Vandel, 1964; Dudich, 1932; Ruffo, 1957; Barr, 1968; Clergue-Gazeau, 1974; Peck & Thayer, 2003). For aquatic subterranean animals equivalent terms stygoxen, stygophile, and stygobite are used. Sket (2008) made a review of historic classifications and standardized the definitions of categories deriving from the classic Schiner-Racovitza terminology (Table 1): ”(1) troglobiont is a species or population, strictly bound to a hypogean habitat; (2) eutroglophile is an essentially epigean species, but able to maintain a permanent subterranean population; (3) subtroglophile is inclined perpetually or temporarily to inhabit a subterranean habitat but is bound to the surface for some biological functions (e.g. feeding); (4) trogloxene is a species only occurring sporadically underground”.

Proposed terminology	**Definition**
Troglobite= troglobiont	Strongly bound to hypogean habitats
Eutroglophile	Essentially epigean species able to maintain a permanent subterranean population
Subtroglophile	Species inclined to perpetually or temporarily inhabit a subterranean habitat but is intimately associated with epigean habitats for some biological functions (daily e.g. feeding, seasonally, or during the life history e.g. reproduction)
Trogloxene	Species only occurring sporadically in a hypogean habitat and unable to establish a subterranean population

Table 1. Ecological classification of subterranean biota according to Sket (2008), modified.

It is important to emphasize that not all troglobites are necessarily troglomorphic. Large communities of organisms living on guano piles in caves encompass animals which are often not troglomorphic, probably due to a large food availability (Gnaspini, 2005). For example, guanobitic springtails exhibit only slight morphological modifications in relation to subterranean life (Deharveng, 2004). Still, they are troglobites since their entire biological cycle takes place inside caves (Gnaspini, 2005). Also, species recently isolated in subterranean habitats may not have had enough time to evolve troglomorphic features. Inversely, not all troglomorphic animals are troglobites. A few species of the entirely troglomorphic genus *Niphargus* are found in surface streams – probably because of recent secondary invasion of surface habitats from subterranean habitats.

0.4.2. Troglomorphy

The term troglomorphy (gr. *troglos* = cave, *morphos* = form), sometimes replaced by the synonymous terms troglomorphy or troglomorphism, was coined by Christiansen (1962) to define features associated with cave life. In the beginning the usage of this term referred only to morphological features, but subsequent usage was expanded to include any morphological, physiological, or behavioural feature that characterizes terrestrial cave organisms (Christiansen, 2005). For aquatic forms inhabiting subterranean waters, the equivalent term stygomorph is used (Christiansen, 2005). However, not every cave animal displays a complete set of troglomorphic traits. Some troglomorphic traits are general and widespread, such as the absence of pigment, anophthalmy and elongation of appendages, while others are limited to certain groups, like apterism, pseudo-physogastry in some groups of cave beetles.

The troglomorphic phenotype shared by unrelated, cave-adapted organisms is considered to be a classical example of convergence, resulting from evolution under similar, severe selection pressures in a subterranean environment (Christiansen, 1961; Hedin & Thomas, 2010; Protas et al., 2006). As an example, Christiansen (1961) stressed similar foot modifications in the Collembolan genera *Sinella* and *Pseudosinella* of the family Entomobryidae in unrelated, geographically remote linages. The foot complex underwent changes from being adapted to adhesion on smooth, wet rock surfaces and penetration into wet clay, to being adapted to movement over water surfaces in caves

(Christiansen, 1961; Christiansen, 2004). Besides foot modifications and loss of eyes and pigment, other morphological adaptations shared by different species belonging to these genera include: increased size of the adult, expansion of mesothorax and elongation of furcula (all associated with increased jumping ability), elongation of legs (associated with increased predation escape), elongation of antennae, and enlargement of antennal sensory organs (associated with increased olfactory and tactile sensitivity) (Christiansen, 1961; Christiansen, 2004). For a few troglobitic springtails, other troglomorphic traits were also documented: lower fecundity than in related epigean species, longer development time, decrease of the ability to regulate water exchange, slower respiratory metabolism, and higher resistance to starvation associated with an increase in the lipid fraction of the tissues (Thibaud, 1970; Thibaud & Deharveng, 1994). All these features are usually considered as evolutionary adaptations under the strong selective pressure of the cave environment (Deharveng, 2004).

Physiological adaptations of cave organisms are mostly influenced by darkness, food scarcity and hypoxia (Mathieu & Hervant, 2004). Different groups of troglobites, both vertebrates and invertebrates, have been used as model organisms for studies of physiological adaptations. Several experimental studies on cave crustaceans (*Stenasellus virei, Niphargus virei*, and *N. rhenorhodanensis*) and cave salamanders (*Proteus anguinus*) showed that troglobites generally have lower metabolic rates than their epigean relatives (Hervant et al., 1998; Hervant et al., 2000; Mathieu & Hervant, 2004). It has also been reported that their activity is considerably reduced, which minimizes oxygen consumption in periods of hypoxic stress (Hervant et al., 1996; Hervant et al., 1999).

As a response to ecological conditions in the subterranean environment, changes in behaviour are noted in many groups of troglobites. Most behavioural modifications related to life in subterranean habitat affect feeding, reproduction, social behaviour, photoresponses, and circadian rhythms (Romero, 2004). Changes in feeding behaviour are well-documented in cave fishes (Schemmel, 1980; Wilkens, 1988). Experimental studies with *Astyanax* revealed that cave populations of this genus are bottom feeders which rely on an enlarged taste bud area on the ventral side of the head in finding food. In contrast, surface populations of the same genus rely on vision in locating food and they do not collect food from the bottom but from the water column. In order to maximize food finding, cave-dwelling *Astyanax* swim continuously while decreasing the angle of the body relative to the bottom. This way, they are increasing the contact area between chemoreceptors in their skin and food on the bottom (Schemmel, 1980; Wilkens, 1988). Aggressive behaviour and aggregation/schooling are generally reduced in many troglobites, from insects (Parzefall, 2000) to fish (Romero, 2001a) because of low predation. Surprisingly, photoresponses are reported in some troglobitic fishes (Romero, 2001a). Experiments showed that cave fishes have the tendency to stay in the dark side of the aquarium, which is considered as a behaviour inherited from epigean ancestors (Romero, 2001a).

Epigean organisms show a series of physiological and behavioural responses known as circardian and seasonal rhythms, mediated by a "biological clock", which are activated by changes of climate. However, in subterranean habitats these variations do not occur, and such cycles are absent in troglobites (Lamprecht & Weber, 1992; Romero, 2004). Acoustic communication is a crucial component of the courtship behavior in some insects, e.g. planthoppers (Claridge, 1985). In troglobitic planthoppers of the family Cixiidae from Havaiian lava tubes, changing of courtship pattern was documented as part of adaptation to the subterranean environment (Hoch, 2000). Contrary to surface-dwelling close ancestors, in populations of cave planthoppers it is the females who initiate the calling and they never rejected a responding male (Hoch, 2000). These changes may even be the result of increased competition among females for access to males. Such a sex role reversal has also been reported for cave psocopteran genus *Neotrogla* known not only for reversed sex roles but also sex organs (Yoshizawa et al., 2014).

0.4.3. Evolution of regressive traits (selective vs. neutral hypothesis)

Since the discovery of the first troglobitic species (*Proteus anguinus* Laurenti, 1768), the bizarre morphology of cave organisms has provoked the interest of evolutionary biologists. Different hypotheses have been proposed to explain the evolution of elaborated and reduced features. Elaborated traits are generally considered to be adaptations since they are clearly beneficial in cave environment (Kane et al., 1990; Culver et al., 1995). Reduced metabolic rate, for example, is adaptive in the cave environment: organisms that do not exhibit reduction in metabolic rate occupy niches that are relatively resource-rich (Kane et al., 1990). Elaboration of sensory organs such as antennae (in arthropods) and lateral line (in fish) facilitates effective foraging when eyes cannot be used, and so this is also generally treated as adaptation (Sket, 1985; Kane et al., 1990; Culver, 1995). More difficult is to explain the reduction or loss of traits which were present in the presumed surface-dwelling ancestors of cave animals.

At least 14 hypotheses have been proposed to explain the mechanisms of regressive changes (in Christiansen, 2005) and some of them date back to the earliest theories of evolution. Lamarck explained the reduced eyes of *Proteus* as a result of lack of use, and drew a parallel to what he assumed had happened in moles (*Talpa*). According to Lamarck, in the absence of light in caves, disuse of eyes will lead to their degeneration and this then be passed on to offspring. He surmised that the ancestor of *Proteus* had fully developed eyes but, over time, repeated cycles of disuse, degeneration and inheritance, led to anophthalmy (in Kane & Richardson, 2005). Darwin, like Lamarck, attributed the blindness of cave organisms to disuse, but for other non-cave organisms (e.g. wingless island-dwelling beetles) he argued that losses are the result of natural selection (in Culver et al., 1995). Regarding the eye loss of cave fishes, he wrote: “As it is difficult to imagine that eyes, although useless, could be in any way injurious to animals living in darkness, I attribute their loss wholly to disuse” (Darwin, 1985). Vandel held to the theory of organicism, stating that lineages, like individuals, go through three stages: birth, specialization, and senescence (Vandel, 1964). He hypothesized that regressive traits are actually degenerative changes, generated as a result of lineage senescence, and not influenced by environment or selection (Vandel, 1964). Even though they were never widely supported, and often sharply criticized, Lankester’s escape hypothesis (Lankester, 1893) and Ludwig’s trap hypothesis (in Christiansen, 2005) are interesting from a historical aspect. Lankester believed that within any animal population there are individuals with weakly developed eyes. If, by chance, individuals from that population fall or are swept into caves, individuals with normally developed eyes will be able to see the light and escape while those with degenerated eyes will remain trapped in a cave (Lankester, 1893). Ludwig hypothesized that weakly pigmented surface-dwelling organisms would be sensitive to light and thus would concentrate in caves (in Christiansen, 2005). This hypothesis was disproved by Fong and Culver (1985) who showed that inside the populations of some amphipods, individuals with larger eyes are actually more photophobic.

Neo-Lamarckians like Packard attributed both elaborations and reductions of traits to the effects of use and disuse and the hereditary transmissions of these acquired changes (in Culver et al., 1995). Neo-Darwinians, particularly Christiansen (1961, 1965) and Poulson (1963), have studied not only reduced, but also the elaborated characters of cave animals. They agreed that natural selection was the major factor moulding the morphology of cave animals and they both used the degree of degeneration as a rough estimate of the time of isolation in caves (in Culver, 1995).

Although many of the aforementioned theories are no longer tenable in a modern evolutionary-biological framework, the question regarding the evolution of regressive traits remains controversial. Modern theories of the evolution of reduced characters may generally be divided into two main streams: one invoking natural selection (selective hypothesis), and the other invoking genetic drift and neutral mutations (neutral hypothesis). The argument in favour of the first theory is that in oligotrophic environment, where energy economy is important, any mutation that reduces a redundant structure provides a selection advantage. Thus, individuals that invest less energy in the development of "functionless" structures like eyes and pigmentation may have better chances to survive and produce offspring (Mitchell, 1969; Culver, 1982; Poulson, 1985, Sket, 1985; Culver and Wilkens, 2000). Moreover, reductions of these structures may represent a trade-off for the elaboration of extra-optic sensory structures such as antennae (Sket, 1985; Kane & Richardson, 2005). On the other hand, according to the neutral hypothesis, regressive traits do not have an impact on the fitness of cave organisms, so they will not be removed by natural selection. Since mutations generally tend to be degenerative, over time they will accumulate and eventually will lead to the reduction or loss of these structures. There are studies that support each of these two hypotheses.

Extensive studies on a crustacean, *Gammarus minus*, showed that eye reduction is the result of natural selection, producing adaptation to the subterranean environment (Culver et al., 1995). The authors noted that morphological differences between cave-dwelling and surface-dwelling populations of *G. minus* have a genetic basis and are therefore heritable. Studies demonstrated that more strongly troglomorphic individuals (with smaller eyes, larger antennae, and larger body size) from cave populations mate more frequently and produce more offspring in comparison to the less troglomorphic individuals (with larger eyes, smaller antennae and smaller body size) within the same population. Therefore, the more troglomorphic individuals in the population are more fit. However, this hypothesis is weakened by the fact that reductions do not only occur in food-limited caves, but are also observed in food-rich underground environments (Mejía-Ortíz & Hartnoll, 2005).

Other studies performed on different model organisms, however, suggest that regressive traits are of no selective value and that mutation pressure and genetic drift play the major role (Kimura, 1983; Wilkens, 1988). Wilkens (1988) investigated the effects of neutral mutation on eye and pigment reduction in cave fishes. He made a comparative study of the surface and cave populations of the characid fish *Astyanax mexicanus* (De Filippi, 1853) (=*Astyanax fasciatus*), distributed in northeast Mexico. Surface and cave populations of this species are interfertile, which makes them an excellent model system for evolutionary research (Wilkens, 1988; Culver and Wilkens, 2000). Troglobitic *A. mexicanus* in underground streams descends from the surface species, which has normally developed eyes (Wilkens, 1988). Genetic analysis of eye reduction shows that this process is genetically based but involves relatively few genes (Wilkens, 1988). In darkness, eyes and pigmentation are useless, so stabilizing selection will no longer act on such features. Recessive mutations of genes responsible for the development of these structures will become neutral and are therefore no longer eliminated by natural selection but accumulated randomly. Over time, accumulation of a large number of recessive mutations will lead to reduction of structures (Wilkens, 1988).

The main argument against the role of selection in eye regression of cave fishes is that the cost of making an eye is insignificant in comparison to the cost of its replacement by the tissue in the socket (Eigenmann, 1909; Culver, 1982). Furthermore, Mitchel et al. (1977) argued that the development cost is paid anyway because the eyes start developing and only degenerate after many cell cycles of tissue growth and replacement. Protas et al. (2007), however, showed that even though the energetic cost of developing an eye may be trivial, the expense of maintaining a functional eye is much greater. They concluded that regression can be achieved in a variety of ways — it may be effected by active selection as well as by the passive accumulation and fixation of damaging mutations (Protas et al., 2007). Similarly, a recent literature review by Rétaux & Casane (2013) shows that both genetic drift (neutral hypothesis) and direct and indirect selection (selective hypothesis) occur together during the loss of eyes in cave animals.

0.5. BIOGEOGRAPHIC AND PHYLOGEOGRAPHIC PATTERNS IN TROGLOBITES

Species richness in the subterranean environment is generally smaller compared to surface habitats. Several decades ago, Sket (1999) proposed a variety of possible reasons for this pattern: reduced area of ecotonal regions between surface and subsurface, reduced subterranean habitat diversity, and reduced food resources. Cave fauna is well known for its extremely high degrees of short-range endemism (Gittenberger, 1975; Haase & Schilthuizen, 2007; Schilthuizen et al., 2012). Due to the fragmented nature of subterranean habitats leading to restricted dispersal, most of its diversity is manifested regionally and not locally (Culver & Sket, 2000a). However, little is known about the processes that shape subterranean biodiversity, as studies have only recently begun to emerge (Bregović & Zagmajster, 2016).

Our knowledge about the diversity of troglobites in different geographical regions is rather uneven; subterranean habitats in temperate regions are generally better explored than in the tropics. Also, some taxonomic groups are better studied than others. This is probably because some organisms are easier to spot and collect in caves or simply because they are more popular among scientists. The first biospeleological explorations started in the Dinaric Mountains in Europe, and caves in this region are still the most comprehensively studied ones (particularly in Slovenia), but caves in the French Alps and Pyrenees are nowadays also well explored. Until the 1980s, it was believed that obligate cave species are absent or rare in the tropics (Howarth, 1987; Deharveng, 2005). This view was supported by the fact that in the humid tropics, where thermal amplitudes are low, the cave environment does not differ much from the surface environment. Moreover, as caves form and disappear much faster under humid tropical conditions, there is not enough time for organisms to develop troglomorphic adaptations and become confined to subterranean habitats. However, as biospeleological research in the tropics continued, a number of troglobites have, in fact, been discovered. It became evident that troglobites are not restricted to the temperate zone but also exist in the tropics and sometimes even form rich communities there (Deharveng, 2005).

0.5.1. Diversity of the Dinaric cave fauna

The Dinaric Mountains in South-Eastern Europe are recognized as the world's major biodiversity hotspot for subterranean fauna (Sket et al., 2004a; Culver et al., 2006; Deharveng et al., 2012). From 20 caves in the world which have more than 20 troglo- and stygobites, 15 are located in the Dinaric Mountains. (Culver & Sket, 2000). This does not seem to be an artifactual consequence of a longer history of investigation. The position on the edge of formerly glaciated regions allowed the Dinaric Mountains to be a refuge for thermophile elements during the Pleistocene (Sket, 2005). This has resulted in a cave fauna that is second to none in diversity world-wide. According to the latest census of the Dinaric obligate subterranean fauna, this mountain range, comprising an area of 60,000 km^2, hosts 600 troglobites and 330 stygobites (Sket et al., 2004b). For comparison, in the whole of North America, comprising about 24,709,000km^2, 930 troglobites and 430 stygobites were recorded (Sket, 2005). The highest species richness of stygobites is in north-western part of the Dinaric range (i.e., Slovenia) while the situation is different for the terrestrial cave fauna, where the highest diversity is attained in the south-eastern range (Bosnia and Herzegovina, Montenegro, Croatia) (Sket et al., 2004b). Not only does this region have the highest numbers of subterranean species, it also comprises the greatest phylogenetic diversity, with a broad assortment of animal orders (Sket, 2004b). Some animal groups such as freshwater sponges (*Eunapius subterraneus*), cnidarians (*Velleovrhia enigmatica*), clams (*Congeria kusceri*), and tubeworms (*Marifugia cavatica*) do not occur anywhere else in the world except in caves of the Dinaric range. Moreover, the only non-American stygobiont amphibian, *Proteus anguinus*, inhabits the Dinaric karst. Surprisingly, however, troglobitic fishes are not recorded in this region nor in other mountain ranges in Europe (Romero, 2001b; Rantin & Bichuette, 2013).

With more than 250 species, the most diverse group of troglobites in the Dinaric Mountains is beetles (Sket et al., 2004b). Cave Coleoptera of the subfamily Cholevinae, with its tribe Leptodirini are the most species-rich group, comprising 175 species of 50 genera, almost entirely endemic to the Dinarides (Sket, 2005). Next in diversity is the family Carabidae with 12 genera and 80 troglobitic species of its subfamily Trechinae (Sket, 2005). The subfamily Pselaphinae is represented by only 20 troglobiotic species (Sket, 2005), but, due to a very small body size and lifestyle predominantly in cave soil and under the rocks, this group is understudied and more species are expected. Two centers of troglobitic beetle richness are recognized in Dinaric karst. The north-western part exhibits high richness of Trechinae, while in the south-eastern Dinarides, the troglobitic Leptodirini are the most diverse (Zagmajster et al., 2008). The second most diverse group after Coleoptera are spiders. Most troglobitic spiders distributed in the Dinaric Mountains belong to the families Dysderidae and Linyphiidae. The other groups represented by troglobiotic species are (in descending order): false scorpions (Pseudoscorpiones), millipedes (Diplopoda), woodlice (Isopoda: Oniscidea), snails (Gastropoda), centipedes (Chilopoda), harvestmen (Opiliones), and planarians (Turbellaria: Tricladida) (Sket, 2005).

Among the stygobites, the predominant group in Dinaric hypogean waters are crustaceans, particularly copepods and amphipods, with approximately 60 species each (Sket, 1999b; Sket, 2005). With about 45 species, the genus *Niphargus* is the most diverse among amphipods (Sket, 2005). Another highly diverse group, with around 130 species, are aquatic snails. The Dinaric aquatic gastropods (almost entirely belonging to the superfamily Hydrobioidea) represent close to half of the world's known stygobitic snail fauna (Sket, 2005). The only stygobitic vertebrate in Europe is the cave salamander *Proteus anguinus* with holodinaric distribution. *Proteus anguinus* comprises two subspecies, one troglomorphic (*P. anguinus anguinus* with reduced eyes, depigmented skin and elongated head) and one nontroglomorphic (*P. anguinus parkelj*) with pigmented skin and normally developed eyes.

0.5.2. Biogeographic and phylogeographic patterns in the Dinaric subterranean habitats

A large number of obligate cave species and subspecies, but also some genera, are known from a small number of localities, sometimes even from a single cave or spring. A good example of this striking subterranean endemism is *Troglomysis vjeternicensis*, the cave mysid, known only from a single lake in Vjeternica cave in Bosnia and Herzegovina (Ozimec & Lučić, 2009). These small distribution areas are difficult to explain. They might be a consequence of locally limited immigrations from surface to cave habitats or they might present the remains of formerly wider area (Sket, 2005). Most subterranean taxa are endemic for the Dinaric region, but endemism is also high across the different regions within the Dinarides. For example, the list of Croatian cave fauna comprises 338 taxa, predominantly Dinaric endemics (330), of which the majority are Croatian endemics (298) (Bedek et al, 2006).

Distribution patterns of many taxa do not follow the recent hydrological divisions and there are examples of both species which are limited to a certain part of the drainage area and species which cross the hydrological divides (Sket, 2005). The possible explanation is that these distribution areas were attained in geologically past drainage areas and preserved until today (Sket, 2005).

Fig. 1. A selection of Dinaric troglobites. A. *Proteus anguinus* Laurenti, 1768; B. ordo Palpigradi; C. *Roncus sp.*; D. *Hadesia vasiceki* Müller, 1911; E. *Echinarmadillidium fruxgalli* (Verhoeff, 1900); F. *Machaerites* sp.; G. *Meledella werneri* Sturany, 1908.

Some taxa, however, have holodinaric distribution. There are cases of several species of the same genus distributed in almost the entire Dinarides and cases of apparently single species or genera occurring over wide Dinaric range. Nevertheless, in the latter case, they are never contiguously distributed over the Dinaric region, but split into different populations in hydrographically isolated karst areas (Sket, 2005). This distribution pattern is noted for some of the most remarkable representatives of Dinaric cave fauna such as the amphibian *Proteus anguinus*, the clam *Congeria kusceri*, the tubeworm *Marifugia cavatica*, and the cnidarian *Velkovrhia enigmatica* (Sket, 2005). A negligible number of taxa have very wide distribution areas, extending beyond the boundaries of the Dinaric Mountains. Some examples of generalists with wide distribution area across Europe that have penetrated to subterranean environment are the isopod *Asellus aquaticus*, the malacostracans *Synurella ambulans* and *Troglocaris anophthalmus,* and the gastropods *Ancylus fluviatilis* and *Zospeum* sp. (Sket, 2005). However, where molecular techniques have been applied, these species have turned out to be species aggregates with very similar morphology caused by troglomorphic convergence (Zakšek et al., 2007, Weigand et al., 2011).

0.5.3. Biogeographic and phylogeographic patterns in subterranean habitats in the tropics

Subterranean habitats in the tropics are generally poorly explored; data are scarce and taxonomically and geographically uneven. Some regions are better explored than others and certain groups are better studied in certain tropical regions. For instance, freshwater stygobites outside micro-crustaacea have been relatively well sampled in Cuba or parts of Thailand, but data are lacking for most other karstic areas in the tropics (Deharveng, 2005). Anchialine stygobites have been extensively studied in the Neotropics while in Southeast Asia only few species have been described (Deharveng, 2005). Interstitial habitats are generally poorly investigated since sampling is more difficult. In Southeast Asia these habitats are undersampled and data on interstitial fauna are lacking (Deharveng, 2005). On the other hand, they have been extensively studied in parts of tropical America (Iliffe, 2000). Many basic data illustrating this uneven knowledge can be found in the Encyclopaedia Biospeologica (Juberthie & Decu, 1994).

Caves in the Malaysian state of Sarawak have the largest number of named troglobites, but the diversity of cave fauna is still far from definite since many groups like Collembola and microarthropods have been completely neglected (Deharveng, 2005). Conversely, Collembola are among the best studied troglobites in Thailand (Deharveng, 2005).

During the last three decades, explorations in the tropical caves have been intensified and the richness of troglobites has been confirmed in many parts of the tropics. Deharveng (2005) pointed out the biodiversity patterns in the tropics that have recently begun to emerge, as follows:

1. The diversity of terrestrial fauna is higher in the Oriental and Australian regions than in the Neotropics and Africa, while the highest diversity of stygobites is recorded in anchialine caves of tropical America and Australia.
2. Large karstic areas are richer in troglobites than small karsts.
3. "Most diversity in caves is expressed regionally rather than locally'' (Culver & Sket, 2000a).
4. Troglomorphy increases with seasonality, with decreasing mean annual temperature, and with increasing drought.
5. Contrary to deeply rooted opinion, climatic relics are present in some tropical caves; for instance, caves in the Cape Range of western Australia or in dry or strongly seasonal tropical regions.

High fragmentation of limestone areas in many tropical karsts causes the isolation of populations of troglobites. Therefore, habitat fragmentation and selective constraints imposed by the underground ecological conditions are the main factor which generate high species richness and endemism (Jeannel 1924, Gibert & Deharveng 2002).

0.6. SPECIATION AND RADIATION IN CAVE ORGANISMS

Speciation, the evolutionary process by which new species are formed has long intrigued biologists working on cave fauna. This is not surprising since extreme and isolated habitats such as subterranean habitats are considered to be ''natural laboratories'' for the study of speciation (Barr & Holsinger, 1985; Schluter, 2000). General speciation models of obligate cave animals were summarized by Barr & Holsinger (1985), Coineau & Boutin (1992), Sbordoni at al. (2000), Holsinger (2000), Schilthuizen et al. (2005).

For more than a century, it was assumed that troglobites were present only in temperate regions that have been affected by glaciation and that they evolved in isolation while the epigean ancestors became extinct. Early works were, therefore, in favor of a strictly allopatric model of speciation (Jeannel, 1943; Vandel, 1964; Barr, 1968), implying that troglobitic species are relicts of formerly widespread surface populations (Jeannel, 1943). Among cave biologists, this speciation model is known as the "Climatic Relict Hypothesis" (Peck & Finston 1993). According to this model, evolution of troglobites from ancestral epigean populations resulted from their isolation caused by severe climatic changes and subsequent extinction of surface ancestors. This view was supported by the fact that many troglobites in temperate zone do not have closely related epigean ancestors. Moreover, it was considered that troglobites are absent in tropical regions since these regions were not affected by glaciation (Jeannel, 1950).

When the presence of troglobitic communities in tropical caves became evident (Howarth, 1973; Chapman, 1980; Gaspini & Trajano, 1994; Deharveng & Bedos, 2000), and more epigean-endogean species pairs were discovered, the allopatric model could no longer remain as the single universal explanation for the origin of troglobites. As an alternative, the parapatric "Adaptive Shift Hypothesis" of cave speciation was proposed. In the last three decades, many authors have expressed themselves in favor of this adaptive shift model of cave speciation (Barr & Holsinger, 1985; Howarth, 1987; Culver, 1987; Howarth 1982; Peck & Finston 1993; Rivera et al. 2002). According to this model, colonisation of the subterranean medium is driven by the opportunity to exploit new resources. After colonisation of the new habitat, the new population may undergo morphological, behavioural, and physiological changes, while still engaging in gene-flow with the epigean ancestor, and become a distinct species. Drastic change in habitat (from epigean to subterranean) is considered to be the driving force for genetic divergence and speciation (Nosil, 2012). As a result, cave and surface populations will initially have sympatric or parapatric distributions. Adaptive shift occurs in three steps. First, a new ecological niche opens when new resources become available for exploitation. This can occur either when a population is expanding its range or during the formation of subterranean habitats by successive geological processes (Howarth & Hoch, 2005). The second step is the shift in behaviour, physiology, or morphology, to exploit the new resources or to survive under new environmental stress, as demonstrated in a comparative study of cave and surface Collembola (Christiansen, 1965).

The final step includes the establishment of a barrier to gene flow between surface and cave population. There are several possible (not mutually exclusive) ways to facilitate the formation and maintenance of such a barrier: selection against intermediate phenotypes leading to assortative mating or shifting the cave population away from the hybrid zone (Howarth & Hoch, 2005). It should be pointed out that both modes of speciation, although often presented as alternatives, are the extremes of a continuum and phases of allopatry may be important even in an adaptive shift process, and, conversely, resource-based selection may play a role in climate relict situations.

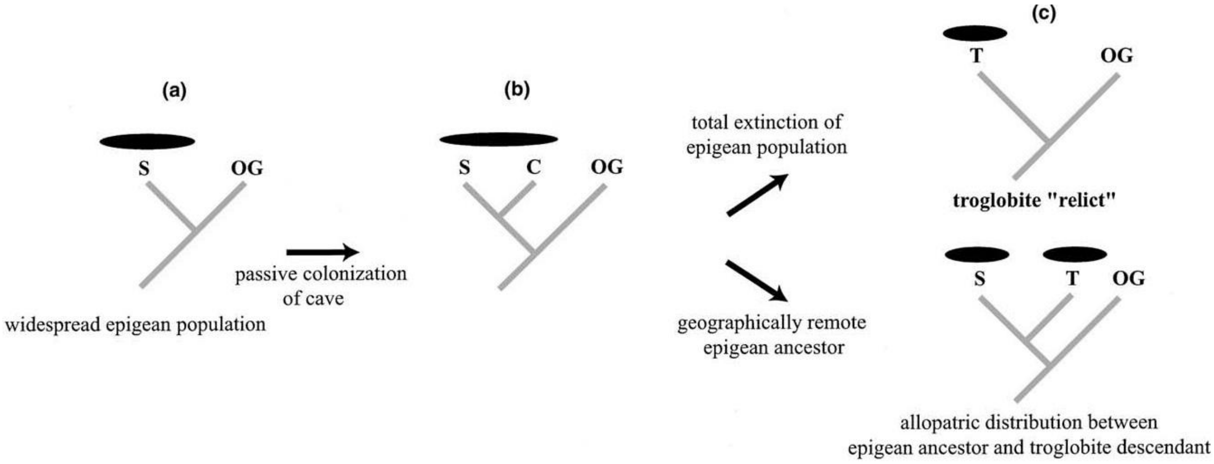

Fig. 2. Phylogenetic prediction of speciation by the Climatic Relict Hypothesis (CRH). OG. outgroup; S. surface; C. cave; T. troglobite. Ellipses represent geographic distribution of populations (Rivera et al., 2002).

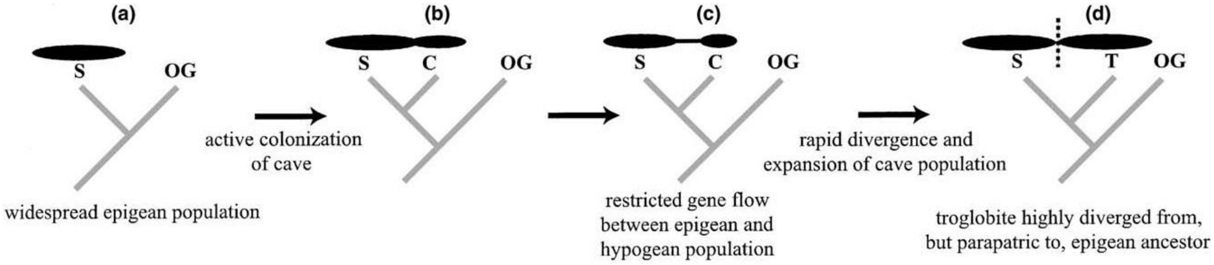

Fig. 3. Phylogenetic prediction of speciation by the Adaptive Shift Hypothesis (ASH). Symbols are as in Fig. 1. (Rivera et al., 2002).

Until recently, the prevailing view regarding the radiation of troglobites was that once a lineage has adapted to the subterranean environment, it is unable to expand or diversify over a larger area because of environmental constraints and, as a result, it remains restricted to a small geographical area (Barr & Holsinger, 1985; Culver & Pipan, 2009). The existence of widespread ancient troglobitic lineages was usually interpreted as a result of multiple independent colonization followed by subsequent extinction of the ancestors. Some studies support this hypothesis (for Pyrenean troglobitic Trechini and Leptodirini; Faille et al., 2011; Fresneda et al., 2011), others suggest a single subterranean colonization event for these clades (Faille et al., 2010; Ribera et al., 2010). This last studies suggest that troglobites would be able to disperse across unfavourable habitats, diversify and persist in subterranean habitats for long periods (Cieslak et al., 2013), which raise a number of problems. Moreover, some studies show that troglobites are even able to expand their range over non-karstified areas through MSS (Rizzo et al., 2013), but these cases are exceptional and rare.

CHAPTER I

INTRODUCTION TO THE MODEL SYSTEM

1.1. OBJECTIVES OF THE STUDY

As explained in the previous chapter, two separate phases may be distinguished in the evolution of subterranean taxa. First, one or multiple independent adaptation events to a subterranean environment; second, subsequent radiation within the troglobitic clade. The aim of this work is to focus on this second phase, and to study the evolutionary radiation in the subterranean environment using the strongly-adapted beetle genus *Anthroherpon* as a model. This genus is a suitable model because it exhibits the most pronounced troglomorphic characters among Coleoptera with adaptations to subterranean environment, while also having diversified into a multitude of species. To understand the evolutionary diversification and biogeographical history of this genus, I combined traditional taxonomy with molecular and biometric approaches.

The specific questions I aim to answer in this work are:

1. Do the previous, qualitative and purely morphology-based species and subspecies delimitations and classifications stand up to objective and quantitative analyses based on several independent data sets?
2. How are its species phylogenetically inter-related to one another and within what time frame did this phylogenesis take place?
3. How has the group developed biogeographically?
4. Have the species evolved morphologically in a neutral manner or are there indications of adaptive divergence?
5. What are the origin/relationships of the genus?

To answer these questions, I have adopted the following research strategy: The first step is to build a phylogeny of the subtribe Anthroherponina and reveal the phylogenetic relations between its genera to test the validity of taxonomic groups defined by previous authors based on morphology. Monophyly of this subtribe, presumed exclusively on morphology by Perreau & Pavićević (2008), and monophyly of the genus *Anthroherpon*, is also tested. Phylogenetic reconstruction of *Anthroherpon* allows me to test if characters that have been traditionally accepted as indicative for its classification are indeed reliable. I clarify the status of the species and subspecies of the genus and study the inter- and intra-populational morphological variability to identify the most relevant characters for taxonomy. I use a molecular clock technique to date the divergence of lineages within the obtained phylogenetic tree. This allows me to answer the first two questions.

The second step is to use Bayesian methods to search for the biogeographic origin of the genus on the basis of the obtained phylogeny and the limits of its geographical area. This gives me the answer to the third question.

In the last step, I combine morphometric analysis of morphological features considered to be adaptations to subterranean habitats, with a molecular phylogenetic reconstruction of the genus. This allows me to answer the final question, on the evolutionary trajectories of these characters. This is particularly interesting because so far, geometric morphometric approaches have rarely been used in the study of troglobitic beetles.

Finally, the overall results of the study are applied in the context of current ideas on the evolution of Dinaric troglobites.

1.2. INTRODUCTION TO LEPTODIRINI (COLEOPTERA, LEIODIDAE, CHOLEVINAE)

The beetle family Leiodidae is a worldwide group of staphylinoid beetles organized into six subfamilies and with about 4,135 described species (Newton, 2016). The two most diverse subfamilies, Leiodinae and Cholevinae, are distributed in both tropical and temperate regions while the remaining four subfamilies are much smaller and with mainly temperate distribution (Newton, 2016). Monophyly of the subfamilies is generally accepted (Newton, 1998, 2005) although there is no formal comprehensive phylogenetic analysis. Leiodidae is a very old group, with fossils dating back to the late Jurassic (Perkovsky, 1999, 2002), showing ancient ("Gondwanan") vicariance patterns (Newton, 2016).

The tribe Leptodirini Lacordaire, 1854 is the most species-rich tribe in the subfamily Cholevinae, comprising 229 genera and 911 (mostly polytypic) species (Perreau, 2000). It includes 7 subtribes: Anthroherponina Jeannel, Bathysciina Horn, Bathysciotina Guéorguiev, Leptodirina Lacordaire, Pholeuina Reitter, Platycholeina Horn, and Speleobatina Guéorguiev (Perreau, 2000). Leptodirini have a Palearctic distribution: Iberian Peninsula, Balkan Peninsula, southern Alps, Romania, southern

Russia, the Caucasus, Middle East, and Iran, with the highest diversity in the Mediterranean basin (Perreau, 2000, 2004; Ribera et al., 2010). Six genera distributed outside of this core area were considered closely related to Leptodirini. However, after molecular phylogenetic analysis, two of these (*Fusi* Perkovsky, 1989 and *Sciaphyes* Jeannel, 1910, both from East Asia) were excluded from the tribe (Fresneda et al., 2011). The North American *Platycholeus* Horn, 1880 has proved to be the sister group of the remaining Leptodirini with an estimated age of vicariant separation in the Early Eocene (Fresneda et al., 2011). The remaining three monotypic genera were found in Far East Asia: *Coreobathyscia* Szymczakowski, 1975 in South Korea, *Proleptodirina* Perkovsky, 1998 in eastern Russia, and *Sinobathyscia* Perreau, 1999 in the Hubei region in eastern China (Newton, 1998; Perreau, 2000, 2004). Their phylogenetic position and presumed close relation to Leptodirini is also uncertain due to potentially convergent morphological characters resulting from the adaptation to the subterranean environment (Fresneda et al., 2011). The easternmost geographic limit of the undisputed Leptodirini is therefore considered to lie at the Zagros and Elburz mountains in Iran (Fresneda et al., 2011).

Fig. 1. Distribution of Leptodirini, excluding the *Platycholeus* Horn and genera with uncertain phylogenetic position (modified from Fresneda et al., 2011).

Leptodirini is one of the largest group of insects living in the subterranean environment, surpassed only by the tribe Trechini (Carabidae) (Fresneda et al., 2007; Ribera et al., 2010). All species are more or less adapted to the subterranean environment (except two termitophilous species) and all are fully anophtalmic (except fewer than 15 species that have extremely reduced eyes). All Leptodirini are considered to be scavengers (Newton, 2016), but food preferences are inadequately known, at least for the most strongly troglomorphic species of the subtribe Anthroherponina. The characters shared by all members of the tribe Leptodirini are: female protarsi tetramere, spiculum gastrale of the abdominal IX segment in males fused with lateral epipleurites; most genera have complete mesoventral processus separating the two mesocoxal cavities (except a few species of Anthroherponina in which this structure is assumed to have secondarily disappeared) (Perreau, 2008b).

The following morphological characters have been used (Jeannel, 1924; Guéorguiev, 1976; Perreau, 2000) to define subtribes of Leptodirini: i) the reduction of male protarsi from five to four tarsomeres (as in females of all Leptodirini) characterizes Bathysciotina and Bathysciina; ii) the presence of an external row of protibial spines correlated to a comb of spines of equal length at the apex of the meso- and metatibia characterizes Bathysciotina and Leptodirina. However, their phylogenetic significance is highly questionable. For example, Perreau & Pavićević (2008 a, b) noted the absence of the protibial row of spines in several genera which are nevertheless generally considered as belonging to Leptodirina: *Nonveilleriella* Perreau & Pavićević, 2008, *Rozajella* S. Ćurčić, Brajković & B. Ćurčić, 2007, *Parapropus* Ganglbauer, 1869, *Leptostagus* Karaman, 1954, *Petkovskiella* V. B. Guéorgiev, 1976, etc.

The monophyly of Leptodirini is morphologically supported by the reduction of the number of protarsomeres in females from five to four (Jeannel, 1924; Giachino et al., 1998; Newton, 1998). However, the study of Carvalho & Gnaspini (2015), based on morphological characters on the pretarsus and terminal tarsomere, challenge the previously suggested monophyly of Leptodirini. All Leptodirini have empodial setae fused basally and arranged diagonally or vertically relative to one another except the subtribe Anthroherponina, whose empodial setae are aligned laterally to each other and do not seem to have the same degree of basal fusion as in other Leptodirini (Carvalho & Gnaspini, 2015). This implies a possible polyphyly of Leptodirini that needs to be investigated by a more comprehensive approach. Recent molecular studies have focused mostly on the internal phylogenetic relationships of Leptodirini (Fresneda et al., 2007, 2011; Ribera et al., 2010). The monophyly of the western Mediterranean Leptodirini is well supported by both morphological (Fresneda et al. 2007) and molecular (Ribera et al., 2010) evidence, but no phylogenetic studies have been performed on Leptodirini from other parts of their areal.

Most species of Leptodirini show all the typical modifications associated with the subterranean life and have very restricted geographical distributions (Salgado et al., 2008; Cieslak et al., 2014a). Some genera, in addition to common troglomorphic morphological traits, have developed exceptional features (see below: Subtribe Anthroherponina Jeannel).

1.3. TRENDS IN THE EVOLUTION OF TROGLOMORPHY IN LEPTODIRINI

Beetles of the tribe Leptodirini have evolved numerous features associated with life in the subterranean environment. Troglomorphic traits in Leptodirini include, apart from anophthalmy and melanin pigment loss: cuticle thinning, loss of membranous wings, elongation of appendages, narrowing of the pronotum, pseudo-physogastry or narrowing of the abdomen (Vandel, 1964; Peck, 1973; Sbordoni, 1980; Ribera et al., 2010). Increase in appendage length is correlated with the increase of the number of sensory organs (Culver, 1982) and is not due to positive allometric effects (i.e. differential growth of different body parts). Increase in number of sensory organs is particularly beneficial in a subterranean environment because it ensures better ability to detect food, potential mates or predators in the absence of light. The increase of antennae length is correlated with the increase in the number of olfactory receptors (Lucarelli & Sbordoni, 1978; Casale et al., 1998; Decu & Juberthie, 2004) which allows sensing the olfactory information from longer distances.

1.3.1. Morphological types in Leptodirini

Within the Leptodirini, four morphological types are recognized: bathyscioid, pholeunoid, scaphoid, and leptodiroid (Fig. 2). They are considered to be the progressive stages in adaptation to subterranean environments (Jeannel, 1924), but since there is no evidence based on the molecular phylogenetics, we cannot be sure if this is indeed true. The bathyscioid morphological type is characterized by a small, egg-shaped body and short appendages. It is present in the muscicolous or endogeous species such as *Bathyscia montana* Schiødte, 1848, distributed in the Dinaric Mountains. Beetles with this type of external morphology are considered to be the least adapted to the subterranean environment because this type of morphology is similar to the morphology of epigean Cholevinae and is also encountered in many troglophiles. Species with pholeunoid body-shape are larger and have more elongated body and appendages, such as the genera *Pholeuon* Hampe, 1856 and *Parapropus* Ganglbauer, 1899. The scaphoid morphological type was named after *Remyella scaphoides* Jeannel, 1931, a species with very elongated body and appendages, and slender, spindle-shaped elytra. The leptodiroid morphological type, named after *Leptodirus hochenwartii* Schmidt, 1832 from Slovenia and Croatia, exhibit the highest degree of troglomorphy. Species belonging to this morphological type have extremely long appendages, extremely elongated head and pronotum, and hemispherical elytra.

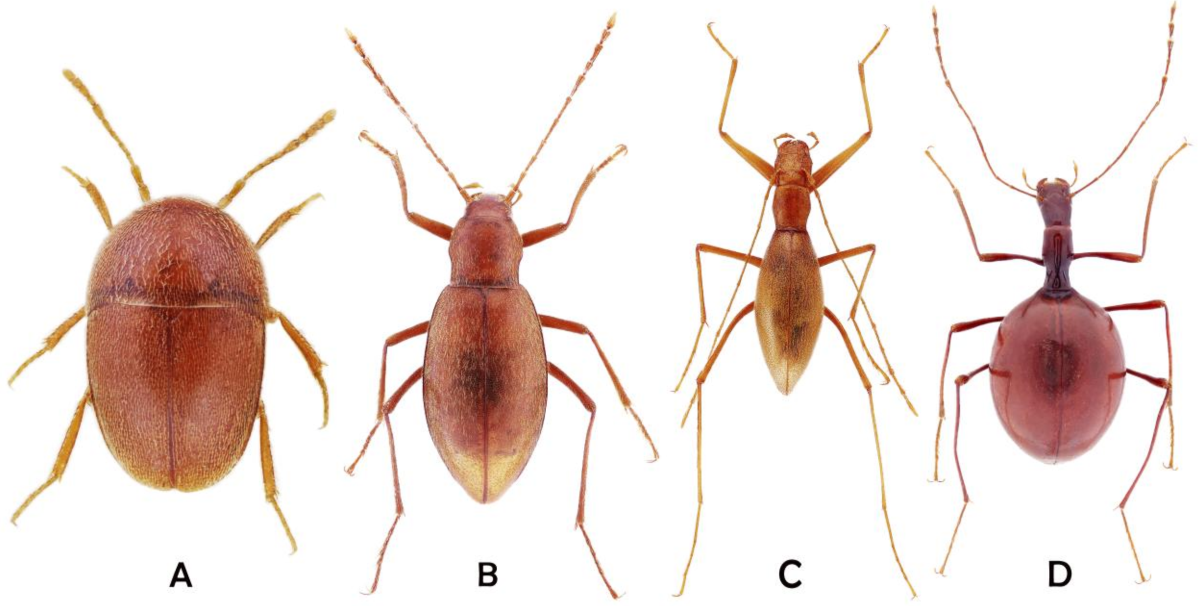

Fig. 2. Four morphological types in Leptodirini: A. bathyscioid– *Bathyscia montana montana* Schiødte, 1848; B. pholeunoid– *Pholeuon (s. str.) knirschi* Breit, 1911; C. scaphoid– *Remyella scaphoides scaphoides* Jeannel, 1931; D. leptodiroid– *Leptodirus hochenwarti croaticus* Pretner, 1955 (Hlaváč et al., 2016, modified) (photos are not at the same scale).

Genera with this type of external morphology (e.g. *Anthroherpon* Reitter, 1889, *Graciliella* Njunjić, Perreau, Hendriks, Schilthuizen & Deharveng, 2016, and *Leptomeson*

Jeannel, 1924) are typical representatives of the Dinaric subterranean leiodid fauna, mostly belonging to the subtribe Anthroherponina.

It is interesting to mention that, due to troglomorphic convergence, one monotypic species (*Glacicavicola bathyscioides* Westcott, 1968) of a different leiodid subfamily (Catopocerinae) has evolved a very similar morphology. Like the Anthroherponina, the Glacicavicolini is a highly-modified troglobite, but distributed in North American lava tubes and ice caves, and clearly not closely related to the group of troglobitic leiodid beetles inhabiting the Dinaric Mountains (Seago, 2008).

1.3.2. Pseudo-physogastry

An interesting feature associated with this morphological type is pseudo-physogastry: the abdomen is greatly enlarged and appears to be swollen due to an empty space between the elytra and the thin abdominal membrane. It has been hypothesized that this adaptation has a respiratory function, increasing the air exposure of the thin abdominal membrane (Vandel, 1964; Sbordoni, 1980). On the other hand, according to Decu & Juberthie (1998, 2004), this subelytral cavity has a role in regulating the hydric equilibrium of the body, or serves as a bubble lung during flooding, or even as a flotation device (Müller, 1904; Jeannel, 1924). Unfortunately, no definitive evidence for any of these functions is available.

1.3.3. K-selection strategy

Leptodirini, like many other subterranean species, have a K-selected life history strategy: they produce fewer but much larger eggs, have reduced numbers and durations of larval instars and longer life expectancy. The life cycle of Leptodirini has undergone extreme modifications compared with that of epigean Cholevinae: the initial reduction of the number of larval instars from the ancestral 3 to 2, and finally a single instar in the more highly troglomorphic species (Deleurance, 1963; Cieslak et al., 2014). Coleoptera normally have a life cycle with three to five instars. They lay a hundreds of small eggs and their larvae feed actively, growing and moulting until they pupate

(Powell, 2009). However, in some subterranean species the number of larval instars is reduced to 2 (the "intermediate" type of life cycle): females lay a reduced number of medium-sized eggs, the larva is mobile and has a free-living phase (from 50 to 65 days in *Speonomus delarouzei*) during which it is actively feeding (Deleurance, 1963). The most modified life cycle is the one with a single larval instar ("contracted" type), which has been observed only in subterranean, highly troglomorphic species (Deleurance, 1963). Females lay only one macrolecital egg which takes a long time to develop. The large yolk provides sufficient nutrients to complete the larval development without feeding. The larva is mobile only very shortly (a few hours or days in *Speonomus longicornis*) after hatching to build a solid chamber ('logette') of sand and clay in which the rest of the larval development takes place. This short larval phase may explain why Leptodirini larvae have never been found in situ. During this period, the larva stays immobile in its clay capsule which provides protection from potential predators (Deleurance, 1963; Cieslak et al., 2014). The larva remains in a state of diapause for five to six months, and then pupates. It has been hypothesized that the reduction in the number of larval instars could be a strategy to escape predation during the most vulnerable developmental stage (Culver, 1982). Nevertheless, even though nobody has ever analyzed predation rate in caves compared with epigean habitats, we could expect that predation is reduced in the subterranean environment because of less energy input and therefore fewer trophic levels. Another explanation is that a non-feeding larval phase may allow the beetles to survive the lack of food in subterranean environments, while the long-legged, mobile adult, able to forage over a large area, is adapted to provide its offspring with enough nutrients to complete the whole development (Poulson & White, 1969). Breeding experiments with some Leptodirini (*Speonomus*) showed that their lifespan is notably long, with life expectancy being estimated between four and six years (Deleurance, 1963; Decu & Juberthie, 2004).

It was assumed that these modifications result from multiple, independent evolutionary processes within the same lineage (sometimes even in each species of the same genus) and geographic area (Cieslak et al., 2014). However, recent studies revealed a different pattern. Cieslak et al. (2014) studied the evolution of the reduction of the number of larval instars in Pyrenean Leptodirini within a phylogenetic framework. Contrary to the aforementioned assumption, their study showed a single transition from 3 to 2 larval instars, dated to Oligocene–Miocene, and a subsequent single transition to 1 instar in the Early Miocene. They concluded that this transition to a single instar was not an evolutionary dead-end for Pyrenean Leptodirini and that they were able to radiate once they have colonized subterranean environment.

1.3.4. Mouthparts of hygropetric species

Some exceptional genera of the tribe Leptodirini have, in addition to the aforementioned troglomorphic characters, strongly modified mouthparts adapted to their specific microhabitat (Fig. 3). Namely, the biotope of these beetles differs in many aspects from most other troglobitic species of Leptodirini as they inhabit a semi-aquatic cave habitat called "hygropetric" (Sket, 2004). They are always found in the vicinity of strong flows of running water on cave walls and even inside the water film itself (Jeannel, 1924; Remy, 1940; Sket, 2004; Perreau & Pavićević, 2008).

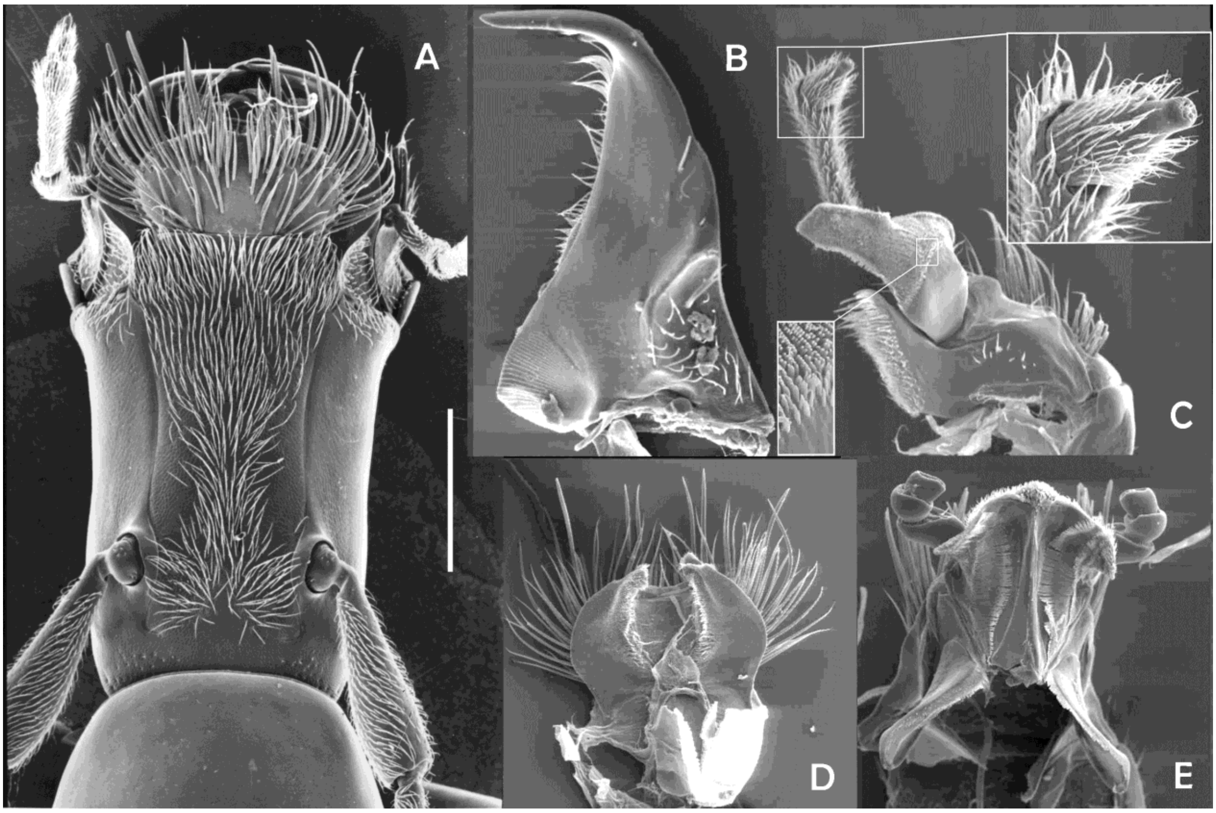

Fig. 3. Genus *Hadesia*, head (*H. asamo*) and mouthparts (*H. vasiceki*); A. head dorsal view (scale bar = 0.5 mm); B. right mandibule dorsal view; C. right maxilla and maxillary palp, ventral side, with details of the apical palpomere and sensillae of the galea; D. labrum and epipharynx ventral side; E. labium and hypopharinx dorsal side (Fig. A. in: Perreau & Pavićević, 2008, Figs. B–E in: Casale et al., 2000.).

The following Leptodirini are known to live in this type of habitat: *Hadesia* Müller, 1911; *Croatodirus* Casale, Giachino & Jalžić, 2000; *Nauticiella* Moravec & Mlejnek, 2002; *Velebitodromus* Casale, Giachino & Jalžić ,2004, and *Kircheria* Giachino & Vailati, 2006 (all from the subtribe Anthroherponina Jeannel), *Radziella* Casale & Jalžić, 1988 (subtribe *Leptodirina* Lacordaire), *Tartariella* Nonveiller & Pavićević, 1999, *Cansiliella* Paoletti, 1972 (both from the subtribe Bathysciina Horn) (Jeannel, 1924; Casale *et al.*, 2000; Moravec & Mlejnek, 2002; Moldovan et al., 2004; Perreau & Pavićević, 2008; Paoletti et al., 2011; Casale et al., 2012; Lohaj et al., 2012). All representatives of these genera have very dense hairs on labrum, labium, and maxillary palps, which enable filtering of the organic matter from water (Sket, 2004) or might serve as "brushing mouthparts" for brushing the deposits on the surface of the stalagmitic walls (Perreau & Pavićević, 2008). Furthermore, they have a very short last maxillary palpomere (the penultimate maxillary palpomere is approximately 2.5 times longer than the last one, versus less than 1.8 times in other Leptodirini). The hoe-shaped, truncated mandibles and spoon-shaped galea enable the water to flow into the mouth (Perreau & Pavićević, 2008; Lohaj et al., 2012). This unusual morphology has been illustrated for the first time by Jeannel (1924), and more recently, using detailed SEM photographs, by Casale et al. (2000), Moldovan et al. (2004), and Perreau & Pavićević (2008). The tarsal claws of hygropetric species are strongly curved, apically blunt, and wider than the usual conformation in Leptodirini (Perreau & Pavićević, 2008; Lohaj et al., 2012). It is assumed that this allows them to obtain better grip on the surface of rocks while they are moving through the water stream (as observed in *Hadesia* and *Nauticiella*) (Lohaj et al., 2012).

The diet of these species is still understudied. They are not attracted to bait in pitfall traps, nor have they been observed on deposits of guano. Recent investigations on *Cansiliella servadeii* Paoletti, 1980, suggest that these beetles are probably associated with moonmilk, a particular speleothem on which they browse on the bacterial biofilm (Paoletti et al., 2011; 2012; 2013).

1.4. SUBTRIBE ANTHROHERPONINA JEANNEL

Within Leptodirini, the subtribe Anthroherponina Jeannel, 1910 (= *Antroherpon* phyletic series of Jeannel, 1924) comprises most taxa with a leptodiroid habitus, showing the most pronounced troglomorphic characters among the Leptodirini. The Anthroherponina currently comprises nine genera: *Anthroherpon* Reitter, 1889; *Leptomeson* Jeannel, 1924; *Parantrophilon* Noesske, 1914; *Hadesia* Müller, 1911; *Kircheria* Giachino & Vailati, 2006; *Nauticiella* Moravec & Mlejnek, 2002; *Croatodirus* Casale, Giachino & Jalžić, 2000, *Velebitodromus* Casale, Giachino & Jalžić, 2004 (Perreau, 2000), and *Graciliella* Njunjić, Perreau, Hendriks, Schilthuizen & Deharveng, 2016 (see: Chapter III). They are distributed in almost the entire Dinaric range (Fig. 4.) from northwestern Croatia (*Croatodirus*, *Velebitodromus*) to northern Albania (*Kircheria*, *Anthroherpon*) (Casale et al., 2000; 2004; Giachino & Vailati, 2006; Perreau & Pavićević, 2008; Lohaj et al., 2012; Njunjić et al., 2015).

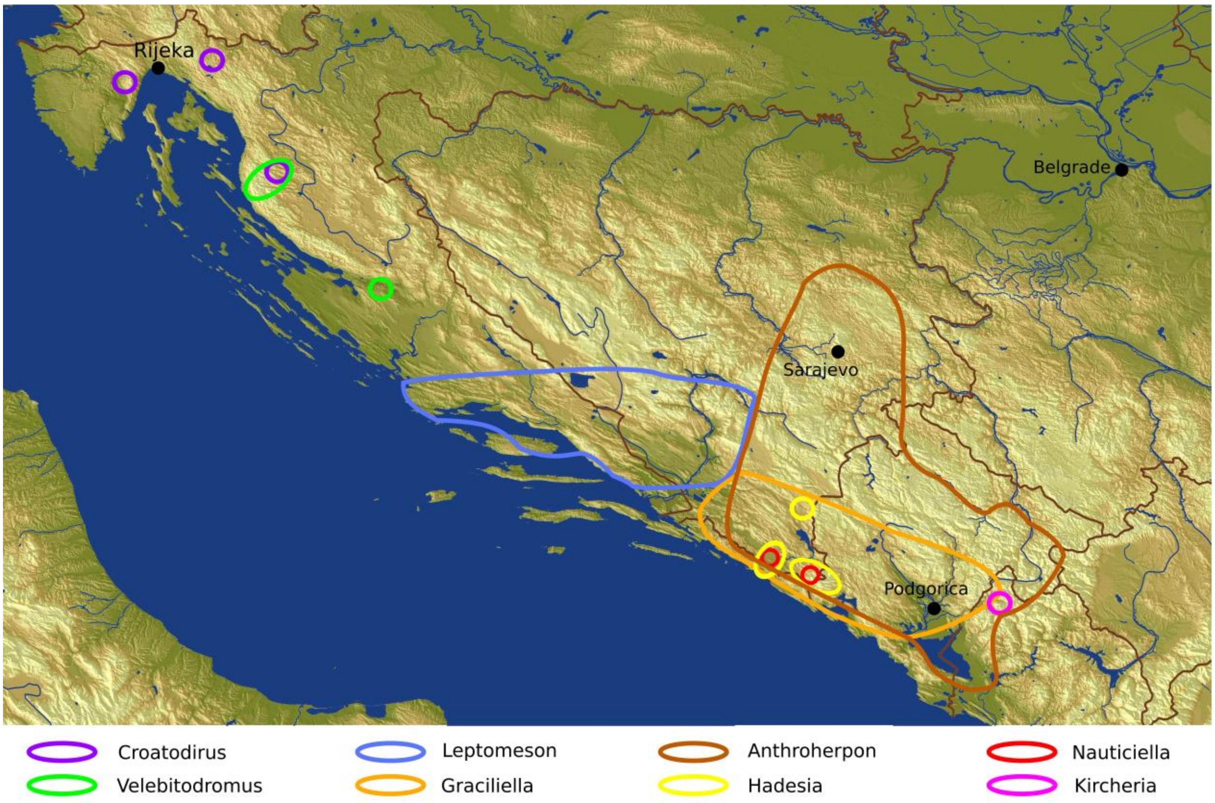

Fig. 4. Distribution of Anthroherponina.

The subtribe Anthroherponina was traditionally defined by the position of the antennal insertions on the posterior third of the head length and the enlarged and apically blunt claws (Jeannel, 1924). Jeannel used this combination of characters to characterize the tribe "Anthroherponini" composed of "phyletic line of *Anthroherpon*" (today: subtribe Anthroherponina) and "phyletic line of *Spelaeobates*" (today: subtribe Spelaeobatina). Even though several other genera of Leptodirini with similar conformation of the claws were subsequently described (*Cansiliella* Paoletti, 1972, *Razdiella* Casale & Jalžić, 1988), only genera from the subtribe Anthroherponina have this combination of claw morphology and position of antennal insertions (Perreau & Pavićević, 2008). Laneyrie (1967) and Guéorguiev (1976) strictly followed Jeannel's point of view on this subject. Cladistic analysis of Leptodirini obtained by Casale et al. (1991) disproved the previously assumed monophyly of Anthroherponina and the monophyly of Spelaeobatina which decreased the phylogenetic importance of the position of the antennal insertions. The position of antennal insertions in the posterior third of the length of the head in Anthroherponina could be linked to the extreme dilation of the abdomen correlated to the position of the insertion of the prothorax on the abdomen (Perreau & Pavićević, 2008). In Anthroherponina, the prothorax is very elongated and inserted in an upper position, which necessitates the elongation of the anterior part of the head in order to reach the food on the surface of the rocks.

Perreau & Pavićević (2008) reconstructed the phylogeny of the subtribe Anthroherponina based on 11 morphological characters. Their study suggested the monophyly of Anthroherponina. In addition to the characters previously observed by Jeannel (1924), they introduced several new features that define the subtribe Anthroherponina (Fig. 5.): (i) female urite IX reduced and without the appendicular parts, (ii) second antennomere much shorter than the first one, (iii) wide and apically blunt claws. The authors emphasized that the absence of the appendicular parts of the female urite IX is the strongest apomorphy of Anthroherponina. This character was previously noted in *Croatodirus* and *Velebitodromus* by Casale et al. (2000, 2004), in *Kircheria* by Giachino & Vailati (2006) and *Anthroherpon*, *Leptomeson*, *Paranthrophilon,* and *Hadesia* by Perreau & Pavićević (2008), but it was never described in *Nauticiella*. However, it is assumed that the same conformation of the female urite IX also occurs in this genus (Perreau & Pavićević, 2008). It should be noted that a similar absence of the appendicular parts also occurs in the Italian genus *Sinuicollia* Piva, 2008 (Bathysciotina) but this is probably due to a convergence. The second antennomere is clearly shorter in Anthroherponina compared to other Leptodirini (Fig. 5A, B) in which it is at least nearly as long as the first one (Perreau & Pavićević, 2008). However, the validity of the conformation of the claws (wide and apically blunt in Anthroherponina vs. narrow and acute in other Leptodirini, Fig. 5. C, D) as taxonomic character for Anthroherponina classification is uncertain (Perreau & Pavićević, 2008). Immature specimens of this subtribe sometimes have unusual conformation of the claws: long and acute rather than short and blunt, but still wide (Giachino & Vailati, 2006). The reason for this difference between adults and immature individuals (with thin and yellowish cuticula) is not yet fully understood but it is presumed to be a consequence of attrition.

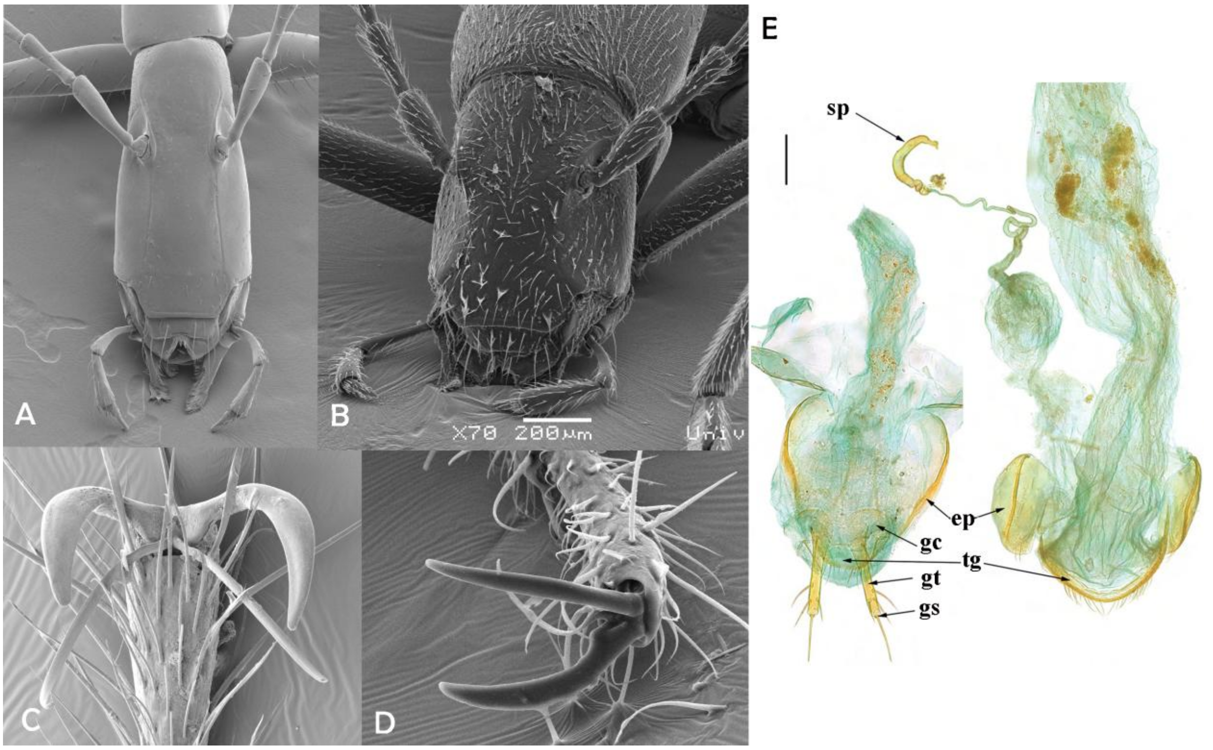

Fig. 5. Characters of the subtribe Anthroherponina vs. other Leptodirini: A. head of *Leptomeson radjai* Giachino, Bregović & Jalžić (Anthroherponina); B. head of *Rozajella deelemani* Perreau & Pavićević (Leptodirini); C. claws of *Anthroherpon cylindricolle cylindricolle* Reitter; D. claws of *R. deelemani*; E. female urite IX (genital segment), left: *Remyella* sp., right: *Hadesia asamo* Perreau & Pavićević (ep: epipleurites; gc: gonocoxite; tg: tergite; gt: gonostyles; gs: gonosubcoxite; sp: spermatheca), scale bar is 0.2 mm (Fig. E. in: Perreau & Pavićević, 2008).

Jeannel (1931) placed the genus *Remyella* in the Anthroherponini based on the insertion of the antennae on the posterior third of the head, and despite it showing a claw conformation different from other genera of this subtribe (narrow in *Remyella* and wide in Anthroherponina). The work of Perreau & Pavićević (2008) led to the exclusion of the genus *Remyella* Jeannel from the subtribe Anthroherponina and its tentative placement in Leptodirini. The study of Carvalho & Gnaspini (2015) based on the characters on the pretarsus and distal margin of the terminal tarsomere confirmed the exclusion of *Remyella* from the Anthroherponina, but this is yet to be confirmed by molecular analyses (see: Chapter V). The relations of genera inside the subtribe

Anthroherponina are far from evident, especially because many species are highly troglomorphic and many characters are potentially homoplasious. Further investigations of the phylogeny of this group are necessary to test the monophyly presumed by Perreau & Pavićević (2008) and to test the validity of morphological characters of Anthroherponina classification.

1.5. GEOLOGICAL AND GEOMORPHOLOGICAL HISTORY OF THE DINARIC KARST

Geographically and geologically, the Dinaric karst is a part of the Dinaric Mountains (=Dinarides), on the Balkan Peninsula between the Adriatic Sea and the Pannonian Basin. It is the largest continuous karst landscape in Europe, covering approximately 60,000 km^2 (Mihevc & Prelovšek, 2010). Dinaric karst covers an area 800 km long and up to 150 km wide, extending in NW – SE direction along the Adriatic Sea; from the Julian Alps on the west, across the western side of Balkan Peninsula, to the Šar-Korab massif on the southeast (Fig. 6.)(Roglić, 1965). The Dinaric chain consists of more than 200 mountains, stretching through seven countries: Italy, Slovenia, Croatia, Bosnia and Herzegovina, Montenegro, Serbia, and Albania. It was named after Dinara (1,913 m), a mountain located on the border of Croatia and Bosnia and Herzegovina. The average elevation is between 1.000 and 2.000 and the highest mountain of the Dinarides is Mount Prokletije, located on the border of eastern Montenegro and northern Albania, with the peak Maja Jezerce at an altitude of 2.694 m.

Characteristic karst relief forms, such as high karst plateaus, poljes, uvalas, dolines, and a network of caves, underground lakes and rivers are widespread in the Dinaric karst. The most prominent relief features in the Dinaric karst are karst poljes (in Slavic languages polje = field). These are elongated, flat bottom depressions, oriented in NW – SE, so called “Dinaric” direction without any superficial waterway connections. Drainage is achieved by sinkholes in the lower part of the polje and ponors (ponor = sinkhole, a place where a surface stream or lake disappears underground) while many horizontal caves are situated in the slopes bordering the poljes. Underground connections between different poljes are present (Milanović, 2006).

Climate is generally of a continental, temperate type with high precipitation (an average of about 3000 mm per year) which falls mostly in autumn and winter. Despite such a high amount of rain, there are only few surface streams because water is typically drained underground, towards the Adriatic and the Black Sea. Most rivers are forming big canyons and some of them are partly subterranean (Piva, Trebišnica–Ombla).

Based on different geological, hydrological, climatic, and geomorphological characteristics, the Dinaric karst is divided into three belts parallel to the Adriatic Sea: the Low coastal Adriatic karst, the High mountain karst, and the Low continental interior karst (Hajna, 2012). Gams (1974) differentiates only two belts: the Internal Dinarides (which correspond to the High mountain karst), and the External Dinarides (which correspond to the Low coastal Adriatic karst). The Dinaric karst is composed predominantly of limestone and dolomite, mostly of Mesozoic and Cenozoic age (Mihevc & Prelovšek, 2010). The youngest limestones (Eocene, Paleocene, and Cretaceous in age) are found along the Adiatic coast and on the islands (Mihevc & Prelovšek, 2010). Carbonate sedimentary rocks were formed on the Adriatic carbonate platform from the Upper Paleozoic to the Paleocene (Hajna, 2012). The existence of this platform is estimated to be from the Early Jurassic to the Late Cretaceous (Hajna, 2010). The final fragmentation of the platform culminated in the formation of flysch in the late Cretaceous and Paleogene and the subsequent uplift of the Dinarides (Hajna, 2010). Only after regression and denudation of the flysch could karstification commence (Deeleman-Reinhold, 1978), but these events are not yet dated (Trontelj et al., 2007). It is still uncertain when the relief rose to its present altitude, but undoubtedly, it happened in several phases, and uplifting and karstification of the Dinarides are still active today (Schmid, 2008).

During the Pleistocene glaciation, the higher parts the Dinaric Mountains were glaciated and the snow line was at about 1,100 m (Mihevc & Prelovšek, 2010). Glaciation partially blocked or interrupted karst drainage and slightly modified the karst features (Mihevc & Prelovšek, 2010). After the retreat of the glaciers, the whole Dinaric karst, except for the highest mountains and a narrow coastal area, was covered with forest. During the Neolithic period, human-caused deforestation took place, and gradually the whole coastal plateau was deforested and the landscape was transformed into a bare rocky country, a real stone desert.

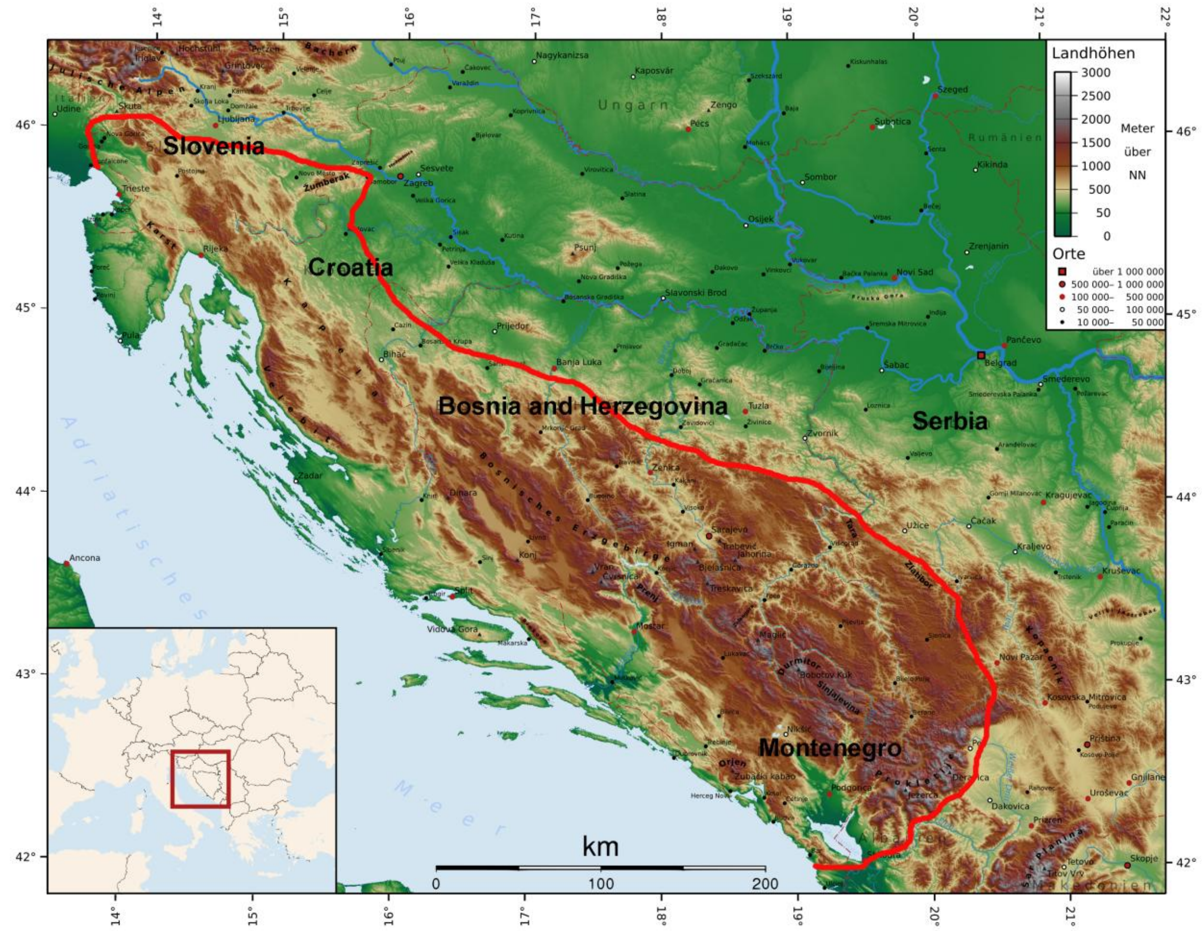

Fig. 6. Delimitation of the Dinaric karst toward the northeast after Roglić (1965).

The geological evolution of the Dinaric Mountains is very complex and there is no general, comprehensive and widely accepted theory for their formation. The geological evolution is largely determined by the history of the Tethys, a large ocean that lay between Gondwana and Laurasia during the Mesozoic and disappeared in the late Mesozoic and Cenozoic because the African and Eurasian Plates moved closer together. Today's geological structures were caused by post-collision processes in the orogenic system (which started before about 35 Ma) that created the Alps (Vrabec & Fodor, 2006). The area's main thrusting and folding events took place in post-Eocene times. The latest phase of tectonic movement in the region began with counter-clockwise rotation of the Adria microplate around 6 MYA (Hajna, 2012). This caused reactivation of already existing Dinaric faults.

The extensive karstic area of the Dinaric Mountains has provided uninterrupted conditions for subterranean life for millions of years, and hosts one of the most complex and diverse subterranean faunas in the world (Culver & Sket, 2000; Deharveng et al., 2012). The position of Dinaric mountains on the edge of formerly glaciated areas and NW – SE orientation, together with the complex geological history and intensive karstification, enabled a high invasion rate into the subterranean realm that resulted in a cave fauna which is second to none in the world (see: Introduction, Diversity of the Dinaric cave fauna).

CHAPTER II

THE GENUS *ANTHROHERPON*

2.1. INTRODUCTION

The genus *Anthroherpon* Reitter, 1889 belongs to the Leiodidae family and is the most species-rich genus of the exclusively subterranean subtribe Anthroherponina, comprising 26 species and 55 subspecies (after excluding *A. absoloni* and *A. apfelbecki ssp*. from the genus *Anthroherpon*, see: Chapter III). *Anthroherpon* ise the epitome of the Dinaric cave fauna; all species have leptodiroid body shape and exhibit the most pronounced troglomorphic morphological characters among Coleoptera (Fig. 1).

The first three species of *Anthroherpon* to be described were *A. cylindricolle, A. pygmaeum*, and *A. hoermanni*, originally published as *Leptoderus* species by Apfelbeck (1889). In the same year Reitter (1889) elevated this group of three species to genus rank and named it *Anthroherpon* (the type species of the genus is *A. cylindricolle*, as designated by Perreau, 2000). Most taxa, were described in the late 19th century and in the first half of the 20th century (Apfelbeck, 1889; 1894; 1907; Reitter, 1903; 1908; 1911; 1913; Müller, 1910; Matcha, 1916; Jeannel, 1924; 1930; 1934; 1947; Winkler, 1925; 1938; Knirsch, 1927, 1929; Zariquiey, 1927). These old descriptions were mostly based on small numbers of specimens and often without explicit diagnoses. Consequently, many species and subspecies were subsequently synonymized, but the status of many taxa remains controversial. Moreover, the taxonomy of the genus has always been difficult due to troglomorphic convergence and high intrapopulational variability. After a break of several decades due to political conditions, new collecting was performed and several new species and subspecies were described (Giachino & Guéorguiev, 1993; Giachino & Vailati, 2005).

Anthroherpon is widely distributed in subterranean habitats of the Dinaric range: from Gornja Oćevija (Bosnia and Herzegovina) on the north, to the maritime mountains of Montenegro on the south, and from Prenj Mt. (Bosnia and Herzegovina) on the west, to Prokletije Mt. (Montenegro, Albania) and Mokra planina Mt. (Montenegro, Metohija) on the east (Fig. 2). Most species are short-range endemics restricted to a single karstic unit (e.g. a single hill, mountain or a karstic plateau), and often known only from a single cave. However, the geographical distribution of some taxa shows surprising patterns and the distribution of some species (*A. latipenne, A. taxi*) is surprisingly disjunct (Fig. 3). To date, despite the revisions by Jeannel (1924, 1930) and Guéorguiev (1990), a synthesis of the information and a proper classification are emerging only very slowly and incompletely from the multitude of small taxonomic papers that have been published.

Within this chapter, I clarify the status of the species and subspecies of the genus *Anthroherpon* including their barcoding characterization and I analyze the inter- and intra-populational morphological variability to identify the most relevant characters for taxonomy.

Fig. 1. *A. cylindricolle cylindricolle* ex situ (photo: Dragiša Savić).

2.2. GENERAL MORPHOLOGY

As mentioned above, the genus *Anthroherpon* comprises taxa of exclusively leptodiroid body shape, with the most troglomorphic phenotype noted among Coleoptera (together with the Trechinae *Giraffaphaenops* Deuve, 2002): complete anophthalmy, extremely elongated appendages, head and pronotum, and physogastric elytra. Total body length ranges from 3.8 mm (*A. weiratheri*) to 8.4 mm (*A. latipenne latipenne*). Coloration is variable, from light yellowish to dark brown, depending on the age of the animal (juvenile specimens are normally yellowish or light brown).

Head (Figs. 1, 4A): elongated and hypognathous; wider in the anterior part and wider than the pronotum; without occipital carina. Clypeus trapezoid, wider in the posterior part. Maxillae long, both galea and lacinia elongated and with numerous setae. Maxillary palps with 4 articles; first one extremely small, second maxillary palpomere arched; terminal maxillary palpomere conical and with a dorso-lateral comb of very short bristles (Fig. 4A). Labium with trapezoid submentum. Mandibles angulate in dorsal view, with sharp apex and with several small teeth between the apex and the last big tooth. Antennal insertions located in the posterior third of the length of the head (measured from the posterior edge of the clypeus). Antennae as long as (in some females) or longer than the whole body; second antennomere is the shortest.

Pronotum longer than wide and always narrower in the posterior part; shape variable; with or without a constriction in the posterior part; maximal width in the anterior part or in the middle.

Mesothoracic pedunculus variable: absent, very short, or long*;* when present always wider than long, narrowest in the anterior part or at mid-section.

Elytra elliptic, physogastric, wide or very narrow; with pointed or widely rounded apex; punctuation absent (if so, then elytra are granulated) or very variable—from superficial and indistinct punctures to deep, wide, and clear punctures; elytral surface covered with short, medium-long or long hairs; hairs can be sparse or very dense, covering the entire elytral surface, or just the dorsal part while lateral sides are hairless.

Abdomen. All species except some individuals of *A. stenocephalum* have a mesoventral processus on the transversal carina on the posterior margin of mesoventrum (for details, see: Taxonomic overview–*Anthroherpon stenocephalum*); mesocoxal cavities confluent; metacoxal cavities separated by an intercoxal apophysis. Mesosternum without a ridge in all species except *A. weiratheri* (Fig. 15C, D). Metendosternite “Y”-shape.

Legs. Long, slender, and pubescent. Femora enlarged in the basal parts, in some species profemora thick along the full length; protibiae straight or slightly curved inwards. Male protarsi with five protarsomeres of equal width, female protarsi with four protarsomeres, mesotarsi and metatarsi with five tarsomeres in both sexes. Claws wide and apically blunt, tarsal empodium with two long setae which are fused at the base and positioned laterally to each other (Fig. 4B).

Aedeagus ventrally strongly or only slightly curved; endophallus without sclerotized structures, but some species with a thickened sclerotized strip along the length of the medioventral side of the median lobe. Jeannel (1924:403) mentions this structure as being unique among all Leptodirini. Parameres (setae excluded) can be as long as, shorter, or longer than the median lobe. Each paramere bears three apical setae which can be of equal length or with one seta shorther than the other two. Urite IX reduced to a ring, as in most Leptodirini.

Female genitalia. Abdominal ventrite VIII with an anterior expansion. Appendicular parts (gonocoxites and gonosubcoxites, cf. Deuve, 2001 for terminology) of the urite IX absent. Spermatheca weakly sclerified.

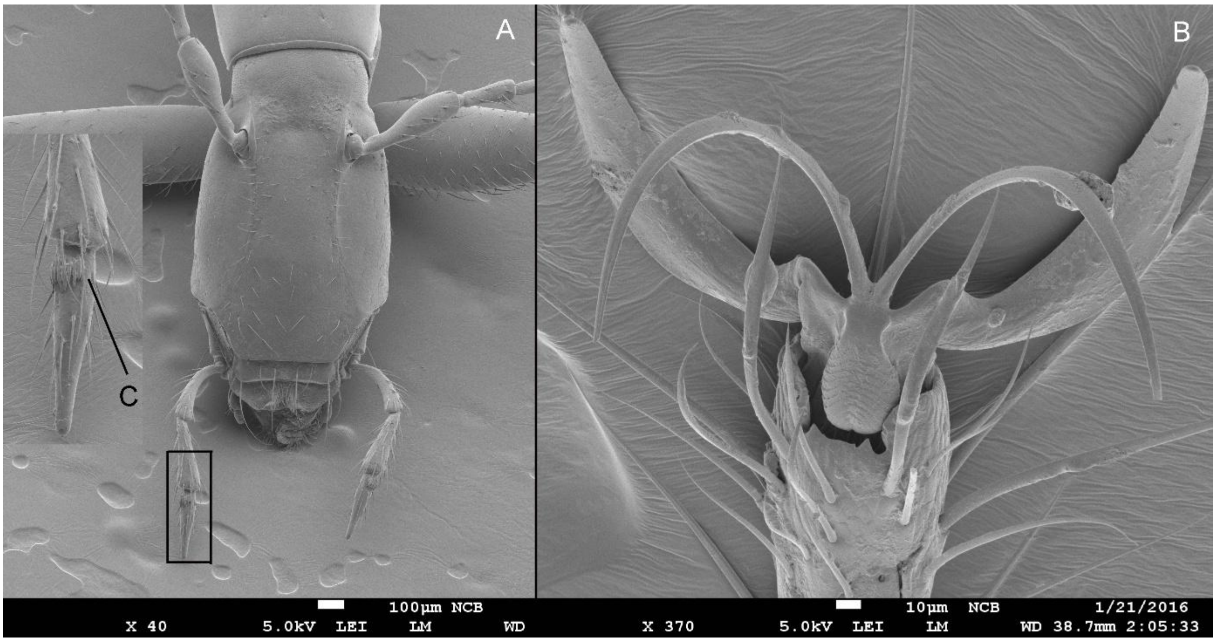

Fig. 4. *A. cylindricolle cylindricolle*; A. Head and dorso-lateral comb of bristles on the terminal palpomere (C); B. Claws and pretarsus of the second pair of legs (ventral view).

2.2.1. Sexual dimorphism

The most obvious difference between males and females is the number of protarsomeres: females have protarsi with four tarsomeres while males have five protarsomeres. In many species there is also a difference between males and females in the shape of elytra, with females having wider elytra, but sometimes also a slightly wider pronotum and often a larger body size. Moreover, in some species, females have shorter antennae relative to the body length in comparison with the males. For instance, the antennae of males of *A. sinjajevina* are 1.2 times longer than the body, while in females, antennae are only 1.05 longer than the body. Furthermore, all species of the "*ganglbaueri*" species group have the last antennomere longer than the penultimate, but this difference is more pronounced in males: the last antennomere in males of *A. sinjajevina* is approximately 2 times longer than the penultimate while in females it is 1.7 times longer.

Due to sexual dimorphism, it is very difficult to identify specimens if only females are availale. For a reliable identification and the description of new taxa it is essential to examine specimens of both sexes.

2.2.2. Pseudo-physogastry

The inflated abdomen and elytra of *Anthroherpon* is shared with most other genera with a "leptodiroid" body shape, and is termed "pseudo-physogastry" (Jeannel, 1924). True physogastry refers to an inflation of the abdomen due to large numbers of ovarioles and eggs while it is assumed that pseudo-physogastry has a different role. In *Anthroherpon*, indeed, the abdomen appears not to be swollen for the benefit of reproductive tissue, but rather to store fat tissue. It has been hypothesized that this adaptation has a respiratory function, increasing the air exposure of the thin abdominal membrane (Vandel, 1964; Sbordoni 1980). On the other hand, according to Decu & Juberthie (1998, 2004), this subelytral cavity has a role in regulating the hydric equilibrium of the body, or serves as a bubble lung during flooding, or even as a flotation device (Müller, 1904) but no definitive evidence for any of these functions is available.

Upon dissection of *Anthroherpon*, I find, under the membraneous tergites, a thick layer of adipose tissue, or even loose droplets of oil, presumably the result of adipocytes ruptured due to the preservation in ethanol. As explained in the next section (see: 3.3.2. *Anthroherpon* behavior regarding water), *Anthroherpon* specimens do float on the water surface, but are unable to swim and very quickly drown. Therefore, it seems very unlikely that pseudophysogastry serves as a buble lung, or a flotation device. To me, more plausible explanation is that pseudophysogastry is an adaptation to irregular feeding in cave environment and has a role in storing adipose tissue.

2.3. GNERAL ECOLOGY, BEHAVIOUR, AND BIOGEOGRAPHY

Virtually nothing is known beyond the taxonomy of *Anthroherpon*, and the fact that they are all cave-restricted. Though I did not carried out an experimental approach, I report here various observations that I made that bring the first data on on the biology, ecology and behaviour of *Anthroherpon*.

Specimens of the genus *Anthroherpon* are always found exclusively in caves, never in MSS (though very little sampling in MSS has been performed in the Balkans). They are normally found on humid cave walls, stalactites, and stalagmites, and they have never been observed on deposits of guano. They are rarely found under the rocks on the cave floor, but they can sometimes be found under stones forming a scree. They inhabit the dark zone of the caves. However, in some rare cases, specimens were found on cave walls close to the entrance (e.g. *A. hoermanni hoermanni* in Borija pećina, and *A. ganglbaueri ganglbaueri* in Novakuša), but always in such places where there is no light. In the presence of humans and light *Anthroherpon* specimens begin to move fast (it is uncertain whether this is due to the light, the radiated heat, or the exhaled CO2), but otherwise they are slowly climbing the cave walls or not moving at all. Males and females were found in about equal numbers in all populations that I have sampled. Temperature in caves where *Anthroherpon* was found ranges from 0°C to about 9°C, with the relative humidity above 87%. Among all caves inhabited by *Anthroherpon*, the caves on Durmitor Mt. and Prokletije Mt. (MNE) are located at the highest elevation—from about 1800 m to more than 2000 m, while Banja Stjena cave in Prača canyon (BiH) is probably the one situated at the lowest elevation—639 m.

The diet of *Anthroherpon*, like that of many other cave beetles, is understudied but it is assumed that they are saprophagous like many other Cholevinae (Beutel & Leschen, 2016). They are attracted by pitfall traps with decaying meat and vinegar. The individuals that I am rearing (see below) are fed with cheese and chicken.

2.3.1. Rearing of *Anthroherpon*

In June 2013, I took the opportunity to test if it is possible to bring *Anthroherpon* specimens alive out of the cave to another place where they could be kept in laboratory conditions. During the visit of Kečina Stena cave (Bosnia and Herzegovina) I placed two specimens of *A. harbichi* in a 50 ml vial together with some mud from the cave. Since it was a hot summer day, with the temperature above 27°C, I was afraid that the beetles might die before reaching Sarajevo where they could be placed in a refrigerator. Suprisingly, however, both specimens survived the transportation from the cave to the city of Sarajevo which in total lasted around 2 hours. I then transferred them to a small plastic box together with a bit of mud I took from the cave and placed them in a refrigerator at 5°C. The beetles lived only for three days. I presume they did not survive longer because the quantity of mud in the box was too small to retain enough moisture, so that the beetles died as soon as the mud dried up (I did not add any water). Although unsuccessful, this "experiment" showed that *Anthroherpon* can survive sudden temperature changes and being exposed to a high temperature for several hours. However, they seem to be very sensitive to the lack of humidity.

The second attempt to keep *Anthroherpon* alive in laboratory conditions was more successful. In March 2014, I collected three live specimens of *A. taxi remyi* in Županska pećina (Montenegro). I transported them for 7 hours by car from the cave to my home in Belgrade in a plastic box that was kept in a portable refrigerator. In Belgrade, I transfered them to a large jar with several holes in the lid. On the bottom of the jar I placed some mud and a stone taken from the cave. The beetles were fer with cheese and water was added to the jar whenever the mud appeared to be drying out. Two beetles died after 4 weeks when the mud in the jar dried out on one occasion. The last individual stayed alive for 3 more weeks but died soon after it became infected with a fungus (see below: Parasites of *Anthroherpon*).

In June 2014, I collected around 30 specimens of *A. cylindricolle cylindricolle* in Golubovića pećina (Bosnia and Herzegovina) together with some stones and mud from the cave and again transported them to my home in Belgrade. For transportation, I used a plastic box with several tiny holes in the lid that was placed in a portable refrigerator. The trip lasted about 7 hours. Immediately upon arrival in Belgrade, I separated the specimens: more than a half of them were placed in a terrarium made of glass (25 cm long, 18 cm wide, and 20 cm deep) and the remaining were placed in a plastic box (20 cm long, 13 cm wide, and 10 cm deep). Both terrariums with *Anthroherpon* were kept in a refrigerator at a temperature of +5°C. Once per week I added about 50 ml of water to each terrarium: using a pipette the water was dripped the water on to the stones and mud, taking care not to disturb the beetles. I fed the beetles irregularly, no more than once per month, with old cheese and boiled or fried chicken meat. When taking the terrariums out of the refrigerator I noticed that the beetles were mostly hiding between the rocks or between the rock and the mud. However, they would start to move after being exposed to room temperature and light for several minutes. They were unable to walk on vertical glass and plastic surfaces and had difficulties to walk on smooth horizontal surfaces. On the other hand, they could very quickly climb the vertical rocks of which the rough surface ensures good grip to their claws. Occasionally, I had to remove some fungus that would start growing on the rocks inside the terrariums. (My previous attempt to rear two specimens of *Anthroherpon taxi remyi* failed because the beetles became infected with a gray fungus which started growing on their elytra, legs, and antennae; see above: Fig. 6)

After 2 years in the refrigerator, almost all specimens from the glass terrarium died while 4 specimens of those kept in the plastic terrarium survived. I suppose that the reason for this unequal survival in laboratory conditions is due to the different distribution of humidity in the two terrariums: the plastic terrarium is more shallow, so the moisture was concentrated more closely to the bottom while the glass terrarium is more than two times deeper so the bottom of the terrarium would dry up more quickly, with water condensing on the inside of the lid. In July 2016, more than two years after I started rearing *Anthroherpon*, I noticed what appears to be an egg in the plastic terrarium (Fig. 5). The egg is about 1.5 mm in diameter and located on the horizontal surface of the rock. It did not hatch until the present day (more than two months after it was laid) and I did not notice any change in its size or colour. One month after I noticed the first egg, the second egg was laid hanging on the vertical surface of the rock.

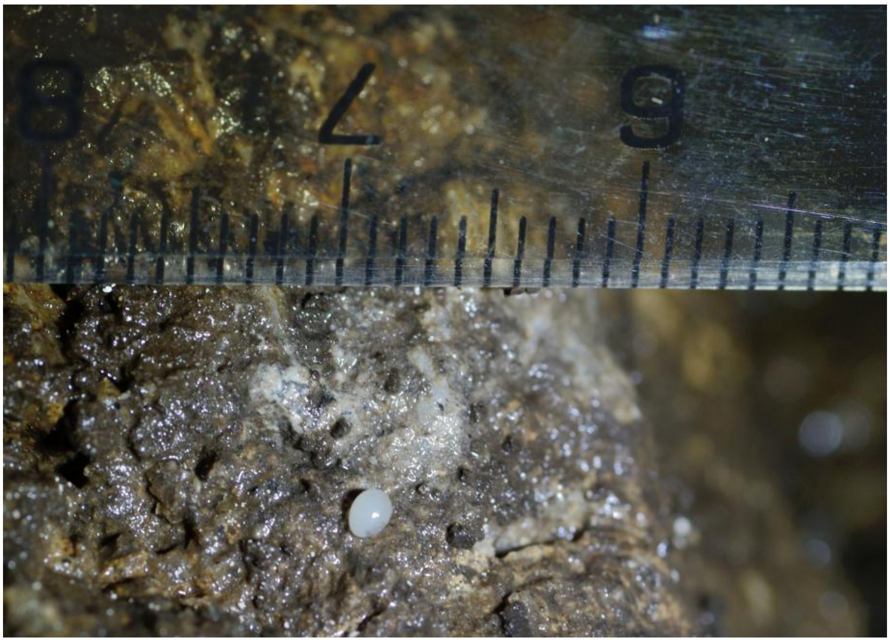

Fig. 5. The egg of *A. cylindricolle cylindricolle* ex situ.

2.3.2. *Anthroherpon* behavior regarding water

While rearing *A. cylindricolle cylindricolle*, I have noticed that their long and thin legs easily get stuck in water films and that they had difficulties to move after that happens. This is surprising since *Anthroherpon* is mostly found on humid cave walls, where the risk of being exposed to running or dripping water is high. Contrary to hygropetric species (e.g. *Hadesia*, *Croatodirus, Nauticiella,* etc.), which inhabit semi-aquatic habitat, *Anthroherpon* has never been found inside the water films and are unable to walk inside the film of running water.

After being dropped down or gently placed on a still water surface, individuals of *A. cylindricolle cylindricolle* are behaving in the following way:

1. They usually fall on the lateral or dorsal side of their body and immediately start to move their legs energetically, like they are trying to escape.

2. Sometimes they move from the dorsal to the lateral side of their body, or from the lateral to the ventral side; their head and pronotum are placed inside the water while a part of their abdomen is floating on the water surface.

3. They are struggling on the water surface for no more than 5 min and remain immobile afterwards.

4. After that, upon being removed from the water they do not move. Hovewer, if they are being removed from the water while they are still alive and quickly dried out

on a tissue, they can recover completely. Nevertheless, this can never happen in their natural environment which is very humid.

Based on these observations, and observations mentioned in the section 3.3.3. Rearing of *Anthroherpon*, I can conclude the following:

1. *Anthroherpon* can survive sudden temperature changes.
2. They are very sensitive to decrease of humidity in their habitats.
3. They can be kept in laboratory conditions with minimal care at 5°C.
4. *Anthroherpon* have difficulties to move after their legs get in touch with water; they are unable to swim and they cannot survive on the surface of water for more than 5 min. Therefore, water can be a barrier to their dispersal.
5. It is unlikely that peudophysogastric elytra of *Anthroherpon* serve as a buble lung, or a flotation device. More plausible explanation is that this subelytral cavity has a role in storing the adipose tissue. However, its potential role in regulating the hydric equilibrium of the body cannot be excluded.
6. They can live at least two years at the adult stage.

2.3.3. Parasites of *Anthroherpon*

Little is known about the parasites of *Anthroherpon*. Fungi of the order Laboulbeniales were recorded as parasites of of this genus (Rossi, 2006). Laboulbeniales are obligate parasites on insects, mites, and millipedes. They display a high degree of host specificity, and if they also regularly infect cave beetles, it is likely that many cave beetle species will carry undescribed, host-specific species of this group of Fungi. One new species of Laboulbeniales, *Diphymyces spelaei* (Ascomycota, Laboulbeniales), was found on head, mesosternum, and legs of *Anthroherpon taxi trezzii* and *Anthroherpon latipenne latellai* and recently described (Rossi, 2006). The constant temperature and high humidity in caves are favorable conditions for Laboulbeniales, so these Fungi are expected to thrive well in these habitats. For a successful infection by Laboulbeniales, besides these abiotic conditions, it is also important that the arthropod hosts are long-lived and have a soft cuticle (Scheloske, 1969; Balazuc, 1988; Weir & Hammond, 1997).

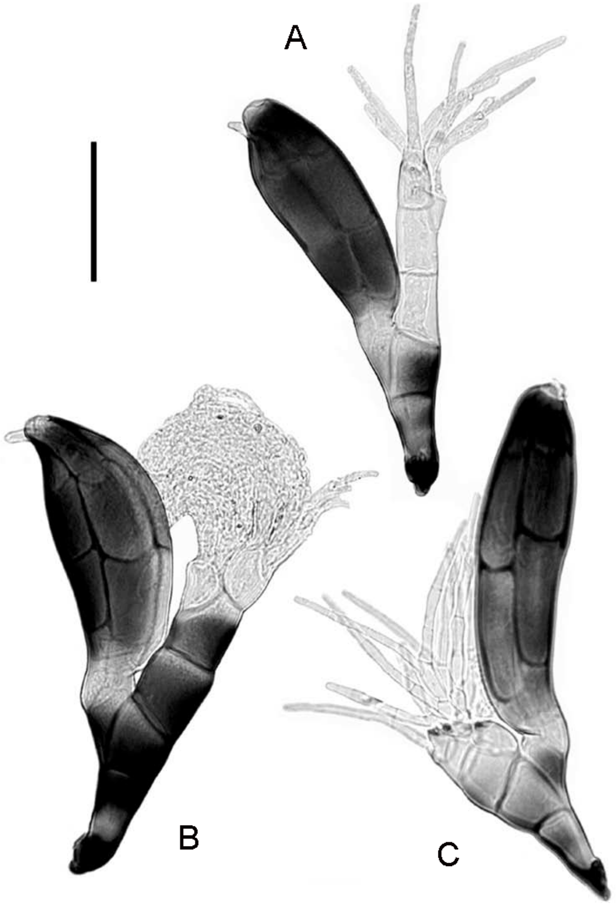

Fig. 6. *Diphymyces spelaei*: A. Thallus from mesosternum of the host; B. Thallus from head; C. Thallus from hind femur. Scale bar = 50 µm (Rossi, 2006, modified).

Another group of organisms sometimes associated with *Anthroherpon* are mites (Arachnida, Acari). They are found externally, superficially attached to the body of these beetles, usually no more than ten individuals on one beetle. I noticed many mites, mostly on the ventral side of the body, on individuals of *A. taxi albanicum* from several sinkholes on Prokletije Mt. (Fig. 7). They were identified as immature stages (deutonymphs) of *Histiostoma* sp. (Astigmata, Histiostomatidae) by Dr. Wayne Knee, a specialist for mite systematics from Canadian National Collection of Insects, Arachnids and Nematodes. The Astigmata is a diverse and widely distributed group of mites and the most successful group of mites in establishing symbiotic relationships with both vertebrates and invertebrates (Houck & O'Connor, 1991). The family Histiostomatidae is unique among Acari due to their mode of feeding: they mostly forage by filtering a bacterial film formed at the surface of fluid covering rotten organic matter (Hughes, 1976; Wirth, 2004; O'Connor, 2009). Their developmental stages similar to other astigmatic mites: egg, larva, protonymph, deutonymph, tritonymph and adult. All specimens that were so far found on *Anthroherpon* were deutonmyphs. The deutonymphal stage actively disperses but does not feed, i.e. it is adapted to phoresy. Phoresy is defined as a form of commensalism facilitating the physical transport of one organism on the body of another, during which no feeding or reproduction occurs in the phoretic organism (Binns, 1982; O'Connor, 1982). Jeannel (1911:549) also mentions the presence of immature mites on *Anthroherpon* on ventral side of profemora and prosternum. The fact that only immatures have been found so far, may suggest that the adults are either free-living or have a different host.

Fig. 7. An immature stage of *Histiostoma* sp. (Arachnida: Astigmata: Histiostomatidae) found on *Anthroherpon taxi albanicum*.

One individual of *A. taxi remy* that I was rearing in a refrigerator in my home (see above) became infected with a fungus after 4 weeks. The fungus grew on the legs, antennae, and elytra of the beetle, which became gray due to the presence of the fungus on their surface (Fig. 8). This fungus probably caused the death of the animal within 3 weeks.

Fig. 8. *A. taxi remyi* infected with a fungus (ex situ) (photo: Dragiša Savić).

2.3.4. Biogeography

Anthroherpon is distributed in subterranean habitats of the Dinaric Mountains, covering a latitudinal range of more than 200 km and longitudinal range of more than 170 km (Fig. 2). It is present in all three belts of the Dinaric karst, determined on different geological, hydrological, climatic, and geomorphological characteristics (Hajna, 2012). If we disregard the highly doubtful distribution of *A. brckoensis* (see below: Article 1), the northernmost finding of *Anthroherpon* is in Banja pećina near Gornja Oćevija (Bosnia and Herzegovina), the type locality of *A. stenocephalum noesskei*. This area roughly overlaps with the aproximative border of the Dinaric karst (Roglić, 1965; Gams, 1974). To the south, the range of the genus spreads throughout the mountains of the Maritime belt of the Dinarides. The discovery of *A. scutariensis* in an unnamed cave on the Albanian side of Scutari lake (=Skadarsko jezero) is the southernmost finding of

Anthroherpon. The western border of *Anthroherpon* distribution is a line extending from Bjelašnica Mt. (*A. pygmaeum* ssp.), over Prenj Mt. (*A. ganglbaueri alticola*), to the eastern side of Velež Mt. (*A. ganglbaueri ganglbaueri*) to Grebci (*A. primitivum primitivum*) (Bosnia and Herzegovina). To the east, *Anthroherpon* is distributed to Mokra Mt. near Drelje on Kosovo and Metohija and to the southeast to Prokletije Mt. on the border of Montenegro and Albania.

Regarding the three main geomorphological units of the Dinaric karst (three belts parallel to the Adriatic Sea: Adriatic karst, the High mountain karst, and the Low continental interior karst) (Hajna, 2012), the highest number of species of the genus *Anthroherpon* is in the High mountain karst, namely 50% of all species. In the Adriatic karst, 30.7 % of the species occur, and 26.9 % of the species is present in the Low continental interior karst. Two species—*A. taxi* and *A. latipenne*—are present in both, the High mountain karst, and the Low continental interior karst, while others are limited to a single belt. Throughout the whole distribution area of *Anthroherpon*, many mountains host more than one species (e.g. Jahorina, Trebević, Baba, Bjelasnica, Moračke planine, Prokletije, Orjen, Lovćen, etc.). There are several caves in which two species occur in syntopy, but only in Ericova cave on Orjen Mt., the maximal number of three syntopic species of *Anthroherpon* (*A. taxi taxi, A. matulici,* and *A. latipenne latipenne*) has been recorded (Eric Quéinnec, pers. comm. cited in: Kurtović et al., 2008).

Many taxa are short-range endemics: from 26 species and 55 subspecies of *Anthroherpon*, 16 species and 24 subspecies are known only from a single cave. On the other hand, two species which morphologicaly closely resemble each other (*A. taxi* senso lato and *A. latipenne* senso lato), have very wide distribution areas: *A. latipenne* (with 4 subspecies) ranges throughout an area of 60 x 117 km, and *A. taxi* (with 11 subspecies) covers an area of approximately 60 x 150 km (Fig. 3). However, the distribution of both species is mysteriously disjunct. They are known from several mountain ranges which are tens of kilometres far from each other, with the most distant ones being more than a hundred kilometers apart. The karst is continuous between most of these areas so it is possible, that gaps in the distribution represent insufficiently explored areas. What is particularly interesting is that both species show a very similar distribution pattern. They both occur on the same mountain ranges: Baba Mt., Golija Mt., Trubjela, Orjen Mt., Moračke planine Mt., and Prokletije Mt. The only places where their distribution area does not overlap are Lovćen Mt. and Kučka Korita (each mountain range hosts one subspecies of *A. latipenne*), and Mokra Mt., Visitor Mt., and Lubnice, where three subspecies of *A. taxi* occur (one in each area). In numerous caves on Baba Mt. (e.g. Jama u Vukovijeh koliba, Zubovića pećina, Vučja Bara, Ledenica jama, Pećina u stare kolibe) and Bjelasnica Mt. (e.g. Pećina u Klačinama, Pećina u Vali pod sedlom, Agova pećina, Snježnica u Tisovom kršu, Nova jama u Tisovom kršu) *A. latipenne latipenne* and *A. taxi hercegovinum* were found in syntopy. In Ledena pećina na V. Kiti on Trubjela, *A. latipenne latipenne* lives in syntopy with *A. taxi pretneri* and also in several caves on Orjen Mt. (Pećina u Kučericama, Ericova jama, Izeta pećina) with *A. taxi taxi*.

Before we excluded *Graciliella* from the genus *Anthroherpon* (see: Chapter III), it was assumed that a group of species, previously considered as *Anthroherpon absoloni* and subspecies of *Anthroherpon apfelbecki*, also had a very wide distribution area. Our study, based on morphological and molecular approaches, revealed that *Graciliella* comprises six species and four subspecies distributed from Crvanj Mt. (Bosnia and Herzegovina) in the north, to Trnovo (Montenegro) in the south, and from Žaba Mt. (Croatia) in the west to Prokletije Mt. (Montenegro) in the east. In comparison to *Anthroherpon*, *Graciliella* occurs usually at much lower altitude: from 50 m to 1270 m above sea level, while the lowest altitude where *Anthroherpon* was found is 639 m and the highest around 2000 m. Moreover, the distribution of these two genera differs in relation to the three main geomorphological units of the Dinaric karst. The genus *Graciliella* is present only in the Low coastal Adriatic karst and the High mountain karst while *Anthroherpon* occurs in much larger area, and in all three belts.

2.4. HISTORY OF THE STUDY OF *ANTHROHERPON*

2.4.1. Pre-World War I period

The genus *Anthroherpon* was established in 1889, after Reitter elevated to genus rank three species described by Viktor Apfelbeck: *Leptoderus cylindricollis*, *L. pygmaeus*, and *L. hoermanni*. Edmund Reitter (1845-1920) was one of the most influential European coleopterologists in the 19th century (Nonveiller, 1999), working on Palearctic beetles. He was actively collecting beetles during his numerous field trips throughout the Balkans and described an impressive number of taxa from different families of

Coleoptera including many subterranean beetles from the Balkan Peninsula. Reitter described 4 species and one subspecies of the genus *Anthroherpon.* As biospeleological investigations on Balkan Peninsula continued, numerous new species and subspecies of this genus were subsequently described by various other authors.

The most productive contributor to the knowledge of *Anthroherpon* diversity was Viktor Apfelbeck (1859-1934). He was curator of the Entomological Collection of the National Museum of Bosnia and Herzegovina in Sarajevo and one of the most important Balkan entomologists at that time. During his career, Apfelbeck explored numerous caves in the entire Balkan region and described a significant number of new taxa. In 1889 he described 3 species of cave beetles from Bosnia and Herzegovina: *Leptoderus cylindricollis, L. hoermanni* and *L. pygmaeus*. These species were the first cave beetles to be described from Bosnia and Herzegovina. They were subsequently placed in the genus *Anthroherpon* when this genus was established by Reitter. Throughout his career, Apfelbeck described 8 species and 3 subspecies of the genus *Anthroherpon*.

2.4.2. Interbellum

Between the two world wars, three renowned entomologists, Josef (Giuseppe) Müller (1880 – 1964), Karel Absolon (1877 – 1960), and René Jeannel (1879 – 1965) were actively working on the Dinaric cave fauna. These scientists were, without any doubt, among the most important contributors to our knowledge of the subterranean Coleoptera of the Dinaric Mountains and the Balkans. They described several species and subspecies of the genus *Anthroherpon*; Müller: 3 species, Absolon: 2 species, and Jeannel: 2 species and 6 subspecies. Jeannel was one of the most influential coleopterologists of the 20th century and a father of modern biospeleology. He worked intensively on the genus *Anthroherpon* and was the first to subdivide the genus into species groups: first into three (1911) groups named “Sectiones”, then into five (1924) Sectiones, and finally (1930) he created two subgroups in his “Sectio III” and “Sectio IV”:

i) Sectio I: *A. cylindricolle*
ii) Sectio II: *A. harbichi*
iii) Sectio III: 1st subgroup: *A. matulici, A. echinatum* (now *A. matulici*), *A. augustae* (now *A. matzenaueri augustae*), *A. augustae havelkai* (now *A. matzenaueri matzenaueri*), *A. augustae taliensis* (now *A. matzenaueri taliensis*), *A. ganglbaueri ganglbaueri, A.*

ganglbaueri distinguendum, A. ganglbaueri alticola; 2nd subgroup: *A. hossei, A. primitivum, A. piesbergeni, A. matzenaueri matzenaueri, A. matzenaueri Luciani* (now *A. latipenne latipenne*), *A. matzenaueri punctipennis* (now *A. latipenne punctipennis*), *A. matzenaueri Göttli* (now *A. latipenne goettli*), *A. albanicum albanicum* (now *A. taxi albanicum*), *A. albanicum Mülleri* (now *A. taxi muelleri*), *A. albanicum Boschi* (now *A. taxi boschi*), *A. albanicum Winkleri* (now *A. taxi winkleri*), *A. albanicum Sydowi* (now *A. taxi sydowi*), and *A. taxi.*

iv) Sectio IV: 1st subgroup: *A. stenocephalum, A. weiratheri;* 2nd subgroup: *A. erebus, A. winneguthi, A. charon, A. subalpinum, A. pygmaeum, A. pozi,* and *A. zariquieyi*.

v) Sectio V: *A. hörmanni* (now *A. hoermanni*), *A. apfelbecki apfelbecki* (now genus *Graciliella*), *A. apfelbecki sculptifrons* (now *G. apfelbecki sculptifrons*), and *A. lahneri* (now *G. lahneri*).

Jeannel (1924) described the genus *Leptomeson* Jeannel, 1924 as a subgenus of *Anthroherpon* in order to separate three species with a different conformation of the aedeagus (with sclerotized structures in the endophallus). *Leptomeson* was subsequently elevated to genus level by Guéorguiev (1990), which proved to be in the accordance with molecular phylogenetics (see: Chapter III). In 1927, Richard Zariquiey (1897–1965), doctor and collector from Barcelona, described 1 new species and 8 new subspecies of the genus *Anthroherpon* from Montenegro, which significantly expanded our knowledge about the diversity and the distribution of this genus. Other entomologists who were describing new taxa of the genus *Anthroherpon* in this period were Matcha, Knirsch, and Breit.

Many species described in the 20th century by different authors (Jeannel, Knirsch, Reitter, Winkler, etc.) were collected by a single person: Leo Weirather (1887–1965), a commercial insect collector and one of the greatest biospeleologists working on the Balkans. He was employed as a postman in the Austro-Hungarian army in Trebinje (Bosnia and Herzegovina) from where he moved to Innsbruck (Austria) after the First World War but continued his commercial activity as insect collector. Only in Dalmatia, he explored around 500 caves and based on his collecting, 5 new genera and 79 lower taxa of Coleoptera from this region were described (Pretner, 1974). Weirather tried to protect his discoveries from competition by disguising the collecting sites under false names ("Decknamen") and code numbers, although the locality information at the larger scale was always correct (Pretner, 1974). For decades, this controversial approach created confusion regarding the type localities. However, Weirather kept notes during his fieldwork in shorthand German. These were later deciphered by Egon Pretner (1896

– 1982), a famous Slovenian biospeleologist and a friend of Weirather who eventually sold the notebooks to the National Science Museum of Geneva (Pipan, 1997). The notes were translated into English and published in Giachino & Lana (2005b), and Hauser (2011).

Fig. 9. Contributors to the knowledge of *Anthroherpon.* A. Viktor Apfelbeck, B. René Jeannel.

2.4.3. Post-World War II

During the Second World War and several decades after, there were no cave explorations on the Balkans: after Winkler's description of *A. ganglbaueri intermedium* in 1938, there was a break for more than a half of a century until any new species of the genus were described again (Giachino & Guéorguiev, 1993; Giachino & Vailati, 2005). However, in 1967, Laneyrie published a paper on a new classification of Bathysciinae (today tribe Leptodirini), in which he listed all species of the genus *Anthroherpon* that were known at that time. He strictly followed Jeannel's division into 5 species groups. The first synthetic revision of the genus came more than 20 years later: in the 1990 the paper "Recherches sur les Bathysciinae (Coleoptera: Catopidae) de Yougoslavie" by Vassil Borislav Guéorguiev (1935-1996), a well-known Bulgarian biospeleologist. In this work, he proposed a new identification key and divided the genus *Anthroherpon* into seven species groups:

i) Groupe “cylindricolle”: *A. cylindricolle*
ii) Groupe ''harbichi'': *A. harbichi*
iii) Groupe ''ganglbaueri'': *A. matulici, A. matzenaueri, A. ganglbaueri*
iv) Groupe “latipenne”: *A. hossei, A. primitivum, A. piesbergeni, A. latipenne, A taxi*
v) Groupe ''stenocephalum'': *A. stenocephalum*, *A. weiratheri*
vi) Groupe ''pygmaeum'': *A erebus, A. winneguthi, A. charon, A. subalpinum, A. pygmaeum, A. pozi, A. zariquieyi*
vii) Groupe “hoermanni”: *A. hoermanni, A. absoloni* (now *G. absoloni*), *A. apfelbecki* (now *G. apfelbecki*).

A few years after this work was published, coleopterologists Pier Mauro Giachino and V. B. Guéorguiev jointly described 4 new species and 1 new subspecies of the genus *Anthroherpon* from Montenegro and Bosnia and Herzegovina (1993). Giachino continued to work on the genus and together with Dante Vailati and they together described 2 species and 5 subspecies from Montenegro and Albania (Giachino & Vailati, 2005). These newly described species were integrated in the 7 species groups introduced by Guéorguiev. After the description of two new species from northern Montenegro (Njunjić et al., 2015), and the exclusion of *A. apfelbecki* and *A. absoloni* from the genus *Anthroherpon* (see: Chapter III), the intrageneric species groups underwent significant changes in numbers of taxa:

i) Groupe “*cylindricolle*”: *A. cylindricolle cylindricolle, A. cylindricolle thoracicum, A. cylindricolle scaphium*
ii) Groupe “*harbichi*”: *A. harbichi*
iii) Groupe “*ganglbaueri*”: *A. matulici, A. matzenaueri matzenaueri, A. matzenaueri augustae, A. matzenaueri taliensis, A. ganglbaueri ganglbaueri, A. ganglbaueri alticola, A. ganglbaueri distinguendum, A. ganglbaueri intermedium, A. sinjajevina, A. udrzali, A. brckoensis*
iv) Groupe “*latipenne*”: *A. elongatum*, *A. garbellii*, *A. gueorguievi*, *A. hossei, A. latipenne latipenne, A. latipenne attenuatum, A. latipenne goettli, A. latipenne latelai, A. latipenne punctipennis, A. piesbergeni, A. primitivum primituvum, A. primituvum jeanneli, A taxi taxi, A. taxi albanicum, A. taxi boschi, A. taxi hercegovinum, A. taxi lemur, A. taxi muelleri, A. taxi pretneri, A. taxi remyi, A. taxi sydowi, A. taxi terezzii, A. taxi winkleri*
v) Groupe “*stenocephalum*”: *A. stenocephalum stenocephalum, A. stenocephalum noesskei*, *A. weiratheri*
vi) Groupe “*pygmaeum*”: *A. charon, A erebus erebus, A. erebus scheibeli, A. pozi, A. pygmaeum pygmaeum, A. pygmaeum stricticolle, A. subalpinum, A. winneguthi, A. zariquieyi*

vii) Groupe “*hoermanni*”: *A. hoermanni hoermanni, A. hoermanni hoffmanni, A. hoermanni hypsophilum, A. hoermanni orlovacensis, A. hoermanni sericeum, A. scutariensis*

However, since our molecular phylogenetic analysis showed that some of these species groups are artificial (see: Chapter IV), a revision of the genus is required to rearrange the species groups and create a classification that corresponds better to the phylogeny.

2.5. MATERIALS AND METHODS

2.5.1. Taxon sampling and morphological study

Specimens were collected in caves of the Dinaric range, in Montenegro and Bosnia and Herzegovina, and stored in 70% ethanol for morphological studies and in 96% ethanol for molecular analyses. Material for morphological studies was collected by hand or using pitfall traps with decaying meat as bait. The external morphology of specimens was examined using Leica MZ 75 and Leica M10 stereomicroscopes. Microphotographs were taken on a Leica DIAPLAN or a Zeiss AXIOLAB microscope with a camera diagnostic instrument Spot INSIGHT IN1820. Macrophotographs were taken using a Canon Eos 70D camera with Canon EF MP-E 65mm f/2.8 macro lens and Canon macro twin lite mt-24ex flash. Digital pictures were processed using the Helicon Focus software 5.3 (http://www.heliconsoft.com/heliconsoft-products/helicon-focus/). Male genitalia were cleared in clove oil, mounted in Euparal on glass slides, and pinned beneath the specimens. Scanning electron microscopy was performed at Naturalis Biodiversity Center (Leiden) on a Hitachi SU3500 after gold coating with a Jeol JFC 1200 sputter coater.

2.5.2. DNA extraction, PCR amplification, and sequencing

DNA was extracted from whole specimens or from one leg with a standard phenol–chloroform extraction (Blin & Stafford, 1976) or the DNeasy Tissue Kit (Qiagen GmbH, Hilden, Germany). Voucher specimens are stored in the CINJ (Iva Njunjić collection, Belgrade, Serbia) and DNA aliquots are kept in the tissue collections of Naturalis (Leiden, Netherlands). The barcoding region of COI was amplified by polymerase chain reaction (PCR) using the standard invertebrate primer pair LCO1490– 5’GGTCAACAAATCATAAAGATATTGG3’ and HCO2198– 5’TAAACTTCAGGGTGACCAAAAAATCA3’ (Folmer et al., 1994). Each 25 µl PCR mixture included 1µl (10 pmol) of each primer, 2.5 µl 10x PCR buffer, 0.5 µl dNTPs, 0.25 µl Taq-polymerase, 18.8 µl ddH2O and 5 µl template DNA. PCR cycles were run at the following conditions: 3 min at 94 °C, followed by 40 cycles of 15 s at 94 °C, 30 s at 54 °C and 40 s at 72 °C, and finally, 5 min at 72 °C. Sequences were assembled and edited using Geneious version 8.0.5 (http://www.geneious.com/, Kearse *et al.*, 2012) and subsequently deposited in GenBank. Sequences were aligned using MAFFT version 7 (Katoh & Standley, 2013).

2.5.3. Distribution maps

The distribution maps were built using SRTM3 data (Shuttle Radar Topography Mission) of NASA with the software QGIS. GDAM (global administrative area: www.gdam.org) data were used for country frontiers and Natural Earth data (http://www.naturalearthdata.com/) were used for hydrographic features: rivers and lakes. Location of species were inserted with Photoshop CS2.

2.5.4. Acronyms

BiH: Bosnia and Herzegovina; MNE: Montenegro; Lct: locus typicus; nfw: nomen fictum Weirather.

2.6. TAXONOMIC OVERVIEW

For the polytypic species I provide a diagnosis only for the nominal subspecies. For the remaining subspecies, see the key of Gueorguiev (1990). Barcode characterization of most species is also provided.

Genus *Anthroherpon* Reitter

Anthroherpon Reitter, 1889: 294, type species: *Leptoderus cylindricollis* Apfelbeck, 1889.
Antroherpon: Müller, 1904: 40. Synonym: *Eumecosoma*
Antroherpon Reitter: Müller, 1914: 1029. Synonym: *Antrophilon*
Antroherpon Reitt.: Jeannel, 1914: 54. Synonym: *Euantroherpon, Protantroherpon*

The following key to *Anthroherpon* species groups is a modified version of the key given by Guéorguiev (1990). It is dedicated to the practical identification of species and does not correspond to the phylogeny of the group.

1 (8) Dorsal surface of pronotum (in lateral view) regularly convex in full length, without an annular constriction in the posterior part.
2 (3) Lateral sides of pronotum (in dorsal view) without a constriction in the posterior part, gradually narrowing posteriorly………………………………………… "*cylindricolle*" group I
3 (2) Lateral sides of pronotum (in dorsal view) with a constriction in the posterior part.
3 (5) Profemora thick in the posterior part. Body size < 5 mm…………"*harbichi*" group II
5 (4) Profemora thin in the posterior part. Body size > 5 mm.
6 (7) Last antennomere longer than the penultimate…………………"*ganglbaueri*" group III
7 (6) Last antennomere shorter than or as long as the penultimate
…………………………………………………………………………………………………"*latipenne*" group IV
8 (1) Dorsal surface of pronotum (in lateral view) with a depression in the posterior part.
9 (12) Body size 3.8–5.0 mm. Without or with short mesothoracic pedunculus.
10 (11) Protibiae 1.5 to 1.6 times longer than protarsi. Protibiae slender, straight or slightly curved outwards in the dorsal view. Profemora thin in the posterior part, tapering towards the apical part ……………………………………… "*stenocephalum*" group V
11 (10) Protibiae 1.8 to 2 times longer than protarsi. Protibiae thick and curved inwards in dorsal view. Profemora very thick in the posterior part and slightly tapered in the apical part ……………………………………………………………………………… "*pygmaeum*" group VI

12 (9) Body size > 5.5 mm. Mesothoracic pedunculus long, about 3 times wider than long (width measured at the narrowest part); pronotum about 2 times longer than wide (width measured at the widest part) .... “*hoermanni*” group VII

2.6.1. I “*cylindricolle*” group V. B. Guéorguiev, 1990

Anthroherpon cylindricolle **(Apfelbeck, 1889)**

Anthroherpon cylindricolle cylindricolle **(Apfelbeck, 1889)**

= *Leptoderus cylindricollis* Apfelbeck, 1889: 61. Lct: “Pećina kod Golubovća”

Type locality: Bosnia and Herzegovina, Rogatica, village Golubovići, Golubovića pećina, 823 m, N 43°47'1.62" E 18°54'46.80".

Distribution: known only from the type locality. Locality indicated as „Glasinac, Lakatoš – Čaplina“ (Guéorguiev, 1990) is probably a mistake.

Diagnosis: Habitus illustrated in Fig.10A. Body length from 5.5 mm to 6 mm. This species can be easily distinguished from other species of the genus by the shape of the pronotum, which is cylindrical, without the constriction in the posterior part, lateral sides gradually narrowing posteriorly. Mesothoracic pedunculus long, with a constriction in the middle. Elytra with long, erected and sparse hairs. Aedeagus illustrated in Fig. 11A: parameres slightly longer than the median lobe (setae excluded), each with a pointed, triangular apex, bearing three setae; median lobe thicker at the base and with lanceolate apex in dorsal view, without the thickened sclerotized strip along the length of the medioventral side of the median lobe.

DNA-barcode: IE3 (Rogatica, village Golubovići, Golubovića pećina; type locality):

```
1        10        20        30        40        50
|        |         |         |         |         |
GTACTTTATACTTCTTATTAGGAGCTTGATCTGGTATAATTGGAACCTCA
CTAAGATTACTAATTCGAGCTGAATTAGGAAATCCAGGAAGATTAATCAT
AAATGATCAAATTTATAACGTTATTGTAACAGCCCATGCTTTTATTATAA
TTTTTTTTATAGTCATACCAATTATAATTGGAGGGTTTGGAAACTGATTA
GTTCCCTTAATATTAGGAGCTCCTGATATAGCATTCCCTCGAATAAATAA
CATAAGATTTTGATTATTGCCTCCTTCTTTAACTCTATTGCTTATAAGAA
GTATAGTAGAAAGAGGAGCTGGAACAGGATGAACTGTATATCCACCTCTA
TCAGCTAATATTGCCCATAGAGGACCATCAGTAGATTTAGCAATTTTTAG
CCTTCATTTAGCAGGAATCTCTTCTATTTTAGGAGCAGTAAATTTTATTA
CTACTATCATTAATATACGATCTCCTGGAATATCTTTTGATAAAATACCA
TTATTTGTATGATCTGTAGCTATTACTGCATTATTATTATTACTTTCATT
ACCAGTATTGGCTGGTGCAATTACTATACTATTAACAGATCGAAATCTAA
ATACCTCTTTCTTTGACCCAGCAGGTGGAGGGGACCCAATTTTATATCAA
CATTTATTT
```

Anthroherpon cylindricolle scaphium Reitter, 1908

Anthroherpon cylindricolle ssp. scaphium Reitter, 1908: 20. Lct: “Grotte von Banja stjena“

Type locality: Bosnia and Herzegovina, Prača, kanjon Prače, Banja Stjena (=Mračna pećina= Govednica), 639 m, N 43°46'20.3" E 18°53'13.6".

Distribution: It is known only from the type locality and Pogorelica pećina (Jeannel, 1930). Three different names for a single cave (type locality) have often caused confusion regarding the distribution of this subspecies.

DNA-barcode: VA14 (Prača, kanjon Prače, Banja Stijena; type locality):

```
1        10        20        30        40        50
|        |         |         |         |         |
GTACTTTATACTTCTTATTAGGAGCTTGATCTGGTATAATTGGGACCTCA
CTAAGATTACTAATTCGAGCTGAATTAGGAAATCCAGGGAGATTAATCAT
AAATGATCAAATTTATAACGTTATTGTAACAGCCCATGCTTTTATTATAA
TTTTTTTTATAGTCATACCAATTATAATTGGAGGATTTGGAAACTGATTG
GTTCCCTTAATATTAGGAGCCCCTGACATAGCATTCCCTCGAATAAATAA
CATAAGATTTTGATTATTGCCTCCTTCTTTAACTTTATTGCTTATAAGAA
GTATAGTAGAAAGAGGAGCTGGAACAGGATGAACTGTATACCCACCTCTA
TCAGCTAATATTGCCCATAGAGGACCATCAGTAGATTTAGCAATTTTTAG
TCTTCATTTAGCAGGAATCTCTTCTATTTTAGGGGCAGTAAATTTTATTA
CTACTATCATTAATATACGATCTCCTGGAATATCTTTTGATAAAATACCA
TTATTTGTATGATCTGTAGCTATTACTGCATTATTATTATTACTCTCATT
ACCAGTATTGGCTGGTGCAATTACTATACTATTAACAGATCGAAATCTAA
ATACCTCTTTCTTTGATCCAGCAGGTGGAGGGGACCCAATTTTATATCAA
CATTTATTT
```

Anthroherpon cylindricolle thoracicum (Apfelbeck, 1907)

Antroherpon cylindricolle thoracicum Apfelbeck, 1907: 402. Lct: “Hercegovina: in antro montis Romanja prope Pale“

Type locality: Bosnia and Herzegovina, Pale, Romanija Mt., Careve vode, Novakova pećina, 1424 m, N 43°50'39.54", E 18°38'19.14".

Distribution: Caves on Romanija Mt. in Bosnia and Herzegovina: Careve vode, Novakova pećina; Lupoglav, Zečeva ledenica, 1508 m, N 43°51'36.84" E 18°40'41.58" (new locality).

DNA-barcode: VA9 (BiH, Pale, Romanija Mt., Careve vode, Novakova pećina; type locality):

```
1        10        20        30        40        50
|        |         |         |         |         |
GTACTTTATACTTCTTATTAGGAGCTTGATCTGGTATAATTGGGACCTCA
CTAAGATTACTAATTCGAGCTGAATTAGGAAATCCAGGAAGATTAATCAT
AAACGATCAAATTTATAACGTTATTGTAACAGCCCATGCTTTTATTATAA
```

```
TTTTTTTTATAGTCATACCAATTATAATTGGAGGATTTGGAAACTGATTG
GTTCCCTTAATATTAGGAGCCCCTGATATAGCATTCCCTCGAATAAATAA
CATAAGATTTTGATTATTGCCTCCTTCTTTAACTTTATTGCTTATAAGAA
GTATAGTAGAAAGAGGGGCTGGAACAGGATGAACTGTATATCCACCTCTA
TCAGCTAATATTGCCCATAGAGGACCATCAGTAGATTTAGCAATTTTTAG
CCTTCATTTAGCAGGAATCTCTTCTATTTTAGGAGCAGTAAATTTTATTA
CTACTATCATTAATATGCGATCTCCTGGAATATCTTTTGATAAAATACCA
TTATTTGTGTGATCTGTAGCTATTACTGCATTATTATTATTACTTTCATT
ACCAGTATTGGCTGGCGCAATTACTATACTATTAACAGATCGAAATCTAA
ATACCTCTTTCTTTGATCCAGCAGGTGGAGGAGACCCAATTTTATATCAA
CATTTATTT
```

Remarks: Genetic distances in the barcoding region between different subspecies of *A. cylindricolle* are almost the same and range from 1.7% to 1.8%. This is surprising regarding the geographical location of caves inhabited by these taxa: Golubovića pećina (type locality of A*. cylindricolle cylindricolle*) and Novakova pećina (type locality of *A. cylidricolle thoracicum*) are 23 km from each other, while Golubovića pećina and Banja Stijena (type locality of *A. cylindricolle scaphium*) are only 2.5 km apart. Subtle differences in the shape of pronotum and shape and punctuation of elytra do exist between these taxa, but the differences in the shape of the aedeagus between them are negligible and are similar to those we normally find at the intrapopulational level.

Table 1. Genetic distances in barcoding region between *A. cylindricolle* subspecies*.*

	A. cylindricolle cylindricolle	*A. cylindricolle scaphium*	*A. cylindricolle thoracicum*
A. cylindricolle cylindricolle		1.7 %	1.8 %
A. cylindricolle scaphium	1.7 %		1.7 %
A. cylindricolle thoracicum	1.8 %	1.7 %	

2.6.2. II "*harbichi*" group V. B. Guéorguiev, 1990

2. *Anthroherpon harbichi* (Reitter, 1913)

Antroherpon Harbichi Reitter, 1913: 153. Lct: "Höhle der Kječina Stjena"

Type locality: Bosnia and Herzegovina, Sarajevo, Crepoljsko polje, Kečina Stena (=Pećina u Kečinoj Steni), 1445 m, N 43°55'53.16" E 18°29'0.72".

Distribution: BiH, Pale, village Sumbulovac, Orlovača, 950 m, N 43°52'59.46" E 18°35'3.53" (new locality); BiH, Pale, Kadino selo, Pećina izvora Mokranjske Miljacke, 1023 m, N 43°55'30.12" E 18°35'44.64" (new locality).

Diagnosis: Habitus illustrated in Fig. 10B. Body length 4 – 4.3 mm. Dorsal surface of pronotum (in lateral view) regularly convex in full length, without a depression in the posterior part. Lateral sides of the pronotum (in dorsal view) weakly rounded in the apical part and gradually narrowing posteriorly; lateral sides are almost parallel in the posterior third of the length of the pronotum. Without mesothoracic pedunculus. Elytra elongated and narrow, but wider in the posterior part, with long and sparse hairs and superficial and sparse punctuation. Profemora thick in the posterior part; protibiae slightly curved outwards in dorsal view. Aedeagus illustrated in Fig. 11B: median lobe very elongated, of uniform width nearly in the full length, and much longer than parameres, with rounded apex, and without the thickened sclerotized strip along the length of the medioventral side of the median lobe; parameres thin, almost straight in dorsal view and of uniform width along their entire length, bearing three long setae in the apical part.

DNA-barcode: VA1 (BiH, Sarajevo, Crepoljsko polje, Pećina u Kečinoj steni; type locality):

```
1        10        20        30        40        50
|        |         |         |         |         |
GTACTCTATACTTTCTCCTAGGAACTTGATCAGGAATAATTGGAACCTCA
TTAAGATTATTAATTCGAGCTGAATTAGGGACTCCGGGAAGATTAATCAT
AAATGATCAAATTTATAATGTTATTGTAACAGCTCATGCTTTTATTATAA
TTTTTTTTATGGTCATACCAATCATAATTGGAGGATTTGGAAATTGATTA
GTCCCTTTAATGCTAGGGGCCCCTGATATAGCTTTCCCACGTATAAATAA
TATAAGATTTTGATTATTACCCCCTTCCTTAACTTTATTACTTATAAGAA
GAATGGTAGAAAGAGGGGCTGGGACAGGATGAACAGTTTATCCGCCTTTA
TCAGCTAATATTGCTCACAGCGGCCCGTCAGTTGACTTAGCAATCTTTAG
TCTTCATTTAGCTGGAATCTCTTCAATTCTAGGGGCAGTAAATTTTATTA
CTACTATTATTAATATACGGTCTCCAGGTATATCTTTTGATAAAATACCA
TTATTTGTATGATCTGTTGCTATTACTGCTTTATTACTACTACTCTCACT
ACCAGTATTAGCAGGTGCAATTACTATATTATTAACAGATCGAAATTTAA
ATACTTCTTTTTTTGATCCTGCAGGAGGTGGGGATCCAATTTTATATCAA
CATTTATTT
```

Remarks: Genetic distances between three different populations of this species range from 0.6 % to 2.4 %: the populations from Kečina Stena and Orlovača differ by 0.6%, the populations from Kečina Stena and Pećina izvora Mokranjske Miljacke by 2.4%, and the populations from Orlovača and Pećina izvora Mokranjske Miljacke by 2.1%.

2.6.3. III “*ganglbaueri*” group V. B. Guéorguiev, 1990

For more details regarding this species group, see below: Article 1: Two new species of the genus *Anthroherpon* Reitter, 1889 from northern Montenegro with notes on the “*A. ganglbaueri*” species group (Coleoptera: Leiodidae: Leptodirini).
Aedeagi illustrated in Fig. 12.

3. *Anthroherpon brckoensis* Giachino & V. B. Géorguiev, 1993

Antroherpon brckoensis Giachino & V. B. Guéorguiev, 1993: 293. Lct: “Bosnia, Brčko”

Type locality: The exact type locality is unknown. On the original label, the locality is given as “’Bosnia, Brčko”.

Distribution: known only from the type locality.

Remarks: We suggested a synonymy of this species with *A. ganglbaueri alticola* (Njunjić et al., 2015) (for details, see below: Article 1).

4. *Anthroherpon cecai* Njunjić, Perreau & Pavićević, 2015

Anthroherpon cecai Njunjić, Perreau & Pavićević, 2015: 403. Lct: “Montenegro, Durmitor Mt., village Mala Crna Gora, Poda, Crna jama (=Jama u Podu), 1900 m“

Type locality: Montenegro, Durmitor Mt., village Mala Crna Gora, Poda, Crna jama (=Jama u Podu), 1900 m.

Distribution: known only from the type locality.

Diagnosis: Habitus illustrated in (see below: Article 1). Distinct from the other species of the group by the following characters: second antennomere approximately two times shorter than the first one. Lateral sides of the pronotum parallel on the basal fifth of the pronotum length. Pronotum with two symmetric dorsal impressions on the anterior half of the pronotum length (with variable extension and depth, absent in some specimens). Elytral surface microreticulated between the punctuation. Parameres as long as the median lobe (setae excluded), with preapical constriction and with rounded apex (Fig. 12F).

5. *Anthroherpon ganglbaueri* (Apfelbeck, 1894)

Anthroherpon ganglbaueri alticola **(Knirsch, 1927)**

Antroherpon alticola Knirsch, 1927: 45. Lct: “Grotten der Prenj planina”

Type locality: Caves on Prenj Mt. on southern Bosnia and Herzegovina. The exact locality is unknown.

Distribution: Snežnica and Vučica pećina near Rećica; Cave in Crno Polje (Jeannel, 1930); Cave on Sirova gora; Cave on mountain Lupoglav; Vučica jama, Rečica, Prenj (Guéorguiev, 1990).

Anthroherpon ganglbaueri distinguendum **(Müller, 1913)**

Antroherpon ganglbaueri distinguendum Müller, 1913: 160. Lct: “Ulog Obrnja”

Type locality: Insurgenten jama (= Morinje pećina) in the vicinity of the village Obrnja, near Ulog in southern Bosnia and Herzegovina (Guéorguiev, 1990).

Distribution: Caves on Crvanj Mt., near the village Obrnja (Jeannel, 1930); Deverdela pećina on Crvanj Mt. (Perreau, 2000).

Anthroherpon ganglbaueri ganglbaueri **(Apfelbeck, 1894)**

Antroherpon Ganglbaueri Apfelbeck, 1894: 513. Lct: ”Novakuša, Nevesinje“

Type locality: Bosnia and Herzegovina, Nevesinje, Velež Mt., village Bišina, Novakuša, 1021 m, N 43°14'57.18" E 18° 4'35.52".

Distribution: known only from the type locality.

DNA-barcode: VA15 (BiH, Nevesinje, Velež Mt., Bišina, Novakuša; type locality):

```
  1        10        20        30        40        50
|        |         |         |         |         |
GTACTTTATATTTTTTAATGGGAACTTGATCAGGAATAATTGGAACCTCA
TTAAGGTTATTGATTCGATCCGAATTAGGAAATCCAGGAAGATTAATTAT
AAATGATCAAATTTATAATGTTATTGTAACAGCTCATGCTTTTATTATAA
TTTTTTTTATAGTTATACCAATTATAATTGGAGGATTTGGAAATTGATTG
GTTCCCTTAATATTAGGGGCCCCTGATATAGCTTTCCCACGTATAAATAA
TATAAGATTCTGACTATTGCCCCCTTCTTTAACTTTATTATTAATAAGAA
GAATCGTAGAAAGAGGGGCCGGAACAGGATGAACTGTTTATCCTCCCCTA
TCTGCTAATATTGCCCATAGCGGCCCATCAGTTGATCTCGCTATTTTTAG
CCTCCATTTAGCTGGAATCTCTTCTATCTTAGGAGCAGTAAATTTTATTA
CTACTATCATCAACATACGATCCCCTGGCATATCTTTTGATAAAATACCT
CTATTTGTATGATCTGTAGCTATTACTGCCTTACTATTATTACTCTCATT
ACCAGTATTAGCTGGGGCAATTACTATATTATTAACTGACCGTAATTTAA
ACACTTCTTTTTTTGACCCTGCTGGAGGAGGAGATCCAATTTTATACCAG
CATTTATTT
```

Diagnosis: Habitus illustrated in Fig. 10C. Body length from 5.4 mm to 5.8 mm. Distinct from the other species of the group by the following combination of characters: first antennomere approximately 1.5 times longer than the second one; the largest width of the pronotum is at the anterior quarter of its length, the lateral sides parallel on the basal third of its length; elytra with short hairs, punctuation small and dense. Aedeagus illustrated in Fig. 12D. Median lobe much longer than parameres and thick at the base, with pointed apex in dorsal view; parameres regularly arcuate near the apex, without the preapical constriction.

Anthroherpon ganglbaueri intermedium **(Winkler, 1938)**

Antroherpon Ganglbaueri ssp. *intermedium* Winkler, 1938: 219. Lct: ”Ost-Herzegowina, Siljevi kom“

Type locality: Bosnia and Herzegovina, Avtovac, Baba mountain, Vučja bara, Pećina Šiljevi Kom.

Distribution: Baba Mt. in BiH: Jama Sniježnica Čančarica, Baba mounatain (Guéorguiev, 1990).

Remarks: In the identification key of Guéorguiev (1990), *A. ganglbaueri ganglbaueri* and *A. ganglbaueri alticola* on one hand, and *A. ganglbaueri intermedium* and *A. ganglbaueri distinguendum* on the other, are separated by the difference in elytra punctuation (more vs. less pronounced). Due to intrapopulation variability of this character, it is extremely difficult to separate subspecies on this ground. However, the differences in the shape of pronotum between *A. ganglbaueri ganglbaueri* and *A. ganglbaueri distinguendum* are clearly noticeable, as previously indicated by Jeannel (1924, 1930). In addition, the differences in aedeagus shape between these subspecies can help to distinguish them (Fig. 12 C, D).

6. *Anthroherpon matulici* (Reitter, 1903)

Antroherpon Matuliči Reitter, 1903: 216. Lct: ”Bukova rupa (Büchehöhle) bei Ubli“
= *Anthroherpon* (s.str.) *echinatum* Jeannel, 1924: 1924 : 416. synonymy in Guéorguiev, 1990: 249.
Lct: ”Herzégovine: Rajčeva jama, sur le Troglav à la frontière du Monténégro“

Type locality: Bosnia and Herzegovina, Orjen, Ubla, Bukova rupa, 1259 m, N 42°36'34.20" E 18°30'11.22".

Distribution: This species is distributed in the Mediterranean range: in southern Montenegro, Bosnia and Herzegovina, and Croatia. It is known from several caves on Orjen Mt. in Montenegro: Izeta pećina in the village Knezlaz; Gubar jama; Bezimena jama in Koprivno Do; Krivošije, Vilina pećina (Giachino & Guéorguiev, 1993); Crkvice, Ericova jama, 1030 m, 42°33'52.56" E 18°38'32.04" (Eric Quéinnec, pers. comm. cited in: Kurtović et al., 2008), and several other localities: Rajčeva jama, Troglav; Pit on the mountain Gumbar, Montenegro (Guéorguiev, 1990); Southeastern Croatia, Sniježnica plateau above Konavle polje, Glogova jama (Lohaj & Jalžić, 2009); Rumija Mt. in Montenegro (in: Lohaj & Jalžić, 2009); Krivošije, Buzina, Sinja jama, 1019 m, N 42°31’45.7'' E 18°44’24.7'' (new locality).

Diagnosis: Habitus illustrated in Fig. 10D. Body length from 6.7 mm to 7.1 mm. Distinct from other species of the group by the extremely long and erect hairs on the elytra. Elytral surface with profound and sparse punctuation. Aedeagus illustrated in Fig. 12G: median lobe as long as parameres (setae excluded), with drawn-out apex. Parameres slightly concave at 2/3 of their length but somewhat curved in the apical part, bearing three apical setae.

Partial DNA-barcode: I5 (MNE, Orjen, Ubla, Bukova rupa; type locality):

```
1               5                   10                  15
|               |                   |                   |
C   G   G   C   A   A   T   T   A   T   T   T   C   G   T
G   C   C   G   C   T   A   C   T   A   A   T   A   G   G
C   G   C   A   C   C   G   G   A   T   A   T   G   G   C
T   T   T   T   C   C   A   C   G   T   C   T   T   A   A
C   A   A   T   A   T   C   A   G   C   T   T   C   T   G
G   C   T   A   T   T   A   G   T   C   C   C   T   G   C
T   T   T   T   C   T   A   T   T   A   C   T   T   A   T
G   T   T   G   T   C   T   G   C   T   T   T   C   G   T
T   G   A   C   G   G   C   G   G   T   C   C   C   G   G
C   A   C   G   G   G   C   T   G   G   A   C   T   C   T
A   T   A   C   C   C   C   C   C   T   T   T   A   A   G
T   A   G   C   C   T   T   A   C   G   G   G   G   C   A
T   C   C   G   G   G   A   G   C   A   G   C   A   G   T
T   G   A   T   A   T   G   G   C   T   A   T   T   T   T
C   A   G   C   T   T   G   C   A   T   T   T   A   A   C
G   G   G   G   C   T   T   T   C   C   T   C   G   A   T
T   C   T   C   G   G   C   T   C   G   A   T   T   A   A
C   T   T   A   A   T   T   G   T   T   A   C   T   A   T
A   T   T   T   A   A   T   A   T   G   C   G   C   G   C
C   C   C   G   G   G   C   A   T   G   G   G
```

Remarks: This species has a disjunct distribution: it is distributed in caves on Orjen Mt. and in the surrounding area, but it was also found on Rumija Mt. in southeastern Montenegro, approximately 70 km from Orjen Mt.

7. *Anthroherpon matzenaueri* (Apfelbeck, 1907)

Anthroherpon matzenaueri augustae (Zariquiey, 1927)

Antroherpon Augustae Zariquiey, 1927: 160. Lct: "Höhle 20, Gola P., Vojnik Gau"
= *Antroherpon Augustae* subsp. *Bocki* Zariquiey, 1927: 161. synonymy in Jeannel, 1930: 139
Lct: "Höhle 21, Rapta J., Vojnik Gau"
= *Antroherpon peduncularium* Knirsch, 1928: 114. synonymy in Jeannel, 1929: 297
Lct: "Gola planina. Vojnik. Montenegro"
= *Antroherpon peduncularium* subsp. *discrepans* Knirsch, 1928: 121
Lct: "Rapta J. Vojnik-Gau (Montenegro)"

Type locality: Montenegro, Vojnik Mt., Bezimena pećina u Prosijanom Brijegu, 1410 m.

Distribution: It is distributed on Vojnik and Maganik Mt. in Montenegro (Zariquiey, 1927; Knirsch, 1928; Jeannel, 1930; Guéorguiev, 1990): Pećina u Prosijanom Brijegu (=nfw. Gola pećina), Praga, Vojnik; Jamica pri Han Gvozdu (=nfw. Rapta jama); Vojnik, Ridezina jama (=nfw. Zasad jama)(Guéorguiev, 1990).

Anthroherpon matzenaueri matzenaueri (Apfelbeck, 1907)

Antroherpon Matzenaueri Apfelbeck, 1907: 401. Lct: "in antro montis Ledenica"
= *Antroherpon Hawelkai* Knirsch, 1929: synonymy in Müller, 1937: 117
Lct: "Ein Schacht des Bjelodol, Umgebung von Gačko, Montenegro"
= *Antroherpon augustae havelkai* Jeannel, 1930: 139. (change of status)
= *Antroherpon matzenaueri* ab. *dilatatus* Apfelbeck: J. Müller, 1913: 129. (*nomen nudum* as ab.)

Type locality: Montenegro, Golija Mt., Jama u Certov Do.

Distribution: Caves on Golija Mt. in Montenegro (Guéorguiev, 1990; Giachino & Gueorguiv, 1993): Montenegro, pećine na Goliji. (nfw: Heta jama, Ovdo pećina, Leo pećina, Vela jama, Hazda pećina) (Perreau, 2000).

Diagnosis: Habitus illustrated in Fig. 10E. Pronotum with its largest width at the anterior third of its length and with its lateral sides parallel at most on the basal fourth of its length. Elytral surface with supperficial and uniformly dispersed punctuation. Elytra less elongate in males, less than 1.75 longer than wide. Aedeagus illustrated in Fig. 12I.

DNA-barcode: VA24 (MNE, Golija, Latično, Jama u Rudinskom Dolu; possible type-locality):

```
1         10        20        30        40        50
|         |         |         |         |         |
GTACTTTATATTTTTTAATAGGAACTTGATCAGGAATAATTGGAACCTCA
TTAAGATTATTGATTCGGTCTGAATTAGGAAATCCAGGAAGATTAATTAT
AAATGATCAAATTTATAACGTTATTGTAACAGCTCACGCTTTTATTATAA
TTTTTTTTATGGTTATACCAATTATAATTGGAGGATTTGGAAATTGATTA
GTTCCCTTAATGTTAGGGGCTCCTGATATAGCTTTCCCACGTATAAATAA
TATAAGATTCTGACTATTACCTCCTTCTTTAACCCTACTATTAATAAGAA
GAATCGTAGAAAGAGGAGCCGGAACAGGATGAACTGTTTACCCTCCCCTA
TCTGCTAATATTGCACATAGCGGCCCATCAGTAGATCTTGCTATTTTTAG
CCTCCATTTAGCTGGAATCTCTTCTATCTTAGGGGCAGTAAATTTTATTA
CTACTATCATCAACATACGATCCCCTGGTATATCTTTTGATAAAATACCT
CTATTTGTATGATCTGTAGCTATTACTGCCTTACTATTACTGCTCTCATT
ACCAGTATTAGCTGGGGCAATTACTATATTATTAACTGACCGTAATTTAA
ATACTTCTTTTTTTGACCCTGCTGGAGGAGGAGATCCAATTTTATATCAG
CAT
```

Remarks: It should be pointed out that even though according to some literature data (Guéorguiev, 1990; Perreau, 2000) this species is located on Ledenica (or Ledenice) mountain in Montenegro, Ledenice is a peak on Golija Mt., not a mountain. This can cause confusion because „Ledenica“ is a comon toponym in Montenegro, and also a common name for a cave or a pit. Giachino & Guéorguiev (1993) explained that three caves named as „Jama u mjestu Goliji“ (Pretner, 1977), „Jama u Certov do“ (Absolon & Lang, 1993; Absolon, 1943) and „Jama u Crtovom dolu“ are synonyms for a single cave located 5 km SW from the village Goransko in western Montenegro. Jama u Rudinskom dolu is located 5 km south from the village Goransko, so it is either very close to the type locality or it is perhaps the type locality itself of *A. matzenaueri matzenaueri*.

Anthroherpon matzenaueri taliensis **(Zariquiey, 1927)**

Antroherpon Augustae subsp. *taliensis* Zariquiey, 1927: 160. Lct: “Höhle 23, Mika J., Tali Gruppe“

Type locality: Montenegro, Moračke Mt., Tali, Žurim, Gornja jama u Cetkov vrh.

Distribution: Several caves on Tali in central Montenegro: Gornja jama u Četovom vrhu (nfw: Mika jama) and jama Pječaljina (nfw: Maj jama) in the vicinity of Zvornik in the Tali planina (Guéorguiev, 1990; Perreau, 2000); Moračke planine, Kapetanovo jezero, Dola pećina, 1788 m, N 42°48.520' E 19°14.438' (new locality); Moračke planine, Kapetanovo jezero, Mlječikova pećina, 1811 m, N 42°48.643' E 19°14.388' (new locality).

DNA-barcode: IE2 (MNE, Moračke Mt., Kapetanovo jezero, Dola pećina):

```
1         10        20        30        40        50
|         |         |         |         |         |
```

```
TTTAATAGGAACTTGATCGGGAATAATTGGAACCTCATTAAGATTATTGA
TTCGGTCCGAATTAGGAAATCCAGGAAGATTAATTATAAACGATCAAATT
TATAATGTTATTGTAACAGCTCATGCTTTTATTATAATTTTTTTTATAGT
TATACCAATTATAATTGGAGGATTTGGAAATTGATTGGTTCCCTTAATAT
TAGGGGCTCCTGATATAGCTTTTCCGCGTATAAATAATATAAGATTCTGA
CTATTACCTCCTTCTTTAACCCTACTATTAATAAGAAGAATCGTAGAAAG
AGGGGCTGGAACAGGGTGAACTGTTTACCCCCCCCTATCTGCTAATATTG
CCCACAGTGGTCCATCAGTAGATCTTGCTATTTTTAGCCTCCATTTAGCT
GGAATCTCTTCTATCTTAGGAGCAGTAAATTTTATTACTACTATCATCAA
CATACGATCCCCCGGTATATCTTTTGATAAAATACCTTTATTTGTGTGAT
CTGTAGCTATTACTGCCTTATTATTACTACTCTCATTACCAGTATTAGCT
GGAGCAATTACTATGTTATTAACTGACCGTAATTTAAATACTTCTTTTTT
TGACCCTGCTGGAGGAGGAGATCCAATTTTATACCAGCATTTATTT
```

Remarks: Lives in sympatry with *A. taxi sydowi* in Mlječikova pećina.

Genetic distance between *A. matzenaueri matzenaueri* and *A. matzenaueri taliensis* is 4.2%. Morphologically these taxa differ in the shape of the pronotum: in *A. matzenaueri matzenaueri*, the pronotum is much wider and rounded in the anterior part in comparison to *A. matzenaueri taliensis*. Moreover, the mesothoracic pedunculus of *A. matzenaueri matzenaueri* is slightly longer than in *A. matzenaueri taliensis*. Aedeagi of *A. matzenaueri* ssp. are illustrated in Fig. 12 H, I, G.

8. ***Anthroherpon sinjajevina*** **Njunjić, Perreau, Pavićević, 2015**

Anthroherpon sinjajevina Njunjić, Pavićević & Perreau, 2015: 403. Lct: “Montenegro, Sinjajevina Mt, Njegovuđe, village Rudanca, Blažova pećina, 1470 m“

Type locality: Montenegro, Sinjajevina Mt, Njegovuđe, village Rudanca, Blažova pećina, 1470m, N 43°4'10.99" E 19°21'57.42.

Distribution: known only from the type locality.

Diagnosis: Habitus illustrated in (see below: Article 1). Distinct from the other species of the group by the following characters: second antennomere approximately two times shorter than the first one. Sides of pronotum convergent behind the anterior third of the pronotum length, the sides parallel on the basal fourth of the pronotum length. Elytral surface with microreticulation between punctures. Pronotum without symmetrical dorsal impressions. Parameres shorter than median lobe (setae excluded), with a constriction in the preapical region and with acute apex (Fig. 12K).

DNA-barcode: VA6 (Montenegro, Sinjajevina Mt, Njegovuđe, village Rudanca, Blažova pećina; type locality):

```
1        10        20        30        40        50
|        |         |         |         |         |
GTACTTTATATTTTTTAATAGGAACTTGATCAGGGATAATTGGAACCTCA
TTAAGACTACTAATTCGGTCCGAATTAGGAAATCCAGGAAGATTAATTAT
AAATGATCAAATTTATAATGTTATTGTAACAGCTCACGCTTTTATTATAA
TTTTTTTTATGGTTATACCAATTATAATTGGAGGATTTGGAAATTGATTG
GTTCCCTTAATGCTAGGGGCTCCTGATATAGCTTTCCCGCGTATAAATAA
TATAAGATTCTGATTATTGCCTCCTTCTTTAACCCTATTATTAATAAGAA
GAATCGTAGAAAGAGGGGCCGGAACAGGATGAACTGTTTACCCCCCCTTA
TCTGCTAATATTGCCCATAGCGGCCCATCAGTAGATCTCGCTATTTTTAG
CCTCCACTTAGCTGGAATCTCTTCTATCTTAGGGGCAGTAAATTTTATTA
CCACTATCATCAACATACGATCCCCTGGCATATCTTTTGATAAAATACCT
CTATTTGTATGATCTGTAGCTATTACTGCCTTACTATTACTACTCTCATT
ACCAGTATTAGCTGGAGCAATTACTATATTATTAACTGACCGTAACTTAA
ATACTTCTTTTTTTGACCCTGCTGGGGGAGGAGATCCAATTTTATACCAG
CATTTATTT
```

9. *Anthroherpon udrzali* Giachino & Vailati, 2005

Anthroherpon matzenaueri udrzali Giachino & Vailati, 2005: 150. Lct: "Montenegro, Orjen planina, pećina u Kučericama"

Type locality: Montenegro, Orjen planina, pećina u Kučericama.

Distribution: known only from the type locality.

Diagnosis: Body length 7.05 mm. Elytral surface with stronger punctuation and with clear traces of transversal alignments in the humeral region (anterior third of the elytral length). Elytra very elongate in males, more than 1.9 times longer than wide. Aedeagus with median lobe triangularly narrowed at the apex in dorsal view (Fig. 12L), strongly curved in the basal region, then more or less straight and flat in the central and apical regions, the dorsal side sinuate in lateral view. Parameres thick, slightly shorter than the median lobe (apical setae excluded), with three apical setae.

2.6.4. IV "*latipenne* " group V. B. Guéorguiev, 1990

10. *Anthroherpon elongatum* Giachino & V. B. Guéorguiev, 1993

Antroherpon elongatum Giachino & V. B. Guéorguiev, 1993: 298. Lct: "Erzegovina, Baba planina, S. Gacko, grotta Snijeznica Cancarica"

Type locality: Bosnia and Herzegovina, Gacko, Baba Mt., cave Snježnica Čančarica.

Distribution: known only from the type locality.

Diagnosis: species was not examined in this study.

Remarks: This species was described on the basis of a single male. Habitus and the aedeagus are illustrated and show clear differences from other species of "*latipenne* " group.

11. ***Anthroherpon garbellii*** **Giachino & Vailati, 2005**

Anthroherpon garbellii Giachino & Vailati, 2005: 157. Lct: "Albania, Boga, Mt. Çardak, Spella e Pucit, 1600m"

Type locality: Albania, Boga, Mt. Çardak, Spella e Pucit, 1600 m.

Distribution: known only from the type locality.

Diagnosis: species was not examined in this study.

Remarks: Lives in sympatry with *A. taxi terezzi* in Spella e Pucit.

12. ***Anthroherpon gueorguievi*** **Giachino & Vailati, 2005**

Anthroherpon Guéorguievi Giachino & Vailati, 2005: 157. Lct: "Serbia-Montenegro, Bundas (Golija) Bajovo polje, Dukova ledenica"

Type locality: Montenegro, village Bajovo Polje, Golija Mt., Bundas, Dukova ledenica.

Distribution: known only from the type locality.

Diagnosis: species was not examined in this study.

Remarks: This species was described on the basis of a single female, so the description is lacking information about the habitus of the male and the morphology of the aedeagus. Due to intrapopulational variability and sexual dimorphism, descriptions based on a single female are not reliable.

13. ***Anthroherpon hossei*** **(Winkler, 1925)**

Antroherpon Hossei Winkler, 1925: 143. Lct: „Schachthöhle des Gebirgszuges nordwestlich von Trebinje"

Type locality: Bosnia and Herzegovina, Trebinje, Turica, Mravinjac (=Pećina u Mravinjac), 994 m, N 42°50'2.40" E 18° 8'35.46".

Distribution: known only from the type locality.

Diagnosis: Habitus illustrated in Fig. 10F. Body length from 5.4 to 5.6 mm. Pronotum two times longer than wide, slightly narrowing in the anterior part, and with slightly concave to parallel lateral sides in the posterior third of the length; without mesothoracic pedunculus; elytra wide, with deep punctuation, and medium-long and sparse hairs. Aedeagus was not examined in this study.

Remarks: Lives in sympatry with *A. primitivum jeanneli* in Pećina u Mravinjac.

14. *Anthroherpon latipenne* (Apfelbeck, 1907)

Anthroherpon latipenne attenuatum (Jeannel, 1934)

Antroherpon (s. str.) *Matzenaueri* subsp. *attenuatum* Jeannel, 1934: 160. Lct: "Tomo pećina dans le massif du Lovćen, Montenegro occidental"

Type locality: Montenegro, Lovćen Mt., Ledenica kod Lipske ploče (Perreau, 2000).

Distribution: known only from the type locality.

Anthroherpon latipenne goettli (Zariquiey, 1927)

Antroherpon Matzenaueri subsp. *Gottli* Zariquiey, 1927: 163. Lct: "Höhle 29, Niva J., Ziovo Gruppe"
= *Antroherpon brevipenne* Knirsch, 1928

Type locality: Montenegro, Žiovo, Zatrijebački katun, Korita, 1360 m, Sniježnica jama.

Distribution: caves on Žiovo Mt. in Montenegro: Kučka Korita, Jama u Borovima, 1253 m, N 42°28'50.40" E 19°30'4.20" (new locality); Korita, Kišna jama, 1364 m, N 42°29'33.60" E 19°32'52.60" (new locality).

DNA-barcode: VA28 (MNE, Kučka Korita, Jama u Borovima)

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1        10        20        30        40        50
|        |         |         |         |         |
GCACTTTATATTTTTTACTAGGAACCTGATCAGGAATAATTGGAACTTCA
TTAAGATTATTGATTCGAGCTGAACTAGGAAACCCAGGAAGTTTAATTAT
AAATGATCAAATTTATAATGTTATTGTAACAGCTCATGCTTTTATTATAA
TTTTTTTTATAGTTATACCAATTATAATTGGAGGATTTGGAAATTGACTA
GTTCCTTTAATACTAGGGGCTCCTGATATAGCTTTCCCACGCATAAATAA
TATAAGATTTTGATTACTACCCCCCTCTCTAACTCTCTTACTTATGAGAA
GTATAGTAGAAAGGGGAGCTGGAACAGGATGAACTGTTTACCCACCTTTA
TCAGCTAATATTGCTCATAGTGGTCCCTCTGTGGACTTAGCAATTTTTAG
CCTTCATTTAGCTGGAATCTCTTCTATTTTAGGCGCAGTAAATTTTATCA
CTACTATTATTAATATACGATCTCCTGGAATATCTTTTGACAAAATACCC
CTATTTGTATGATCTGTAGCTATTACTGCCTTATTATTACTACTTTCATT
GCCTGTATTAGCTGGCGCAATTACTATACTACTAACAGATCGAAACTTAA
ATACTTCTTTTTTTGATCCTGCAGGAGGCGGAGATCCAATTTTATACCAA
CATTTATTT
```

Remarks: Genetic distances between this subspecies and 7 different populations of *A. latipenne latipenne* range from 1.1 to 1.3 %. However, these two subspecies differ in the shape of the pronotum: part of the pronotum posterior of the constriction is wider in *A. latipenne goettli* which makes it look more robust. Moreover, the sides of the median lobe of the aedeagus of *A. latipenne goettli* is more convex in dorsal view in comparison to *A. latipenne latipenne*.

***Anthroherpon latipenne latellai* Giachino & Vailati, 2005**

Anthroherpon latipenne latellai Giachino & Vailati, 2005: 153. Lct: "Albania, Massiccio del Kollatës, Mt. Mali i Thatë, shpella e Haxhisë, 1720 m"

Type locality: Albania, Massiccio del Kollatës, Mt. Mali i Thatë, shpella e Haxhisë, 1720m.

Distribution: Albania, massive Kollatës: Mt. Mali i Thatë, cave shpella e Haxhisë, 1720m; cave Rrethit të Bardhë, 1850 m (Giachino & Vailati, 2005).

***Anthroherpon latipenne latipenne* (Apfelbeck, 1907)**

Antroherpon Matzenaueri var. *latipenne* Apfelbeck, 1907: 401. Lct: "in antro montis Ledenica"
= *Antroherpon Luciani* Müller, 1913
= *Antroherpon* (s. str.) *Matzenaueri* Apfelbeck: Jeannel, 1924

Type locality: The exact locality is unknown (cave on Golija Mt.).

Distribution: Caves on Bjelasnica Mt. and Baba Mt. (both in BiH, near Gacko), and Orjen Mt., Moračke planine, Dobreljica Mt, and Golija Mt. (all in MNE). Caves on Bjelasnica Mt.: Jama pod Strucom Čolovisem, Vučja bara; Jama Torina vući, Vučja bara; Sniježnica (=Ajba jama), Tisovi krš; Pećina u Tišovom kršu; Sniježnica više Jelova dola; Velika pećina u Bašte; Jaskovac jama; Ledenica u Komovima; Agova pećina; Pećina u vali pod sedlom; Pećina u čistoj strani; Nova jama u Tisovom kršu; Stari pećina; Snježnica pri šumarksoj kuču, Vučja bara; Bezimena jama (=Gova jama), Vučja bara (Guéorguiev, 1990), Sari pećina; Pećina u Klacinama (Giachino & Vailati, 2005). Caves on Baba Mt.: Pećina u stare kolibe; Jama kod Vukovljevih koliba; Ledenica jama; Zubovića pećina; pećina u Jasenovom dolu; Pećina u Siljevom komu; Jama u Ivovom dolu; Cekova jama; Torica pećina; Sniježnica Čančarica (Guéorguiev, 1990); Golubača jama (Giachino & Vailati, 2005). Caves on Orjen Mt.: Pećina u Kučericama; cave on Lisac near Dvrsno polje; Rajčeva jama; Izeta pećina, Knezlaz, 711 m, N 42° 32' 38.2'' E 18° 41' 20.2''; Pećina kod Grkovca; Jelova jama, Koprivni Do, Ubli; Sniježna jama (=Eishohle); Orjen- Sattel; Snježnica na dnu Koprivnih Aluga, Kuk (Guéorguiev, 1990); Crkvice, Ericova jama, 1030 m, 42°33'52.56" E 18°38'32.04" (Eric Quéinnec, pers. comm. cited in: Kurtović et al., 2008); Ubla, Bukova rupa, 1259 m, N 42°36'34.20" E 18°30'11.22". Cave on Moračke Mt.

(MNE): Kapetanovo jezero, Dola pećina, 1788 m, N 42°48.520' E 19°14.438' (new locality). Caves on Dobreljica Mt.: Kazanci, Bijela pećina (new locality); Nikšić, Donje Čarađe, Prljača, 1150 m, N 43° 1'4.01" E 18°40'5.01" (new locality). Caves on Golija Mt.: Jama u Bijelom dolu (=Hava pećina), Ledenice (Guéorguiev, 1990); Ćurilo, Snežnica (new locality).

Diagnosis: Body length from 7.6 to 8.4 mm. Distinct from other species of the „*latipenne*“ group by the following combination of characters: pronotum about 1.7 times longer than wide, particularly elongated in the posterior part where the lateral sides of the pronotum are slightly divergent in dorsal view; elytra slightly depressed along the sutural line, with extremely short hairs which are very dense but present only dorsally, while the lateral sides of the elytra are hairless. Aedeagus illustrated in Fig. 11F. Median lobe wedge-shaped, much longer than the parameres (setae excluded), with very elongated and pointed apex, and with a thickened sclerotized strip along the length of the medioventral side of the median lobe; parameres convex, but more straight in the apical part, bearing two long apical and one short subapical setae.

DNA-barcode: VA29 (MNE, Dobreljica, Bijela pećina)

```
1        10        20        30        40        50
|        |         |         |         |         |
GTACTTTATATTTTTTACTAGGAACCTGATCAGGAATAATTGGAACTTCA
TTAAGATTATTGATTCGAGCTGAACTAGGAAACCCAGGAAGTTTAATTAT
AAATGATCAAATTTATAATGTTATTGTAACAGCTCATGCTTTTATTATAA
TTTTTTTTATAGTTATACCAATTATAATTGGAGGATTTGGAAATTGACTA
GTTCCTTTAATACTAGGAGCTCCTGATATAGCTTTCCCACGCATAAATAA
TATAAGATTTTGATTACTACCCCCCTCTCTAACTCTCTTACTTATAAGAA
GTATAGTAGAAAGGGGAGCTGGAACAGGATGAACTGTTTACCCACCTTTA
TCAGCTAATATTGCTCATAGTGGTCCTTCTGTGGACTTAGCAATTTTTAG
CCTTCATTTAGCTGGAATCTCTTCTATTTTAGGCGCAGTAAATTTTATCA
CTACTATTATTAATATACGATCTCCTGGAATATCTTTTGACAAAATACCC
CTATTTGTATGATCTGTAGCTATTACTGCCTTATTATTATTACTTTCATT
GCCTGTATTAGCTGGCGCAATTACTATACTATTAACAGATCGAAACTTAA
ATACTTCTTTTTTTGATCCTGCGGGAGGCGGAGATCCAATTTTATACCAA
CATTTATTT
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Remarks: This species lives in sympatry with four other species of the genus *Anthroherpon*: with *A. taxi hercegovinum* and *A. elongatum* on Baba Mt. and Bjelasnica Mt., with *A. taxi taxi* and *A. matulici* in several caves on Orjen Mt., with *A. taxi pretneri* in Ledena pećina na V. Kiti on Trubjela, and with *A. matzenaueri taliensis* in Dola pećina near Kapetanovo jezero on Moračke planine. By the shape of the pronotum and conformation of the aedeagus this species mostly resembles *A. taxi*, but it can be easily distinguished from it by the absence of hairs on the lateral side of the elytra. Genetic distances between *A. taxi* senso lato and *A. latipenne* range from 4% to 6.4%.

Anthroherpon latipenne punctipennis **(Jeannel, 1930)**

Anthoherpon (s. str.) *matzenaueri* subsp. *punctipennis* Jeannel, 1930: 142. Lct: "Montenegro central, massif du Maganik, nwf: Lug jama"

Type locality: Montenegro, Maganik Mt., Koprivni Do, Bezimeno brezno.

Distribution: known only from the type locality.

Remarks on *A. latipenne* senso lato: This species has a very large distribution area: from Orjen Mt. (MNE) close to the Adriatic Sea on the south, to Golija Mt. (MNE) on the north (a territory more than 60 km long), and from Baba Mt. and Bjelasnica Mt. (both in BiH) on the east to Žiovo Mt. (MNE) on the west (approximately 116 km apart). Specimens from different populations have somewhat different shape of pronotum and elytra. These differences also exist between males and females of the same population, with females having wider pronotum and wider elytra than males. Specimens from different populations have a different colour of the integument: specimens from Orjen Mt. have the entire body very dark, reddish-brown and very shiny, while specimens from the northernmost part of the distribution area (Golija Mt., and Dobreljica) are almost matte, lighter and have yellowish-brown elytra and dark brown pronotum. Despite these morphological differences, genetic distances between 8 different populations of *A. latipenne* (Orjen Mt., Bukova rupa, Orjen Mt., Ericova jama; Trubjela, Ledena pećina na Kiti; Žiovo, Jama u Borovima; Moračke Mt., Dola pećina; Dobreljica, Prljača; Dobreljica, Kazanci, Bijela pećina, and Golija, Ćurilo, Snežnica) are small, in most cases less than 1%, and maximally 1.4 %.

15. *Anthroherpon piesbergeni* (Zariquiey, 1927)

Antroherpon Piesbergeni Zariquiey, 1927: 161. Lct: "Höhle 51, Otvor J., Lovcen Gaú"

Type locality: Montenegro, Lovćen Mt., Njeguši, east of Vršanj, Bezdan jama.

Distribution: Montenegro, Podbožur, Bijele Rudine, Bajovo polje, Manja pećina sa vodom, 913 m, N 42°43'39.12" E 18°43'6.30".

Diagnosis: Habitus illustrated in Fig. 10G. Body length 5.7 mm. It resembles *A. primitivum* but *A. piesbergeni* has a slightly longer mesothoracic pedunculus and a different shape of the pronotum: wider and more rounded in the anterior part, and with lateral sides almost parallel posterior of the constriction, like a light bulb. Punctuation of the elytra is deep and dense (punctures almost touching each other); elytra are covered with short and sparse hairs. Aedeagus illustrated in Fig. 11G: very big and robust, only very slightly curved in lateral view; median lobe longer than parameres, thick in dorsal and in lateral view, and with broad, rounded apex, and without the thickened sclerotized strip along the length of the medioventral side of the median lobe; parameres thin, very convex and somewhat wider in the apical part, and bearing two setae.

16. *Anthroherpon primitivum* (Absolon, 1913)

Anthroherpon primitivum jeanneli (Winkler, 1925)

Antroherpon Jeanneli Winkler, 1925: 143. Lct: "Schachthöhle des Gebirgszuges nordwestlich von Trebinje"

Type locality: The exact locality in unknown.

Distribution: Caves on Bjelasnica Mt. and Ilijina Mt. in Bosnia and Herzegovina: Golubnja jama (=Oskar jama), Turica, Bjelasnica; Schacht, Tuhalska Bjelina, Bjelasnica; Bisačina jama (=Sači jama), Drapići, Ilijina planina; Javorova rupa, Drapići, Ilijina planina (Gueoruiev, 1990); Bjelasnica Mt., Motka, Turica, Jama u Motki (=Velika lazina), 1040 m (Pavićević & Perreau, 2000); Bosnia and Herzegovina, Trebinje, Turica, Mravinjac (=Pećina u Mravinjac), 994 m, N 42°50'2.40" E 18° 8'35.46".

Diagnosis: Habitus illustrated in Fig. 10H. Body length 6.7 – 7 mm. Pronotum 1.6 times longer than wide, lateral sides in dorsal view very convex in the anterior part, and convergent posteriorly; without mesothoracic pedunculus; elytra wide, with deep and clear punctuation (punctures are touching each other), and very short hairs. Aedeagus illustrated in Fig. 11H. Median lobe thick and robust, longer than parameres, with lanceolate apex, and with a thickened sclerotized strip along the length of the medioventral side of the median lobe; parameres almost straight, thick in the basal part, and bearing two apical and one subapical setae.

DNA-barcode: VA18 (BiH, Turica, Pećina u Mravinjac)

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1        10        20        30        40        50
|        |         |         |         |         |
GTACTTTATATTTTTTATTGGGAACTTGATCAGGAATAATTGGGACATCA
CTTAGATTACTAATTCGAGCTGAATTAGGTAACCCAGGTAGCCTAATTAT
AAATGACCAAATCTACAATGTTATTGTAACAGCTCATGCTTTTGTGATAA
TTTTTTTCATGGTCATGCCTATTATAATCGGAGGATTTGGAAATTGACTA
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GTTCCTTTAATATTAGGAGCCCCTGATATAGCCTTTCCACGCATAAATAA
TATAAGATTTTGACTATTACCCCCTTCTTTAACTTTATTACTCATAAGAA
GAATGGTAGAAAGAGGGGCTGGAACAGGATGAACTGTTTACCCTCCCCTA
TCTGCCAATATCGCCCATAGAGGCCCATCAGTAGACTTAGCTATTTTTAG
GCTTCATCTAGCTGGAATCTCTTCTATTTTAGGAGCAGTAAATTTTATTA
CTACTATTATCAATATACGAGCTCCTGGTATATCTTTCGATAAAATATCA
CTATTTGTATGATCCGTAGCTATTACTGCTCTATTGCTACTACTTTCATT
ACCAGTATTAGCCGGTGCAATTACTATACTATTAACAGACCGAAATTTAA
ATACCTCTTTCTTTGACCCCGCAGGGGGAGGAGACCCAATTCTATACCAG
CATTTATTT
```

Remarks: Lives in sympatry with *A. hossei* in Pećina u Mravinjac.

Anthroherpon primitivum primitivum **(Absolon, 1913)**

Antrophilon primitivum Absolon, 1913: 101. Lct: "Höhle an der Zaton-(Malfi)-Bucht, hart an der herzegovinisch-dalmatinischen Grenze"

Type locality: Bosnia and Herzegovina, Grepci, Kali pećina (Perreau, 2000).

Distribution: known only from the type locality.

17. ***Anthroherpon taxi*** **(Müller, 1913)**

Anthroherpon taxi albanicum **(Apfelbeck, 1919)**

Antroherpon albanicum Apfelbeck, 1919: 271. Lct: "Nordalbanische Alpen. In einem Schneeloch der Čafa Brojs (Hochgebirge)"

Type locality: unnamed cave (snow cave) on Ćaf Bora (=Ćaf Borit) on Prokletije Mt.

Distribution: known from several caves on Prokletije Mt., on the border of Montenegro and Albania: Troposa, Valbona, Massiccio del Kollatës, pozzo di Rrethit të Bardhë, 1850 m; Troposa, Valbona, Mt. Lugu i Cirkut, Grotta presso la Miniera di bauxite, 1850 m (Giachino & Vailati, 2005); Prokletije, Ćaf Bora, several vertical caves: Maja Kolata, Kolektor, 1252 m, N 42°29'47.44" E 19°53'49.26"; Bjelič, Babina sisa, 1965 m, N 42°30'0.90" E 19°53'57.54"; Bjelič, Jama ispod Maja Nike, 2000 m; Kolata, Oktopod (=03 139-1), 2008 m; Dmuhava (all new localities).

DNA-barcode: VA40 (Montenegro, Prokletije, Ćaf Bora, Kolektor)

```
1        10        20        30        40        50
|        |         |         |         |         |
GTACTTTATATTTTTTACTAGGAACCTGATCAGGAATAATTGGAACTTCA
TTAAGATTATTGATTCGAGCTGAATTAGGAAATCCAGGAAGTTTAATCAT
AAATGATCAAATTTATAACGTTATTGTAACAGCCCATGCTTTTATTATAA
TTTTTTTTATAGTTATACCAATTATAATCGGAGGATTTGGAAATTGACTA
GTTCCTTTAATACTGGGAGCCCCTGATATAGCTTTCCCACGTATAAATAA
TATAAGATTTTGATTATTACCCCCCTCTTTAACTCTCTTACTGATAAGAA
GTATAGTAGAAAGAGGAGCCGGAACAGGATGAACTGTTTACCCCCCTTTA
TCAGCTAATATTGCCCATAGAGGTCCTTCTGTGGACTTAGCAATTTTTAG
CCTCCATTTAGCTGGGATCTCTTCTATTTTAGGAGCAGTGAATTTTATCA
CTACTATTATTAATATACGATCTCCTGGAATATCTTTTGACAAGATACCC
CTATTTGTATGATCTGTAGCAATTACTGCCTTATTATTATTGCTTTCATT
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ACCTGTATTAGCTGGCGCAATTACTATACTACTAACAGATCGAAACTTAA
ACACTTCTTTTTTTGATCCTGCAGGAGGTGGAGATCCAATTTTATACCAA
CATTTATTT
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Remarks: Specimens from Ćaf Bora were described as *A. albanicum* by Apfelbeck (1919) and then lowered to a subspecies rank by Giachino & Vailati (2005) but without any explanation: they simply listed this species as a subspecies of *A. taxi*. Interestingly, this taxon was not included in the list of taxa belonging to the genus *Anthroherpon* neither by Guéorguiev (1990), nor by Giachino & Guéorguiev (1993) even before it was lowered to a subspecies rank by Giachino & Vailati (2005). Perhaps this was due to an oversight on the authors' part. On the other hand, in papers of Jeannel (1910; 1924; 1930), this taxon is listed as the species *A. albanicum*. Due to the great genetic distances between this taxon and *A. taxi* (ranging from 5.7 % to 6.2 %) (see Table 2) and the different morphology of the pronotum, this taxon should indeed be elevated to species level. Aedeagus illustrated in Fig. 11M. Median lobe longer than parameres (setae excluded), with drawn-out, pointed apex, and with a thickened sclerotized strip along the length of the medioventral side of the median lobe which is gradually tapering towards the apical part; parameres convex in dorsal view, but almost straight in the apical part, bearing three long setae.

Anthroherpon taxi boschi **(Zariquiey, 1927)**

Antroherpon Winkleri subsp. *Boschi* Zariquiey, 1927: 162. Lct: "Höhle 30, Dúboka J., Mokra pl. Gau"
= *Antroherpon montenegrinum* Knirsch, 1928: 118. synonymy in Jeannel, 1929: 297
Lct: "Duboka, Mokra planina. Montenegro"
= *Antroherpon albanicum gracile* Winkler, 1933: 77; synonymy in V. Guéorguiev, 1990: 258
Lct: "Montenegro: Mokra planina, pečina u Bukovini"

Type locality: Mokra planina Mt. (border of Montenegro and Serbia), Čakor, Čavčina jama.

Distribution: Kosovo, Peć, Alta Valle Rugova, Grotta presso Drelje, 1129 m (Giachino & Guéorguiev, 2005); Mokra planina Mt., Pećina u Bukovini (Perreau, 2000).

Anthroherpon taxi hercegovinum **(Winkler, 1938)**

Antroherpon Taxi hercegovinum Winkler, 1938: 220. Lct: "Höhle Pod Colovišem strugom"

Type locality: Bosnia and Herzegovina, Baba Mt., cave Pod Colovišem strugom.

Distribution: Caves on Baba Mt. in Bosnia and Herzegovina: pecina Torina; Čenova pećina; Bukovica pećina; Jama u Jasenovom Dolu; Jama u Jovovom dolu; Jama u

Vukovijeh koliba; Zubovića pećina; Vučja Bara; Ledenica jama; Čavčevica jama; Pećina u stare kolibe, Orlovica pećina; Caves on Bjelasnica Mt. in Bosnia and Herzegovina: Pećina u Klačinama; Pećina u Čistoj strani; Sniježnica više Jelova dola; Velika pećina u Baste; Jasikovac jama; Bukova jama; Pećina u Vali pod sedlom; Agova pećina; Ledenica u Komovima; Sniježnica u Tisovom kršu; Nova jama u Tisovom kršu; Jegikovac jama.

Remarks: Lives in sympatry with *A. latipenne latipenne* in numerous caves on Baba Mt. and Bjelasnica Mt.

Anthroherpon taxi lemur **(Knirsch, 1929)**

Antroherpon lemur Knirsch, 1929: 96. Lct: "Bei Kula Šejtan, westlich von Gacko. Montenegro"
= *Antroherpon (s. str.) albanicum* subsp. *ledenicense* Jeannel, 1934: 159. synonymy in V. Guéorguiev, 1990: 258
Lct: "Hadža pećina, dans les monts Ledenica, nord du Monténégro [= Ljutska pećina, Šejtan Kula]"

Type locality: Montenegro, Goransko, Kula Šejtan, Hadža pećina (=Ljutska pećina-Ledenice) (Guéorguiev, 1990).

Distribution: Caves on Golija Mt. in Montenegro (Jeannel, 1934); Ledenice, Golija Mt., Bezimena snežna jama (Giachino & Guéorguiev, 1993).

Anthroherpon taxi muelleri **(Zariquiey, 1927)**

Antroherpon Winkleri subsp. *Mülleri* Zariquiey, 1927: 162. Lct: "Höhle 35, nfw: Kuna P., Visitor Gruppe"
= *Antroherpon grebense* Knirsch, 1928: 119. synonymy in Jeannel, 1929: 297
Lct: "Kuna, Greben. Montenegro"

Type locality: Montenegro, Visitor Mt., unnamed cave (Perreau, 2000).

Distribution: known only from the type locality.

Anthroherpon taxi pretneri **Giachino & Vailati, 2005**

Anthroherpon taxi pretneri Giachino & Vailati, 2005: 156. Lct: "Crna Gora, Trubleja, Ledenica na V. Kiti"

Type locality: Montenegro, Nikšić, Trubjela, Ledenica na V. Kiti

Distribution: known only from the type locality.

DNA-barcode: VA47 (MNE, Nikšić, Trubjela, Ledenica na V. Kiti; type locality)

```
1        10        20        30        40        50
|        |         |         |         |         |
GTACTTTATACTTTTTACTAGGAACCTGGTCAGGGATAATTGGAACTTCA
TTAAGATTATTAATTCGAGCTGAACTAGGGAACCCGGGAAGTTTAATTAT
GAATGATCAAATTTATAATGTTATTGTAACAGCTCATGCTTTTATTATAA
TTTTTTTTATAGTTATACCAATTATAATCGGAGGATTTGGAAATTGACTA
```

```
GTTCCTTTAATGCTAGGAGCCCCTGATATAGCTTTCCCACGTATAAATAA
CATAAGATTTTGATTATTACCCCCTTCTCTTACTCTTTTACTTATAAGAA
GCATAGTAGAAAGAGGAGCTGGAACAGGATGAACTGTTTACCCCCCTTTA
TCAGCTAATATTGCTCATAGTGGCCCTTCTGTAGACTTAGCAATTTTTAG
CCTTCATTTAGCTGGAATCTCTTCTATTTTAGGAGCAGTAAATTTTATCA
CTACTATTATTAATATACGATCTCCTGGAATATCTTTTGACAAAATACCC
TTATTTGTATGATCTGTAGCTATTACTGCCTTACTACTATTACTTTCATT
ACCTGTATTAGCTGGCGCAATTACTATGCTACTAACAGATCGAAACTTAA
ATACTTCTTTTTTTGATCCTGCAGGAGGCGGAGATCCAATTTTATACCAA
CACTTGTTT
```

Remarks: This subspecies was described on the basis of a single female so the description is lacking information about the habitus of the male and the morphology of the aedeagus. Two males and two females from the type locality were examined and one male was barcoded. It differs in barcoding region by 1.6 % from *A. taxi sydowi* and by 2% from *A. taxi taxi*. However, genetic distances between *A. taxi pretneri* and *A. taxi albanicum* range from 6 % to 7.4 % which is the percentage that is usually found above the species level in Leiodidae. This justifies raising *A. taxi albanicum* to species rank. Lives in sympatry with *A. latipenne latipenne* in Ledenica na V. Kiti on Trubjela.

Description of the male.

Head: hypognathous, wider in the anterior part and wider than the pronotum. Surface matte, microreticulated, with sparse and short hairs (shorter than on the elytra). Last maxillary palpomere very short in comparison to the penultimate one, with a dorso-lateral comb of very short bristles. Second antennomere approximately 1.8 times shorter than the first one, last antennomere shorter than the penultimate.

Pronotum: about 2 times longer than wide, with the largest width in the anterior third of the length, and lateral sides slightly diverging posteriorly in dorsal view; dark coloured, matte, with microreticulated dorsal surface. Only on the right, lateral side of the pronotum there is a single, very long and erected bristle.

Elytra: elliptic, light-brown coloured and lighter than the rest of the body, with medium-long, semi-erect hairs along the whole elytral surface, and shallow, indistinct punctures.

Legs: very long, slender, femora gradually thickened in their basal parts, tibiae straight.

Aedeagus: illustrated in Fig. 11L. Very curved in lateral view; median lobe robust, longer than parameres (setae excluded), with convex lateral sides in dorsal view, gradually narrowing towards the apex; apex pointed; parameres convex, of uniform width along their full length and with three setae in the apical part; two located on the apex are longer than the one located subapically.

Anthroherpon taxi remyi **(Jeannel, 1931)**

Antroherpon (s. str.) *albanicum* ssp. *Remyi* Jeannel, 1931: 265. Lct: "Yougoslavie: Zupanska pecina près de Lubnice"

Type locality: Montenegro, Berane, Lubnice, Županska pećina (=Župan pećina, Velika Braćanovića), 1205 m, N 42°51'17.20" E 19°44'50.10".

Distribution: known only from the type locality.

DNA-barcode: VA11 (Montenegro, Berane, Lubnice, Županska pećina; type locality):

```
1        10        20        30        40        50
|        |         |         |         |         |
GTACTTTATATTTTTTACTAGGAGCCTGATCAGGAATAATTGGAACTTCA
TTAAGATTATTGATTCGAGCTGAATTAGGAAACCCAGGAAGGTTAATTAT
AAATGATCAAATTTATAATGTTATTGTAACAGCCCATGCTTTTATTATAA
TTTTTTTTATAGTTATACCAATTATAATCGGAGGATTTGGAAATTGACTA
GTTCCTTTAATACTAGGAGCCCCTGATATAGCTTTCCCACGTATAAATAA
TATAAGATTTTGATTATTACCCCCATCTTTAACTCTCTTACTTATAAGTA
GTATAGTAGAAAGAGGAGCCGGAACAGGATGAACTGTTTATCCTCCTTTA
TCAGCTAATATTGCCCATAGAGGTCCTTCTGTGGACTTAGCAATTTTTAG
CCTCCATTTAGCTGGAATCTCTTCTATTTTAGGGGCAGTGAATTTTATCA
CTACTATTATTAATATACGATCTCCTGGAATATCTTTTGACAAAATACCC
CTATTTGTATGATCTGTAGCTATTACTGCCTTATTATTATTGCTTTCATT
ACCTGTATTAGCTGGCGCAATTACTATACTGCTAACAGATCGAAACTTAA
ACACTTCTTTTTTCGATCCTGCAGGAGGCGGAGATCCAATTTTATACCAA
CATTTATTT
```

Remarks: Habitus illustrated in Fig. 10I. This taxon was initially described as a subspecies of *A. albanicum* (Jeannel, 1931). However, in the paper of Guéorguiev (1990) it is listed as a subspecies of *A. taxi*, but without any explanation for this change. Based on the genetic distances in barcoding region, this taxon appears more closely related to *A. taxi albanicum* (= *A. albanicum*) than to *A. taxi*. It shows from 4.3 to 5.9 % of genetic distance with *A. taxi* senso lato, and 2.7 % of genetic distance with *A. taxi albanicum*. There is confusion regarding the distribution of this subspecies that needs to be clarified. Guéorguiev (1990) mentions that it subspecies is known from 3 caves on Bjelasica Mt. in Montenegro, but all the names he listed are synonyms for a single cave.

Anthroherpon taxi sydowi **(Zariquiey, 1927)**

Antroherpon Winkleri subsp. *Sydowi* Zariquiey, 1927: 162. Lct: "Höhle 23, nfw: Mika J., Tali Gruppe"
= *Antroherpon Winkleri Sydowi* Zariquiey: Jeannel, 1929:297
= *Antroherpon albanicum Sydowi* Zariquiey: Jeannel, 1947:48

Type locality: Montenegro, Moračke planine., Žurim, Zvornik, Četkov vrh, Gornja jama.

Distribution: caves near Žurim on Moračke planine in Montenegro: Čavčina jama, Zvornik; Pječaljina jama, Zvornik (Guéorguiev, 1990); Nikšić, Trubjela, Ledena pećina na Kiti; Kapetanovo jezero, Mlječikova pećina, 1811 m, N 42°48.643' E 19°14.388' (new locality).

DNA-barcode: IE14 (Moračke planine, Kapetanovo jezero, Mlječikova pećina)

```
1         10        20        30        40        50
|         |         |         |         |         |
TGGAACTTCATTAAGATTATTAATTCGAGCTGAACTAGGAAACCCGGGAA
GTTTAATTATAAATGACCAAATTTATAATGTTATTGTAACAGCTCATGCT
TTTATTATAATTTTTTTTATAGTTATACCAATTATAATCGGAGGATTTGG
AAATTGACTAGTTCCTTTAATGCTAGGAGCCCCTGATATAGCTTTCCCGC
GTATAAATAATATAAGATTTTGATTATTACCCCCTTCTCTTACTCTTTTA
CTTATAAGAAGCATAGTAGAAAGAGGAGCTGGAACAGGATGAACTGTTTA
CCCTCCTTTATCAGCTAATATTGCTCATAGTGGCCCTTCTGTAGACTTAG
CAATTTTTAGCCTTCATTTAGCTGGAATCTCTTCTATTTTAGGAGCAGTA
AATTTTATCACTACTATTATTAATATACGATCTCCTGGAATATCTTTTGA
CAAAATACCCTTATTTGTATGATCTGTAGCTATTACTGCCTTACTATTAT
TACTTTCATTACCTGTATTAGCTGGCGCAATTACTATACTACTAACAGAT
CGAAACTTAAATACTTCTTTTTTTGATCCTGCGGGAGGTGGAGATCCAAT
TTTATACCAACACTTGTTT
```

Remarks: Lives in sympatry with *A. matzenaueri taliensis* in Mlječikova pećina.

Anthroherpon taxi taxi **(Müller, 1913)**

Antroherpon Taxi Müller, 1913: 159. Lct: "Höhlen im Orjengebiet (Krivošije, herzegowinisch-dalmatiniches Grenzgebiet)"

Type locality: Montenegro, Orjen Mt, Pećina u Kučericama.

Distribution: numerous caves on Orjen Mt. and Krivišije on Montenegro: Grab, Pećina u Kučericama; Vrbanje, Ledenica na Kalupnoj gredi; Štirovnik, Zla jama (Guéorguiev, 1990); Knezlaz, Izeta pećina, 711 m, N 42° 32' 38.2'' E 18° 41' 20.2''; Crkvice, Křižna jama (=Križeva jama), 1055 m, N 42° 33' 53.39'' E 18° 38' 31.08''; Devočka jama (=CO1), 1460 m, N 42° 33' 35'' E 18° 35' 58''; Jama u Malov Do, 1114 m, N 42° 34' 24'' E 18° 38' 15''; Crkvice, Ericova jama, 1030 m, N 42°33'52.56'' E 18°38'32.04'' (Kurtović et al., 2008); Kameno more, Dvestotka, 965 m, N 42° 33' 20.9'' E 18° 40' 16.1'' (new locality).

DNA-barcode: VA25 (Montenegro, Orjen, Križeva jama)

```
1         10        20        30        40        50
|         |         |         |         |         |
GTACTTTATACTTTTTACTAGGAACCTGATCAGGGATAATTGGAACTTCA
TTAAGATTATTAATTCGAGCTGAACTAGGAAACCCGGGAAGTTTAATTAT
AAATGATCAAATTTATAATGTTATTGTAACAGCTCATGCCTTTATTATAA
TTTTTTTTATGGTTATACCAATTATAATCGGAGGATTTGGAAATTGACTA
GTTCCTTTAATGCTAGGAGCCCCTGATATAGCTTTCCCACGTATAAATAA
TATAAGATTTTGATTATTACCCCCCTCTCTTACTCTTTTACTTATAAGAA
GCGTAGTAGAAAGAGGAGCTGGAACAGGATGAACTGTTTACCCTCCTTTA
TCAGCTAATATTGCTCATAGTGGCCCTTCTGTAGACTTAGCAATTTTTAG
CCTTCATTTAGCTGGAATCTCTTCTATTTTAGGGGCAGTAAATTTTATCA
CTACTATTATTAATATACGATCCCCTGGAATATCTTTTGACAAAATACCC
```

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TTATTTGTATGATCTGTAGCTATTACTGCCTTACTATTATTACTTTCATT
ACCTGTATTAGCTGGCGCAATTACTATGCTACTAACAGATCGAAACTTAA
ATACTTCTTTTTTTGATCCTGCGGGAGGCGGAGATCCAATTTTATACCAA
CACTTGTTT
```

Diagnosis: Habitus illustrated in Fig. 10J. Body length 7.2 – 8 mm. Pronotum elongated, 1.9 times longer than wide, very narrow in the anterior half, wide before the mid-section, and very narrow posterior of the mid-section, with lateral sides slightly divergent after the posterior quarter. Mesothoracic pedunculus short, 2.5 times wider (measured at the narrowest section) than long, narrowest in the anterior part. Elytra wide, without a depression along the sutural line, surface entirely covered with hairs, including the lateral side. Aedeagus illustrated in Fig. 11I. Median lobe longer than parameres (setae excluded), gradually narrowing towards the apical part, with very drown-out, pointed apex, and with a thickened sclerotized strip along the length of the medioventral side of the median lobe; parameres of uniform width along their entire length, slightly convex, and bearing three long setae.

Anthroherpon taxi trezzii **Giachino & Vailati, 2005**

Anthroherpon taxi trezzii Giachino & Vailati, 2005: 154. Lct: "Albania, Boga, Mt. Çardak, shpella e Pucit, 1600 m"

Type locality: Albania, Boga, Mt. Çardak, shpella e Pucit, 1600 m.

Distribution: known only from the type locality.

Remarks. It was found in the same cave as *A. garbellii*.

Anthroherpon taxi winkleri **(Zariquiey, 1927)**

Antroherpon Winkleri Zariquiey, 1927: 162. Lct: "Höhle 34, Poda J., Pajs Gruppe"
= *Antroherpon velare* Knirsch, 1928: 118 synonymy in Jeannel, 1929a: 297
Lct: "Poda I. Pays-Gruppe, Montenegro" [= a pit, Katun Gropa Bajrović, Trojan, Prokletije]
= *Antroherpon velare ssp. divergens* Knirsch, 1928: 122; synonymy in Pretner, 1974: 11
Lct: "Kriva J. Kolac-Gruppe (Montenegro)" [= Špela Koruns, Katun Bjelić (Prokletije)]= *Antroherpon albanicum malissorum* Winkler, 1933: 77. synonymy in Pretner, 1974: 11
Lct: "Nordalbanische Alpen: Maja Rosit an der Montenegrischen Grenze, Spela Karon dei Bjelič" [= Špela Koruns, Katun Bjelić (Prokletije)]

Type locality: Albania, Prokletije Mt, Alm Katun Gropa Bajrović below Karaula Trojan, Unnamed pit, 1760 m.

Distribution: Prokletije Mt on the border of Montenegro and Albania: Špela Koruns (=Kriva jama), Bjelić, Prokletije (Guéorguiev, 1990); Prokletije, caves on Trojan and Greben (nfw: Poda jama, Stražna jama, Snijeg jama, Kriva jama) (Perreau, 2000).

Remarks on *A. taxi* senso lato: Genetic distances between different subspecies of *A. taxi* are ranging from 1.6 to 6.2% (Table 2). The highest genetic distance is between *A. taxi taxi* and *A. taxi albanicum* (6.2 %) which is not surprising since collecting sites of these two subspecies are more than 100 km from each other. The lowest genetic distance is between *A. taxi taxi* and *A. taxi sydowi* (1.6%) even though the collecting sites are about 70 km apart. *A. taxi albanicum* was described as a new species by Apfelbeck (1919) and then lowered to subspecies rank by Giachino & Vailati (2005), but without further explanation: they simply listed this species as a subspecies of *A. taxi*. Interestingly, this taxon is not included in the list of taxa belonging to the genus *Anthroherpon* by Guéorguiev (1990), nor by Giachino & Guéorguiev (1993).

Table 2. Genetic distances in barcoding region among *A. taxi* subspecies.

	A.taxi pretneri	*A.taxi taxi*	*A.taxi sydowi*	*A.taxi remyi*	*A.taxi albanicum*
A.taxi pretneri		2 %	1.6 %	5.9 %	6.1 %
A. taxi taxi	2 %		1.6 %	5.6 %	6.2 %
A.taxi sydowi	1.6 %	1.6 %		4.3 %	5.7 %
A.taxi remyi	5.9 %	5.6 %	4.3 %		2.7 %
A.taxi albanicum	6.1 %	6.2 %	5.7 %	2.7 %	

2.6.5. V “*stenocephalum*” group V. B. Guéorguiev, 1990

18. *Anthroherpon stenocephalum* (Apfelbeck, 1901)

***Anthroherpon stenocephalum noesskei* (Jeannel, 1924)**

Antroherpon (s. str.) *stenocephalum* Subsp. *Noesskei* Jeannel, 1924: 417. Lct: “Banja pećina“

Type locality: Bosnia and Herzegovina, Olovo, Gornja Oćevija, Banja pećina, 830 m, N 44° 9'50.85" E 18°27'21.05".

Distribution: known only from the type locality.

DNA-barcode: VA16 (BiH, Olovo, Gornja Oćevija, Banja pećina; type locality)

```
1        10        20        30        40        50
|        |         |         |         |         |
TAATTGGGACTTCCCTAAGATTATTAATTCGGGCTGAATTAGGAAATCCA
GGTAGTTTAATTATAAATGATCAAATTTACAATGTAATTGTCACAGCTCA
TGCTTTTATTATAATTTTTTTTATAGTTATACCAATTATAATTGGAGGAT
TTGGAAACTGATTAGTTCCTTTAATATTAGGGGCTCCTGATATAGCTTTC
CCACGTATAAATAATATAAGATTTTGATTATTGCCTCCGTCTCTTACTTT
ATTACTTATAAGAAGAATAGTAGAAAGTGGAGCAGGGACAGGATGAACAG
TCTATCCTCCTTTATCAGCTAATATTGCTCATAGTGGCCCATCAGTAGAT
TTAGCAATCTTTAGCCTTCATTTAGCTGGAATCTCTTCTATTTTAGGGGC
```

```
AGTAAATTTTATCACTACTATTATTAATATGCGATCTCCTGGCATATCTT
TTGATAAAATACCATTATTTGTCTGATCTGTAGCTATTACTGCCTTATTA
TTATTACTTTCATTACCAGTACTTGCTGGTGCAATTACTATATTATTAAC
AGATCGAAATTTAAATACTTCTTTTTTTGACCCAGCAGGGGG
```

Anthroherpon stenocephalum stenocephalum (Apfelbeck, 1901)

Antroherpon stenocephalum Apfelbeck, 1901:15. Lct: “Höhlen bei Olovo“

Type locality: Bosnia and Herzegovina, Olovo, Krivajevići, Bijambare (=Čevljanovića pećina), 967 m, N 44° 5'41.40" E 18°29'56.94".

Distribution: BiH, caves near Olovo on Ljubina Mt.: Zvijezda Mt., Jama Delinuša (Guéorguiev, 1990).

Diagnosis: Habitus illustrated in Fig. 10K. Body size 4.6 mm. Distinct from *A. weiratheri* by the following combination of characters: pronotum cylindrical in the anterior part, with lateral sides almost parallel anterior of the constriction, and very divergent posteriorly; pronotum hairless; protibiae slender, straight in the dorsal view; profemora thick in the posterior part, narrower the apical part; elytra very elongated and narrow; larger body size.

Aedeagus illustrated in Fig. 13E. Median lobe much longer than parameres, thin and elongated, with slightly convex lateral sides in dorsal view and lanceolate apex, without the thickened sclerotized strip along the length of the medioventral side of the median lobe; parameres almost straignt but convex in the apical part, of uniform width along their entire length, and bearing three setae.

DNA-barcode: IE29 (BiH, Olovo, Krivajevići, Bijambare; type locality)

```
1        10        20        30        40        50
|        |         |         |         |         |
GAACTTTATATTTTTTATTAGGGGCCTGATCAGGAATAATTGGGACTTCC
CTAAGATTATTAATTCGGGCTGAATTAGGAAATCCAGGTAGTTTAATTAT
AAATGATCAAATTTACAATGTAATTGTCACAGCTCATGCTTTTATTATAA
TTTTTTTTATAGTTATACCAATTATAATTGGAGGATTTGGAAACTGATTA
GTTCCTTTAATATTAGGGGCTCCTGATATAGCTTTCCCGCGTATAAATAA
TATAAGATTTTGATTATTGCCTCCGTCTCTTACTTTATTACTTATAAGAA
GAATAGTAGAAAGTGGAGCAGGGACAGGATGAACAGTCTATCCTCCTTTA
TCAGCTAATATTGCTCATAGTGGTCCATCAGTAGATTTAGCAATCTTTAG
CCTTCATTTAGCTGGAATCTCTTCTATTTTAGGGGCAGTAAATTTTATCA
CTACTATTATTAATATGCGATCTCCTGGCATATCTTTTGATAAAATACCA
TTATTTGTCTGATCTGTAGCTATTACTGCCTTATTATTATTACTTTCATT
ACCAGTACTTGCTGGTGCAATTACTATATTATTAACAGATCGAAATTTAA
ATACTTCTTTTTTTGACCCAGCAGGGGGTGGAGACCCTATTTTATACCAG
CATTTATTT
```

Remarks: In all species of the genus *Anthroherpon*, the transversal carina on the posterior margin of the mesoventrum has a mesoventral processus. Only in *A. stenocephalum stenocephalum*, an intrapopulational variability of this character has been noted: some specimens have a mesoventral processus between the mesocoxae while it is absent in others (Fig. 14). This structure is, however, absent in all specimens of *A. stenocephalum noesskei* that I have examined.

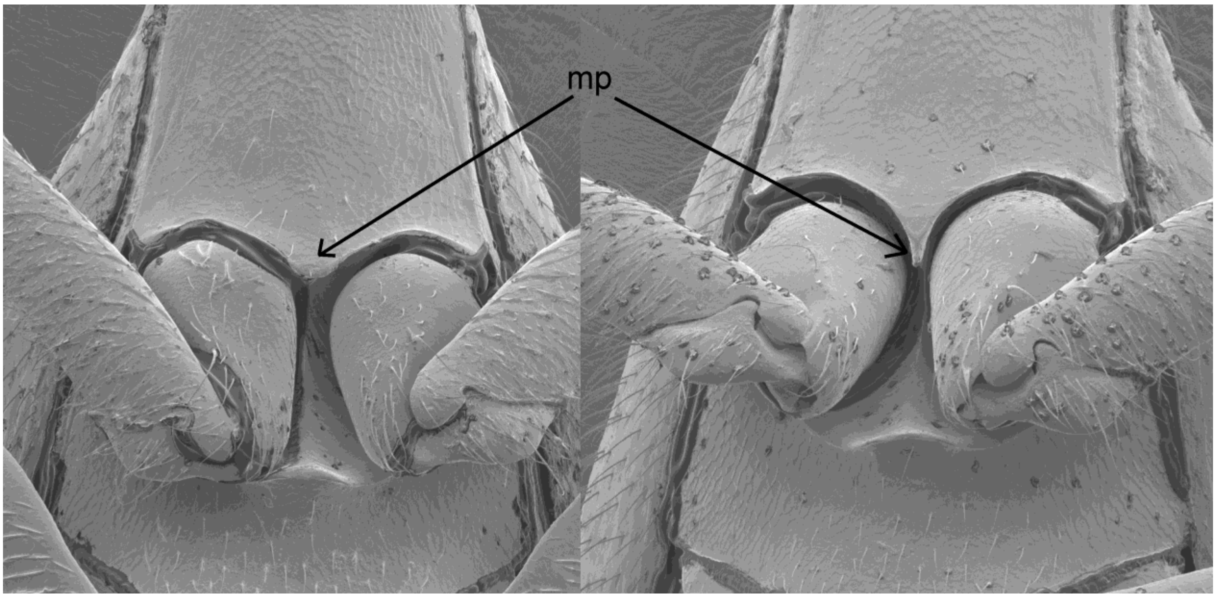

Fig. 14. Intra-populational differences in the length of mesoventral processus in *A. stenocephalum stenocephalum.*

19. ***Anthroherpon weiratheri*** **(Reitter, 1913)**

Antroherpon Weiratheri Reitter, 1913: 154. Lct: „Höhle der Kječina stiena“

Type locality: Bosnia and Herzegovina, Sarajevo, Crepoljsko polje, Kečina Stena (=Pećina u Kečinoj Steni), 1445 m, N 43°55'53.16" E 18°29'0.72".

Distribution: BiH, Pale, village Sumbulovac, Orlovača, 950 m, N 43°52'59.46" E 18°35'3.53" (new locality).

Diagnosis: Body size from 3.5 to 4 mm. Distinct from *A. stenocephalum* by the following combination of characters: lateral sides of pronotum convex anterior of the constriction, and slightly divergent posteriorly in dorsal view, with a thin ridge on the constriction in the lateral view; pronotum dorsaly covered with sparse hairs; protibia slightly curved outwards in dorsal view; profemora thin; elytra less elongated; smaller body size. Aedeagus illustrated in Fig. 13F: robust, only slightly curved and very thick in the lateral view; median lobe longer than parameres, thick, with convex lateral sides in dorsal view, wide in the apical part, and without a thickened sclerotized strip along the length of the medioventral side of the median lobe; parameres very thin, strongly convex in dorsal view, with a constriction in the apical part, and bearing two very short setae.

DNA-barcode: IE21 (BiH, Sarajevo, Crepoljsko polje, Kečina Stena; type locality)

```
1        10        20        30        40        50
|        |         |         |         |         |
GTACTATATATTTTTTATTAGGAACTTGATCGGGAATAATTGGAACTTCA
TTAAGATTATTAATCCGAGCTGAATTAGGAAGCCCAGGAAGTTTAATTAT
AAATGATCAAATTTATAATGTTATTGTAACAGCTCATGCTTTCATTATAA
TTTTTTTTATAGTCATACCAATCATAATTGGAGGATTCGGAAATTGACTA
GTCCCATTAATATTAGGAGCCCCTGACATGGCATTTCCTCGTATAAATAA
CATAAGATTTTGACTATTACCCCCTTCTTTAATTTTATTACTTATAAGAA
GTATAGTAGAAAGAGGAGCAGGAACAGGATGAACTGTTTATCCACCATTA
TCTGCTAATATCGCTCATAGTGGTCCATCTGTAGACTTAGCAATTTTTAG
CCTTCATTTAGCTGGAATTTCTTCTATTTTAGGAGCAGTAAATTTTATTT
CTACTATCATTAATATACGATCTCCTGGTATATCTTTTGATAAAATACCT
TTATTTGTTTGATCGGTAGCTATTACTGCCTTATTACTATTACTTTCATT
ACCCGTATTAGCAGGAGCTATTACTATATTATTAACAGATCGAAATCTAA
ATACTTCTTTTTTTGACCCTGCTGGGGGTGGAGATCCAATTTTATACCAG
CACTTATTT
```

Remarks: This species has a peculiar morphology and some unusual features unique in the genus *Anthroherpon*. These include: presence of a ridge on the mesosternum (Fig. 14); and unusual antennae, slightly shorter than the body (antennae are longer than the body in all other species of *Anthroherpon*). Antennomeres 8, 9,10, and 11 very short; antenomere 8 very thin, only slightly longer than the 2nd; antennomere 7 very wide in the apical part. Genetic distances in barcoding region around 11 % are noted between *A. weiratheri* and *A. stenocephalum*, and also between *A. weiratheri* and *A. harbichi.*
Lives in sympatry with *A. harbichi* in both caves where it was collected.

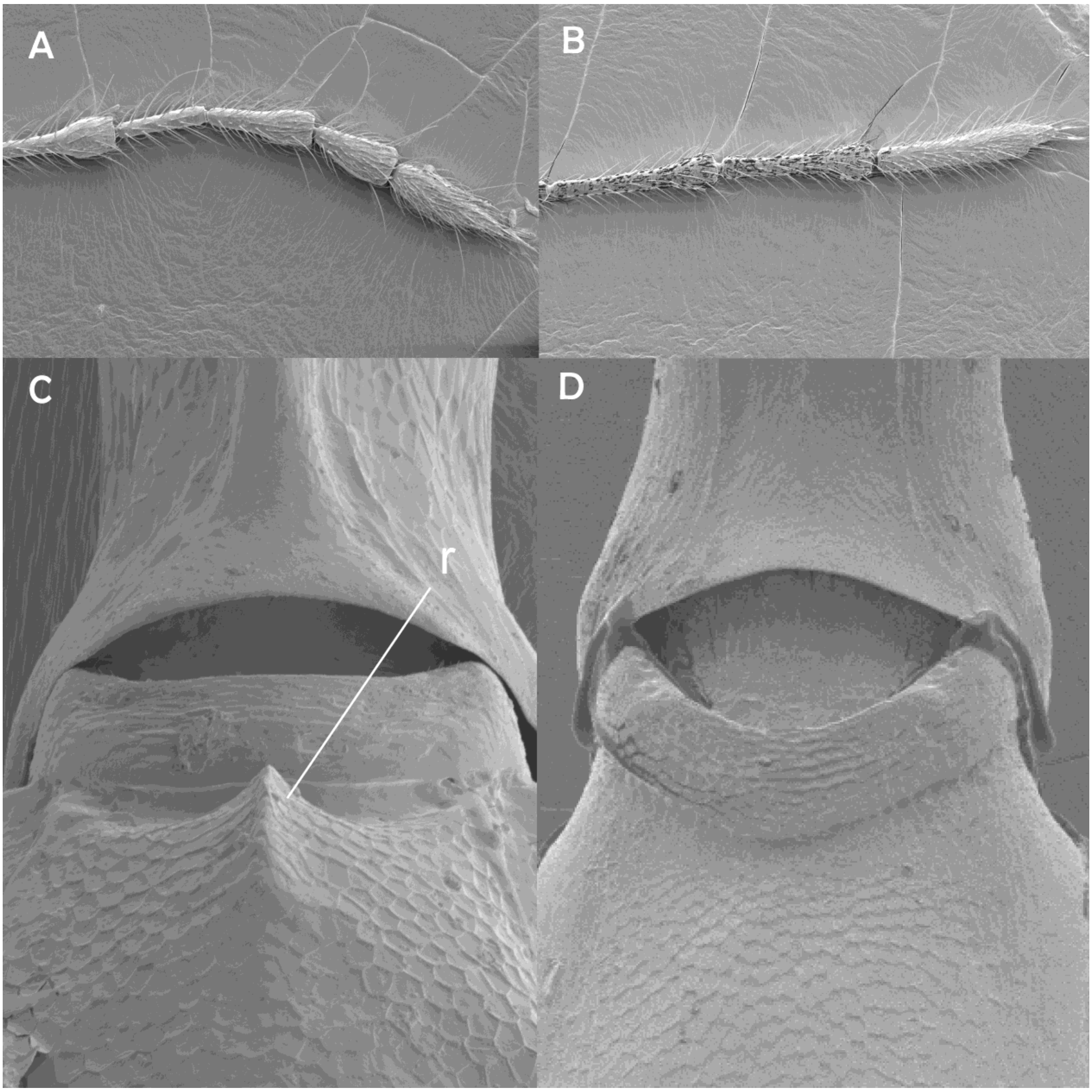

Fig. 15. A. *A. weiratheri*, antennomerae 7, 8, 9, 10, 11; B. *A. zariquieyi*, antennomerae 9, 10, 11; C. *A. weiratheri*, mesosternum with a ridge (r); D. *A. stenocephalum stenocephalum*, mesosternum without a ridge.

2.6.6. VI "*pygmaeum*" group V. B. Guéorguiev, 1990

18. ***Anthroherpon charon*** **(Reitter, 1911)**

Antroherpon Charon Reitter, 1911: 173. Lct: "Grotte bei Luki, Lipa und Pavlovac, im Gebirgsstocke der Joharina"

Type locality: Bosnia and Herzegovina, Trebević Mt., village Luke, Ledenica, 1146 m, N 43°47'38.16" E 18°29'26.40".

Distribution: BiH, caves on Trebević Mt. and Jahorina Mt.: Pašina dolina; Dasidoli, 8 km SSO from Pale (=Kasidol, Bosnia) (Guéorguiev, 1990); Pale, Omladinska pećina, 1326 m, N 43°46'13.08" E 18°36'37.80"; cave between Sarajevo and Pale, 872 m, N 43°51'2.32" E 18°32'19.91".

Diagnosis: Habitus illustrated in Fig. 10L. Body size 4.3 – 4.6 mm. Distinct from other species of the group by the following combination of characters: constriction of the pronotum less pronounced, lateral sides almost parallel posterior of the constriction, dorsal surface of the pronotum with very short and sparse hairs; elytra elongated, narrow in the anterior part, with the largest width anterior of the mid-section, with superficial and indistinct punctuation; aedeagus curved in an „S" shape in lateral view. Aedeagus illustrated in Fig. 13A. Median lobe longer than parameres (setae excluded), with slightly drown-out apex, and with the thickened sclerotized strip along the length of the medioventral side of the median lobe; parameres only in the apical part strongly curved inwards, bearing three setae.

DNA-barcode: IB2 (BiH, Trebević Mt., village Luke, Ledenica; type locality)

```
1        10        20        30        40        50
|        |         |         |         |         |
GTACTTTATATTTTTTACTAGGAGCTTGATCAGGAATAATTGGGACCTCA
TTAAGACTATTAATTCGAGCTGAATTAGGAAATCCAGGAAGTTTAATCAT
AAATGATCAAATTTACAATGTTATTGTAACTGCTCATGCTTTTATTATAA
TTTTTTTTATAGTTATACCAATTATAATTGGAGGATTTGGAAATTGATTA
ATTCCTTTAATATTAGGGGCCCCTGATATAGCTTTCCCGCGTATAAATAA
TATAAGATTTTGACTATTACCCCCTTCTTTAACTTTATTACTTATAAGAA
GTATAGTAGAAAGAGGGGCTGGAACGGGATGAACTGTTTATCCCCCCTTA
TCAGCTAATATCGCTCATAGCGGCCCATCAGTAGATTTAGCAATTTTTAG
CCTCCATTTAGCCGGAATTTCTTCTATTTTAGGTGCAGTAAACTTTATTA
CTACTATTATAAATATACGATCTCCTGGTATATCTTTTGATAAAATACCA
TTATTTGTATGATCTGTGGCTATTACTGCCCTATTATTATTACTTTCATT
GCCTGTATTAGCTGGTGCAATTACTATGCTATTAACAGATCGAAATTTAA
ACACTTCTTTTTTTGACCCTGCGGGGGGAGGAGACCCTATTTTATATCAA
CATTTGTTT
```

19. *Anthroherpon erebus* (Breit, 1913)

Anthroherpon erebus erebus (Breit, 1913)

Antroherpon Erebus Breit, 1913: 237. Lct: "In einer namenlosen Höhle im Bjelasnicagebiet"
= *Antroherpon affinis* Breit, 1913

Type locality: Bosnia and Herzegovina, Treskavica Mt., Unnamed cave.

Distribution: caves on Treskavica Mt. in BiH: Jama alpinska No 2 (Guéorguiev, 1990).

Anthroherpon erebus scheibeli (Jeannel, 1924)

Antroherpon (s. str.) *Erebus* Subsp. Scheibeli Jeannel, 1924: 418. Lct: "Bosnie: grotte de Trnovo, vallée de la Zeljeznica, au pied du Treskavica"

Type locality: Bosnia and Herzegovina, Trnovo, cave in the valley of Željeznica river.

Distribution: BiH, Trnovo, Bjeličina pećina, 990 m, N 43°40'56.28" E 18°27'35.94" (new locality).

Diagnosis: Distinct from other species of the group by the following combination of characters: pronotum with less pronounced constriction, lateral sides slightly diverging posterior of the constriction (in dorsal view), and with few, extremely short and thin hairs on the dorsal surface; elytra with clear punctuation, short hairs, and rhomboid shape.

Aedeagus illutrated in Fig. 13B. Median lobe robust, wedge-shaped, longer than parameres (setae excluded), with rounded apex, and with a thickened sclerotized strip along the length of the medioventral side of the median lobe; parameres very curved in dorsal view and particularly in the apical part, bearing two apical setae.

DNA-barcode: VA5 (BiH, Trnovo, Bjeličina pećina)

```
1        10        20        30        40        50
|        |         |         |         |         |
GTACTTTATATTTTTTATTAGGAACTTGATCAGGAATAATTGGAACCTCA
TTAAGGCTATTAATTCGGGCTGAATTAGGAAATCCAGGAAGTTTAATCAT
AAATGATCAAATTTACAATGTTATTGTAACTGCTCATGCTTTTATTATAA
TCTTTTTTATAGTTATACCAATTATAATTGGAGGATTTGGAAATTGATTA
ATTCCTTTAATGTTAGGAGCTCCTGATATAGCTTTCCCTCGTATAAATAA
TATAAGATTTTGACTATTACCTCCTTCTTTAACTTTATTACTTATAAGAA
GTATAGTAGAAAGAGGAGCTGGAACGGGATGAACTGTTTATCCACCCTTA
TCAGCTAATATCGCTCATAGTGGTCCATCAGTAGATTTAGCAATTTTTAG
CCTTCATTTAGCCGGAATTTCTTCTATTTTAGGTGCAGTAAACTTTATTA
CTACTATTATTAATATACGATCTCCTGGTATATCTTTTGATAAAATACCA
TTATTTGTGTGATCTGTAGCTATTACTGCCTTATTATTATTACTTTCATT
ACCAGTATTAGCTGGTGCGATTACTATATTATTAACAGATCGAAATTTAA
ACACTTCTTTTTTTGACCCTGCAGGAGGAGGAGATCCTATTTTATACCAA
CATTTGTTT
```

Remarks: This subspecies is missing from the list of species of *Anthroherpon* given by Guéorguiev (1990) and Giachino & Guéorguiev (1993).

20. *Anthroherpon pozi* (Absolon, 1913)

Antroherpon poži Absolon, 1913: 1. Lct: “Sudbosnien: Eishöhle bei Kalinovik“

Type locality: Bosnia and Herzegovina, Kalinovik, Treskavica Mt., Ledenica.

Distribution: known only from the type locality.

Diagnosis: Habitus illustrated in Fig. 10M. Distinct from other species of the group by the following combination of characters: pronotum with pronounced constriction, lateral sides distinctively diverging posterior of the constriction, very rounded in the anterior part, and with sparse hairs; integument matte; elytra elongated, wide and broadly rounded in the posterior part, with short, dense hairs; surface matte, without punctuation but granulated; profemora thick. Aedeagus was not examined in this study.

21. *Anthroherpon pygmaeum* (Apfelbeck, 1889)

Anthroherpon pygmaeum pygmaeum (Apfelbeck, 1889)

Leptoderus pygmaeus Apfelbeck, 1889: 61. Lct: "Megara pećina u Preslici-planini (kotar Kenjički)"

Type locality: Bosnia and Herzegovina, Bjelašnica, Opančak, Megara pećina (=Mijatova pećina/Hajdučka špilja na Ivan planini/Kuvija), 1302 m, N 43°42'51.90" E 18° 5'4.62".

Distribution: known only from the type locality.

Diagnosis: Distinct from other species of the group by the following combination of characters: pronotum with pronounced constriction, lateral sides distinctively diverging posterior of the constriction, less rounded in the anterior part, with smooth and shiny dorsal surface; elytra elliptic, surface shiny, with wide but shallow and sparse punctuation and long hairs. Aedeagus was not examined in this study.

Remarks: I visited Megara on June 15th, 2013, with a team of five biospeleologists but we failed to find this species. The cave has undergone extreme devastation, particulatly in the Final hall, due to illegal excavation of cave bear skeletal remains (Bilela et al., 2013). It is possible that this activity had a negative impact on the cave fauna but this has never been investigated. However, it shoud be noted that we collected 10 specimens of *Apholeuonus longicollis* Reitter, 1904 which is also a troglobite of the tribe Leptodirini.

Anthroherpon pygmaeum stricticolle (Jeannel, 1930)

Antroherpon (s. str.) *pygmaeum* subsp. *stricticolle* Jeannel, 1930: 144. Lct: "Bosnie méridionale: Sudarova pećina sur le plateau de Radopolje, partie occidentale du Bjelašnica planina"

Type locality: Bosnia and Herzegovina, Bjelašnica, Radopolje, Sudarova pećina (=Serdareva pećina), 1301 m, N 43°41'6.84" E 18° 4'58.74".

Distribution: Bosnia and Herzegovina, Bjelašnica Mt.: Jama u Observatorije, Bezimenska jama, Čančarica jama, (Guéorguiev, 1990); Opančak, Ledenjača, 1301 m, N 43°43'14.46" E 18°6'5.64".

DNA-barcode: VA35 (BiH, Bjelašnica, Sudarova pećina; type locality)

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 1        10        20        30        40        50
```

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|        |         |         |         |         |
GTACTTTATATTTTTTATTGGGAACTTGATCAGGAATAATTGGAACATCA
TTAAGATTATTAATTCGAGCTGAGCTAGGAAACCCAGGAAGCTTAATTAT
AAATGATCAAATTTATAATGTTATTGTAACAGCTCATGCTTTTATTATAA
TCTTTTTTATAGTTATACCAATCATAATTGGAGGATTCGGAAATTGATTA
GTTCCTTTAATATTAGGAGCCCCTGACATAGCTTTTCCACGTATAAATAA
TATAAGATTTTGATTATTACCCCCATCTTTAACTTTATTACTAATAAGAA
GTATAGTAGAAAGAGGGGCTGGAACAGGATGAACTGTATACCCTCCCTTA
TCAGCCAATATTGCTCATAGAGGTCCATCAGTAGATTTAGCAATTTTTAG
TCTTCATCTAGCAGGAATCTCTTCTATTTTAGGAGCAGTAAATTTTATTA
CTACTATTATTAATATACGATCTCCTGGCATATCTTTTGATAAAATACCA
TTATTTGTGTGATCAGTAGCTATTACAGCCTTATTATTATTACTTTCATT
ACCGGTATTAGCTGGTGCTATTACAATACTATTAACAGATCGAAATTTAA
ATACTTCTTTTTTTGACCCTGCGGGGGGAGGAGATCCTATTTTATACCAA
CATTTATTT
```

Remarks: Aedeagus illustrated in Fig. 13C. Median lobe much shorter than parameres, with drown-out apex, and without the thickened sclerotized strip along the length of the medioventral side of the median lobe; parameres thicker and strongly convex in the apical part, bearing three setae.

22. *Anthroherpon subalpinum* (Jeannel, 1924)

Antroherpon subalpinum Jeannel, 1924: 418. Lct: "grotte de Stambulčić, massif du Kodža planina"

Type locality: Bosnia and Herzegovina, Stambulčić, Jahorina Mt., Kodža, unnamed cave.

Ditribution: known only from the type locality.

Diagnosis: Body length 4.2–4.5 mm. Distinct from other species of the group by the following combination of characters: pronotum with less pronounced constriction, less rounded anterior of the constriction and with slightly convex and slightly divergent lateral sides posterior of the constriction; elytra elongated, widest in the mid-section; profemora thick, protibia curved outwards in dorsal view; aedeagus not curved in „*S*" shape in lateral view.

23. *Anthroherpon winneguthi* (Apfelbeck, 1919)

Antroherpon Winneguthi Apfelbeck, 1919: 271. Lct: "in einer kleiner Höhle bei Pale (bezirk Sarajevo)"

Type locality: Bosnia and Herzegovina, Pale, unnamed cave near Pale.

Distribution: known only from the type locality.

Diagnosis: Body length 3.8 mm. Pronotum short, lateral sides anterior of the constriction very convex. Constriction not very pronounced, but more so than in *A. erebus*; elytra short, with broadly rounded apex in dorsal view, and convex dorsal surface in lateral view, with superficial punctuation and short hairs. Aedeagus was not examined in this study.

24. *Anthroherpon zariquieyi* (Jeannel, 1930)

Antroherpon (s. str.) *Zariquieyi* Jeannel, 1930: 145. Lct: "Massif du Durmitor, nfw. Nade pećina"

Type locality: Montenegro, Durmitor Mt., Savin kuk, Vodeni do, Vodena pećina, 1684 m, N 43°7'1.56" E 19°5'42.00".

Distribution: MNE, Durmitor Mt.: Sedlo, Pećina u Sedlenoj gredi, 2031 m, N 43°5'37.55" E 19°3'15.34"; Surutka, Jama na Vjeterenim brdima; Zeleni vir, Zelenovirska pećina; Obla glava, Ledena pećina, 2184 m (Pavićević, 2016, pers. comm.).

Diagnosis: Habitus illustrated in Fig. 10N. Body size 4.8– 5.1 mm. Distinct from other species of the group by the following combination of characters: integument dark reddish-brown; pronotum less convex in the anterior part, with a pronounced constriction, lateral sides distinctively diverging posterior of the constriction (in dorsal view), with short and sparse hairs on the dorsal surface; mesothoracic pedunculus long and constricted in the mid-section in dorsal view; elytra elongated, wide and broadly rounded in the posterior part, surface matte, without punctuation but granulated, and with short, very dense hairs; profemora thin. Aedeagus illustrated in Fig. 13D. Median lobe longer than parameres (setae excluded), thin in the posterior and wide and triangular in the anterior part, with lanceolate apex; parameres thin, slightly convex, bearing three medium-long setae in the apical part.

DNA-barcode: VA13 (MNE, Durmitor Mt., Sedlo, Pećina u Sedlenoj gredi)

```
1        10        20        30        40        50
|        |         |         |         |         |
GTACTTTATATTTCTTACTAGGAGCTTGATCAGGAATAATTGGAACATCA
TTAAGATTACTAATTCGAGCTGAATTAGGAAATCCAGGAAGTTTAATCAT
AAACGATCAAATTTATAATGTTATTGTAACAGCTCATGCTTTTATTATAA
TTTTTTTTATAGTTATGCCAATTATAATTGGAGGTTTTGGAAATTGACTA
GTTCCATTAATATTAGGAGCCCCTGATATAGCTTTCCCTCGTATAAATAA
TATAAGATTTTGATTATTACCTCCTTCCCTAACCTTACTTCTTATAAGAA
GAATAGTAGAAAGCGGAGCTGGAACAGGATGAACTGTTTATCCCCCTTTA
TCTGCTAATATCGCTCATAGTGGCCCATCAGTAGACTTAGCTATTTTTAG
CCTTCATTTAGCTGGAATCTCCTCTATTTTAGGAGCAGTAAATTTTATTA
CCACTATTATTAATATACGATCTCCTGGTATATCTTTTGATAAAATACCA
CTATTTGTATGATCTGTAGCTATTACTGCCCTATTATTATTACTTTCTTT
GCCAGTATTAGCTGGTGCAATTACCATATTATTAACAGACCGTAACTTAA
ATACATCTTTTTTTGATCCCGCAGGAGGCGGAGACCCAATTTTATATCAA
CATTTATTT
```

Remarks: The large genetic distances in the barcoding region between different species of this group (Table 3) indicate that the classification is probably artificial. All species of the „*pygmaeum*“ group except *A. zariquieyi* are distributed in Bosnia and Herzegovina, on the northeastern part of the distribution area of the genus, while *A. zariquieyi* is located in Montenegro, and more towards the southwest and about 80 km far from the nearest species of this group, so it is very likely that this species doesn't belong to this species group.

Table 3. Genetic distances in the barcoding region between species of the „*pygmaeum*“ species group.

	A. zariquieyi	*A. pygmaeum stricticolle*	*A. erebus scheibeli*	*A. charon*
A. zariquieyi		10.9 %	9 %	8.8 %
A. pygmaeum stricticolle	10.9 %		7.4 %	8.3 %
A. erebus scheibeli	9 %	7.4 %		4.4 %
A. charon	8.8 %	8.3 %	4.4 %	

In 1991, during speleological expedition on Durmitor jointly organized by the Montenegrin Academy of Sciences and Arts and the Yugoslav Entomological Society, *A. zariquieyi* was found in the deepest pit on Durmitor– Jama na Vjetrenim Brdima, on the deepth of -700 m (Pavićević, 2016, pers. comm.). No other *Anthroherpon* has ever been found as deep as this. Caves and pits on Durmitor are located on the altitude of more than 2000 m and together with caves on Prokeltije Mt. present the greatest elevation of all *Anthroherpon* inhabited caves.

During the exploration of Ledena pećina, a cave with permanent ice stalactites and stalagmites, located underneath Obla Glava, an interesting observation was made by a biospeleologist S. Ognjenović– he found one specimen of *A. zariquieyi* under the thick layer of ice after breaking the ice with a hack.

2.6.7. VII "*hoermanni*" group V. B. Guéorguiev, 1990

25. ***Anthroherpon hoermanni*** **(Apfelbeck, 1889)**

Anthroherpon hoermanni hoermanni **(Apfelbeck, 1889)**

Leptoderus Hoermanni Apfelbeck, 1889: 62. Lct: "Ustaška pećina kod Krbljine“
= *Anthroherpon hörmanni* Jeannel, 1910
= *Antroherpon Bokori* Csiki, 1912: 512. synonymy in Jeannel, 1924: 369
Lct: "Bosnia: in antro prope Županjac“

Type locality: Bosnia and Herzegovina, Dobro polje, Ustaška pećina (=Insurgenten Höhle), 1050 m, N 43°35'32'' E 18°29'59''.

Distribution: Numerous caves on Treskavica Mt. and in the vicinity of Kalinovik in eastern Bosnia and Herzegovina (Reitter, 1906; Jeannel, 1911, 1924; Hoffman, 1929; Pretner, 1969; Guéorguiev, 1990, Perreau, 2000): Podkraj pećina; Ledenica; Kalinovik, pećina u Hotovlje (Guéorguiev, 1990); Kalinovik, Borija, Borija pećina (=Pećina u Glavičinama), 1170 m, N 43°29'55.68" E 18°32'57.96"; BiH, Nedavić, Vareničina pećina, 1056m, N 43°26'29.76" E 18°19'36.06" (new locality).

Diagnosis: Habitus illustrated in Fig. 10O. Body length 6.2–7 mm. Dorsal surface of pronotum (in lateral view) with a depression in the posterior part. Pronotum 2 times longer than wide (measured at the widest section), widest and rounded in the anterior third, and with a strong constriction in the posterior third; Mesothoracic pedunculus elongated, approximately 2 times wider than long (measured at narrowest section), narrowest in the anterior part. Elytra narrow, widest slightly posterior of the middle, with short and dense hairs, and distinct punctuation. Aedeagus illustrated in Fig. 11C; curved under an almost 90° angle in lateral view, median lobe very thin in the anterior third of its length and curved in an „S“ shape (in lateral view); median lobe in dorsal view much shorter than parameres, with triangular apex, and without a thickened sclerotized strip along the length of the medioventral side of the median lobe; parameres of uniform width in full length, with three apical setae, and in the upper position of the median lobe in the lateral view.

DNA-barcode: IB7 (BiH, Kalinovik, Borija, Borija pećina):

```
1        10        20        30        40        50
|        |         |         |         |         |
GCACTTTATACTTCTTATTAGGAACTTGATCAGGAATGATTGGAACCTCA
CTAAGACTATTAATTCGGGCAGAATTAGGAAATCCCGGTAGATTAATTAT
AAATGATCAAATTTACAATGTTATTGTAACAGCCCACGCTTTTATTATAA
TTTTTTTCATGGTCATACCTATTATAATTGGAGGATTTGGAAATTGATTA
GTTCCATTAATATTAGGAGCCCCTGACATGGCTTTTCCCCGTATAAATAA
TATAAGATTTTGACTATTACCTCCTTCTTTAACCCTTCTACTTATAAGAA
GCATAGTAGAAAGGGGAGCTGGAACAGGATGAACTGTTTATCCCCCTTTA
TCGGCCAATATTGCCCACAGTGGCCCATCAGTAGATTTAGCCATTTTTAG
ATTACACTTAGCTGGAATCTCTTCTATTTTAGGAGCAGTAAATTTTATTA
CTACTATTATTAATATACGATCTCCCGGAATATCTTTCGATAAAATACCC
TTATTTGTATGGTCTGTAGCTATTACTGCTCTATTATTACTACTATCATT
ACCCGTACTTGCTGGGGCAATTACTATACTATTAACAGATCGTAACCTAA
ATACTTCTTTTTTTGACCCCGCAGGAGGTGGAGACCCGATTTTATACCAA
CATTTATTT
```

Anthroherpon hoermanni hoffmanni **Giachino & V. B. Guéorguiev, 1993**

Antroherpon hoermanni hoffmanni Giachino & V. B. Guéorguiev, 1993: 310. Lct: "Bosnia, Visocica planina"

Type locality: Bosnia and Herzegovina, Visočica Mt.

Distribution: known only from the type locality.

Anthroherpon hoermanni hypsophilum **(Apfelbeck, 1907)**

Antroherpon Hoermanni hypsophilum Apfelbeck, 1907: 402. Lct: "in antro montis Lebršnik"

Type locality: Bosnia and Herzegovina, Lebršnik Mt., Vilina pećina.

Distribution: Caves on Lebršnik Mt. and Volujak Mt., on the border of Bosnia and Herzegovina and Montenegro: BiH, Lebršnik Mt., Đatlo, 1547 m, N 43°12'14.04" E 18°38'4.32"; Volujak, Jama u Marcelov do (Guéorguiev, 1990); Dvogrla jama, Čavčarica jama, 1675 m.

DNA-barcode: VA36 (BiH, Lebršnik, Đatlo)

```
1        10         20         30         40         50
|        |          |          |          |          |
GTACTCTATACTTCTTACTGGGAACTTGATCAGGAATAATTGGAACCTCG
CTAAGACTATTAATTCGGGCAGAATTAGGAAATCCCGGCAGGTTAATTAT
AAACGATCAAATTTATAATGTTATTGTAACCGCCCACGCTTTCATTATAA
TTTTTTTTATGGTCATGCCTATTATAATTGGAGGATTTGGAAACTGATTA
GTTCCACTAATATTAGGGGCTCCTGATATGGCTTTCCCCCGTATAAATAA
TATAAGATTTTGACTATTACCACCTTCTTTAACCCTTTTACTTATAAGAA
GTATGGTAGAAAGGGGGGCCGGAACAGGGTGAACTGTTTATCCCCCCCTA
TCTGCTAACATTGCCCATAGCGGACCCTCAGTAGATTTGGCTATTTTTAG
GCTACATCTAGCTGGAATCTCTTCTATTTTAGGAGCAGTAAATTTTATTA
CCACTATTATTAATATGCGATCTCCTGGCATATCCTTTGATAAAATACCT
TTATTTGTATGGTCTGTAGCTATCACTGCCCTATTACTACTACTCTCACT
TCCGGTATTAGCTGGAGCAATTACTATACTATTAACAGACCGTAATCTAA
ATACTTCTTTTTTTGACCCTGCAGGAGGTGGAGACCCAATCTTATATCAA
CACTTATTC
```

Anthroherpon hoermanni orlovacensis **(V. B. Guéorguiev, 1990)**

Antroherpon hoermanni orlovacensis V. B. Guéorguiev, 1990: 264. Lct: "Montenegro, Orlovac Geb., Teufelshöhle (=Šejtana pećina)"

Type locality: Montenegro, Maglić Mt., Orlovac, Šejtan jama (=Šejtan pećina; Devil's cave; Orlova cave).

Distribution: Maglić Mt. on the borded of BiH and MNE. Montenegro, Maglić, Trnovačko jezero, Katuni, Pećina iznad katuna, N 43°14'47.29" E 18°42'50.53", 1779 m (new locality).

DNA-barcode: IE50 (MNE, Maglić, Trnovačko jezero, Katuni, Pećina iznad katuna):

```
1        10        20        30        40        50
|        |         |         |         |         |
GCACTCTATACTTCTTACTAGGAACTTGATCAGGAATAATTGGAACCTCA
CTAAGACTATTAATTCGGGCAGAATTAGGAAACCCAGGTAGGTTAATTAT
AAATGATCAAATTTATAATGTTATTGTAACAGCCCACGCTTTCATTATAA
TTTTTTTTATGGTGATACCTATTATAATTGGAGGATTTGGAAACTGATTA
GTTCCGTTAATATTAGGGGCCCCTGATATGGCTTTCCCCCGTATAAATAA
TATAAGATTTTGACTATTACCCCCTTCTTTAACCCTTTTACTTATAAGGA
GCATAGTAGAAAGGGGAGCCGGGACAGGATGAACTGTTTATCCTCCCCTA
TCTGCCAACATTGCCCATAGTGGGCCATCAGTAGATTTAGCTATTTTTAG
GCTACACTTAGCTGGAATCTCTTCTATTTTAGGGGCAGTAAATTTTATTA
CTACTATTATTAATATACGATCTCCTGGCATATCCTTTGATAAAATACCT
TTATTTGTATGGTCTGTAGCTATTACTGCCTTATTACTACTACTCTCACT
TCCGGTATTAGCAGGAGCAATTACTATGCTATTAACAGACCGGAATCTAA
ATACTTCTTTTTTTGACCCTGCGGGAGGCGGAGACCCAATCTTAT
```

***Anthroherpon hoermanni sericeum* (Jeannel, 1930)**

Antroherpon Hörmanni ssp. *sericeum* Jeannel, 1930:146. Lct: "Oede pećina"

Type locality: Bosnia and Herzegovina, Zelengora Mt., Jabuka, Jabučke stijene, Pećina u Jabučkim stijenama.

Distribution: Bosnia and Herzegovina, caves on Zelengora Mt.: Zelengora, Pećina na Bregoču, 1544 m, N 43°21'18.62" E 18°34'46.24".

DNA-barcode: IR49 (BiH, Zelengora, Pećina na Bregoču):

```
1        10        20        30        40        50
|        |         |         |         |         |
GTACTTTATACTTCTTATTGGGAACTTGATCAGGAATAATTGGAACCTCT
CTAAGACTACTAATTCGAGCAGAATTAGGAAACCCCGGTAGACTAATTAT
AAATGATCAAATTTACAATGTTATTGTAACAGCCCACGCTTTTATTATAA
TTTTTTTTATGGTCATACCTATTATAATTGGGGGGTTTGGAAATTGATTA
GTTCCGTTAATATTAGGAGCTCCTGATATGGCTTTCCCTCGTATAAATAA
TATAAGATTTTGACTACTACCTCCTTCTTTAACCCTTCTACTTATAAGAA
GTATGGTAGAAAGGGGAGCTGGAACAGGATGAACTGTTTACCCCCCTTTG
TCAGCTAATATTGCTCACAGTGGCCCATCAGTAGATTTAGCCATTTTTAG
GCTACACTTAGCTGGAATCTCTTCTATTTTAGGGGCAGTAAATTTTATTA
CTACAATTATTAATATACGATCTCCTGGTATATCTTTCGATAAAATACCC
TTATTTGTATGATCTGTGGCTATTACTGCCTTATTATTACTACTCTCATT
ACCAGTACTTGCCGGAGCAATTACTATACTATTAACAGACCGTAATCTAA
ACACTTCTTTTTTCGACCCCGCTGGGGGTGGGGATCCAATTTTATACCAA
CATTTATTT
```

Remarks on *A. hoermanni* senso lato: High genetic distances in barcoding region (Table 4) indicate that three subspecies: *A. hoermanni sericeum, A. hoermanni hypsophylum,* and *A. hoermanni orlovacensis* could be elevated to species level. *A. hoermanni sericeum* differs from other *A. hoermanni* subspecies by the following combination of characters: body size 5.9 to 6 mm; lateral sides of the pronotum less convex in the anterior part and distinctively diverging posterior of the (very narrow) constriction; elytra elliptic, short, widest in the mid-section, with distinct punctuation, and short hairs. Aedeagus illustrated in Fig. 11E. Median lobe much shorter than parameres, thin and elongated and slightly thinner in the middle than in the apical part, and with lanceolate apex; parameres convex, slightly thicker in the appical part, and bearing three setae: two long and one short. *A. hoermanni hypsophylum* differs from other *A. hoermanni* subspecies by the following combination of characters: lateral sides of pronotum less convex in the anterior part; elytra with superficial and indistinctive punctuation. *A. hoermanni orlovacensis* differs from other *A. hoermanni* subspecies by the following combination of characters: lateral sides of pronotum very convex in the anterior part; elytra with superficial and indistinctive punctuation.

Table 4. Genetic distances in barcoding region between *A. hoermanni* subspecies.

	A. hoermanni sericeum	*A. hoermanni hoermanni*	*A. hoermanni hypsophilum*	*A.hoermanni orlovacensis*
A. hoermanni sericeum		7.1 %	10.9 %	10 %
A. hoermanni hoermanni	7.1 %		10.2 %	8.6 %
A. hoermanni hypsophilum	10.9 %	10.2 %		5.9 %
A.hoermanni orlovacensis	10 %	8.6 %	5.9%	

26. *Anthroherpon scutariensis* Giachino & V. B. Guéorguiev, 1993

Antroherpon scutariensis Giachino & V. B. Guéorguiev, 1993: 308. Lct: “Albania, Scutari“

Type locality: Albania, Scutari (=Skadarsko jezero); the exact locality is unknown.

Distribution: known only from the type locality.

Remarks: Species was not examined in this study.

2.7. ARTICLE 1: Two new species of the genus *Anthroherpon* Reitter, 1889 from northern Montenegro with notes on the "*A. ganglbaueri*" species group (Coleoptera: Leiodidae: Leptodirini)

by Iva Njunjić (1,2), Michel Perreau (3) & Dragan Pavićević (4)

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Abstract:

Two new species of *Anthroherpon* belonging to the "*ganglbaueri*" species group are described from northern Montenegro: *Anthroherpon sinjajevina* n. sp. and *Anthroherpon cecai* n. sp. Based on morphological investigations of other species of this group, we elevate *A. udrzali* Giachino & Vailati from subspecies to species rank, and suggest the synonymy of *A. brckoensis* Giachino & Guéorguiev with *A ganglbaueri alticola* Knirsch. An identification key of the *ganglbaueri* species group (*sensu* Guéorguiev, 1990) is given and the distributions of species and subspecies are mapped and discussed.

Key words: Coleoptera, Anthroherponina, Leptodirini, *Anthroherpon, sinjajevina* sp. n, *cecai* sp. n, troglobitic species, taxonomy, Montenegro.

Introduction:

The genus *Anthroherpon* Reitter, 1889 belongs to the tribe Leptodirini, subtribe Anthroherponina which was recently revised (Perreau & Pavićević, 2008). The genus comprises 27 species and 62 subspecies (Perreau, 2004) divided into seven species groups (Guéorguiev, 1990). Most of species have been described in the late XIXth century or the first half of the XXth century (Apfelbeck, 1889; 1894; 1907; Reitter, 1903; 1908; 1911; 1913; Müller, 1910; Matcha, 1916; Jeannel, 1924; 1930; 1934; 1947; Winkler, 1925; 1938; Knirsch, 1927, 1929; Zariquiey, 1927). After a break of several decades due to political conditions, new collections were performed and several new species and subspecies were subsequently described (Giachino & Guéorguiev, 1993; Giachino & Vailati, 2005). Synthetic revisions of *Anthroherpon* have been undertaken by Jeannel (1924, 1930) and Guéorguiev (1990).

The genus is widely distributed in subterranean habitats of the southern Dinaric range: South Croatia, Bosnia and Herzegovina, Montenegro, south Serbia and north Albania. This European region is known as the major hotspot of biodiversity in the world for subterranean fauna, not only for Coleoptera but also for most of zoological groups (Deharveng & al., 2012). The genus *Anthroherpon* shows the most pronounced troglobiomorphic morphological characters not only among Leptodirini but also among subterranean Coleoptera: extremely long appendages, extreme elongation of the fore-body (head and pronotum), hemispherical elytra, and complete anophthalmy.

The "*ganglbaueri*" species group was introduced by Guéorguiev (1990) from the reinterpretation of Jeannel's "sectio I" and "sectio II" (Jeannel, 1924). The group is defined by the following set of characters: pronotum without annular constriction in the last third of its length, sides of pronotum sinuate in the basal half, protarsi more than half as long as protibiae, profemorae not enlarged in the basal parts, last antennomere longer than the penultimate and body length above 5 mm. Presently it contains four species: *A. brckoensis* Giachino & Guéorguiev, 1993; *A. ganglbaueri* Apfelbeck, 1894, with four subspecies (including *alticola* Knirsch, 1927, *distinguendum* Müller, 1913; *intermedium* Winkler, 1938); *A. matulici* Reitter, 1903; *A. matzenaueri* Apfelbeck, 1907 with four subspecies (including *augustae* Zariquiey, 1927; *taliensis* Zariquey, 1927); *A. udrzali* Giachino & Vailati, 2005. The monophyly of this group is not formally established, just as the other species groups of the genus, since the phylogeny of *Anthroherpon* is poorly understood. A detailed investigation of the phylogeny of *Anthroherpon*, combining morphological and molecular methods, is currently being undertaken by one author of this paper (I. Njunjić).

This paper gives a short overview on the *ganglbaueri* species group, excluding the subspecies, describing two new species: *A. sinjajevina* and *A. cecai*, raising *A. udrzali* from subspecies of *A. matzenaueri* to species rank, and suggesting the new synonymy of *A. brckoensis* with *A. ganglbaueri alticola*.

Material and methods

The external morphology of specimens was examined using Leica MZ 75 and Leica M10 stereomicroscopes. Male and female genitalia were cleared in KOH 0.1 N, female genitalia have been stained with Azoblack (Carayon, 1969), both sexes mounted in Euparal on glass slides, and fixed on the pin under the specimen. Microphotographs were taken on a Leica DIAPLAN or a Zeiss AXIOLAB microscope with a camera diagnostic instrument Spot INSIGHT IN1820. Macrophotographs were taken using a Nikon SMZ 1000 stereomicroscope with a Canon EOS 60D camera (Fig. 1) and a digital microscope Keyence Z20 (Fig. 2). Digital pictures were processed using the Helicon Focus software 5.3 (http://www.heliconsoft.com/heliconsoft-products/helicon-focus/) (except Fig. 2). Scanning electron microscopy was performed at the National Museum of Natural History (Paris) on a Hitachi SU3500 after gold coating with a Jeol JFC 1200 sputter coater.

The following abbreviations for collections and institutions are used: CDPV: Dragan Pavićević Collection, Belgrade, Serbia; CINJ: I. Njunjić Collection, Belgrade, Serbia; CJLK, Jan Lakota (Ružomberok, Slovakia); CMPR: Michel Perreau Collection, Paris, France; MNHN: Muséum National d'Histoire Naturelle, Paris, France; NMPC: Entomologické Oddělení Národního Muzea, Prague, Czech Republic; MZBS: Museo de Zoologîa, Barcelona, Spain.

The following abbreviations for morphological details are used: HL: head length (measured from the posterior margin of clypeus); HW: maximum width of head; HL/HW: ratio length of head/maximum width of head; PL: pronotum length (measured along the median line); Pwmax: maximum width of pronotum (largest transverse width); Pwmin: minimum width of pronotum (shortest transverse width); EL: elytral length (as linear distance measured along the median line from base to the apex); EW: maximum width of elytra.

References to type material are abbreviated as HT: holotype; PT: paratype.

TAXONOMY

Anthroherpon sinjajevina **sp. n.**

Type material: Holotype male labeled as follows: "MONTENEGRO, Sinjajevina Mt, Njegovuđe, village Rudanca, Blažova pećina, 1470 m, 25.06.2013, leg. I. Njunjić" (white label, printed) / "HOLOTYPUS *Anthroherpon sinjajevina* sp.n. I. Njunjić, M. Perreau & D. Pavićević det. 2014." (red label, printed), (MNHN). Paratypes: 5♂ 10♀, 18.07.2010. (MNHN, CINJ, CMPR, CDPV), 7♂ and 12♀, same cave, 25.06.2013, leg. I. Njunjić (MNHN, CINJ, CMPR, CDPV). All paratypes are labeled with white, printed locality labels and with red printed labels "PARATYPUS *Anthroherpon sinjajevina* sp.n. I. Njunjić, M. Perreau & D. Pavićević det. 2014".

Figs. 1–2. Habitus of male. 1: *A. sinjajevina* n. sp.2: *A. cecai* n. sp. Scale bar is 1 mm.

Diagnosis: Habitus illustrated in Fig. 1. Distinct from the other species of the group by the following characters: second antennomere approximately two times shorter than the first one. Sides of pronotum convergent behind the anterior third of the pronotum length, the sides parallel on the basal fourth of the pronotum length. Elytral surface with microreticulation between punctures. Pronotum without symmetrical dorsal impressions. Parameres shorter than median lobe (setae excluded) with a constriction in the preapical region and with acute apex (Fig. 6c).

Description: Body length 5.6 to 5.9 mm (HT 5.71 mm). General morphology leptodiroid; head wider than pronotum, elytra physogastric; eyeless; wingless, and depigmented.

Head: hypognathous, wider in the anterior part and wider than the pronotum, HL 1.13-1.22 mm (HT 1.19 mm), HW 0.73-0.76 mm (HT 0.73 mm). Surface rough with evident microreticulation visible without scanning electron microsopy and with sparse and short pubescence. Penultimate maxillary palpomere approximately 1.2 times longer than the last one. Antennae 1.20 (males) or 1.05 (females) longer than the whole body. Second antennomere very short in comparison to the other antennomeres, approximately 2 times shorter than the first one. Last antennomere approximately 2 times (male) or 1.7 times (females) longer than the penultimate.

Table 2. Antennal formula of the holotype of *Anthroherpon sinjajevina* sp. n (mm).

Article	I	II	III	IV	V	VI	VII	VIII	IX	X	XI
Length	0.42	0.2	0.8	0.55	0.8	0.66	0.69	0.42	0.51	0.52	1.02

Pronotum: distinctly longer than wide with maximum width in anterior third, mean Pwmax/Pwmin: 1.6 (HT 1.61 mm). Sides convergent behind the anterior third of the pronotum length, the sides parallel on the basal fourth of the pronotum length. Dorsal surface, microreticulated and with a few punctures randomly disposed. Short and very sparse setae are randomly disposed mostly in the posterior part.

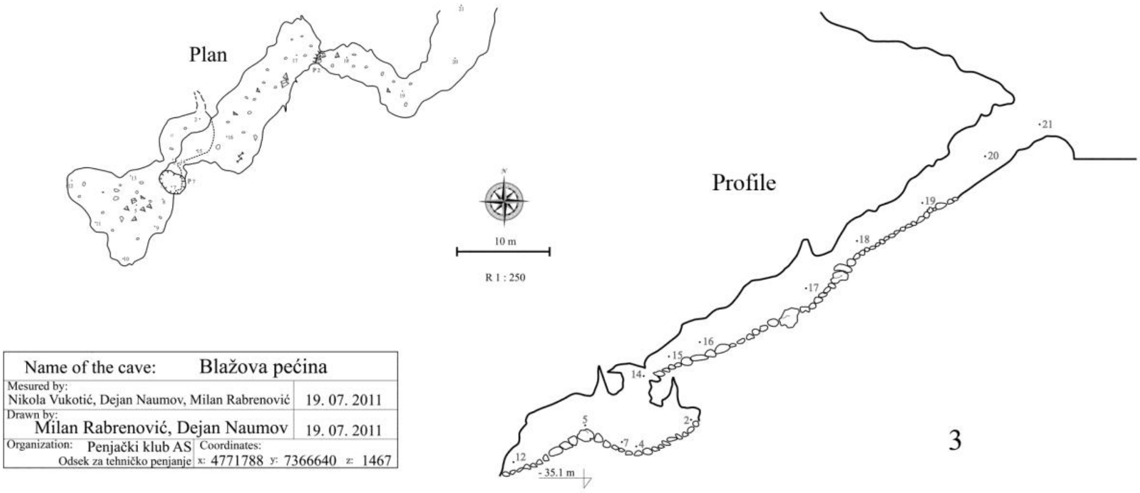
Mesothoracic pedunculus around 2 times wider than long and with stronger microreticulation than the pronotum.

Elytra elliptic, with maximum width in the mid-section, mean EL/EW 1.70 (males; HT 1.73), 1.60 (females). Surface with regularly spaced punctation and with microreticulation between the punctation. Pubescence short, dense and semi-erected.

Legs: long, slender, femora gradually thicken in the basal parts, tibias straight. Male protarsi with five protarsomeres, female protarsi with four protarsomeres, mesotarsi and metatarsi with five tarsomeres in both sexes. Tarsi not dilated, tarsal empodium with two setae.

Aedeagus: median lobe thicker at the base and with triangular apex in dorsal view (Fig. 6a), strongly curved in the basal region then more or less straight and flat in the central and apical region in lateral view (Fig. 6b). Parameres shorter than the median lobe (setae excluded) with a constriction in the preapical region, with acute apex and three apical setae (Fig. 6c). Endophallus without sclerotized structures.

Female genitalia: abdominal ventrite VIII with a short and narrow anterior expansion (fig. 6d). Urite IX without appendicular parts. Spermatheca: weakly sclerified, C shaped, short and slightly widened in the apical region.



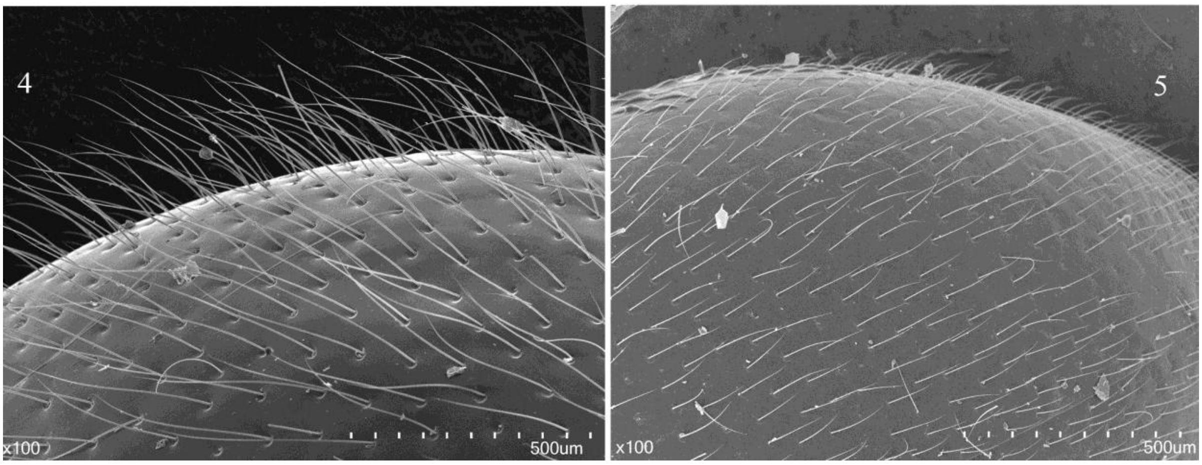


Fig. 3. Map of Blažova pećina.
Figs. 4–5. Elytral punctation and setation. 4: *A. matulici*; 5: *A. ganglbaueri ganglbaueri*

Distribution: *A. sinjajevina* n. sp. is presently known only from the type locality Blažova pećina, situated near the village Rudanca near Njegovuđe, on Sinjajevina Mt, Montenegro.

Blažova pećina is a small and simple cave consisting in two chambers divided by a 9 m deep vertical section (Fig. 3). The bottom of the first chamber is covered with leaf litter, branches and old trunks while the rest of the channel is covered with rock debris and blocks of 0.3 to 0.8 m.

Etymology: From Sinjajevina Mountain, where the type locality is situated.

Anthroherpon cecai **sp. n.**

Type material: Holotype male labeled: „MONTENEGRO, Durmitor Mt, village Mala Crna Gora, Poda, Crna jama (= Jama u Podu), 1900 m, 07.08.1993, leg. A. Milosavljević" (white label, printed) / „HOLOTYPUS *Anthroherpon cecai* sp.n. I. Njunjić, M. Perreau & D. Pavićević det. 2014" (red label, printed), (MNHN). Paratypes: 3♀, same date as holotype (MNHN, CDPV); 1♂, same locality, 03.08.1992, leg. A. Milosavljević, 1♂, same locality, 05.07.1991, leg. S. Ognjenović (MNHN, CDPV). All paratypes are labeled with white, printed locality labels and with red printed labels „PARATYPUS *Anthroherpon cecai* sp.n. I. Njunjić, M. Perreau & D. Pavićević det. 2014".

Diagnosis: Habitus illustrated in Fig. 2. Distinct from the other species of the group by the following characters: second antennomere approximately two times shorter than the first one. Lateral sides of the pronotum parallel on the basal fifth of the pronotum length. Pronotum with two symmetric dorsal impressions on the anterior half of the pronotum length (with variable extension and deepness, absent in some specimens). Elytral surface microreticulated between the punctation. Parameres as long as the median lobe (setae excluded), with preapical constriction and with rounded apex (fig. 7c).

Description: Body length from 6.59 to 7 mm. (HT 6.59 mm). General morphology leptodiroid; pronotum slightly wider than the head, wide elytra; eyeless; wingless and depigmented.

Head: hypognathous, wider in the anterior part, HL 1.34- 1.35 mm (HT 1.35 mm), HW 0.81- 0.85 mm (HT 0.81 mm). Surface shiny, microreticulated and with sparse and short pubescence. Penultimate maxillary palpomere approximately 1.4 times longer than the last one. Antennae 1.20 (males) or 1.00 (females) longer than the whole body. Second antennomere approximately 2 times shorter than the first one. Last antennomere approximately 1.7 times (males) or 1.6 times (females) longer than the penultimate. Antennal formula: see Table 1.

Table 1. Antennal formula of the holotype of *Anthroherpon cecai* sp. n (mm).

Article	I	II	III	IV	V	VI	VII	VIII	IX	X	XI
Length	0.48	0.27	0.93	0.6	0.95	0.8	0.86	0.86	0.61	0.67	1.07

Pronotum strongly narrowing towards the base with maximum width in anterior third, Pwmax/Pwmin: 1.65-1.73 (HT: 1.65 mm). Sides convergent in the anterior third and parallel on the basal fifth of the pronotum length. Dorsal area with two symmetric impressions more or less extended transversally (the conformation is variable according to the specimen and absent in some specimens), located on the anterior quarter of the pronotum length. Surface microreticulated, with short and sparse setae regularly disposed.

Mesothoracic pedunculus short, around 2.8 times wider than long and with stronger microreticulation than the pronotum.

Elytra elliptical, with maximum width in the mid-section, EL/EW approximately 1.6 (males; HT 1.74), 1.65 (females). Surface with regularly spaced punctation and with microreticulation between the punctation. Pubescence short, dense and semi-erected.

Legs long, slender, femora gradually thicken in the basal parts, tibias straight. Male with protarsi five protarsomeres, female with four protarsomeres, mesotarsi and metatarsi with five tarsomeres in both sexes. Tarsi not dilated, tarsal empodium with two setae.

Aedeagus with median lobe elongated, slightly wider in basal part and with rounded apex in dorsal view (Fig. 7a), strongly curved in the basal region then more or less straight and flat in the central and apical region in lateral view (Fig. 7b). Parameres as long as the median lobe (apical setae excluded), with a preapical constriction near, rounded at the apex, and with three long apical setae (Fig. 7c). Endophallus without sclerotized structures.

Female genitalia: abdominal ventrite VIII with a short and narrow anterior expansion (Fig. 7d). Urite IX without appendicular parts. Spermatheca: weakly sclerified, C shaped, short and slightly widened in the apical region.

Figs 6–11. Genital structures of *Anthroherpon* of the *ganglbaueri* group. a: aedeagus, dorsal view; b: aedeagus, lateral view with profile of the median lobe; c: apex of parameres; d: abdominal ventrite VIII of females. 6: *A. sinjajevina* n. sp.; 7: *A. cecai* n. sp.; 8. *A. matulici*; 9: *A. ganglbaueri ganglbaueri*; 10. *A. matzenaueri matzenaueri*; 11. *A. udrzali*.

Etymology: After our colleague and friend Aleksandar Milosavljević- Ceca, speleologist (Belgrade) and finder of the first specimens of this species.

Associated fauna: *Leonhardella (L.) antennaria antennaria* Apfelbeck 1907 and *Neobisium* sp. (Pseudoscorpiones).

Distribution: *A.cecai* sp. is presently known only from the type locality: Crna jama pit (-121 m). All *Anthroherpon* specimens were collected on the very bottom while *Leonhardella* and Pseudoscorpiones were found on the walls from the entrance to the bottom of the pit.

Anthroherpon udrzali **Giachino & Vailati, 2005 n. stat.**

Anthroherpon matzenaueri udrzali Giachino & Vailati, 2005: 150. (holotype *in* CJLK)
type locality: Montenegro, Orjen planina, pećina u Kučericama
Material examined: 1♂: Orjen planina, pećina u Kučericama (CMPR)

Further description:
Body length: 7.05 mm. Pronotum with strong microreticulation, sparse and regularly spaced punctation. Pwmax/Pwmin: 1.87. Elytra in an elongated oval shape: EL/EW: 1.93. Elytral surface with strong and dense punctation, with traces of transversal alignments of the punctation in the anterior third of the elytral length, without microreticulation. Aedeagus with median lobe triangularly narrowed at the apex in dorsal view (Fig. 11a), strongly curved in the basal region then more or less straight and flat in the central and apical region, the dorsal side sinuate in lateral view (Fig. 11b). Parameres thick, slightly shorter than the median lobe (apical setae excluded) with three apical setae.
The thickness of parameres and the disposition of the elytral punctation with traces of transversally aligned punctation (unusual in the subtribe Anthroherponina) support the change of status from subspecies to species rank. This status is consistent with the geographic location of this population in the maritime belt of Dinarides (Orjen) rather than in the central belt as the other subspecies of *A. matzenaueri* (Fig. 12).

REMARKS

Both new species occur along the southern banks of the Tara river (Fig. 12), they share two characters which seem to be synapomorphies: the second antennomere approximately two times shorter than the first one and the external side of parameres preapically sinuated (Figs. 6a, 7a).

The aedeagi of the other species of the group are illustrated to aid identification and complete the identification key: *A. matulici* (Figs. 8a, 8b), *A. ganglbaueri ganglbaueri* (Figs. 9a, 9b) and *A. matzenaueri matzenaueri* (Figs. 10a, 10b). *A. brckoensis*, reported from Brčko in northern Bosnia (Giachino & Guéorguiev, 1993) is evidently a mistake. Brčko is located far outside the distribution area of the genus *Anthroherpon*, and even more located outside the Dinarides range (Hajna, 2012). The exact wording of the label is „brčka" without anymore detail. The general morphology shows strong similarity with *A. ganglbaueri* and especially with *A. g. alticola*. As the revision of subspecies is not the topic of this paper, we suggest here the possible synonymy of *A. brckoensis* with *A. g. alticola* without establishing it formally.

IDENTIFICATION KEY

The following identification key of the *A. ganglbaueri* species group modifies and updates the key given by Guéorguiev (1990). It is dedicated to practical identification of species and does not correspond to the phylogeny of the group, which has not yet been addressed.

1 – Pubescence of elytra extremely long and erect. Elytral surface with very strong and sparse punctation, without microreticulation between punctation (Fig. 4) ……………………………………………………………………………*matulici* Reitter

– Pubescence of elytra shorter and semi-erected or recumbent. Elytral surface with smaller and denser punctuation (Fig. 5) ……………………………………………………2

2 – First antennomere approximately two times longer than the second one. Parameres with a preapical constriction (Figs. 6c, 7c) ……………………………………………3

– First antennomere approximately 1.5 times longer than the second one. Parameres regularly arcuate near the apex (Figs. 8a-11a) ……………………………………………4

3 – Size smaller: less than 6.1 mm (Fig. 1). Lateral sides of the pronotum parallel on the posterior fourth of the pronotum length. Penultimate maxillary palpomere approximately 1.2 times longer than the last one. Parameres of the aedeagus shorter than the median lobe (apical setae excluded) and with an acute apex (Fig. 6c) ……………………………………………………………………*sinjajevina* n. sp.

– Size larger: more than 6.4 mm (Fig. 2). Lateral sides of the pronotum parallel on the posterior fifth of the pronotum length. Penultimate maxillary palpomere approximately

1.4 times longer than the last one. Parameres of the aedeagus as long as the median lobe (apical setae excluded) and with a rounded apex (Fig. 7c) ……*cecai* n. sp.

4 – Pronotum with its largest width at the anterior quarter of its length, and with the lateral sides parallel on the basal third of its length………………………*ganglbaueri* Apfelbeck

– Pronotum with its largest width at the anterior third of its length and with its lateral sides parallel at most on the basal fourth of its length ………………………………………………5

5 – Elytral surface with weaker and uniformly dispersed punctation. Elytra less elongate in males, less than 1.75 longer than wide……………………………………*matzenaueri* Apfelbeck

– Elytral surface with stronger punctation and with clear traces of transversal alignments in the humeral region (anterior third of the elytral length). Elytra very elongate in males, more than 1.9 longer than wide. Body length ~7 mm …………………………………………………………………………………*udrzali* Giachino & Vailati

BIOGEOGRAPHY

Fig. 12 shows the geographic distribution of the species of the *A. ganglbaueri* species group, in relation to the main geomorphological units of Dinarides according to http://www.summitpost.org/dinaric-alps/155326#chapter_14. Dotted lines separate the three main belts, from south to north: maritime belt; central belt or High Dinaric Alps; northeastern belt. Different colors indicate the three main massifs where *Anthroherpon* of the *ganglbaueri* species group live: high Herzegovina mountains; maritime and central Montenegro; high Montenegrian plateaus (for exhaustive nomenclature of all massifs of the Dinarides, cf. above cited URL). The distribution areas of species are roughly aligned on the main geomorphological and tectonic alignments, parallel to the coast line: In the maritime belt: *A. udrzali* in Orjen and *A. matulici* in Orjen and in Garač. In the central belt: *A. ganglbaueri*, *A. matzenaueri*, *A.* and *A. sinjajevina*. *A. ganglbaueri* is located in high Herzegovina, each one of the subspecies occurring in one of the main massifs of this region (except Čvrsnica and Lebršnik): *A. g. alticola* in Prenj; *A. g. ganglbaueri* in Velez; *A. g. distinguendum* in Crvanj; *A. g. intermedium* in Baba. *A. matzenaueri* is located in the high Montenegrian massifs, the three subspecies are aligned along the Golija-Vojnik-Maganik line: *A. m. matzenaueri* in Golija; *A. m. taliensis* in Žurim; *A. m. augustae* in Maganik and Vojnik. It should be noticed that Maganik and

Žurim are contiguous and belong to the same Morača massif, but accomodate two different subspecies while Maganik and Vojnik, are not contiguous and accomodate the same subspecies. The new species *A.* and *A. sinjajevina* are aligned along massifs located on the south bank of Tara river (respectively Mala Crna Gora and Rudanci).

However this correlation between tectonics of Dinarides and species distribution leaves opened some problems as the questionable disconnected distribution of *A. matzenaueri augustae* in the two distant massifs of Maganik and Vojnik. More generally, the relevance of the subspecific taxa which is not adressed in this paper. Without additional experimental data, it is untimely to make further hypothesis on the phylogeny of taxa of this group and the relations between phylogeny and distribution. Molecular investigations of all taxa of *Anthroherpon* are in progress and will help to clarify the distributional and evolutionary patterns of the genus.

Fig. 12. Distribution map of the species of the *A. ganglbaueri* group. Legends of colors are given only for regions where Anthroherpon of the *ganglbaueri* species group live. Map after Wikipedia © Sémhur / Wikimedia Commons / CC-BY-SA-3.0 (modified); data after http://www.summitpost.org/dinaric-alps/155326#chapter_14

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2.8. RESULTS AND DISCUSSION

COI amplicons were recovered from 75 individuals, and no indels or stop codons were encountered. Sequence length averaged 650 bp (range = 297–659 bp), and 97.4% of the records were above 650 bp. Overall nucleotide frequencies were as follows: A (29.5%), C (17.1%), G (15.4%), and T (36.8%). The barcodes of 15 species and 26 subspecies were obtained, representing more than half of *Anthroherpon* diversity. Mean intraspecific genetic distance in the barcoding region was 1.6% (min 0%, max 6.8%), and mean interspecific genetic distance was 12% (min 3.6%, max 20.3%). However, if we elevate the subspecies *A. taxi albanicum*, *A. hoermanni sericeum*, *A. hoermanni hypsophylum,* and *A. hoermanni orlovacensis* to species level, and treat *A. taxi remyi* as a subspecies of *A. albanicum*, mean intra-specific genetic distance changes to 0.8% (min 0%, max 3%), while mean interspecific genetic distance remains almost the same: 11.9% (min 2%, max 20.3%). Under the latter scenario (respective subspecies elevated to species level), this division between intra- and interspecific sequence variation is further illustrated through the histogram in Fig. 15. A threshold value below 3% COI sequence variation (i.e., a barcoding gap) can be detected, which separates intra- and interspecific sequence pairs in most of the cases. Interestingly, a second gap is seen within the interspecific distances around 8% divergence, which may suggest a recent burst of simultaneous speciation followed by a period with less speciation; or, alternatively, the second gap may signify a period of extinction.

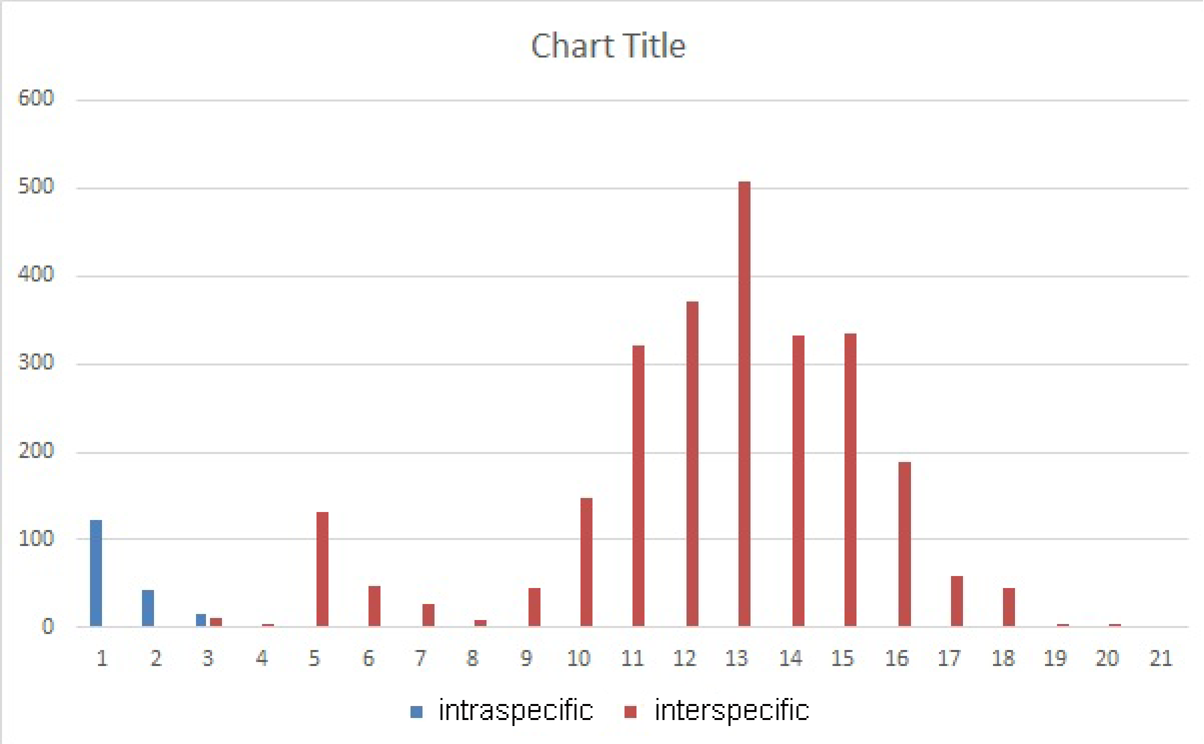

Fig. 16. Distribution of pairwise simple genetic distances between COI-sequences, revealing a distinct "barcoding gap" at 3% divergence.

In subterranean environment morphological traits often undergo strong selection due to extreme abiotic factors and may not entirely reflect the evolutionary history of the species. Therefore, the discrepancy between morphological and molecular analyses is often found in cave organisms (Faille et al., 2010; Juan & Emerson, 2010). In this study, we demonstrated that the application of DNA barcoding can alleviate these problems. Due to troglomorphic convergence, taxonomic designation of taxa within *Anthroherpon* has always been difficult. The previous, qualitative and purely morphology-based species delimitation and classification proved to be supported in the case of most of the *Anthroherpon* taxa. However, our results suggest that at least four subspecies (*A. taxi albanicum*, *A. hoermanni sericeum, A. hoermanni hypsophylum,* and *A. hoermanni orlovacensis*) should be elevated to species level, and that *A. taxi remy* should be treated as a subspecies of *A. albanicum*. Elevating *A. taxi albanicum* to species level, and placing *A. taxi remyi* as its subspecies is in the acoordance with the classification of *Anthroherpon* proposed by Jeannel (1930) based on morphological characters.

In the case of two species with very wide, and mostly overlaping distribution areas (*A. taxi* and *A. latipenne*), our results revealed two different patterns. Despite mostly large geographical distance between 8 different populations of *A. latipenne,* genetic distances are small, in most cases less than 1 %, and maximally 1.4 %. On the other hand, genetic distances between different populations of *A. taxi* are higher and ranging from 1.6% to 6.2%. This indicates that one subspecies of *A. taxi* (*A. taxi albanicum*) could be elevated to species level. Interestingly, it seems that there is not much correlation between genetic and geographic distance since the highest genetic distance is between subspecies which live more than 100 km (*A. taxi taxi* and *A. taxi albanicum*) apart while the lowest genetic distance is recorded between subspecies found 70 km apart (*A. taxi taxi* and *A. taxi sydowi*). At present, these patterns remain mysterious and hard to interpret given our current knowledge of dispersal and endemism in *Anthroherpon*.

2.9. APPENDIX

Fig. 2. Approximate distribution of the genus *Anthroherpon*

Fig. 3. Distribution of *A. taxi* and *A. latipenne*.

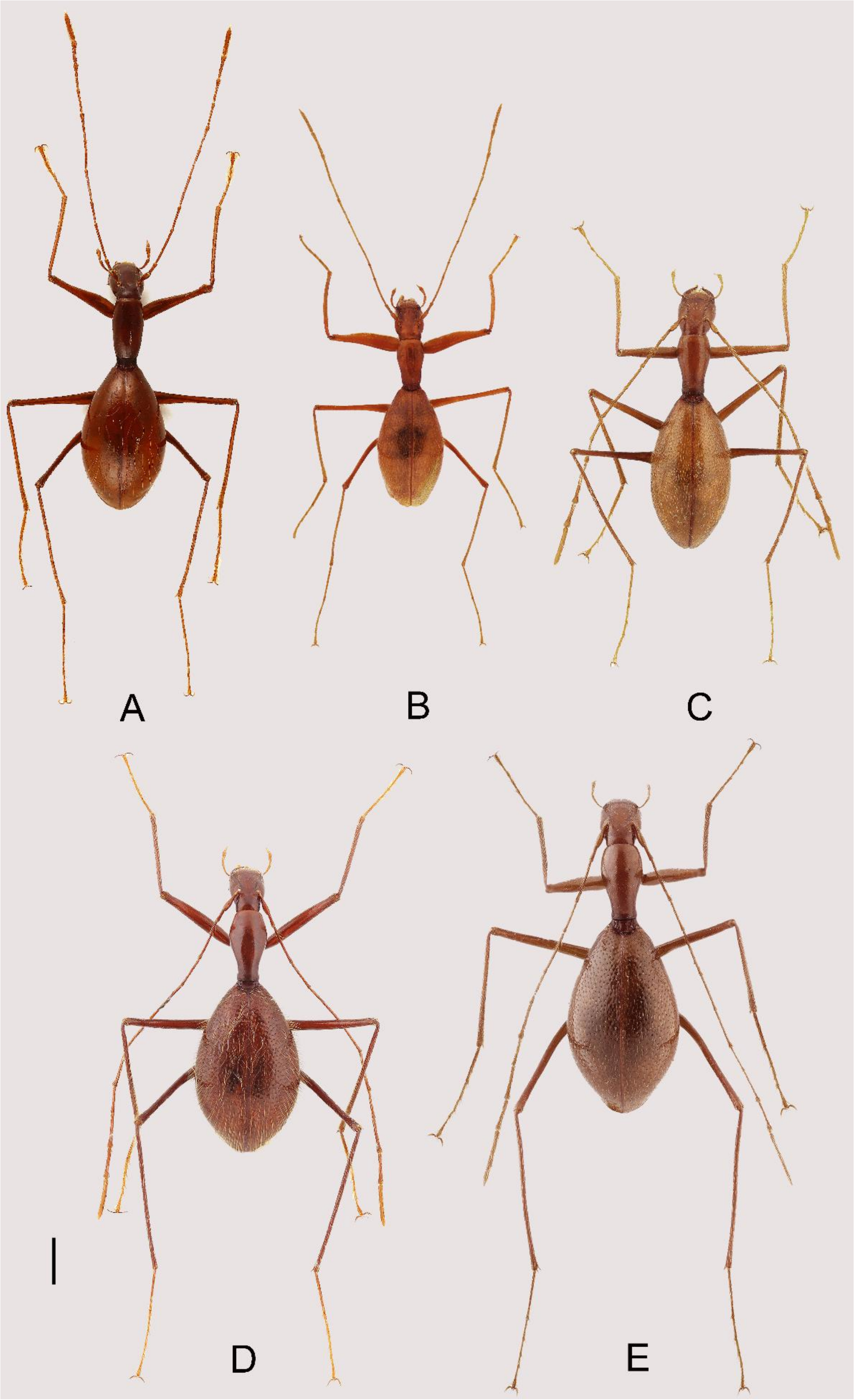

Fig. 10A. Habitus. A. *A. cylindricolle cylindricolle*; B. *A. harbichi*; C. *A. ganglbaueri ganglbaueri*; D. *A. matulici*; E. *A. matzenaueri matzenaueri*. Scale bar is 1 mm. (Figs. B, C, D, E from Hlaváč et al., 2016, used with permission).

Fig. 10B. Habitus. F. *A. hossei*; G. *A. piesbergeni*; H. *A. primitivum jeanneli*; I. *A. taxi remyi*; J. *A. taxi taxi*; K. *A. stenocephalum stenocephalum*. Scale bar is 1 mm. (Figs. F, G, H, J from Hlaváč et al., 2016, used with permission).

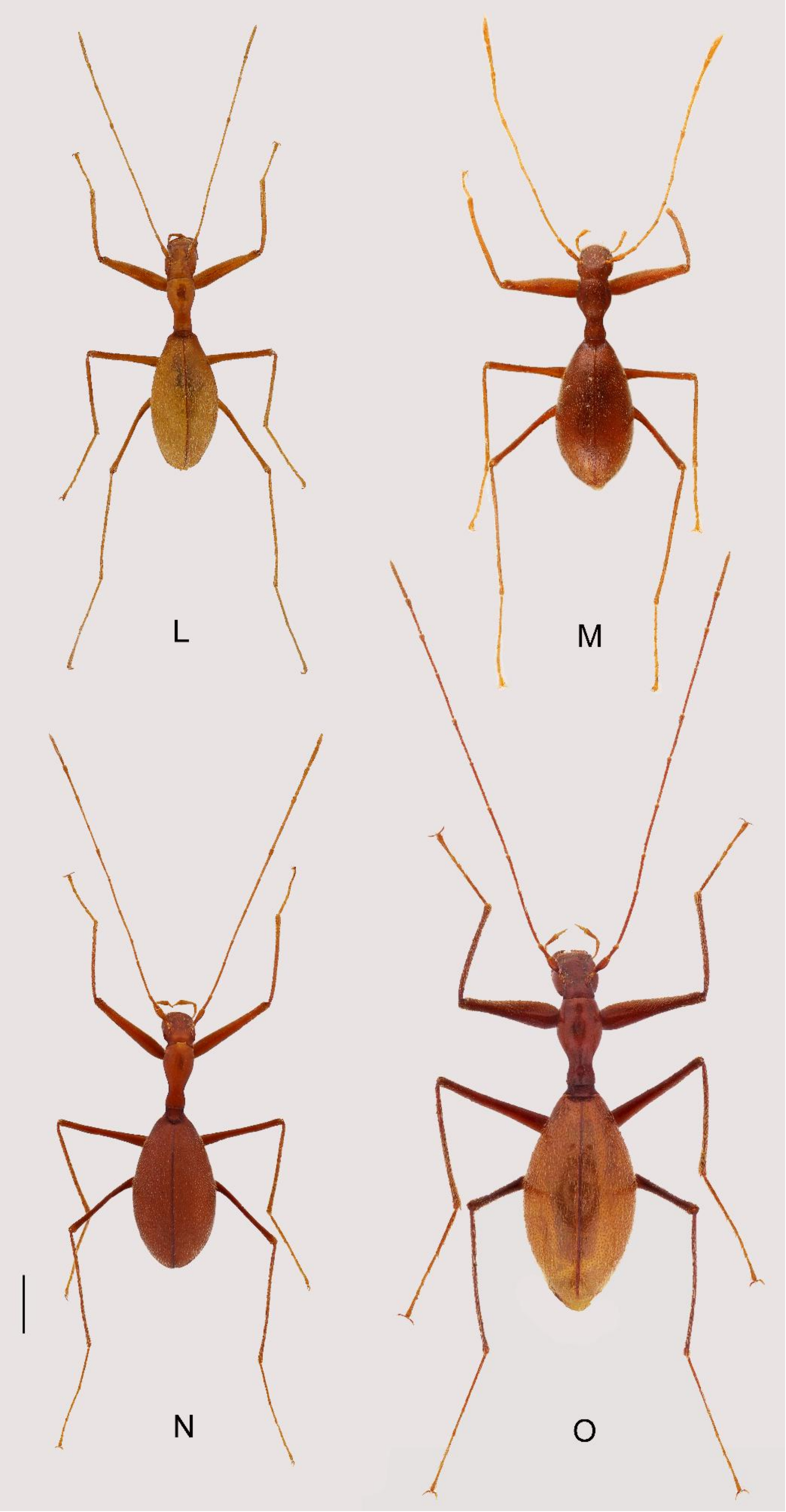

Fig. 10C. Habitus. L. *A. charon*; M. *A. pozi*; N. *A. zariquieyi*; O. *A. hoermanni hoermanni*. Scale bar is 1 mm. (Figs. L, N, O from Hlaváč et al., 2016, used with permission).

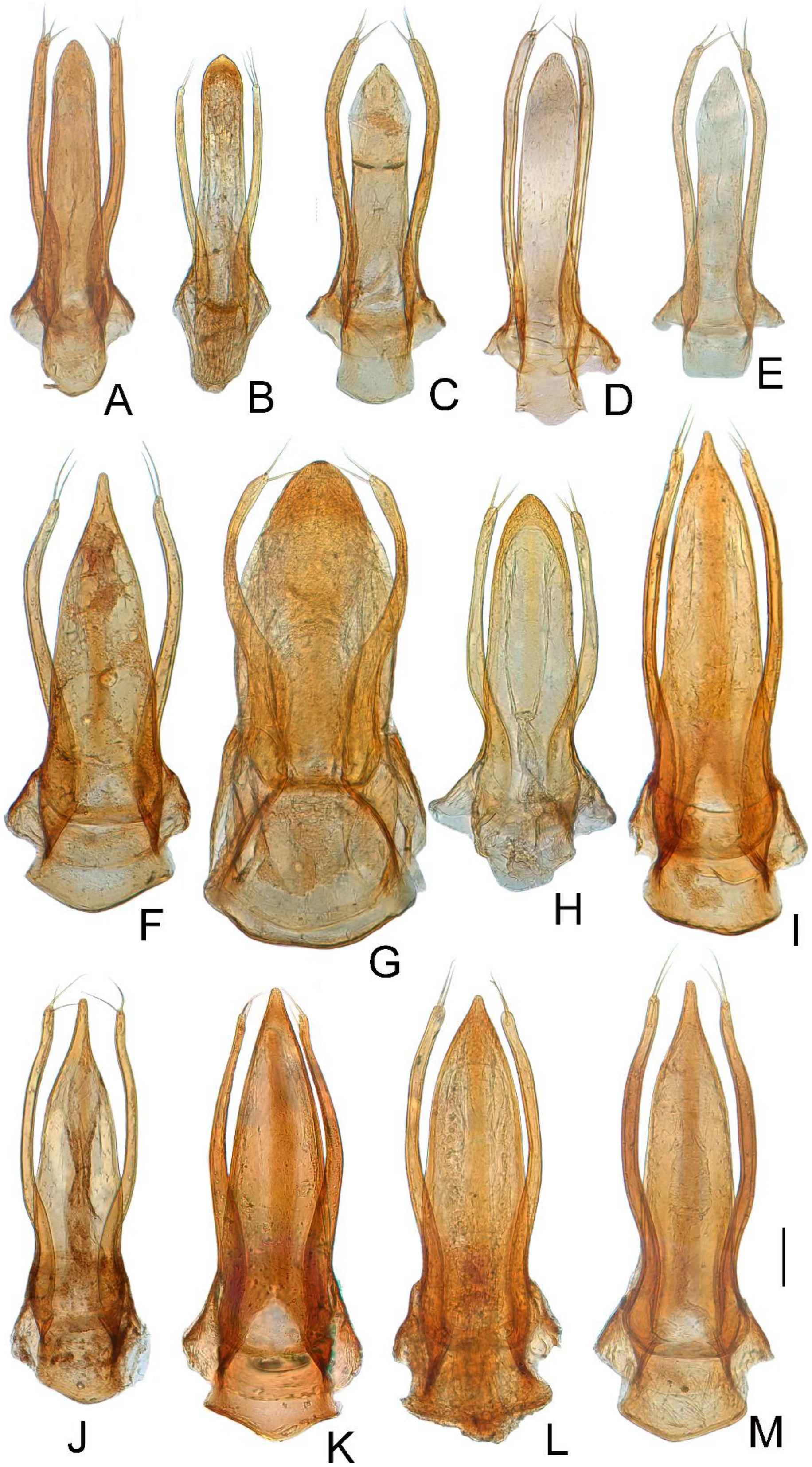

Fig. 11. Aedeagi of the genus *Anthroherpon*. A. *A. cylindricolle s. str.*; B. *A. harbichi*; C. *A. hoermanni s. str.*, D. *A. hoermanni hypsophilum*; E. *A. hoermanni sericeum*; F. *A. latipenne s. str.*; G. *A. piesbergeni*; H. *A. primitivum jeanneli*; I. *A. taxi taxi*; J. *A. taxi remyi*; K. *A. taxi sydowi*; L. *A. taxi pretneri*; M. *A. taxi albanicum.* Scale bar 0.1mm.

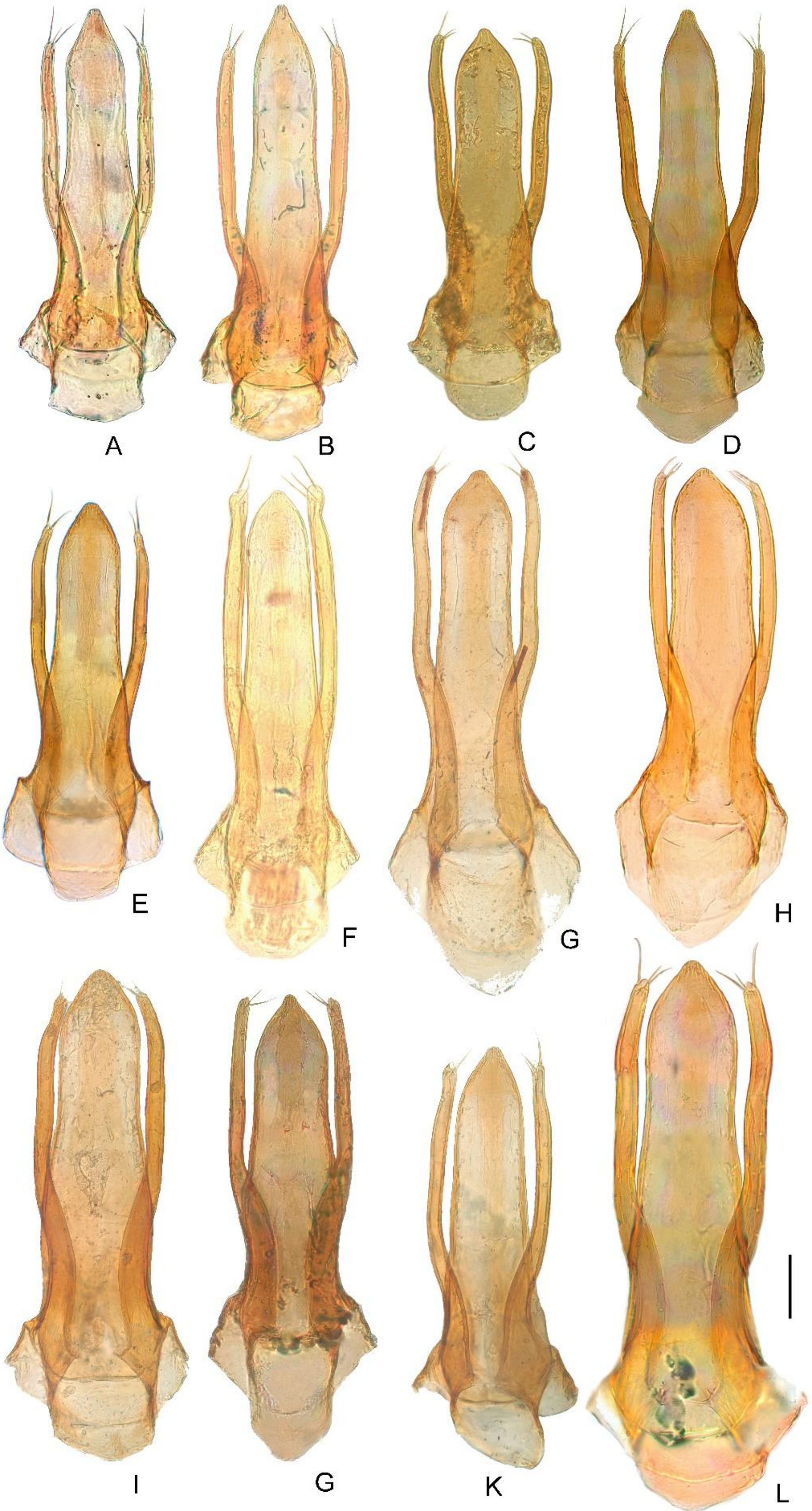

Fig. 12. Aedeagi of *Anthroherpon* of "*ganglbaueri*" group. A. *A. brckoensis*; B. *A. ganglbaueri alticola*; C. *A. ganglbaueri distinguendum*; D. *A. ganglbaueri ganglbaueri*; E. *A. ganglbaueri intermedium*; F. *A. cecai*; G. *A. matulici*; H. *A. matzenaueri augustae*; I. *A. matzenaueri matzenaueri*; G. *A. matzenaueri taliensis*; K. *A. sinjajevina*; L. *A. udrzali*. Scale bar 0.1mm.

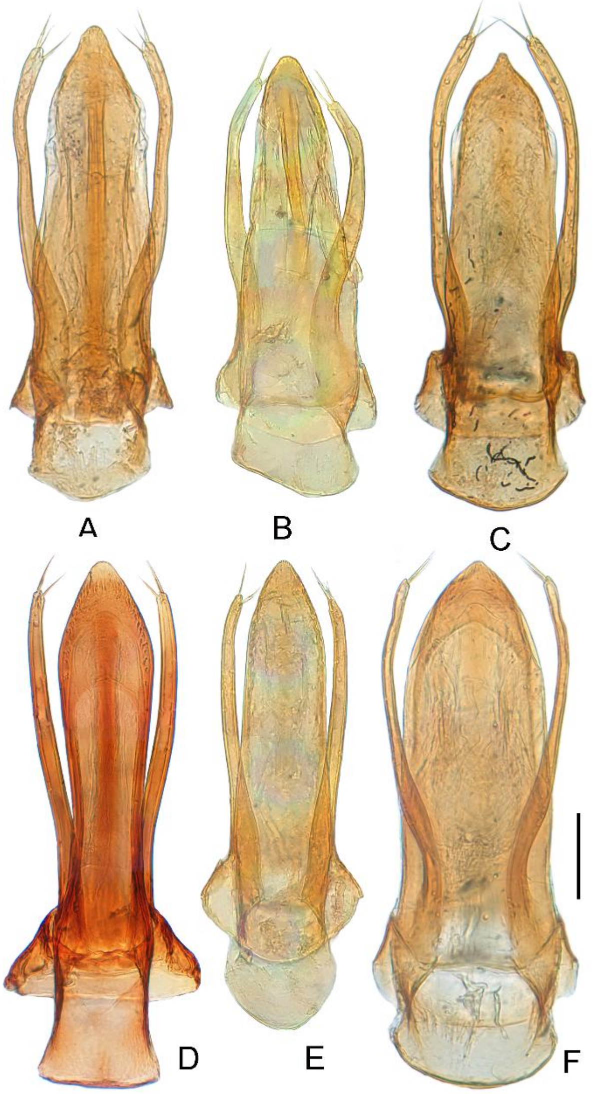

Fig. 13. Aedeagi of *Anthroherpon* of "*pygmaeum*" and "*stenocephalum*" group. A. *A. weiratheri*; B. *A. erebus scheibeli*; C. *A. pygmaeum stricticolle*; D. *A. zariquieyi*; E. *A. stenocephalum s. str.*; F. *A. weiratheri*. Scale bar 0.1mm.

CHAPTER III

ARTICLE 2:

THE CAVE BEETLE GENUS *ANTHROHERPON* IS POLYPHYLETIC; MOLECULAR PHYLOGENETICS AND DESCRIPTION OF *GRACILIELLA* N. GEN. (LEIODIDAE, LEPTODIRINI)

The cave beetle genus *Anthroherpon* is polyphyletic; molecular phylogenetics and description of *Graciliella* n. gen. (Leiodidae, Leptodirini)

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Key words: Coleoptera, convergent evolution, Dinaric Mountains, morphometrics, troglobites

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Abstract

The subtribe Anthroherponina form an iconic group of obligate cave beetles, typical representatives of the Dinaric subterranean fauna, which is considered to be the richest in the world. Phylogenetic studies within this subtribe are scarce and based only on morphological characters, which, due to troglomorphic convergence, are frequently unreliable. Moreover, morphological stasis and morphological polymorphism make classification of taxa difficult. To test if characters that have traditionally been accepted as informative for Anthroherponina classification are indeed reliable, we evaluated the monophyly of the most species-rich genus of this subtribe - *Anthroherpon* Reitter, 1889. Our study, based on a molecular phylogenetic analysis of fragments of the 18S, 28S, and COI (both 5' and 3' end) loci revealed that the genus *Anthroherpon* as conventionally defined is polyphyletic. To resolve this polyphyly, we defined one new additional genus, *Graciliella* n. gen., for which we then examined the intrageneric diversity using molecular and morphometric approaches. Molecular phylogenetic analysis of two COI mitochondrial gene fragments revealed the presence of four species inside *Graciliella* n. gen., including two new species, which we here describe as G. *kosovaci* n. sp. and *G ozimeci* n. sp. To analyze interspecific morphological differences within *Graciliella* we performed a discriminant analysis based on 40 linear morphometric measurements. The results showed that differences between species and subspecies inside *Graciliella*, however subtle they may seem, are measurable and reproducible. All species of the genus are briefly diagnosed, an identification key is proposed and a distribution map of all taxa of *Graciliella* is provided.

3.1. INTRODUCTION

Unrelated organisms that are placed under similar, severe selection pressures often converge on (or evolutionarily remain trapped in) a limited set of body shapes, and may therefore be hard to classify morphologically. Extreme environments (*e.g.* subterranean habitats, deep sea, polar regions, deserts) exert strong directional selection upon their inhabitants, leading to convergent and generally predictable outcomes (Cloudsley-Thompson, 1988; Trontelj et al., 2012). As a consequence, extremophiles found in Arctic tundra (Grundt et al., 2006), subterranean habitats (Lefébure et al., 2006) and deep-sea environments (Vrijenhoek et al., 1994) are well-known for their morphological uniformity and cryptic diversity.

Subterranean environments are considered to be extreme because of highly stable abiotic conditions and low nutrient availability. Species that have colonized these habitats evolve a somewhat predictable suite of morphological, physiological and behavioural characteristics (Gross, 2012; Trontelj et al., 2012), including eye degeneration, depigmentation, development of sensory organs, longer life cycles, lower metabolic rate, and body shape modifications (Racovitza, 1907; Vandel, 1964; Culver et al., 1990; Faille et al., 2009). This set of characteristics is known as ‘troglobiomorphy’ or ‘troglomorphy’ and organisms characterized by these features are often restricted to subterranean environment (troglobites).

The troglomorphic phenotype shared by diverse cave-adapted animals is often regarded as a classical example of convergent evolution (Stemmer, 2015). Many studies have shown troglomorphic convergence in different taxa (Christiansen, 1961; Hedin and Thomas, 2010; Protas et al., 2006), which may complicate phylogenetic reconstruction by morphological approaches. Molecular techniques have become a crucial tool to overcome this problem and to efficiently reveal phylogenetic relationships among different taxonomic categories. They have been recently involved in elucidating the phylogeny of different groups with subterranean representatives such as Bivalvia (Stepien et al., 2001), Dytiscidae (Leys et al., 2003), Decapoda (Zakšek et al., 2007), Parabathynellidae (Abrams et al., 2012), Trechini (Faille et al., 2009; 2013) and Leptodirini (Caccone and Sbordoni, 2001; Ribera et al., 2010; Fresneda et al., 2011;

Cieslak et al., 2014a; 2014b). Molecular techniques have helped in distinguishing morphological characters inherited from a common ancestor from those resulting from recent, independent adaptation to a specific biotope.

Among Insecta, Coleoptera are the dominant group that have colonized subterranean habitats (Sket, 2005). Two groups of Coleoptera are particularly diverse in caves: the subfamily Cholevinae in the suborder Polyphaga, and the subfamily Trechinae in the suborder Adephaga (Casale et al., 1998; Faille et al., 2009). With more than 230 genera and 900 (mostly polytypic) species, the tribe Leptodirini (Cholevinae) forms one of the most species-rich tribes of cave-adapted Coleoptera, surpassed only by Trechini (Carabidae). Most are subterranean, living in caves and fissures, except a few species inhabiting underground termite nests or superficial litter layers. Within the Leptodirini, four morphological types are recognized: bathyscioid, pholeuonoid, scaphoid, and leptodiroid (Jeannel, 1924). The leptodiroid morphological type, named after *Leptodirus hochenwartii* Schmidt, 1832 from Slovenia, Croatia, and Italy, is the most derived morphologically, compared to the presumed ancestral body plan (Sket, 2005). Species belonging to this morphological type have extremely long appendages, extremely elongated head and pronotum, hemispherical elytra, and are fully anophthalmic. The subtribe Anthroherponina comprises taxa of exclusively leptodiroid morphological type, showing the most pronounced troglomorphic characters among Leptodirini (Njunjić et al., 2015). To date, taxonomic studies of Anthroherponina were based exclusively on morphological traits, but possible phenotypic convergence leaves these conclusions doubtful.

To test if characters that have traditionally been accepted as indicative of Anthroherponina classification are indeed reliable, we evaluated the monophyly of the most species-rich genus of this subtribe - *Anthroherpon* Reitter, 1889. Taxonomy of this genus has always been difficult. Most taxa were described in the late 19th century and in the second half of the 20th century, on a small number of specimens and without an explicit diagnosis, so many were subsequently synonymized. Moreover, the geographical distribution of some taxa is mysteriously disjunct (Njunjić et al., 2015).

The present work provides a first phylogenetic analysis of three closely related genera of the subtribe Anthroherponina: *Anthroherpon*, *Leptomeson* Jeannel, 1924, and *Hadesia* Müller, 1911, combining morphological and molecular approaches. We clarify their generic status and we reveal the existence of the fourth, new genus, *Graciliella*, for which we then examine the intrageneric diversity. Additionally, two new species belonging to this new genus are described.

3.2. MATERIAL AND METHODS

3.2.1. Acronyms

The following abbreviations for collections and institutions are used: CNHM: Croatian Natural History Museum, Zagreb, Croatia; MNHN: Muséum National d'Histoire Naturelle, Paris, France; Naturalis: Naturalis Biodiversity Center, Leiden, The Netherlands; NMP: Narodni Museum, Prague, Czech Republic; NHM: Naturhistoriches Museum Wien; CINJ: Iva Njunjić Collection, Belgrade, Serbia; CMPR: Michel Perreau Collection, Paris, France; CDPV: Dragan Pavićević Collection, Belgrade, Serbia.

The following abbreviations for morphological details are used: HL: head length (measured from the posterior margin of the clypeus); HW: maximum width of the head; PL: pronotum length (measured along the median line); Pwmax: maximum width of the pronotum (largest transverse width); Pwmin: minimum width of the pronotum (shortest transverse width), PL/Pwmax: length of the pronotum divided by maximum width of the pronotum; MPl: mesothoracic pedunculus length, MPwmin: minimal width of the mesothoracic pedunculus; PL/MPl: length of the pronotum divided by the length of mesothoracic pedunculus; EL: elytra length (as linear distance measured along the median line from the base to the apex); EW: maximum width of the elytra; EL/EW: length of the elytra divided by maximum width of the elytra; MP2/MP3: length of the second maxillary palp divided by the length of the third maxillary palp. Reference to type material is abbreviated as HT: holotype.

3.2.2. Taxon sampling and morphological study

Specimens were collected in caves of the Dinaric range, in Montenegro, Bosnia and Herzegovina, and Croatia as listed in S1. For amplification and sequencing we used two specimens per population of the genus *Graciliella* n. gen. and *Anthroherpon*, and one specimen per population of the genus *Hadesia* Müller, 1911. Since specimens from the same population had almost identical 28S, 18S and COI gene fragments, we used only one individual per population for the phylogenetic tree. A total of 44 specimens representing 27 taxa from three genera (*Anthroherpon*, *Hadesia*, and *Leptomeson*) were included in the molecular analysis. We chose these genera because they are the most species-rich Anthroherponina and because major taxonomic issues need to be resolved among them. As outgroups we chose several subterranean species or subspecies from other genera of the tribe Leptodirini: *Parapropus* *sericeus muelleri* Jeannel, 1924, *Charonites* Apfelbeck, 1907 sp., and *Apholeuonus nudus sturanyi* Apfelbeck, 1906 of which we sequenced a single individual per population. To root the tree, we used *Speonesiotes* Jeannel, 1910 sp. belonging to the subtribe Bathysciotina. In total we sampled 36 specimens of 18 species of *Anthroherpon*, 4 specimens of 4 species of *Hadesia*, and 4 specimens of 4 species of *Leptomeson* (Table 1; S1).

After extraction, male genitalia were cleared in clove oil, mounted in Euparal on glass slides and pinned beneath the specimens. The external morphology of specimens was examined using Leica MZ 75 and Leica M10 stereomicroscopes. Microphotographs were taken on a Leica DIAPLAN or a Zeiss AXIOLAB microscope with a camera diagnostic instrument Spot INSIGHT IN1820. Macrophotographs were taken using a Canon Eos 70D camera with Canon EF MP-E 65mm f/2.8 macro lens and Canon macro twin lite mt-24ex flash. Digital pictures were processed using the Helicon Focus software 5.3 (http://www.heliconsoft.com/heliconsoft-products/helicon-focus/).

Table 1. Genera with total number of species and number of species included in the study. Taxonomy follows Perreau (2004), updated.

Genus	**N. spp.**	**Sampled spp.**
Anthroherpon	30	18
Hadesia	4	4
Leptomeson	9	4

3.2.3. DNA extraction, PCR amplification, and sequencing

The specimens used in the study were collected alive in the field and preserved in 96% ethanol. DNA was extracted from whole specimens or from one leg with a standard phenol–chloroform extraction (Blin and Stafford, 1976) or the DNeasy Tissue Kit (Qiagen GmbH, Hilden, Germany). Voucher specimens are stored in the MNHN (Paris, France), CNHM (Zagreb, Croatia) and DNA aliquots are kept in the tissue collections of Naturalis (Leiden, Netherlands).

We amplified fragments of two nuclear genes: 5’ end of the small ribosomal unit, 18S rRNA (SSU) and an internal fragment of the large ribosomal unit, 28S rRNA (LSU), and of two non-overlapping sections of mitochondrial gene fragments– the 5’ and 3’ halves of cytochrome c oxidase subunit 1 (which we here term COIa and COIb, respectively). Primers used are given in Table 2, and PCR protocols are given in S2. Sequences were assembled and edited using Geneious version 8.0.5 (http://www.geneious.com/, Kearse et al., 2012). DNA sequences obtained for each genetic marker were aligned separately using MAFFT version 7 (Katoh and Standley, 2013). Sequences have been deposited in GenBank with Acc. Nos (S1).

Table 2. Primers used in the study.

Fragment	**Name**	**Sense**	**Sequence 5'-3'**	**Reference**
COIa	LCOI-1490	F	GGTCAACAAATCATAAAGATATTG	Folmer et al. (1994)
COIa	HCOI-2198	R	TAAACTTCAGGGTGACCAAAAAATCA	Folmer et al. (1994)
COIb	Jerry	F	CAACATTTATTTTGATTTTTTGG	Simon et al. (1994)
COIb	Pat	R	TCCAATGCACTAATCTGCCATATTA	Simon et al. (1994)
18S	5'	F	GACAACCTGGTTGATCCTGCCAGT	Shull et al. (2001)
18S	b5.0	R	TAACCGCAACAACTTTAAT	Shull et al. (2001)
28S	Ka	F	ACACGGACCAAGGAGTCTAGCATG	Ribera et al. (2010)
28S	Kb	R	CGTCCTGCTGTCTTAAGTTAC	Ribera et al. (2010)

3.2.4. Phylogenetic analyses

For each sequence alignment, the optimal model of nucleotide substitution was determined using jModelTest2 (Darriba et al., 2012; Guindon and Gascuel, 2003), run on the CIPRES webportal (Miller et al., 2010). Selection was based on the Akaike Information Criterion (AIC), and resulted in selection of TIM2ef+I, GTR+G, TPM1uf+I+G and TPM3uf+I+G substitution models for 18S, 28S, COIa and COIb, respectively. We did two separate phylogenetic analyses- one at the genus level, using all three loci, and one at the species level within *Graciliella* using only COI. Phylogenetic analysis was performed using MrBayes 3.2.2 (Ronquist and Huelsenbeck, 2003) on CIPRES (Miller et al., 2010), with settings for multiple markers unlinked, running two replicates of 15 x 10^6 generations each. Sample frequency was set to 5000. Convergence diagnostics were run using Tracer version 1.5 (Rambaut et al., 2014), where ESS values for all parameters were >>200. After discarding a 25% burn-in, the resulting majority-rule consensus tree was visualized using FigTree version 1.4 (Rambaut, 2012).

3.2.5. Morphometric analysis

We subjected a total of 41 individuals, all males, belonging to *G. apfelbecki apfelbecki* (14 specimens), *G. apfelbecki scutulatum* (1 specimen), *G. metohiensis* (14 specimens), *G. absoloni* (1 specimen), *G. lahneri* (2 specimens), *G. ozimeci* (9 specimens), and *G. kosovaci* (1 specimen) to linear morphometric measurement with a Micro-Vu Vertex 251HC (https://www.microvu.com/), three-dimensional set-up, using Inspec Metrology Software (https://www.inspec-inc.com/). We used 40 landmarks on antennae, maxillary palps, head, thorax, abdomen, and legs (S4). Each individual was measured three times. In a small number of cases, obvious measurement errors were detected a posteriori (values differing by a factor of >2 from the other two replicates of the same individual). These were then replaced by the average value of the other two replicates. The full list of measured material and their respective morphometrics is given in S3. We first checked, for those species for which we had DNA extracts, whether the species that had been recognized by traditional, qualitative morphological study, could also be recovered as monophyletic groups in the molecular phylogeny (see below). As this appeared to be the case, we then subjected this set of independently obtained quantitative measurements to a Discriminant Analysis in PAST 3.10 (Hammer et al., 2001), to investigate whether a more objective morphometric analysis would allow us to recognize the same groups.

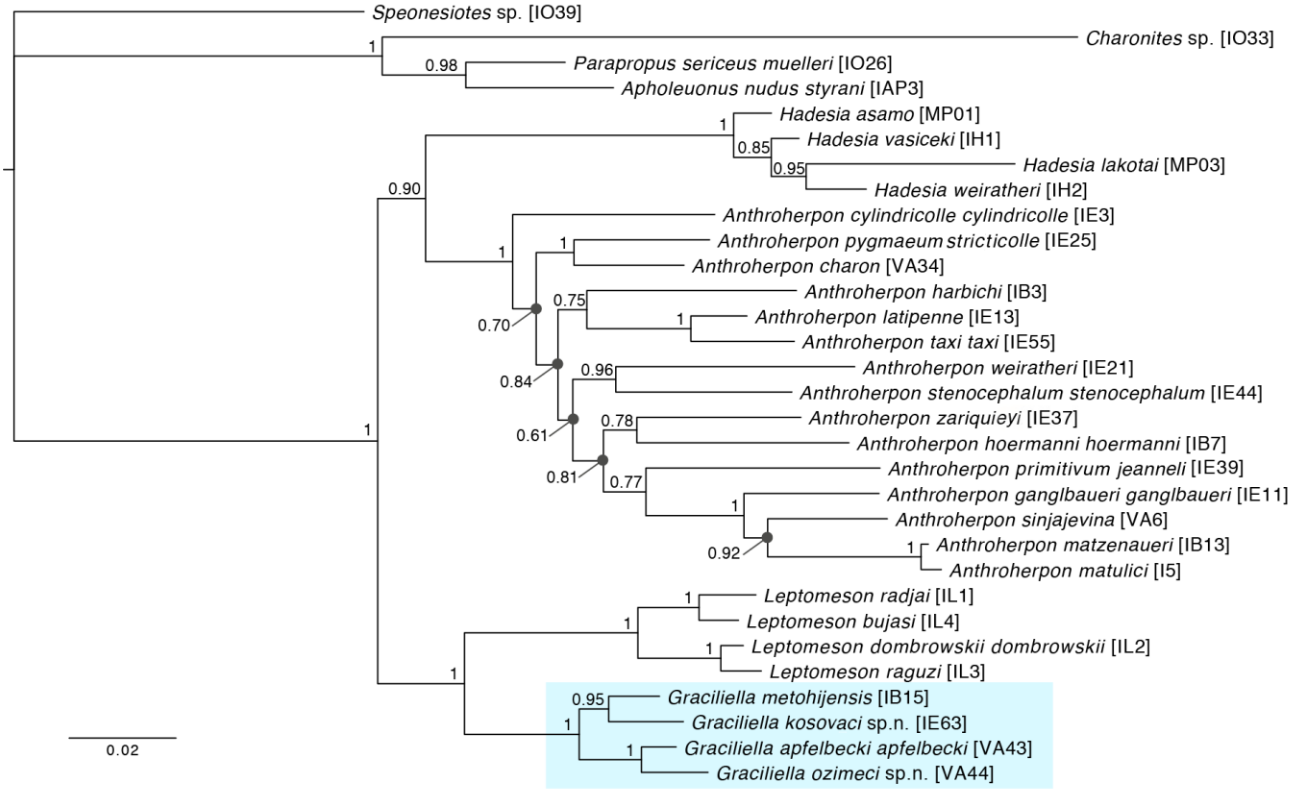

Fig 1. Bayesian inference of combined 18S, 28S, COIa, and COIb data sets for *Hadesia*, *Anthroherpon, Leptomeson*, and *Graciliella*. Numbers above nodes are posterior probabilities.

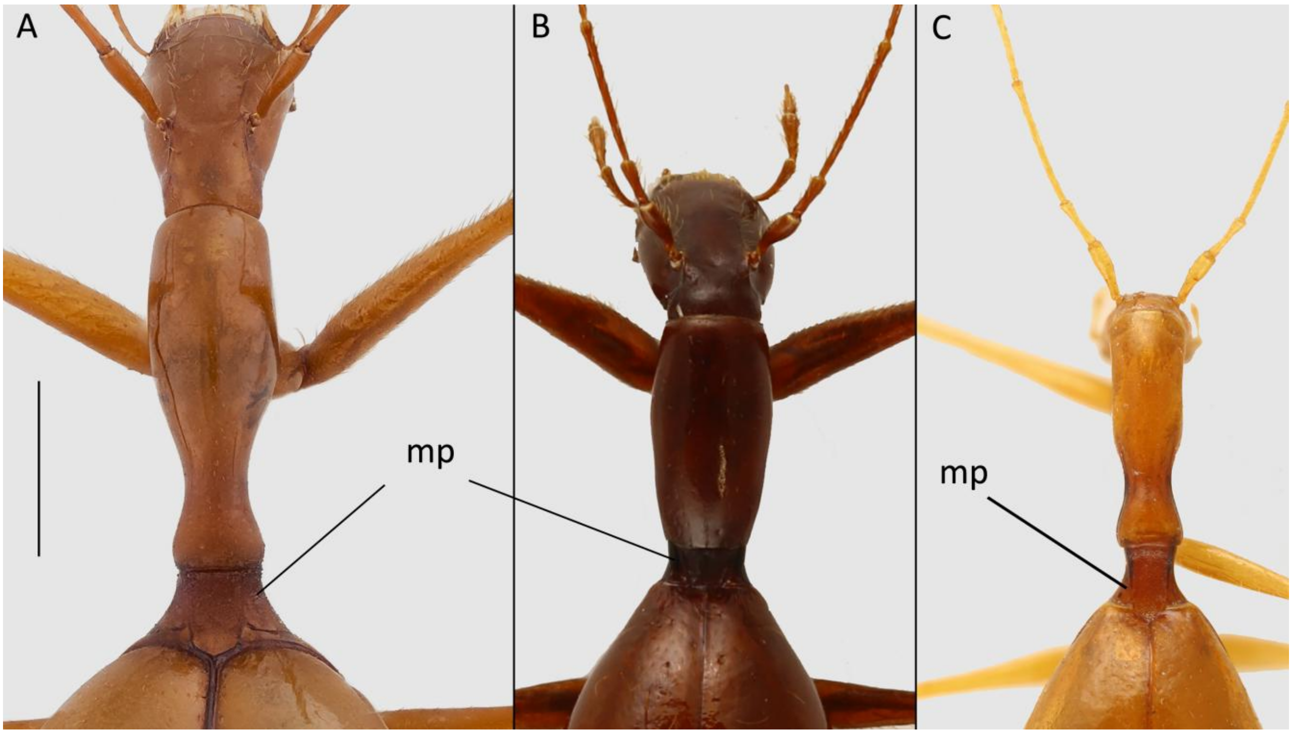

Fig. 3. Pronotum and mesothoracic pedunculus: A. *Graciliella metohijensis*; B. *Anthroherpon cylindricolle cylindricolle*; C. *Leptomeson bujasi*; mp. mesothoracic pedunculus. Scale bar is 1 mm.

3.3. RESULTS AND DISCUSSION

Our study, based on morphological and molecular approaches, reveals that the genus *Anthroherpon* as conventionally defined (Jeannel, 1924; Guéorguiev 1990; Giachino and Gueorguiev, 1993; Giachino and Vailati, 2005; Perreau and Pavićević, 2008) is polyphyletic (Fig. 1). The genus *Leptomeson* forms the highly supported sister clade of one polytypic *Anthroherpon* species, namely *A. apfelbecki* sensu lato and the genus *Hadesia* forms the sister clade of the remaining *Anthroherpon* species. To resolve this polyphyly, we defined one new additional genus, *Graciliella*, comprising *A. apfelbecki* sensu lato and one species, *A. absoloni* (Guéorguiev, 1990), for which we did not have genetic data, but which morphologically closely resembles *A. apfelbecki* (see below under the genus description). Morphological phylogenetic analysis of the species of *Hadesia* was obtained by Perreau and Pavićević (2008). Their parsimony analysis gave a well-supported monophyletic group for *H. vasiceki* Müller, 1911 and *H. lakotai* Perreau and Pavićević, 2008 (bootstrap value 100 %), but the possible sister group *Hadesia asamo* Perreau and Pavićević, 2008 + *H. weiratheri* Zariquiey, 1927 was weakly supported (bootstrap value 50 %). However, analysis based on distances as optimality criterion supported both groups: *H. vasiceki* + *H. lakotai* and *H. asamo* + *H. weiratheri* as sister groups at the same level of bootstrap value (Perreau and Pavićević, 2008). Our phylogenetic tree based on molecular data has a different topology--*H. asamo* and *H. vasiceki* are early-branching and *H. lakotai* and *H. weiratheri* are forming a clade. These results are surprising regarding the distribution of the species. Namely, *H. asamo* and *H. weiratheri* are both located in the Orjen massif in Montenegro while *H. weiratheri* and *H. lakotai* were found in remote massifs, in caves which are more than 50 km apart. Moreover, *H. asamo* and *H. weiratheri* are morphologically extremely similar (Perreau and Pavićević, 2008) which can easily mislead phylogenetic reconstruction by morphological approaches.

The genus *Leptomeson* was originally established as a subgenus of *Anthroherpon* (Jeannel, 1924) and then subsequently raised to genus rank by Guéorguiev (1990). This separation was based on the following characters of *Leptomeson*: more developed mesothoracic pedunculus, with constriction in the middle part and wider in the base, endophallus with differentiated basal 'armature' (sclerified stylet-shaped phanera, Giachino et al., 2011) and parameres bearing 4 setae. Further investigations and discoveries of several new species belonging to this genus have shown that number of setae on parameres varies in fact from 3 to 4 (Giachino et al., 2011). The molecular phylogenetic reconstruction is in accordance with the morphology regarding the separation of the genus *Leptomeson* from *Anthroherpon*. The clade *Graciliella + Leptomeson*, in addition to being clearly supported by the molecular analysis, is also morphologically supported by the following characters that are likely synapomorphies: very elongated pronotum and mesothoracic pedunculus, and mesoventrum without processus between mesocoxae. Mesoventral processus is normally entire (it completely separates the two mesocoxal cavities) in Leptodirini, except in some highly evolved groups such as Anthoherponina in which it is reduced or absent (Jeannel, 1911). We observed that the degree of reduction of this processus is a distinguishing character among Anthroherponina: it is present in the genus *Anthroherpon*, and absent in *Leptomeson* and *Graciliella*. However, this character is still insufficiently explored in other Anthroherponina to assess its relevance in non-Anthroherponine Leptodirini. For instance, *Anthroherpon stenocephalum* (Apfelbeck, 1910) is the only species of the genus *Anthroherpon* in which we noticed intrapopulation variability of this character: some specimens have a mesoventral processus between the mesocoxae while it is absent in others, but no intermediate state was observed. The monophyly of the genus *Leptomeson* is morphologically supported by the presence of the sclerotized structures in the endophallus (Fig. 2O), which are absent in *Graciliella,* and the shape of mesothoracic pedunculus with the constriction in the mid-section or posterior of the mid-section (Fig. 3C).

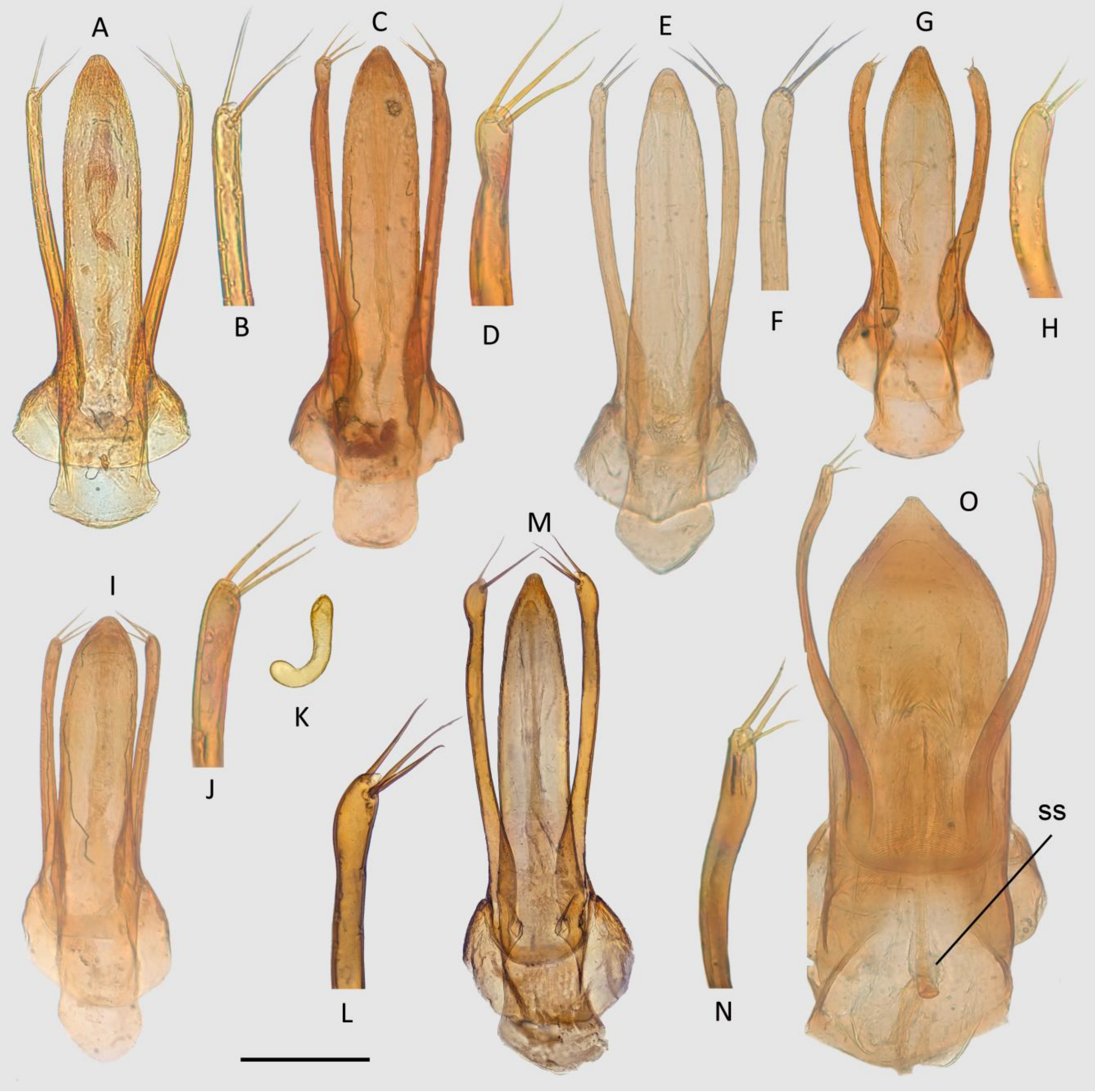

Fig. 2. Genital structures of *Graciliella* and *Leptomeson*, complete aedeagus + detail of paramere apex: A-B. *G. apfelbecki apfelbecki*; C-D. *G. metohijensis*; E-F. G. absoloni; G-H. *G. kosovaci* sp. n.; I-J. *G. ozimeci* sp. n.; L-M. *G. lahneri*; N-O. *L. leonhardi*; ss. sclerified structure in the endophallus; K. spermatheca of *G. ozimeci* sp. n. Scale bar is 0.1 mm.

A dorsal comb of very short bristles on the terminal maxillary palpomere is present in three genera that we analyzed in this study (*Anthroherpon, Leptomeson,* and *Graciliella*). However, it is absent in *Hadesia*. We could find no previous literature regarding this character so that we believe it was not previously observed. Given the scarcity of nutrients in the subterranean environment, this comb is possibly an extra sensory organ that has a yet to be determined role in detecting food.

Our molecular analysis shows that the four *Graciliella* species are genetically well-separated, with COI-differences between 3.9 and 12.4%, values that are normally found at or above the species level in Cholevinae (Schilthuizen et al., 2011) (Fig. 4). To confirm that our mostly genetically circumscribed species indeed correspond with morphologically recognizable units, we carried out a linear discriminant analysis (LDA) based on 40 linear morphometric measurements. An LDA is especially suited for maximizing inter-group separation and minimizing intra-group separation for pre-defined units (McLachlan, 2004). This LDA confirmed that the interspecific morphological differences, however subtle and minute as they may seem, are measurable and reproducible. Along the first two axes (with major loadings for tibia lengths, lengths of antennomeres 4 and 11, and elytra length), all species can be distinguished, except the two new species, *G. kosovaci* and *G. ozimeci* (Fig. 5A). These latter two species, however, are well-separated along the third discriminant axis, in which there are additional loadings for the first meso- and metatarsomere (Fig. 5B). Based on this independent, initial morphometric separation, we then detected additional qualitative diagnostic characters in setation, shape of pronotum, aedeagus, and microsculpture. The diagnoses and identification keys are largely based on these qualitative characters, as they are more easily accessible.

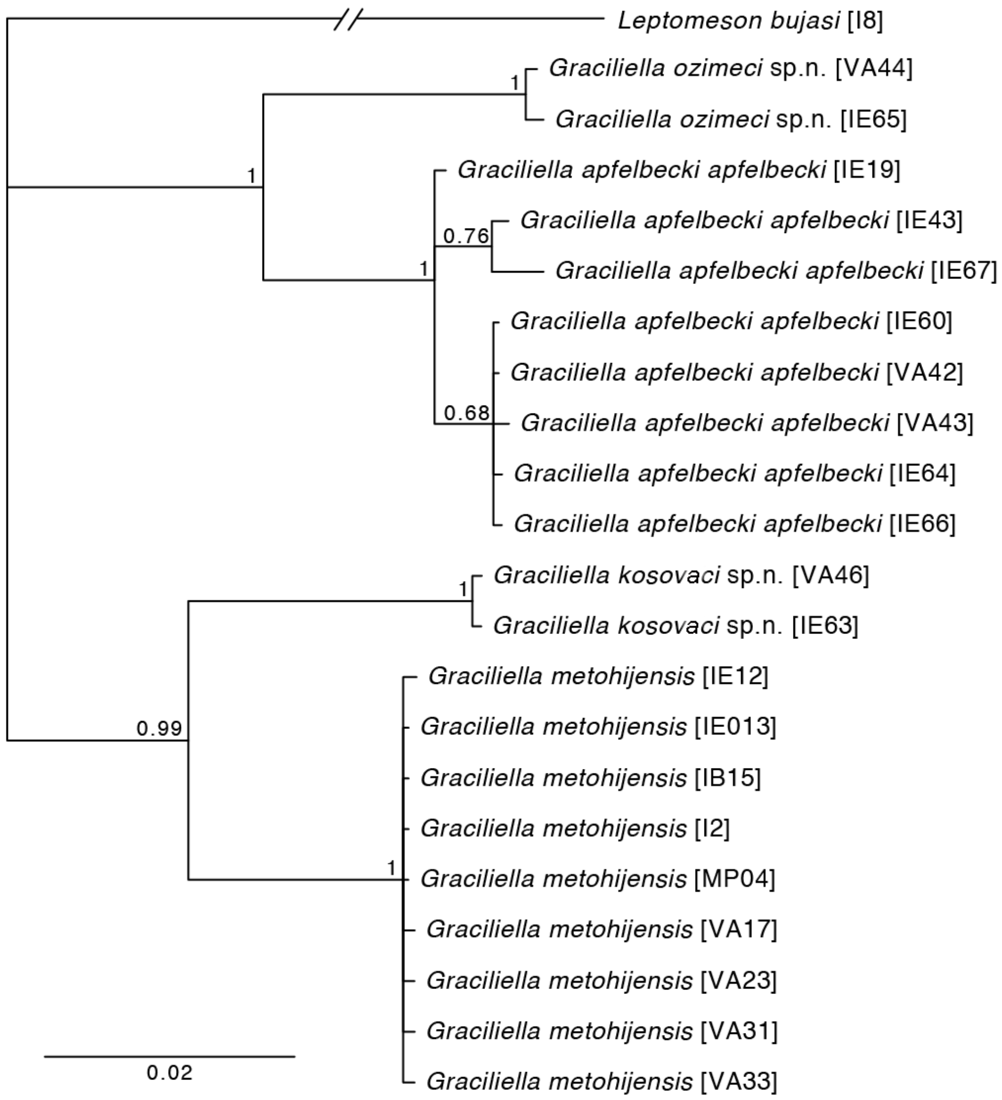

Fig. 4. Bayesian inference of combined COIa and COIb data sets for *Graciliella*. Numbers above nodes are posterior probabilities.

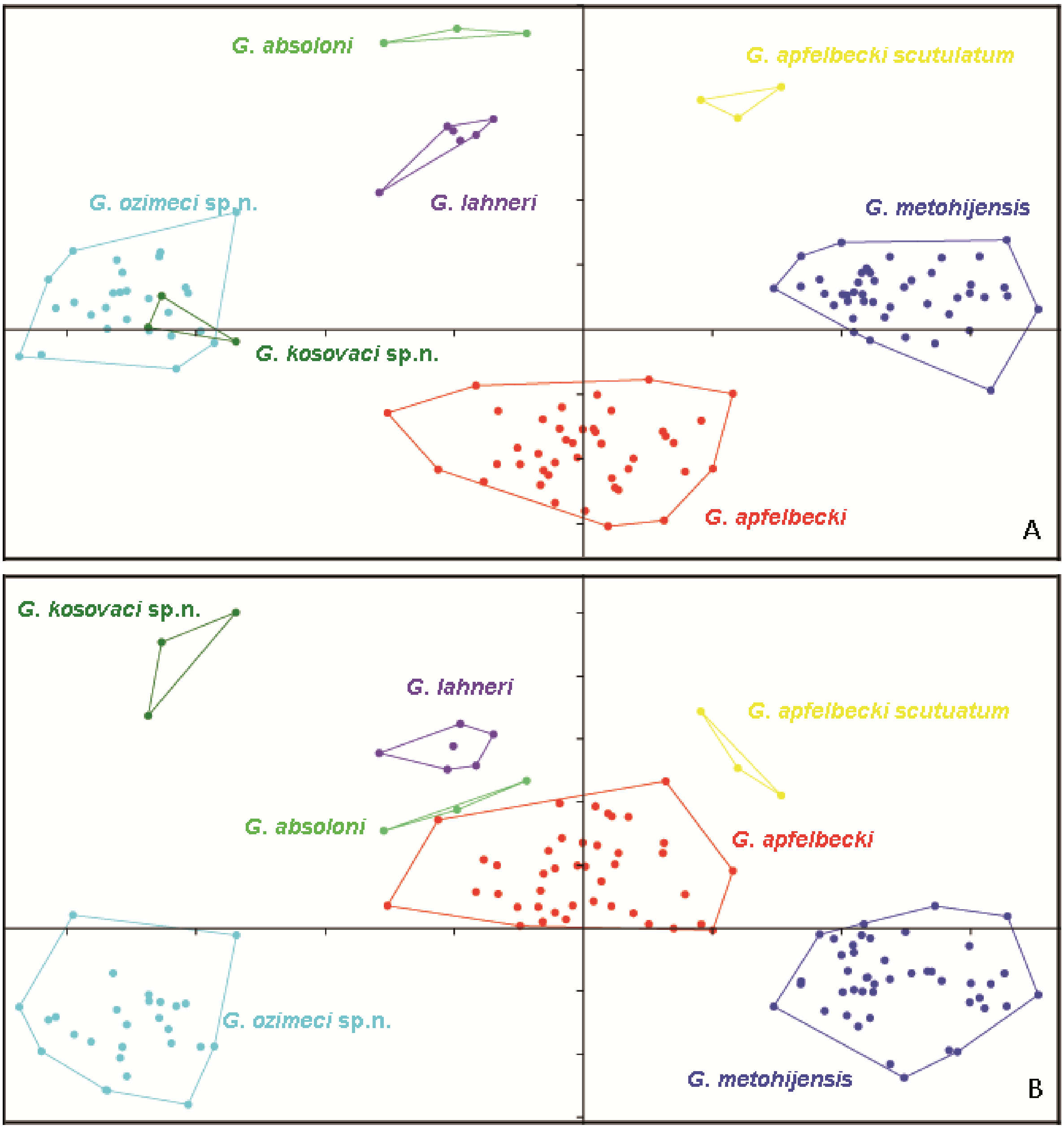

Fig. 5. Linear discriminant analysis (LDA) based on 40 linear morphometric measurements. A. Axes 1 and 2; B. Axes 1 and 3.

Perreau and Pavićević (2008) reconstructed the phylogeny of the subtribe Anthroherponina based on 11 morphological characters. Their most parsimonious tree shows *Leptomeson* and *Anthroherpon* as sister genera, and *Hadesia* distantly related, as sister genus of *Kircheria* Giachino and Vailati (2006). The character states that Perreau and Pavićević defined for *Anthroherpon* also apply to all species of *Graciliella*. If we rather accept our molecular phylogeny, in which *Graciliella* is a sister clade of *Leptomeson*, and *Anthroherpon* s.str. sister clade of *Hadesia*, this would imply that a large number of morphological characters are strongly homoplasious. These characters are: female ventrite VIII with an anterior apophysis versus without, aedeagus with internal sclerified structure versus without, abdominal ventrites with glabrous plates versus without glabrous plates, female first ventrite with lateral hollows versus without, third maxillary palpomere short versus long, claws narrow versus wide. Although many of these characters seem reliable and have traditionally been accepted as indicative of Anthroherponina classification, our results suggest that perhaps this could be attributed to convergence due to shared selective pressures rather than to shared ancestry.

3.4. SYSTEMATIC PART

Above, where we discuss the results of molecular phylogenetics, we show that the genus *Anthroherpon* is polyphyletic. For this reason we erect a new genus, *Graciliella*. In this systematic section, we describe and diagnose this genus and its constituent species. For full treatment of molecular phylogenetics we refer to the Results section.

http://zoobank.org/urn:lsid:zoobank.org:pub: 0A490485-E40F-4422-8D34-D78A92716FB0

Graciliella **n. gen.**

urn:lsid:zoobank.org:act:B0FA4AC2-76D4-4208-8053-317D44E61E42

Type species: *Graciliella apfelbecki apfelbecki* (Müller, 1910)

Description. With a body length from 7.09 to 8.63 mm (HT 8.33 mm), this new genus is one of the largest-bodied Leptodirini known from the Dinaric Mountains. General morphology leptodiroid; pronotum and mesothoracic pedunculus very elongated, elytra physogastric; eyeless, wingless, and depigmented.

Head. Elongated and hypognathous, wider in the anterior part and wider than the pronotum, HL 1.33-1.58 mm (HT 1.55 mm), HW 0.75-0.93 mm (HT 0.83 mm). Dorsal surface shiny or matte, with superficial microreticulation visible at 40 x magnification under a stereomicroscope and with sparse and short hairs. Few random punctures present on the dorsal surface of the head and most numerous on the frons. Dorsal side of the occiput bearing two patches of punctures with very short hairs on both sides of the head. Labrum trapezoid with setae longer than on the frons. Maxillae long, both galea and lacinia elongated and with brushes of setae. Maxillary palps with 4 articles; second maxillary palpomere arched and about the same length as the third one; terminal maxillary palpomere is the shortest and conical in shape; mean MP2/MP3: 1.33. The base of the terminal maxillary palpomere with a dorsal comb of very short bristles (Fig. 6). Labium with trapezoid submentum bearing two long setae. Mandibles angulate in dorsal view, with sharp apex and with 3 or 4 small teeth between the apex and the last big tooth. Antennal insertions are located in the posterior quarter of the length of the head. Antennae longer than the body, second antennomere is the shortest and last one is longer than the penultimate in both sexes. All antennomeres are covered with hairs of medium length, but only antennomeres 7-11 bear up to 8 long, erected setae arranged circularly and symmetrically in apical part (Fig. 7A). Three last antennomeres (9, 10, 11) bear very short and thin hairs inserted between the hairs of medium length and long apical setae. These short, thin hairs are the most numerous on the last antennomere where they form dense covering (Fig. 7B).

Pronotum. Distinctly longer than wide, narrow, and with a strong constriction in the posterior quarter. Maximal width is in the second third of its length; Pwmax: 0.6-0.8 mm (HT 0.72 mm); Pwmin: 0.3-0.45 mm (HT 0.36); PL: 1.77-2.27 mm (HT 2.18). Sides divergent in the anterior part of the pronotum, before its maximal width and convergent in the posterior quarter of the length of the pronotum. Surface shiny or matte, with evident microreticulation visible at 40 x magnification under a stereomicroscope. Without or with few very short hairs, mostly in the posterior part.

Mesothoracic pedunculus. Elongated, PL/MPl: 4-5.3 mm (HT 5.06); slightly wider (minimal width) than long or as long as wide; minimal width in the anterior part; MPl: 0.31-0.48 mm (HT 0.43), MPwmin: 0.42-0.55 mm (HT 0.51). Ecusson (scutum+scutellum; Jeannel, 1911) rough, 'U'-shaped and with microsculptural mesh resembling honeycomb pattern.

Elytra. Elliptic, physogastric, and wide; maximum width in the anterior part of the mid-section, at the mid-section, or slightly posterior of the mid-section, EL: 3.48-4.54 mm (HT 4.16), EW: 2.19-3.42 (HT 2.78). Surface smooth, shiny or matte, generally with no microreticulation except in some specimens of *Graciliella ozimeci* sp. n. Punctuation superficial, spaced, and irregular; if present, hairs are sparse, short or long.

Abdomen. Transversal carina on the posterior margin of mesoventrum non-interrupted, without mesoventral processus; mesocoxal cavities widely confluent (Fig. 8B). Abdominal ventrites densely covered with short hairs except in the anterior part where they are smooth.

Legs. Long, slender, and pubescent. Femora enlarged in the basal parts, tibiae straight or slightly curved inwards (protibiae). Male protarsi with five protarsomeres, female protarsi with four protarsomeres, mesotarsi and metatarsi with five tarsomeres in both sexes. Tarsi not dilated, tarsal empodium with two long setae.

Aedeagus (Fig. 2A-J, L, M). Slender, median lobe with rounded, lanceolate or acute apex, endophallus without 'sclerified stylet-shaped phanera in the median part' (Giachino et al., 2011) *i.e.*, endophallus without sclerotized structures. Parameres as long as or shorter than the median lobe (setae excluded), bearing three apical setae. Urite IX reduced to a ring, as in most Leptodirini.

Female genitalia. Abdominal ventrite VIII with a long and narrow anterior expansion. Appendicular parts (gonocoxites and gonosubcoxites, cf. Deuve, 2001 for terminology) of the urite IX absent. Spermatheca: weakly sclerified, 'J'-shaped, short and slightly widened in the apical region (Fig. 2K).

The genus comprises the following taxa that were previously placed in '*hoermanni'* species group of the genus *Anthroherpon*:

Graciliella absoloni (Guéorguiev, 1990) n. comb.
Graciliella apfelbecki apfelbecki (Müller, 1910) n. comb.
Graciliella apfelbecki scutulatum (Giachino and Guéorguiev, 1993) n. comb.
Graciliella apfelbecki schwienbacheri (Giachino and Vailati, 2005) n. comb.
Graciliella apfelbecki sculptifrons (Winkler, 1925) n. comb.
Graciliella lahneri (Matcha, 1916) n. comb., n. stat
Graciliella metohijensis (Zariquiey, 1927) n. comb., n. stat
Graciliella kosovaci sp. n.
Graciliella ozimeci sp. n.

After removing these taxa, the '*hoermanni'* species group comprises: *Anthroherpon hoermanni hoermanni* (Apfelbeck, 1889), *A. hoermanni hoffmanni* Giachino and Guéorguiev, 1993, *A. hoermanni hypsophilum* Apfelbeck, 1907, *A. hoermanni orlovacensis*, Guéorguiev, 1990, *A. hoermanni sericeum* Jeannel, 1930, *A. scutariensis* Giachino and Guéorguiev, 1993.

Cross diagnosis. The new genus can be easily distinguished from the genus *Anthroherpon*, even with the naked eye, by the very elongated mesothoracic pedunculus, narrowest in the anterior part, shape of pronotum (Fig. 3) and more spherical-shaped elytra. Additional difference is in the mesoventral processus which is absent in *Graciliella* and present in *Anthroherpon* (Fig. 8). It differs from the genus *Leptomeson* by the following combination of characters: larger body size (from 7.09 to 8.63 mm in *Graciliella* and from 4.63 to 7.22 mm in *Leptomeson*), shape of mesothoracic pedunculus and elongated male genitalia without sclerotized structures in the endophallus (Fig. 2A-J, L, M). In *Graciliella*, the narrowest part of the mesothoracic pedunculus is in the anterior part while in *Leptomeson* it is in the mid-section or posterior of the mid-section (Fig. 3).

Distribution. The distribution areas are shown on the map of Fig. 9.

Etymology. The new generic name derives from the Latin word "*gracilis*" which means slender and refers to the habitus of the genus.

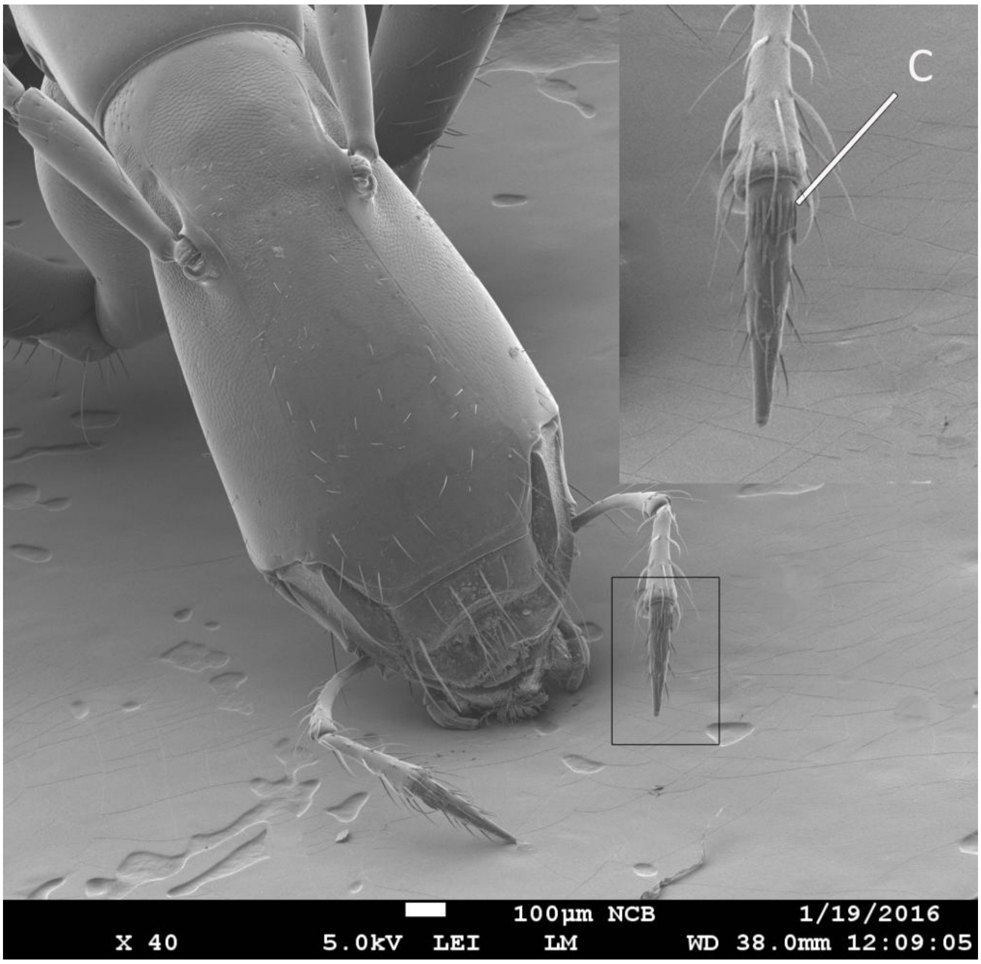

Fig. 6. Terminal maxillary palpomere with a dorsal comb of bristles (c).

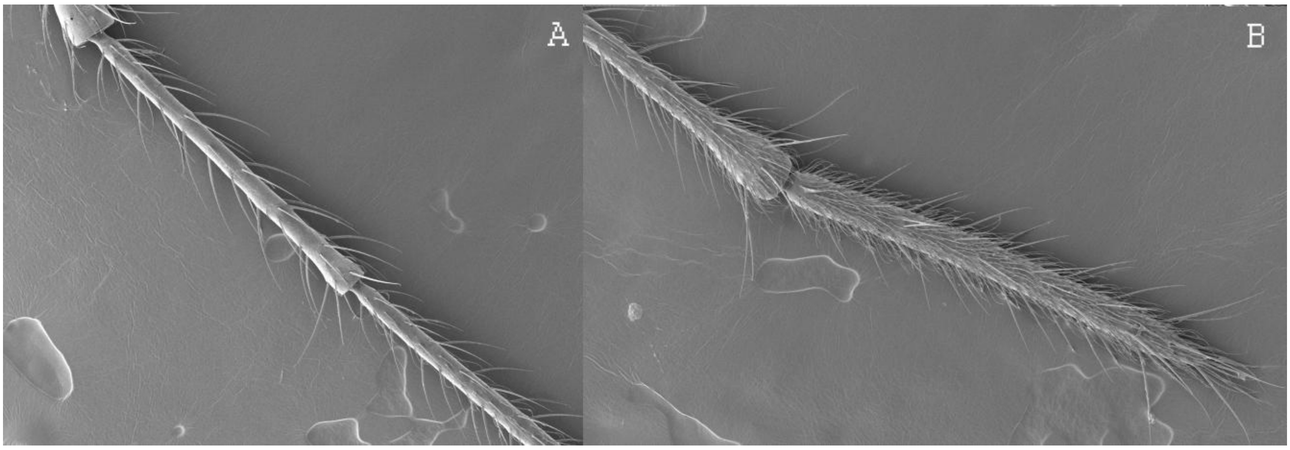

Fig. 7. Antennomeres: A. antennomeres 7 and 8; B. antennomeres 10 and 11.

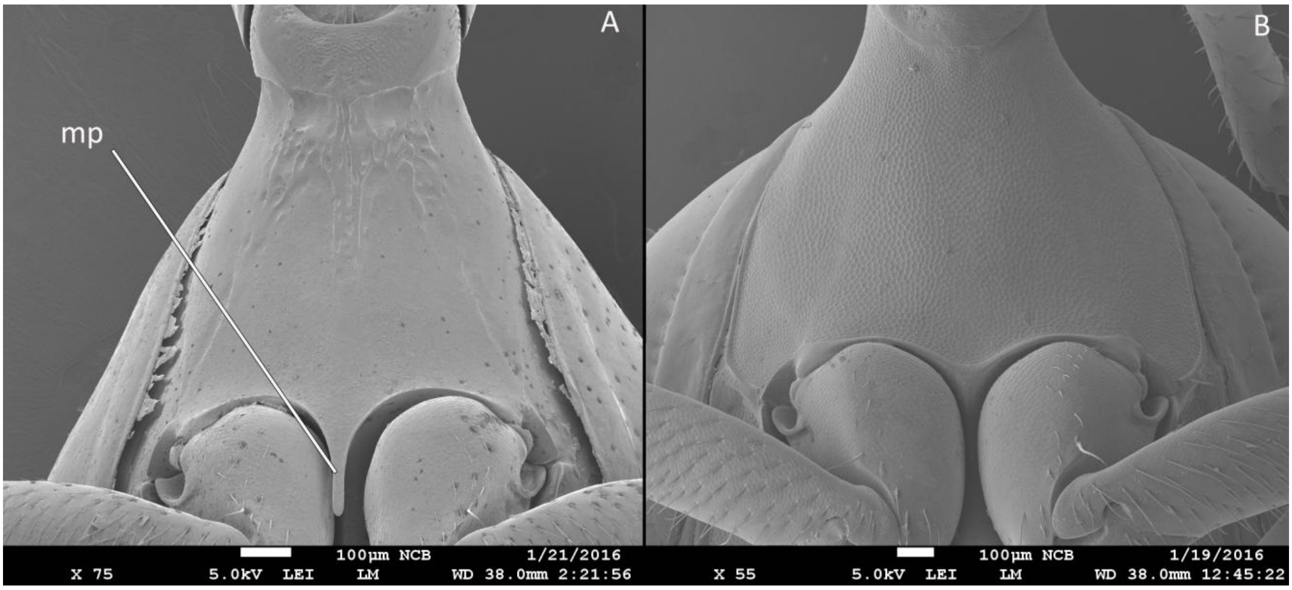

Fig. 8. Mesoventrum of *Anthroherpon* and *Graciliella*: A. *Anthroherpon cylindricolle cylindricolle*; B. *Graciliella metohijensis*; mp. mesoventral processus.

3.4.1. Identification key to separate genera

The following identification key of the genera of the subtribe Anthroherponina modifies and updates the respective section of the key given by Perreau and Pavićević (2008):

8. Pronotum and mesothoracic pedunculus less elongated. Mesothoracic pedunculus short or absent; PL/MPl > 5.3 mm (Fig. 3B). Transversal carina on the posterior margin of mesoventrum interrupted, mesoventrum with long processus between mesocoxae (there is an exception; see: Discussion) (Fig. 8A) ………………………………………………… *Anthroherpon* Reitter

Pronotum and mesothoracic pedunculus very elongated. Mesothoracic pedunculus long, always present; PL/MPl < 5.3 mm. Transversal carina on the posterior margin of mesoventrum non-interrupted, mesoventrum without processus between mesocoxae 9

9. Mesothoracic pedunculus narrowest in the anterior part (Fig. 3A). Aedeagus slender, without sclerotized structures in the endophallus, parameres bearing 3 setae, (Fig. 2A-J, L, M), body size from 7.09 to 8.63 mm .... *Graciliella* n. gen.

Mesothoracic pedunculus narrowest in the mid-section or posterior of the mid-section (Fig. 3C). Aedeagus robust, with sclerotized structures in the endophallus, parameres bearing 3 or 4 setae, (Fig. 2N,O), body size from 4.63 to 7.22 mm .... *Leptomeson* Jeannel

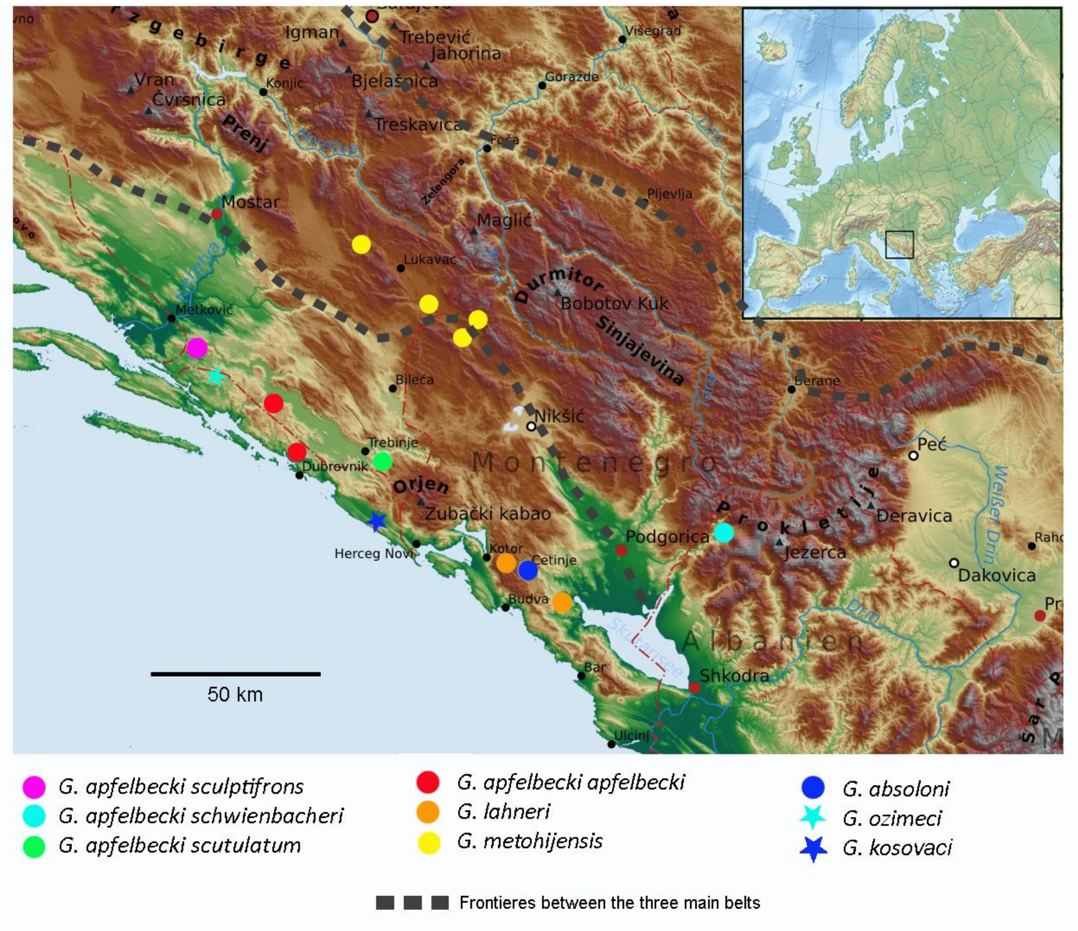

Fig. 9. Distribution map of the species of *Graciliella* in relation to the main geomorphological units of the Dinaric Mountains. Dotted lines separate the three main belts, from south to north: Maritime Belt or Southwestern Belt, the Central Belt or High Dinaric Mountains, and the Northeastern Belt.

***Graciliella kosovaci* sp. n.**

urn:lsid:zoobank.org:act:CCB91AA9-E5D4-4DCE-9070-C5A3A89C2BFB

Type series. Holotype male labeled as follows: 'CROATIA, Popovići, Gruda, Konavle, Kaverna 781, 50 m, 09.11.2014, leg. R. Ozimec' (white label, printed)/ 'HOLOTYPUS *Graciliella kosovaci* sp. n. I. Njunjić, M. Perreau, K. Hendriks, M. Schilthuizen and L. Deharveng det. 2015' (red label, printed), (CNHM). Paratype: 1♀, 09.11.2014, same cave, leg. R. Ozimec, (CNHM). Paratype is labeled with white, printed locality labels and with red printed labels 'PARATYPUS *Graciliella kosovaci* sp. n. I. Njunjić, M. Perreau, K. Hendriks, M. Schilthuizen and L. Deharveng det. 2015'.

Type locality. Croatia, Popovići, Gruda, Konavle, Kaverna 781, 50 m a.s.l.

Diagnosis. Habitus illustrated in Fig. 10A. Distinct from the other species of the genus by the following characters: small size of the body (7.09-7.36 mm); lateral sides of pronotum slightly concave in the anterior part before the maximal width; lateral sides of the mesothoracic pedunculus almost parallel on the anterior third of its length; long hairs on the dorsal surface of the elytra; aedeagus small, median lobe with acute apex, parameres strongly arched inwards, of uniform width, bearing very short setae (Fig. 2G, H).

Description. Body length from 7.09 to 7.36 mm (HT 7.09). General morphology leptodiroid; head wider than pronotum; eyeless; elytra physogastric; wingless, and depigmented.

Head: hypognathous, wider in the anterior part and wider than the pronotum, HL 1.34-1.39 mm (HT 1.39 mm), HW 0.75-0.78 mm (HT 0.78 mm). Surface smooth and shiny with superficial microreticulation and with sparse and short hairs. Few random punctures present on clypeus and along both sides of fronto-genal sulcus.

Pronotum. Narrow and elongated, approximately three times longer than wide, mean PL/Pwmax 3.06 (HT 3.11). Sides divergent and slightly concave in the anterior part before the maximal width, and convergent between the maximal and minimal width of the pronotum. After the minimal width of pronotum, sides are divergent and with rounded posterior margin. Dorsal surface shiny, with superficial microreticulation, and numerous, random and shallow punctures; hairs are not present.

Mesothoracic pedunculus. 1.16 times (HT 1.09) wider than long; lateral sides parallel in the anterior third of the length (measured from the posterior margin of pronotum to the posterior margin of écusson). Ecusson rough, 'U'-shaped and with microsculptural mesh resembling honeycomb pattern.

Elytra. Physogastric and thick in lateral view, maximal width slightly posterior of the mid-section; mean EL/EW: 1.45 mm (HT 1.52). Surface smooth and shiny, microreticulation not visible; punctuation sparse and superficial. Hairs, sparse and present mostly on the dorsal surface, very rare laterally; as long as the medium-sized hairs on the antennae.

Legs. Like in other species of this genus (see genus diagnosis), protibia straight.

Table 3. Antennal formula of the holotype of *Graciliella kosovaci* sp. n.

Article	I	II	III	IV	V	VI	VII	VIII	IX	X	XI
Length (mm)	0.47	0.30	1.02	0.86	1.21	1.08	1.00	0.79	0.88	0.75	1.03

Aedeagus. Small, median lobe longer than parameres (setae included) and with acute apex in dorsal view (Fig. 2G, H). Parameres strongly arched inwards, of uniform width and bearing three short apical setae.

Distribution. Known only from the type locality (Fig. 9). Kaverna 781 is 142 m long and located in south Croatia, in the tunnel 'Konavosko polje-Sea' and is probably a part of the upper floor system of underground channels which naturally drain the water of Konavle polje to the sea. Lack of stalactite deposits indicates that the cave is a relatively young foundation, and the remains of the freshwater cave-dwelling tube worm *Marifugia clavatica* observed on the rock indicates its recent hydrological activity (Cvitanović, 2015).

Etymology. We are glad to dedicate this interesting species to a dear friend, speleologist Petar Kosovac from Belgrade, in recognition for his enthusiastic help in the field and in collecting material.

***Graciliella ozimeci* sp. n.**

urn:lsid:zoobank.org:act:37DB2F08-D301-4637-9023-E948491ABEAA

Type series. Holotype male labeled as follows: 'CROATIA, Dubrovačko primorje, Točionik, Vranja peć, 230 m, 08.11.2014., leg. R. Ozimec' (white label, printed)/ 'HOLOTYPUS *Graciliella ozimeci* sp. n. I. Njunjić, M. Perreau, K. Hendriks, M. Schilthuizen and L. Deharveng det. 2015' (red label, printed), (CNHM). Paratypes: 27♂ 30♀, 08.11.2014, same cave, leg. R. Ozimec, (CNHM, MNHN, CINJ, CMPR, Naturalis). Paratypes are labeled with white, printed locality labels and with red printed labels 'PARATYPUS *Graciliella ozimeci* sp. n. I. Njunjić, M. Perreau, K. Hendriks, M. Schilthuizen and L. Deharveng det. 2015'.

Type locality. Croatia, Dubrovačko primorje, Točionik, Vranja peć, 230 m a.s.l.

Diagnosis. Habitus illustrated in Fig. 10B. Distinct from the other species of the genus by the following characters: body size 7.13-7.5 mm; elytra more elongated, EL/EW: 1.47-1.64 mm; shorter mesothoracic pedunculus (1.38 times wider than long); surface of the body matte. Aedeagus with rounded apex in dorsal view and almost straight parameres.

Description. Body length from 7.13 to 7.50 mm (HT 7.45). General morphology leptodiroid; head wider than pronotum; eyeless; elytra physogastric; wingless, and depigmented.

Head. Hypognathous, wider in the anterior part and wider than pronotum, HL 1.35-1.46 mm (HT 1.46 mm), HW 0.75-0.81 mm (HT 0.81 mm). Surface matte, with superficial microreticulation and with sparse and short hairs on clypeus, genae, and frons, particularly along the dorsal side of fronto-genal sulcus. Last antennomere is shorter in females.

Pronotum. Narrow and elongated, mean PL/Pwmax 2.85 (HT 3.01). Sides almost straight in the anterior part before the maximal width; posterior margin almost straight. Dorsal surface matte and hairless, with superficial microreticulation, and numerous, random and shallow punctures. Ventral side of the pronotum completely smooth behind the first pair of legs.

Mesothoracic pedunculus. 1.38 times (HT 1.53) wider than long; lateral sides parallel in the anterior quarter of the length (measured from the posterior margin of pronotum to the posterior margin of écusson). Ecusson rough, 'U'-shaped and with microsculptural mesh resembling honeycomb pattern.

Elytra. Physogastric but narrow; maximal width slightly posterior of the mid-section; mean EL/EW: 1.47-1.64 mm (HT 1.49). Surface irregular and matte, microreticulation and punctures visible in some specimens. Hairs very sparse and present mostly on the dorsal surface, very rare or absent laterally.

Legs. Like in other species of the genus (see genus description), protibia straight.

Aedeagus. Median lobe longer than parameres (setae excluded) and with rounded apex in dorsal view (Fig. 2I, J). Parameres almost straight, of uniform width and bearing three apical setae.

Table 4. Antennal formula of the holotype of *Graciliella ozimeci* sp. n.

Article	I	II	III	IV	V	VI	VII	VIII	IX	X	XI
Length (mm)	0.51	0.37	1.00	0.92	1.28	1.11	1.07	0.85	0.93	0.76	1.06

Distribution. Known only from the type locality (Fig. 9). Vranja peć is situated in south Croatia, in the municipality Dubrovačko primorje, close to the village Točionik. It is a cave with a vertical entrance, 188 m long and 50 m deep. The bottom of the channel is covered with rock debris and large blocks of rocks broken off from the ceiling. The cave is hydrologically inactive except the small drippings at the bottom (Cvitanović, 2015).

Graciliella apfelbecki apfelbecki **(Müller, 1910) n. comb.**

Antroherpon apfelbecki Müller, 1910: 186. *Type locality.* Höhle zwischen Jasenica und Zavala (Herzegowina) [= pećina Vjetrenica; N 42° 50' 28.34'' E 17° 59' 11.87'', alt. 268 m]
Antroherpon Apfelbecki J. Müller: Jeannel, 1911: 560.
Antroherpon (s. str.) *Apfelbecki* J. Müller, 1910: Jeannel, 1924: 420.
Antroherpon Apfelbecki Müll: Winkler, 1925: 144.
Antroherpon (s. str.) *Apfelbecki* subsp. *Apfelbecki* Müller: Jeannel, 1930: 147.
Antroherpon apfelbecki J. Müller: V. B. Guéorguiev, 1990: 264.
Antroherpon apfelbecki apfelbecki J. Müller, 1910: Giachino and Guéorguiev, 1993: 316.
Antroherpon apfelbecki J. Müller: Perreau and Pavićević, 2008: 211.
Antroherpon Kauti Apfelbeck, 1911: 216. (synonymy in Jeannel, 1914).
Type locality: Herzegowina: in der Vjeternica höhle bei Zavala.
Antroherpon Kauti Apfelbeck, 1916: 346.

Material examined. 1♂: LECTOTYPE (here designated), Hercegovina, 'Vjeternica H.', 11.07.1914., leg. K. Absolon, RMNH.INS.627472 (Naturalis); 1♀: PARALECTOTYPE (here designated), Herzegovina Vjeternica, 1931, leg. K. Absolon (MNHN); 1♂: COTYPE, Höhle bei Savala, Herzegovina (MNHN); 1♂, 1♀: Herzegovina Vjeternica, leg. K. Absolon (MNHN); 1♀: Zavala, Hr., leg. Novak, 5.9.1925 (MNHN); 1♀: Zavala, Herzeg. (MNHN); 1♂: Hercegovina, Neretva, Ravno, Zavala, Vjeternica pećina, Popovo polje, 42.88-17.98 03/VI/2011., leg. M. Perreau, RMHN.INS.550268 (Naturalis); 1♀: same data, RMNH.INS.550269 (Naturalis); 1♀: Vjeternica pećina, Collectie G. Paganetti-Hummler acq. 1937, ZMA.INS.1236005 (Naturalis); 1♀: same data, ZMA.INS.1236007 (Naturalis); 1♂: Hercegovina, Zavala, Dr. Grabowski, Coll. Dr. S. Breuning, ZMA.INS.1235988 (Naturalis); 2♀: BiH, Zavala, Vjeternica., leg. R. Ozimec (CNHM); 1♀: same locality, 21.08.2008., leg. Bakšić Darko (CNHM); 2♂: same locality, 20.08.2007., leg. M. Perreau (CMPR); 1♀: Zavala, Popovo polje, Vjetrenica, Hercegovina, 12.09.1975., leg. Pretner (CMPR); 1♀: Dr. Absolon, Vjeternica (CMPR); 1♂: Vjeternica, Absolon, 1912, Hercegovina (CMPR); 1♀: Kali pećina apud Grebci, Herzegovina, Absolon (CMPR); 3♀: Grabovica bei Grepci, Herzegovina, Absolon (CMPR); 3♂ 3♀: Gjurkovina pećina apud Grepci, Herzegovina, Absolon (CMPR); 2♂ 4♀: Močiljska pec, Rjeka-Dubrov., Svircev, 5.1.931.

(MNHN); 1♂ 1♀: Močiljska pec. Dubrovnik, Svircev, 1.9.923. (MNHN); 2♂: Croatia, Dubrovačko primorje, Dubrovnik, Osojnik, Močiljska špilja, 12.11.2014., leg. Hanžek Nikola (CNHM); 2♀: same locality, leg., R. Ozimec (CNHM); 1♀: Močiljska pećina, Močilje, Ombla, Dalmatia, leg. E. Pretner (CMPR); 1♂: Croatia, Dubrovačko primorje, Dubrovnik, Gromača, Špilja za Gromačkom vlakom, 28.08.2014., leg. Slapnik Rajko (CNHM); 1♀: same locality, 25.09.2014., leg. Slapnik Rajko (CNHM); 1♀: Bosnia and Herzegovina, Bobani, Grabovica, 08.05.2011., leg. R. Ozimec (CNHM); 1♀: Grotte Grabovica, Herzegovina Weirather, 1-14 (MNHN); 1♂: Kali pećina, Grebci, Herceg., Svircev, 2.1.931. (MNHN); 1♂: Gr. Močilje, pr. Ragusa, 30.11.921., G. Müller (MNHN); 2♀: Orlica pecina, Popovo polje, Herzegovina, Absolon (CMPR); 1♀: Ravno Hercegov. Vl. Zoufal (CMPR); 1♂: Bihovo, Trebinje, Herz, Collectie G. Paganetti-Hummler acq. 1937, ZMA.INS.1236013 (Naturalis).

Diagnosis. Habitus illustrated in Fig. 11A. Body length from 7.3 to 8.63 mm. Median lobe with lanceolate apex in dorsal view and considerably longer than parameres (setae excluded). Parameres of uniform width, slightly curved inwards in the apical part and bearing three long apical setae (Fig. 2A, B).

Remarks. Two specimens, one in Naturalis and the other one in MHNH were labelled as types, so we designated the male specimen from Naturalis as lectotype and female specimen from MNHN as paralectotype.

We do not have molecular data for the three following subspecies of *G. apfelbecki,* so we follow the classification of Giachino and Vailati (2005) on this point.

Fig. 10. Habitus of *Graciliella*. A. *G. kosovaci* sp. n.; B. *G. ozmeci* sp. n. Scale bar is 1 mm.

Fig. 11. Habitus of *Graciliella*. A. *G. apfelbecki apfelbecki*; B. *G. metohijensis*. Scale bar is 1 mm.

Graciliella apfelbecki scutulatum (Giachino and Guéorguiev, 1993) n. comb.

Antroherpon scutulatum Giachino and V. B. Guéorguiev, 1993: 317. Type locality: Erzegovina, Bravenik [= Hercegovina, Zupci, Bravenik; N 42°37.143' E 18°25.969', alt. 740 m a.s.l.]
Antroherpon apfelbecki scutulatum Giachino and Guéorguiev: Perreau and Pavićević, 2008: 212.

Material examined: 1♂ 3♀: BiH, Trebinje, Zupci, Bravenik (=Jama u Braveniku), alt. 740m a.s.l., 19.06.2003., leg. M. Perreau (CMPR).

Graciliella apfelbecki schwienbacheri (Giachino and Vailati, 2005) n. comb.

Anthroherpon apfelbecki schwienbacheri Giachino and Vailati, 2005: 159. Type locality: Albania, Kelmend, Tamara umg., Shpella Siper Rruges, 185m a.s.l.

Material examined: We did not examine this subspecies.

Graciliella apfelbecki sculptifrons (Winkler, 1925) n. comb.

Antroherpon Apfelbecki sculptifrons Winkler, 1925: 144. Type locality: In einem Hallenschacht bei der Žaba planina südlich von Metković [=Žaba Mt, Golubinka]
Antroherpon (s. str.) *Apfelbecki* subsp. *sculptifrons* Winkler: Jeannel, 1930: 147.
Antroherpon apfelbecki sculptifrons Winkler: Pretner, 1974: 11.
Material examined: 1♂: TYPE, Žaba plan., Golubinka (NHM).

Graciliella metohijensis (Zariquiey, 1927) n. stat., n. comb.

Antroherpon Apfelbecki subsp. *Metohijensis* Zariquiey, 1927: 163. Type locality: Höhle 216, Urvol P., Nordostherzeg., Karstlande [=Đatlo pećina, Korita, Bosnia and Herzegovina; N 43°03.710' E 18°29.678', alt. 1013 m a.s.l.]
Antroherpon (s. str.) *Apfelbecki* subsp. *metohiensis* Zariquiey: Jeannel, 1930: 147.
Antroherpon apfelbecki metohijense Zariquiey: Pretner, 1974: 11.
Antroherpon apfelbecki metohijensis Zariquiey: Guéorguiev, 1990: 265.
Antroherpon apfelbecki metohijensis Zariquiey, 1928: Giachino and Guéorguiev, 1993, Natura bresciana: 317.

Material examined. 1♂: TYPE, H 216 Urvol p. Weirather, NordostHerzeg. 1922., Karstlande (MNHN); 1♂: BiH, Korita, Kobilja Glava, Đatlo, alt. 1013m, 13.09.2006., leg. M. Perreau (CMPR); 1♀: same locality, 05.08.2013., leg. M. Perreau (CMPR); 1♀: same locality, 15.03.2008, leg. S. Ognjenović (CMPR); 1♀: same locality, 13.07.2007., leg. M. Đokić (CMPR); 3♂: same locality, 23.06.2003., leg. M. Perreau (CMPR); 3♂ 3♀: BiH, Korita, Brestica, Jametina, 14.07.2007., leg. M. Perreau (CMPR); 10♂ 8♀: BiH, Balabani, Drvendžina pećina, 26.08.2013., leg. I. Njunjić (CMPR, CINJ); 3♂ 3♀: Montenegro, Golija Mt, Donje Čarađe, Prljača, 07.08.2010., leg. S. Ognjenović (CDPV); 2♂, same locality, 28.08.2010., leg. I. Njunjić (CINJ); 2 ♀, same locality, 13.08.2010, M. Plećaš (CDPV); 1♂, same locality, same date, leg. M. Perreau (CMPR); 3♀: Montenegro, Kazanci, Bijela pećina, 10.08.2010, leg., M. Perreau; 1♀: Hercegovina, grot

Provalija, 22.07.1962., RMNH.INS.627543 (Naturalis); 1♀: same data, Hercegovina, grot Provalija, 22.07.1962., RMNH.INS.627700 (Naturalis); 1♂ 5♀: Montenegro, Stozi, Stoška pećina, 10.08.2010., leg. I. Njunjić (CINJ); 2♀: same data, leg. S. Ognjenović (CDPV).

Diagnosis. Habitus illustrated in Fig. 11B. Large species, body length from 7.76 to 8.53 mm. Distinct from the other species of the genus by the following characters: elytra strongly physogastric and very wide, mean EL/EW: 1.33, with the largest width slightly before the mid-section and dense but shallow punctuation. Sparse hairs are present mostly on the dorsal surface of the elytra. Pronotum with rounded anterior margin. Parameres shorter than the median lobe (setae excluded), with strong constriction in the preapical region (Fig. 2C, D) in some populations, or without the constriction. Median lobe of the aedeagus with lanceolate apex in dorsal view. This combination of characters supports the change of the status from subspecies to species rank. New status is consistent with the molecular phylogeny, morphometrics and geographic location of this species mostly in the Central Belt of the Dinaric Mountains (Golija Mt., Vojnik Mt., Crvanj Mt., Somina Mt., Korita), rather than in the Southwestern (Maritime) Belt as most of the other species and subspecies of the genus *Graciliella*.

Remarks. There is inter- and intrapopulation variability in the shape of parameres: In some populations (*e.g.*, Drvendžina pećina in Balabani) the parameres have a strong preapical constriction while in others (*e.g.*, Prljača on Golija Mt., Stoška pećina on Somina Mt.), the parameres are of equal width in full-length. In the population from Drvendžina pećina there is a gradient in this character: from evident preapical constriction to parameres without constriction. Some specimens from this population have asymmetric aedeagi: one paramere with constriction and other without. Since we have sequences from only two specimens from the population of Drvendžina pećina, we are unable to assess the taxonomic significance of this variability.

Graciliella lahneri **(Matcha, 1916) n. stat. n. comb**

Antroherpon Lahneri Matcha, 1916: 63. Type locality: Montenegro: Lottspeich Grotte bei Njeguši
Antroherpon (s. str.) *Lahneri* Matcha, 1916: 420.
Antroherpon Lahneri Matcha: Fagniez, 1927: 23.
Antroherpon (s. str.) *Lahneri* Matcha: Jeannel, 1930: 147.
Antroherpon apfelbecki lahneri Matcha: Guéorguiev, 1990: 265.
Antroherpon apfelbecki lahneri Matcha, 1916: Giachino and Guéorguiev, 1993: 317.

Material examined. 1♀: TYPE, Lovćen, Geb. Mal. 1916, Montenegro, Rittm. Matcha, Duboki do (MNHN); 1♀: Montenegro, Lovćen Mt, Bjeloši, Jankova jama, 27.06.2013., leg. I. Njunjić (CINJ); 2♂: Montenegro, Virpazar, Trnovo, Grbočica, 30.05.2004., leg. R. Mlejnek (CMPR); 1♂: Montenegro, Virpazar, Trnovo, Grbočica pećina, cave traps, 21.03-16.07.2008., leg. R. Lohaj, ZMA.INS.1236016 (Naturalis); 1♀: same data, ZMA.INS.1235992 (Naturalis).

Diagnosis. Elytra slightly convex on the disc. Median lobe of the aedeagus elongated and narrow, with acute apex. Parameres as long as the median lobe, with weak preapical constriction and significantly wider in the apical part. Apex of the parameres acute and bearing three setae (Fig. 2L, M). The distinctive shape of the aedeagus allows the change of the status from subspecies to species rank. New status is consistent with the morphometrics and the geographic location of this species in Mountains of Maritime Montenegro.

Graciliella absoloni **(Guéorguiev, 1990) n. comb.**

Antroherpon absoloni Guéorguiev, 1990: 265. Type locality: Montenegro, Lipska pećina près de Cetinje [N 42° 22.433' E 018° 57.207', alt. 456 m a.s.l.]

Material examined. 1♂: TYPE, Montenegro, Lipska pećina près de Cetinje, 10.VIII 1912., leg. J. Matcha (NMP); 1♀: PARATYPE, same data (NMP).

Diagnosis. Body length 7.53 mm (HT). Elongated mesothoracic pedunculus, narrowest in the anterior part, PL/MPl: 4.13 (HT). Pronotum distinctly longer than wide, narrow, and with a strong constriction in the posterior quarter. Maximal width is in the second third of its length; PL: 1.98 mm (HT). Elytra elliptic and physogastric; EL: 3.5 mm (HT). Aedeagus slender, endophallus without sclerotized structures, elongated and with lanceolate apex in dorsal view. Parameres shorter than the median lobe, with weak preapical constriction and slightly wider in the apical part, bearing three setae (Fig. 2E, F). These characters fit in the description of *Graciliella* which allows us to place this species in this new genus.

Identification key

Here we present the identification key to the genus *Graciliella*, excluding the subspecies. It is dedicated to practical identification of species and does not correspond to the molecular phylogeny. Although we have used a few characters previously mentioned in the

Anthroherpon key of Giachino and Vailati (2005), the present key is based chiefly on the diagnoses provided above.

1. Elytra strongly physogastric and very wide, EL/EW: 1.18-1.43 mm, with the largest width slightly before the middle, depressed and attenuated towards the apex ……………………………………………………………………………………………… *metohijensis* (Zariquiey)

Elytra less physogastric and less wide, EL/EW: 1.3-1.64 mm, with the largest width in the mid-section or slightly in the posterior part of the mid-section, not depressed and not attenuated towards the apex ………………………………………………………………………………… 2

2. Anterior part of the pronotum ovoid. Parameres as long as the median lobe, significantly wider in the apical part……………………………………………………………………… *lahneri* (Matcha)

Anterior part of the pronotum subcylindrical. Parameres shorter than the median lobe, same width in the full length or slightly wider in the apical part ……………………………………………3

3. Protibiae weakly curved inwards. Elytra with rounded apex and with the largest width in the middle. Parameres with weak preapical constriction and slightly wider in the apical part. (6.7-7.1 mm) ……………………………………………………………………………*absoloni* (Guéorguiev)

Protibiae straight. Elytra with acute apex and with the largest width slightly posterior of the mid-section. Parameres of uniform width, without constriction in the apical part ……… 4

4. Lateral sides of pronotum convex in the anterior part. Median lobe with lanceolate apex in dorsal view and considerably longer than parameres (setae excluded). Parameres of uniform width, slightly curved inwards in the apical part. Body length 7.3-8.63 mm. ………………………………………………………………………………………………… *apfelbecki* (Müller)

Lateral sides of pronotum concave or almost straight in the anterior part. Median lobe with acute apex or rounded in the apical part, longer than parameres (setae excluded). Parameres strongly arched inwards or almost straight. Body length 7.09-7.5 mm ………5

5. Lateral sides of the pronotum concave in the anterior part before the maximal width. Dorsal surface of the body smooth and shiny. Long hairs on the dorsal surface of the elytra. Aedeagus with acute apex in dorsal view, parameres strongly arched inwards, bearing very short setae………………………………………………………………………………………… *kosovaci* sp.n.

Lateral sides of pronotum almost straight in the anterior part before the maximal width. Dorsal surface of the body matte. Short hairs on the dorsal surface of the elytra. Aedeagus with rounded apex in dorsal view, parameres almost straight, bearing long setae ……………………………………………………………………………………………………… *ozimeci* sp.n.

3.4.2. Biogeography and ecology

The genus *Graciliella* is distributed in subterranean habitats of the Dinaric Mountains, from Crvanj mountain (Bosnia and Herzegovina) in the north, to Trnovo (Montenegro) in the south, and from Žaba mountain (Croatia) in the west to Prokletije mountain (Montenegro) in the east. It is distributed from an altitude of 50 m (*G. kosovaci* in Croatia, Konavle, Kaverna 781) to 1270 m (*G. metohijensis* in Montenegro, Somina, Stoška pećina) above sea level.

Figure 9 shows the geographic distribution of the species of the genus *Graciliella*, in relation to the three main geomorphological units of the Dinaric Mountains: the Maritime Belt or Southwestern Belt, the Central Belt or High Dinaric Mountains, and the Northeastern Belt (Hajna, 2012). The genus *Graciliella* is present only in the Maritime and Central Belt. Even though the cave fauna of the Northeastern Belt is fairly well known, *Graciliella* have never been found in this region. The distribution of *G. apfelbecki schwienbacheri* far from other *apfelbecki* subspecies and in the Central Belt rather than in the Maritime Belt of the Dinaric Mountains where other *apfelbecki* subspecies are distributed indicates that the systematic position of this subspecies should be analyzed using molecular approaches.

In the region of Velež Mt. in Bosnia and Herzegovina all three genera were found--*Leptomeson loreki* Zoufal, 1904, *Anthroherpon ganglbaueri ganglbaueri* (Apfelbeck, 1894) and *Graciliella metohijensis* (Zariquiey, 1927), but never in syntopy. However, in a few caves in Bosnia and Herzegovina (*e.g.* Vjeternica in Zavala, Bravenik in Zupci, Veliko Đatlo in Korita) and Croatia (Špilja za Gromačkom Vlakom in Dubrovačko primorje), *Graciliella* and *Hadesia* coexist in the same cave. The biotope of *Hadesia* differs in many aspects from most other troglobitic species of Leptodirini--these beetles inhabit a semi-aquatic cave habitat called "hygropetric"--they are always found in the vicinity of strong flows of running water on stalagmitic walls and often inside the film of running water (Jeannel, 1924, Remy, 1940, Sket, 2004, Perreau and Pavićević, 2008). *Graciliella*, *Anthroherpon* and *Leptomeson,* however, prefer cave walls without water flow and they have never been found inside the water film itself. Also, they have never been observed on deposits of guano and almost never under the rocks on the cave floor. The representatives of these three genera inhabit humid cave walls, stalactites and stalagmites with almost no visible siltation. Their diet is understudied but it is assumed that they are saprophagous like many other Cholevinae (Beutel and Leschen, 2016) while *Hadesia* is considered to filter water or brush the surface of stalagmitic walls with its specialized mouthparts (Perreau and Pavićević, 2008). The different ecological niches in *Hadesia* and *Graciliella* may allow coexistence without competition.

Online supplementary information

S1. Sequenced specimens, with depository, locality, collectors, and sequence accession numbers.

S2. PCR cycling conditions.

S3. The list of material included in the morphometric analysis (Sheet 1) and list of measured traits (Sheet 2).

S4. The derivations of the 40 linear measurements.

S5. Landmarks recorded on the body of *Graciliella* spp.

3.5. ACKNOWLEDGEMENTS

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3.6. APPENDIX

S1. Sequenced specimens, with depository, locality, collectors, and sequence accession numbers.

Species	Voucher code	Locality	Source	GenBank accession			
				18S	28S	COI (Folmer)	COI (Simon)
Overview of specimens used for figure 1							
Anthroherpon charon (Reitter, 1911)	VA34	BiH, Pale, Ravna pl, Omladinska pecina	I. Njunjić	KU980188	KX121059	KX121088	KX121109
Anthroherpon cylindricolle cylindricolle (Apfelbeck, 1889)	IE3	BiH, Pale, Rogatica, Golubovića pećina	I. Njunjić	KU980189	KX121055	KX121089	KX121110
Anthroherpon ganglbaueri ganglbaueri (Apfelbecki, 1894)	IE11	BiH, Nevesinje, Novakuša	I. Njunjić	KU980190	KX121077	KX121090	KX121111
Anthroherpon harbichi (Reitter, 1913)	IB3	BiH, Sarajevo, Crepoljsko, Kečina stena	I. Njunjić	KU980191	KX121060	KX121091	KX121112
Anthroherpon hoermanni hoermanni (Apfelbeck, 1889)	IB7	BiH, Kalinovik, Miljevina, Borija pećina	I. Njunjić	KU980192	KX121061	KX121092	KX121113
Anthroherpon latipenne latipenne (Apfelbeck, 1907)	IE13	MNE, Orjen, Ericova jama	M. Perreau	KU980184	KX121062	KX121093	KX121114
Anthroherpon matulici (Reitter, 1903)	I5	BiH, Gubar Mt, Bukova rupa	M. Perreau	KU980186	KX121056	-	KX121115
Anthroherpon matzenaueri matzenaueri (Apfelbeck, 1907)	IB13	MNE, Golija, Latično, Jama u Rudinskom dolu	I. Njunjić	KU980193	KX121057	KX121086	KX121116
Anthroherpon primitivum jeanneli (Winkler, 1925)	IE39	BiH, Hercegovina, Turica, Motka, Mravinjac	M. Perreau	KU980187	KX121063	KX121094	KX121117
Anthroherpon pygmaeum stricticolle (Jeannel, 1930)	IE25	BiH, Bjelašnica, Opančak, Ledenjača	I. Njunjić	KU980178	KX121064	KX121085	KX121118
Anthroherpon sinjajevina Njunjić, Perreau, Pavićević, 2015	VA6	Crna Gora, Sinjajevina, Rudnica, Blažova pećina	I. Njunjić	KU980194	KX121065	KX121097	-
Anthroherpon stenocephalum stenocephalum (Apfelbeck, 1910)	IE44	BiH, Olovo, Bijambare	I. Njunjić	KU980195	KX121066	KX121096	KX121119
Anthroherpon taxi taxi (Müller, 1913)	IE55	Crna Gora, Kameno more, Dvestotka	I. Njunjić	KU980185	KX121067	KX121097	KX121120
Anthroherpon weiratheri (Reitter, 1913)	IE21	BiH, Sarajevo, Crepoljsko, Kečina stena	I. Njunjić	KU980179	KX121068	KX121098	KX121121

Anthroherpon zariquieyi (Jeannel, 1930)	IE37	MNE, Durmitor, Vodeni krš, Vodena pećina	I. Njunjić	KU980196	KX121069	KX121099	KX121122
Apholeuonus nudus styrani Apfelbecki, 1906	IAP3	BiH, Kalinovik, Borija, Borija pećina	I. Njunjić	KU980171	KX121082	KX121100	-
Charonites sp. Apfelbeck, 1907	IO33	BiH, Pale, Omladinska pećina	I. Njunjić	KU980172	KX121083	KX121101	KX121123
Graciliella apfelbecki apfelbecki (Müller, 1910)	VA43	CRO, Bobani, Grabovica	R. Ozimec	KU980180	KX121070	KX015840	KX011091
Graciliella kosovaci sp. n.	IE63	CRO, Tunel Konavle, Kaverna 781	R. Ozimec	KU980181	KX121071	-	KX121124
Graciliella metohijensis (Zariquiey, 1927)	IB15	BiH, Nevesinje, Balabani, Drvendžina pećina	I. Njunjić	KU980182	KX121058	KX015829	KX011080
Graciliella ozimeci sp. n.	VA44	CRO, Dubrovnik, Točionik, Vranja pećina	R. Ozimec	KU980183	KX121072	KX015841	KX011092
Hadesia asamo Perreau & Pavićević, 2008	MP01	BiH, Trebinje, Zupci, Bravenik	M. Perreau	KU980174	KX121073	-	KX121125
Hadesia lakotai Perreau & Pavićević, 2008	MP03	BIH, Hercegovina, Korita, Kobilja glava, Đatlo	M. Perreau	KU980175	KX121074	KX121102	KX121126
Hadesia vasiceki Müller, 1911	IH1	BiH, Popovo polje, Vjeternica	M. Perreau	KU980176	KX121075	KX121103	KX121127
Hadesia weiratheri Zariquiey, 1927	IH2	Crna Gora, Orjen, Kameno more, PT4	M. Perreau	KU980177	KX121076	KX121104	KX121128
Leptomeson bujasi Giachino, Bregović, Jalžić, 2011	IL4	Croatia, Brač, Jama kod Matešića stana	B. Jalžić & P. Kutleša	KU980200	KX121080	KX121105	KX121129
Leptomeson dombrowskii dombrowskii (Apfelbeck, 1907)	IL2	Croatia, Dalmacija, Mosor, Vranjača špilja kod Dugopolja	B. Jalžić & H. Bilandžija	KU980198	KX121078	KX121106	KX121130
Leptomeson radjai Giachino, Bregović, Jalžić, 2011	IL1	Croatia, Šibenik, Perković, Zvekača	P. Bregović	KU980197	KX121081	KX121107	KX121131
Leptomeson raguzi Giachino, Bregović, Jalžić, 2011	IL3	BIH, zapadna Hercegovina, Grude, Drinovci, Majića ponor	H. Bilandžija	KU980199	KX121079	-	KX121132
Parapropus sericeus muelleri Jeannel, 1924	IO26	BIH, Sanski most, Hrustovačka pećina	I. Njunjić	KU980173	KX121084	KX121087	KX121133
Speonesiotes sp. Jeannel, 1910	IO39	Hercegovina, Trebinje, Zupci, Vilina pećina	I. Njunjić	KU980170	-	KX121108	KX121134

Overview of additional specimens used for figure 4

Gracieliella apfelbecki apfelbecki (Müller, 1910)	IE60	CRO, Dubrovnik, Osojnik, Močiljska špilja	N. Hanžek			KX015832	KX011084
Graciliella apfelbecki apfelbecki (Müller, 1910)	IE19	BiH, Popovo polje, Vjeternica	M. Perreau			KU958638	-
Graciliella apfelbecki apfelbecki (Müller, 1910)	IE43	BiH, Popovo polje, Vjeternica	R. Ozimec			KU958639	KX011083
Graciliella apfelbecki apfelbecki (Müller, 1910)	IE64	CRO, Dubrovnik, Osojnik, Močiljska špilja	R. Ozimec			-	KX011085
Graciliella apfelbecki apfelbecki (Müller, 1910)	IE66	CRO, Dubrovačko primorje, Špilja za Gromačkom vlakom	R. Slapnik			-	KX011087
Graciliella apfelbecki apfelbecki (Müller, 1910)	IE67	CRO, Bobani, Grabovica	R. Ozimec			KX015833	KX011088
Graciliella apfelbecki apfelbecki (Müller, 1910)	VA42	CRO, Dubrovačko primorje, Špilja za Gromačkom vla	R. Slapnik			KX015839	KX011090

Graciliella kosovaci sp. n.	VA46	CRO, Tunel Konavle, Kaverna 781	R. Ozimec	KX015842	KX011093
Graciliella metohijensis (Zariquiey, 1927)	I2	Crna Gora, Golija, Donje Čarađe, pećina Prljača	M. Perreau	KU958640	KX011078
Graciliella metohijensis (Zariquiey, 1927)	IE013	BiH, Hercegovina, Korita, Kobilja glava, Đatlo	M. Perreau	KX015831	KX011082
Graciliella metohijensis (Zariquiey, 1927)	IE12	BiH, Korita, Jametina	M. Perreau	KX015830	KX011081
Graciliella metohijensis (Zariquiey, 1927)	MP04	BiH, Hercegovina, Korita, Kobilja glava, Đatlo	M. Perreau	KX015834	KX011089
Graciliella metohijensis (Zariquiey, 1927)	VA17	BiH, Korita, Jametina	M. Perreau	KX015835	-
Graciliella metohijensis (Zariquiey, 1927)	VA23	BiH, Nevesinje, Balabani, Drvendžina pećina	I. Njunjić	KX015836	-
Graciliella metohijensis (Zariquiey, 1927)	VA31	BiH, Hercegovina, Korita, Kobilja glava, Đatlo	S. Ognjenović	KX015837	-
Graciliella metohijensis (Zariquiey, 1927)	VA33	Crna Gora, Golija, Donje Čarađe, pećina Prljača	M. Perreau	KX015838	-
Graciliella ozimeci sp. n.	IE65	CRO, Dubrovnik, Točionik, Vranja pećina	R. Ozimec	-	KX011086
Leptomeson bujasi Giachino, Bregović, Jalžić, 2011	I8	Hrvatska, Brač, Selca, Nagorinac, Ješkalovica	B. Jalžić & P. Kutleša	KX015828	KX011078

S2. PCR cycling conditions.

Number of cycles: 40

Temperature (C)	Time (min)
95	3:00
94	0:15
54	0:30
72	0:40
72	5:00
12	∞

S3. The list of material included in the morphometric analysis.

Voucher	Species	Depository	Locality	Date of collecting	Legator
NMP	*Graciliella absoloni*	NMP	Montenegro, Lipska pećina près de Cetinje	10.08.2012.	J. Matcha
IE19	*Graciliella apfelbecki*	CINJ	BIH, Zavala, Vjeternica	20.08.2007.	R. Ozimec
INJ58	*Graciliella apfelbecki*	CNHM	CRO, Bobani, Grabovica	08.05.2011.	R. Ozimec
JC10	*Graciliella apfelbecki*	MNHN	Popovo polje, Grabovica	unknown	unknown
JC12	*Graciliella apfelbecki*	MNHN	BIH, Zavala, Vjeternica	unknown	unknown
JC14	*Graciliella apfelbecki*	MNHN	Dubrovnik, Močiljska pećina	unknown	unknown
JC15	*Graciliella apfelbecki*	MNHN	Rijeka, Dubrov, Močiljska pećina	unknown	unknown
JC16	*Graciliella apfelbecki*	MNHN	Rijeka, Dubrov, Močiljska pećina	unknown	unknown
JC17	*Graciliella apfelbecki*	MNHN	Hercegovina, Grebci, Kali pećina	unknown	unknown
JC9	*Graciliella apfelbecki*	MNHN	BIH, Zavala, Vjeternica	unknown	unknown
MP1	*Graciliella apfelbecki*	CMPR	BIH, Zavala, Vjeternica	20.08.2007.	M. Perreau
MP2	*Graciliella apfelbecki*	CMPR	BIH, Zavala, Vjeternica	20.08.2007.	M. Perreau
MP5	*Graciliella apfelbecki*	CMPR	BIH, Brestica near Korita, Jametina jama	14.08.2007.	M. Perreau
MP6	*Graciliella apfelbecki*	CMPR	BIH, Brestica near Korita, Jametina jama	14.08.2007.	M. Perreau
VA43	*Graciliella apfelbecki*	MNHN	BIH, Zavala, Vjeternica	20.08.2007.	R. Ozimec
INJ96	*Graciliella apfelbecki scutulatum*	CMPR	BIH, Trebinje, Zupci, Bravenik	29.07.2013.	M. Perreau
INJ54	*Graciliella kosovaci*	CNHM	CRO, Tunel Konavle, Kaverna 781	09.11.2014.	R. Ozimec
INJ95	*Graciliella lahneri*	CMPR	CG, Virpazar, Trnovo, Grbočica	28.04.2004.	R. Mlejnek
INJ128	*Graciliella metohijensis*	CMPR	BIH, Korita, Kobilja glava, Veliko Đatlo	13.09.2006.	M. Perreau
INJ32	*Graciliella metohijensis*	CMPR	BIH, Korita, Kobilja glava, Veliko Đatlo	13.09.2006.	M. Perreau
INJ34	*Graciliella metohijensis*	CDPV	BIH, Nevesinje, Balabani, Drvendžina pećina	21.12.2003.	M. Đokić
INJ55	*Graciliella metohijensis*	CINJ	BIH, Nevesinje, Balabani, Drvendžina pećina	26.08.2013.	I. Njunjić
INJ59	*Graciliella metohijensis*	CINJ	BIH, Nevesinje, Balabani, Drvendžina pećina	26.08.2013.	I. Njunjić
INJ60	*Graciliella metohijensis*	CINJ	BIH, Nevesinje, Balabani, Drvendžina pećina	26.08.2013.	I. Njunjić
INJ61	*Graciliella metohijensis*	CINJ	BIH, Nevesinje, Balabani, Drvendžina pećina	26.08.2013.	I. Njunjić

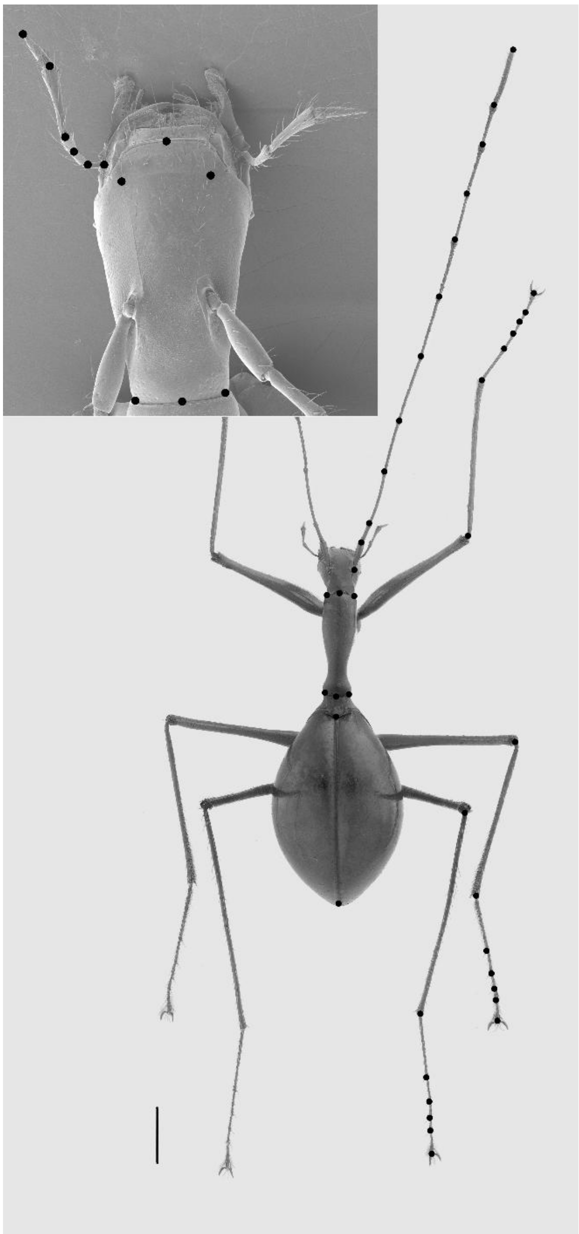
INJ62	*Graciliella metohijensis*	CINJ	BIH, Nevesinje, Balabani, Drvendžina pećina	26.08.2013.	I. Njunjić
INJ63	*Graciliella metohijensis*	CINJ	BIH, Nevesinje, Balabani, Drvendžina pećina	26.08.2013.	I. Njunjić
INJ64	*Graciliella metohijensis*	CINJ	BIH, Nevesinje, Balabani, Drvendžina pećina	26.08.2013.	I. Njunjić
INJ65	*Graciliella metohijensis*	CINJ	BIH, Nevesinje, Balabani, Drvendžina pećina	26.08.2013.	I. Njunjić
INJ66	*Graciliella metohijensis*	CINJ	BIH, Nevesinje, Balabani, Drvendžina pećina	26.08.2013.	I. Njunjić
INJ67	*Graciliella metohijensis*	CINJ	BIH, Nevesinje, Balabani, Drvendžina pećina	26.08.2013.	I. Njunjić
INJ68	*Graciliella metohijensis*	CINJ	BIH, Nevesinje, Balabani, Drvendžina pećina	26.08.2013.	I. Njunjić
INJ89	*Graciliella metohijensis*	CINJ	CG, Somina, Donje Čarađe, Prljača	06.08.2010.	S. Ognjenović
INJ45	*Graciliella ozimeci*	CNHM	CRO, Dubrovnik, Točionik, Vranja pećina	08.11.2014.	R. Ozimec
INJ46	*Graciliella ozimeci*	CNHM	CRO, Dubrovnik, Točionik, Vranja pećina	08.11.2014.	R. Ozimec
INJ47	*Graciliella ozimeci*	CNHM	CRO, Dubrovnik, Točionik, Vranja pećina	08.11.2014.	R. Ozimec
INJ48	*Graciliella ozimeci*	CNHM	CRO, Dubrovnik, Točionik, Vranja pećina	08.11.2014.	R. Ozimec
INJ49	*Graciliella ozimeci*	CNHM	CRO, Dubrovnik, Točionik, Vranja pećina	08.11.2014.	R. Ozimec
INJ51	*Graciliella ozimeci*	CNHM	CRO, Dubrovnik, Točionik, Vranja pećina	08.11.2014.	R. Ozimec
INJ52	*Graciliella ozimeci*	CNHM	CRO, Dubrovnik, Točionik, Vranja pećina	08.11.2014.	R. Ozimec
INJ69	*Graciliella ozimeci*	CNHM	CRO, Dubrovnik, Točionik, Vranja pećina	08.11.2014.	R. Ozimec
INJ70	*Graciliella ozimeci*	CNHM	CRO, Dubrovnik, Točionik, Vranja pećina	08.11.2014.	R. Ozimec

S4. List of measured traits.

Palp2	length of first maxillary palp: sum of three separate distances (see Suppl. Fig. 1)
Palp3	length of second maxillary palp
Palp4	length of third maxillary palp
Ant1	length of first antennomere
Ant2	length of antennomere 2
Ant3	length of antennomere 3
Ant4	length of antennomere 4
Ant5	length of antennomere 5
Ant6	length of antennomere 6
Ant7	length of antennomere 7
Ant8	length of antennomere 8
Ant9	length of antennomere 9
Ant10	length of antennomere 10
Ant11	length of antennomere 11
L1tib	length of protibia
L2tib	length of mesoprotibia
L3tib	length of metaprotibia
L1tarsus1	length of protarsomere 1
L1tarsus2	length of protarsomere 2
L1tarsus3	length of protarsomere 3
L1tarsus4	length of protarsomere 4

L1tarsus5	length of protarsomere 5
L2tarsus1	length of mesotarsomere 1
L2tarsus2	length of mesotarsomere 2
L2tarsus3	length of mesotarsomere 3
L2tarsus4	length of mesotarsomere 4
L2tarsus5	length of mesotarsomere 5
L3tarsus1	length of metatarsomere 1
L3tarsus2	length of metatarsomere 2
L3tarsus3	length of metatarsomere 3
L3tarsus4	length of metatarsomere 4
L3tarsus5	length of metatarsomere 5
Head.Lght.	length of the head
Head.antW	anterior width of the head
Head.postW	posterior width of the head
Pntm.Lgth	length of pronotum
Pntm.antW	anterior width of pronotum
Pntm.postW	posterior width of pronotum
Elyt.Lgth	lenght of elytra
Meso.Lgth	length of mesothoracical pedunculus

S5. Landmarks recorded on the body of *Graciliella* spp.



CHAPTER IV

COMPREHENSIVE EVOLUTIONARY ANALYSIS OF THE *ANTHROHERPON* RADIATION

4.1. INTRODUCTION

The subterranean environment -- isolated, oligotrophic, and ecologically simpler than many other types of habitats -- has long been considered as a "natural laboratory" for evolutionary studies, particularly for the study of speciation and processes of adaptation (Poulson & White 1969; Culver, 1982). Moreover, terrestrial cave animals are also excellent models for biogeographical studies since their present distribution tends to reflect those of ancestral surface ancestors (Ribera et al., 2010; Faille et al., 2010), given that subterranean dispersal is much more restricted. For these reasons, subterranean cave animals have figured prominently in studies about phenotypic adaptation, speciation, endemism, and evolutionary radiation.

Organisms inhabiting the subterranean environment evolve similar suites of morphological, physiological, and behavioural characteristics, known as troglomorphy or troglobiomorphy (Christiansen, 1962). Troglomorphic characteristics include: eye degeneration, apterism, depigmentation, extreme development of sensory organs, longer life cycles, lower metabolic rate, and certain body shape modifications (Racovitza, 1907; Vandel, 1964; Culver et al., 1990). Many evolutionary studies have focused on these peculiar characteristics of cave animals, and different hypotheses have been proposed to explain the evolution of elaborated as well as "regressive" morphological features.

In addition, questions regarding the colonisation of the subterranean environment and the origin of troglobites have been raised since the early development of biospeleology. Various evolutionary scenarios have been proposed to explain the events that have led to the colonisation of these habitats. There are two general hypotheses of speciation in the subterranean environment: the "Climatic Relict Hypothesis", implying an allopatric speciation model (Jeannel, 1943; Vandel, 1964; Barr, 1968; Peck & Finston 1993), and the "Adaptive Shift Hypothesis", implying a parapatric speciation model (Barr & Holsinger, 1985; Howarth, 1982, 1987; Culver, 1987; Peck & Finston 1993; Rivera et al. 2002). Phylogenetic evidence to support or refute these hypotheses has begun to emerge only recently, especially for the most diverse lineages of troglobites such as beetles of the tribe Leptodirini (Caccone & Sbordoni, 2001; Ribera et al., 2010). Although each of these hypotheses could be supported by evidence from certain animal groups or geographic areas, it appears that no unique explanation can encompass all evolutionary transitions of epigean to obligate subterranean species.

Cave fauna is well known for its extremely high degrees of short-range endemism. Many troglobites are strongly and irreversibly adapted to cave conditions (Schiner, 1854; Racovitza, 1907; Sket, 2008). This, plus the fact that cave systems are often geologically isolated, has caused many cave organisms to be restricted to very small ranges. This has usually been explained by their highly reduced dispersal abilities because of intrinsic factors (low vagility, high specialization, small size) as well as extrinsic factors (habitat fragmentation) (Crouau-Roy, 1989; Gibert et al., 1994). However, the relative importance of dispersal and vicariance in the biogeography of subterranean animals is still a matter of debate (Culver et al., 2009). The most recent studies of cave beetles have shown that their present distribution could have been caused by ancient vicariance events (Faille et al., 2010; Ribera et al., 2010).

Until recently, the widely accepted view regarding the radiation of troglobites was that once a lineage has adapted to the subterranean environment within a karst unit, it is unable to expand or diversify over a larger area because of environmental constraints and, as a result, it remains restricted to a small geographical area (Poulson & White, 1969; Barr & Holsinger, 1985; Culver & Pipan, 2009). The existence of widespread ancient troglobitic lineages was usually interpreted as the result of multiple independent colonisations followed by extinction of the ancestors (Jeannel, 1943; Poulson & White, 1969; Culver & Pipan, 2009). Some studies support this hypothesis (for Pyrenean troglobitic Trechini and Leptodirini; Faille et al., 2011; Fresneda et al., 2011), while others suggest a single subterranean colonization event (Faille et al., 2010; Ribera et al., 2010). Still, the evolutionary dynamics and the origin of strictly subterranean lineages with multiple species are understudied.

One group that has undergone extensive diversification in the subterranean environment are beetles of the tribe Leptodirini (Coleoptera: Leiodidae: Cholevinae), one of the largest groups of underground insects (Ribera et al., 2010). Leptodirini have a Palearctic distribution, with the highest diversity in the Mediterranean basin (Perreau, 2000, 2004; Ribera et al., 2010). To resolve their phylogeny, molecular approaches have been initiated by Sbordoni et al. (1980). Applying the technique of the molecular clock, Caccone & Sbordoni (2001) gave a first estimation of a date of separation of the Sardinian from the Iberian-Pyrenean fauna. After this pioneering work, the next study on the molecular phylogenetics of Leptodirini was that of Ribera et al. (2010). The phylogeny of the Western Mediterranean species of Leptodirini including the Pyrenean fauna revealed that the main subterranean lineages became separated before the Early Oligocene (Ribera et al., 2010).

These molecular studies focused on the western Mediterranean. Further east, the Dinaric Mountains have provided uninterrupted conditions for subterranean life for millions of years, and host a rich and diverse cave fauna, with complex, "hotspot-within-hotspot" patterns (Zagmajster et al., 2008). This mountain chain is recognized as having the world's greatest species richness for the subterranean fauna (Sket et al., 2004a; Culver et al., 2006; Deharveng et al., 2012). The Leptodirini are the most species-rich group, comprising 175 species in 50 genera, almost entirely endemic to the Dinarides (Sket, 2005). Within Leptodirini, the subtribe Anthroherponina Jeannel, 1910 (= *Antroherpon* phyletic series of Jeannel, 1924) shows the most pronounced troglomorphic characters and comprises species of exclusively leptodiroid habitus. Studies of these beetles have so far been based only on morphological characters (Jeannel, 1930; Pavićević & Perreau, 2008) and phylogenetic relationships inferred from morphology have not yet been tested with molecular data. The most species-rich genus of the subtribe is *Anthroherpon* Reitter, comprising 26 species and 55 subspecies, and showing some of the most remarkable radiations among all Leptodirini. All species of this genus have developed typical troglomorphic modifications: complete anophthalmy, apterism, extreme elongation of appendages, head, and pronotum, and physogastric elytra. Clearly, to understand the evolutionary history of this group, the troglomorphic adaptations need to be studied in a phylogenetic framework.

We provide here a comprehensive evolutionary analysis of the *Anthroherpon* radiation, using a dated molecular phylogeny as framework for understanding *Anthroherpon* diversification, reconstructing the ancestral range, and exploring troglomorphic diversity.

4.2. MATERIALS AND METHODS

4.2.1. Acronyms

The following abbreviations for collections and institutions are used: CINJ: Iva Njunjić Collection, Belgrade, Serbia; CDPV: Dragan Pavićević Collection, Belgrade, Serbia; CMPR: Michel Perreau Collection, Paris, France; CNHM: Croatian Natural History Museum, Zagreb, Croatia; MNHN: Muséum National d'Histoire Naturelle, Paris, France; Naturalis: Naturalis Biodiversity Center, Leiden, The Netherlands.

4.2.2. Multi-locus molecular phylogenetics

4.2.2.1. Taxon sampling

Specimens were collected in caves and sinkholes of the Dinaric range, in Montenegro, Bosnia and Herzegovina, and Croatia, with the focus on the type localities (Suppl. Table 1). In total, we sampled 45 species of Leptodirini belonging to 12 genera, among which 16 species and 23 subspecies of the genus *Anthroherpon*, covering its entire distribution range. For *Anthroherpon*, two individuals per population were used for amplification and sequencing. We included 8 outgroups from Ribera et al. (2010) belonging to different tribes of Cholevinae (Anemadini, Ptomaphagini, Cholevini). To root the tree we used *Agathidium* from the subfamily Leiodinae. The final data matrix comprised 4143 bp of 113 species from 51 genera.

4.2.2.2. DNA extraction, PCR amplification, and sequencing

The specimens used in the study were collected alive in the field and preserved in 96% ethanol. DNA was extracted from whole specimens or from one leg with a standard phenol–chloroform extraction (Blin & Stafford, 1976) or the DNeasy Tissue Kit (Qiagen GmbH, Hilden, Germany). Voucher specimens are stored in CINJ and CNHM, and DNA aliquots are kept in the tissue collections of Naturalis.

We amplified six fragments of five genes, three mitochondrial and two nuclear loci: (i) two non-overlapping sections of mitochondrial cytochrome c oxidase subunit 1, the 5' and 3' halves, which we here term COIa and COIb, respectively; (ii) the 5' end of the mitochondrial large ribosomal unit plus the Leucine transfer RNA plus the 3' end of NADH dehydrogenase subunit 1 (rrnL-nad1); (iii) an internal fragment of cytochrome b (cob); (iv) the 5' end of the small ribosomal unit, 18S rRNA (SSU); (v) an internal fragment of the large ribosomal unit, 28S rRNA (LSU). Primers used are given in Table 1. Each 25 µl PCR mixture included 1 µl (10 pmol) of each primer, 2.5 µl 10x PCR buffer, 0.5 µl dNTPs, 0.25 µl Taq-polymerase, 18.8 µl ddH2O and 5 µl template DNA. PCR cycles were run at the following conditions: 3 min at 94 °C, followed by 40 cycles of 15 s at 94

°C, 30 s at 54 °C and 40 s at 72 °C, and finally, 5 min at 72 °C. Sequences were assembled and edited using Geneious version 8.0.5 (http://www.geneious.com/, Kearse *et al.*, 2012). DNA sequences obtained for each genetic marker were aligned separately using MAFFT version 7 (Katoh & Standley, 2013).

Table 1. Primers used in the study.

Fragment	**Name**	**Sense**	**Sequence 5’-3’**	**Reference**
COIa	LCOI-1490	F	GGTCAACAAATCATAAAGATATTG	Folmer et al. (1994)
COIa	HCOI-2198	R	TAAACTTCAGGGTGACCAAAAAATCA	Folmer et al. (1994)
COIb	Jerry	F	CAACATTTATTTTGATTTTTTGG	Simon et al. (1994)
COIb	Pat	R	TCCAATGCACTAATCTGCCATATTA	Simon et al. (1994)
cob	CB3	F	GAGGAGCAACTGTAATTACTAA	Barraclough et al. (1999)
cob	CB4	R	AAAAGAAA(AG)TATCATTCAGGTTGAAT	Barraclough et al. (1999)
rrnL-nad1	16Sbi F	F	ACATGATCTGAGTTCAAACCGG	Simon et al. (1994)
rrnL-nad1	FawND1 R	R	TAGAATTAGAAGATCAACCAGC	Simon et al. (1994)
SSU	5’	F	GACAACCTGGTTGATCCTGCCAGT	Shull et al. (2001)
SSU	b5.0	R	TAACCGCAACAACTTTAAT	Shull et al. (2001)
LSU	Ka	F	ACACGGACCAAGGAGTCTAGCATG	Ribera et al. (2010)
LSU	Kb	R	CGTCCTGCTGTCTTAAGTTAC	Ribera et al. (2010)

4.2.2.3. Multi-locus molecular phylogenetics analysis

DNA sequences from the current study (Suppl. Table 1.) and a selection of data from Ribera et al. (2010) were combined using Geneious v9.1.2 (http://www.geneious.com, Kearse et al. 2012) as a workbench. For each genetic marker, an alignment was made using MUSCLE (Edgar, 2004). For LSU and SSU, in some regions, alignment was ambiguous due to large indels; these regions were removed from the final data matrix. Translations of coding genes were checked for consistency and validity.

Prior to the analysis in BEAST we performed Bayesian phylogenetic analyses on the entire dataset, to clarify the taxonomic status of all taxa included in the analyses. The analysis was conducted using MrBayes 3.2.2 (Ronquist & Huelsenbeck, 2003) on the CIPRES webportal (Miller et al., 2010), while data for all markers were linked and the most general substitution model (gamma, with all substitutions possible) was set. Two replicates of 10 x 10^6 generations each were run, sampling values and trees every 5000 generations. Convergence diagnostics were run using Tracer version 1.5 (Rambaut et al., 2014), where ESS values for all parameters were >>200. After discarding a 25% burn-in, the resulting majority-rule consensus tree was visualized using FigTree version 1.4 (Rambaut, 2012). Intersubspecific genetic distances among subspecies in three *Anthroherpon* species (*A. taxi, A. hoermanni*, *A. matzenaueri*) were high (genetic distance in barcoding region >3%), meaning that these may be used as species level units in the BEAST analysis. For this, the specimen-level phylogenies were pruned so that a single operational taxonomic unit (OTU) remained per species.

Phylogenetic analysis was performed in BEAST2 v2.3.2 (Bouckaert et al., 2014) on the CIPRES web portal (Miller et al., 2010), using a single individual per species. Site models for all six sequence alignments were unlinked (i.e. six partitions), with the most general GTR model selected for each partition. A relaxed lognormal clock was chosen, where all partitions were linked together. The Yule model was chosen for the tree prior. The analysis was run twice to check for consistency, each time with a chain length of 100 million generations, sampled and stored every 10 000 generations. Convergence diagnostics were performed using Tracer v1.5 (Rambaut et al., 2014), where full sampling of parameter space and ESS values for all parameters >200 were confirmed. After discarding 25% burn-in, the resulting majority-rule consensus tree was visualized using FigTree v1.4 (Rambaut 2012).

Reliable dating for vicariant events or the age of subterranean habitats of the Dinaric Mts. has not yet been established (Trontelj et al., 2007). Ribera et al. (2010) used the separation of the Sardinian microplates from mainland Europe to calibrate the phylogeny of Western Mediterranean Leptodirini. In line with their findings, we calibrated our phylogeny by setting an expected age of 37.8MY for the sister clade of the Sardinian taxa ("Western Mediterranean Leptodirini"). To this end, we defined a calibration prior on the clade of "Western Mediterranean Leptodirini" with a normal distribution (mean of 37.8 MY, standard deviation of 2 MY) in our BEAST2 run.

4.2.3. Reconstruction of the ancestral range

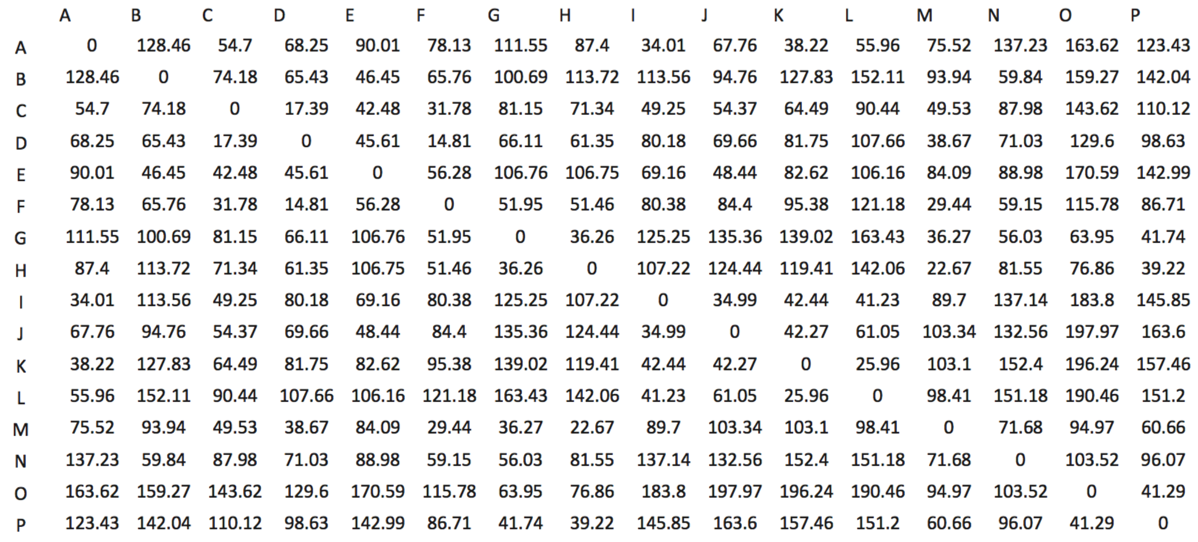
The ancestral geographic range of *Anthroherpon* was inferred using the R package BioGeoBEARS 0.2.1 (Matzke, 2013, 2014a). This model approach directly tests the fit of commonly used biogeographical inference models: dispersal-extinction-cladogenesis model (DEC) (Ree & Smith, 2008), maximum likelihood versions of dispersal-vicariance analysis (DIVALIKE) (Ronquist, 1997), and Bayesian biogeographical inference (BAYAREALIKE) (Landis et al., 2013). The ultrametric tree from the Bayesian relaxed molecular clock analysis, which includes one member per (sub)species, was used as input tree. We identified 16 different geographic areas, corresponding to mountain ranges or caves, inhabited by *Anthroherpon* species. We could not include larger number of areas due to computational limitations. Each species was coded as being present or absent in each of these areas (Table 2), and a maximal number of areas occupied by a single species was set to 3. For a detailed list of the localities see Suppl. Table 1. For all three models, we compared the fit with and without a founder event parameter– "j", which describes a speciation event where a "jump dispersal" event quickly results in an evolutionarily independent lineage (Matzke, 2014a). The "j" parameter allows for a daughter lineage to immediately occupy via long-distance dispersal a new area that is different from the parental lineage (ABCD > ABCD, E). Finally, we tested a novel distance-based dispersal model (+x) where dispersal probability is multiplied by distance to the power "x" (Van Dam & Matzke, 2016). For this purpose we created a matrix indicating distances between selected geographical areas (Table 3). Distances between the area centroides were measured in kilometres on Google Earth (Google Earth, September 2016). These distances were used in the constrained distance-dependent dispersal matrix. In total, we implemented 12 models in BioGeoBEARS (Table 4, 5). The models were compared to each other using two different methods: (1) the Likelihood of all models were compared to each other with Akaike Information Criterion (AIC). This was done in two blocks: all the models without "x" compared to each other, and all the models with "x" compared to each other; (2) the nested models were compared with each other using a chi-squared test. This is mainly to see if models with "j" parameter are preferred or not.

Table 2. List of taxa and their geographic distributions, as included in the biogeographic analysis. Abbreviations of geographic areas as follows: A. Golubovića pećina; B. Mravinjac; C. Zelengora Mt.; D. Lebršnik Mt.; E. Velež Mt.; F. Dobreljica Mt.; G. Moračke planine; H. Sinjajevina Mt.; I. Tebević Mt.+Jahorina Mt.; J. Bjelašnica Mt.; K. Kečina stena; L. Banja pećina; M. Durmitor Mt., N. Orjen Mt.; O. Prokletije Mt., P. Županska pećina. Details of the localities are given in Suppl. Table 1.

Areas in Fig. 3.	A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P
A.cylindricolle cylindricolle	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
A.primitivum jeanneli	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
A.hoermanni hoermanni	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
A.hoermanni sericeum	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
A.hoermanni hypsophilum	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0
A.hoermanni orlovacensis	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0
A.ganglbaueri	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0

ganglbaueri																
A.matulici	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0
A.matzenaueri matzenaueri	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
A.matzenaueri taliensis	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0
A. sinjajevina	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0
A.charon	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0
A.erebus scheibeli	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0
A.pygmaeum stricticolle	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0
A.harbichi	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0
A.weiratheri	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0
A.stenocephalum stenocephalum	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
A.zariquieyi	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
A.latipenne	0	0	0	0	0	1	1	0	0	0	0	0	0	1	0	0
A.t. Taxi + a. T. Sydowi	0	0	0	0	0	0	1	0	0	0	0	0	0	1	0	0
A.taxi albanicum	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0
A.taxi remyi	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1

Table 3. Distances between areas in km. Abbreviations of geographic areas as in Fig. 3.



	A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P
A	0	128.46	54.7	68.25	90.01	78.13	111.55	87.4	34.01	67.76	38.22	55.96	75.52	137.23	163.62	123.43
B	128.46	0	74.18	65.43	46.45	65.76	100.69	113.72	113.56	94.76	127.83	152.11	93.94	59.84	159.27	142.04
C	54.7	74.18	0	17.39	42.48	31.78	81.15	71.34	49.25	54.37	64.49	90.44	49.53	87.98	143.62	110.12
D	68.25	65.43	17.39	0	45.61	14.81	66.11	61.35	80.18	69.66	81.75	107.66	38.67	71.03	129.6	98.63
E	90.01	46.45	42.48	45.61	0	56.28	106.76	106.75	69.16	48.44	82.62	106.16	84.09	88.98	170.59	142.99
F	78.13	65.76	31.78	14.81	56.28	0	51.95	51.46	80.38	84.4	95.38	121.18	29.44	59.15	115.78	86.71
G	111.55	100.69	81.15	66.11	106.76	51.95	0	36.26	125.25	135.36	139.02	163.43	36.27	56.03	63.95	41.74
H	87.4	113.72	71.34	61.35	106.75	51.46	36.26	0	107.22	124.44	119.41	142.06	22.67	81.55	76.86	39.22
I	34.01	113.56	49.25	80.18	69.16	80.38	125.25	107.22	0	34.99	42.44	41.23	89.7	137.14	183.8	145.85
J	67.76	94.76	54.37	69.66	48.44	84.4	135.36	124.44	34.99	0	42.27	61.05	103.34	132.56	197.97	163.6
K	38.22	127.83	64.49	81.75	82.62	95.38	139.02	119.41	42.44	42.27	0	25.96	103.1	152.4	196.24	157.46
L	55.96	152.11	90.44	107.66	106.16	121.18	163.43	142.06	41.23	61.05	25.96	0	98.41	151.18	190.46	151.2
M	75.52	93.94	49.53	38.67	84.09	29.44	36.27	22.67	89.7	103.34	103.1	98.41	0	71.68	94.97	60.66
N	137.23	59.84	87.98	71.03	88.98	59.15	56.03	81.55	137.14	132.56	152.4	151.18	71.68	0	103.52	96.07
O	163.62	159.27	143.62	129.6	170.59	115.78	63.95	76.86	183.8	197.97	196.24	190.46	94.97	103.52	0	41.29
P	123.43	142.04	110.12	98.63	142.99	86.71	41.74	39.22	145.85	163.6	157.46	151.2	60.66	96.07	41.29	0

4.2.4. Linear morphometrics and three-dimensional geometric morphometrics

4.2.4.1. Data collection

Specimens of the genus *Anthroherpon* were mounted on a cardboard on the ventral side of the body and labelled. We subjected a total of 102 specimens belonging to 16 species and 20 subspecies of *Anthroherpon* to morphometric analyses, of which 92 specimens are stored at CINJ, and 10 specimens were on loan from MNHN. To prevent sexual dimorphism from complicating the analyses, we used only males.

Linear and 3D morphometric measurements were obtained with a Micro-Vu Vertex 251HC (https://www.microvu.com/), three-dimensional set-up, using Inspec Metrology Software (https://www.inspec-inc.com/). In total, we recorded 79 landmarks (Fig. 1). Linear measurements were taken on antennae, maxillary palps, head, pronotum, elytra, and legs, using 53 landmarks. Shape analyses were based on 40 landmarks: 6 on the head, 18 on the pronotum, and 16 on the elytra. Each individual was measured three times. In a small number of cases, obvious measurement errors were detected a posteriori (values differing by a factor of >2 from the other two replicates of the same individual). These were then replaced by the average value of the other two replicates. The full list of measured material is given in Suppl. Table 2.

4.2.4.2. *Analysis of morphometric data*

We used the morphometric data in a series of analyses, with the following aims: (i) morphological separation of the genera *Anthroherpon* and *Graciliella*; (ii) exploration of troglomorphic diversity within *Anthroherpon*; (iii) exploration of character evolution within *Anthroherpon*; (iv) character divergence in two syntopic species, *A. harbichi* and *A. weiratheri*.

To compare the shapes of the different individuals, the 3D coordinates were first superimposed for each body part using a Generalized Procrustes Superimposition (GPA) (Dryden & Mardia, 1998). Because the three shapes present an axis of symmetry, they were symmetrized during the superimposition to remove the asymmetric variation (Klingenberg & McIntyre, 1998). The resulting aligned coordinates were then projected from the raw shape space to the tangent shape space before using them as variables to quantify the shapes. The GPA and subsequent analyses of shapes and linear measurements were performed in R (R development core team, 2016), with the packages “ape”, “geomorph”, and “phytools” (Paradis et al., 2004, Adams & Otarola-Castillo, 2013, Revell, 2012).

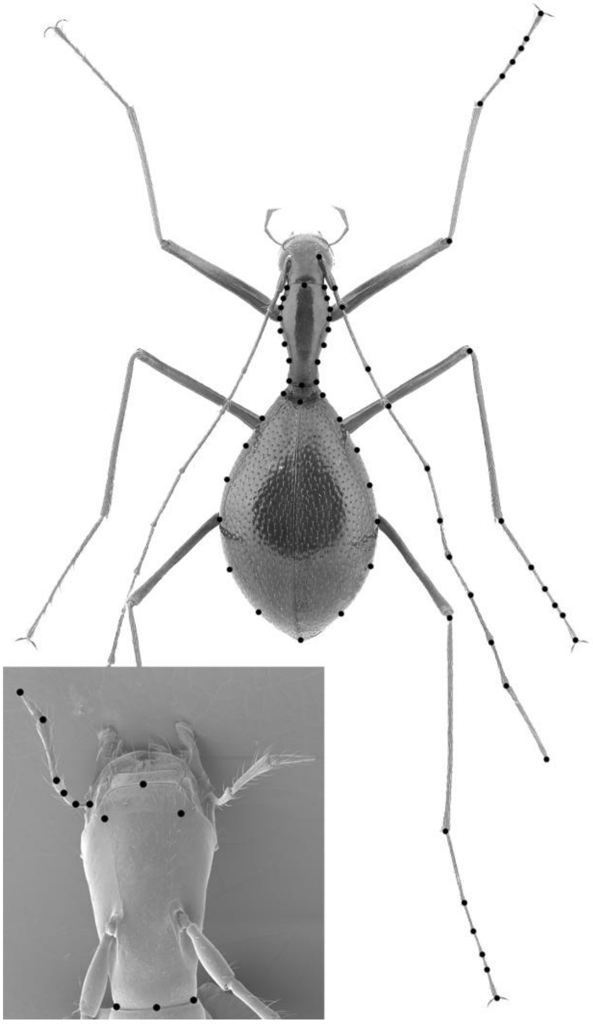


Fig. 1. Landmarks recorded on the body of *Anthroherpon*.

1. *Morphological separation of the genera* Anthroherpon *and* Graciliella

To quantify the habitus shape differences between the newly-described genus *Graciliella* (see Chapter 4) and the genus *Anthroherpon* (s. str.), we analyzed shape differences in the three major body parts (head, pronotum, and elytra). These differences were visualized using deformation vectors from the mean for *Anthroherpon* s. str. (reference) to the mean for *Graciliella* (target). The shape variation among individuals was also explored using Principal Component Analyses on the aligned coordinates of the shapes of each body part.

2. *Exploration of troglomorphic diversity within* Anthroherpon

Degree of troglomorphy in arthropods is often evaluated on the basis of relative elongation of the appendages (palps, antennae, legs). To quantify the degree of troglomorphy within *Anthroherpon* based on these proxies, we compared the lengths of these appendages and their segments relative to body length between species. We also studied the evolution of appendage length in the clade.

3. *Exploration of character evolution within* Anthroherpon

To quantify the habitus shape variation within the genus *Anthroherpon* (s. str.), we analyzed shapes of the three main body parts (head, pronotum, and elytra), using geometric morphometrics. Mean specimens were selected for each species or subspecies to represent the average shapes of the taxa. The data were subjected to character evolution analyses by testing for phylogenetic signal in each of the three body part datasets separately, using the K of Blomberg (Blomberg et al., 2003). Tests were performed using 10 000 permutations, and a risk alpha of 5%. The variation was then explored in the phylomorphospace, which combined a Principal Component Analysis of shape space with the information of shape evolution from the phylogeny (as derived from the molecular phylogenetic analysis—see above).

4. *Character divergence in two syntopic species,* A. harbichi *and* A. weiratheri

Since *A. harbichi* and *A. weiratheri* are two sister species living in syntopy, we aimed at testing whether their speciation may have been accompanied by greater morphological divergence between these two species than expected under morphological drift. To test this hypothesis, we compared the evolution rates of these two species with that of the rest of the tree (Denton & Adams 2015).

4.3. RESULTS

4.3.1. Phylogeny of the subtribe Leptodirina

Our phylogenetic analyses (Fig. 2) revealed that the subtribe Leptodirina is polyphyletic: three genera of this subtribe (*Charonites, Apholeuonus,* and *Parapropus*) form a highly-supported clade which is a sister clade to other members of the tribe Leptodirini, while two genera tentatively placed in Leptodirina (*Remyella* and *Rozajella*) (Perreau & Pavićević, 2008) form a clade with Bathysciina+Bathysciotina. The latter clade is weakly supported (posterior probability 0.76) and also unlikely on morphological grounds. The origin of the clade comprising *Rozajella* and *Remyella* was estimated to have occurred in the Early Oligocene (ca. 32 MYA) while “true” Leptodirina (*Charonites, Apholeuonus,* and *Parapropus*) are of more recent age: Late Oligocene (ca. 25 MYA). For details regarding the phylogeny of *Rozajella* and *Remyella*, see Chapter VI.

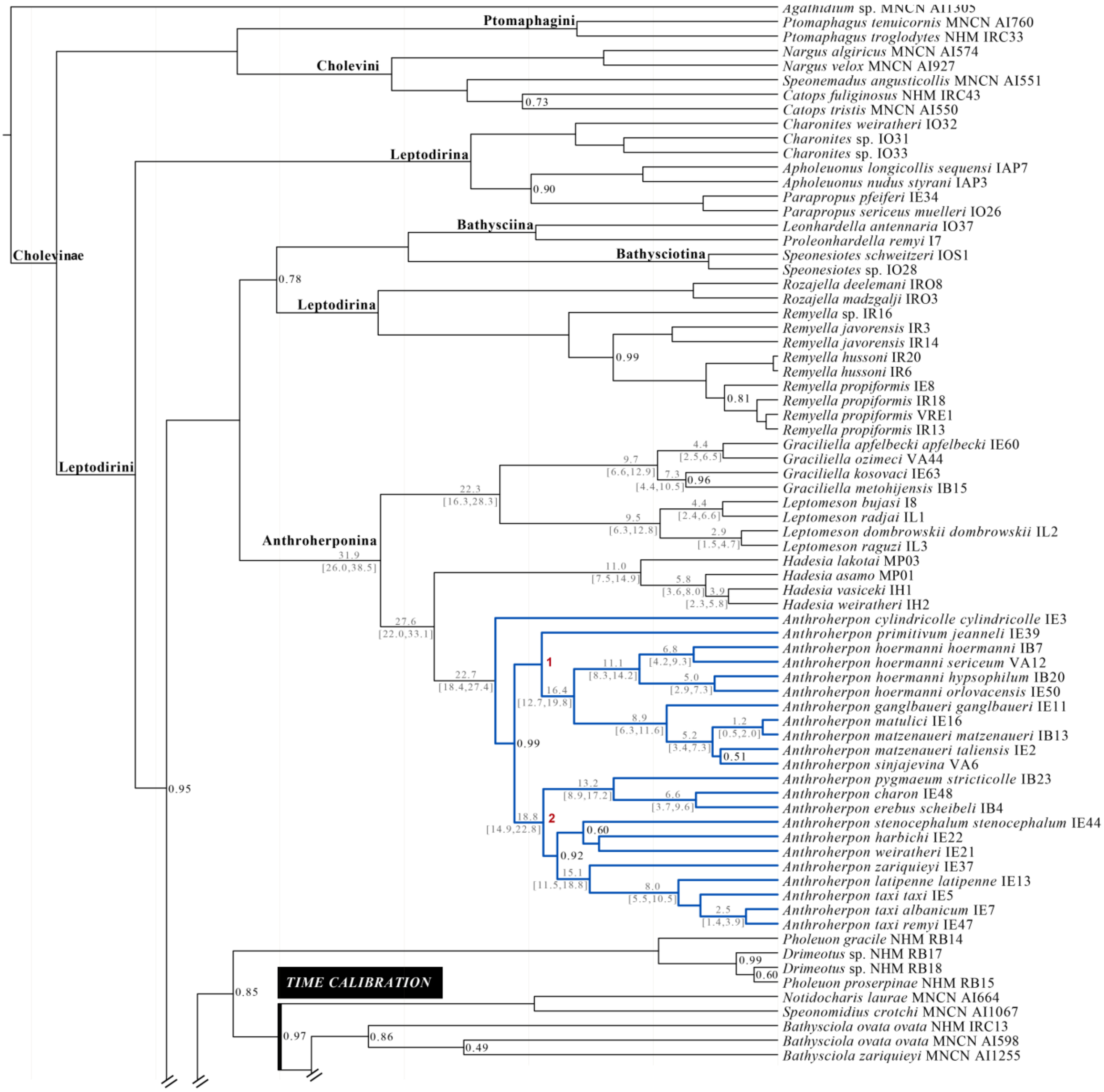
4.3.2. Phylogeny of the subtribe Anthroherponina

The study revealed the monophyletic origin of the analysed taxa of the subtribe Anthroherponina (Fig. 2), which can be dated to the Late Oligocene (ca. 32 MYA). The origins of the four analysed genera of this subtribe (*Graciliella, Leptomeson, Hadesia*, and *Anthroherpon*) were estimated to a relatively narrow time window in the Late Oligocene - Early Miocene. Two major sister clades can be recognized: one comprising the genera *Graciliella* and *Leptomeson* and one comprising *Anthroherpon* and *Hadesia*.

The separation of *Graciliella* and *Leptomeson* was estimated to have taken place in the Early Miocene (ca. 22 MYA), while *Anthroherpon* and *Hadesia* split earlier, in the Late Oligocene (ca. 27 MYA). The four *Graciliella* species are genetically well-separated, with *G. apfelbecki* being a sister species to *G. ozimeci*, and *G. kosovaci* being a sister species to *G. metohijensis*. The diversification of species occurred between 7 and 5 MYA, in the Late Miocene or Pliocene. The start of diversification of the genus *Leptomeson* was estimated to the Late Miocene (ca. 9 MYA), with *L. bujasi* being a sister clade to *L. radjai*, and *L. dombrowski dombrowski* being a sister clade to *L. raguzi.* All four species have diverged in the Pliocene: the first two 4 MYA, and the latter two 3 MYA. Within the genus *Hadesia*, the first species that branches off is *H. lakotai*, which is a sister clade to the three remaining species: *H. asamo, H. vasiceki*, and *H. weiratheri.* The two latter species have split relatively recently, in the Pliocene (ca. 3 MYA).

4.3.3. Phylogeny of the genus *Anthroherpon*

The phylogenetic analysis shows the monophyletic origin of the genus *Anthroherpon*, which was estimated to have started to diverge in the Early Miocene (ca. 22 MYA) (Fig. 2). The most basal clade that branches off contains a single species: *A. cylindricolle s. str*. The genus is otherwise split into two main, highly supported (pp 100%) clades defined by nodes 1 and 2. Both clades started to diverge approximately around the same time (ca. 19 MYA) in the Early Miocene. The clade defined by node 1 is composed of two monophyletic clades comprising species of the "*hoermanni*" and "*ganglbaueri*" species group, and the single species *A. primitivum jeanneli*, which forms a separate clade. All three subclades within the clade defined by node 1 are well-supported (pp 100%). The clade defined by node 2 contains three main clades: one highly supported clade (pp 100%) comprising three species of the "*pygmaeum*" species group, and two sister clades containing the rest of *Anthroherpon* species belonging to the "*pygmaeum*", "*harbichi*", "*stenocephalum*", and "*latipenne*" species groups.



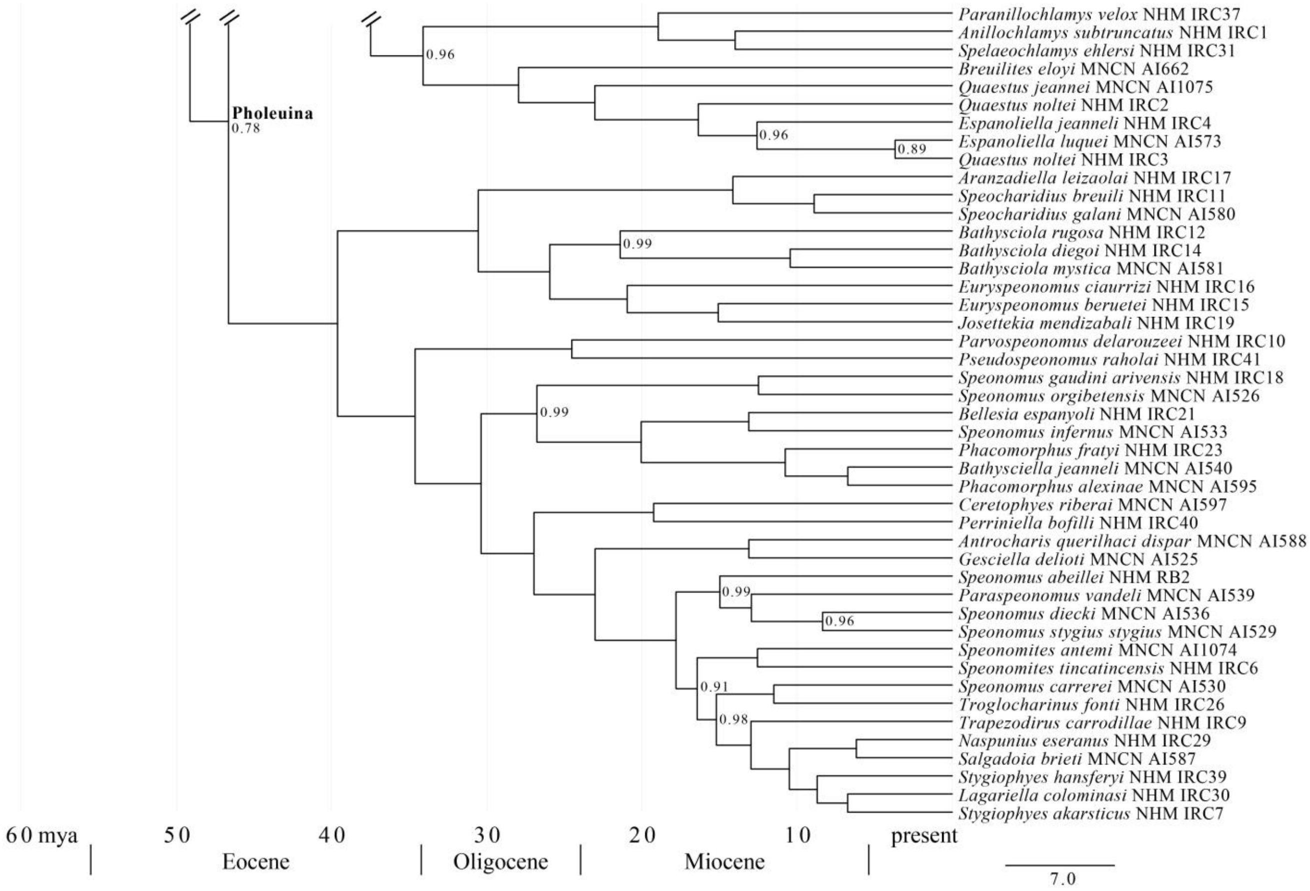


Fig. 2. Ultrametric tree of the phylogeny of the genus *Anthroherpon* obtained with Beast using as calibration the separation of *Bathysciola zariquieyi* from its sister obtained by Ribera et al. (2010). Numbers above nodes, estimated age (in MYA); numbers inside nodes are posterior probabilities<95%; where the number is not indicated pp are >95%. Numbers 1 and 2 in red indicate two major clades of *Anthroherpon*.

4.3.4. Ancestral range reconstruction

The results of ML inference of each biogeographical model are given in Tables 4 and 5. Pairwise likelihood ratio test detected a significantly ($p < 0.05$) better fit of models with a founder effect (j) (results not shown). Model comparisons based on AIC consistently favoured the BAYAREALIKE model with a founder effect (j), either with and without the distances between the areas taken into account. Under this model the most likely ancestral range of the *Anthroherpon* clade consists of the areas F, G, and N (Golija Mt, Moračke planine Mt, and Orjen Mt.) (Figs. 3, 4). From this ancestral range, several single dispersal events into areas A, B, I, K, M, and O occurred. Subsequent dispersal events from these areas explains the occurrence of *Anthroherpon* in the remaining areas.

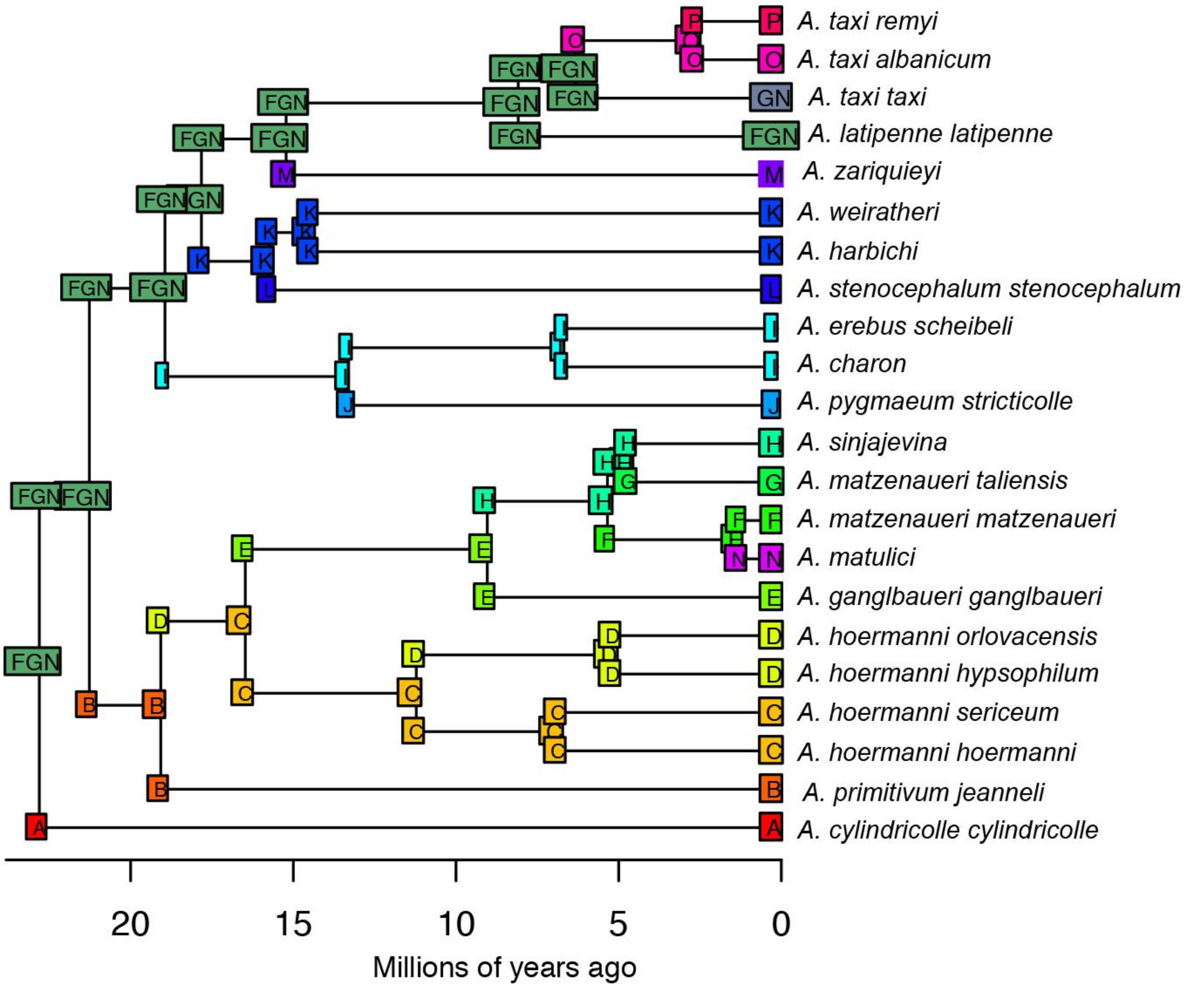


Fig. 3. Preferred model of biogeographic reconstruction (BAYAREALIKE+*j*), according to biogeographic analysis of *Anthroherpon* distribution. Abbreviations of geographic areas as follows: A. Golubovića pećina; B. Mravinjac; C. Zelengora Mt.; D. Lebršnik Mt.; E. Velež Mt.; F. Dobreljica Mt.; G. Moračke planine; H. Sinjajevina Mt.; I. Tebević Mt.+Jahorina Mt.; J. Bjelašnica Mt.; K. Kečina stena; L. Banja pećina; M. Durmitor Mt., N. Orjen Mt.; O. Prokletije Mt., P. Županska pećina. *A. taxi taxi* also includes *A. taxi sydowi* as indicated in the Table 2.

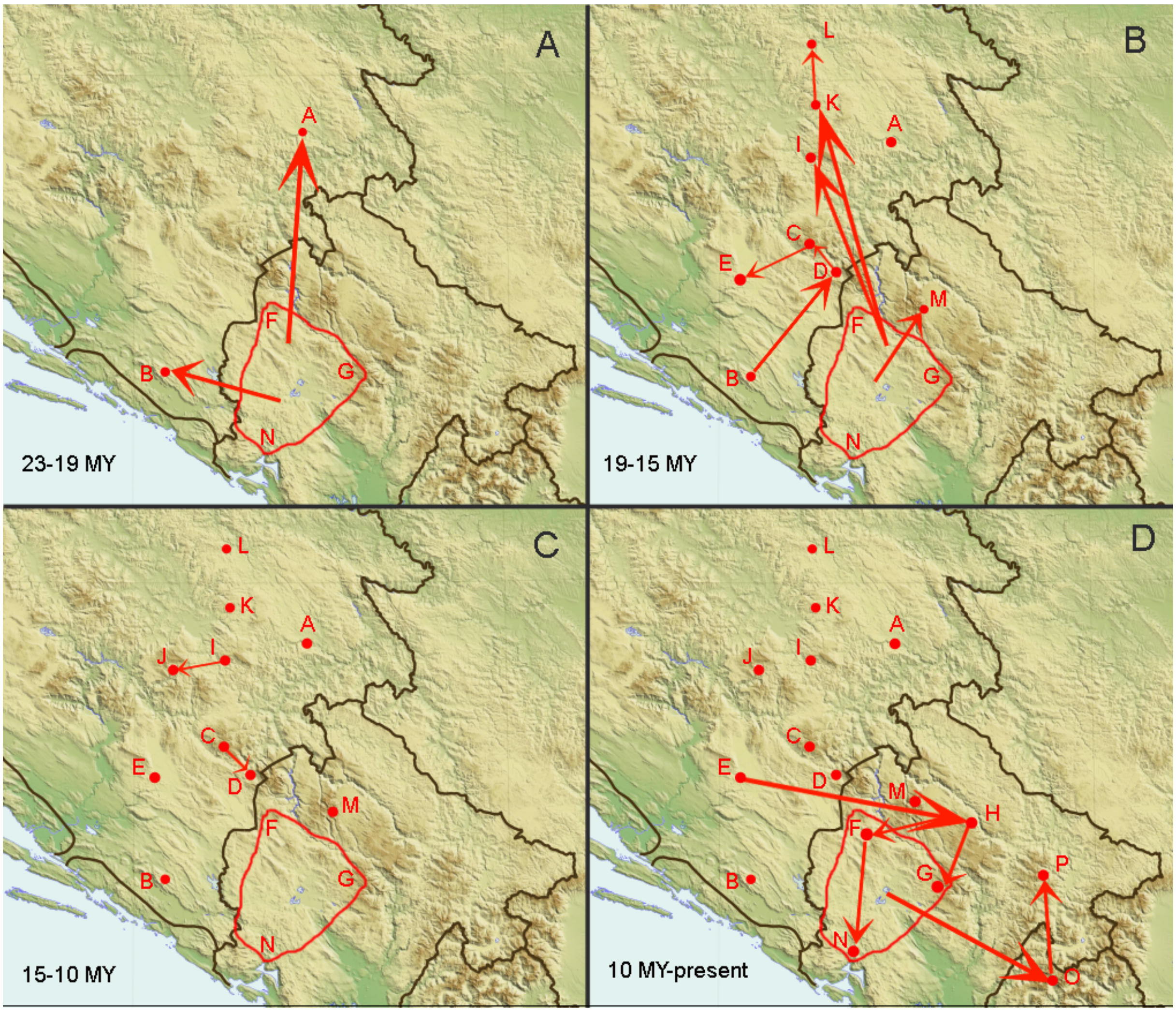


Fig. 4. Inferred biogeographic history of *Anthroherpon* using the BAYAREALIKE+*j* model. Geographical areas used in the analyses are marked with same letters as in Fig. 3. Arrows denote dispersal directions.

Table 4. BioGeoBEARS results for the genus *Anthroherpon* based on BEAST topology. Models tested without the distances between the areas taken into account.

Model	LnL	Degrees of freedom	d	e	j	AIC	AIC wt
BAYAREALIKE+j	-63.06	3	1.0e-07	0.0031	0.10	132.1	0.98
BAYAREALIKE	-94.97	2	0.0079	0.11	0	193.9	3.7e-14
DIVALIKE+j	-67.13	3	0.0009	1.0e-12	0.087	140.3	0.017
DIVALIKE	-85.67	2	0.0044	0.0094	0	175.3	4.0e-10
DEC+j	-68.59	3	0.0008	1.0e-12	0.07	143.2	0.0039
DEC	-90.7	2	0.0057	0.043	0	185.4	2.6e-12

Table 5. BioGeoBEARS results for the genus *Anthroherpon* based on BEAST topology. Models tested with the distances between the areas (+x) taken into account.

Model +x	LnL	Degrees of freedom	d	e	x	j	AIC	AIC wt
BAYAREALIKE+j	-63.94	4	1.0e-07	0.0029	0.086	0.11	135.9	0.96
BAYAREALIKE	-93.75	3	0.05	0.11	-0.43	0	193.5	2.9e-13
DIVALIKE+j	-67.39	4	0.0011	1.0e-12	-0.0030	0.064	142.8	0.03
DIVALIKE	-84.99	3	0.0079	0.014	-0.13	0	176	1.9e-09
DEC+j	-68.48	4	0.0007	1.0e-12	0.029	0.081	145	0.010
DEC	-90.64	3	0.0061	0.044	-0.014	0	187.3	6.6e-12

4.3.5. Results of morphometric analyses

1. *Morphological separation of the genera* Anthroherpon *and* Graciliella

The deformation vectors confirmed that the major difference in habitus between the two genera is in the shape of the pronotum, which is more strongly constricted in *Graciliella*, and elytra which are more globular in *Graciliella*. There is very little difference in the head shape between these two genera (Fig. 4). The differences in pronotum and elytra shape were considered diagnostic in the taxonomic cross diagnosis for the new genus (see Chapter 4). The PCAs for the partial warp scores (data not shown) confirmed that the two genera are most clearly separated in pronotum shape.

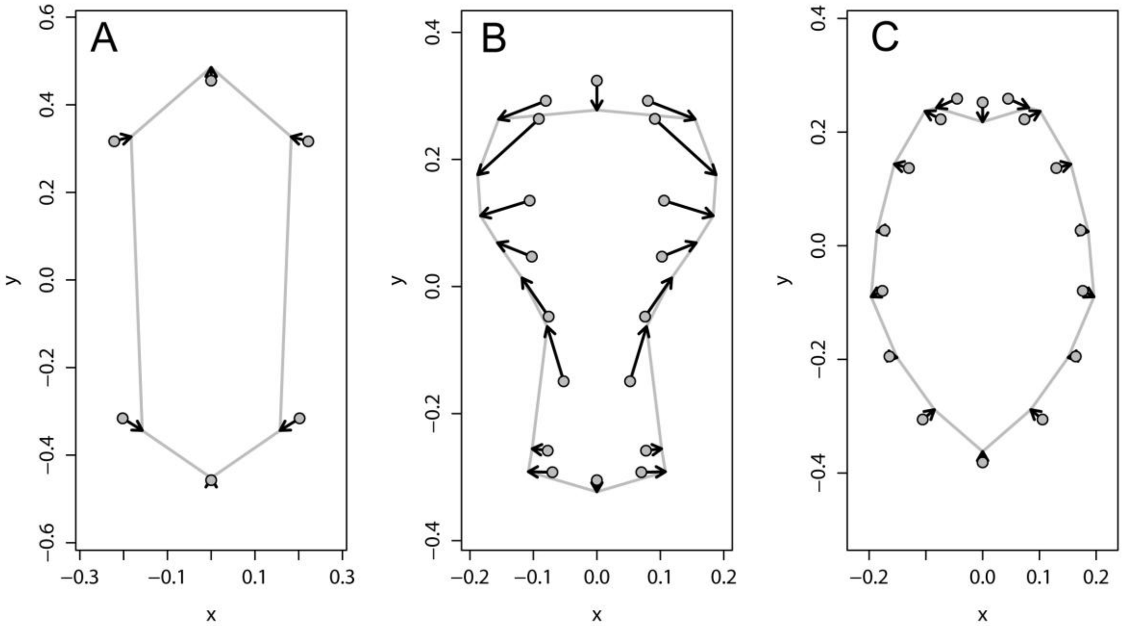


Fig. 4. X,Y transformation vectors (exaggeration 2x) for mean *Anthroherpon* shape (represented by the grey line) towards mean *Graciliella* shape (represented by the arrow heads) for head (A), pronotum (B), and elytra (C). Rostral is at the top of each figure.

2. *Exploration of troglomorphic diversity within* Anthroherpon

Figures 5 and 6 show the relative lengths of appendages of all studied *Anthroherpon* species. Stronger elongation of antennae and legs is seen in several species, particularly *A. hoermanni*, *A. sinjajevina*, *A. pygmaeum*, *A. harbichi*, *A. latipenne*, and *A. taxi*. Interestingly, the elongation of palps follows a different pattern. Here, the greatest relative lengths are found in *A. cylindricolle*, *A. ganglbaueri*, and *A. zariquieyi*. See further below.

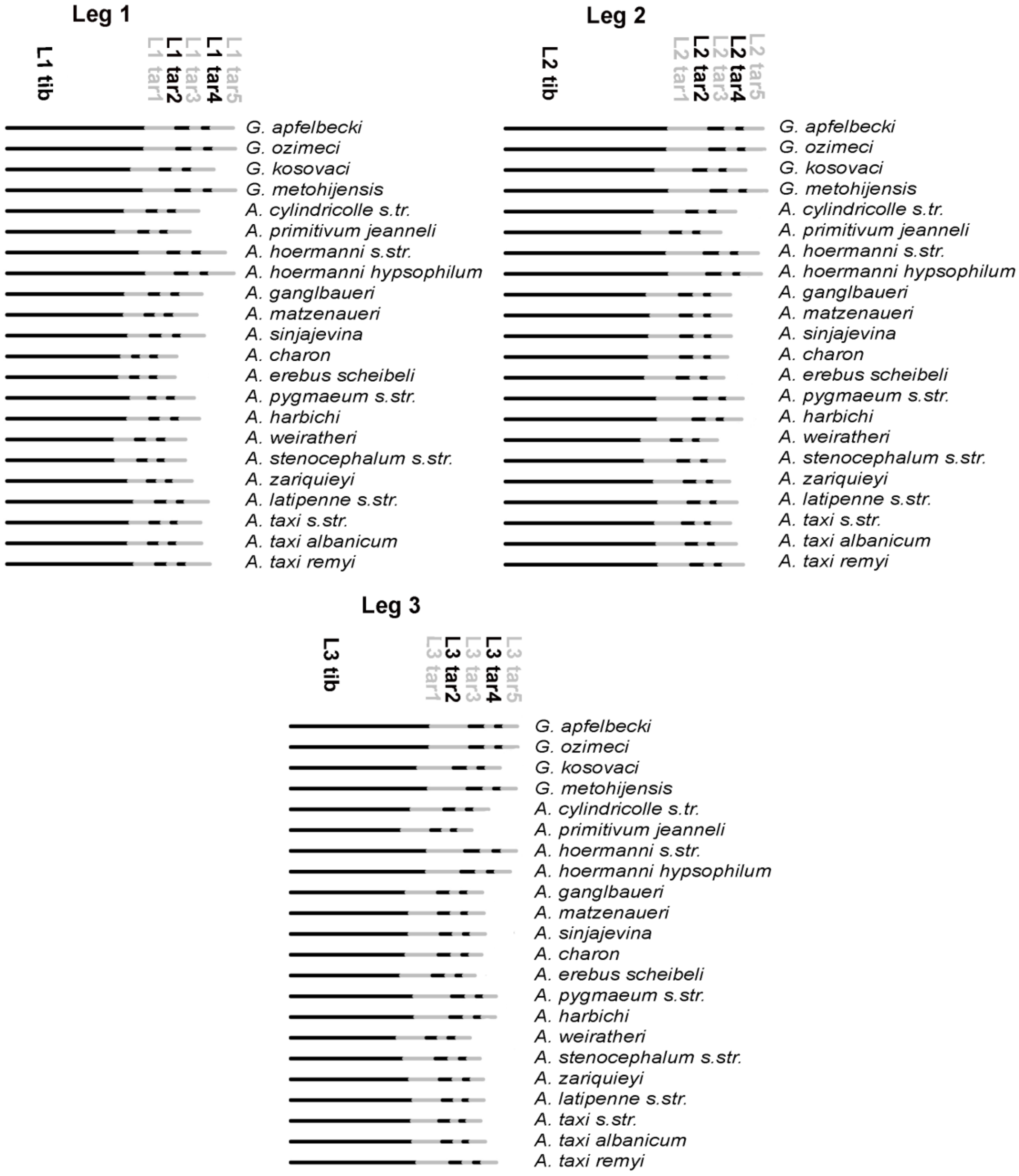


Fig. 5. Legs lengths, relative to body length, for *Anthroherpon* and *Graciliella* species.

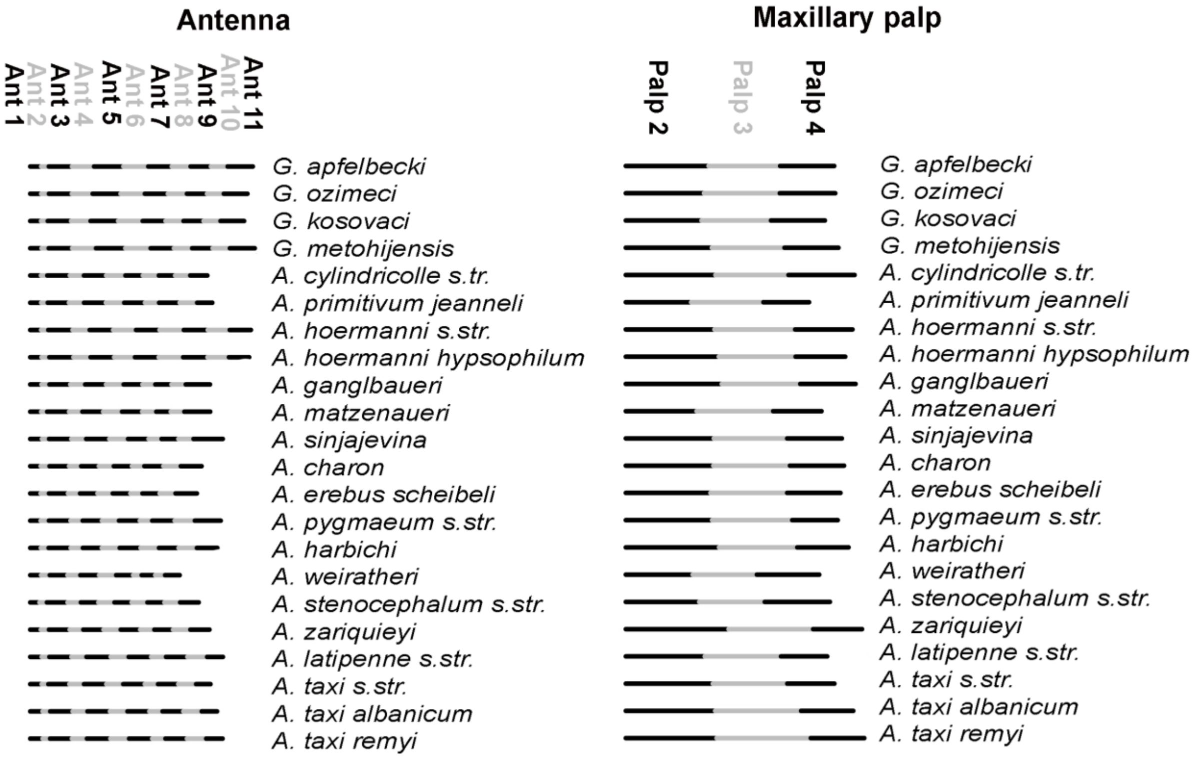


Fig. 6. Antennomerae and maxillary palpomerae lengths, relative to body length, for *Anthroherpon* and *Graciliella* species.

3. *Exploration of character evolution within* Anthroherpon

We found the following values for Blomberg's *K*. Head: *K*=0.57 (not significant, *P*=0.3924); Pronotum: *K*=0.65 (significant, *P*=0.0014); Elytra: *K*=0.75 (significant, P=0.003). The phylomorphospace graphs are shown in Fig. 7. These suggest that most species present a similar head shape, except for *A. stenocephalum*, *A. weiratheri*, and *A. taxi remyi*; pronotum shape appears to vary homogeneously, with the exception of *A. weiratheri* and *A. harbichi* (see below under 4.); elytra shape variation also seems relatively homogeneous in the phylomorphospace, with no apparent trend in shape variation except for the influence of the phylogeny.

Phylomorphospace for the lengths of the appendages is shown in the Fig. 8. The evolution of the relative lengths of antennae and legs (Fig. 8 B, C) mainly follows the phylogeny, while the evolution of maxillary palps (Fig. 8 A) shows a different pattern, with many branches in phylomorphospace crossing each other and the two analysed genera (*Graciliella* and *Anthroherpon*) not clearly separated.

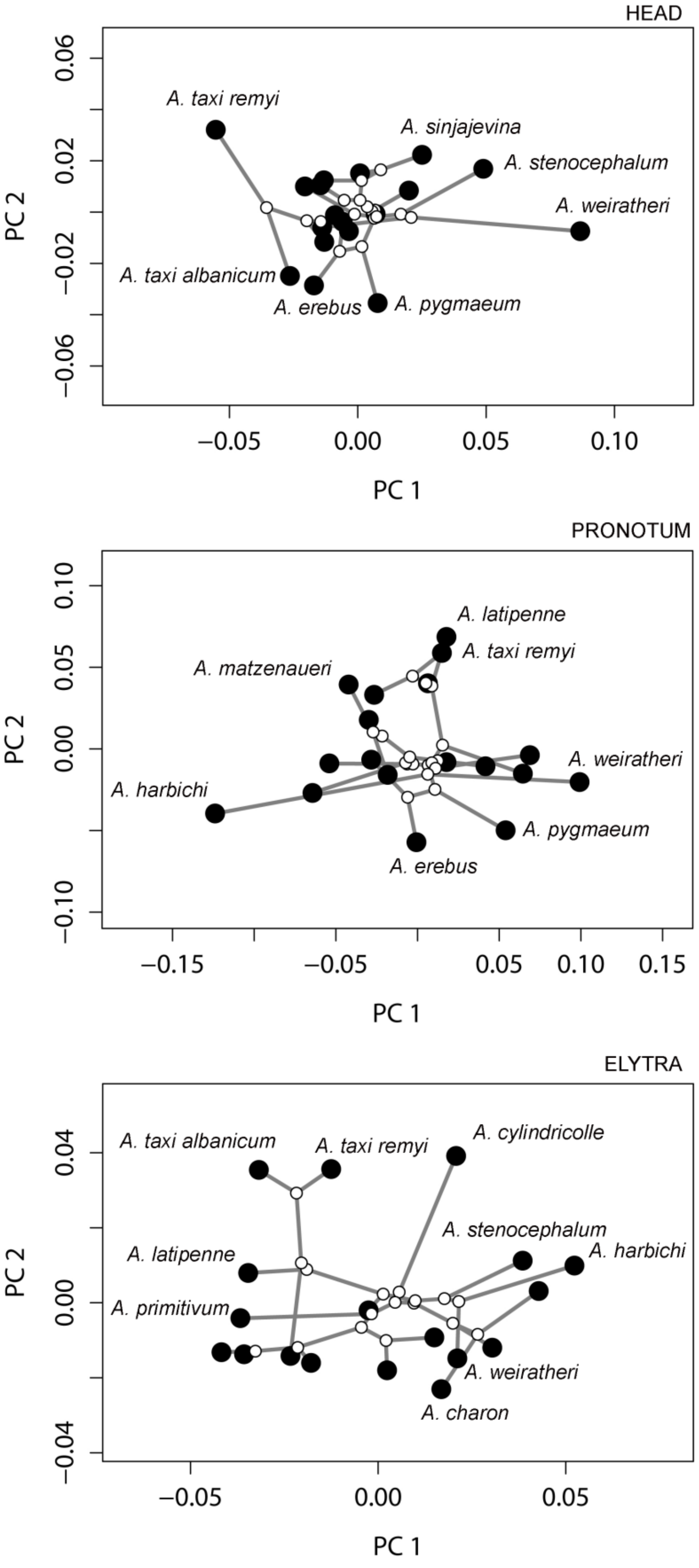
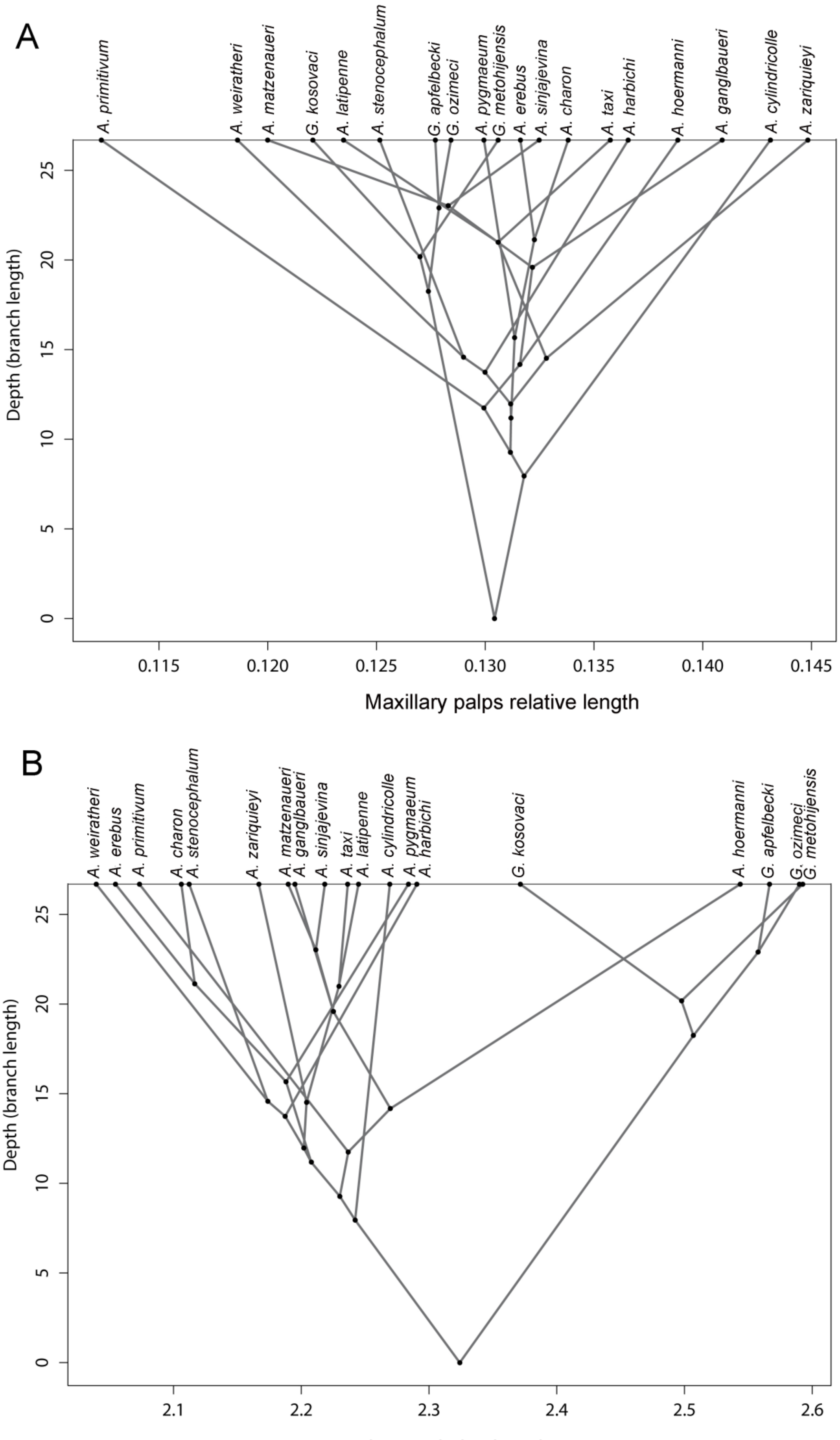


Fig. 7. Phylomorphospace for the genus *Anthroherpon*, shown for each body part separately. Open symbols are internal nodes; filled symbols are terminal nodes.


A
A. primitivum
A. weiratheri
A. matzenaueri
G. kosovaci
A. latipenne
A. stenocephalum
G. apfelbecki
G. ozimeci
A. pygmaeum
G. metohijensis
A. erebus
A. sinjajevina
A. charon
A. taxi
A. harbichi
A. hoermanni
A. ganglbaueri
A. cylindricolle
A. zariquieyi
Depth (branch length)
0
5
10
15
20
25
0.115
0.120
0.125
0.130
0.135
0.140
0.145
Maxillary palps relative length
B
A. weiratheri
A. erebus
A. primitivum
A. charon
A. stenocephalum
A. zariquieyi
A. matzenaueri
A. ganglbaueri
A. sinjajevina
A. taxi
A. latipenne
A. cylindricolle
A. pygmaeum
A. harbichi
G. kosovaci
A. hoermanni
G. apfelbecki
G. ozimeci
G. metohijensis
Depth (branch length)
0
5
10
15
20
25
2.1
2.2
2.3
2.4
2.5
2.6
Legs relative length

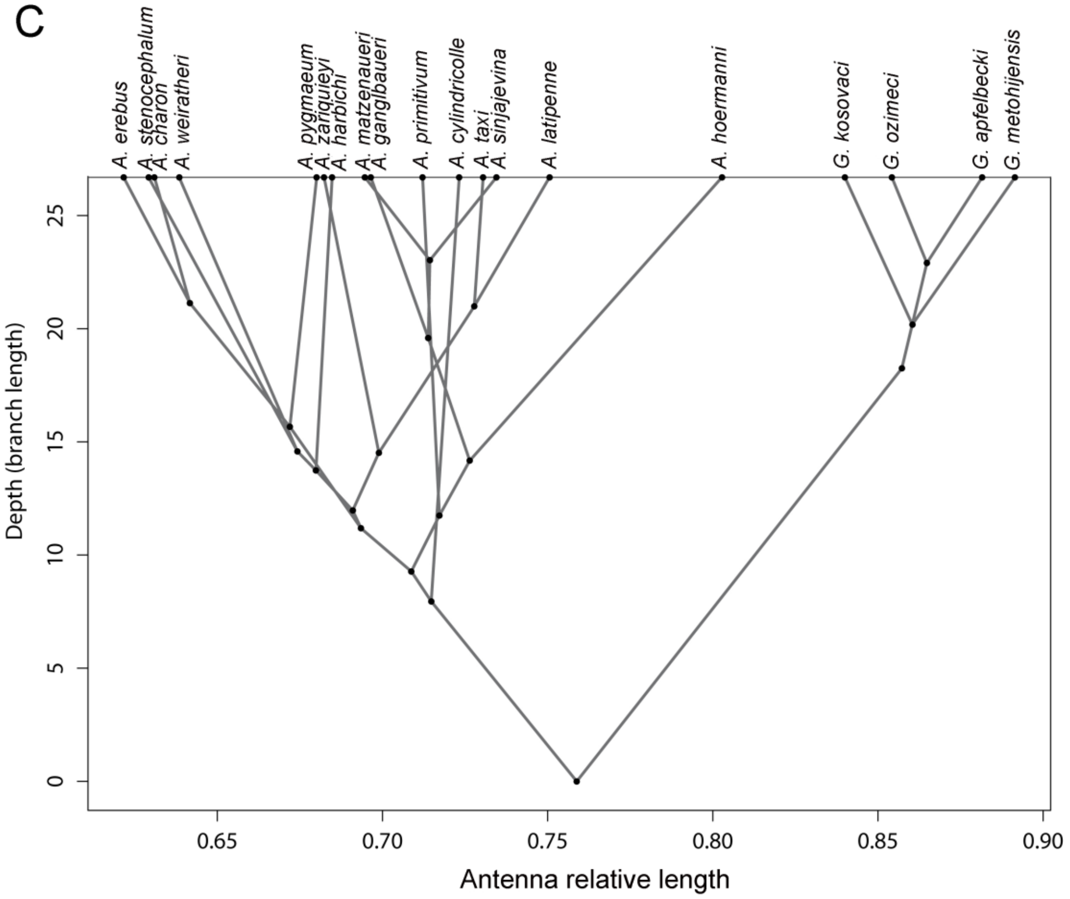


Fig. 8. Phylomorphospace for the lengths of the appendages (completeness sake, the genus *Graciliella* is also included). A. Maxillary palp relative length; B. Legs relative length; C. Antenna relative length.

4. *Character divergence in two syntopic species,* A. harbichi *and* A. weiratheri

The evolutionary rates found in *A. harbichi* & *A. weiratheri* were significantly different for the pronotum (2.6 times higher; *P*=0.014; see Fig. 9). All other body parts (except relative palp length; see Fig. 8 A) showed morphological divergence that was in line with that in the rest of the genus.

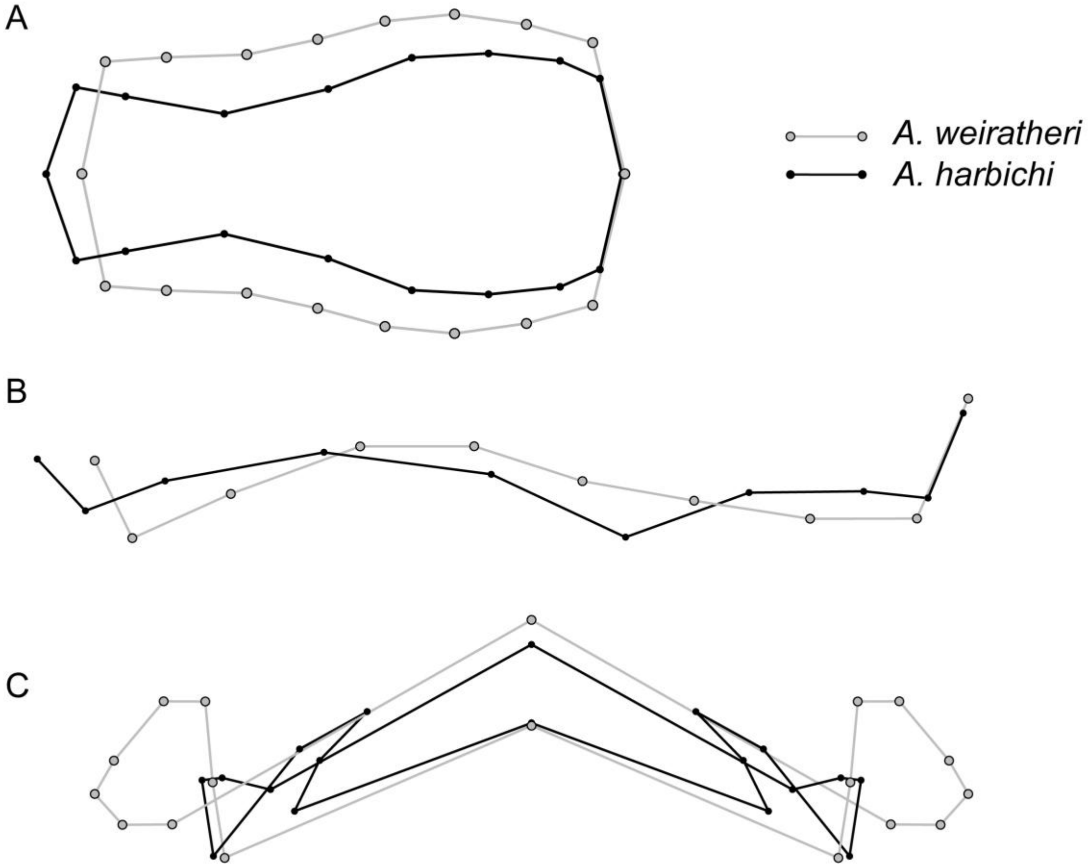


Fig. 9. Non-amplified mean differences in pronotum shape between *A. weiratheri* and *A. harbichi*, shown in dorsal (A), lateral (B), and frontal (C) view.

4.4. DISCUSSION

4.4.1. Subterranean compartment as a natural laboratory

We reconstructed the phylogeny of *Anthroherpon* and related genera, and used this to analyse the genus's biogeographical history and morphological evolution. Beyond this taxon-focused study, it appears that it was indeed of particular interest to make use of a subterranean taxon as *Anthroherpon,* prone to endemism and highly diversified, as a natural model for studying evolutionary radiations. Although many questions remain and data need to be completed in several fields, our results show that by such a comprehensive analysis, we may draw inferences about the evolution of troglomorphic features, as well as about subterranean colonization routes and endemism. Our study represents the first comprehensive analysis of Dinaric terrestrial troglobites that involves dated phylogeny, ancestral range reconstruction, and morphometric approaches. Enlarging the scope of future phylogenetic analyses around *Anthroherpon*, a step towards a complete phylogeny of Cholevinae, would allow better understanding of important evolutionary and biogeographical questions.

4.4.2. Phylogeny and taxonomy

Although the genus *Anthroherpon* is the focus of this Chapter, our results also show the polyphyly of the subtribe Leptodirina which includes it, suggesting that the tribal assignation of *Remyella* Jeannel, 1931, *Rozajella* S. Ćurčić, Brajković & B. Ćurčić, 2007 and also *Nonveilleriella* Perreau & Pavićević, 2008 should be reconsidered. However, the clarification of this question requires to take into consideration the whole tribe Leptodirina and not only the small number of genera used in this work (*Apholeuonus* Reitter, 1889, *Charonites* Apfelbeck, 1907, and *Parapropus* Ganglbauer, 1899). Enlarging the scope of future phylogenetic analyses around *Anthroherpon* would allow better understanding important evolutionary and biogeographical questions. That would be another step towards a complete phylogeny of Cholevinae.

The dated phylogeny we reconstructed for *Anthroherpon* and related genera (Fig. 2), shows that the large genus *Anthroherpon* began to diverge approximately in the early Miocene (22 MYA), ca. 5 million years after breaking away from a lineage to a divergent, hygropetric genus also endemic of the Dinarides, *Hadesia*. Although we did not do a formal lineages-through-time analysis, the branching points within *Anthroherpon* are spaced quite evenly through time, suggesting that the evolutionary radiation of the genus has not been marked by any major burst of radiation or extinction.

Even when disregarding the polyphyly of *Anthroherpon* that necessitated the erection of the new genus, *Graciliella* (Njunjić et al., 2016), our results only partly support the traditional, morphology-based, subdivision of the genus *Anthroherpon* into species groups (Jeannel, 1930; Guéorguiev, 1999). Our phylogenetic reconstruction shows that only the "*ganglbaueri*" and "*cylindricolle*" species groups are monophyletic, while all others show polyphyly.

4.4.3. Historical biogeography

The phylogenetic reconstruction shows a certain degree of geographic structuring (Fig. 10). Namely, the clade defined by node 1 chiefly contains the "*hoermanni*" species group and the "*ganglbaueri*" species group, distributed in the High mountain karst (The only exceptions are *A. matulici* and *A. primitivum jeanneli* from the Low coastal Adriatic karst). In contrast, the clade defined by node 2 contains species from very different parts of the range.

This geographic structuring allows formal and informal analyses of the biogeographic history. Our formal reconstruction of the biogeographic history shows four successive phases (Fig. 4), characterized by: (i) an origin in western Montenegro and dispersal of the common ancestor of *Anthroherpon* into eastern Bosnia and Herzegovina; (ii) further, multiple movements through western Montenegro and eastern Bosnia and Herzegovina, mostly towards the north and northeast; (iii) a period of dispersal stagnation; (iv) further dispersals, mostly in a south-eastern direction, into eastern Montenegro and Albania. Dispersal events occurred mostly during the Miocene, when the mountains ranges within the Dinarides already had the present spatial distribution (Grubić, 1980; Marović et al., 2007), so we may assume that the large-scale tectonic events that formed these mountain ranges preceded the *Anthroherpon* radiation.

Our biogeographic reconstruction of the ancestral range (Fig. 4) shows the origin of the genus in the area comprising three high mountains in western Montenegro: Orjen Mt. (1894 m), Dobreljica Mt. (1834 m), and Moračke planine Mt. (2226 m). From this area the presumed *Anthroherpon* ancestor dispersed to the other parts of its present range. The origin of the genus in western Montenegro is in the accordance with the available data on the paleogeography of the Balkan Peninsula. In the Lower Miocene, the northeastern part of the Dinarides was covered by the Dinaric Lake System that extended north to the Lower Austria (Krstić et al., 2003; Krstić et al., 2012). Western and southern Montenegro were not covered by water during that time, which is consistent with an initiation of *Anthroherpon* radiation there. During the Middle and Late Miocene and the Pliocene lakes remained only in the depressions (Mirković, 1980; Buzaljko & Pamić, 1982), which may have limited dispersal routes to the dry areas in between. It has been argued that the recent lineages of *Proteus* and *Troglocharis* originate from the Dinaric Lake System (Trontelj et al., 2007). This would imply very different diversification patterns between terrestrial and aquatic cave species in the Dinarides, hence probably very different distribution patterns. This opens major perspectives in the understanding of global subterranean biodiversity patterns that deserve to be addressed.

4.4.4. Evolution of discrete and continuous morphological features and troglomorphy

Our phylogeny allows tracing and analysing the evolution of both discrete and continuous morphological characters. Although we did not subject the discrete characters to a formal analysis, we addressed their evolution (both within and above the level of the genus *Anthroherpon*) on the basis of a visual inspection of their distribution across the phylogeny. The continuous (morphometric) characters were subjected to a formal character evolution analysis.

As for discrete characters, the monophyly of the four most species-rich genera of the subtribe Anthroherponina (*Anthroherpon, Hadesia, Graciliella*, and *Leptomeson*) suggests a single origin of their shared character states (Jeannel, 1924; Perreau & Pavićević, 2008), namely: (i) position of the antennal insertions on the posterior third of the head length; (ii) enlarged and apically blunt claws; (iii) female urite IX reduced and without the appendicular parts; (iv) second antennomere much shorter than the first one; (v) wide and apically blunt claws. However, since four other genera of the Anthroherponina were not available for molecular phylogenetic analysis (they are extremely rare stenoendemics and mostly monospecific), conclusions on evolutionary conservation of these traits are preliminary.

Our morphometric analysis confirmed a strong separation between the genera *Anthroherpon* and *Graciliella*, as recently proposed (Njunjic et al., 2016). The separation appears to be most distinct in the shape of the pronotum, with *Graciliella* displaying a more slender, much more strongly constricted pronotum than *Anthroherpon*.

Within *Anthroherpon*, our phylogeny implies a single origin for only two character states, namely: (i) pronotum without a constriction in the posterior part (in the "*cylindricolle*" species group), and (ii) the last antennomere longer than the penultimate (in the "*ganglbaueri*" species group). Most other taxonomically relevant discrete characters have multiple independent origins, namely: (i) protibiae 1.8 to 2 times longer than protarsi, thick and curved inwards in dorsal view; (ii) profemora very thick in the posterior part; (iii) aedeagus with a thickened sclerotized strip along the length of the medioventral side of the median lobe; and (iv) last antennomere shorter than or as long as the penultimate.

To obtain some insight into the forces driving speciation in *Anthroherpon*, we conducted a formal evolutionary analysis of morphometric (continuous) traits. Within *Anthroherpon*, it appears that body shape (except head shape, which is more conserved) evolves neutrally: K-values are close to 1.0 and display significant phylogenetic signal. This suggests that in most species, presumably after isolation in allopatry, body shape evolves slowly following a Brownian motion model: body shape differences between species are roughly proportional to their phylogenetic distance and there is no strong divergence (K>1.0) or stasis (K<1.0).

Where troglomorphy in the appendages is concerned, we see that some species have more strongly elongated antennae and legs, but the projections in phylomorphospace suggest that this pattern mainly follows the phylogeny (Figs. 8B, 8C), which may mean that elongations in general are also neutral and not the result of rapid adaptation. If the latter were the case, we would expect to see many branches crossing each other in the phylomorphospace, which is not the case.

However, there are a few indications that, under certain circumstances, morphometric traits may evolve adaptively. Firstly, our analyses of the syntopic sister species pair *A. harbichi* & *A. weiratheri* suggests that these two species have undergone strong divergent evolution in pronotum shape (2.6 times greater than in the rest of the genus; P=0.014), with *A. harbichi*, which has a more strongly constricted pronotum (Fig. 9), showing the greatest disparity in phylomorphospace (Fig. 7). This suggests strong character divergence during or after the speciation process. Character divergence between two closely related species in syntopy could be caused either by (i) reproductive character displacement (if it takes place as part of the speciation process, termed reinforcement; Butlin, 1987), or (ii) ecological character displacement. Since relative palp length in these two species is more different than expected (Fig. 8 A), it could imply possibly different food uptake (in cave Cholevinae, maxillary palp length and structural features are considered indicative of diet; Moldovan et al., 2004). Pronotum shape, on the other hand, is unlikely to play a role in food or microhabitat specialization, but might conceivably play a role during copulation: in Cholevinae, copulation often (but not always, see Juberthie-Jupeau, 1988) involves the male holding the female's pronotum with his protarsi (Schilthuizen pers. comm.). For these reasons, we would tentatively suggest that both ecological and reproductive character displacement has occurred in this case.

Second, with regard to troglomorphy, *A. hoermanni* evolved distinctly increased leg and antenna length relative to the body size, compared with the other members of *Anthroherpon* (Figs. 5, 6). This may mean that in this particular species, extreme troglomorphy may have been selected for. It is difficult to speculate on the causes for this, since the exact selection pressures driving the elongation of appendages in troglobites is not fully understood. It might be that the caves where *A. hoermanni* live are particularly poor in nutrients, which require the beetles to move faster and travel longer distances, selecting for longer legs and antennae (the former for increased locomotion, the latter for surface increase of sensillae for long-distance detection of food).

Third, in contrast with the elongation of the other appendages (i.e. antenna and legs), the evolution of palp length relative to the body length is not reflecting the phylogeny. Since palps are directly involved with food uptake (Moldovan et al., 2004), this may mean that this morphometric character evolves under the influence of functional constraints linked to nutrition. Some species (*A. primitivum*, *A. weiratheri*) evolved relatively short palps compared with their sister species, whereas others (*A. zariquieyi*, *A. taxi remyi*) evolved much longer palps. No link with environmental parameters has been detected so far, and palp length has not been either analysed in the literature about cave beetles, but investigations on the diet of these species would bring interesting insight into the adaptive nature of palp elongation.

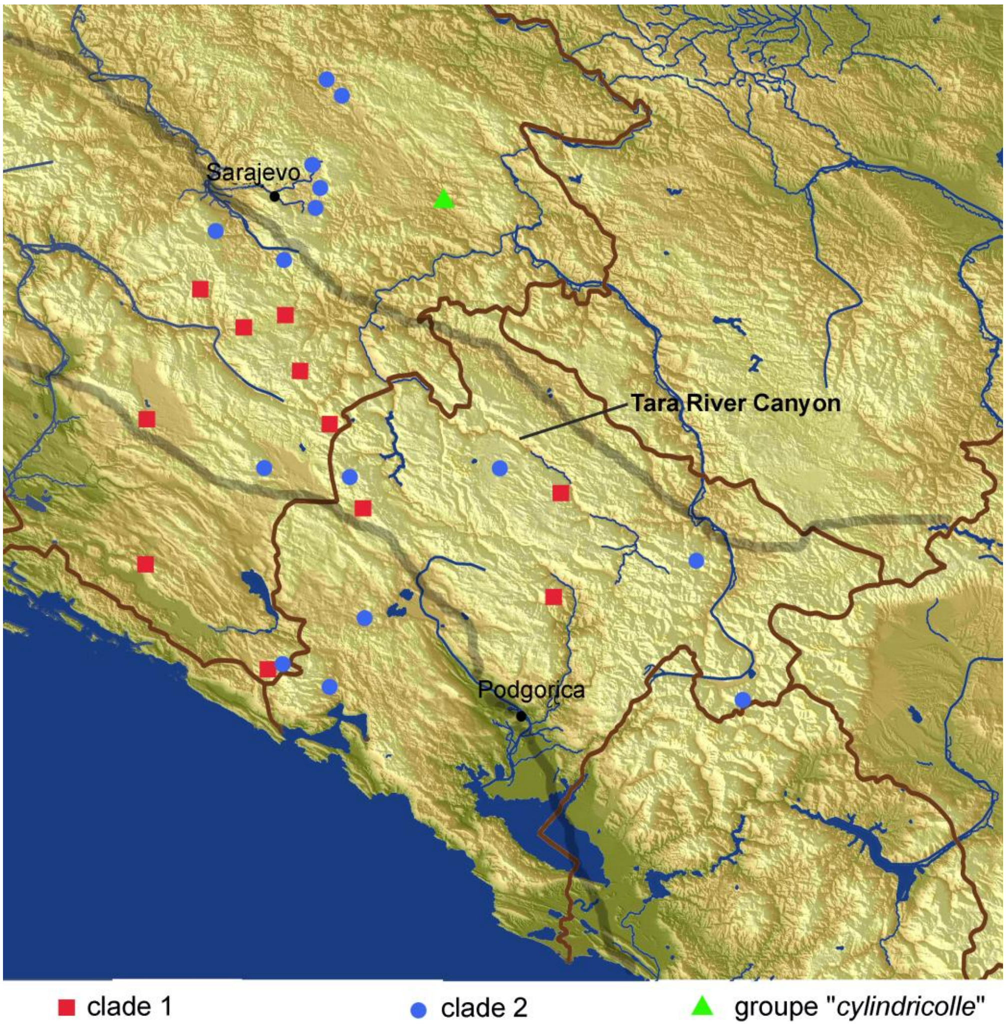


Fig. 10. Distribution of *Anthroherpon* species included in the phylogenetic reconstruction obtained with BEAST (Fig. 2). Different colours and symbols denote main clades of the phylogenetic tree. Thick grey lines show delimitation of the Dinaric karst in three main belts, from the southwest to northeast: the Low coastal Adriatic karst, the High mountain karst, and the Low continental interior karst (Hajna, 2012).

4.4.5. Evolutionary scenarios

Anthroherpon is an entirely troglobitic genus, and it is most parsimonious to presume that its ancestor also was a troglobite, since the genus is embedded in a larger clade consisting of entirely troglobitic genera (*Hadesia, Leptomeson*, and *Graciliella*). This implies that troglobitic lineages can disperse and diversify over a large geographic area during long periods of time.

In *Anthroherpon*, this certainly is the case. It is the most widely distributed genus of the subtribe Anthroherponina, covering a latitudinal range of more than 200 km and a longitudinal range of more than 170 km. Such "wide" distribution ranges of ancient troglobitic lineages might imply multiple independent colonisations and subsequent extinction of the epigean ancestors (Jeannel, 1943; Poulson & White, 1969; Culver & Pipan, 2009). Regarding *Anthroherpon*, however, there is no evidence for that: evidence rather suggests that the genus originated from a single colonization event. Similar conclusions were obtained by Faille et al. (2010) for Trechini and by Ribera et al. (2010) for Western Mediterranean Leptodirini.

Moreover, until recently, the widely accepted view regarding the radiation of troglobites was that once a lineage has adapted to the subterranean environment within a karst unit, it is unable to expand or diversify over a larger area and, as a result, it remains restricted to a very small range (Poulson & White, 1969; Barr & Holsinger, 1985; Culver & Pipan, 2009). Many taxa within the genus *Anthroherpon* are short-range endemics: from 26 species and 55 subspecies of *Anthroherpon*, 16 species and 24 subspecies are only known from a single cave. This endemicity, paired with the wide range of the genus as a whole, is a paradox: highly reduced dispersal abilities within the subterranean realm (Crouau-Roy, 1989; Gibert et al., 1994) seem to be balanced by occasional long-distance dispersal.

Dispersal of terrestrial troglobites can be limited by intrinsic factors (low vagility, high specialization, small size) and extrinsic factors (e.g., fluvial and stratigraphic barriers) (Barr, 1985). The present distribution of the genus *Anthroherpon* shows that large rivers, deep canyons and non-karstic terrain are indeed potential barriers to the dispersal of terrestrial troglobites.

Anthroherpon is widely distributed in subterranean habitats of the Dinaric range: from Gornja Oćevija (Bosnia and Herzegovina) on the north to the maritime mountains of Montenegro on the south, and from Prenj Mt. (Bosnia and Herzegovina) on the west to Prokletije Mt. (Montenegro, Albania) and Mokra planina Mt. (Montenegro, Metohija) on the east. If we disregard the highly doubtful distribution of *A. brckoensis* (see Chapter III: Article 1), the northernmost finding of *Anthroherpon* is in Banja pećina near Gornja Oćevija (Bosnia and Herzegovina), the type locality of *A. stenocephalum noesskei*. This area roughly overlaps with the approximate border of the Dinaric karst (Roglić, 1965; Gams, 1974). The easternmost finding of *Anthroherpon* in its northern part of the distribution area is in Golubovića pećina near Rogatica (*A. cylindricolle cylindricolle*). Further east, the area where no *Anthroherpon* has been found, was a depression during the Miocene (when the dispersal of *Anthroherpon* was the most intensive, see above), where freshwater sediments were deposited (Vujnović, 1981). It is possible that this stratigraphic barrier, though discontinuous, limited the dispersal of *Anthroherpon* further to the east. This has however not been investigated so far as a barrier to dispersal for other groups of cave fauna. Sampling focussed on this particular area should be performed in the future to address this question.

To the south, the range of the genus spreads throughout the mountains of the Maritime belt of the Dinarides. Although karst in south Montenegro almost reaches sea level, *Anthroherpon* was found only at higher elevations. This southern limit of *Anthroherpon* distribution may reflect the hydrographical history of this area. During the Pliocene and Pleistocene, alternations of transgressive and regressive phases influenced the coastline (De Giuli et al., 1987; Marović et al., 1999; Kuhlemann, 2003, 2007) and may have prevented dispersal further to the south. Another explanation could be in the specific microhabitat requirements: highly specialized species of the genus *Anthroherpon* originating from mountainous areas may not have been able to survive in warmer and less humid caves at lower elevations.

In the north-eastern Montenegro, what appears to be a clear geomorphological barrier to *Anthroherpon* dispersal is the 82 km long and 1.333 m deep Tara River Canyon, the deepest canyon in Europe. Near the southern border of the Canyon, two species of *Anthroherpon* occur: *A. zariquieyi* on Durmitor Mt. and *A. sinjajevina* on Sinjajevina Mt., but so far no *Anthroherpon* has been found on the northern side of the Tara River Canyon (Fig. 10). The Tara river is known as a barrier for troglobitic harvestmen of the genus *Cyphophthalmus* belonging to the "*minutus*" group (Murienne et al., 2010), which have similar distribution area as *Anthroherpon*.

One of the eastern limits of *Anthroherpon* distribution is Velež Mt. in Bosnia and Herzegovina. Here, two more genera of the subtribe Anthroherponina occur – *Graciliella* and *Leptomeson* – but so far they were never found in syntopy. It is possible that *L. loreki* and *G. metohijensis*, which are larger species (around 7 mm) and have more physogastric elytra (i.e., they can store more fat) are more competitive than *A. ganglbaueri ganglbaueri* that coexists in the same mountain range.

With the above discussion, we have given some insights into the intrinsic and extrinsic features of *Anthroherpon* that may have influenced their (in)ability for dispersal. The biogeographic analysis in BioGeoBears shows that models that include founder events (specified by the parameter *j*) have a better fit with the data. Overall, the phylogenetic and biogeographic reconstructions are consistent with the idea that *Anthroherpon* radiated underground from an already troglobitic common ancestor. The founder event signal suggests that speciation was often initiated by long-distance dispersal of one or a few colonists.

Such a scenario of evolutionary radiation accompanied by long-distance founder-events in troglobites has traditionally been interpreted as signifying a peripatric speciation process, involving the genetic revolutions in the small founder populations, followed by stasis when the population size had grown (Barr, 1967; Barr & Holsinger, 1985). On the other hand, this need not be the case: (weak) selection in large, isolated populations, may, over the long time periods that our dated phylogeny implies, also cause speciation (Feder et al., 2012).

4.4.6. Comparison with other subterranean taxa and conclusions

The observed phylogeographical patterns in *Anthroherpon* reflect a complex of paleo-biogeographical factors, involving the geological history of the Dinaric Mountains. So far, similar studies of Dinaric terrestrial troglobites are lacking, so we cannot make a comparison with other groups to check for concordance with our results. However, studies on Dinaric stygobites indicate that their distribution patterns do not correspond to recent hydrological divisions, but were attained in past drainage areas in geological history and preserved until today (Sket, 2005). A recent study of *Congeria* Partsch, 1835, the only known troglobiotic bivalve, suggests that it separated from its closest relative approximately at the same time as *Anthroherpon* began to diverge (22-23 MYA; Bilandžija et al., 2013). Since there is no reliable dating for vicariant events or the age of subterranean habitats of the Dinaric Mts. that could be used as a calibration point, dating phylogenies of Dinaric troglobites is problematic. Trontelj et al. (2007) have estimated the timeframe of the cladogenetic events for the aquatic isopod *Asellus aquaticus* Linnaeus, the cave salamander *Proteus anguinus* Laurenti, and the cave shrimp *Troglocharis* Dormitzer. They also encountered an inability to find reliable time estimates for paleogeographic events to calibrate local molecular clocks for different lineages (Trontelj et al., 2007). In our case, we alleviated this problem by using the separation of the Sardinian microplates from mainland Europe to calibrate the phylogeny of Western Mediterranean Leptodirini (Ribera et al., 2010).

Although our comprehensive study has allowed a more focused evaluation of competing hypotheses about the evolutionary radiation in *Anthroherpon*, additional targeted studies will be needed to confirm some of the conclusions reached here. Further molecular studies may be particularly helpful. For example, studies of genetic polymorphisms in large population samples will allow calculations of ancestral population sizes for endemic species, enabling an evaluation of the severity of founder events and bottlenecks (Juan et al., 2010). Also, niche differentiation in syntopic species pairs could be assessed with metabarcoding of gut contents (Kress et al., 2015). An interesting possibility is that antennae elongation, classically considered as a troglomorphic character, may have reached a maximum in these extremely troglomorphic species. Further elongation might be simply biophysically impossible, so in *Anthroherpon* an end-point may have been reached at the most advanced stage known among Coleoptera.

Suppl. Table 1. Sequenced specimens, with depository, locality, and collectors. All vouchers are stored at CINJ.

Species	**Voucher code**	**Locality**	**Legator**
Anthroherpon charon (Reitter, 1911)	VA34	BiH, Pale, Ravna pl, Omladinska pecina	I. Njunjić
Anthroherpon charon (Reitter, 1911)	IB17	BiH, Pale, Ravna pl, Omladinska pecina	I. Njunjić
Anthroherpon charon (Reitter, 1911)	IE48	BIH, Pale, Pećina na Palama	I. Njunjić
Anthroherpon charon (Reitter, 1911)	IB2	BIH, Sarajevo, Luke, Ledenica	I. Njunjić
Anthroherpon charon (Reitter, 1911)	VA2	BIH, Sarajevo, Luke, Ledenica	I. Njunjić
Anthroherpon charon (Reitter, 1911)	IE48	BIH, Pale, Pećina na Palama	I. Njunjić
Anthroherpon cylindricolle cylindricolle (Apfelbeck, 1889)	IE3	BiH, Pale, Rogatica, Golubovića pećina	I. Njunjić
Anthroherpon cylindricolle cylindricolle (Apfelbeck, 1889)	VA3	BiH, Pale, Rogatica, Golubovića pećina	I. Njunjić
Anthroherpon cylindricolle scaphium Reitter, 1908	IE10	BIH, Prača, Mračna pećina	I. Njunjić
Anthroherpon cylindricolle scaphium Reitter, 1908	VA14	BIH, Prača, Mračna pećina	I. Njunjić
Anthroherpon cylindricolle thoracicum (Apfelbeck, 1907)	VA9	BIH, Pale, Romanija, Novakova pećina	I. Njunjić
Anthroherpon cylindricolle thoracicum (Apfelbeck, 1907)	IB12	BIH, Pale, Romanija, Novakova pećina	I. Njunjić
Anthroherpon cylindricolle thoracicum (Apfelbeck, 1907)	IE24	BIH, Romanija, Zečeva ledenica	I. Njunjić
Anthroherpon erebus scheibeli (Jeannel, 1924)	IB4	BIH, Trnovo, Bjeličina pećina	I. Njunjić
Anthroherpon erebus scheibeli (Jeannel, 1924)	VA5	BIH, Trnovo, Bjeličina pećina	I. Njunjić
Anthroherpon ganglbaueri ganglbaueri (Apfelbecki, 1894)	VA15	BiH, Nevesinje, Novakuša	I. Njunjić
Anthroherpon ganglbaueri ganglbaueri (Apfelbecki, 1894)	IE11	BiH, Nevesinje, Novakuša	I. Njunjić
Anthroherpon harbichi (Reitter, 1913)	IB3	BiH, Sarajevo, Crepoljsko, Kečina stena	I. Njunjić
Anthroherpon harbichi (Reitter, 1913)	VA1	BiH, Sarajevo, Crepoljsko, Kečina stena	I. Njunjić
Anthroherpon harbichi (Reitter, 1913)	IB22	BIH, Pale, Mokro, Kadino selo, Vrelo Miljacke	I. Njunjić
Anthroherpon harbichi (Reitter, 1913)	IB16	BIH, Pale, Mokro, Kadino selo, Vrelo Miljacke	I. Njunjić
Anthroherpon harbichi (Reitter, 1913)	IE56	BIH, Pale, Orlovača	I. Njunjić
Anthroherpon hoermanni hoermanni (Apfelbeck, 1889)	IB7	BiH, Kalinovik, Miljevina, Borija pećina	I. Njunjić
Anthroherpon hoermanni hoermanni (Apfelbeck, 1889)	VA8	BiH, Kalinovik, Miljevina, Borija pećina	I. Njunjić
Anthroherpon hoermanni hoermanni (Apfelbeck, 1889)	IB8	BIH, Kalinovik, Nedavić, Vareničina pećina	I. Njunjić
Anthroherpon hoermanni hypsophilum (Apfelbeck, 1907)	IB20	BIH, Lebršnik, Đatlo	I. Njunjić
Anthroherpon hoermanni hypsophilum (Apfelbeck, 1907)	VA36	BIH, Lebršnik, Đatlo	I. Njunjić
Anthroherpon hoermanni sericeum (Jeannel, 1930)	VA12	BIH, Zelengora, Pećina na Bregoču	I. Njunjić
Anthroherpon hoermanni sericeum (Jeannel, 1930)	IE49	BIH, Zelengora, Pećina na Bregoču	I. Njunjić
Anthroherpon hoermanni orlovacensis (V. B. Guéorguiev, 1990)	IE50	MNE, Maglić, Trnovačko jezero, Katuni, Pećina	I. Njunjić
Anthroherpon latipenne latipenne (Apfelbeck, 1907)	IE13	MNE, Orjen, Ericova jama	M. Perreau

Anthroherpon latipenne latipenne (Apfelbeck, 1907)	VA30	BIH, Gubar Mt, Bukova rupa	M. Perreau
Anthroherpon latipenne latipenne (Apfelbeck, 1907)	IE40	MNE, Maganik, Kapetanovo jezero, Dola pećina	M. Perreau
Anthroherpon latipenne latipenne (Apfelbeck, 1907)	VA29	MNE, Dobreljica, Kazanci, Bijela pećina	I. Njunjić
Anthroherpon latipenne latipenne (Apfelbeck, 1907)	IB14	MNE, Nikšič, Dobreljica, Prljača	I. Njunjić
Anthroherpon latipenne latipenne (Apfelbeck, 1907)	IE54	Crna Gora, Nikšić, Trubjela, Ledena pećina na kiti	M. Komnenov
Anthroherpon matulici (Reitter, 1903)	I5	BiH, Gubar Mt, Bukova rupa	M. Perreau
Anthroherpon matulici (Reitter, 1903)	IE16	BiH, Gubar Mt, Bukova rupa	M. Perreau
Anthroherpon matzenaueri matzenaueri (Apfelbeck, 1907)	IB13	MNE, Golija, Latično, Jama u Rudinskom dolu	I. Njunjić
Anthroherpon matzenaueri matzenaueri (Apfelbeck, 1907)	VA24	MNE, Golija, Latično, Jama u Rudinskom dolu	I. Njunjić
Anthroherpon matzenaueri taliensis (Zariquiey, 1927)	IE2	MNE, Maganik, Kapetanovo jezero, Dola pećina	M. Perreau
Anthroherpon matzenaueri taliensis (Zariquiey, 1927)	VA19	MNE, Maganik, Kapetanovo jezero, Dola pećina	M. Perreau
Anthroherpon primitivum jeanneli (Winkler, 1925)	IE39	BiH, Hercegovina, Turica, Motka, Mravinjac	M. Perreau
Anthroherpon primitivum jeanneli (Winkler, 1925)	IE1	BiH, Hercegovina, Turica, Motka, Mravinjac	I. Njunjić
Anthroherpon pygmaeum stricticolle (Jeannel, 1930)	IE25	BiH, Bjelašnica, Opančak, Ledenjača	I. Njunjić
Anthroherpon pygmaeum stricticolle (Jeannel, 1930)	IB23	BiH, Bjelašnica, Opančak, Ledenjača	I. Njunjić
Anthroherpon sinjajevina Njunjić, Perreau, Pavićević, 2015	VA6	MNE, Sinjajevina, Rudnica, Blažova pećina	I. Njunjić
Anthroherpon sinjajevina Njunjić, Perreau, Pavićević, 2015	I3	MNE, Sinjajevina, Rudnica, Blažova pećina	I. Njunjić
Anthroherpon stenocephalum stenocephalum (Apfelbeck, 1910)	IE44	BiH, Olovo, Bijambare	I. Njunjić
Anthroherpon stenocephalum stenocephalum (Apfelbeck, 1910)	IE4	BiH, Olovo, Bijambare	I. Njunjić
Anthroherpon stenocephalum noesskei (Jeannel, 1924)	IE9	BIH, Olovo,Očevlje, Banja pećina	I. Njunjić
Anthroherpon stenocephalum noesskei (Jeannel, 1924)	IB19	BIH, Olovo,Očevlje, Banja pećina	I. Njunjić
Anthroherpon taxi taxi (Müller, 1913)	IE55	MNE, Kameno more, Dvestotka	I. Njunjić
Anthroherpon taxi taxi (Müller, 1913)	IE5	MNE, Orjen, Križeva jama	I. Njunjić
Anthroherpon taxi albanicum (Apfelbeck, 1919)	IE7	MNE, Prokletije, Caf Borit, Kolektor	M. Perreau
Anthroherpon taxi albanicum (Apfelbeck, 1919)	IE17	MNE, Prokeltije, Ćaf Borit, Dmuhava	I. Njunjić
Anthroherpon taxi albanicum (Apfelbeck, 1919)	IE32	MNE, Prokletije, Oktopod	I. Njunjić
Anthroherpon taxi albanicum (Apfelbeck, 1919)	I1	MNE, Prokletije, Ćef Borit, Nibu Čarna	I. Njunjić
Anthroherpon taxi remyi (Jeannel, 1931)	IE47	MNE, Berane, Lubnice, Župan pećina	I. Njunjić
Anthroherpon weiratheri (Reitter, 1913)	IE21	BiH, Sarajevo, Crepoljsko, Kečina stena	I. Njunjić
Anthroherpon weiratheri (Reitter, 1913)	IE23	BIH, Pale, Orlovača	I. Njunjić
Anthroherpon zariquieyi (Jeannel, 1930)	IE37	MNE, Durmitor, Vodeni krš, Vodena pećina	I. Njunjić
Anthroherpon zariquieyi (Jeannel, 1930)	VA13	MNE, Durmitor, Peć. u Sedlenoj gredi	I. Njunjić
Anthroherpon zariquieyi (Jeannel, 1930)	IB6	MNE, Durmitor, Peć. u Sedlenoj gredi	I. Njunjić
Apholeuonus nudus styrani Apfelbecki, 1906	IAP3	BiH, Kalinovik, Borija, Borija pećina	I. Njunjić
Apholeuonus nudus styrani Apfelbecki, 1906	IAP4	BiH, Kalinovik, Borija, Borija pećina	I. Njunjić
Apholeuonus longicollis sequensi Reitter, 1906	IAP6	BIH, Bjelašnica, Opančak, Megara	I. Njunjić
Apholeuonus longicollis sequensi Reitter, 1906	IAP7	BIH, Bjelašnica, Opančak, Megara	I. Njunjić

Charonites sp. Apfelbeck, 1907	IO33	BiH, Pale, Omladinska pećina	I. Njunjić
Charonites sp. Apfelbeck, 1907	IO31	BIH, Pale, Mokro, Kadino selo, vrelo Miljacke	I. Njunjić
Graciliella apfelbecki apfelbecki (Müller, 1910)	VA43	CRO, Bobani, Grabovica	R. Ozimec
Graciliella kosovaci Njunjić, Perreau, Hendriks, Schilthuizen, Deharveng, 2016	IE63	CRO, Tunel Konavle, Kaverna 781	R. Ozimec
Graciliella metohijensis (Zariquiey, 1927)	IB15	BiH, Nevesinje, Balabani, Drvendžina pećina	I. Njunjić
Graciliella metohijensis (Zariquiey, 1927)	MP04	BIH, Hercegovina, Korita, Kobilja glava, Đatlo	M. Perreau
Graciliella ozimeci Njunjić, Perreau, Hendriks, Schilthuizen, Deharveng, 2016	VA44	CRO, Dubrovnik, Točionik, Vranja pećina	R. Ozimec
Graciliella ozimeci Njunjić, Perreau, Hendriks, Schilthuizen, Deharveng, 2016	IE65	CRO, Dubrovnik, Točionik, Vranja pećina	R. Ozimec
Graciliella apfelbecki apfelbecki (Müller, 1910)	IE60	CRO, Dubrovnik, Osojnik, Močiljska špilja	N. Hanžek
Graciliella apfelbecki apfelbecki (Müller, 1910)	IE19	BiH, Popovo polje, Vjeternica	M. Perreau
Graciliella apfelbecki apfelbecki (Müller, 1910)	IE43	BiH, Popovo polje, Vjeternica	R. Ozimec
Graciliella apfelbecki apfelbecki (Müller, 1910)	IE64	CRO, Dubrovnik, Osojnik, Močiljska špilja	R. Ozimec
Graciliella apfelbecki apfelbecki (Müller, 1910)	IE66	CRO, Dubrovačko primorje, Špilja za Gromačkom vlakom	R. Slapnik
Leonhardella atennaria Apfelbeck, 1907	IO37	MNE, Durmitor, Savin kuk, Vodena pećina	I. Njunjić
Leptomeson bujasi Giachino, Bregović, Jalžić, 2011	I8	Hrvatska, Brač, Selca, Nagorinac, Ješkalovica	B. Jalžić & P. Kutleša
Leptomeson bujasi Giachino, Bregović, Jalžić, 2011	IL4	Croatia, Brač, Jama kod Matešića stana	B. Jalžić & P. Kutleša
Leptomeson dombrowskii dombrowskii (Apfelbeck, 1907)	IL2	Croatia, Dalmacija, Mosor, Vranjača špilja kod Dugopolja	B. Jalžić & H. Bilandžija
Leptomeson radjai Giachino, Bregović, Jalžić, 2011	IL1	Croatia, Šibenik, Perković, Zvekača	P. Bregović
Leptomeson radjai Giachino, Bregović, Jalžić, 2011	IL5	Croatia, Šibenik, Perković, Zvekača	P. Bregović
Leptomeson raguzi Giachino, Bregović, Jalžić, 2011	IL3	BIH, zapadna Hercegovina, Grude, Drinovci, Majića ponor	H. Bilandžija
Hadesia asamo Perreau & Pavićević, 2008	MP01	BiH, Trebinje, Zupci, Bravenik	M. Perreau
Hadesia lakotai Perreau & Pavićević, 2008	MP03	BIH, Hercegovina, Korita, Kobilja glava, Đatlo	M. Perreau
Hadesia vasiceki Müller, 1911	IH1	BiH, Popovo polje, Vjeternica	M. Perreau
Hadesia weiratheri Zariquiey, 1927	IH2	MNE, Orjen, Kameno more, PT4	M. Perreau
Parapropus sericeus muelleri Jeannel, 1924	IO26	BIH, Sanski most, Hrustovačka pećina	I. Njunjić
Parapropus pfeiferi Apfelbeck, 1908	IE34	BIH, Sanski most, Donji kamičak	I. Njunjić
Proleonhardella (s.str) remyi Jeannel, 1934	I7	Srbija, Kamena gora, Guvniste, Mujova jama	I. Njunjić
Remyella sp. Jeannel, 1910	IR16	Serbia, Pešter, Đerekare, Vrelo Đerekarske reke	S. Ognjenović
Remyella javorensis S. Ćurčić & B. Ćurčić, 2008	IR3	Serbia, Sjenica, Ursule, Baždarska pećina	S. Ognjenović
Remyella javorensis S. Ćurčić & B. Ćurčić, 2008	IR14	Serbia, Donje Lopiže, Ušačka pećina	I. Njunjić
Remyella hussoni Jeannel, 1934	IR20	Serbia, Pešter, Đerekare, Sjera jama	S. Ognjenović

Remyella hussoni Jeannel, 1934	IR6	Serbia, Pešter, Đerekare, Piskova livada	P. Lazarević
Remyella propiformis Winkler, 1933	IE8	Serbia, Pešter, Lopužina jama	M. Popović
Remyella propiformis Winkler, 1933	IR18	Serbia, Pešter, Boljare, Kaćunova jama	S. Ognjenović
Remyella propiformis Winkler, 1933	VRE1	Serbia, Pešter, Crvsko, Jagoševa pećina	M. Popović
Remyella propiformis Winkler, 1933	IR13	Serbia, Pešter, Dolići, Jama bez dna	S. Ognjenović
Rozajella deelemani Perreau & Pavićević, 2008	IR08	MNE, Berane, Petnjik, Građa pećina	I. Njunjić
Rozajella madzgalji sp.n.	IR03	MNE, Berane, Goražde, Velika pećina na Gareškom kršu	I. Njunjić

Suppl. Table 2. The list of material included in the morphometric analyses. Abbreviations: CINJ (collection Iva Njunjić), CMPR (collection Michel Perreau), MNHN (collection Muséum national d'histoire naturelle)

Col. Number.	**Genus**	**Species**	**Subspecies**	**Locality and legator (where available)**
INJ1 (CINJ)	*Anthroherpon*	*stenocephalum*	*stenocephalum*	BIH, Olovo, Bijambare, I. Njunjić
INJ2 (CINJ)	*Anthroherpon*	*taxi*	*remyi*	CG, Berane, Lubnice, Županska pećina, 23.03.2014, I. Njunjić
INJ3 (CINJ)	*Anthroherpon*	*taxi*	*remyi*	CG, Berane, Lubnice, Županska pećina, 23.03.2014, I. Njunjić
INJ4 (CINJ)	*Anthroherpon*	*taxi*	*remyi*	CG, Berane, Lubnice, Županska pećina, 23.03.2014, I. Njunjić
INJ5 (CINJ)	*Anthroherpon*	*taxi*	*remyi*	CG, Berane, Lubnice, Županska pećina, 23.03.2014, I. Njunjić
INJ6 (CINJ)	*Anthroherpon*	*taxi*	*remyi*	CG, Berane, Lubnice, Županska pećina, 23.03.2014, I. Njunjić
INJ7 (CINJ)	*Anthroherpon*	*cylindricolle*	*cylindricolle*	BIH, Rogatica, Golubovići, Golubovića pećina, 02.05.2013, P. Kosovac
INJ8 (CINJ)	*Anthroherpon*	*cylindricolle*	*cylindricolle*	BIH, Rogatica, Golubovići, Golubovića pećina, 02.05.2013, P. Kosovac
INJ9 (CINJ)	*Anthroherpon*	*cylindricolle*	*cylindricolle*	BIH, Rogatica, Golubovići, Golubovića pećina, 02.05.2013, P. Kosovac
INJ10 (CINJ)	*Anthroherpon*	*cylindricolle*	*cylindricolle*	BIH, Rogatica, Golubovići, Golubovića pećina, 02.05.2013, P. Kosovac
INJ11 (CINJ)	*Anthroherpon*	*cylindricolle*	*cylindricolle*	BIH, Rogatica, Golubovići, Golubovića pećina, 02.05.2013, P. Kosovac
INJ12 (CINJ)	*Anthroherpon*	*stenocephalum*	*stenocephalum*	BIH, Olovo, Bijambare, 04.05.2013, I. Njunjić
INJ13 (CINJ)	*Anthroherpon*	*stenocephalum*	*stenocephalum*	BIH, Olovo, Bijambare, 04.05.2013, I. Njunjić
INJ14 (CINJ)	*Anthroherpon*	*stenocephalum*	*stenocephalum*	BIH, Olovo, Bijambare, 04.05.2013, I. Njunjić
INJ15 (CINJ)	*Anthroherpon*	*stenocephalum*	*stenocephalum*	BIH, Olovo, Bijambare, 04.05.2013, I. Njunjić
INJ16 (CINJ)	*Anthroherpon*	*cylindricolle*	*thoracicum*	BIH, Pale, Careve vode, Novakova pećina, 30.08.2013.
INJ17 (CINJ)	*Anthroherpon*	*cylindricolle*	*thoracicum*	BIH, Pale, Careve vode, Novakova pećina, 30.08.2013.

INJ18 (CINJ)	*Anthroherpon*	*cylindricolle*	*thoracicum*	BIH, Pale, Careve vode, Novakova pećina, 30.08.2013.
INJ19 (CINJ)	*Anthroherpon*	*cylindricolle*	*thoracicum*	BIH, Pale, Careve vode, Novakova pećina, 30.08.2013.
INJ20 (CINJ)	*Anthroherpon*	*cylindricolle*	*thoracicum*	BIH, Pale, Careve vode, Novakova pećina, 30.08.2013.
INJ21 (CINJ)	*Anthroherpon*	*sinjajevina*		CG, Sinjajevina, Rudanca, Blažova pećina, 24.06.2013. I. Njunjić
INJ22 (CINJ)	*Anthroherpon*	*sinjajevina*		CG, Sinjajevina, Rudanca, Blažova pećina, 24.06.2013, I. Njunjić
INJ23 (CINJ)	*Anthroherpon*	*sinjajevina*		CG, Sinjajevina, Rudanca, Blažova pećina, 24.06.2013, P. Kosovac
INJ24 (CINJ)	*Anthroherpon*	*cylindricolle*	*cylindricolle*	BIH, Rogatica, Golubovići, Golubovića pećina, 30.08.2013, P. Kosovac
INJ25 (CINJ)	*Anthroherpon*	*cylindricolle*	*cylindricolle*	BIH, Rogatica, Golubovići, Golubovića pećina, 30.08.2013, P. Kosovac
INJ26 (CINJ)	*Anthroherpon*	*cylindricolle*	*cylindricolle*	BIH, Rogatica, Golubovići, Golubovića pećina, 30.08.2013, P. Kosovac
INJ27 (CINJ)	*Anthroherpon*	*cylindricolle*	*cylindricolle*	BIH, Rogatica, Golubovići, Golubovića pećina, 30.08.2013, P. Kosovac
INJ28 (CINJ)	*Anthroherpon*	*hoermanni*	*hoermanni*	BIH, Kalinovik, Borija, Borija pećina, 20.06.2013, I. Njunjić
INJ29 (CINJ)	*Anthroherpon*	*hoermanni*	*hoermanni*	BIH, Kalinovik, Borija, Borija pećina, 20.06.2013, I. Njunjić
INJ30 (CINJ)	*Anthroherpon*	*hoermanni*	*hoermanni*	BIH, Kalinovik, Borija, Borija pećina, 20.06.2013, I. Njunjić
INJ31 (CINJ)	*Anthroherpon*	*charon*		BIH, Trebević, Luke, Ledenica,17.06.2013, I. Njunjić
INJ32 (CINJ)	*Graciliella*	*metohijensis*		Hercegovina, Korita, Kobilja glava, Veliko Đatlo, 13.09.2006, M. Perreau
INJ33 (CINJ)	*Graciliella*	*metohijensis*		Hercegovina, Nevesinje, Balabani, Drvendžina pećina, 21.12.2003, M. Đokić
INJ34 (CINJ)	*Graciliella*	*metohijensis*		Hercegovina, Nevesinje, Balabani, Drvendžina pećina, 21.12.2003, M. Đokić
INJ35 (CINJ)	*Anthroherpon*	*charon*		BIH, Trebević, Luke, Ledenica,17.06.2013, I. Njunjić
INJ36 (CINJ)	*Anthroherpon*	*charon*		BIH, Trebević, Luke, Ledenica,17.06.2013, I. Njunjić
INJ37 (CINJ)	*Anthroherpon*	*charon*		BIH, Trebević, Luke, Ledenica,17.06.2013, I. Njunjić
INJ38 (CINJ)	*Anthroherpon*	*harbichi*		BIH, Sarajevo, Crepoljsko polje, Kečina stena, 01.05-18.06.2013, I. Njunjić
INJ39 (CINJ)	*Anthroherpon*	*harbichi*		BIH, Sarajevo, Crepoljsko polje, Kečina stena, 01.05-18.06.2013, I. Njunjić
INJ40 (CINJ)	*Anthroherpon*	*harbichi*		BIH, Sarajevo, Crepoljsko polje, Kečina stena, 05.2013, I. Njunjić
INJ41 (CINJ)	*Anthroherpon*	*harbichi*		BIH, Sarajevo, Crepoljsko polje, Kečina stena, 05.2013, I. Njunjić
INJ42 (CINJ)	*Anthroherpon*	*harbichi*		BIH, Sarajevo, Crepoljsko polje, Kečina stena, 01.09.2013, I. Njunjić
INJ43 (CINJ)	*Anthroherpon*	*sinjajevina*		MNE, Sinjajevina, Rudanca, Blažova pećina, 24.06.2013, I. Njunjić
INJ44 (CINJ)	*Anthroherpon*	*sinjajevina*		MNE, Sinjajevina, Rudanca, Blažova pećina, 24.06.2013, I. Njunjić
INJ45 (CINJ)	*Graciliella*	*ozimeci*		CRO, Dubrovnik, Točionik, Vranja pećina, 08.11.2014, R. Ozimec
INJ46 (CINJ)	*Graciliella*	*ozimeci*		CRO, Dubrovnik, Točionik, Vranja pećina, 08.11.2014, R. Ozimec
INJ47 (CINJ)	*Graciliella*	*ozimeci*		CRO, Dubrovnik, Točionik, Vranja pećina, 08.11.2014, R. Ozimec
INJ48 (CINJ)	*Graciliella*	*ozimeci*		CRO, Dubrovnik, Točionik, Vranja pećina, 08.11.2014, R. Ozimec
INJ49 (CINJ)	*Graciliella*	*ozimeci*		CRO, Dubrovnik, Točionik, Vranja pećina, 08.11.2014, R. Ozimec
INJ50 (CINJ)	*Graciliella*	*ozimeci*		MNE, Orjen, Grahovo, Vojvode Dakovića, 22.08.2007, M. Perreau
INJ51 (CINJ)	*Graciliella*	*ozimeci*		CRO, Dubrovnik, Točionik, Vranja pećina, 08.11.2014, R. Ozimec
INJ52 (CINJ)	*Graciliella*	*ozimeci*		CRO, Dubrovnik, Točionik, Vranja pećina, 08.11.2014, R. Ozimec
INJ53 (CINJ)	*Graciliella*	*ozimeci*		CRO, Dubrovnik, Točionik, Vranja pećina, 08.11.2014, R. Ozimec
INJ54 (CINJ)	*Graciliella*	*kosovaci*		CRO, Tunel Konavle, Kaverna 781, 09.11.2014, R. Ozimec

INJ55 (CINJ)	*Gracileilla*	*metohijensis*		Hercegovina, Nevesinje, Balabani, Drvendžina pećina, 26.08.2013, I. Njunjić
INJ56 (CINJ)	*Graciliella*	*metohijensis*		Hercegovina, Nevesinje, Balabani, Drvendžina pećina, 26.08.2013, I. Njunjić
INJ57 (CINJ)	*Graciliella*	*metohijensis*		Hercegovina, Nevesinje, Balabani, Drvendžina pećina, 26.08.2013, I. Njunjić
INJ58 (CINJ)	*Graciliella*	*apfelbecki*		CRO, Bobani, Grabovica, 08.05.2011, R. Ozimec
INJ59 (CINJ)	*Graciliella*	*metohijensis*		Hercegovina, Nevesinje, Balabani, Drvendžina pećina, 26.08.2013, I. Njunjić
INJ60 (CINJ)	*Graciliella*	*metohijensis*		Hercegovina, Nevesinje, Balabani, Drvendžina pećina, 26.08.2013, I. Njunjić
INJ61 (CINJ)	*Graciliella*	*metohijensis*		Hercegovina, Nevesinje, Balabani, Drvendžina pećina, 26.08.2013, I. Njunjić
INJ62 (CINJ)	*Graciliella*	*metohijensis*		Hercegovina, Nevesinje, Balabani, Drvendžina pećina, 26.08.2013, I. Njunjić
INJ63 (CINJ)	*Graciliella*	*metohijensis*		Hercegovina, Nevesinje, Balabani, Drvendžina pećina, 26.08.2013, I. Njunjić
INJ64 (CINJ)	*Graciliella*	*metohijensis*		Hercegovina, Nevesinje, Balabani, Drvendžina pećina, 26.08.2013, I. Njunjić
INJ65 (CINJ)	*Graciliella*	*metohijensis*		Hercegovina, Nevesinje, Balabani, Drvendžina pećina, 26.08.2013, I. Njunjić
INJ66 (CINJ)	*Graciliella*	*metohijensis*		Hercegovina, Nevesinje, Balabani, Drvendžina pećina, 26.08.2013, I. Njunjić
INJ67 (CINJ)	*Graciliella*	*metohijensis*		Hercegovina, Nevesinje, Balabani, Drvendžina pećina, 26.08.2013, I. Njunjić
INJ68 (CINJ)	*Graciliella*	*metohijensis*		Hercegovina, Nevesinje, Balabani, Drvendžina pećina, 26.08.2013, I. Njunjić
INJ69 (CINJ)	*Graciliella*	*ozimeci*		CRO, Dubrovnik, Točionik, Vranja pećina, 08.11.2014, R. Ozimec
INJ70 (CINJ)	*Graciliella*	*ozimeci*		CRO, Dubrovnik, Točionik, Vranja pećina, 08.11.2014, R. Ozimec
INJ71 (CINJ)	*Graciliella*	*apfelbecki*	*apfelbecki*	CRO, Gromača, Špilja za Gromačkom vlakom, 28.08.2014, R. Ozimec
INJ72 (CINJ)	*Anthroherpon*	*primitivum*		BIH, Trebinje, Turica, Mravinjac, 31.07.2013, M. Perreau
INJ73 (CINJ)	*Anthroherpon*	*primitivum*		BIH, Trebinje, Turica, Mravinjac, 31.07.2013, M. Perreau
INJ74 (CINJ)	*Anthroherpon*	*primitivum*		BIH, Trebinje, Turica, Mravinjac, 31.07.2013, M. Perreau
INJ75 (CINJ)	*Anthroherpon*	*primitivum*		BIH, Trebinje, Turica, Mravinjac, 31.07.2013, M. Perreau
INJ76 (CINJ)	*Anthroherpon*	*taxi*	*albanicum*	Prokletije, Ćaf Borit, Kolektor, 24.07.2012, M. Perreau
INJ77 (CINJ)	*Anthroherpon*	*taxi*	*albanicum*	Prokletije, Ćaf Borit, Kolektor, 24.07.2012, M. Perreau
INJ78 (CINJ)	*Anthroherpon*	*taxi*	*albanicum*	Prokletije, Ćaf Borit, Kolektor, 24.07.2012, M. Perreau
INJ79 (CINJ)	*Anthroherpon*	*zariquieyi*		MNE, Durmitor, Pećina u Sedlenoj gredi, 15.07.2014, I. Njunjić
INJ80 (CINJ)	*Anthroherpon*	*zariquieyi*		MNE, Durmitor, Pećina u Sedlenoj gredi, 15.07.2014, I. Njunjić
INJ81 (CINJ)	*Anthroherpon*	*zariquieyi*		MNE, Durmitor, Pećina u Sedlenoj gredi, 15.07.2014, I. Njunjić
INJ82 (CINJ)	*Anthroherpon*	*hoermanni*	*hypsophilum*	Hercegovina, Lebršnik, Dvogrla jama, 18.09.2006, M. Perreau
INJ83 (CINJ)	*Anthroherpon*	*hoermanni*	*hypsophilum*	Hercegovina, Lebršnik, Dvogrla jama, 18.09.2006, M. Perreau
INJ84 (CINJ)	*Anthroherpon*	*hoermanni*	*hypsophilum*	Hercegovina, Lebršnik, Vilina pećina,09.09.2006. M. Perreau
INJ85 (CINJ)	*Anthroherpon*	*hoermanni*	*hypsophilum*	Hercegovina, Lebršnik, Dvogrla jama, 18.09.2006, M. Perreau
INJ86 (CINJ)	*Anthroherpon*	*hoermanni*	*hypsophilum*	Hercegovina, Lebršnik, Dvogrla jama, 18.09.2006, M. Perreau
INJ87 (CINJ)	*Anthroherpon*	*hoermanni*	*hypsophilum*	Hercegovina, Lebršnik, Čavčarica jama, 09.09.2005, M. Perreau
INJ88 (CINJ)	*Anthroherpon*	*hoermanni*	*hoermanni*	BIH, Zelengora, Nedavići, Vareničina pećina, 21.06.2013, I. Njunjić
INJ89 (CINJ)	*Graciliella*	*metohijensis*		CG, Somina, Donje Čarađe, Prljača, 06.08.2010, S. Ognjenović
INJ90 (CINJ)	*Graciliella*	*apfelbecki*		Hercegovina, Bileća, Bijele Rudine, jama Vranjača, 07.06.2005, M. Perreau
INJ91 (CINJ)	*Graciliella*	*apfelbecki*		Hercegovina, Bileća, Bijele Rudine, jama Vranjača, 07.06.2005, M. Perreau

INJ92 (CINJ)	*Graciliella*	*ganglbaueri*	*ganglbaueri*	BIH, Nevesinje, Bišina, Novakuša, 21.06.2013, I. Njunjić
INJ93 (CINJ)	*Anthroherpon*	*ganglbaueri*	*ganglbaueri*	BIH, Nevesinje, Bišina, Novakuša, 21.06.2013, I. Njunjić
INJ94 (CINJ)	*Anthroherpon*	*ganglbaueri*	*ganglbaueri*	BIH, Nevesinje, Bišina, Novakuša, 21.06.2013, I. Njunjić
INJ95 (CINJ)	*Graciliella*	*apfelbecki*	*lahneri*	CG, Virpazar, env. Trnovo
INJ96 (CINJ)	*Graciliella*	*apfelbecki*	*scutulatum*	Bravenik
INJ97 (CINJ)	*Anthroherpon*	*matzenaueri*	*taliensis*	CG, Maganik, Kapetanovo jezero, Dola pećina, 01.08.2013, Mp. Perreau
INJ98 (CINJ)	*Anthroherpon*	*cecai*		CG, Durmitor, Mala Crna Gora, Jama u Podu, 03.08.1992, A. Milosavljević
INJ99 (CINJ)	*Anthroherpon*	*taxi*	*sydowi*	CG, Tali Mt, Mlječikova pećina, 03.08.2013, M. Perreau
INJ100 (CINJ)	*Anthroherpon*	*latipenne*		CG, Somina, Donje Čarađe, Prljača, 06.08.2010, S. Ognjenović
INJ101 (CINJ)	*Anthroherpon*	*latipenne*	*punctipennis*	CG, Maganik, Kapetanovo jezero, Dola pećina, 01.08.2013, Mp. Perreau
INJ102 (CINJ)	*Anthroherpon*	*taxi*	*albanicum*	CG, Prokletije, ćaf Borit, Jama kolektor, 25.07.2011-24.07.2012, M. Popović
INJ103 (CINJ)	*Anthroherpon*	*taxi*	*albanicum*	CG, Prokletije, ćaf Borit, Jama kolektor, 25.07.2011-24.07.2012, M. Popović
INJ104 (CINJ)	*Anthroherpon*	*taxi*	*taxi*	CG, Orjen, Kameno more, Dvestotka, 13.08.2009, I. Njunjić
INJ105 (CINJ)	*Graciliella*	*apfelbecki*	*lahneri*	CG, Trnovo, Grbočica, 28.04.2004.
INJ106 (CINJ)	*Anthroherpon*	*ganglbaueri*	*ganglbaueri*	BIH, Nevesinje, Bišina, Novakuša, 21.06.2013, I. Njunjić
INJ107 (CINJ)	*Anthroherpon*	*ganglbaueri*	*ganglbaueri*	BIH, Nevesinje, Bišina, Novakuša, 21.06.2013, I. Njunjić
INJ108 (CINJ)	*Anthroherpon*	*ganglbaueri*	*ganglbaueri*	BIH, Nevesinje, Bišina, Novakuša, 21.06.2013, I. Njunjić
INJ109 (CINJ)	*Anthroherpon*	*ganglbaueri*	*ganglbaueri*	BIH, Nevesinje, Bišina, Novakuša, 21.06.2013, I. Njunjić
INJ110 (CINJ)	*Anthroherpon*	*latipenne*		CG, Orjen, Križeva jama
INJ111 (CINJ)	*Anthroherpon*	*latipenne*		CG, Orjen, Križeva jama
INJ112 (CINJ)	*Anthroherpon*	*latipenne*		CG, Somina, Donje Čarađe, Prljača, 06.08.2010, S. Ognjenović
INJ113 (CINJ)	*Anthroherpon*	*latipenne*		CG, Somina, Donje Čarađe, Prljača, 06.08.2010, S. Ognjenović
INJ114 (CINJ)	*Anthroherpon*	*taxi*	*taxi*	CG, Orjen, Kameno more, Dvestotka, 13.08.2009, I. Njunjić
INJ115 (CINJ)	*Anthroherpon*	*taxi*	*taxi*	CG, Orjen, Kameno more, Dvestotka, 13.08.2009, I. Njunjić
INJ116 (CINJ)	*Anthroherpon*	*pygmaeum*	*stricticolle*	BIH, Bjelašnica, Sudareva pećina, 03.09.2013, I. Njunjić
INJ117 (CINJ)	*Anthroherpon*	*pygmaeum*	*stricticolle*	BIH, Bjelašnica, Sudareva pećina, 03.09.2013, I. Njunjić
INJ118 (CINJ)	*Anthroherpon*	*pygmaeum*	*stricticolle*	BIH, Bjelašnica, Sudareva pećina, 03.09.2013, I. Njunjić
INJ119 (CINJ)	*Anthroherpon*	*pygmaeum*	*stricticolle*	BIH, Bjelašnica, Sudareva pećina, 03.09.2013, I. Njunjić
INJ120 (CINJ)	*Anthroherpon*	*pygmaeum*	*stricticolle*	BIH, Bjelašnica, Sudareva pećina, 03.09.2013, I. Njunjić
INJ121 (CINJ)	*Anthroherpon*	*taxi*	*taxi*	CG, Orjen, Dvestotka, 13.08.2009, I. Njunjić
INJ122 (CINJ)	*Anthroherpon*	*taxi*	*taxi*	CG, Orjen, Dvestotka, 13.08.2009, I. Njunjić
INJ123 (CINJ)	*Anthroherpon*	*hoermanni*	*hoermanni*	BIH, Kalinovik, Borija, Borija pećina, 20.06.2013, I. Njunjić
INJ124 (CINJ)	*Anthroherpon*	*taxi*	*taxi*	CG, Orjen, Dvestotka, 13.08.2009, I. Njunjić
INJ125 (CINJ)	*Anthroherpon*	*taxi*	*taxi*	CG, Orjen, Dvestotka, 13.08.2009, I. Njunjić
INJ126 (CINJ)	*Anthroherpon*	*taxi*	*albanicum*	Prokletije, Ćaf Borit, Babina sisa, 15.07.2012, M. Perreau
INJ127 (CINJ)	*Anthroherpon*	*taxi*	*albanicum*	Prokletije, Ćaf Borit, Babina sisa, 15.07.2012, M. Perreau
INJ128 (CINJ)	*Graciliella*	*metohijensis*		BIH, Hercegovina, Korita, Kobilja glava, Đatlo, 05.08.2013, M. Perreau

INJ129 (CINJ)	*Anthroherpon*	*zariquieyi*		CG, Durmitor, Sedlo, Pećina u Sedlenoj gredi, 24.06.2013, I.Njunjić
IE44 (CINJ)	*Anthroherpon*	*stenocephalum*	stenocephalum	BIH, Olovo, Bijambare, 04.05.2013, I. Njunjić
VA5 (CINJ)	*Anthroherpon*	*erebus*	scheibeli	BIH, Trnovo, Bjeličina pećina
MP1 (CMPR)	*Graciliella*	*apfelbecki*		BIH, Popovo polje, Vjeternica, M. Perreau
MP2 (CMPR)	*Graciliella*	*apfelbecki*		BIH, Popovo polje, Vjeternica, M. Perreau
MP3 (CMPR)	*Anthroherpon*	*latipenne*	*latipenne*	Hercegovina, Gubar Mt, Bukova rupa, 11.09.2008. M. Perreau
MP4 (CMPR)	*Anthroherpon*	*latipenne*	*latipenne*	Hercegovina, Gubar Mt, Bukova rupa, 11.09.2008. M. Perreau
MP5 (CMPR)	*Graciliella*	*metohijensis*		BIH, Brestica near Korita, Jametina jama, 14.08.2007, M. Perreau
MP6 (CMPR)	*Graciliella*	*metohijensis*		BIH, Brestica near Korita, Jametina jama, 14.08.2007, M. Perreau
MNHN	*Anthroherpon*	*charon*		Trebević, Reitter
MNHN	*Anthroherpon*	*charon*		Trebević, Reitter
MNHN	*Anthroherpon*	*charon*		Ledenjača, Trebević, Svirčev 11.29.
MNHN	*Anthroherpon*	*erebus*	*scheibeli*	Trnovo (type)
MNHN	*Anthroherpon*	*erebus*	*scheibeli*	Trnovo (cotype)
MNHN	*Anthroherpon*	*erebus*	*scheibeli*	Trnovo
MNHN	*Anthroherpon*	*primitivum*		Heregovina, Nevada, Grebci, Čelina pećina, 16.07.1930, Svirčev
MNHN	*Anthroherpon*	*weiratheri*		BIH, Sarajevo, Kečina stena
MNHN	*Anthroherpon*	*weiratheri*		BIH, Sarajevo, Kečina stena
MNHN	*Graciliella*	*apfelbecki*		BIH, Zavala, Vjeternica (cotype)
MNHN	*Graciliella*	*apfelbecki*		Popovo polje, Grabovica
MNHN	*Graciliella*	*apfelbecki*		Dubrovnik, Močiljska pećina
MNHN	*Graciliella*	*apfelbecki*		Dubrovnik, Močiljska pećina
MNHN	*Graciliella*	*apfelbecki*		Dubrovnik, Močiljska pećina
MNHN	*Graciliella*	*apfelbecki*		Hercegovina, Grebci, Kali pećina

CHAPTER V

ARTICLE 3 (submitted):

MOLECULAR PHYLOGENETICS AND SYSTEMATICS OF THE CAVE BEETLE GENERA *REMYELLA* AND *ROZAJELLA* (COLEOPTERA: LEIODIDAE: CHOLEVINAE: LEPTODIRINI)

To investigate the phylogenetic placement of the genus *Anthroherpon* among related genera, we used molecular and morphological approaches to examine the phylogenetic position of *Remyella* Jeannel, 1931 and *Rozajella* S. Ćurčić, Brajković & B. Ćurčić, 2007, two genera that were considered to be closely related to Anthroherponina. The genus *Remyella* was previously placed in the Anthroherponini (today: subtribe Anthroherponina) based on morphology (Jeannel, 1931). Perreau and Pavićević (2008) reconstructed the phylogeny of this subtribe based on morphological characters and excluded the genus *Remyella* from this subtribe. Presently, both genera are tentatively placed in Leptodirina.

ARTICLE 3: Unravelling phylogenetic position and clarifying the puzzling taxonomic situation of cave beetle genera *Remyella* and *Rozajella* (Coleoptera: Leiodidae: Cholevinae: Leptodirini)

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Key words
Leptodirini, Mantel test, Montenegro, Serbia, troglobites.

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Abstract

The subtribe Leptodirina is one of the most species-rich subtribes of the tribe Leptodirini, comprising 31 genera of beetles adapted to subterranean environment and distributed in West Paleartic. The relationships among genera of this subtribe are far from evident due to potentially convergent morphological characters resulting from the adaptation to the subterranean environment. Two genera with uncertain systematic position– *Rozajella* S. Ćurčić, Brajković & B. Ćurčić, 2007 and *Remyella* Jeannel, 1931 are tentatively placed to the subtribe Leptodirina based on morphological characters. However, no molecular phylogenetic studies have been performed within this subtribe to support or refute the phylogenetic position of these genera. Moreover, the

morphological distinctions of species and subspecies of the genus *Remyella* are tenuous, their geographical distribution hardly coherent, and their validity disputable.

To examine the phylogenetic position of these two genera showing shared morphological characters and living in neighbouring geographic areas (Pešter polje and surrounding massifs), we performed a molecular phylogenetic analysis. Detailed molecular and morphological investigation of the taxa within *Remyella* is also provided to clarify the puzzling taxonomic situation of the existing taxa. The speciation events within *Remyella* occurred approximately from 5 to 2 million years ago which corresponds to the end of the Miocene and the Pliocene, periods when the Pešter field was periodically filled by shallow lakes. The distribution area of the genus, confined to the periphery of Pešter is probably contingent to the presence of these lakes, preventing the species to spread more uniformly over the entire area of the field. Additionally, the correlations between genetic and geographic distances were investigated using the Mantel test. Four synonymies inside *Remyella* are established and all species of the genus are briefly diagnosed. Identification tables and distribution maps of species of *Remyella* and *Rozajella* are provided, and a new species of the genus *Rozajella* is described.

5.1. INTRODUCTION

The tribe Leptodirini Lacordaire, 1854 is the most speciose tribe in the subfamily Cholevinae of the family Leiodidae (Coleoptera) and comprises 195 genera and 925 (mostly polytypic) species (Newton, 2016). All species are more or less adapted to subterranean environment (except two termitophilous species) and all are fully anophtalmic (except some 15 species that have extremely reduced eyes). The characters shared by all members of this tribe are: female protarsi tetramerous and spiculum gastrale of the abdominal IX segment in males fused with lateral epipleurites. Most genera have complete mesoventral processus separating the two mesocoxal cavities (except a few species of Anthroherponina in which this structure secondarily disappeared) (Perreau, 2008). Leptodirini have a Palearctic distribution: Iberian Peninsula, Balkan Peninsula, southern Alps, Romania, southern Russia, the Caucasus, Middle East, and Iran, with the highest diversity in the Mediterranean basin (Perreau, 2000, 2004; Ribera et al., 2010).

The phylogeny of Leptodirini remains poorly understood, although several systematic and evolutionary studies were recently performed on this group (Ribera et al., 2010; Fresneda et al., 2011; Cieslak et al., 2015). Apart from the pioneering work of Caccone & Sbordoni (2001), molecular approaches of the phylogeny of Leptodirini are recent and concern Western Mediterranean genera only (Ribera et al., 2010; Fresneda et al., 2011). Comprehensive studies of Leptodirini distributed in the Eastern Mediterranean have started to emerge only recently (Perreau & Pavićević, 2008b; Njunjić et al., 2016). Among the Leptodirini, but also among all Coleoptera with troglomorphic adaptations, the subtribe Anthroherponina shows the most pronounced troglomorphic morphological characters: extremely long appendages, extreme elongation of the head and pronotum, and hemispherical elytra (Njunjić et al., 2015). The subtribe Anthroherponina was traditionally defined by the position of the antennal insertions on the posterior third of the head and the enlarged and apically blunt claws (Jeannel, 1924). Jeannel (1924) used this combination to characterize the "Anthroherpona" composed of the "phyletic series of *Anthroherpon*" (today: subtribe Anthroherponina) and the "phyletic series of *Spelaeobates*" (today: subtribe Spelaeobatina). The latter is nowadays generally considered to be phylogenetically more closely related to Bathysciina (Casale et al. 1991) than Anthroherponina. Jeannel (1931) placed the genus *Remyella* in the Anthroherponini based on the insertion of the antennae on the posterior third of the head, and despite its claw conformation different from other genera of this subtribe (narrow in *Remyella* and wide in Anthroherponina). Perreau and Pavićević (2008b) reconstructed the phylogeny of Anthroherponina based on morphological characters and excluded the genus *Remyella* from this subtribe, placing it tentatively into the subtribe Leptodirina. The monophyly of Leptodirina, however, is highly questionable (Perreau & Pavićević, 2008b), especially because many species are highly troglomorphic and many characters are potentially homoplasious.

To examine the phylogenetic position of *Remyella* and *Rozajella*, two genera having shared morphological characters and living in neighbouring geographic areas (Pešter polje and surrounding massifs), we performed a molecular phylogenetic analysis. A detailed molecular and morphological investigation of the taxa within *Remyella* is also provided, since up to now the morphological distinctions of species and subspecies are tenuous, their geographical distribution hardly coherent, and their validity disputable. We also show a strong correlation between genetic and geographic distances among the different populations of *Remyella*. After this revision, *Remyella* contains five species, without subspecies. The genetic analysis reveals the close phylogenetic relationship of *Rozajella* and *Remyella*. Taking into account morphology, the set *Nonveilleriella*+*Rozajella*+*Remyella* is likely monophyletic. Additionally we describe a new species: *Rozajella madzgalji* **n. sp.** and the male of *R. deelemani*, of which only females were previously known.

5.2. MATERIAL AND METHODS

5.2.1. Acronyms

The following abbreviations for collections and institutions are used: CDPV: Dragan Pavićević Collection, Belgrade, Serbia; CINJ: Iva Njunjić Collection, Belgrade, Serbia; CMPR: Michel Perreau Collection, Paris, France; MNHN: Muséum National d'Histoire Naturelle, Paris, France; Naturalis: Naturalis Biodiversity Center, Leiden, The Netherlands.

The following abbreviations for morphological details are used: HT: holotype; PT: paratype. In table 3, TL means: type locality.

5.2.2. Taxon sampling

A total of 51 specimens representing 24 taxa from 12 genera were collected for molecular analysis in caves of the Dinaric range, in Montenegro, Serbia, and Bosnia and Herzegovina as listed in supporting information Table S1. For amplification and sequencing we used seven specimens of *R. madzgalji* n. sp., three specimens of *R. deelemani* and one or two specimens per population of other genera. Ten populations of *Remyella* were available for molecular investigations (24 for morphological investigations). For outgroup rooting we chose three species of the subtribe Pholeuina: *Antrocharis querilhaci* (Lespes, 1857), *Stygiophyes akarsticus* (Escolà, 1980), and *Speonomus diecki* (Saulcy, 1872), of which sequences are available from Ribera et al.

(2010). All populations examined (including molecular and morphological studies) of the genera *Remyella*, *Rozajella*, and *Nonveilleriella* are listed in Table 3.

5.2.3. Morphological study

After extraction, male genitalia were cleared in clove oil, mounted in Euparal on glass slides and pinned beneath the specimens. The external morphology of specimens was examined using Leica MZ75 and Leica M10 stereomicroscopes. Microphotographs were taken on a Leica DIAPLAN or a Zeiss AXIOLAB microscope with a camera diagnostic instrument Spot INSIGHT IN1820. Photographs of habitus (Figs. 2, 3) were taken using a Nikon SMZ 1000 stereomicroscope with a Canon EOS 60D camera. Digital pictures were processed using the Helicon Focus software 5.3 (http://www.heliconsoft.com/heliconsoft-products/helicon-focus/). High resolution pictures of external morphology were taken with a microscope Keyence VHX5000 with lenses VH-Z20T (Figs. 28-35) and VH-Z250T (Figs. 4-7; 36-37).

5.2.4. DNA extraction, PCR amplification, and sequencing

The specimens used in the study were collected alive in the field and preserved in 96% ethanol. DNA was extracted from whole specimens or from one leg with a standard phenol–chloroform extraction (Blin & Stafford, 1976) or the DNeasy Tissue Kit (Qiagen GmbH, Hilden, Germany). Voucher specimens are stored in the MNHN (Paris, France) and DNA aliquots are kept in the tissue collections of Naturalis (Leiden, The Netherlands).

We amplified fragments of two nuclear genes: 5’ end of the small ribosomal subunit, 18S rRNA (SSU), and an internal fragment of the large ribosomal subunit, 28S rRNA (LSU), and of two non-overlapping sections of mitochondrial gene fragments: the 5’ and 3’ halves of cytochrome c oxidase subunit 1 (which we here term COIa and COIb, respectively). Primers used are given in Table 1, and PCR protocols are given in Table 2. Sequences were assembled and edited using Geneious version 8.0.5 (http://www.geneious.com/, Kearse et al., 2012). DNA sequences obtained for each genetic marker were aligned separately using MAFFT version 7 (Katoh & Standley, 2013). Sequences have been deposited in GenBank with Accession Numbers (will be provided) given in supporting information Table S1.

Table 1. Primers used in the study.

Fragment	**Name**	**Sense**	**Sequence 5’-3’**	**Reference**
COIa	LCOI-1490	F	GGTCAACAAATCATAAAGATATTG	Folmer et al. (1994)
COIa	HCOI-2198	R	TAAACTTCAGGGTGACCAAAAAATCA	Folmer et al. (1994)
COIb	Jerry	F	CAACATTTATTTTGATTTTTTGG	Simon et al. (1994)
COIb	Pat	R	TCCAATGCACTAATCTGCCATATTA	Simon et al. (1994)
18S	5’	F	GACAACCTGGTTGATCCTGCCAGT	Shull et al. (2001)
18S	b5.0	R	TAACCGCAACAACTTTAAT	Shull et al. (2001)
28S	Ka	F	ACACGGACCAAGGAGTCTAGCATG	Ribera et al. (2010)
28S	Kb	R	CGTCCTGCTGTCTTAAGTTAC	Ribera et al. (2010)

Table 2. PCR cycling conditions.

Number of cycles: 40

Temperature (C)	Time (min)
95	3:00
94	0:15
54	0:30
72	0:40
72	5:00
12	∞

5.2.5. Phylogenetic analyses

For each sequence alignment, the optimal model of nucleotide substitution was determined using jModelTest2 (Darriba et al., 2012; Guindon & Gascuel, 2003), run on the CIPRES webportal (Miller et al., 2010). Selection was based on the Akaike Information Criterion (AIC), and resulted in selection of SYM+I, GTR+G, HKY+I+G, and GTR+I+G substitution models for 18S, 28S, COIa and COIb, respectively. Phylogenetic analysis was performed using MrBayes 3.2.1 (Ronquist & Huelsenbeck, 2003) on CIPRES (Miller et al., 2010), with settings for multiple markers unlinked, running two replicates of 15x10^{6} generations each. Sample frequency was set to 5000. After discarding a 25% burn-in, the resulting majority-rule consensus tree was visualized using FigTree version

1.4 (Rambaut, 2012). The correlations between genetic and geographic distances were investigated using the Mantel test through the software XLSTAT.

5.2.6. Distribution map

The map of Fig. 39 was built using SRTM3 data (Shuttle Radar Topography Mission) of NASA with the software QGIS. GDAM (global administrative area: www.gdam.org) data were used for country frontiers and Natural Earth data (http://www.naturalearthdata.com/) were used for hydrographic features: rivers and lakes. Other marks (location of species and caves, geographical names, and delimitations of distributions areas) were inserted with Photoshop CS2.

5.3. RESULTS AND DISCUSSION

5.3.1. Phylogenetic relationships and systematic placement of *Remyella, Rozajella*, and *Nonveilleriella*

The molecular investigations based on 18S, 28S, and COI gene fragments reveal the close relationship of *Remyella* and *Rozajella* (posterior probability = 1.00) (Fig. 1). Three genera of the subtribe Anthroherponina (*Leptomeson* Jeannel, 1924, *Anthroherpon* Reitter, 1889, and *Hadesia* Müller, 1911) form a highly supported clade (posterior probability = 1.00). This clade appears as the sister group of the genus *Speonesiotes* (subtribe Bathysciotina). However, this relationship is weakly supported (posterior probability = 0.56) and is unlikely on morphological grounds.

Rozajella and *Remyella* were previously (and tentatively) placed in Leptodirina (Perreau & Pavićević, 2008a, 2008b). On the basis of our phylogenetic analysis, the polyphyly of Leptodirina as presently defined appears clearly, and the tribal assignation of *Remyella*, *Rozajella* (and also *Nonveilleriella*) should be reconsidered. However, the clarification of this question requires to take into consideration the whole tribe Leptodirina and not only the small number of genera used in this work (*Apholeuonus*, *Charonites*, and *Parapropus*). That is, however, out of the scope of this paper.

Even though we do not have genetic data for the genus *Nonveilleriella* Perreau & Pavićević, 2008, based on its morphology, this genus is clearly closely related to the genus *Rozajella*. They share the following apomorphic character states: (i) strong and dense microsculpture of the pronotum; (ii) explanate surface of the pronotum near the posterior angles (Fig. 6); (iii) punctuation of elytra transversely aligned with transverse strigae (Fig. 7) (also sporadically aligned in some specimens of *Remyella*, but without strigae). Punctuation of elytra is never transversally aligned in other troglomorphic species of Leptodirina or Anthroherponina (except some traces of alignments in *Anthroherpon udrzali* Giachino & Vailati). With *Remyella,* these two genera share the "V"-shape of the metendosternite (Figs. 10; 12; 13) rather than the "Y"-shape (with a longer and thinner basal part) that is found in most other troglomorphic species of Leptodirini, for instance *Anthroherpon primitivum* Absolon (Fig. 8) and *Parapropus sericeus* Schmidt (Fig. 9).

These three genera (*Remyella*, *Rozajella*, and *Nonveilleriella*) also share a continuous geographical area in Sandžak (a historical geo-political region straddling the

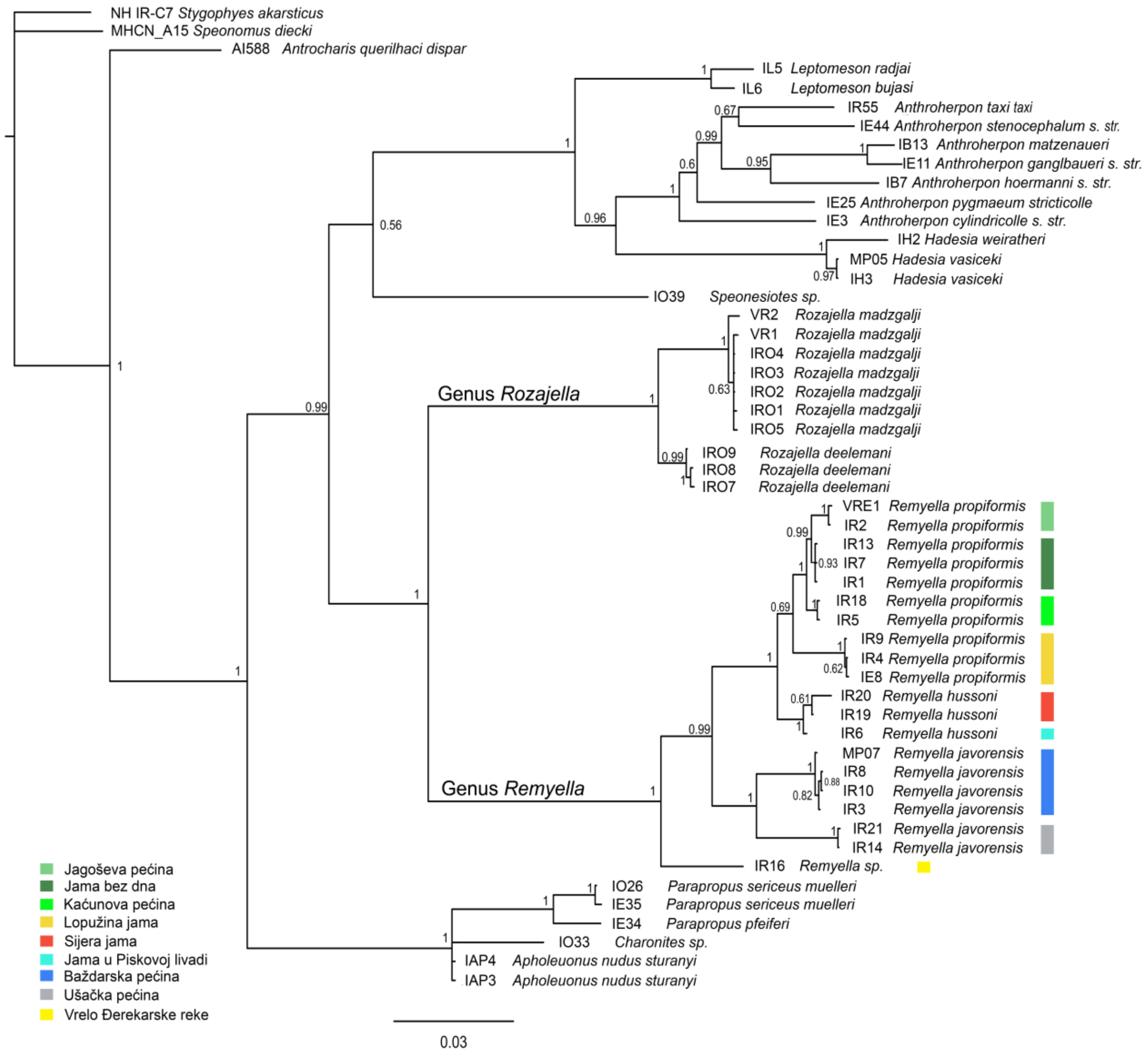


Fig. 1. Bayesian inference of combined 18S, 28S, COIa, and COIb data sets for *Remyella* and *Rozajella*. Numbers above nodes are posterior probabilities.

border between Serbia and Montenegro). *Remyella* lives in Pešter polje and the following surrounding massifs: Jadovnik (1734 m), Javor (1520 m), Giljeva (1617 m), Ninaja (1358 m), Žilindar (1616 m); *Rozajella* in karstic area located south from Pešter; *Nonveilleriella* in massifs located on the northern border of the Kosovo plain (*Nonveilleriella*) (Fig. 39). They can be distinguished with the following identification key:

1 – Elytral punctuation not transversally aligned or only sporadically so, without transversal strigae. Posterior angles of the pronotum not explanate. Profemora with a dense coating of tiny setae on the ventral surface (Fig. 4). Genital segment not reduced

to a narrow ring encircling the aedeagus, clearly separated in one dorsal and one ventral part (Figs. 14; 15). Anterior border of the female abdominal ventrite VIII angulate, the apophysis located at the top of the angle (Fig. 26)..*Remyella* Jeannel
– Elytral punctuation transversally aligned and with transverse strigae (Fig. 6), at least in a significant part of the elytral surface. Posterior angles of the pronotum shortly explanate (Fig. 7). Profemora with only sparse, tiny setae on the ventral surface (Fig. 5). Genital segment reduced to a narrow ring around the aedeagus as in most Leptodirini. Anterior border of the female abdominal ventrite VIII approximately straight (Fig. 27)...2

2 – Antennae very long relative to the body length, much longer than the body..*Nonveilleriella* Perreau & Pavićević
– Antennae short relative to the body length, shorter than or as long as the body (Figs. 2; 3) ....*Rozajella* S. Ćurčić & B. Ćurčić

5.3.2. Phylogeny of the genus *Remyella*

The genus *Remyella* was described by Jeannel in 1931 with the single species R. *scaphoides*. Two additional taxa were subsequently introduced by Winkler (1933): *R. propiformis* and *R. borensis.* The genus was reinvestigated by Jeannel (1934), adding two taxa: *R. hussoni* and *R. puncticollis*, and later revised by Giachino & Vailati (1995) with one new taxon, *R. droveniki*, and the synonymy of *R. puncticollis* with *R. borensis*. Three new taxa were subsequently described by S. Ćurčić et al. (2008; 2013). Four species are presently recognized: *R. javorensis*, *R. montenegrina*, *R. raskae*, and *R. scaphoides*, with the latter one comprising five subspecies: *R. s. borensis*, *R. s. droveniki*, *R. s. hussoni*, *R. s. propiformis*, and *R. s. scaphoides*.

Figs. 2–7. 2: *Rozajella madzgalji* n. sp. holotype, habitus. 3: *R. deelemani* male, habitus. 4: *Remyella javorensis*, profemur. 5: *Rozajella madzgalji* n. sp., profemur. 6: *Nonveilleriella ognjenovici*. explanate posterior angle of pronotum. 7: *Rozajella madzgalji* n. sp., elytral punctuation.

The distribution area of the genus *Remyella* is located on the borders of the Pešter polje (Fig. 39). It is divided into three main areas significantly remote from each other and corresponding to the three disconnected karstic areas (Fig. 39):

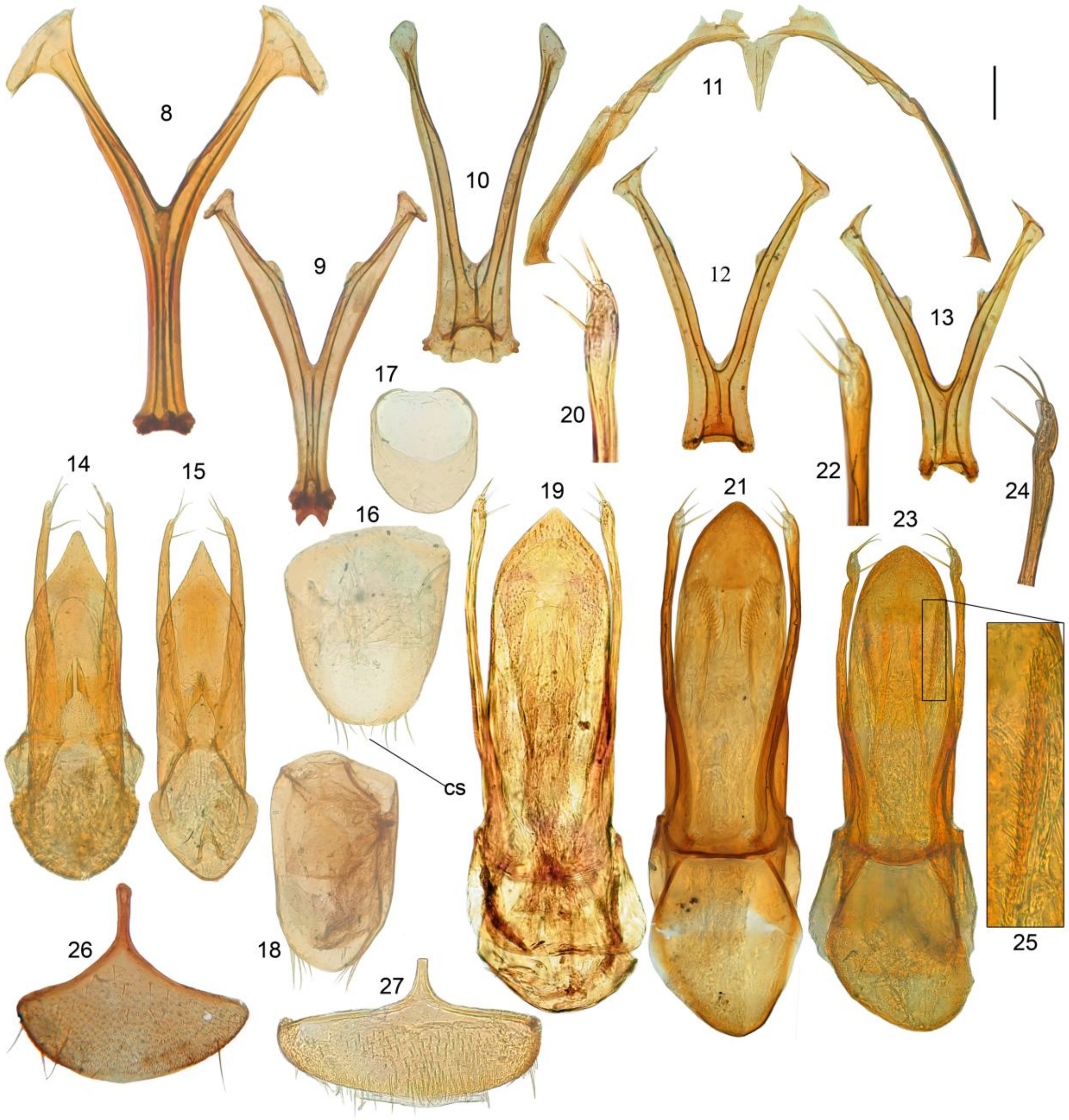
– In the surroundings of Sjenica, on the north-western side of Pešter polje, two caves hosting *R. javorensis*: Ušačka pećina (= Ušački pećinski sistem) in Gornje Lopiže and Baždarska pećina (the type locality) near Ursule.

– In the surroundings of Novi Pazar, on the north-eastern side of Pešter polje, Pećina u vrelu Raške (the springs of the Raška river), the type locality and only known locality of *R. raskae*.

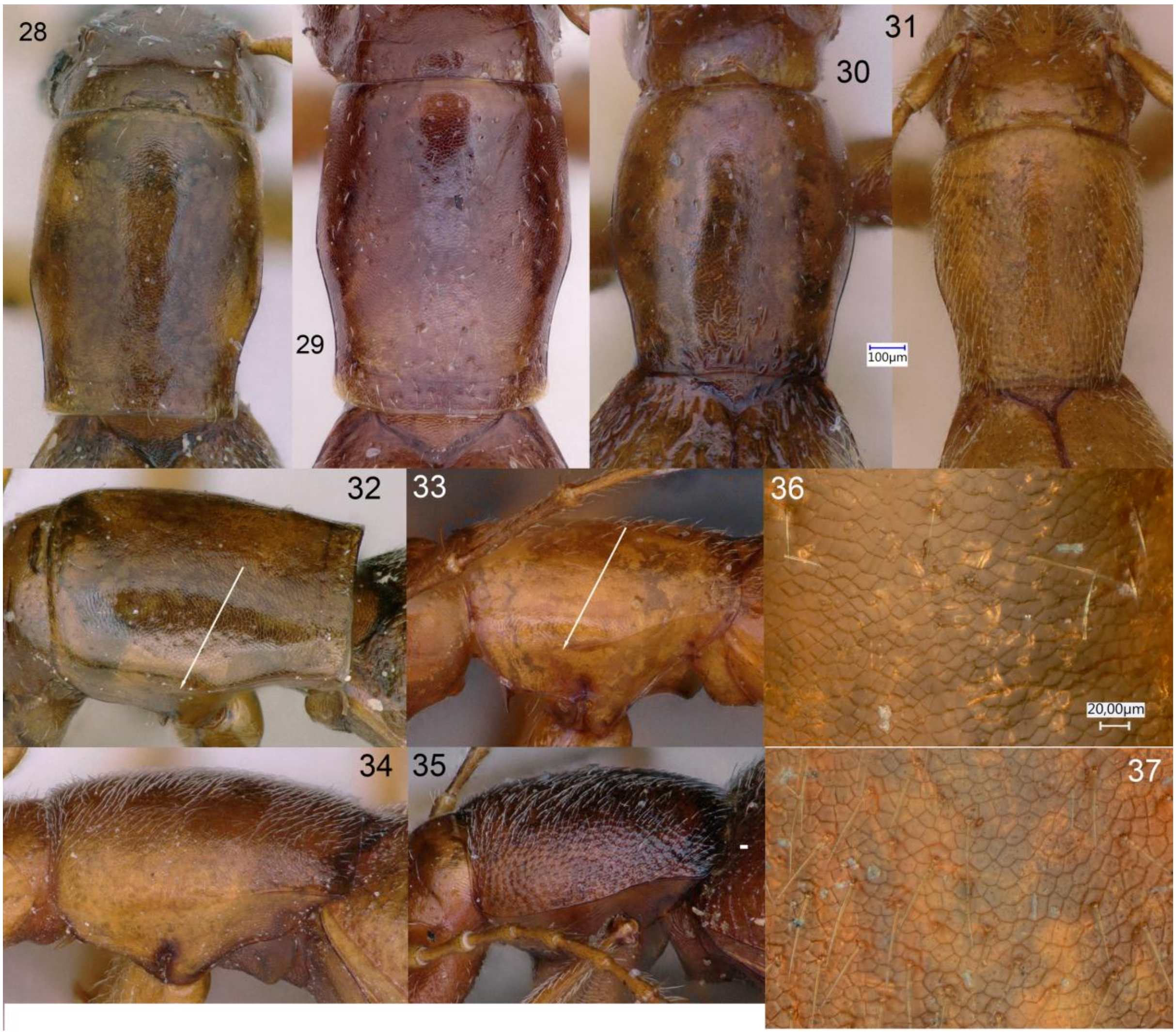
– The third area is a zone of 25 km long and 10 km wide, oriented NW-SE on the southern border of the Pešter polje, along the frontier between Serbia and Montenegro.

In this area, 20 caves host populations assigned presently to six taxa: *R. borensis*, *R. droveniki*, *R. montenegrina*, *R. propiformis*, *R. puncticollis*, and *R. scaphoides*. Velika pećina near Grgaje, the type locality of *R. scaphoides*, is the westernmost population of this area. Then, from west to east we find the northernmost population in Lopužino brdo on Giljeva Mountain. Continuing along the Serbian side of the border, three caves are located in the vicinity of Kapeš, including Uleva pećina III, the type locality of *R. droveniki*, and three in the vicinity of Boljare and Doliće. After a gap of 3.5 km, three caves are located in the vicinity of Ugao, including Špela Hajnet (= Špela Hajnit, = Špela Maja Hajnet), the type locality of *R. scaphoides propiformis* and Špela Bores, the type locality of *R. scaphoides borensis*. After another gap of 4.5 km, four caves are located in the vicinity of Đerekare, including Pećina u Hamidovoj vrtači, the type locality of *R. scaphoides hussoni* and Đerekarsko vrelo: the spring of the Boroštica river (= Đerekarska river (Nešić, 2015)). Four other caves are located on the Montenegrian side of the border: two near Đalovići, including Đjalovića pećina, the type locality of *R. montenegrina*, and two in the vicinity of Korita, including Ledenica pećina, the type locality of *R. puncticollis*. The latter two caves are the southernmost localities of the genus *Remyella*.

Except for the two geographically clearly remote (Fig. 39) and morphologically distinct species, *R. javorensis* and *R. raskae*, the populations of other taxa, especially the subspecies of *R. scaphoides*, but also *R. montenegrina,* are geographically and morphologically very close together. They are distributed in caves only a few kilometres apart from each other, on a single karstic plateau without any obvious geological barriers able to induce such a high level of endemism. Moreover, the distributions of some subspecies are overlapping. For example, Giachino and Etonti (1995) reported two subspecies located in the village Ugao: *R. scaphoides borensis* from Špela Hajnit and *R. scaphoides propiformis* from Špela Maja Hajnet. During our field work, local people from this village explained to one of the authors (D. Pavićević) that these are different names for the same cave: Špela Hanjet, meaning that two subspecies are reported from a single cave. Even though this situation clearly comes from a confusion in the denomination of the cave, it casts a serious doubt on the validity of the morphological criteria used to distinguish these subspecies.



Figs. 8–27. Internal characters. 8: *Anthroherpon primitivum*, metendosternite. 9: *Parapropus sericeus*, metendosternite. 10: *Remyella javorensis*, metendosternite. 11: *Remyella javorensis*, metatergum. 12: *Rozajella deelemani*, metendosternite. 13: *R. madzgalji* n. sp., metendosternite. 14: *Remyella scaphoides*, aedeagus dorsal view. 15: *R. propiformis* (from Špela Bores), aedeagus dorsal view. 16: *R. propiformis* (from Špela Bores), male genital segment, dorsal part (cs=central setae). 17: *R. propiformis* (from Špela Bores), male genital segment, ventral part. 18: *R. raskae*, male genital segment, dorsal part. 19: *Rozajella jovanvladimiri*, aedeagus dorsal view. 20: *R. jovanvladimiri*, apex of paramere. 21: *R. deelemani*, aedeagus dorsal view. 22: *R. deelemani*, apex of paramere. 23: *R. madzgalji*, aedeagus dorsal view. 24: *R. madzgalji*, apex of paramere. 25: *R. madzgalji*, longitudinal row of spines of the endophallus. 26: *R. deelemani*, abdominal ventrite VIII. 27: *Remyella raskae*, abdominal ventrite VIII.



Figs. 28–37. Pronotum. 28–31: Dorsal view. 32–35: Lateral view (the arrow shows the maximal extension of the lateral rim). 36-37: Detail of the dorsal punctuation. 28: *Remyella raskae*. 29, 32: *R. javorensis*. 30, 36: *R. scaphoides*. 31: *R. hussoni* (from Ledenica pećina. 34: *Rozajella madzgalji*. 35: *R. jovanvladimiri.* 37: *R. hussoni* (from Pećina u Hamidovoj vrtači).

Morphological characters originally used to distinguish *Remyella* species (Jeannel, 1931; 1934; Winkler, 1933) have been further discussed by Giachino and Etonti (1995). The measurements given by the latter authors show that the differences in the length/width ratios of the head, pronotum, and elytra between taxa are small and do not exceed intrapopulational variations. Giachino and Etonti based their taxonomical divisions on a single character, i.e. the relative location and relative distances of the three apical setae of the parameres. But looking at their illustrations (Giachino & Etonti 1995: 85), it is also clear that these differences do not exceed usual intrapopulational variability.

To clarify this intricate situation, we performed a molecular genetic analysis and reinvestigated the morphology. The populations of *Remyella* are listed in Table 3, including already known localities, as well as newly explored caves or pits. Table 3 also gives taxonomic names used by previous authors and the taxonomic assignations as resulting from our present work.

The results of the molecular phylogenetic analysis are shown in Fig. 1, and the genetic distances are given in Fig. 38. The populations are initially identified by their locality rather than by taxonomic names, as their final taxonomic assignation will be contingent on the results of the combined phylogenetic analysis and morphological characterization.

Our phylogenetic tree first confirms the monophyly of the genus *Remyella* and reveals the following nesting of populations. For simplicity, we start with the terminal branches containing the populations near Doliće (jama Bez Dna) and those near Đalovići (Jagoševa pećina). These form the sister group of the populations near Boljare (genetic distances 0.6 % to 1.5 % from the population of Jagoševa pećina), and subsequently these three populations are (although with very low support) sister to the population from Lopužino brdo (genetic distance 2.9 % from population of Doliće). The sister clade of the preceding group are the two populations of Sijera jama and Jama u Piskovoj livadi near Đerekare (genetic distance of 3.6 % to 4.4 % from populations of Jagoševa, Doliće and Boljare. More basal in the tree we find the separation of the populations from Baždarska pećina and Ušačka pećina, genetically divergent by 7.3% but morphologically identical (see below). The earliest separation within *Remyella* leads to the population from the spring of Boroštica (Đerekarsko vrelo), which shows a genetic distance of more than 9% from the other populations located also near Đerekare, despite their geographical proximity. This population of Đerekarske vrelo is therefore enigmatic, and we exclude it presently from this study awaiting new material for further investigations.

We have no available specimens for molecular analysis of the following populations: the single known locality of *R. scaphoides* (Velika pećina); the single known locality of *R. raskae*; populations located near Ugao (Špela Hajnet, Špela Bores, Špela Vogel, and the Sinkhole of Boroštica), of Pećina u Hamidovoj vrtači (the type locality of *R. hussoni*); and populations of Ledenica pećina and Pećina u vrh Livade Radojeve (both being assigned to *R. puncticollis* by Jeannel).

R. raskae and *R. scaphoides* both known from a single and geographically isolated location are also morphologically well characterized. Populations of Ušačka pećina and Baždarska pećina correponding to *R. javorensis* are morphologically similar and also geographically isolated. The specific status of the three preceding populations is undisputable. Based on the morphological similarity and biogeographic coherence, the specific assignation of the other populations is done in the following way: Populations near Ugao are not morphologically distinct from the populations located near Doliće, Boljare, Đalovići and Lopužino, so we consider them as conspecific, the senior names for them are *R. propiformis* or *R. borensis* (described in the same paper of Winkler, 1933), and we choose *R. propiformis*. The population from the surroundings of Đerekare, Pećina u Hamidovoj vrtači, Ledenica pećina, and Pećina u vrh Livade Radojeve are morphologically similar, we consider them as conspecific, the senior names are *R. hussoni* and *R. puncticollis* (described in the same paper of Jeannel, 1934), and we choose *R. hussoni*.

We should notice that an uncertainty remains regarding the exact type locality of *R. puncticollis*, namely Ledenica pećina. A cave with the same name is cited in Nešić (2015), located close, but slightly to the east of the location given by Giachino & Etonti (1995). However, since the location of this cave in Giachino & Etonti (1995) is given approximately (Giachino, pers. comm.), we assume that it is probably the same.

The taxonomical consequences of the preceding analysis are given in the following section.

5.3.3. Morphological characterization and taxonomical status of *Remyella* species

Diagnosis: genus with a scaphoid body shape, length: 3.8-4.9 mm (males) 4.0-5.4 mm (females). Anophthalmic, depigmented. All external parts of the body microreticulated. Antennae sexually dimorphic, approximately 1.35 times as long as the body in males, 1.1 times as long as the body in females. Punctuation of the dorsal parts always associated with setae. Punctuation of pronotum distributed in variable patterns according to species, but generally always present and/or denser along the posterior margin. Punctuation of the elytra always dense and with few specific variations, sometimes with sporadic traces of transversal alignments, but without transversal striae. Sutural stria absent. Apex of the elytra widely rounded and widely separated in males, shortly rounded and more closely joined in females (but yet with a small gap). Mesoventral process developed but not fused with the metasternum. Metatergum with a short posterior expansion (Fig. 11). Metendosternite "V"-shaped (Fig. 10). Profemur with a dense coating of tiny setae (Fig. 4). Protibiae without a lateral external row of spines. Protarsi five-segmented in males, four-segmented in females, not dilated in both sexes. Female genital segment with distinct appendicular parts, spermatheca weakly sclerotized. Male genital segment much more developed than generally in other Leptodirini (Figs. 16-18), with setae either distributed along the entire width of the posterior edge, or restricted to the sides, according to species. Aedeagus with a median lobe parallel from base to two third of its length, then terminated in equilateral triangle either with a sharp or blunt apex according to species. Parameres with two apical and one subapical setae.

The morphological characters which appear to be relevant to distinguish the five species are the following: body size; length of the lateral pronotal marginal rim (Figs. 32, 33); shape of lateral sides of the pronotum (Figs. 28-31); density of punctuation of the dorsal pronotum surface (Figs. 36; 37); shape of the apex of the median lobe of the aedeagus; setation of the apical edge of the male genital segment.

Using these morphological characters we recognize five relevant species (without any subspecies) in agreement with the phylogenetic analysis of the previous section and coherent with the geographical distribution (Fig. 39). They are listed below, and in Table 3, with the appropriate synonymies. We give a short diagnosis, the list of caves where the species were recorded, and the numbers of specimens examined in addition to those used for DNA extraction. An identification key is also given.

Remyella javorensis S. Ćurčić & B. Ćurčić, 1995
Remyella javorensis S. Ćurčić & B. Ćurčić, 1995: 112.
loc. typ.: Baždarska pećina Cave, village of Ursule, Mt. Javor near Sjenica, Southwest Serbia

Diagnosis: Large size, males 4.6-4.8 mm; females: 4.5-5.2 mm. The largest species of the genus. Pronotum long: 1.25 times as long as wide, the maximum width at the middle, the anterior half linearly convergent from the middle to the anterior edge, abruptly narrowed at the middle, then parallel on the posterior half (Fig. 29). Lateral marginal rim of the pronotum limited to the basal half of the pronotum (Fig. 32). Punctuation sparse, denser along the posterior edge (Fig. 29). Dorsal part of the male genital segment with lateral and central setae. Apex of the median lobe of the aedeagus sharp, and slightly bent upward.

Distribution: known only from two caves: Baždarska pećina near Ursule (type locality) [5 m, 13 f] and Ušačka pećina near Ušak (new locality) [1 m].

Remyella raskae S. Ćurčić & B. Ćurčić, 2008
Remyella raskae S. Ćurčić & B. Ćurčić, 2008: 110.
loc. typ.: Pećina u Vrelu Raške cave near Novi Pazar, Pešter Polje, Southwest Serbia

Diagnosis: length: males: 4.6-4.8 mm; females: 4.5-5.2 mm. Pronotum long: 1.25 times as longer as wide, widest at the posterior third, the anterior two-thirds linearly convergent towards the anterior edge, abruptly narrowed at the basal third, then parallel towards the hind angles (Fig. 28). Lateral marginal rim of the pronotum limited to the basal half of the pronotum. Punctuation sparse, denser along the posterior edge (Fig. 24). Dorsal part of the male genital segment with lateral setae only, central setae absent (Fig. 18). Apex of the median lobe of the aedeagus sharp.

Distribution: known only from the type locality, the spring of river Raška (Pećina u Vrelu Raške) near Novi Pazar, Serbia [3 m, 6 f].

Remyella scaphoides Jeannel, 1931
Remyella scaphoides Jeannel, 1931: 263.
Remyella scaphoides scaphoides Jeannel, 1931: Giachino & Etonti, 1995: 88.
loc. typ.: Yougoslavie: Velika pećina près du hameau de Grgaje, opstina Bare

Diagnosis: length: males: 4.1-4.5 mm; females 4.5-4.75 mm. Pronotum 1.15 times as long as wide, with the maximum width approximately in the middle. Pronotal lateral sides convex in the anterior half, convergent in the posterior half until the base, or shortly parallel just before the hind angles. Lateral rim of the pronotum extending from the base to 2/3 or ¾ of the pronotum. Pronotal punctuation sparse (Fig. 30). Apex of the aedeagus blunt (Fig. 14).

Distribution: known only from the type locality: Serbia, Velika pećina near Grgaje [9m, 8f].

Remyella propiformis Winkler, 1933
Remyella propiformis propiformis Winkler, 1933: 78.
loc. typ.: Spela Maja Hejne, nordöstlich von Ugo im Sandjak Novipazar
Remyella scaphoides propiformis Winkler, 1933: Giachino & Vailati, 1995: 88.
Remyella propiformis borensis Winkler, 1933 **n. syn.**
loc. typ.: Spela Bor, nordwestlich von Ugo im Sandjak Novipazar
Remyella scaphoides borensis Winkler, 1933: Giachino & Vailati, 1995: 90. (partim)
Remyella scaphoides droveniki Giachino & Etonti, 1995: 91 **n. syn.**
loc. typ.: Serbia occ.: Dolić, Pešter, Kapeš, Uleva pećina III
Remyella montenegrina S. Ćurčić, Antić, N. Ćurčić & B. Ćurčić, 2013: 1218 **n. syn.**
loc. typ.: Đalovića Pećina Cave (= Pećina nad Vražjim Firovima Cave), Đalovića Gorge, village of Đalovići near Bijelo Polje, Pešter Polje, northeastern Montenegro

Diagnosis: length males: 3.8-4.6 mm, females: 4.0-4.8 mm. Pronotum 1.2 times as long as wide, with the maximum width approximately in the middle. Pronotal lateral sides convex in the anterior half, convergent in the posterior half until the base, or shortly parallel just before the hind angles. Lateral rim of the pronotum extending from the base to 2/3 or ¾ of the pronotum length. Pronotal punctuation sparse. Apex of aedeagus sharp (Fig. 15).

Distribution: Serbia: caves located in the surroundings of Ugao: Špela Hajne [21 m, 9 f], Špela Vogel [1 m, 5 f], sinkhole of Boroštica river (Giachino & Etonti, 1995), Špela Bores [14 m, 16 f]. Caves on Giljeva: Lopužino brdo. Caves in the surroundings of Doliće, Boljare and Kapeš: pećina Uleva III [13 m, 11 f], Pećina u Anin Kapeš [13 m, 15 f], Pećina Ispad Gluare [13 m, 14 f], Jama Bez Dna, Pećina kod Jagodina Dola [4 m, 10 f], Kaćunova Jama [1 m, 4 f]. Montenegro: caves located in the surroundings of Đalovići: Jagoševa pećina, Đjalovića pećina (=Pećina nad Vražjim Firovima) [12 m, 22 f].

Remyella hussoni Jeannel, 1934

Remyella hussoni Jeannel, 1934: 103

loc. typ.: U Hamidova vrtaca pećina à Krnja Jela, Sandjak de Novi-Pazar

Remyella scaphoides hussoni Jeannel, 1934: Giachino & Vailati, 1995: 91.

Remyella puncticollis Jeannel, 1934: 103. **n. syn.**

loc. typ.: Sandjak de Novi Pazar, Ledenica pećina, opština de Korito, srez de Bijelopolje

Remyella scaphoides borensis Winkler, 1933: Giachino & Vailati, 1995: 90 (partim)

Diagnosis: length males: 3.8-4.4 mm, females: 4.2-5 mm. Pronotum 1.1 times as long as wide, with the maximum width approximately in the middle. Pronotal lateral sides arcuate in the anterior half, convergent in the posterior half until the base, or shortly parallel just before the hind angles. Lateral rim of the pronotum extending from the base to 2/3 or ¾ of the pronotum. Pronotal punctuation dense and more or less uniformly distributed. Apex of the aedeagus sharp (Fig. 15).

Distribution: Serbia: cave in Hamidova vrtača (type locality) [2 m, 3 f]; caves in the surroundings of Đerekare, on the left side of the Boroštica river: Sijera jama, Jama u Piskovoj livadi, Ponor Ledenice. Montenegro: caves located on the Montenegrian side of the frontier: Ledenica pećina [1 m, 3 f], Pećina u vrh Livade Radojeve (Jeannel, 1934).

The five species can be identified with the following identification key:

1 – Lateral sides of pronotum convergent in a straight line between the maximum width and the anterior edge (Figs. 28; 29). Lateral marginal rim of the pronotum extending from the posterior edge to barely more than a half of the pronotum length (Fig. 32)...2

– Lateral sides of the pronotum convex in the anterior part (Figs. 30; 31). Lateral marginal rim of the pronotum extending from the posterior edge to 2/3 to ¾ of the pronotum length (Fig. 33)..3

2 – Larger body size: 4.6-4.9 mm (males); 4.5-5.2 mm (females). Pronotum widest at the middle. (Fig. 29). Male genital segment with lateral and central setae (Fig. 16)..*javorensis* S. Ćurčić & B. Ćurčić

– Smaller body size: 4.1-4.4 mm (males); 4.1-4.5 mm (females). Pronotum widest at the basal third (Fig. 28). Male genital segment only with lateral setae (Fig. 18)..*raskae* S. Ćurčić & B. Ćurčić

3 – Pronotal punctuation dense and more or less uniformly distributed (Fig. 37)..*hussoni* Jeannel

– Pronotal punctuation very sparse and sporadic, generally denser along the base, and sometimes along the median line of the dorsal surface (Fig. 36)...4

4 – Median lobe of the aedeagus wide, and with a blunt apex (Fig. 14)…………………………………………………………………….……………………......*scaphoides* Jeannel

– Median lobe of the aedeagus narrow, and with a sharp apex (Fig. 15) ……………………………………………………..…………………………………......*propiformis* Winkler

5.3.4. Biogeography of *Remyella*

The table of Fig. 38 contains above the diagonal the COI genetic distances, and below the diagonal their geographical distances. Using a Mantel test, we find a high correlation of 0.86 between geographic and genetic distances (Fig. 38). The single pair of populations that do not conform are the two populations of *R. javorensis* from Baždarska pećina and Ušačka pećina, geographically distant by less than 4 km, but with a genetical distance of 7.3 %. Actually, these two populations are separated by the deep valley of the Vapa river which may be a strong barrier to gene flow and could explain this high genetic divergence. However, we find no significant morphological differences between these two populations and we consider them as conspecific.

According to the genetic divergences given in Fig. 38, which range from 3.5% to 10%, and using as a preliminary approach the divergence rate of 2% per million years obtained by Ribera et al. (2010) for mitochondrial genes of the western Mediterranean Leptodirini, the speciation events in *Remyella* occurred approximately from 5 to 2 million years ago. This corresponds to the end of the Miocene and the Pliocene, periods when the Pešter field was periodically filled by shallow lakes forming sedimentary deposits (Mojsilović et al., 1973). The distribution area of the genus, confined to the periphery of Pešter (Fig. 39), is probably contingent to the presence of these lakes, preventing species to spread more uniformly over the entire area of the field.

Correlation between genetic and geographic distances: 0.86

Genetic distance (%) Geographic distance (km)	Jagoševa pećina	Jama bez dna	Kaćunova pećina	Lopužina jama	Ponor Ledenice	Sjera jama	Ušačka pećina	Baždarska pećina
Jagoševa pećina	0	0,6	1,5	3,5	3,9	4,2	9,15	8,35
Jama bez dna	9,24	0	1,2	2,9	3,6	3,9	8,65	8,45
Kaćunova pećina	6,55	2,67	0	3,8	4,25	4,4	8,8	8,575
Lopužina jama	8,95	8,45	7,2	0	5,3	5,6	9,75	8,8
Ponor Ledenice	18,25	11,87	11,55	20,2	0	0,3	9,95	9,1
Sjera jama	8,95	8,45	7,2	20,2	1,47	0	9,45	9,15
Ušačka pećina	29,51	30,55	29,6	22,35	42,45	41	0	7,3
Baždarska pećina	32,83	31,9	32,8	25,38	45,1	43,62	3,65	0

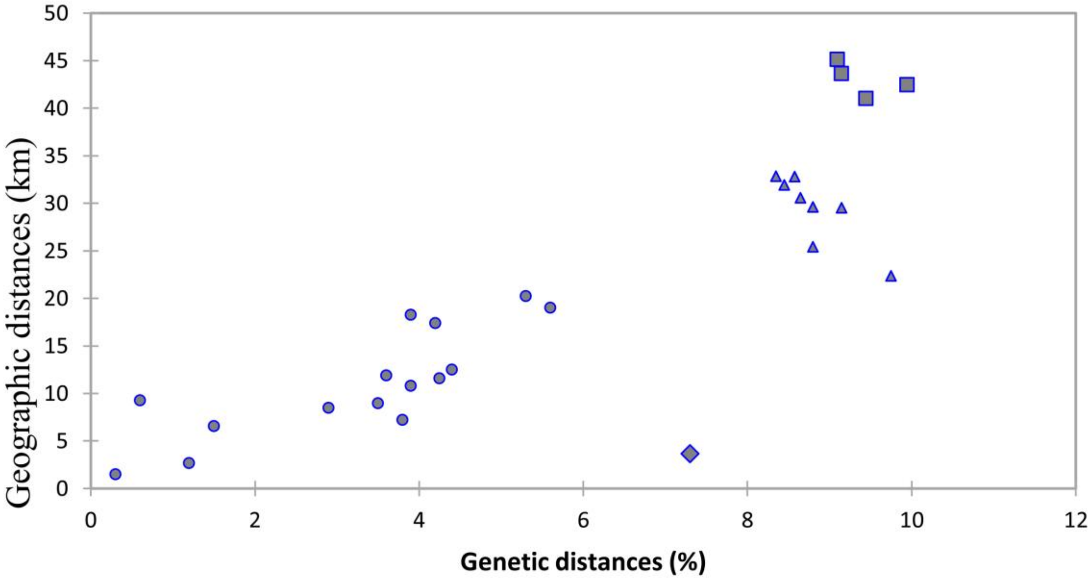


Fig. 38. Table of the genetic (barcoding region) and geographic distances with the plot of correlations resulting from the Mantel test. Squares correspond to pairs of populations *R. javorensis* – *R. hussoni*; Triangles correspond to pairs of populations *R. javorensis* – *R. propiformis*; the diamond corresponds to the two known populations of *R. javorensis*; circles are other pairs of populations.

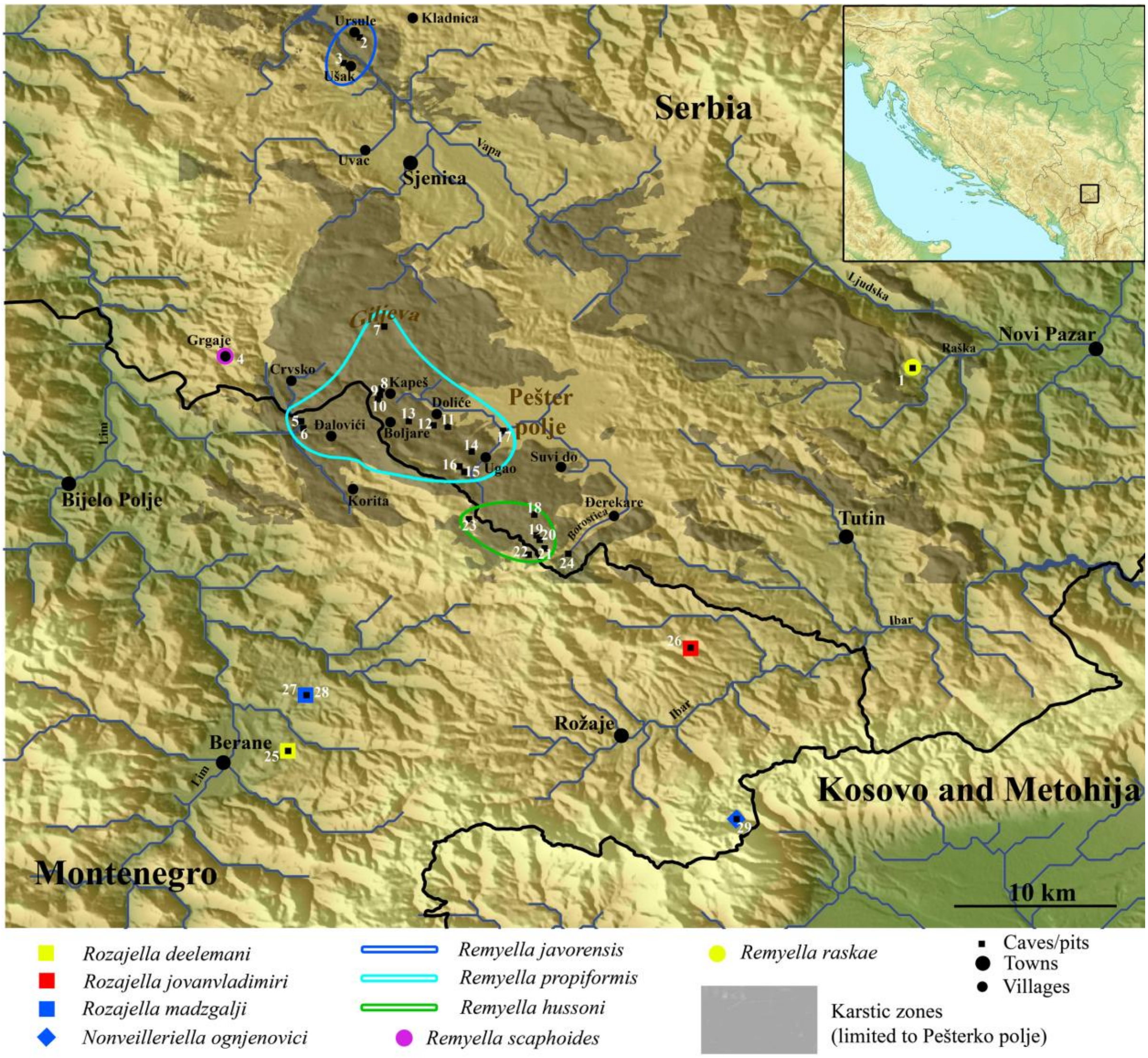


Fig. 39. Distribution map of *Remyella* and *Rozajella*. Shaded areas are karstic zones of the Pešter polje (after Mojsilović & al., 1973).

5.3.5. The genus *Rozajella*

The genus *Rozajella* S. Ćurčić, Brajković & B. Ćurčić, 2007 comprises two obligate cave species: *R. jovanwladimiri* S. Curčić, Brajković & B. Curčić, 2007 and *R. deelemani* Perreau & Pavićević, 2008, distributed in the eastern range of the Dinaric Mountains in Montenegro. Both species are short-range endemics, known only from a single cave, one near Rozaje and one near Berane in Montenegro. A third species, *Rozajella madzgalji* n. sp., is described below and the male of *R. deelemani* is described for the first time.

Rozajella madzgalji n. sp.

Type material: Holotype male labeled as follows: “MONTENEGRO, Berane, Goražde, Mala pećina u Garaškom kršu”, 860 m, 19.07.2014, leg. I. Njunjić” (white label, printed)/“HOLOTYPUS *Rozajella madzgalji* sp.n. I. Njunjić, M. Perreau & D. Pavićević det. 2016.“ (red label, printed), (MNHN). Paratypes: 4♂ 6♀, 19.07.2014. (MNHN, CINJ, CMPR, CDPV), 5♂ and 12♀, same cave, 22.11.2014, leg. I. Njunjić (MNHN, CINJ, CMPR, CDPV); 2♂ 4♀ Montenegro, Berane, Goražde, Velika pećina u Garaškom kršu, 860 m, 22.11.2014, leg. I. Njunjić (MNHN, CINJ, CMPR, CDPV). All paratypes are labeled with white, printed locality labels and with red printed labels “PARATYPUS *Rozajella madzgalji* sp.n. I. Njunjić, M. Perreau & D. Pavićević det. 2016”.

Type locality: Mala pećina u Garaškom kršu, Goražde, Berane, Montenegro; N 42° 52′ 15.7″ E 19° 55′ 10.5″.

Diagnosis. Habitus illustrated in Fig. 2. Distinct from the other species of the genus by the following combination of characters: medium size of the body, shape of pronotum, parameres with constriction in the apical part, sharp and pointed apex of the parameres.

Description

Body length 3.29 to 4.32 mm (HT 4.09 mm). General morphology leptodiroid, eyeless, wingless, and depigmented.

Head narrower than pronotum. Dorsal surface rough with dense, erected hairs. Occiput with deep, random punctures. The base of the terminal maxillary palpomere with a dorsal comb of very short hairs. Antennae inserted slightly after the middle of the length of the head (measured from the posterior margin of clypeus) and shorter than the body in both sexes. Second antennomere slightly longer than the first one; 8th antennomere is the shortest; last antennomere longer than the penultimate in both sexes, but slightly more so in males. All antennomeres covered with hairs of medium length, but only antennomeres 5-11 bear up to 8 long, erected setae arranged circularly and symmetrically in the apical part. Hairs of medium length are particularly dense on the two last antennomeres.

Pronotum approximately 1.31 times as long as wide (HT 1.32). The maximal width approximately at the middle of the length. Lateral margins sinuous towards the head and towards the posterior angles, with a lateral rim interrupted in the middle (Fig. 33). Posterior lateral angles acute or right. Posterior edge with a marginal carina (Fig. 34), anterior edge with a thin marginal carina. Dorsal surface rough with dense, short hairs (Fig. 34). Ventral surface with a microsculptural mesh resembling a honeycomb pattern, hairless, except a few sparse hairs on the ventrolateral side. Metendosternite in "V"-shape (Fig. 12).

Abdomen. Ventral surface with microsculptural mesh resembling a honeycomb pattern. Mesosternum and mesepisternum hairless, other ventrites covered with dense hairs.

Elytra elliptic, approximately 1.6 times as long as wide (HT 1.75), without sutural striae (Fig. 6). Dorsal surface with punctures and dense hairs transversally aligned and transversal strigae on the anterior part of the elytra (Fig. 6).

Legs. Long and slender, protibia weakly curved inwards, profemora thicker than meso- and metafemora.

Aedeagus. Median lobe thick, slightly curved ventrally in lateral view and with rounded apex in dorsal view (Fig. 23). Parameres slightly shorter than the median lobe, with a strong constriction in the apical part, bearing three setae (Fig. 24). Apex of the parameres pointed and concave on the interior side. Endophallus with armature in two lateral fields with several dozens of short and thin teeth (Fig. 25). For comparison, we figure the aedeagus and apex of parameres of *R. jovanvladimiri* (Figs. 19, 20) and *R. deelemani* (Figs. 21, 22).

Female genitalia. Spermatheca unsclerotised. Abdominal ventrite VIII with a short and narrow anterior expansion. Urite IX with appendicular parts. Gonostyles with 3 setae, one apical and 2 lateral.

TABLE 3. Antennal formula of the holotype of *Rozajella madzgalji* sp. n. (mm).

Article	I	II	III	IV	V	VI	VII	VIII	IX	X	XI
Length	0.23	0.25	0.27	0.29	0.29	0.34	0.35	0.21	0.35	0.32	0.42

Distribution

R. madzgalji n. sp. is known from two caves: Mala pećina u Garaškom kršu (210 m long) and Velika pećina u Garaškom kršu (188 m long), located only 150 meters from each other, close to the village Zagrađe near Berane in Montenegro (Fig. 39). During the second visit to Mala pećina u Garaškom kršu, a large number of individuals was found feeding on a pigeon wing in the dark zone of the cave, approximately 100 m from the entrance.

Etymology

The new species is named after speleologist Zeljko Madžgalj from Bijelo Polje (Montenegro), in recognition for his contribution to speleological investigations of Montenegro and enthusiastic help in the field.

Rozajella deelemani Perreau & Pavićević, 2008 - description of a male

Material examined: 1♀: Holotype, Crna Gora, Građa pećina, Petnik, Ivangrad (= Berane) 5.8.1970. leg. Deeleman (CMPR); 1♀: Paratype, same data (CDPV); 3♂: Crna Gora, Berane, Petnik, Građa pećina, 22.11.2014. leg. I. Njunjić (CINJ); 4♀: same data (CINJ).

Type locality: Crna Gora, Berane, Petnik, Građa pećina; N 42° 50' 06.7'' E 19° 54' 26.1'' (Fig. 39).

Description

Habitus illustrated in Fig. 3. Body length 4.35 mm. General morphology leptodiroid, eyeless, wingless, and depigmented.

Head narrower than pronotum. Dorsal surface rough with dense, erected hairs. Penultimate maxillary palpomere short and thick. Terminal maxillary palpomere thin and with a dorsal comb of very short hairs at the base. Antennae inserted slightly after the middle of the length of the head (measured from the posterior margin of clypeus) and shorter than the body. Second antennomere almost the same length as the first one; 8th antennomere is the shortest; last antennomere is the longest and it is longer than the penultimate. Hairs on the antennae like in other species of the genus, as described above in *R. madzgalji*.

Pronotum 1.34 times as long as wide. The maximal width approximately at the middle of the length. Lateral margins sinuous towards the head and slightly sinuous towards the posterior angles. Posterior lateral angles right. Posterior edge with a thick marginal carina, anterior edge with a thin marginal carina. Dorsal surface rough with short hairs, which are very rare on the dorso-lateral side. Metendosternite "V"-shaped (Fig. 13).

Elytra elliptic, 1.85 times as long as wide, without sutural stria. Dorsal surface with punctures and short, dense hairs transversally aligned on the anterior part of the elytra.

Aedeagus. Median lobe thick, somewhat curved ventrally in lateral view and with lanceolate apex in dorsal view (Fig. 21). Parameres shorter than the median lobe and slightly wider in the apical part. Apex of the parameres pointed, bearing three setae (Fig. 22). Endophallus with armature in two lateral fields with several dozens of thick teeth (Fig. 21).

TABLE 4. Antennal formula of the male of *Rozajella deelemani* (mm).

Article	I	II	III	IV	V	VI	VII	VIII	IX	X	XI
Length	0.27	0.26	0.28	0.29	0.36	0.36	0.36	0.23	0.38	0.36	0,48

The following identification key of the genus *Rozajella* updates the key given by Perreau & Pavićević (2008a).

1 – Size smaller than 3.5 mm. Lateral margins of the pronotum with an uninterrupted carina (Fig. 35). Pronotum at most 1.2 times as long as wide. Elytral punctures with transversal strigae on the whole surface of the elytra……………………………………………………………………………….…………….....*jovanvladimiri*

– Size larger than 3.7 mm. Lateral margins of the pronotum with a carina interrupted at the middle (Fig. 34). Pronotum at least 1.3 times as long as wide. Elytral punctures with transversal strigae only on two third of the length of the elytra.. 2

2 – Median lobe with rounded apex in dorsal view. Parameres with a strong constriction in the apical part. Endophallus with armature in two lateral fields with several dozens of short and thin teeth (Fig. 25)……………………………………………..………….……………… *madgalji*

– Median lobe with lanceolate apex in dorsal view. Parameres slightly wider in the apical part. Endophallus with armature in two lateral fields with several dozens of thick teeth (Fig. 21).. *deelemani*

5.4. ACKNOWLEDGEMENTS

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5.5. ANNEX

Table 3: Populations and localities investigated. The number refers to the map of Fig. 39.

n°	Cave	Location	Coordinates (WGS84)	Altitude (m)	Taxonomy (from litterature)	Taxonomical status from this work
1	Pećina na Vrelu Raške	Novi Pazar	N43°06'57.03" E20°22'15.36"	725	*raskae* (S. Ćurčić et al., 2008) **TL**	*Remyella raskae*
2	Baždarska pećina	Ursule			*javorensis* (S. Ćurčić et al., 2008) **TL**	*R. javorensis*
3	Ušačka pećina	Ušak	N43°20'38.86" E19°57'17.74"		new locality	*R. javorensis*
4	Velika pećina	Grgaje			*scaphoides* (Jeannel, 1931) **TL** *scaphoides* (Jeannel, 1934) *scaphoides scaphoides* (Giachino & Vailati, 1995)	*R. scaphoides*
5	Jagoševa jama (=pecina?)	Crvsko	N43°04'52.9" E19°55'23.2"	1184	new locality	*R. propiformis*
6	Đjalovića pećina =Pećina nad Vražjim Firovima	Đjalovići	N43°04'35.2'' E19°55'21.9''	820	*montenegrina* (S. Ćurčić & al., 2013) **TL**	*R. propiformis*
7	Bezdan na Lopužinom brdu	Lopužino brdo	N43°08'40.4" E19°59'26.8"	1338	new locality	*R. propiformis*
8	Uleva pećina III	Kapeš			*scaphoides droveniki* (Giachino & Vailati, 1995) **TL**	*R. propiformis*
9	Pećina u Anin Kapeš	Kapeš			*scaphoides droveniki* (Giachino & Vailati, 1995)	*R. propiformis*
10	Pećina Ispad Gluare	Kapeš			*scaphoides droveniki* (Giachino & Vailati, 1995)	*R. propiformis*
11	Jama Bez Dna	Doliće	N43°04'39.5" E20°02'09.8"	1205	new locality	*R. propiformis*
12	Pećina kod Jagodina Dola	Doliće			new locality	*R. propiformis*
13	Kaćunova Jama	Boljare	N43°04'49.1" E20°00'11.7"	1237	new locality	*R. propiformis*

14	Špela Bores	Ugao	N43°03'54.0" E20°02'48.2"	1215	*propiformis borensis* (Winkler, 1933) **TL** *propiformis borensis* (Jeannel, 1934) *scaphoides borensis* (Giachino & Vailati, 1995)	*R. propiformis*
15	Špela Hajnet (= Špela Hajnit = (= Špela Maja Hajnet)	Ugao	N43°02'46.5" E20°02'14.0"	1345	*propiformis propiformis* (Winkler, 1933) **TL** *propiformis propiformis* (Jeannel, 1934) *scaphoides propiformis* Giachino & Vailati, 1995) *scaphoides borensis* (Giachino & Vailati, 1995) (pars)	*R. propiformis*
16	Špela Vogel (=Maja Vogelj)	Ugao	N43°02'36.7" E20°02'20.9"	1307	new locality	*R. propiformis*
17	Sinkhole of Boroštica	Ugao			*propiformis propiformis* (Jeannel, 1934) *scaphoides borensis* (Giachino & Vailati, 1995)	*R. propiformis*
18	Pećina u Hamidovoj vrtači	Suvi Do			*propiformis hussoni* (Jeannel, 1934) **TL** *scaphoides hussoni* (Giachino & Vailati, 1995)	*R. hussoni*
19	Sijera jama	Đerekare	N42°59'35.0" E20°06'00.0"	1525	new locality	*R. hussoni*
20	Jama u Piskovoj livadi	Đerekare				*R. hussoni*
21	Ponor Ledenice	Đerekare				*R. hussoni*
22	Ledenica pećina	Korito	N42°58'48.7" E20°05'55.0''	1518	*puncticollis* (Jeannel, 1934) **TL** *scaphoides borensis* (Giachino & Vailati, 1995)	*R. hussoni*
23	Pećina u vrh Livade Radojeve	Korito		1400	*puncticollis* (Jeannel, 1934) *scaphoides borensis* (Giachino & Vailati, 1995)	*R. hussoni*
24	Pećina na Đerekarskom vrelu	Đerekare	N42°58'51.0" E20°07'13.0"	1320	new locality	*Remyella* Enigmatic species
25	Građa Pećina	Petnik	N42°50'06.7'' E19°54'26.1''			*Rozajella deelemani*
26	Pećina u dubokom Potoku	Donje Biševo				*R. jovanvladimiri*
27	Mala pećina u Garaškom kršu	Zagrade	N42°52'15.7" E19°55'10.5"		new locality	*R. madzgalji*
28	Velika pećina u Garaškom kršu	Zagrade			new locality	*R. madzgalji*
29	Jama kod Stovarišta	Šoljani				*Nonveilleriella ognjenovici*

Table S1 : Sequenced specimens, with depository, locality, collectors and sequence accession numbers (will be provided).

Species	Voucher code	Locality	Source
Anthroherpon cylindricolle cylindricolle (Apfelbeck, 1889)	IE3	BiH, Pale, Rogatica, Golubovića pećina	I. Njunjić
Anthroherpon ganglbaueri ganglbaueri (Apfelbecki, 1894)	IE11	BiH, Nevesinje, Novakuša	I. Njunjić
Anthroherpon hoermanni hoermanni (Apfelbeck, 1889)	IB7	BiH, Kalinovik, Miljevina, Borija pećina	I. Njunjić
Anthroherpon matzenaueri matzenaueri (Apfelbeck, 1907)	IB13	MNE, Golija, Latično, Jama u Rudinskom dolu	I. Njunjić
Anthroherpon pygmaeum stricticolle (Jeannel, 1930)	IE25	BiH, Bjelašnica, Opančak, Ledenjača	I. Njunjić
Anthroherpon stenocephalum stenocephalum (Apfelbeck, 1910)	IE44	BiH, Olovo, Bijambare	I. Njunjić
Antrocharis querilhaci dispar Abeille de Perrin, 1878	MNCN-AI588	Ariège, Aigües-Juntes, Grotte Les Cloutets	A. Faille
Apholeuonus nudus sturanyi Apfelbeck, 1906	IAP3	BIH, Kalinovik, Borija, Borija pećina	I. Njunjić
Apholeuonus nudus sturanyi Apfelbeck, 1906	IAP4	BIH, Kalinovik, Borija, Borija pećina	I. Njunjić
Charonites sp. Apfelbeck, 1907	IO33	BIH, Pale, Omladinska pećina	I. Njunjić
Hadesia vasiceki Müller, 1911	MP05	BIH, Popovo polje, Vjeternica	M. Perreau
Hadesia vasiceki Müller, 1911	IH3	BIH, Popovo polje, Vjeternica	M. Perreau
Hadesia weiratheri Zariquiey, 1927	IH2	Crna Gora, Orjen, Kameno more, PT4	M. Perreau
Leptomeson bujasi Giachino, Bregović, Jalžić, 2011	IL6	Croatia, Brač, Jama kod Matešića stana	B. Jalžić & P. Kutleša
Leptomeson radjai Giachino, Bregović, Jalžić, 2011	IL5	Croatia, Šibenik, Perković, Zvekača	P. Bregović
Parapropus sericeus muelleri Jeannel, 1924	IE35	BIH, Sanski most, Hrustovačka pećina	I. Njunjić
Parapropus sericeus muelleri Jeannel, 1924	IO26	BIH, Sanski most, Hrustovačka pećina	I. Njunjić
Parapropus pfeiferi Apfelbeck, 1908	IE34	BiH, Sanski most, Donji Kamičak, pećina u Donjem kamičku	I. Njunjić
Remyella hussoni Jeannel, 1934	IR6	Serbia, Pešter, Đerekare, Piskova livada	P. Lazarević
Remyella hussoni Jeannel, 1934	IR19	Serbia, Pešter, Đerekare, Sjera livada, Ponor Ledenice	D. Pavićević
Remyella hussoni Jeannel, 1934	IR20	Serbia, Pešter, Đerekare, Sjera jama	S. Ognjenović
Remyella javorensis S. Ćurčić & B. Ćurčić, 2008	MP07	Serbia, Sjenica, Ursule, Baždarska pećina	S. Ognjenović

Remyella javorensis S. Ćurčić & B. Ćurčić, 2008	IR10	Serbia, Sjenica, Ursule, Baždarska pećina	S. Ognjenović
Remyella javorensis S. Ćurčić & B. Ćurčić, 2008	IR8	Serbia, Sjenica, Ursule, Baždarska pećina	S. Ognjenović
Remyella javorensis S. Ćurčić & B. Ćurčić, 2008	IR3	Serbia, Sjenica, Ursule, Baždarska pećina	S. Ognjenović
Remyella javorensis S. Ćurčić & B. Ćurčić, 2008	IR21	Serbia, Sjenica, Donje Lopiže, Ušačka pećina	I. Njunjić
Remyella javorensis S. Ćurčić & B. Ćurčić, 2008	IR14	Serbia, Donje Lopiže, Ušačka pećina	I. Njunjić
Remyella propiformis Winkler, 1933	VRE1	Serbia, Pešter, Crvsko, Jagoševa pećina	M. Popović
Remyella propiformis Winkler, 1933	IR2	Serbia, Pešter, Crvsko, Jagoševa pećina	M. Popović
Remyella propiformis Winkler, 1933	IR13	Serbia, Pešter, Dolići, Jama bez dna	S. Ognjenović
Remyella propiformis Winkler, 1933	IR7	Serbia, Pešter, Dolići, Jama bez dna	S. Ognjenović
Remyella propiformis Winkler, 1933	IR1	Serbia, Pešter, Dolići, Jama bez dna	S. Ognjenović
Remyella propiformis Winkler, 1933	IR18	Serbia, Pešter, Boljare, Kaćunova jama	S. Ognjenović
Remyella propiformis Winkler, 1933	IR5	Serbia, Pešter, Kaćunova pećina	M. Popović
Remyella propiformis Winkler, 1933	IR9	Serbia, Pešter, Lopužino brdo, Lopužina jama	M. Popović
Remyella propiformis Winkler, 1933	IR4	Serbia, Pešter, Lopužino brdo, Lopužina jama	M. Popović
Remyella propiformis Winkler, 1933	IE8	Serbia, Pešter, Lopužina jama	M. Popović
Remyella sp. Jeannel, 1910	IR16	Serbia, Pešter, Đerekare, Vrelo Đerekarske reke	S. Ognjenović
Rozajella deelemani Perreau & Pavićević, 2008	IRO7	Crna Gora, Berane, Petnjik, Građa pećina	I. Njunjić
Rozajella deelemani Perreau & Pavićević, 2008	IRO8	Crna Gora, Berane, Petnjik, Građa pećina	I. Njunjić
Rozajella deelemani Perreau & Pavićević, 2008	IRO9	Crna Gora, Berane, Petnjik, Građa pećina	I. Njunjić
Rozajella madzgalji sp.n.	VR1	Crna Gora, Berane, Goražde, Mala pećina na Gareškom kršu	I. Njunjić
Rozajella madzgalji sp.n.	VR2	Crna Gora, Berane, Goražde, Mala pećina na Gareškom kršu	I. Njunjić
Rozajella madzgalji sp.n.	IRO1	Crna Gora, Berane, Goražde, Mala pećina na Gareškom kršu	I. Njunjić
Rozajella madzgalji sp.n.	IRO2	Crna Gora, Berane, Goražde, Velika pećina na Gareškom kršu	I. Njunjić
Rozajella madzgalji sp.n.	IRO3	Crna Gora, Berane, Goražde, Velika pećina na Gareškom kršu	I. Njunjić
Rozajella madzgalji sp.n.	IRO4	Crna Gora, Berane, Goražde, Velika pećina na Gareškom kršu	I. Njunjić
Rozajella madzgalji sp.n.	IRO5	Crna Gora, Berane, Goražde, Velika pećina na Gareškom kršu	I. Njunjić
Speonesiotes sp. Jeannel, 1910	IO39	BIH, Trebinje, Zupci, Vilina pećina	I. Njunjić
Speonomus (Speonomus) diecki (Saulcy, 1872)	MNCN-	Ariège, Moulis, Grotte d'Aubert	A. Faille

	AI536		
Stygiophyes akarsticus (Escolà, 1980)	NHM-IRC7	Lleida, Altron, Forat del Tincatinc	J. Fresneda

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Evolution, adaptation and speciation in *Anthroherpon* Reitter, a genus of highly evolved subterranean Coleoptera

Iva Njunjic

Abstract

The PhD research project focus on the study of evolution, adaptation, and speciation in the subterranean environment using troglobitic Coleoptera of the genus *Anthroherpon* as a model. Genus *Anthroherpon* belongs to the tribe Leptodirini (Coleoptera: Leiodidae: Cholevinae), a group that has undergone extensive diversification in the subterranean environment. All species of this genus have developed typical troglomorphic modifications: complete anophthalmy, apterism, extreme elongation of appendages, head, and pronotum, and physogastric elytra. To understand the evolutionary history of this group, the troglomorphic adaptations need to be studied in a phylogenetic framework. The thesis provides a comprehensive evolutionary analysis of the *Anthroherpon* radiation, using a dated molecular phylogeny as framework for understanding *Anthroherpon* diversification, reconstructing the ancestral range, and exploring troglomorphic diversity. In light of these findings, a new taxonomical organisation of the group has been proposed.

UNIVERZITET U NOVOM SADU
PRIRODNO-MATEMATIČKI FAKULTET
DEPARTMAN ZA BIOLOGIJU I EKOLOGIJU

Evolution, adaptation and speciation in *Anthroherpon* Reitter, a genus of highly evolved subterranean Coleoptera

- doktorska disertacija –

Mentori: Prof. dr Ivo Karaman
Dr Louis Deharveng

Kandidat: Iva Njunjić

Novi Sad, 2016.

KRATKA BIOGRAFIJA

Iva Njunjić rođena je 1987. godine u Beogradu, osnovnu školu i gimnaziju završila je u Lazarevcu. Diplomirala je 2011. godine na Biološkom fakultetu Univerziteta u Beogradu (smer ekologija i zaštita životne sredine) sa prosečnom ocenom 9,37 i stekla zvanje Msc odbranom rada „Kavernikolna fauna Orjena i Krivošija (Crna Gora)" pod mentorstvom prof. dr Slobodana Makarova. Kao stipendista Vlade Republike Francuske, 2012. godine upisala je doktorske studije u komentorstvu na Departmanu za biologiju i ekologiju Prirodno-matematičkog fakulteta Univerziteta u Novom Sadu i Muséum National d'Histoire Naturelle, Sorbonne Universités (Pariz, Francuska) pod mentorstvom prof. dr Ive Karamana i dr Louis Deharveng-a. Laboratorijski deo disertacije vezan za molekularnu filogeniju uradila je pod mentorstvom prof. dr Menno Schilthuizen-a na Univerzitetu u Lajdenu (Holandija). Doktorsku disertaciju brani 2016. godine u Parizu u Muséum National d'Histoire Naturelle.

Volontirala je u Zavodu za zaštitu prirode Srbije (2006–2011) gde po završetku master studija, tokom 2012-2014, radi na poziciji stručnog saradnika za zaštitu prirode. Nakon toga, u periodu 2014–2016, radi kao asistent na Katedri za Zoologiju Departmana za biologiju i ekologiju Prirodno-matematičkog fakulteta Univerziteta u Novom Sadu. Radila je i kao stručni saradnik na programu biologije u Istraživačkoj stanici Petnica, a 2012. godine bila je predstavnik Srbije u Svetskom parlamentu mladih za vode (World Youth Parliament for Water) u Marseju.

Iva Njunjić je aktivan biolog na terenu– učestvovala je u brojnim speleološkim i biospeleološkim ekspedicijama u Srbiji, Crnoj Gori, Bosni i Hercegovini, Hrvatskoj i Maleziji (Borneo, Sabah). Interesuje je evolucija u podzemnim staništima, pećinski insekti, biogeografija i zaštita pećinske faune.

Hovi Sad, novembar 2016

Iva Njunjić

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Doktorska disertacija predstavlja studiju evolucije, adaptacije i specijacije u podzemnim staništima troglobiontnih tvrdokrilaca roda *Anthroherpon*. Pomenuti rod pripada tribusu Leptodirini (Leiodidae, Cholevinae), grupi koja je prošla intenzivnu diverzifikaciju u uslovima podzemnih staništa. Sve vrste pomenutog roda poseduju tipične troglomorfne osobine, kao što su: anoftalmija, apterizam, ekstremno izduženi telesni nastavci, glava i pronotum, i fizogastrija. Radi razumevanja evolucione istorije grupe, troglomorfne adaptacije su studirane u filogenetskom kontekstu. U analizi evolutivne radijacije roda *Anthroherpon* korišćena je datirana molekularna filogenija kao okvir za razumevanje diverzifikacije roda, evolucije troglomorfnih karaktera i rekonstrukciju predačkog areala. U svetlu novih nalaza predložena je nova taksonomska organizacija grupe.

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Abstract:
AB

The PhD research project focus on the study of evolution, adaptation, and speciation in the subterranean environment using troglobitic Coleoptera of the genus *Anthroherpon* as a model organism. Genus *Anthroherpon* belongs to the tribe Leptodirini (Coleoptera: Leiodidae: Cholevinae), a group that has undergone extensive diversification in the subterranean environment. All species of this genus have developed typical troglomorphic modifications: complete anophthalmy, apterism, extreme elongation of appendages, head, and pronotum, and physogastric elytra. To understand the evolutionary history of this group, the troglomorphic adaptations need to be studied in a phylogenetic framework. The thesis provide a comprehensive evolutionary analysis of the *Anthroherpon* radiation, using a dated molecular phylogeny as a framework for understanding *Anthroherpon* diversification, reconstructing the ancestral range, and exploring troglomorphic diversity. In light of these findings, a new taxonomical organisation of the group has been proposed.

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Rezime

Evolucija, adaptacija i specijacija visoko evoluiranih pećinskih koleoptera roda *Anthroherpon* Reitter

Uvod u evoluciju u podzemnim staništima

Podzemna staništa obuhvataju šupljine različitih dimenzija koje se nalaze ispod površine zemlje, a koje mogu biti ispunjene vodom ili vazduhom. Premda su jedna od najslabije istraženih staništa na Zemlji, poznato je da veliki broj životinjskih grupa (od unicelularnih organizama do kičmenjaka) nastanjuje ove prostore. Pećine predstavljaju najbolje proučena podzemna staništa koja se zbog svoje izolovanosti i relativne stabilnosti fizičkih uslova često označavaju kao "prirodne laboratorije" za evoluciona istraživanja. Nastaju procesom speleogeneze i mogu se formirati u različitim tipovima stena. Najveći broj pećina formiran je u krečnjaku i sličnim karbonantnim stenama, ali se pećine takođe mogu formirati i u peščaru, lavi, bazaltu, granitu i ledu.

Sva podzemna staništa poseduju sledeće zajedničke osobine: odsustvo svetlosti, odsustvo primarne produkcije (osim u retkim slučajevima gde je zabeležena hemoautotrofija), relativno konstantna temperatura, visoka relativna vlažnost vazduha i odustvo sezonskih klimatskih varijacija. Navedeni abiotički faktori su glavni izvor selektvnog pritiska u podzemnim staništima. Organizmi koji su ih kolonizovali tokom evolucije razvili su specifične morfološke, fiziološke i bihevioralne karakteristike, kao što su: anoftalmija, depigmentisanost i promena telesne forme, izuzetno razvijeni čulni organi, duži životni vek, niža stopa metabolizma u poređenju sa nadzemnim srodnicima i drugi. Ovaj skup karakteristika označava se kao troglomorfija, a organizmi koji ih poseduju nazivaju se troglobiontima. Biološka disciplina koja proučava živi svet podzemnih staništa naziva se biospeleologija.

Veliki broj evolucionih studija bavi se procesom kolonizacije podzemnih staništa i adaptacijama pećinskih organizama. Predloženo je čak 11 različitih modela kolonizacije podzemnih staništa koji se mogu podeliti u dve grupe– modele aktivne i pasivne kolonizacije. Za svaki od ovih modela postoje primeri iz određenih životinjskih grupa, ali do sada nije predložen ni jedan model kojim bi se obuhvatile sve moguće tranzicije od epigejskih do obligatnih pećinskih organizama.

Akcenat brojnih studija koje se bave problematikom adaptacija pećinskih organizama na uslove u podzemnim staništima je na njihovim regresivnim karakterima, naročito anoftalmiji. Neke studije pokazale su da regresivni karakteri nastaju usled akumulacije neutralnih mutacija pod blagim selekcionim pritiskom, ali ima i onih koje pokazuju da i ovi karakteri mogu evoluirati pod snažnim selekcionim pritiskom sredine.

Veliki broj pećinskih organizama su endemiti vezani za određeno karstno područje. Dugo se smatralo da je ovakav areal rezultat kolonizacije i specijacije na datom lokalitetu jer se pretpostavljalo da troglobionti nemaju mogućnost širenja areala kada se jednom adaptiraju na podzemna staništa. Međutim, postojanje vrsta čiji areali zahvataju šire prostore ukazuje na mogućnost disperzije podzemne faune. Otkrićem mreže malih plitkih podzemnih šupljina (MSS-a) u karstu može se objasniti disperzija podzemne faune, ali su njene razmere još uvek nedovoljno poznate.

Uvod u model sistem

Među insektima, tvrdokrilci (Coleoptera) predstavljaju dominantnu grupu koja je kolonizovala podzemna staništa. Sa više od 230 rodova i oko 900 (uglavnom politipskih) vrsta, tribus Leptodirini (Leiodidae) predstavlja jedan od najraznovrsnijih grupa koleoptera prilagođenih na život u podzemnim uslovima. Većina vrsta živi isključivo u podzemnim staništima, kao što su pećine i fisure u karstu, izuzev nekoloko mirmekofilnih vrsta koje nastanjuju podzemna gnezda mrava ili šumsku stelju. Od svih pripadnika tribusa Leptodirini, subtribus Anthroherponina, a naročito rod *Anthroherpon* Reitter, 1889 poseduje najizraženije troglomorfne osobine: anoftalmija, ekstremno izduženi ekstremiteti i antene, izdužena glava i pronotum, hemisferične elitre. Rod je rasprostranjen u podzemnim staništima južnog dela Dinarida, regionu koji je prepoznat kao vruća tačka biodiverziteta pećinske faune. Rod *Anthroherpon* obuhvata 26 vrsta i 55 podvrsta i po broju vrsta je najbogatiji rod subtribusa Anthroherponina. Međutim, morfološki karakteri na osnovu kojih su ovi taksoni opisani su slabo definisani pa je njihov status kontroverzan. Mnoge vrste i podvrste opisane su na osnovu malog broja primeraka i na osnovu morfoloških razlika koje u većini slučajeva ne prevazilaze intrapopulacione varijacije. Osim toga, pojedine vrste imaju iznenađujuće disjunktnu distribuciju. Filogenetski odnosi o okviru Anthroherponina su nedovoljno proučeni i sve dosadašnje studije su bazirane isključivo na morfologiji.

Predmet istraživanja u okviru doktorske disertacije je evolucija, adaptacija i specijacija u podzemnim staništima na primeru obligatnih pećinskih koleoptera roda *Anthroherpon*. Istraživanje teži razumevanju evolucione radijacije i biogeografske istorije pomenutog roda kroz analizu podataka molekularne filogenije, geometrijske morfometrije, taksonomije i distribucije taksona.

Disertacija daje odgovore na sledeća pitanja:

1. Da li prethodna podela i klasifikacija vrsta i podvrsta, bazirana isključivo na morfologiji, može biti potvrđena objektivnim, kvantitativnim analizama baziranim na nekoliko nezavisnih setova podataka?
2. Kakvi su filogenetski odnosi sa drugim rodovima u okviru subtribusa Anthroherponina?
3. Kako su vrste međusobno filogenetski povezane i u kom vremenskom periodu se odigrala filogeneza?
4. Kakva je biogeografska istorija roda?
5. Da li su morfološki karakteri evoluirali neutralno ili postoje indikacije adaptivne divergencije?

Kako bih dala odgovore na ova pitanja, usvojila sam sledeću strategiju: prvi korak predstavlja izrada filogenetskog stabla subtribusa Anthroherponina da bi se ustanovili filogenetski odnosi između rodova i da bi se testirala validnost taksonomskih grupa prethodno definisanih na osnovu morfologije. Monofiletsko poreklo Anthroherponina i roda *Anthroherpon* pretpostavljeno na osnovu morfologije takođe je testirano u ovoj studiji. Rekonstrukcija filogenije roda *Anthroherpon* omogućila mi je da testiram da li su karakteri koji su tradicionalno prihvaćeni kao indikativni za klasifikaciju taksona ovog roda zaista pouzdani. Status problematičnih taksona je rešen, a proučavanjem inter- i intra-populacione morfološke varijabilnosti utvrđeni su karakteri relevantni za taksonomiju grupe. Takođe, primenjena je i tehnika molekularnog sata kako bi se datirala divergencija evolucionih linija. Ovaj pristup omogućio mi je da odgovorim na prva tri pitanja.

Naredni korak predstavlja primenu Bayesian metoda implementiranih u softver BioGeoBEARS kako bi se na osnovu filogenetskg stabla rekonstruisao predački areal roda. Rezultati ovih analiza daju odgovor na četvrto pitanje.

Poslednji korak je analiza morfoloških karaktera (koji se smatraju adaptacijama na životne uslove u podzemnim staništima) u filogenetskom kontekstu što mi omogućava da odgovorim na poslednje pitanje.

Molekularna filogenija roda *Anthroherpon* i subtribusa Anthroherponina predstavlja prvu filogenetsku analizu ovog tipa jedne grupe Dinarskih pećinskih koleoptera.

Rod *Anthroherpon*

Rod *Anthroherpon* pripada familiji Leiodidae i po broju vrsta je najbogatiji rod troglobiontnih koleoptera subtribusa Anthroherponina. Areal roda obuhvata jugoistočne delove Dinarskih planina– od Gornje Oćevije (Bosna i Hercegovina) na severu do primorskog pojasa planina u Crnoj Gori i Albaniji na jugu i od Prenja (Bosna i Hercegovina) na zapadu do Prokletija (Crna Gora) i Mokre planine (Crna Gora, Metohija) na istoku. Prisutan je u sva tri pojasa Dinarskih planina, a najveći diverzitet (50% taksona) zabeležen je u pojasu visokog planinskog karsta. Veliki broj taksona su endemiti– od 26 vrsta i 55 podvrsta roda *Anthroherpon*, čak 16 vrsta i 24 podvrste zabeleženo je u samo jednoj pećini. Sa druge strane, dve morfološki veoma slične vrste *A. taxi* i *A. latipenne* široko su rasprostranjene i imaju disjunktnu distribuciju.

Sve vrste ovog roda poseduju leptodiroidni oblik tela i odlikuju se najizraženijim troglomorfnim morfološkim karakteristikama među koleopterama. Zbog troglomorfne konvergencije determinacija vrsta i podvrsta je izuzetno zahtevna i status mnogih taksona je kontroverzan. U ovom Poglavlju, primenom molekularnih metoda (DNA barcoding) status kontroverznih taksona je rešen, a studiranjem intra- i inter-populacione morfološke varijabilnosti utvrđeni su karakteri relevantni za taksonomiju. Na osnovu morfoloških karaktera, rod *Anthroherpon* je podeljen u 7 specijskih grupa: *„cylindricolle“*, *„harbichi“*, *„ganglbaueri“*, *„latipenne“*, *„stenocephalum“*, *„pygmaeum“* i *„hoermanni“*. Molekularnim analizama pokazali smo da je ova podela većinom veštačka što ukazuje da je neophodna revizija roda kako bi se vrste pregrupisale i klasifikovale u skladu sa filogenijom. Sa druge strane, većina vrsta i podvrsta opisanih na osnovu morfologije pokazala se kao ispravno definisana. Međutim, rezultati ukazuju da bar tri podvrste (*A. taxi albanicum*, *A. hoermanni sericeum* i *A. hoermanni hypsophylum*) trebalo podići na nivo vrste, a da *A. taxi remyi* treba klasifikovati kao podvrstu *A. albanicum*. Vrednost od 3% divergencije u barkoding regionu COI gena je granična vrednost (barcoding gap) koja predstavlja razliku između inter- i intra-specifične genetičke distance. Interesantno je da postoji još jedna „barcoding gap“ od oko 8% divergencije u okviru interspecifične distance, što može biti indicija da je postojao period intenzivne, simultane specijacije praćen periodom sporije specijacije ili, alternativno, može ukazati na period izumiranja.

Sem taksonomije i činjenice da žive u pećinama o rodu *Anthroherpon* se ništa više ne zna. Ovde su po prvi put izneta zapažanja o biologiji, ekologiji i ponašanju ovih insekata zasnovana na njihovom posmatranju u prirodi i gajenju u laboratorijskim uslovima.

Ovo poglavlje takođe sadrži i rad objavljen u časopisu *Zootaxa* (2015): Članak 1: Two new species of the genus *Anthroherpon* Reitter, 1889 from northern Montenegro with notes on the "*A. ganglbaueri*" species group (Coleoptera: Leiodidae: Leptodirini)

U radu su opisane dve nove vrste roda *Anthroherpon* iz specijske grupe "*ganglbaueri*": *Anthroherpon sinjajevina* n.sp. i *Anthroherpon cecai* n.sp., pronađene u pećinama u severnom delu Crne Gore. Na osnovu morfoloških karakteristika, *A. udrzali* Giachino & Vailati je podignut sa subspecijskog nivoa na nivo vrste i predložena je sinonimija *A. brckoensis* Giachino & Guéorguiev sa *A ganglbaueri alticola* Knirsch. U radu je dat ključ za identifikaciju vrsta, kao i mapa distribucije taksona pomenute specijske grupe.

Članak 2: The cave beetle genus *Anthroherpon* is polyphyletic; molecular phylogenetics and description of *Graciliella* n. gen. (Leiodidae, Leptodirini)

Poglavlje u celini predstavlja rad publikovan u časopisu *Contributions to Zoology* (2016).

Koleoptere subtribusa Anthroherponina simbol su pećinske faune Dinarida koji su prepoznati kao vruća tačka biodiverziteta na svetskom nivou kada je pećinska fauna u pitanju. Dosadašnje filogenetske studije ove grupe bazirane isključivo na morfološkim karakterima smatraju se nepouzdanim zbog mogućnosti troglomorfne konvergencije, a morfološka staza i morfološki polimorfizam otežavaju klaifikaciju taksona. Da bismo testirali da li su karakteri koji su tradicionalno prihvaćeni kao indikativni u klasifikaciji Anthroherponina zaista pouzdani, testirali smo monofiletsko poreklo roda *Anthroherpon* koji u ovoj grupi prednjači po broju vrsta. Osim ovog roda, u molekularne analize uključili smo i rodove *Hadesia* Müller i *Leptomeson* Jeannel, takođe iz subtribusa Anthroherponina.

Naša studija, bazirana na molekularnoj filogentskoj analizi fragmenata 18S, 28S i COI genskih lokusa ukazala je na polifiletsko poreklo roda *Anthroherpon.* Rod *Leptomeson* formira sestrinsku kladu sa jednom politipskom vrstom roda *Anthroherpon* (*A. apfelbecki* sensu lato), a rod *Hadesia* formira sestrinsku kladu sa preostalim vrstama roda *Anthroherpon*. Da bismo rešili polifiliju, definisali smo novi rod *Graciliella* n. gen., a potom smo molekularnim metodama i morfometrijom ispitali njegov intragenerički diverzitet. Molekularnom filogenetskom analizom dva mitohondrijalna genska fragmenta (COI) ustanovili smo da rod *Graciliella* ima četiri vrste, od kojih smo dve opisali u ovom radu– G. *kosovaci* n. sp. i *G. ozimeci* n. sp. Razlike u COI genu među pomenutim vrstama iznose od 3.9% do 12.4%. Da bismo analizirali interspecifične morfološke razlike roda *Graciliella*, sproveli smo linearnu diskriminantnu analizu koristeći 40 linearnih morfometrijskih merenja uzetih sa antenna, maksilarnih palpusa, glave, toraksa, abdomena i nogu. Duž prve dve diskriminantne ose sve vrste roda *Anthroherpon* su razdvojene, izuzev dve nove vrste koje smo opisali u ovom radu. Međutim, ove dve vrste jasno su razdvojene duž treće diskriminantne ose. Polazeći od ove nezavisne morfološke separacije vrsta roda *Anthroherpon*, pronašli smo dodatne kvalitativne karaktere koji se mogu koristiti za determinaciju taksona. Identifikacioni ključ dat u radu zasnovan je upravo na ovim kvalitativnim karakteristikama.

Rod *Leptomeson* prvobitno je opisan kao podrod roda *Anthroherpon*, da bi kasnije dobio status roda. Ovo odvajanje zasnovano je na sledećim morfološkim karakteristikama roda *Leptomeson*: razvijeniji pedunculus mezotoraksa sa suženjem u središnjem delu i nešto širi u osnovi, endofalus sa sklerotizovanom strukturama, i paramere sa 3 do 4 sete. U ovoj studiji, molekularnom rekonstrukcijom filogenije pokazali smo da je ovo odvajanje opravdano. Klada *Leptomeson* + *Graciliella*, pored toga što je podržana molekularnim analizama, takođe je podržana i sledećim sinapomorfnim morfološim karakterima: veoma izdužen pronotum i mezotorakalni pedunkulus, i mezoventrum bez procesusa između mezokoksi. Monofilija roda *Leptomeson* je morfološki podržana prisustvom sklerotivovanih struktura u endofalusu koje ne postoje kod roda *Graciliella*.

Rezultati naših molekularnih analiza nisu u skladu sa prethodnim filogenetskim studijama roda *Hadesia* baziranih na morfološkim karakterima, ali ni sa distribucijom vrsta. Pokazalo se da su dve vrste koje žive u udaljenim krečnjačkim masivima (*H. lakotai* i *H. weiratheri*) sestrinske vrste, a da dve vrste sa Orjena (Crna Gora), koje su pritom i morfološki veoma slične (*H. asamo* i *H. weiratheri*), nisu blisko srodne.

Studija je pokazala da veliki broj morfoloških karaktera koji se tradicionalno koriste u klasifikaciji Anthroherponina (VIII ventrit kod ženki sa anteriornom apofizom/bez anteriorne apofize; endophallus sa sklerotizovanim strukturama/bez sklerotizovanih struktura; abdominalni ventriti sa glatkim pločama/ bez glatkih ploča; prvi ventrit ženki sa lateralnim šupljinama/bez lateralnih šupljina; treći maksilarni palpus kratak/dugačak; kandžice uzane/široke) predstavljaju homoplazije i najverovatnije su rezultat konvergencije.

Sveobuhvatna evolutivna analiza radijacije roda *Anthroherpon*

U ovom poglavlju predstavljamo evolutivnu analizu radijacije roda *Anthroherpon* koristeći datiranu molekularnu filogeniju kao okvir za razumevanje diverzifikacije roda, troglomorfnog diverziteta i rekonstrukciju predačkog areala.

U molekularne analize uključili smo 45 vrsta tribusa Leptodirini iz 12 rodova, od čega 16 vrsta i 23 podvrste roda *Anthroherpon*. Finalni matriks sadrži 4143 baznih parova. Amplifikovali smo šest fragmenata pet mitohondrijalnih gena: COIa, COIb, cob, rrnL-nad1, SSU i LSU. Za filogenetsku analizu koristili smo BEAST v2.3.2. na CIPRES web portalu. Kalibracija filogenije izvršena je na osnovu podataka dotupnih iz studije zapadnomediteranskih Leptodirini (Ribera et al., 2010), tj. postavljanjem očekivane starosti od 37.8 miliona godina za sestrinsku kladu taksona prisutnih na Sardiniji. Rekonstrukcija predačkog areala urađena je u softveru R pomoću paketa BioGeoBEARS 0.2.1. koristeći datirano filogenetsko stablo kao input. U morfometrijske analize uključena su ukupno 102 primerka (16 vrsta i 20 podvrsta) roda *Anthroherpon*, a merenje je izvršeno Micro-Vu Vertex 251 HC. Sa svakog primerka uzeto je 79 mernih tačaka (landmarks) sa antenna, maksilarnih palpusa, glave, pronotuma, elitri i nogu. Svaki primerak izmeren je tri puta. Generalized Procrustes Superimposition (GPA) i ostale analize oblika i linearnih merenja izvedene su u softveru R koristeći pakete: “ape”, “geomorph” i “phytools”. Morfometrija je urađena sa sledećim ciljevima: (i) proučiti razdvajanje rodova *Anthroherpon* i *Graciliella* na osnovu morfologije, (ii) proučiti troglomorfni diverzitet roda *Anthroherpon*, (iii) ispitati evoluciju morfoloških karaktera roda *Anthroherpon*, (iv) istražiti divergenciju karaktera dveju vrsta koje žive u sintopiji (*A. harbichi* i *A. weiratheri*).

Iako je focus ove studije bio na rodu *Anthroherpon*, rezultati do kojih smo došli ukazali su na moguće polifiletsko poreklo subtribusa Leptodirina što navodi na zaključak da bi trebalo razmotriti klasifikaciju rodova *Remyella, Rozajella* i *Nonveilleriella*.

Studija je pokazala monofiletsko poreklo subtribusa Anthroherponina, čije poreklo datira iz Gornjeg Oligocena (32 miliona godina). Četiri analizirana roda ovog subtribusa (*Graciliella, Leptomeson, Hadesia* i *Anthroherpon*) potiču iz perioda Gornji Oligocen– Donji Miocen. U okviru Anthroherponina mogu se razlikovati dve klade: *Anthroherpon* + *Hadesia* i *Leptomeson* + *Graciliella.* Filogenetska analiza pokazala je da rod *Anthroherpon* ima monofiletsko poreklo, a njegova divergencija datira iz Donjeg Miocena (22 miliona godina). Bazalnu kladu čini samo jedna vrsta- *A. cylindricolle cylindricolle*, a preostale vrste ovog roda podeljene su u dve klade definisane nodusima obeleženim brojevima “1” i “2”. Divergencija obeju klada započela je otprilike u istom periodu– Donjem Miocenu, pre oko 19 miliona godina. Klada definisana nodusom “1” obuhvata dve monofiletske klade (jednu čine vrste “*hoermanni*” a drugu vrste “*ganglbaueri*” specijske grupe) i vrsta *A. primitivum jeanneli* koja formira zasebnu kladu. Klada definisana nodusom “2” obuhvata 3 klade od kojih jednu formiraju tri vrste specijske grupe “*pygmaeum*”, a druge dve sestrinske klade obuhvataju preostale vrste roda *Anthroherpon* iz specijskih grupa “*pygmaeum*”, “*harbichi*”, “*stenocephalum*” i “*latipenne*”. Naši rezultati samo su delimično saglasni sa tradicionalnom podelom roda *Anthroherpon* na 7 specijskih grupa. Naime, prema našim nalazima, samo su grupe “*cylindricolle*” i “*ganglbaueri*” monofiletske. Filogenija pokazuje izvestan stepen geografske struktuiranosti– klada definisana nodusom “1” uglavnom obuhvata vrste koje žive u središnjem pojasu Dinarida (visoki planinski karst). Jedini izuzetak su vrste

A. matulici i *A. primitivum jeanneli* koje se javljaju u pojasu niskog priobalnog Jadranskog karsta (Low costal Adriatic karst).

Rekonstrukcija biogeografske istorije roda *Anthroherpon* pokazuje 4 sukcesivne faze: (i) poreklo u zapadnoj Crnoj Gori i disperziju zajedničkog pretka u istočnu Bosnu i Hercegovinu, (ii) višestruke disperzije kroz zapadnu Crnu Goru i Bosnu i Hercegovinu, uglavnom prema severu i severoistoku, (iii) period stagnacije, (iv) dalje disperzije prevashodno u jugoistočnom pravcu u istočnu Crnu Goru i Albaniju. Disperzija se odvijala uglavnom u Miocenu, kada su planine koje pripadaju Dinaridima već zauzele sadašnji prostorni raspored što navodi na zaključak da su tektonski događaji većih razmera koji su zahvatali ovaj prostor u prošlosti prethodili radijaciji roda *Anthroherpon*. Prema našoj biogeografskoj rekonstrukciji, tri planinska venca u zapadnoj Crnoj Gori: Orjen (1894 m), Dobreljica (1834 m) i Moračke planine (2226 m) predstavljaju predački areal roda *Anthroherpon* odakle je otpočela disperzija. Poreklo roda u zapadnoj Crnoj Gori je u skladu sa podacima o paleogeografiji Balkanskog Poluostrva.

Filogenija roda *Anhroherpon* ukazuje na jedinstveno poreklo samo dva morfološka karaktera: (i) pronotum bez posteriornog suženja (sp. grupa "*cylindricolle*") (ii) poslednja antenomera duža od pretposlednje (sp. grupa "*ganglbaueri*"). Većina drugih taksonomski relevantnih karaktera ima višestruko, nezavisno poreklo.

Morfometrijske analize su potvrdile jasno odvajanje rodova *Anthroherpon* i *Graciliella*. Najznačajnija razlika je u obliku pronotuma koji je kod *Graciliella* izduženiji i sa izraženijim suženjem. Takođe, rezultati su pokazali da oblik tela (izuzev oblika glave) kod *Anthroherpon* evoluira neutralno: K-vrednosti su oko 1.0 i pokazuju značajan filogenetski signal. Ovo ukazuje na činjenicu da kod većine vrsta nakon izolacije u alopatriji oblik tela polako evoluira u skladu sa Brownian motion model-om. Što se tiče troglomorfije telesnih nastavaka, nalazimo da neke vrste imaju izduženije antene i noge ali ovaj obrazac uglavnom prati filogeniju što može biti indicija da izduživanje telesnih nastavaka takođe evoluira neutralno i nije rezultat rapidne adaptacije. Međutim, postoje indikacije da u izvesnim slučajevima karakteri koje smo podvrgli morfometrijskim analizama evoluiraju adaptivno. Naše analize pokazale su da je oblik pronotuma dve vrste koje žive u sintopiji (*A. harbichi* i *A. weiratheri*) snažno divergirao (2.6 puta više nego u ostalim vrstama ovog roda; P=0.014) i pokazuje najveću razliku u filomorfoprostoru (phylomorphospace). Ovo ukazuje na snažnu divergenciju karaktera tokom ili nakon procesa specijacije koja može biti posledica: (i) *reproductive character displacement* ili (ii) *ecological character displacement*. Kako je relativna dužina maksilarnih palpusa ove dve vrste više različita nego što je očekivano, moguće je da postoji razlika u njihovoj ishrani. Pronotum sa druge strane ne igra ulogu u specijalizaciji na uslove mikrostaništa, ali je kod Cholevinae poznata njegova uloga u razmnožavanju–mužjak je prilikom kopulacije zakačen protarzama za pronotum ženke. Iz ovih razloga moguće je da u ovom slučaju postoje i reproduktivni i ekološki character displacement.

Članak 3: Molecular phylogenetics and systematics of the cave beetle genera *Remyella* Jeannel, 1931 and *Rozajella* S. Ćurčić, Brajković & B. Ćurčić, 2007 (Coleoptera: Leiodidae: Cholevinae: Leptodirini)

Ovo poglavlje u celini predstavlja rad poslat u časopis *Arthropod Systematics and Phylogeny.*

Studija izložena u prethodnom poglavlju ukazala je na moguće polifiletsko poreklo subtribusa Leptodirina što sugeriše da bi trebalo razmotriti klasifikaciju rodova *Remyella*, *Rozajella* i *Nonveilleriella* koji se smatraju blisko srodnim rodovima subtribusa Anthroherponina.

Subtribus Leptodirina je po broju vrsta najbogatiji subtribus tribusa Leptodirini i obuhvata 31 rod koleoptera zapadnog Paleartika adaptiranih na život u podzemnim staništima. Filogenetski odnosi rodova ovog subtribusa su slabo poznati zbog potencijalno konvergentnih morfoloških karaktera koji mogu biti posledica adaptacije na uslove u podzemnim ekosistemina. Dva roda sa neizvesnom filogenetkom pozicijom– *Rozajella* i *Remyella* su na osnovu morfologije uslovno smeštena u subtribus Leptodirina. Međutim, studije molekularne filogenije koje bi predloženu klasifikaciju potvrdile ili odbacile do sada nisu sprovedene na subtribusu Leptodirina. Vrste i podvrste roda *Remyella* veoma je teško razlikovati na osnovu morfologije a status mnogih taksona je kontroverzan. Da bismo ustanovili filogenetsku poziciju ova dva roda koja dele iste morfološke karaktere i žive u susednim geografskim područjima (Pešter polje i okolni masivi), sproveli smo molekularnu filogenetsku analizu. Taksone roda *Remyella* podvrgli smo detaljnoj molekularnoj i morfološkoj analizi da bismo rasvetlili njihov taksonomski status.

Specijacija roda *Remyella* odigrala se otprilike između pet i dva milliona godina, tokom Miocena i Pliocena, perioda tokom koga je Pešter polje bilo periodično prekriveno plitkim jezerima. Distribucija taksona ograničena na periferiju Pešter polja je najverovatnije posledica prisustva pomenutih jezera koja su sprečila da se vrste ravnomerno raširе kroz karst po čitavoj površini polja. Da bismo testirali korelaciju geografskih i genetičkih distanci kod roda *Remyella* sproveli smo Mantel test. Rezultati su pokazali visoku korelaciju (0.86) između dve matrice.

Dodatno, u radu je predstavljena i revizija rodova *Rozajella* i *Remyella*. Njihova bliska filogenetska srodnost potvrđena je molekularnim analizama za čije smo potrebe sekvencirali dva mitohondrijalna i dva nuklearna gena. Bliska filogenetska veza rodova *Nonveilleriella*, *Rozajella* i *Remyella* podržana je postojanjem istih morfoloških karaktera. Populacije roda *Remyella* sa već poznatih i sa novih lokaliteta su analizirane koristeći molekularne metode i morfologiju, da bi se rešio status problematičnih taksona. Prepoznato je pet vrsta ovog roda: *R. hussoni* Jeannel, 1933; *R. javorensis* S. Ćurčić & B. Ćurčić, 1995; *R. propiformis* Winkler, 1933; *R. raskae* S. Ćurčić & B. Ćurčić, 2008; i *R. scaphoides* Jeannel, 1931. Uspostavili smo četiri sinonimije: *R. propiformis propiformis* Winkler 1933 = *R. propiformis borensis* Winkler 1933 n. syn. = *R. droveniki* Giachino & Vailati, 1995 n. syn. = *R. montenegrina* S. Ćurčić, Antić, N. Ćurčić & B. Ćurčić, 2013 n. syn.; *R. hussoni* Jeannel, 1934 = *R. puncticollis* Jeannel, 1934 n. syn. Opisana je jedna nova vrsta roda *Rozajella*: *R. madzgalji* n. sp. iz istočne Crne Gore, a dat je i opis mužjaka vrste *R. deelemani* Perreau & Pavićević, 2008 pošto je opis vrste baziran na ženkama. Ključ za identifikaciju i mape distribudije vrsta rodova *Remyella*, *Rozajella*, and *Nonveilleriella* su takođe date u radu.