

# 18 Stygofauna of the Fresh Waters in Bulgaria

Ivan Pandourski

*Institute of Zoology, Bulgarian Academy of Sciences, Tsar Osvoboditel Blvd. 1, 1000 Sofia, Bulgaria,  
e-mail: stygologia@gmail.com*

**Abstract:** The species diversity and distribution of freshwater invertebrate stygofauna in Bulgaria, including 194 species belonging to 78 genera, are reviewed. Typical endemic species and genera for each taxonomic group are listed. Main paleogeographic and paleoclimatic events in Bulgaria that have had an important role in the formation of the stygobiotic faunistic complex are discussed

## 1 Introduction

The great diversity of karstic regions located from the sea level to the highest peaks of the Pirin Mts. (2914 m a.s.l.), as well as the paleogeographic and paleoclimatic development of Bulgarian landscapes, are the main factors which formed the relatively high biodiversity of underground water ecosystems in Bulgaria. Stygobiotic crustaceans and water mites are groups with the highest and the second highest species diversity, respectively. However, some large taxonomic groups of invertebrates, which inhabit ground waters, are poorly studied. These are, for example, Nematoda, Oligochaeta, and Ostracoda. Studies on the genus *Niphargus* belonging to the higher crustaceans are still in their initial stages. The possible number of species of these crustaceans, which inhabit caves or other underground habitats, could be several times greater than the number of recently discovered species.

At the same time, the large hydrogeological regions of Bulgaria remain totally unstudied (for example, parts of Western Rhodopes, almost the entire Eastern Rhodopes, as well as Slavyanka, Pirin, Belasitsa, Osogovo, Maleshevska and Vlahina Mts.). In other cases, the investigations were of an incidental nature. Examples of an ecological approach (Cvetkov et al., 1982) are rare. Quantitative data concerning stygofauna are also rare. Below, we concentrate on the composition, distribution, and origin of the known taxonomic groups inhabiting the ground waters of Bulgaria.

## 2 Species Diversity and Distribution

*Protozoa: Ciliata.* Free living in ground waters, unicellular Protozoa have not been studied in Bulgaria. Only two undetermined epibiotic representatives of ciliates from the genera *Vorticella* and *Tokophrya* have been found on stygobiotic higher crustaceans (Golemansky, 1983).

*Platyhelminthes: Turbellaria.* Depigmented planaria have been found in many caves in Bulgaria, but their genus and species have not yet been identified.

*Oligochaeta.* Our studies show that Oligochaeta are found in almost all studied karstic and phreatic habitats, but the collected material has not yet been determined. *Haplotaxis bureschi* (Michaelsen) is frequently found in the karstic waters of Western Stara Planina.

*Hirudinea.* Leeches living in the underground waters of Bulgaria are also poorly studied. The two known species of the genus *Dina* are found mainly in the karstic waters of Western Stara Planina. The classification of *Trocheta bykowskii* Gedroys as a stygobiont is debatable since it also inhabits mountain springs; this species forms isolated populations in some caves in Bulgaria.

*Crustacea: Ostracoda.* Ostracods inhabit ground waters in almost every region in Bulgaria, but they are still poorly studied. The only determined species inhabiting karstic underground habitats is *Pseudocandona eremia* (Vejdovsky). *Kovalevskiella elegans* Danielopol et Cvetkov is widely distributed in Bulgaria in phreatic lowland waters and also in Strandja Mts. The two species of *Kovalevskiella* are endemic for Bulgaria and inhabit phreatic waters.

*Crustacea: Copepoda.* The cyclopoid copepods are frequently found in diverse underground habitats within almost every type of interstitial and karstic waters. Two copepod genera, *Acanthocyclops* and *Diacyclops*, have the greatest number of stygobiotic representatives (seven species each); those from the species group "kieferi" of the genus *Acanthocyclops* inhabit mainly karstic waters. The species from the group "languidoides" of the genus *Diacyclops* have been most frequently found in hyporheic habitats and wells (Pandourski, 1997). *Graeteriella unisetigera* (Graeter) is the only representative of this genus; it mainly inhabits the interstitial habitats. The species from the genus *Speocyclops* are typical for the karstic regions of Western Stara Planina and Predbalkan. *Speocyclops rhodopensis* Pandourski is the only species of this genus found in South Bulgaria and it inhabits karstic crevice waters in the Western Rhodope Mts.

The harpacticoid copepods are the most numerous group of stygobiotic crustaceans found in Bulgaria (48 species) (Apostolov, in press). They have colonized almost all kinds of underground habitats. Mihailova-Neikova (1986) have found 24 harpacticoid species in the karstic waters of Strandja. Mihailova-Neikova (1975) identified 26 taxa from the karstic waters near the village of Bankya, Tran District, in western Bulgaria. The endemic species from the genera *Elaphoidella*, *Stygoelaphoidella*, and *Neoelaphoidella* are typical for the karstic habitats as well as for the interstitial ones.

*Crustacea: Malacostraca.* The higher crustaceans are common in the biota of karstic water ecosystems in Bulgaria. Most numerous are representatives of the amphipod genus *Niphargus*, which are found in almost every karstic region. Sixteen species of *Niphargus* and one species of *Niphargopsis* have been described so far. The amphipod *Bogidiella skopljensis* (Karaman) has been found in the karstic waters of Dobrudja, and *B. albertimaghi* Hertzog, in the phreatic waters of lowlands surrounding Sofia.

Representatives of the amphipod genus *Gammarus* are commonly found in ground waters. The endemic form *G. pulex cognominus* Karaman et Pinkster has been recorded from the Devetashko Karstic Plateau.

Isopods are also represented in underground ecosystems. Their different genera and families are usually adapted to different underground habitats. Microisopods from the genera *Microcharon* and *Microcerberus* are adapted to life in interstitial conditions and wells, and they are rarely found in karst. Conversely, Stenasellidae and Cirolanidae of large size (above 1 cm) inhabit mainly karstic habitats.

The representatives of Syncarida, which are exclusively subterranean inhabitants, are mainly adapted to life in interstitial conditions. We discovered previously unknown populations in the Duhlata Cave (Vitoshka Mts.), in caves of Ponor Mts. (Western Stara Planina), and in the Ponora Cave, near the village of Chiren in Vratsa District. *Bathynella natans natans* Vejdovsky and *Parabathynella stygia* Chappuis are frequently and widely found in Bulgaria, and five species of the genus *Hexabathynella* are typical for the phreatic waters of South Bulgaria.

*Acari*. Underground water mites are a comparatively well-studied group due to the efforts of Petrova (1968, 1984). They comprise the larger part of the stygobiotic complex in Bulgaria (55 species). The generic diversity is very high (32 genera); 23 genera are represented only by one species each. The fauna of stygobiotic mites in the hyporheic and phreatic waters of Veleka River in Strandja Mts. (Petrova, 1968) has been well studied. Numerous new and endemic species have been described throughout Bulgaria.

*Mollusca*. Twelve stygobiotic mollusk species are known from Bulgaria. They belong to eight genera (Table 1). This group of invertebrates is very common in the karstic waters of Western Stara Planina, where they form numerous populations with high density.

### 3 Endemism

In Bulgarian stygofauna we find Carpathian–Balkan, Bulgarian, and local endemics. The specific features of the underground water ecosystems (hydrographic isolation with very restricted possibilities for the migration of stygobionts, etc.) are factors influencing the high level of endemism among the underground water fauna.

The recently discovered underground species of Oligochaeta, *Haplotaxis bureschi* (Michaelsen), is a Balkan endemic species known from the wells near Skopje in Macedonia and the karstic waters of Western Stara Planina. The leech subspecies *Dina lineata arndti* Augener is a regional endemic taxon for Western Stara Planina.

Three species of underground ostracods are endemic: *Kovalevskiella bulgarica* (Danielopol), a regional endemic of western Bulgaria; *K. cvetkovi* (Danielopol), a Balkan endemic; and *Mixtacandona elegans* Danielopol et Cvetkov, a Bulgarian endemic species.

**Table 1** The generic composition of the Bulgarian stygofauna

Taxonomic groups	Genera	Number of stygobiotic species
PROTOZOA	<i>Vorticella</i> Linnaeus	1*
	<i>Tokophrya</i> Bütschli	1*
OLIGOCHAETA	<i>Haplotaxis</i> Hoffmeister	1
HIRUDINEA	<i>Trocheta</i> Dutrochet	1
	<i>Dina</i> Blanchard	2
OSTRACODA	<i>Pseudocandona</i> Kaufmann	2
	<i>Mixtacandona</i> Klie	1
	<i>Kovalevskiella</i> Klein	2
COPEPODA:	<i>Eucyclops</i> Claus	1
CYCLOPOIDA	<i>Acanthocyclops</i> Kiefer	7
	<i>Diacyclops</i> Kiefer	7
	<i>Speocyclops</i> Kiefer	4
	<i>Graeteriella</i> Brehm	1
COPEPODA:	<i>Nitocrella</i> Chappuis	4
HARPACTI-	<i>Nitocrellopsis</i> Petkovski	1
COIDA		
	<i>Elaphoidella</i> Chappuis	20
	<i>Stygoelaphoidella</i>	7
	Apostolov	
	<i>Neoelaphoidella</i>	1
	Apostolov	
	<i>Parastenocaris</i> Kessler	8
	<i>Ceuthonectes</i> Chappuis	2
	<i>Maraenobiotus</i> Scott	5
SYNCARIDA	<i>Bathynella</i> Vejdovsky	1
	<i>Parabathynella</i> Chappuis	1
	<i>Hexabathynella</i> Schminke	5
ISOPODA:	<i>Sphaeromides</i> Dollfus	2
CIROLANIDAE		
ISOPODA:	<i>Asellus</i> E.L. Geoffroy	1
ASELLOTA		
	<i>Proasellus</i> Dudich	1
	<i>Balkanostenasellus</i>	1
	Cvetkov	
	<i>Stenasellus</i> Dollfus	1
	<i>Protelsonia</i> Mehely	2
	<i>Microcharon</i> Karaman	9
ISOPODA:	<i>Microcerberus</i> Karaman	2
MICROCERBERIDAE		
ISOPODA:	<i>Bureschia</i> Verhoeff	1
ONISCIDEA		
AMPHIPODA:	<i>Gammarus</i> Fabricius	1
GAMMARIDAE		
AMPHIPODA:	<i>Niphargus</i> Schiodte	16
NIPHARGIDAE	<i>Niphargopsis</i> Chevreux	1
AMPHIPODA:	<i>Bogidiella</i> Hertzog	2
BOGIDIELLIDAE		
AMPHIPODA:	<i>Ingolfiella</i> Hansen	1
INGOLFIELLIDAE		

continued

Table 1 continued

Taxonomic groups	Genera	Number of stygobiotic species
ACARI	<i>Halacarellus</i> Viets	3
	<i>Lobohalacarus</i> Viets	1
	<i>Soldanellonyx</i> Walter	3
	<i>Parasoldanellonyx</i> Viets	1
	<i>Porolohmanella</i> Viets	2
	<i>Bandakia</i> Thor	1
	<i>Kawamuracarus</i> Uchida	1
	<i>Atractides</i> Koch	11
	<i>Neumania</i> Lebert	1
	<i>Feltria</i> Koenike	1
	<i>Barbaxonella</i> Lundblad	1
	<i>Albaxona</i> Szalay	2
	<i>Axonopsis</i> Piersig	4
	<i>Erebaxonopsis</i> Motaş et Tanasachi	1
	<i>Ljania</i> Thor	1
	<i>Lethaxona</i> Viets	1
	<i>Kongsbergia</i> Thor	4
	<i>Stygomonomia</i> Szalay	1
	<i>Momonisia</i> Petrova	1
	<i>Mideopsis</i> Neuman	1
	<i>Neoacarus</i> Halbert	1
	<i>Chappuisides</i> Szalay	1
	<i>Stygohydracarus</i> Viets	1
	<i>Phreatohydracarus</i> Tanasachi et Orghidan	1
	<i>Hungarohydracarus</i> Szalay	1
	<i>Balcanohydracarus</i> Motaş et Tanasachi	1
	<i>Arrenurus</i> Duges	2
	<i>Charonothrombium</i> Motaş et Tanasachi	1
	<i>Wandesia</i> Schechtel	1
	<i>Frontipodopsis</i> Walter	1
	<i>Torrenticola</i> Piersig	1
	<i>Aturus</i> Kramer	1
	MOLLUSCA	<i>Belgrandiella</i> A.J. Wagner
<i>Cavernista</i> Radoman		1
<i>Hauffenia</i> Pollonera		1
<i>Iglica</i> A.J. Wagner		1
<i>Insignia</i> Angelov		1
<i>Plagigeyeria</i> Tomlin		1
<i>Pontobelgrandiella</i> Radoman		1
<i>Saxurinator</i> Schütt		2

\* epibiotic species on stygobiotic higher crustaceans

High percentage of endemism is found among Copepoda. All endemic species of the order Cyclopoida belong to three genera: *Acanthocyclops*, *Diacyclops*, and *Speocyclops*. In karstic waters we frequently find different local and regional endemics from the species group “*kieferi*” of the genus *Acanthocyclops*. These are *A. chappuisi* (Naidenow et Pandourski) from the caves of Zemen Gorge in Struma Valley, *A. radevi* Pandourski from the karst of Ponor Mts. in Western Stara Planina, and *A. strimonis* (Pandourski) from the karst of Bosnek in Vitoshka Mts. and Lepenitsa Cave near Velingrad in Western Rhodopes. The Carpathian–Balkan endemic taxa are *A. iskrecensis* Pandourski from Ponor Mts. and Lazareva Pečina Cave in Kučaj Mts. (eastern Serbia) and *A. propinquus* (Plesa) from western Bulgaria and Romania. A Bulgarian endemic from the species group “*crassicaudis*” is *A. fontinalis* (Naidenow), but its taxonomic status is unclear (Pandourski, 1997). A local endemic subspecies from the genus *Diacyclops* is *D. pelagonicus saetosus* Pandourski, found so far only in the Dushnika Cave near the village of Iskrets in Western Stara Planina. *Speocyclops rhodopensis* Pandourski is the only local endemic species of its genus inhabiting Sbirkovata Cave and Lednitsata Cave in the Rhodopes. Widely distributed in the karst of Western Stara Planina is the Balkan endemic species *S. lindbergi* Damian, originally described from Romania.

The harpacticoid copepods from the genera *Elaphoidella*, *Stygoelaphoidella*, and *Parastenocaris* show the highest percentage of endemism in the karstic waters of Bulgaria. Particularly rich in endemic species is the karst of Ponor Mts. The local endemic taxa *Parastenocaris bulgarica* Apostolov, *Elaphoidella iskrecensis* Apostolov, *Stygoelaphoidella elegans* Apostolov, and *Maraenobiotus parainsignipes* Apostolov live in the zone of the karstic springs near Iskrets. The local endemic *Elaphoidella pandurskyi* Apostolov and the Balkan endemic *Parastenocaris jeanneli* Chappuis have been found in the karstic area near the village of Bosnek. Many local endemic species are known only from one or more caves or springs: *Neoelaphoidella intermedia* Apostolov from Mishin Kamak Cave in Western Stara Planina (Apostolov, 1999), *Stygoelaphoidella stygia* Apostolov from Vasilyovska Mts. (Central Stara Planina), *S. elegans* Apostolov from Ponor Mts., *Elaphoidella balkanica* Apostolov from Vrachanska Mts. (Western Stara Planina), and *E. cavernicola* Apostolov from Ponor Mts. and the karstic region near Karlukovo (Apostolov, 1992).

Higher crustaceans (Malacostraca) also show a high percentage of endemics among the stygobiotic groups. Five species of the syncarid genus *Hexabathynella* are regional endemics from the phreatic waters of South Bulgaria. In the caves of Duhlata and Marina Dupka (Ponor Mts.) we found some representatives of the order Bathynellacea but the species were impossible to determine.

Isopods, particularly the group Asellota, are poorly studied in Bulgaria. Their species number, including the endemic taxa, is probably higher than that actually known. The Bulgarian endemic species *Balkanostenasellus rumelicus* (Cvetkov) from South Bulgaria and the two local endemics *Protelsonia lakatnicensis* (Buresch et Guéorguiev) and *P. bureschi* (Racovitza) from western Bulgaria belong to this group. Isopods of the family Cirolanidae are the largest in size of all cave water

crustaceans known in Bulgaria. They are also relicts with a supposed ancient origin dating from the end of the Mesozoic. These are *Sphaeromides bureschi* Strouhal from Western Stara Planina and *S. polateni* Angelov from a karstic spring near Teteven in Central Stara Planina.

Both genera *Microcharon* and *Microcerberus* from the microisopods include regional endemic taxa, distributed mainly in interstitial habitats. They are sometimes found in caves and karstic springs (Ponora Cave near Chiren Village, Zemen Gorge, and the areas of Beglezh and Sadovets).

The only local endemic subspecies of the family Gammaridae is *Gammarus pulex cognominus* Karaman et Pinkster inhabiting the caves of Devetashko Plateau.

The amphipod genus *Niphargus* is typical for the karstic water ecosystems of Bulgaria. Unfortunately, in spite of the efforts of Dr. Stoitze Andreev from the National Museum of Natural History, Sofia (Andreev, 1972; Pandourski, 1998), the species of this genus are still poorly studied. Sixteen species of *Niphargus* have been determined so far, and five of them are local endemic taxa. The representatives of the genera *Bogidiella* and *Ingolffiella* have only one Balkan endemic species each; *B. skopljensis* (Karaman) inhabits the karstic waters of Dobrudja.

The high level of endemism is also observed among the stygobiotic water mites. Bulgarian endemic species are *Mideopsis motasi* Petrova, which is found throughout Bulgaria; *Soldanellonyx chappuisi thracicus* Petrova from the Thracian Lowland and Strandja Mts.; and *Halacarellus phreaticus* Petrova from the phreatic waters near to the Black Sea coast.

Regional endemic taxa for Strandja are *Atractides asticae* Petrova, *A. longiporus* Petrova, *Axonopsis orghidani* Petrova, *A. bureschi* Petrova, *Pontarachna valkanovi* Petrova, and *Momonisia phreatica* Petrova. Petrova suggests that the only species of the genus *Momonisia* from Strandja is a relict form descended from the tropical fauna, which inhabited the Bulgarian territory in the Tertiary.

All recently discovered underground mollusks from Bulgaria are local endemic species. Four gastropod species, two from the genus *Belgrandiella* and one each from the genera *Insignia* and *Pontobelgrandiella*, are known from the karstic waters near Teteven. The remaining species are known from caves or karstic springs in Western Stara Planina (the areas of Lakatnik, Iskrets, and Opitsvet).

## 4 Origin

No analysis or discussion has been published concerning the origin of the stygobiotic Mollusca, Oligochaeta, Hirudinea, and Ostracoda in Bulgaria. The origin of the stygobiotic crustaceans has been discussed by Cvetkov (1967, 1972, 1975, 1982), Pandourski and Breskovski (1995), Pandourski (1997), and Pandourski and Ognjanova (2001). In terms of their origin, the stygobiotic crustaceans are divided in two main groups: species with a marine origin and those with a freshwater origin.

To the first group belong mainly the higher crustaceans. The colonization of underground karstic waters by Cirolanidae and Stenasellidae is related to the paleogeographic situation at the time when Stara Planina (the Balkanid arch) was formed during the Upper Cretaceous. During this period, direct contact between the paleokarstic systems and the regressing marine basins occurred. Both Bulgarian species of the genus *Sphaeromides* and both Bulgarian species of the genus *Protelsonia* are relicts, and their ancestors should have inhabited parts of the northern Tethys (Pandourski and Breskovski, 1995).

The formation of the system of Neogene lake basins along the valleys of Struma, Mesta, and Maritsa (Galassi et al., 1995; Pandourski and Ognjanova, 2001) has been important for the origin of the relict endemic species of *Hexabathynella* and *Microcharon* from South Bulgaria. At some stage of their development, these lakes have been affected by marine waters. The brackish Miocene basins have influenced the region of Strandja, located near to the recent south Bulgarian Black sea coast (Pandourski and Ognjanova, 2001).

Two stages of paleogeographic and paleoclimatic development of Bulgarian territories were important for the species with freshwater origin, especially for the lower crustaceans such as copepods. The first stage is connected with the cool climate in Europe in the end of the Tertiary and with the beginning of glacial periods during the late Pliocene and early Quaternary. At that time, the surface populations of many boreal groups of species and genera are believed to have disappeared. The colonization and isolation of part of these populations is a factor in the origin of vicariant species and their divergence (Pandourski, 1997). Such Tertiary paleoendemics are found in the species group "kieferi" of the genus *Acanthocyclops*, in the genera *Speocyclops*, *Elaphoidella*, *Stygoelaphoidella*, etc.

The second stage is related to the retreat of the Würm Glacial Period, accompanied by climate warming. The populations of the cold-preferring species remained isolated in the ground waters. The neoendemics from the genus *Diacyclops* probably have such an origin.

The origin of part of the species and genera of underground water invertebrates is still being debated. For example, there are various opinions about the marine or freshwater origin of the amphipod genus *Niphargus*. Petrova (1968) examines the origin of the stygobiotic water mites of Strandja Mts., which is the most thoroughly studied region in Bulgaria. She concludes that the most ancient group of "paleophreatic" mites originates from the Paleogene, when Strandja had already emerged as land surrounded by sea. After the formation of underground populations they became isolated. As a result, the entire subterranean families and subfamilies evolved: *Kawamuracarus* (Limnesiidae), *Hungarohydracaridae*, and *Chappuisiidae*.

For the so-called "neophreatic" water mites, Petrova (1984) suggests that they originated during the post-Tertiary stage, when the climate was cooler. Part of the populations still exhibits similarities to their surface ancestors. Many transitional species ranging from surface to strictly underground species are typical for these water mites. An example is the genus *Atractides*, for which 22 species are known from Strandja, including 11 stygobionts.



## 5 Conclusions

At this moment, 194 species of stygobiotic animals belonging to 78 genera are known from the underground continental waters of Bulgaria (Table 1). The crustaceans have the highest species diversity with 121 species, followed by the water mites with 55 species. The most ancient relict representatives are the crustaceans whose ancestors are believed to have inhabited the North Tethys during the Upper Cretaceous. Climatic changes leading to a general cooling and mountain formation (orogenesis) in Bulgarian territory during the Tertiary had an important role in the origin of the paleoendemics with freshwater origin. After the retreat of the Würm Glacial Period and warming of the climate many populations of cold-preferring species remained isolated in the underground water habitats and gave rise to many neoendemics.

The Western Stara Planina is characterized by the presence of a relatively high percentage of endemic stygobionts. Particularly rich is the endemic acarofauna of Strandja. Less number of endemics is found in the Rhodopes; however, we have to consider here the fact that large regions of Bulgaria are poorly studied.

The study of biodiversity in the ground waters of Bulgaria is still in its early stages, and is limited to the available taxonomists. Importance of the stygofauna as a part of the natural heritage of Bulgaria has still not been evaluated, and this fauna is excluded unreasonably from the National Action Plan for the Protection of Biodiversity (2000).

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