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LAND SNAILS FROM SUBTERRANEAN TRAPS EXPOSED IN A FORESTED SCREE SLOPE (WESTERN CARPATHIANS, SLOVAKIA)

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The paper is dedicated to the memory of Jozef Šteffek (1952–2013), the famous Slovak malacozoologist who died suddenly during the implementation of this study. The first series of the samples was determined by him.

Abstract: Forested scree slopes represent one of the types of shallow subterranean habitats characterised by several unique traits, such as close contact with the soil surface, better availability of organic matter, and lower seasonal fluctuations in microclimate. Relatively varied assemblages of gastropods were collected using a series of subterranean traps filled with formaldehyde or ethylene glycol, exposed inside a forested scree slope at depths from 5 to 95 cm in the Natural Nature Reserve Sivec (eastern Slovakia). The number of gastropods was smaller compared to arthropods. The material of 120 specimens consisted of 22 gastropod species representing 15 families. Almost 87% of the specimens were captured near the soil surface (5–15 cm) and only few of them penetrated deeper. Ethylene glycol was more effective than formaldehyde in collecting gastropods. No subterranean species were found. Forest species predominated over some hygrophilous and petrophilous ones.

Key words: Gastropoda, superficial subterranean habitat, vertical distribution, subterranean traps

INTRODUCTION

In the central European mountains, scree slopes represent island formations and demonstrate the effect of a periglacial climate. The living conditions inside the stony accumulations depend particularly on stabilisation by the soil and vegetation cover (Růžička 1988, 1993). The loose scree slopes are characterised by a specific microclimate defined by a sharp contrast between temperature and humidity values. The surface tends to overheat enormously during the day and cool off overnight, while a relatively constant low temperature and high air humidity is typical of the inner parts (Růžička 1991). On the other hand, the forested scree slopes are characterised by much more stable temperature and humidity values in the whole profile. Inner zones of such talus deposits represent a peculiar type of hypogean environment known as mesovoid shallow substratum (MSS) (Juberthie et al. 1980). MSS is constituted by a network of empty ventilated voids within rocky debris, which have accumulated on the bedrock in various morphogenetic types. The accumulation of rocks is covered with a layer of soil that isolates MSS from the ground surface (Juberthie 2000, Juberthie & Decu 2004). Two different species communities can be found in MSS. The first one is composed of subterranean forms, among which troglobionts and eutroglophiles may be distinguished. Troglobionts are strongly bound to hypogean environment and possess some typical morphological adaptations to life in darkness. On the other hand, eutroglophiles represent essentially edaphic species; however, they are able to maintain permanent subterranean populations. The second community of species occurring in the MSS includes edaphic species. They either actively migrate to it or are transported by the percolating rainwater (Sket 2008).

Forested talus deposits along with other kinds of shallow underground habitats (e.g. air-filled epikarstic cavities, crevices in bedrock, seepage springs) are generally termed superficial subterranean habitats (SSHs) (Culver & Pipan 2008). They represent a transition between soil and deep subterranean habitats (caves) and have several common characteristics with them. Like the caves, SSHs are completely aphotic, they are spatially large enough for the inhabiting fauna not to be in contact with a solid surface, and they have organisms with character-states related to subterranean life. SSHs share with soil the occurrence of edaphobionts and the close contact with the soil surface (less than 10 m). However, SSHs possess several unique characteristics: minimal seasonal fluctuations of microclimate, better availability of organic matter, and the presence of troglomorphic fauna typical only of SSHs (Culver & Pipan 2010, Pipan et al. 2011, Pipan & Culver 2013).

The studies on Gastropoda in subterranean environment are rather scarce and they are mainly focused on aquatic species (e.g. Georgiev 2011, Pešić & Glöer 2012). In terms of terrestrial gastropods, most of the data come from caves. Dvořák (2002) investigated malacofauna in 14 limestone caves in south-western Romania. The occurrence of most species found was limited to the vicinity of cave entrances. Four species showed a preference to deeper parts of the caves. Among them, *Oxychilus montivagus* and *O. glaber*, regarded as troglophilous species, were recorded. Subsequently, Dvořák (2003) conducted a similar study in 10 limestone caves located in southwest Bohemia. Several forest species were observed regularly at those sites, e.g. *Oxychilus cellarius*, *Helix pomatia*, *Helicigona lapicida*. Slapnik & Ozimec (2004) highlighted the distribution of troglobiont snails of the genus *Zospeum* in the Croatia karst region. There are some data referring to terrestrial gastropods in Slovak caves. Šteffek (1992) mentioned the occurrence of several ecological forms of epigeic gastropod species in some karstic caves in eastern Slovakia. Ložek (1999) and Ložek & Horáček (2007) highlighted the Late Pleistocene and Holocene succession of terrestrial malacofauna when eluting the remains of shells from sediments from a deep research pit excavated in the entrance of Veľká Ružínska cave in eastern Slovakia – the cave situated in the same slope as our study plot. Several Carpathian endemics occurred within some strata of the excavated column. Three endemic species (*Faustina faustina*, *Cochlodina cerata cerata* and *Vestia turgida*) were present in the glacial stratum. Another endemic, *Pseudalinda stabilis*, was found in the Late Holocene stratum. Particular findings of Mollusca suggest the local succession to be more concerned with woodland environments. Šteffek (2000) conducted a survey of gastropod fauna in two karstic caves in central Slovakia. He found several epigeic species using the caves as a shelter from adverse conditions on the surface. Afterwards Papáč et al. (2009) continued with the research of cave fauna in volcanic rocks in south Slovakia. Three surface gastropods species were found: *Aegopinella* cf. *nitens*, *Discus ruderatus* and *Oxychilus glaber*.

In addition to caves, the malacofauna was also studied in artificial underground shelters. Dvořák (2005) pointed out the presence of gastropods in several cellars, abandoned mines, and military bunkers in the Czech Republic. A troglophilous species *Oxychilus depressus* was found in some mines. The occurrence of common forest species was characteristic of other types of artificial cavities. For instance, *Helix pomatia*, *Limax maximus* and *Oxychilus cellarius* were typical inhabitants of cellars, while *Limax cinereoniger*, *Monachoides incarnatus*, *Cochlicopa lubrica* were the most abundant species in bunkers.

Very little is still known about the gastropods inhabiting the MSS habitats. Ortuño at al. (2013) mentioned the presence of terrestrial gastropods in several alluvial MSS sites in southeast Spain. The aims of this study were: (1) to determine the malacofauna composition and vertical distribution of gastropods in different profiles of forested talus deposit and (2) to evaluate the differences in capture efficiency between formaldehyde and ethylene glycol.

MATERIAL AND METHODS

The study was carried out on forested talus deposits (48°50'31.27"N, 21°06'33.97"E) in the National Nature Reserve Sivec (Čierna hora Mts, Western Carpathians, eastern Slovakia). The site covered with the *Tilieto-Aceretum* forest association is situated in the Malý Ružínok Valley under a limestone wall of 10–25° slope facing northeast at the altitude 530 m a. s. l. Scree material is covered by Holocene rendzina (FAO) with a sparse herb layer: *Mercurialis perennis*, *Dentaria* sp., *Lamium* sp., *Urtica dioica*, *Asplenium alternifolium*, young seedlings of *Sambucus* sp., and ferns (Fig. 1). The microclimate of the study plot is characterised by long-lasting thermal fluctuations, but still synchronised by climate dynamics on the soil surface above. The significant diurnal variations are typical of the soil surface above. The average annual tem-

Fig. 1. The studied scree slope and its location in Slovakia

perature (measured from October 2008 to November 2009) on the soil surface was 7.9°C. It increased with the depth and at the depth of 95 cm reached the value of 9.3°C (RENDOŠ et al. 2012).

The material of Gastropoda was collected using a series of subterranean traps (SCHLICK-STEINER & STEINER 2000) primarily installed to collect arthropods. Each trap consisted of a PVC tube (length 110 cm, circumference 35 cm) which accomodated 10 pitfall traps (plastic cups of 500 ml volume) filled with preservatives. The gaps (diameter approx. 7 mm), allowing animals to enter the traps, were drilled at 10 horizontal levels (5, 15, 25, 35, 45, 55, 65, 75, 85 and 95 cm) around the tube. The top of tube was covered with a tin plate (Fig. 2). The traps were dug triplicate in line, 30 cm one from another. Two kinds of preservatives, commonly applied in the studies on Arthropoda, were used: 4% formaldehyde (October 2008 to November 2009) and 50% ethylene glycol (November 2009 to July 2010). The sampling with formaldehyde traps was performed monthly. The ethylene glycol traps were controlled twice: in May and July 2010. The captured individuals were subsequently fixed in 75% ethyl-alcohol and determined to species level. The gastropod nomenclature used in this study follows HORSÁK et al. (2010) and WELTER-SCHULTES (2012).

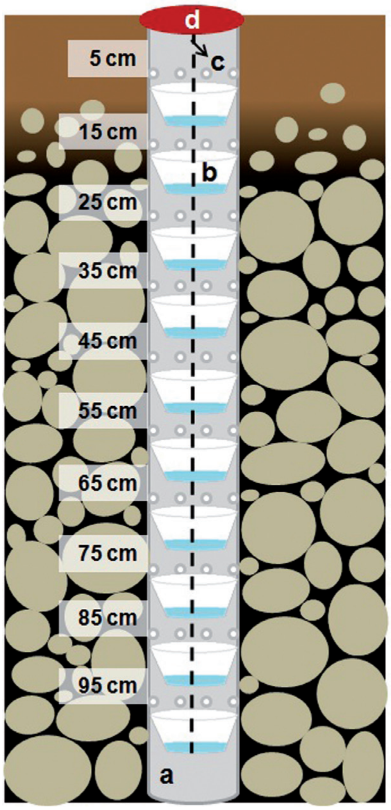

Fig. 2. Subterranean trap design: (a) – PVC tube, (b) – pitfall trap, (c) – helical rod, (d) – metal cover

RESULTS

More than 34,000 individuals of invertebrates were collected, and most of them (99.7%) represented Arthropoda. A total of 120 gastropods (0.35% among all invertebrates) were collected. They represented 15 families and 22 species; 2 young specimens were not identified to the species level (Table 1). The highest species diversity was observed in the family Hygromiidae (4 spp.), followed by Zonitidae (3 spp.). Gastropod activity was concentrated mainly between the depths of 5 and 15 cm under the soil surface,

Table 1. Diversity and spatial distribution of Gastropoda. Traps: F – formaldehyde, E – ethylene glycol

Species	Depth (cm)										Trap			
	5	15	25	35	45	55	65	75	85	95	F	E	Total	D (%)
1. Carychiidae														
Carychium tridentatum (Risso, 1826)	2					3	1				4	2	6	5.0
2. Agriolimacidae														
Deroceras sp.	5	1		1	1							8	8	6.7
Deroceras turcicum (Simroth, 1894)		1										1	1	0.8
3. Clausiliidae														
Alinda cf. *biplicata* (Montagu, 1803)	1	1									2		2	1.7
Vestia gulo (E. A. Bielz, 1859)	7											7	7	5.8
indet. sp.					1					1	2		2	1.7
4. Daudebardiidae														
Daudebardia brevipes (Draparnaud, 1805)	1											1	1	0.8
Daudebardia rufa (Draparnaud, 1805)			1								1		1	0.8
5. Discidae														
Discus perspectivus (M. von Mühlfeld, 1816)	4											4	4	3.3
6. Gastrodontidae														
Zonitoides nitidus (O. F. Müller, 1774)		1									1		1	0.8
7. Helicidae														
Faustina faustina (Rossmässler, 1835)	10											10	10	8.3
8. Hygromiidae														
Monachoides incarnatus (O. F. Müller, 1774)	3	1										4	4	3.3
Perforatella dibotrion (Bielz, 1860)	5											5	5	4.2
Petasina unidentata (Draparnaud, 1805)	6										2	4	6	5.0
Trochulus hispidus (Linnaeus, 1758)	8											8	8	6.7
9. Limacidae														
Malacolimax tenellus (O. F. Müller, 1774)	1											1	1	0.8
10. Orculidae														
Orcula dolium (Draparnaud, 1801)	3										3		3	2.5
11. Punctidea														
Punctum pygmaeum (Draparnaud, 1801)	5								1		5	1	6	5.0
12. Pyramidulidae														
Pyramidula pusilla Gittenberger et Bank, 1996	2		1				1				4		4	3.3
13. Valloniidae														
Acanthinula aculeata (O. F. Müller, 1774)	7										1	6	7	5.8
14. Vitrinidae														
Semilimax semilimax (J. Férussac, 1802)	3											3	3	2.5
15. Zonitidae														
Oxychilus depressus (Sterki, 1880)	8						2					10	10	8.3
Vitrea transsylvanica (Clessin, 1877)		1									1		1	0.8
Total	98	8	2	1	2	3	4	0	1	1	28	92	120	100.0
D (%)	81.7	6.7	1.7	0.8	1.7	2.5	3.3	0.0	0.8	0.8	23.3	76.7	100.0	
Species richness	18	7	2	1	1	1	3	0	1	0	11	16	22	
Number of families	14	5	2	1	2	1	3	0	1	1	11	12	15	

where 106 individuals of 21 species were collected. Deeper, the occurrence of Gastropoda was rather sporadic. Four species constituted more than 5% of the community: *Aegopinella pura* followed by *Faustina faustina, Oxychilus depressus* and *Acanthinula aculeata*. Below the depth of 15 cm, five gastropod species were found: *Carychium tridentatum, Daudebardia rufa, Oxychilus depressus, Punctum pygmaeum* and *Pyramidula pusilla*.

Both individual and species numbers differed considerably between the two types of preservatives used. In comparison with the formaldehyde traps (installed for 13 months), the traps filled with ethylene glycol (installed for 7 months) trapped more individuals of Gastropoda (76.6%). Eleven gastropod species were collected with the formaldehyde traps, while the traps filled with ethylene glycol caught five species more during a similar period. Among the 22 species found in both preservatives, six species were exclusively recorded in the formaldehyde traps and 11 species were exclusively recorded in the ethylene glycol traps (Table 1).

DISCUSSION

Considering the size of the study plot, the spectrum of gastropod species found was relatively rich. Ložek (1952) previously pointed out unusually abundant malacofauna of the National Nature Reserve Sivec in both individual and species numbers when collecting the molluscs from several types of surface habitats, where forest sites predominated. Among the species documented by this author, several Carpathian endemics were recorded (e.g. *Cochlodina cerata, Vestia turgida, Monacha vicina* and others). Compared to the surface, the endemic species were virtually absent in scree slopes, except for some specimens of *Perforatella dibotrion, Vestia gulo* and *Vitrea transsylvanica* trapped at the depth of 5 and 15 cm. No subterranean forms were collected. Presumably, the obligate subterranean species of gastropods are not present in Slovakia and the Western Carpathians (Kováč et al. 2014). Košel (2012) summarised all published data on the occurrence of gastropods in the caves of the Western Carpathians. There is no reference to terrestrial troglobiotic gastropod species. They were not even found among the recent or sub-fossil assemblages in the entrance of Veľká Ružínska cave (Ložek 1999, Ložek & Horáček 2007). Investigated gastropod assemblages consisted predominantly of common woodland species, associated with some hygrophilous species (*Carychium tridentatum, Trochulus hispidus*), species bound to calcareous rocks (*Daudebardia rufa, Orcula dolium, Pyramidula pusilla*), and species preferentially dwelling in debris (*Oxychilus depressus*) (Welter-Schultes 2012). *Oxychilus depressus* and *Daudebardia rufa* could also be regarded as troglophiles. Their carnivory is probably pre-adaptation for colonisation of habitats without direct influence of sunlight (Bernasconi 2004).

Most of the species (over 81%) were active 5 cm below the ground surface. Deeper, the presence of gastropods was sporadic. This pattern reflects the food preferences of woodland species that feed mostly on both living and dead plant material, and also contribute considerably to the plant litter decomposition process (Mason 1970, Fog 1979). The activity of detritivores is directly related to the decay stages of food sources, and thus they are abundant mostly near the ground surface where the largest proportion of organic matter occurs. Deeper, both parameters decrease (Gers 1998). This corresponds with the stratification of the talus deposit investigated in this study where the maximum thickness of organic matter (leaf litter with humus below) exceeds the depth of 5–15 cm. The small size of open spaces within the scree slope is another limiting factor for colonisation of most gastropods. Specimens of sizes 2–50 mm (median 7.5 mm) were observed in the samples. Medium-sized species (4.5–20 mm) with discoid shells predominated (11 species, size 2 to 8.5 mm, median 5.0).

The considerable differences in capture efficiency between formaldehyde and ethylene suggest possible species selectivity of these preservatives. As observed by Schmidt at al. (2006), capture efficiency may vary widely among different kinds of preservatives. It seems that the ethylene glycol traps attract higher numbers of gastropod species compared to formaldehyde. Mock et al. (2014) observed better efficiency of ethylene glycol for trapping saprophagous invertebrates when compared to formaldehyde. Jud & Schmidt-Entling (2008) referred to the high attraction of ethylene glycol to capture slugs. This could explain the presence of slugs (*Deroceras turcicum* and *Malacolimax tenellus*) in traps after the preservatives had been changed. Another important factor that could potentially lead to the increase of gastropods trapped is the weather (Topping & Sunderland 1992). The period when the ethylene glycol traps were set up (year 2010) was characterised by heavy rainfall.

So far, most of the authors (e.g. Tuf et al. 2008, Laška et al. 2011) who applied the methodology of subterranean traps evaluated only some arthropod taxa. Therefore, there is not enough data to generalise the knowledge of the vertical distribution of land snails below the soil surface. Moreover, to gain a more comprehensive view on the preference of gastropod assemblages to particular parts of depth gradient, it will be necessary to supplement the trapping of specimens with subterranean traps with manual collection as well as sieving the substrate from excavated pits in which the subterranean traps are to be buried. In general, this approach might be useful for other invertebrate groups with low trappability.

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