

## Species of the family Lumbricidae (Oligochaeta) found in uranium mining areas in Bulgaria

RALITSA TSEKOVA\*, LIDIA SAKELARIEVA

*Department of Geography, Ecology and Environmental Protection, Faculty of Mathematics and Natural Sciences, South-West University "Neofit Rilski", 66, Ivan Mihailov Street, 2700 Blagoevgrad, Bulgaria, ralitsa.tsekova.is@gmail.com*

**Abstract.** This paper presents the list of earthworms found in five affected by the uranium mining activities areas in Bulgaria. For the whole period of study (2011-2016) ten species belonging to six genera of family Lumbricidae were recorded. The genera *Aporrectodea* and *Lumbricus* were represented by 3 species each, and the other four by one species only. Half of all registered earthworms were endogeic. Three species (*Aporrectodea rosea*, *Eisenia fetida* and *Octolasion lacteum*) were found in all sampling sites.

**Key words:** earthworms, species composition, Rhodope and Stara Planina uranium mining regions.

### Introduction

Earthworms are one of the most important animal groups inhabiting the soil, strongly affecting both the structure and chemical properties of the soil (Darwin 1881; Zicsi 1975). Pollution with radionuclides and heavy metals influences the taxonomic composition and diversity of earthworms in the affected environments. Uranium (U) mining and milling activities produce millions of tons of material contaminated with radionuclides and heavy metals (HMs) (Suzuki *et al.* 2005). Wind and water erosion of waste piles contaminate natural and agricultural ecosystems, even when the mine is no longer active (Mahmoud *et al.* 2005), so the radionuclide and HM contamination appear to be factors influencing the earthworm diversity in these areas. Intensive U mining and milling in Bulgaria, which was performed between 1946 and 1990 with an annual production of about 645 tons, caused significant soil and water pollution. U production was ceased by Government decree in 1992, and mines and tailings were technically liquidated and gradually remediated. Some of their surroundings are still highly contaminated, and additional contamination has been observed from compromised mine and tailings reclamation. The earthworms are poorly studied despite their importance in the anthropogenically affected areas in Bulgaria. Few data have been reported until now (Tsekova & Sakelarieva 2020; Tsekova *et al.* 2022).

The aim of this study is to present and describe the species composition of earthworms inhabiting environments impacted by U mining and milling activities.

### Material and Methods

The study was carried out in the period 2011 - 2016 in 2 stages. The first stage included sampling in 3 mine sites and a control one in the area of Eleshnitsa village and in 4 mine sites and a control one in the area of the U mine "Senokos" (near the village of Senokos), all part of the Rhodope uranium mining region in Bulgaria. A total of 14 field expeditions were conducted in May and October throughout the study period. In 2011, sampling was also done in March and August, but due to the negligibly number of

Lumbricidae specimens collected and the difficult digging conditions, the study focused only on samples taken in May and October.

During the second stage, in the period 2015 - 2016, 6 new sites (3 mine and 3 control) were added and sampled in two areas of Buhovo mines (near the town of Buhovo and Seslavtsi region) and in the U mining area "Iskra" (near the village of Katina), part of the Stara Planina uranium mining region. Two field expeditions were conducted in each year of the study (in May and October).

The taxonomic determination of the collected earthworms is done in laboratories at Sofia University "St. Kliment Ohridski", Bulgaria and University of Ecology and Biology in Kragujevac, Serbia. The systematic status of the species is determined according to: Blakemore (2008), Mršić (1991), Zicsi (1982), Šapkarev (1978), and Csuzdi & Zicsi (2003).

## Results and Discussion

Ten species were identified during the study (Table 1).

**Table 1.** Checklist of the earthworm species found in all sampling sites for the whole period of study (2011-2016).

Species	Eleshnitsa		Senokos		Buhovo		Seslavtsi		Iskra Katina	
	Mine	Contr	Mine	Contr	Mine	Contr	Mine	Contr	Mine	Contr
<i>Al.chlorotica</i> Savigny, 1826			x							
<i>A.caliginosa</i> Savigny, 1826			x	x			X			
<i>A.trapesoides</i> Duges, 1828			x	x						
<i>A.rosea</i> Savigny, 1826	x	x	x	x	x	x	X	x	x	x
<i>B.rubidus</i> Eisen, 1874			x							
<i>E.fetida</i> Savigny, 1826	x	x	x	x	x	x	X	x	x	x
<i>O.lacteum</i> Orley, 1881	x	x	x	x	x	x	X	x	x	x
<i>L.rubellus</i> Hoffmeister, 1843	x	x	x	x			X	x	x	x
<i>L.meliboues</i> Rosa, 1884							X			
<i>L.terrestris</i> Linnaeus, 1758							X	x		
Number of species	4	4	8	6	3	3	7	5	4	4

The number of species varied between 3 (Buhovo) and 8 (Senokos mine). Common to all sampling sites were the endogeic *A. rosea* and *O. lacteum* (very adaptable and in some cases more able to survive in conditions of pesticide residues and heavy metals) and the epigeic *E. fetida* (one of the species most capable of accumulating heavy metals, widely used as a reference in various toxicity tests). These 3 species were the only members of Lumbricidae family found in the Buhovo sampling site. All of them belong to widespread genera, which are numerically dominant in arable lands, agricultural ecosystems, and other

ecosystems with significant anthropogenic impact on soils. The species are cosmopolitan (peregrine) due to their high adaptability and wide tolerance to many environmental factors. Four species - *A. rosea*, *E. fetida*, *O. lacteum*, *L. rubellus* were identified from the Eleshnitsa sampling sites (Table 1) and *E. fetida* was the most abundant one.

The number of Lumbricidae species at the Senokos mine sampling sites was higher (8), compared to the control site (6). This may be due to the reclamation activities carried out by spreading a humus layer, sand and natural fertilizers on the surface to create conditions for the normal development of plants and soil species. The same six species were found in both the mine sites and the control one (Table 1). *Al. chlorotica* and *B. rubidus* were found only on the territory of the mine. Both species prefer substrates rich in plant residues, such as rotting wood and other plant matter, that are not typical of the study area. For the whole study period, *A. rosea* was the most abundant species.

Seven species of earthworms (Table 1) were identified during the two-year study of the mining areas close to Sofia. All of them were found in the Seslavtsi mine sampling sites, and two of them, *A. caliginosa* and *L. meliboues*, were not found in the samples from the control site. Again, the most abundant species was *A. rosea*. The species richness in the mine sites and control sites was equal in the mine of Buhovo (3) and “Iskra” Katina (4) (Table 1).

The most abundant species for the whole study period and for all sampling sites was *A. rosea*. The species can be found among the roots of plants and in the upper soil layer between 2 and 10 cm. It was repeatedly reported for Bulgaria by various authors (Černosvitov 1937; Plisko 1963; Mihailova 1964, 1966; Shapkarev 1986; Stojanović et al. 2012, Valchovski 2014; Valchovski & Szederjesi 2016).

The dominance of species such as *A. rosea*, *E. fetida* and *O. lacteum*, which are tolerant to various environmental conditions and disturbances, can be considered as a result of the intensive anthropogenic activity accompanying the production of U. Communities that include more sensitive species are an indicator of more favorable environmental conditions (Ivask & Kuu 2005). Three of the species identified during the study (*Al. chlorotica*, *B. rubidis*, *L. meliboues*) were found only in mine sampling sites, but with low number of individuals (Tsekova & Lozev 2017, Tsekova & Sakelarieva 2020; Tsekova et al. 2022). Other species were more abundant in the samples taken from the mine sites in comparison with the control ones (Tsekova & Lozev 2017, Tsekova & Sakelarieva 2020; Tsekova et al. 2022). This is consistent with the Krivolutzkii's (1987, 1992) findings, which showed that 2 years after the Chernobyl accident (1988), the number of earthworms in the affected areas was higher than the one in the controls.

The species structure of the earthworm communities from the mines was strongly deformed - a small number of species, presented with low number of individuals (Tsekova & Lozev 2017, Tsekova & Sakelarieva 2020; Tsekova et al. 2022). Only the tolerant ones were more abundantly represented, which indicated that the environmental conditions were unfavorable for the lumbricid species.

Although the study does not report a direct adverse (lethal) effect of heavy metal contamination on earthworms, it raises the question of whether U affected ecosystems can provide optimal conditions for the development of this group of organisms? Due to the limited number of studies on the topic, additional work is needed to contribute to the knowledge on this issue.

## References

- Blakemore, R. (2008) Cosmopolitan earthworms – an eco-taxonomic guide to the species (3rd ed.). *Verm Ecology*, Yokohama, Japan, 1–757.
- Černosvitov, L. (1937) Die Oligochaetenfauna Bulgariens. *Mitteilungen aus den Königlich Naturwissenschaftlichen Instituten in Sofia*, 10: 62–92. (in German, English summary).

- Csuzdi, C. & Zicsi, A. (2003) Earthworms of Hungary. *Pedozoologica Hungarica* No1. Natural History Museum and Hungary Academy of Sciences, Budapest, 271 pp.
- Darwin, C. (1881) *The formation of vegetable mould through the action of worms*. J. Murray, London, 365 pp.
- Krivolutzkii, D. (1987) Radiation ecology of soil animals. *Biology and Fertility of Soils*, 3: 51–55.
- Krivolutzkii, D., Pokarzhevskii, A. & Viktorov, A. (1992) Earthworm populations in soils contaminated by the chernobyl atomic power station accident, 1986–1988. *Soil Biology and Biochemistry*, 24 (12): 1729–1731.
- Mahmoud, K., Leduc, L. G., & Ferroni, G. (2005) Detection of *Acidithiobacillus ferrooxidans* in acid mine drainage environments using fluorescent in situ hybridization (FISH). *Journal of Microbiological Methods*, 61(1): 33–45.
- Mihailova, P. (1964) Njakoj vidove ot semejstvo Lumbricidae (Oligochaeta) novi za faunata na Blgarija. *Annuaire de Université de Sofia*, 57: 163–169.
- Mihailova, P. (1966) Dzdovni cervi Lumbricidae (Oligochaeta) v Trakija. Fauna na Trakja, *Bulgarian Academy of Science*, Sofia, 3: 181–200.
- Mrsić, N. (1991) *Monograph on earthworms (Lumbricidae) of the Balkans*. Slovenian Academy of Sciences and Arts, Ljubljana, 755 pp.
- Plisko, G. (1963) Materialien zur Kenntnis der Regenwürmer (Oligochaeta, Lumbricidae) Bulgariens. *Fragmenta Faunistica*, Warsawa, 10: 425–440.
- Sapkarev, J. (1978) Kišne gliste Jugoslavije. Sadašnja taksonomska proučenost i njihova dalja istraživanja. *Biosistematika*, 4: 293–304.
- Sapkarev, J. (1986) Earthworm fauna of Bulgaria (Oligochaeta: Lumbricidae). *Fragm Balc.* 13: 77–94.
- Stojanovic, M., Tsekova, R. & Milutinovic, T. (2012) Earthworms (Oligochaeta: Lumbricidae) of Bulgaria: Diversity and Biogeographical Review. *Acta zoologica bulgarica*, Supplement 4: 7–15.
- Suzuki, Y., Kelly, S. D., Kemner, K. M., & Banfield, J. F. (2005). Direct microbial reduction and subsequent preservation of uranium in natural near-surface sediment. *Applied Environmental Microbiology*, 71(4): 1790–1797.
- Tsekova, R. & Lozev, R. (2017) The earthworm study from closed uranium mining facilities in Buhovo Region, Bulgaria. *Proceedings ISEM7*: 145–152.
- Tsekova, R. & Sakelarieva, L. (2020) Species Structure of the Earthworm communities (Lumbricidae) in the grounds of two liquidated Uranium Mines (Senokos and Eleshnitsa) in Bulgaria. *Ecologia Balkanica*, Special Edition: 165–179.
- Tsekova, R., Sakelarieva, L. & Varadinova, E. (2022) Impact of uranium mining activities in two abandoned mines (Western Bulgaria) on the earthworm communities. *North-Western Journal Of Zoology*, Article No.: e221301, 18 (1): 17–23.
- Valchovski, H. (2014) Diversity of earthworms (Oligochaeta: Lumbricidae) in Sofia Plain, Bulgaria. *ZooNotes*, 59: 1–9.
- Valchovski, H. & Szederjesi, T. (2016) New and additional records of earthworms (Oligochaeta: Lumbricidae) from Bulgaria: First finding of endemic species *Cernosvitovia munteniana* on the Balkan Peninsula, *North-Western Journal of Zoology* 12(2): 356–360.
- Zicsi, A. (1975) Zootische Einflüsse auf die Streuzersetzung in Hainbuchen-Eichnewald Ungarns. *Pedobiologia* 15: 432–438.
- Zicsi, A. (1982) Verzeichnis der bis 1971 beschriebenen und revidierten *Taxa der Familie Lumbricidae*, *Acta Zoologica Hungarica*, 28: 421–454.