ECOLOGIA BALKANICA
2019, Special Edition 2 pp. 165-179

Species Structure of the Earthworm Communities (Lumbricidae) in the Grounds of Two Liquidated Uranium Mines (Senokos and Eleshnitsa) in Bulgaria

Ralitsa V. Tsekova*[*], *Lidia G. Sakelarieva

South-West University "Neofit Rilski", Faculty of Mathematics and Natural Sciences, Department of Geography, Ecology and Environmental Protection, 66 Ivan Mihailov Street, 2700 Blagoevgrad, BULGARIA
[*]Corresponding author: ralvir@abv.bg

Abstract. The soil earthworm communities (Lumbricidae) in the abandoned uranium mines Eleshnitsa and Senokos, located in the southwestern part of Bulgaria, were studied in 2011 – 2016. The sampling areas have been polluted to varying degrees with heavy metals and radionuclides and disturbed by different mining and reclaiming activities. The species structure of the communities has been determined by the species composition, the number of species, the total abundance, and some diversity indices (species richness, evenness, dominance, total diversity). The number of species identified in the sampling sites is equal (Eleshnitsa mine) or higher (Senokos mine) than the one in the control sites. The total abundance of the earthworm communities is higher in the control areas and in the autumn samples. The number of immature worms is higher than that of the mature ones and more abundant in the autumn. Higher earthworm diversity is evaluated for Senokos mine and it seems that it is under the influence of many factors (environmental characteristics, mining and reclaiming activities), not only the pollution.

Key words: earthworms, diversity, uranium mines, Bulgaria.

Introduction

Soil and its characteristics are key factors that determine biodiversity in terrestrial ecosystems. Soil has abiotic components, and live elements that altogether create the soil environment (COLEMAN *et al.*, 2004). As soil organisms, the earthworms (Lumbricidae) take part in mixing the humus with the mineral soil and provide the soil fertility by enhancing macroporosity, humidification, and mineralization of the organic matter (FRANCIS & FRASER, 1998). Soil earthworm communities encompass a wide range of organisms performing various functions that regulate soil physical properties and chemical processes (STINNER & HOUSE, 1990; LAVELLE *et al.*, 1997). Invertebrates living in soil have often been considered as indicators for soil condition (status) due to their ecological demands (LAVELLE & SPAIN, 2001; GARCIA-RUIZ *et al.*, 2009) and the earthworms are among the invertebrates used most often (FRAGOSO *et al.*, 1999; HOLE *et al.*, 2005). Their popularity as indicator organisms is based on their close connection to the land, limited locomotion, ease of determination and sensitivity to the chemical and physical characteristics of the soil environment

Union of Scientists in Bulgaria – Plovdiv
University of Plovdiv Publishing House

(Paoletti, 1999; Tischer, 2009) and to different human activities, including the anthropogenic pollution (Christensen *et al.*, 1987; Paoletti, 1999; Garbeva *et al.*, 2004). Changes in abundance, biomass, or species richness of natural communities are common ecological endpoints to identify point-sources of soil pollution in the field monitoring (Spurgeon & Hopkin, 1999; Nahmani & Lavelle, 2002; Dunger & Voigtländer, 2005; Vandecasteele *et al.*, 2004). The tolerance of the earthworms to highly metal contaminated soils and the capacity to accumulate elevated concentrations of heavy metals in their tissues led to their use as sentinel species (Lukkari *et al.*, 2004).

Uranium (U) and heavy metals pollution (HMs) of the ecosystems resulting from mine activities affect the individuals, populations, biotic communities and ecosystems as a whole. Bulgaria was one of the biggest producers of U in Europe till 1992 when the U production and milling was ceased by a decree of the Bulgarian government.

The important role of earthworms as bioindicators in a polluted with heavy metals soil is mainly proved in a laboratory setting and for the aim of this investigation, any related reports are missing. There are some data referring the effects and consequences of the two abandoned U mines Eleshnitsa mine and Senokos mine, but they regard the soil characteristics, the plant and bacterial communities (Bogoev *et al.*, 2010; Kenarova *et al.*, 2010; 2014; Radeva *et al.*, 2013; Boteva *et al.*, 2015) and the water condition in the impacted rivers around the mines (Stoyanova *et al.*, 2014, Kolev *et al.*, 2014). The purpose of this study is to assess the species structure of the earthworm communities exposed to long-term U and HMs contamination in the territory of Eleshnitsa and Senokos mines.

Material and Methods

Study areas. The abandoned uranium mines Eleshnitsa (41°51′18.0″ N; 23°38′13.7″ E) and Senokos (41°49′53.0″ N; 23°13′ 11.8″ E) are located in the southwestern part of Bulgaria. The mining operations in Eleshnitsa had been conducted in a conventional underground manner (Fig. 1), while Senokos was an open-cast mine (Fig. 2). Nevertheless, during the operation of the mines and later (since 1992) as a result of the compromised rehabilitation large amounts of mine wastes have been dispersed in the surrounding areas by both surface erosion and wind action (Senokos mine) and by water effluents draining the mine galleries (Eleshnitsa mine).

Experimental design, Sampling and Identification. The sampling took place in May and October from 2011 till 2016. The specimens were collected by digging and hand sorting the 0.25x0.25 m blocks, as well as by turning over rocks, debris, and logs. The sampling sites were located in front of some of the the entrances of the galleries of Eleshnitsa mine and at different spots in the territory of Senokos mine. The control sites were chosen to be far enough from the mines. The Eleshnitsa sampling area contains 3 sampling sites in the territory of the mine (mine gallery 31 Sps, mine gallery 53 Sps and Sps WT close to the Waste water treatment plant) and one control site SpsBE. The Senokos sampling area contains 4 sampling sites (Sps 1, Sps 2, Sps 3 and Sps 4) and a control sampling site Sps 22. In order to get a better idea of a sampling site, 5 subsamples were collected while walking in a zigzag pattern across the area of the field. The approximate depth of 25 cm was chosen to reach all the important soil layers. The worm materials from all the subsamples collected from one site were mixed and filled in sample bags (collectors). The collectors from the different sampling sites were placed separately, labeled to identify and transported to the laboratory. There the earthworms were cleaned, then killed in 70% ethanol, fixed in 4% formalin solution, and stored in 90% ethanol. The identification and nomenclature of taxa were made in the laboratories of Sofia University and University of Kragujevac (Serbia), by R. Tsekova and M. Stojanovic, according to Blakemore (2008), Mršić (1991), Zicsi (1982), Šapkarev (1978), and Csuzdi & Zicsi (2003).

The species structure of the earthworm communities has been determined by the species composition, the number of taxa (S), the frequency of occurrence (pF%), the Sorenson's similarity index (QS%) (SORENSON, 1948), the total abundance (N), and some diversity indices: Margalef index of species richness (d) (MARGALEF, 1958), Simpson index of dominance (c) (SIMPSON, 1949), Pielou index of evenness (e) (PIELOU, 1966), Shannon index (H) of total diversity (SHANNON & WEAVER, 1963). The seasonal distribution of the mature and immature earthworms has also been determined.

Results and Discussion

Species composition and number of species (S)

Eight earthworm species (*Allolobophora chlorotica, Aporrectodea caliginosa, A. trapesoides, A. rosea, Bimastos rubidus, Eisenia fetida, Octolasion lacteum, Lumbricus rubellus)* belonging to six genera of the Lumbricidae family were identified during the study. The genus *Aporrectodea* is represented by the highest number of species – three. The number of species found in all the sampling and control sites for the whole period of study varied between 2 and 7 (on average 4), which corresponds with the statement of EDWARDS & BOHLEN (1996), that the earthworm diversity of all the habitat types ranges from 1 to 15 species (Fig. 3).

One and the same species were found in the territory of Elesnitsa mine and in the control site – *A. rosea, E. fetida, O. lacteum, L. rubellus.* The number of species (S = 4) remained constant for the whole research period both in the spring and autumn samples with two exceptions only (May 2013 and May 2015) (Fig. 3).

All eight species identified during the study were observed in the Senokos mine sampling sites, and two of them*: Al. chlorotica* and *B. rubidus* were found only there. The number of species ranged – between 3 (May 2014) and 7 (October 2012), and in all the years (except in 2011) it was larger in the autumn samples. The number of species from the Senokos control site was six (*A. caliginosa, A. trapesoides*, *A. rosea, E. fetida, O. lacteum, L. rubellus*), and it ranged between 2 and 5. In all cases except in May 2013 the species richness is higher in the mine sampling sites than in the control ones (Fig. 3). The reason may due to the reclamation activities following the liquidation of the mines, done by spreading a humus layer, sands and natural fertilizers on the surface to create conditions for the normal development of the plant and soil species.

For the whole period of study *E. fetida* is the most numerous species in the area of Eleshnitsa mine as well as in the control site both in the spring and autumn samples (Fig. 4).

Two species dominated in numbers in the area of Senokos mine and the control site: *A. rosea* and *O. lacteum.* They were more abundant in the control site both in the spring samples and especially in the autumn ones (almost twice) (Fig. 5). Two other species were present in large numbers in the Senokos control sampling site: *E. fetida* in the spring and *L. rubellus* in the autumn. *B. rubidus* was presented by 2 adults only (Fig. 5). The species is an epigeic species which inhabits and feeds in the litter and organically enriched surface layers of soil (HENDRIX, 1995). The reason for the low number of individuals of the species is its preferences to substrates rich in organic material, such as rotting wood and other plant matter, compost, peat, and manure, which are absent on the territory of the mines.

The values of the frequency of occurrence index show that in Eleshnitsa sampling area all the species identified were constant components of the earthworm communities with very high frequency of occurrence (pF = 100% or pF = 91,6%) both in the mine and control sampling sites.

In Senokos sampling area two species occurred very often both in the mine and control sampling sites: *A. rosea* (pF = 100%) and *O. lacteum* (pF = 75%). *E. fetida* and *L. rubellus* have also high values of pF, varying between 58% and 67% in the mine and control sites. The species *A. caliginosa* (pF = 57%) and *A. trapesoides* (pF = 100%) were constant in the mine samples, but quite rare in the control ones (pF = 25%). The two species that were found only in the territory of the mine have different values of the frequency of occurrence index: pF = 58% for *Al. chlorotica* and *B. rubidus* was the rarest species (pF = 8,3%).

Fig. 1. View from Eleshnitsa mine.

Fig. 2. View from Senokos mine.

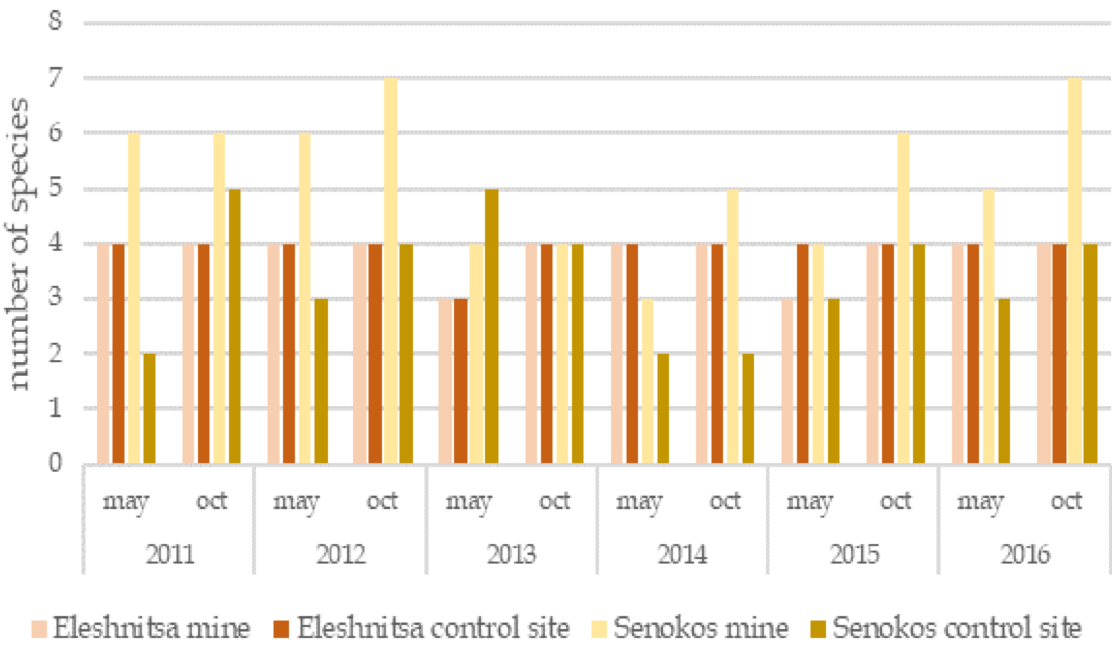

Fig. 3. Number of species collected from all the sampling sites and for the whole period of study (2011-2016).

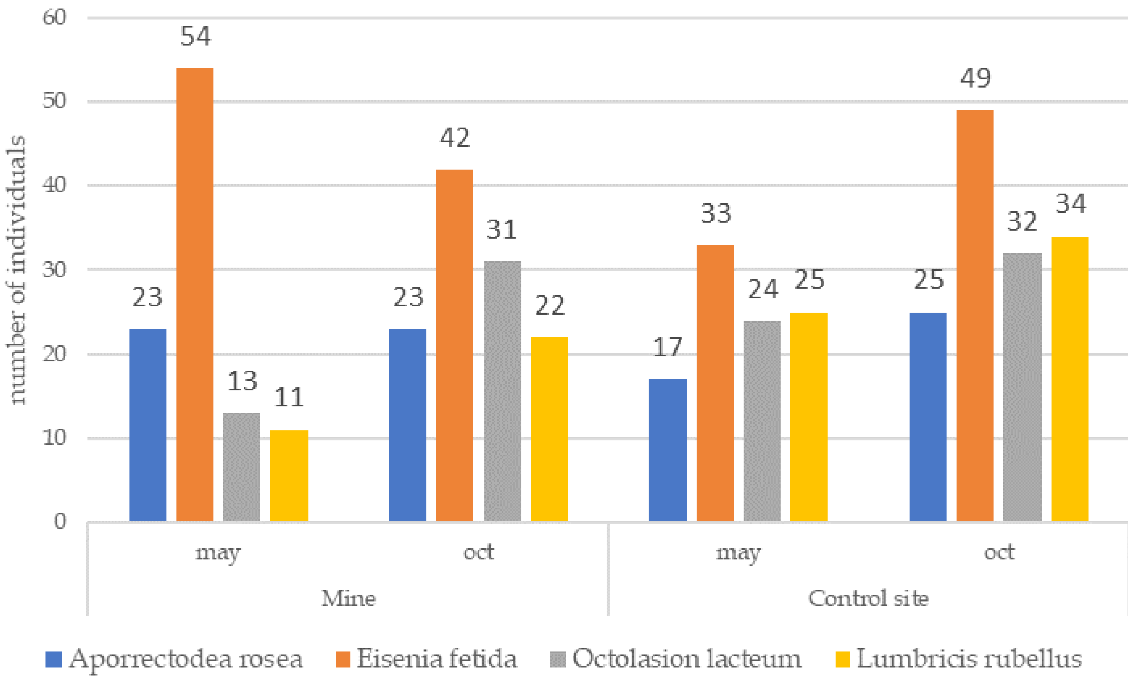

Fig. 4. Number of the adult individuals of the earthworm species collected from the Eleshnitsa mine and the control site (2011-2016).

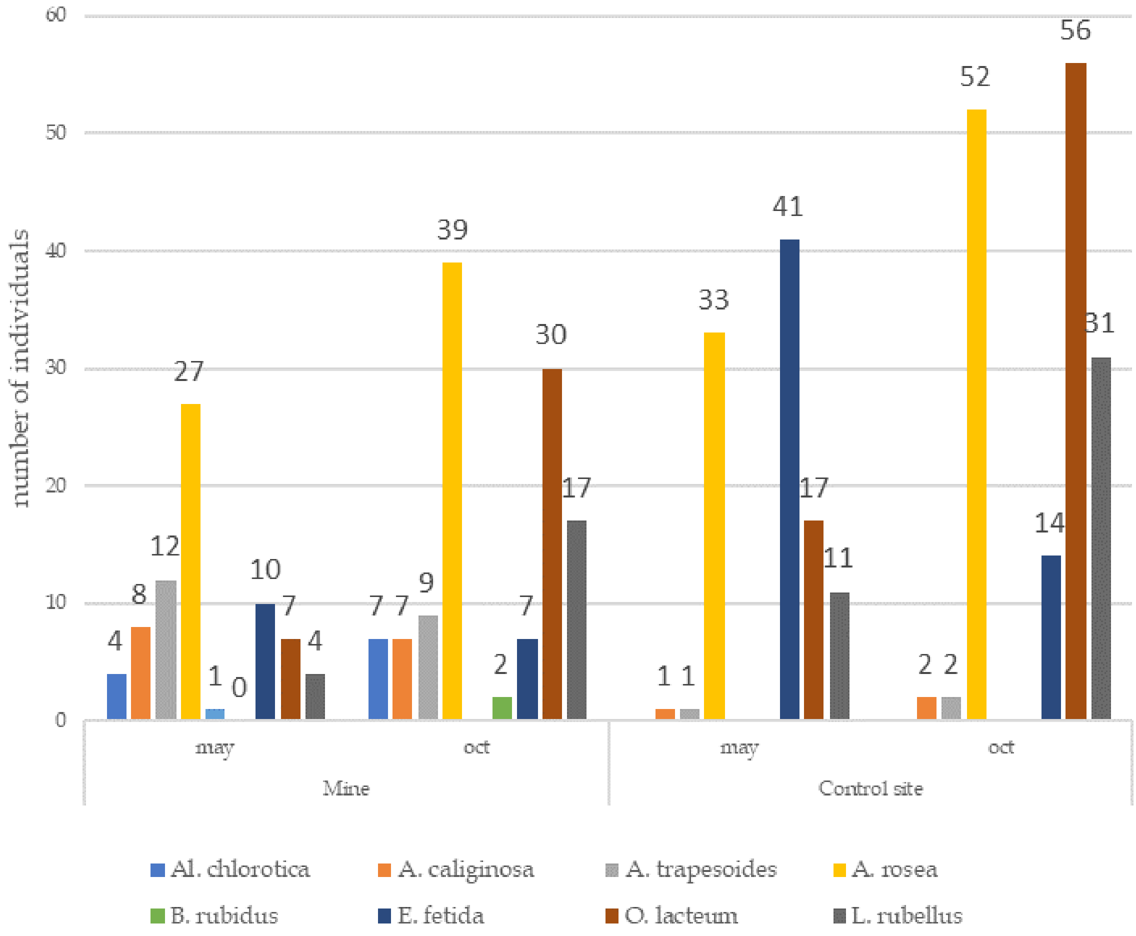

Fig. 5. Number of the adult individuals of the earthworm species collected from the Senokos mine and the control site (2011-2016).

From all the species found *A. rosea, O. lacteum, L. rubellus* and *E. fetida* were the most numerous and with the highest values of the frequency of occurrence index. They belong to four widespread genera of earthworms which are numerically dominants in arable land, agro-ecosystems and other ecosystems with considerable anthropogenic impacts on soils. The taxa are cosmopolitan around the world, because of their high adaptability and wide tolerance to many of the environmental factors.

There is complete similarity of 100% between the species composition of the earthworm communities from the Eleshnitsa mine and control sites and very high similarity (QS = 85%) between the species composition of the communities from Senokos mine and control sites. The similarity between the earthworm communities from both sampling areas (Eleshnitsa and Senokos) is also comparatively high (QS = 66%) although they are situated at a distance and the environmental conditions are different.

Total abundance

A total of 971 individuals were collected from Eleshnitsa sampling area, 462 from the territory of the mine and 509 from the control site, 458 of all individuals were mature and 513 immatures. The abundance of the earthworm communities from the mine and control sites was equal (2013, 2014, 2015) or larger in the control site (2011, 2012, 2016) (Fig. 6).

The number of the individuals collected from Senokos sampling area was 902, 432 from the territory of the mine and 470 from the control site. The number of mature and immature individuals was equal – 451. The abundance of the communities from the mine sampling site was equal (2011), or higher (2012, 2014), or lower (2013, 2015, 2016) than that of the communities from the control site. (Fig. 7) The total abundance of the earthworm communities from both sampling areas had the highest values in the last 2 years (2015, 2016) (Fig. 6 and 7).

Mature earthworms were more abundant in the autumn samples (Fig. 8). The number of immature worms was more abundant in the autumn and the values increase with 25 to 50 % (Fig. 9). Changes in the number of immature earthworms over the time seemed to be in agreement with Watanabe & Tsukamoto (1976) who found that immature worms and cocoons were recorded mainly in autumn, being not affected by the high and low temperatures in the summer and winter, respectively. Juveniles made up the majority of individuals from most of the samples and their proportional abundance (relative to adults) was similar across the sampling sites.

Diversity indices

The values of Margalef index of species richness from Eleshnitsa sampling area (mine and control) are relatively close, which corresponds to the one and the same number of species found in the whole investigated area.

Analysis of data revealed that maximum species richness in term of Margalef index was found in Senokos mine. Senokos mine is the sampling area where all the eight identified in the study earthworm species occurred. The values of the index range from 0.26 (Senokos control site, May 2011) to 1.39 (Senokos mine, Oct. 2012) (Fig. 10). The lower values at the Senokos control site may due to different agricultural human activities, located far enough from the mine, where the control site is situated.

Dominance is inversely proportional with diversity. The Simpson index varies from 0.18 (Senokos mine, May 2011) to 0.82 (Senokos control site, Oct. 2012, where from 22 individuals collected, 17 were belonging to two species *A. rosea* – 8 ind. and *O. lacteum* – 9 ind.*)*. Most of the values are lower or about 0.4 for all the sampling sites in both mines (Fig. 11). The only exceptions are from Senokos control site in May 2011, Eleshnitsa mine and Senokos control site in October 2012, Senokos control site in May and October 2014 and Elesnitsa mine in May 2015. The highest values of the Simpson index correspond to numerically close values of the Pielou index (Fig. 12). This is relatively rare and happens in cases of reduced number of taxa, represented by single individuals. The Pielou index varies between 0.62 (Eleshnitsa mine, October 2012) and 1 (Senokos control site, May 2014). The earthworm communities in Eleshnitsa control sampling site have the lowest variation in the values of both Simpson and Pielou indices, which corresponds with the more stable environmental conditions there.

Shannon diversity index reports the species richness and evenness, that's why it may varies in extended ranges. The calculated values of the diversity index lie between 0.94 (Senokos control site, May 2011 and October 2014, when the species richness was very low (S = 2) and 2.5 (Senokos mine, May 2011 and October 2016), when the number of species was very high (S = 7), and the species were represented by almost equal number of individuals (Fig. 13). The only exception with short range of the diversity values is evaluated for Eleshnitsa control site –

from 1.5 to 1.98, which again confirms the stable environmental conditions on this sampling area. Soil properties are likely to be very influential in determining the earthworm activities. The results are consistent with the findings of HOLLAND (2004) who showed that there is relationship between the environmental conditions, especially the soil structure (texture) and the earthworm biodiversity. The variations of the values of Shannon index are probably result of levels of HMs and the soil texture (sandy soils). Sandy soils are often dry, nutrient deficient and fast-draining. They have little (or no) ability to transport water from deeper layers through capillary transport, that makes the earthworms less numerous in sandy soils.

Conclusion

The earthworm species, identified in the study, were variable presented. The highest number of species were reported for one of the mine areas – Senokos. The abundance of both groups – mature and immature was higher in the autumn samples. The total abundance was higher in the control areas but there was still no clear trend in the number of species, their seasonal abundance or diversity pattern. That is probably due to the different sensitivity of the species to the environmental characteristics of the sampling areas.

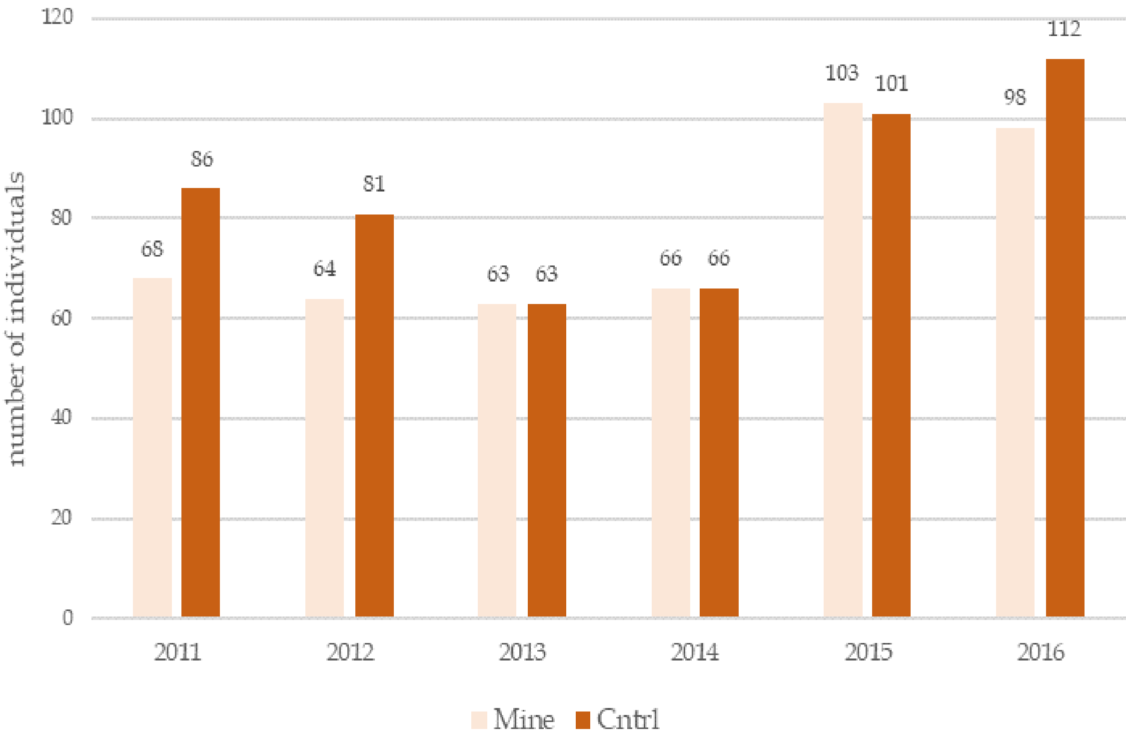

Fig. 6. Total abundance of the earthworm communities from the Eleshnitsa mine and control sampling sites in 2011-2016.

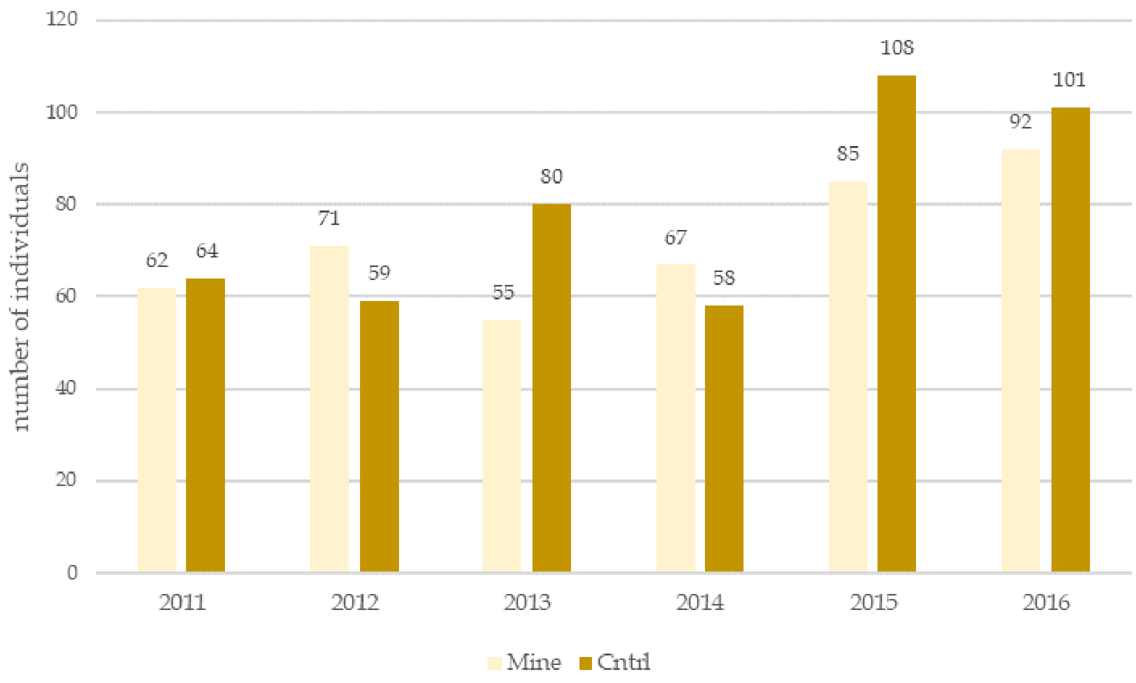

Fig. 7. Total abundance of the earthworm communities from the Senokos mine and control sampling sites in 2011-2016.

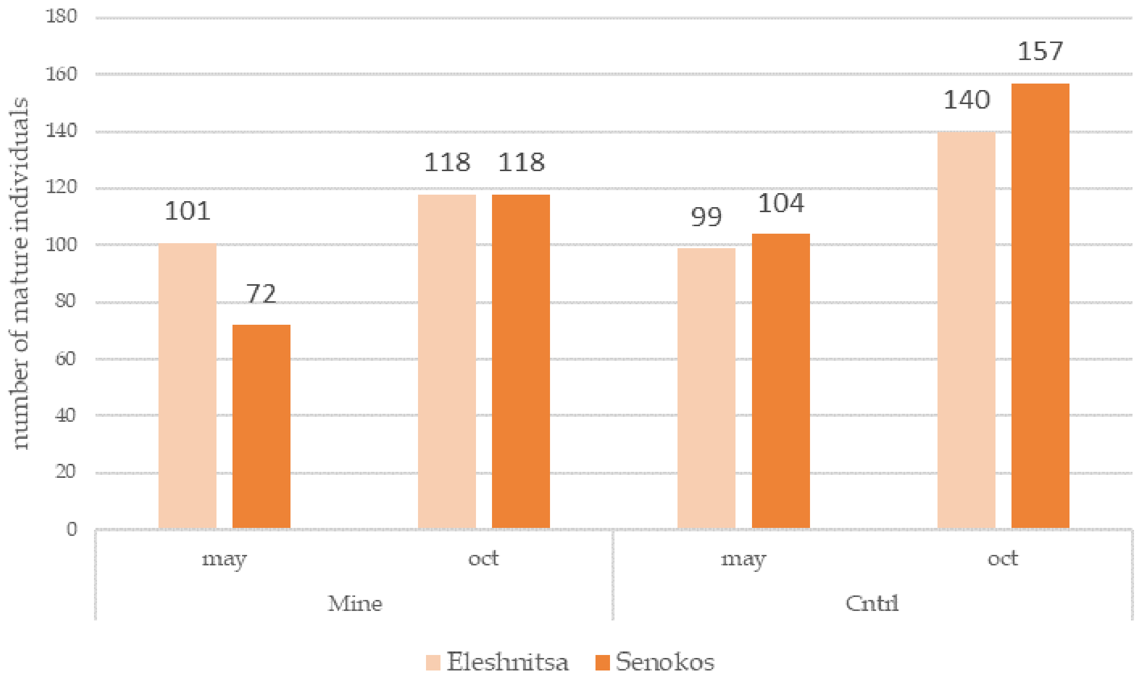

Fig. 8. Total number of mature individuals collected from all the sampling sites and the whole period of study (2011-2016).

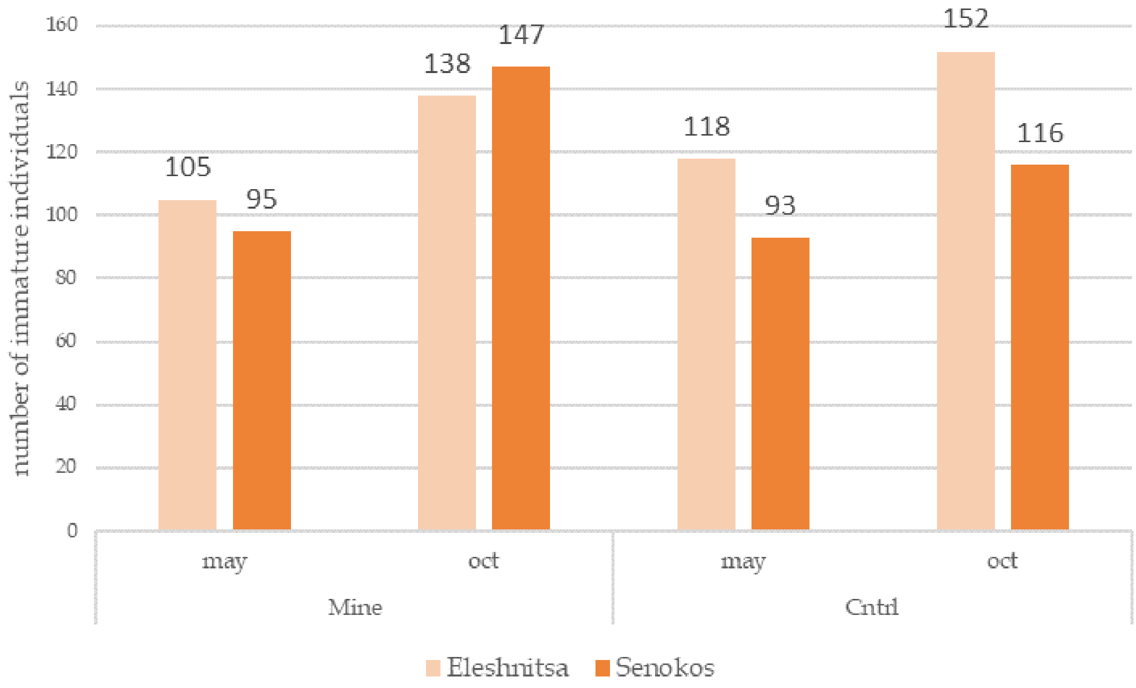

Fig. 9. Total number of immature individuals collected from all the sampling sites and the whole period of study (2011-2016).

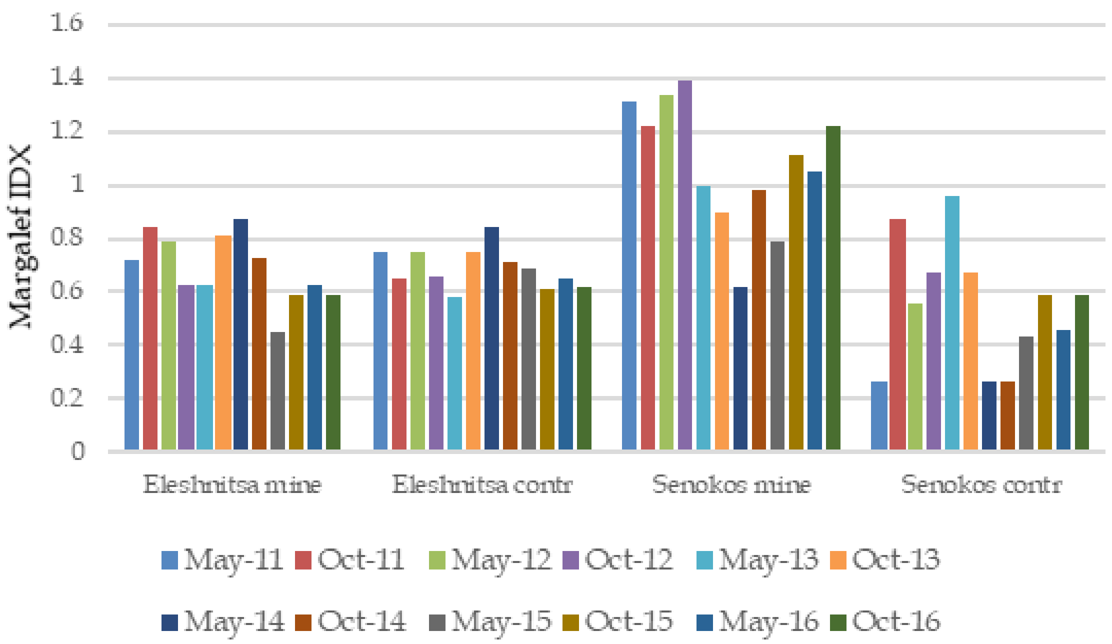

Fig. 10. Values of Margalef index from all the investigated areas and the whole period of study (2011-2016).

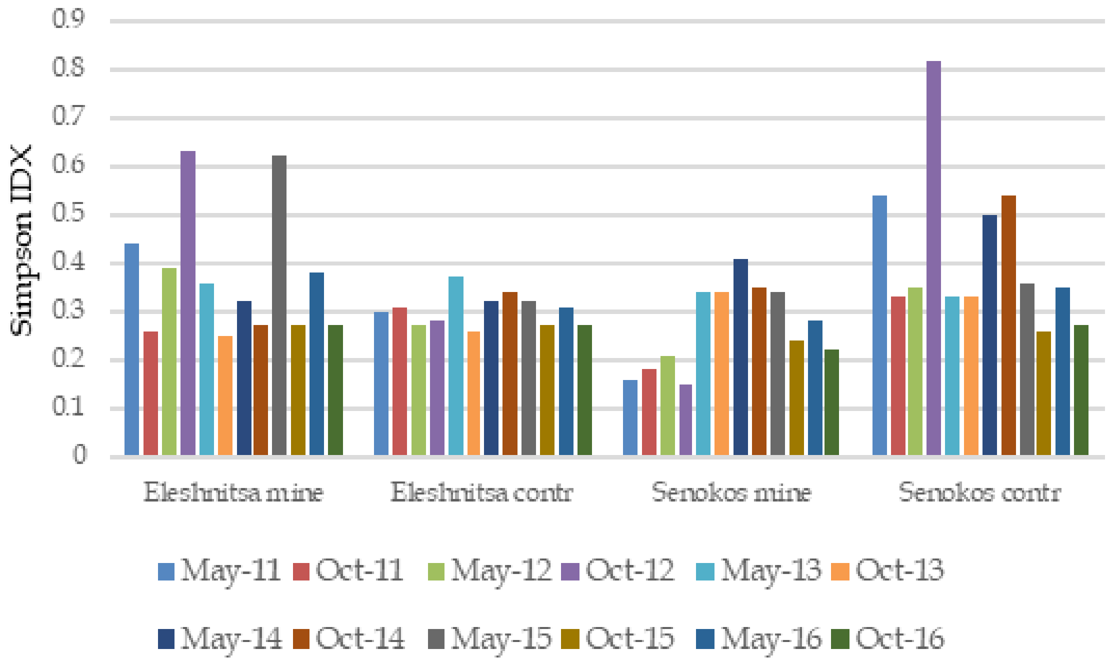

Fig. 11. Values of Simpson index from all the investigated areas and the whole period of study (2011-2016).

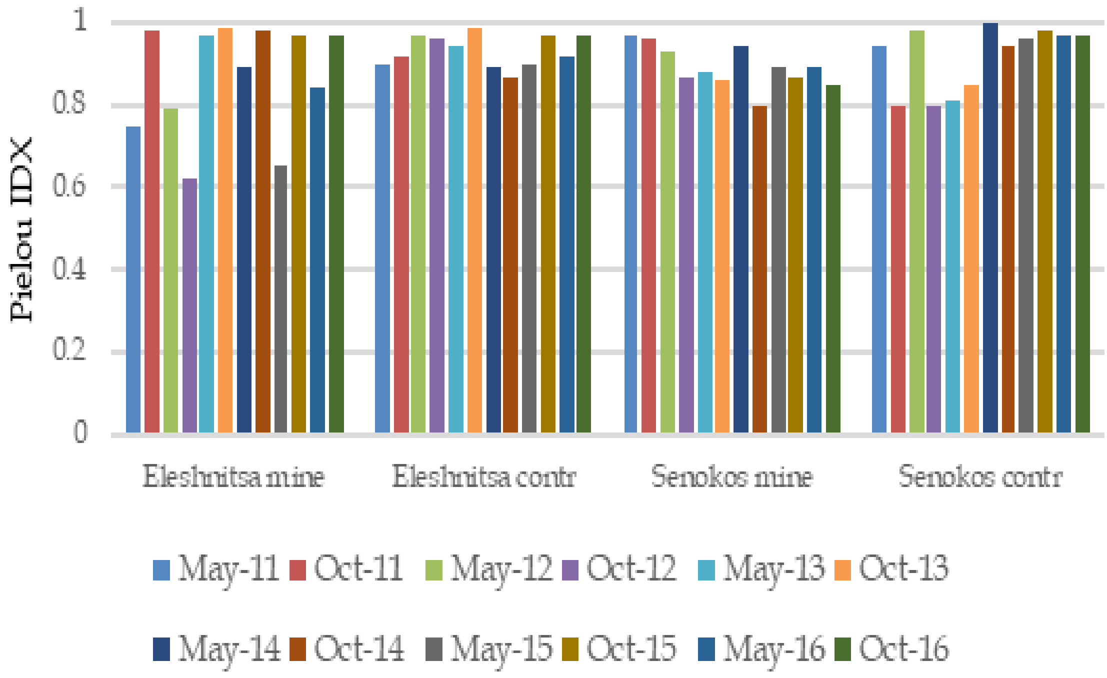

Fig. 12. Values of Pielou index from all the investigated areas and whole period of study (2011-2016).

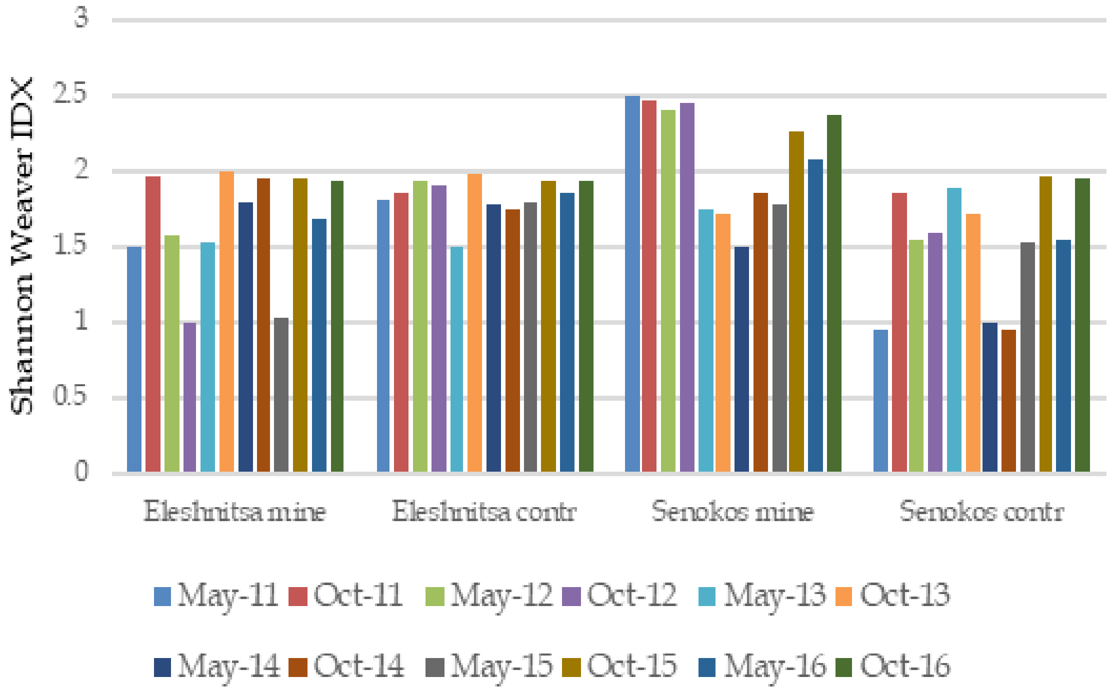

Fig. 13. Values of Shannon-Weaver index from all the investigated areas and the whole period of study (2011-2016).

This is confirmed in the study of LAVELLE & SPAIN (2001) where the response of earthworms is likely to depend mostly on environmental conditions in the field, such as site latitude, soil type, soil texture, sampling season, sampling method, etc. The soils of the investigated samplings from U areas Eleshnitsa and Senokos are heterogeneously loaded from slight to moderate levels of contamination with U and HMs (BOTEVA, *et al.*, 2015). Higher earthworm diversity is evaluated for Senokos mine (in terms of higher number of species represented by almost equal number of individuals) and lower diversity - for Senokos control site. For Eleshnitsa sampling area is opposite, higher value of diversity in general is evaluated for Eleshnitsa control site and lower diversity - for the sampling territory of the mine. The diversity seems to be under the influence of many factors as environmental characteristics, mining and reclaiming activities also, not only from the pollution with varying degrees of heavy metals and radionuclides.

Regarding all these, a clear general response of the earthworm community to the set of uranium mining activities cannot be clarified. However, the study provided some new insights about the existing effects of the anthropogenic mining activities on the soil diversity and the distribution of the earthworm species.

References

BLAKEMORE R.J. 2008. An updated list of valid, invalid and synonymous names of Criodriloidea, (Criodrilidae) and Lumbricoidea (Annelida: Oligochaeta: Sparganophilidae, Ailoscolecidae, Hormogastridae, Lumbricidae and Lutodrilidae). Available at: [annelida.net], Accessed at: 20.10.2010.

BOGOEV V., A. KENAROVA, I. TRAYKOV, R. TZONEV, R. TSEKOVA, T. STOYANOVA, S. BOTEVA, N. PARLEVA. 2010. Natural

Communities of Uranium Mining Impacted Area in The Vicinity of The Senokos Village. - *Biotechnology & Biotechnological Equipment,* SE 24:240-246.

BOTEVA S., A. KENAROVA. 2015. Functional Diversity of Microorganisms in Heavy Metal-Polluted Soils. - In: *Heavy Metal Contamination of Soils*.

BOTEVA S., A. KENAROVA, G. RADEVA, I. TRAYKOV. 2015. Effects of long-term radionuclide and heavy metal contamination on the activity of microbial communities, inhabiting uranium mining impacted soils. - *Environmental Science and Pollution Research,* 23: 6.

CHRISTENSEN O., P. DAUGBJERG, J. HINGE, J.P. JENSES, H. SIGURDARDOTTIR. 1987. Effekten af dyrkningspraksis pa regnorme og deres mulige rolle som bioindikatorer. - *Særtryk af Tidsskrift for Planteavl*, 91: 15-32.

COLEMAN D., D. CROSSLEY, P. HENDRIX. 2004. *Fundamentals of Soil Ecology*. Elsevier, New York, 386 p.

CSUZDI C., A. ZICSI. 2003. *Earthworms of Hungary*, Pedozoologica Hungarica No1, Hungary Natural History Museum and Hungary Academy of Sciences, Budapest.

DUNGER W., K. VOIGTLÄNDER. 2005. Assessment of biological soil quality in wooded reclaimed mine sites. - *Geoderma* 129: 32–44.

EDWARDS C.A., P.J. BOHLEN. 1996. *Biology and Ecology of Earthworms.* Chapman & Hall, London.

FRAGOSO C., P. LAVELLE, E. BANCHART, K.B. SENAPATI, J.J. JIMENEZ, D.L.A.M. MARTINEZ, T. DECAENS, J. TONDOH. 1999. Earthworm communities of tropical agroecosystems: origin, structure and influence of management practices. - In: Lavelle P., L. Brussaard, P. Hendrix (Eds.), *Earthworm management in tropical agroecosystems, CABI, Wallingford*, pp. 27-55.

FRANCIS G.S., P.M. FRASER. 1998. The effects of three earthworm species on soil macroporosity and hydraulic conductivity. - *Applied Soil Ecol*ogy, 10(1-2): 11.

GARBEVA P., J.A. VAN VEEN, J.D. VAN ELSAS. 2004. Microbial diversity in soil: selection of the microbial populations by plant and soil type and implementation for disease suppressiveness. - *Annual Review of Phytopathology*, 42: 243-270.

GARCIA-RUIZ R., V. OCHOA, B. VINEGLA, M.B. HINOJOSA, R. PENA-SANTIAGO, G. LIEBANAS, J.C. LINARES, J.A. CARREIRA. 2009. Soil enzymes, nematode community and selected physico-chemical properties as soil quality indicators in organic and conventional olive oil farming: Influence of seasonality and site features. - *Applied Soil Ecology,* 41(3): 305.

HENDRIX P.F. (Ed.) 1995. *Earthworm Ecology and Biogeography in North America*. Boca Raton (FL): Lewis Publisher.

HOLE G. D., J.A. PERKINS, D.J. WILSON, H.I. ALEXANDER, V.P. GRICE, D.A. EVANS. 2005. Does organic farming benefit biodiversity? - *Biological Conserv*ation, 122(1): 113.

HOLLAND J.M. 2004. The environmental consequences of adopting conservation tillage in Europe: reviewing the evidences. - *Agriculture, Ecosystems, Environment,* 103: 1–25.

KENAROVA A., G. RADEVA, I. DANOVA, S. BOTEVA, I. DIMITROVA. 2010. Soil Bacterial Abundance And Diversity Of Uranium Impacted Area In North Western Pirin Mountain. - *Biotechnology & Biotechnological Equipment*, SE 24: 469-473.

KENAROVA A., G. RADEVA, I. TRAYKOV, S. BOTEVA. 2014. Community level physiological profiles of bacterial communities inhabiting uranium mining impacted sites. - *Ecotoxicology and Environmental Safety*, 100: 226-232.

KOLEV S., N. VAN MEIR, KR. IVANOVA. 2014. Assessment of the current state of radionuclides and metals content in the surface and groundwater at the region of Eleshnitsa former uranium mine. - *Radiation Protection Journal*, 2: 35-42.

LAVELLE P., A. SPAIN. 2001. *Soil Ecology.* Kluwer Scientific Publications. Amsterdam, 684 p.

LAVELLE P., D. BIGNELL, M. LEPAGE, V. WOLTERS, P. ROGER, P. INESON, O.W. HEAL, S. DHILLION. 1997. Soil function in a changing world: The role of invertebrate ecosystem engineers. - *European Journal of Soil Bio*logy, 33(4): 159.

LUKKARI T., M. TAAVITSAINEN, A. VÄISÄNEN, J. HAIMI. 2004. Effects of heavy metals on earthworms along contamination gradients in organic rich soils. - *Ecotoxicology & Environmental Safety,* 59: 340–348.

MARGALEF R. 1958. Information theory in ecology. - *General Systematics,* 3: 36-71.

MRŠIĆ N. 1991. *Monograph on earthworms (Lumbricidae) of the Balkans I-II*, Slovenska Akademija Znanosti in Umetnosti, Ljubljana.

NAHMANI J., P. LAVELLE. 2002. Effects of heavy metal pollution on soil macrofauna in a grassland of Northern France. - *European Journal of Soil Biology*, 38: 297–300.

PAOLETTI M. 1999. The role of earthworms for assessment of sustainability and as bioindicators. - *Agriculture, Ecosystems and Environment*, 74: 137-155.

PIELOU E. 1966. Species diversity and pattern diversity in the study of ecological successions. - *Journal of Theoretical Biology, London*, 10: 370-383.

RADEVA G., S. SELENSKA-POBELL, K. FLEMMING, V. BACHVAROVA, A. KENAROVA, I. POPOV, D. VASSILEV. 2013. Bacterial Diversity at Abandoned Uranium Mining and Milling Sites in Bulgaria as Revealed by 16S rRNA Genetic Diversity Study. - *Water Air & Soil Pollution,* 224:1748.

ŠAPKAREV J. 1978. Kišne gliste Jugoslavije. Sadašnja taksonomska proučenost i njihova dalja istraživanja. - *Biosistematika,* 4: 293–304.

SIMPSON E. 1949. Measurement of diversity. - *Nature, London.* 163, 688.

SORENSEN K. 1948. A method of establishing groups of equal amplitude in plant society based on similarity of species content. - *Danske Vedensk. Selsk*, 5: 1-34.

SHANNON S., W. WEAVER. 1963. *The mathematical theory of communication*. Urbana Univ Illinois Press., 117 p.

SPURGEON D.J., S.P. HOPKIN. 1999. Seasonal variation in the abundance, biomass and biodiversity of earthworms in soils contaminated with metal emissions from a primary smelting works. - *Journal of Applied Ecology,* 36: 173–183.

STINNER B.R., G.J. HOUSE. 1990. Arthropods and Other Invertebrates in Conservation-Tillage Agriculture. - *Annual Review of Entomology,* 35: 299-318.

STOYANOVA T., Y. VIDINOVA, I. YANEVA, V. TYUFEKCHIEVA, D. PARVANOV, I. TRAYKOV, V. BOGOEV. 2014. Ephemeroptera, Plecoptera and Trichoptera as Indicators for Ecological Quality of the Luda Reka River, Southwest Bulgaria. - *Acta zoologica bulgarica*, 66(2): 255-260.

TISCHER S. 2009. Earthworms (Lumbricidae) as bioindicators: the relationship between in-soil and in-tissue heavy metal content. - *Polish Journal of Eco*logy, 57(3): 513.

VANDECASTEELE B., J. SAMYN, P. QUATAERT, B. MUYS, F.M.G. TACK. 2004. Earthworm biomass as additional information for risk assessment of heavy metal biomagnification: a case study for dredged sediment derived soils and polluted floodplain soils. - *Environmental Pollution,* 129: 363–375.

WATANABE H., J. TSUKAMOTO. 1976. Seasonal changes in size class and stage structure of Lumbricid *Eisenia foetida* population in a field compost and its practical application as the decomposter of organic waste matter. - *Revue d"Ecologie et de Biologie du Sol*, 13: 141-146.

ZICSI A. 1982. Verzeichnis der bis 1971 beschriebenen und revidierten Taxa der Familie Lumbricidae (Oligochaeta). - *Acta Zoologica Hungarica*, 28: 421–454.

Received: 05.12.2019
Accepted: 23.12.2019