ISSN 1067-4136, Russian Journal of Ecology, 2025, Vol. 56, No. 5, pp. 385–398. © Pleiades Publishing, Ltd., 2025.

Enchytraeid Communities of Birch Forests in Vicinities of the Middle Ural Copper Smelter

A. V. Nesterkov[a], *, M. I. Degtyarev[b], **, and D. V. Nesterkova[a]

[a] *Institute of Plant and Animal Ecology, Ural Branch, Russian Academy of Sciences, Yekaterinburg, 620144 Russia*

[b] *Institute of Ecology and Evolution, Russian Academy of Sciences, Moscow, 119071 Russia*

**e-mail: nesterkov@ipae.uran.ru*

***e-mail: degtyarevmi@gmail.com*

Received May 3, 2025; revised August 5, 2025; accepted September 15, 2025

Abstract—Enchytraeid communities were studied in the upper soil of birch forests in areas within the pollution gradient of emissions from the Middle Ural Copper Smelter (MUCS). In the unpolluted area, 17 species were identified (from 6 to 13 in different sample plots), the average density was 13666–44903 specimens/m^2, average (raw) biomass 1357–3699 mg/m^2. Near the smelter, population density, biomass and species richness were reduced by 80, 20, and 2.4 times, respectively. However, the diversity of communities changed only slightly, and the diversity structure of enchytraeids in the impact zone is potentially comparable to the background's. In the background zone, the highest values of density and biomass were concentrated in the upper layer (0–2 cm) of soil, corresponding to the forest litter (61 and 63%, respectively). In the impact zone, the vertical distribution of enchytraeids was different: density (59%) and biomass (61%) were redistributed to the underlying (2.1–4 cm) layer (lower part of the litter), which contained the highest concentrations of metals. Changes in the vertical distribution of enchytraeids are not thought to be related to pollution effects.

Keywords: enchytraeidae, oligochaetes, abundance, biomass, species richness, diversity, vertical distribution, industrial pollution, heavy metals

DOI: 10.1134/S1067413625600788

INTRODUCTION

Enchytraeids are small oligochaetes that play an important role in nutrient cycling and organic matter degradation in terrestrial forest ecosystems [1–4]. From an ecological standpoint, enchytraeids serve as a convenient indicator group of organisms [5, 6], suitable for assessing soil quality. Enchytraeids are also useful for monitoring chemical stress, as species-specific differences in pollutant sensitivity can lead to rapid and pronounced changes in community composition and structure [6].

Field studies of enchytraeids under metal pollution conditions are relatively few, and studies focused on point sources of pollution are scarce [7–13]. In almost all cases examined, a decrease in population density and biomass in the contaminated area was noted, and in some examples, a decrease in diversity was also observed.

It is important to note that all currently available data on enchytraeids in impact regions were obtained relatively locally (Europe), almost all in biocenoses of the same type (coniferous forests), and under similar pollution conditions (neutralization of the pH of surrounding soils). Obviously, this is not enough to form a comprehensive picture of the response of enchytraeid communities to industrial pollution.

A separate, poorly studied issue is the effect of pollution on the vertical distribution of enchytraeids. In undisturbed ecosystems, the highest population density is observed in the upper 2–3 cm of soil and decreases significantly with depth [14–16]. There is virtually no information on changes in the vertical distribution of enchytraeids near point sources of pollution. For the area of a copper smelter in Sweden, a shift of a significant proportion of the total number of enchytraeids from the upper soil layer (0–3 cm) to the underlying soil layer (3–6 cm) has been described [14]. We are not aware of any other similar studies, but the available data allowed us to formulate a testable hypothesis. Our work presents the first data for Russia on the composition and structure of enchytraeid communities in the impact region, which allows us to speak about its relevance.

Objective—To analyze changes in the population density, biomass, species structure, and vertical distribution of soil enchytraeids in birch forests of the territory exposed to pollution by Middle Ural Copper Smelter (MUCS) emissions. The hypotheses that in the birch forests of the contaminated area: (1) the population density and biomass of enchytraeids are greatly reduced; (2) species richness is reduced and the community structure is altered; and (3) the density of enchytraeids is shifted from the upper soil layer to the underlying soil.

MATERIALS AND METHODS

Characteristics of the Study Area

The study was conducted in the least elevated part of the Ural Mountains (150–400 m above sea level) in the southern taiga subzone, in the vicinity of MUCS (56.8515° N, 59.9069° E), which has been operating since 1940. The main components of the emissions are sulfur dioxide, fluorine compounds, nitrogen oxides, metals, and metalloids (Cu, Pb, Zn, Cd, Fe, and As). The annual volume of emissions in 1980 reached 225000 tons, after which it began to decline: to 148000 tons in 1990, 63000 tons in 2000, 28000 tons in 2004, and to 3000–5000 tons after 2010 [17]. The plant's activities led to the formation of a man-made geochemical anomaly with a significant accumulation of heavy metals in the original sod-podzolic soils of heavy loamy composition. In the soils of the contaminated area, the eluvial-gley process intensified, the content of exchangeable Ca and Mg decreased, the processes of organic matter destruction slowed down, and the humus state changed [17, 18]. Near the smelter, there is a marked suppression of all layers of vegetation: woody (the stock of forest stands, their density and crown density have been reduced, the proportion of dead wood has increased), grass-shrub, and grassy (species richness and abundance have been reduced) [19, 20]. Significant changes also occurred in the animal population; in particular, in the community of pedobionts in the impact zone, a sharp decrease in the abundance of most groups was noted, as well as a predominance of phytophages and zoophages over saprophages [22].

The sample plots were located in birch forests, derivatives of the original dark coniferous forests. Previously (in 2022), the vegetation condition and several soil parameters were described in the sample plots. The tree layer is dominated by silver birch (*Betula pendula* Roth), Scots pine (*Pinus sylvestris* L.) is found occasionally, and aspen (*Populus tremula* L.). The diameter of trees in the background zone is 6–30 cm, in the impact zone it is 2–30 cm. The herbaceous-dwarf layer of the background zone is dominated by *Calamagrostis arundinacea* (L.) Roth, *Aegopodium podagraria* L., *Rubus saxatilis* L., *Oxalis acetosella* L., *Asarum europaeum* L., and *Stellaria holostea* L. The impact zone at 3.7 km is dominated by *Deschampsia cespitosa* (L.) P. Beauv., *Veratrum lobelia* Bernh., and *Horsetail* L., while the rest of the plots are dominated by *Agrostis tenuis* Sibth. and *D. cespitosa*. The content of metals in the soil of the sample plots (Appendix, Table S1) corresponds to the values specified for the background and impact zones [17].

Sample Collection

Enchytraeids were collected from 10 sample plots (SPs) (Fig. 1) located to the west of the plant: 5 in the impact zone (distance from 1.9 to 3.7 km) and 5 in the background zone (15.9–20.3 km), over the course of 1 day (9/27/2024).

At each SP (20 × 20 m), 6 samples were taken, spaced at least 2 m apart. The samples were collected using a collapsible soil sampler (diameter 5 cm, cross-sectional area 19.6 cm^2). The sampler extracted from the soil was opened into two longitudinal halves, gaining access to the soil core (Fig. 2), after which the thickness of the forest litter was measured with an accuracy of 0.1 cm. Next, using two rounded spatulas and a ruler, the core was divided into 2 cm layers, and the top four layers (0–2, 2.1–4, 4.1–6, and 6.1–8 cm) were each packed into separate zip-lock bags. Immediately after collection, the soil samples were placed in cooler bags, where the temperature was maintained at 10–15°C using water ice (to prevent the samples from freezing too much). A total of 60 soil samples (240 layers) were collected.

Extraction of Enchytraeids

Enchytraeids were extracted by wet extraction without heating [22] in a climate room at a constant temperature (+15°C). Each core layer was processed separately: carefully reduced to fragments by hand and transferred to a sieve with a mesh diameter of 0.7 mm. The sieve with the sample was placed in a plastic funnel, prefilled with cold tap water. A test tube was attached to the bottom of the funnel using an elastic silicone tube, which served as a collection point for the worms leaving the soil. Extraction was carried out for 24 h, since this period is sufficient to account for most of the enchytraeids [23]. After the distillation was completed, the tube was detached and stored in a rack at +15°C. Each extraction cycle involved one complete sample (all four core layers) from each SP; thus, 4 layers × 10 SPs = 40 samples (layers) were simultaneously extracted each time. The total duration of extraction of the collected material was 6 days.

Material Processing

For the majority of the material (30 samples from the impact zone and 20 from the background zone), the species affiliation of enchytraeids was determined, and the abundance of each species (specimens) was calculated. For the remaining 10 samples from the background zone, the total population density of enchytraeids (specimens/m^2) was calculated.

Enchytraeids were identified in vivo using a light microscope according to [24]. Species described later

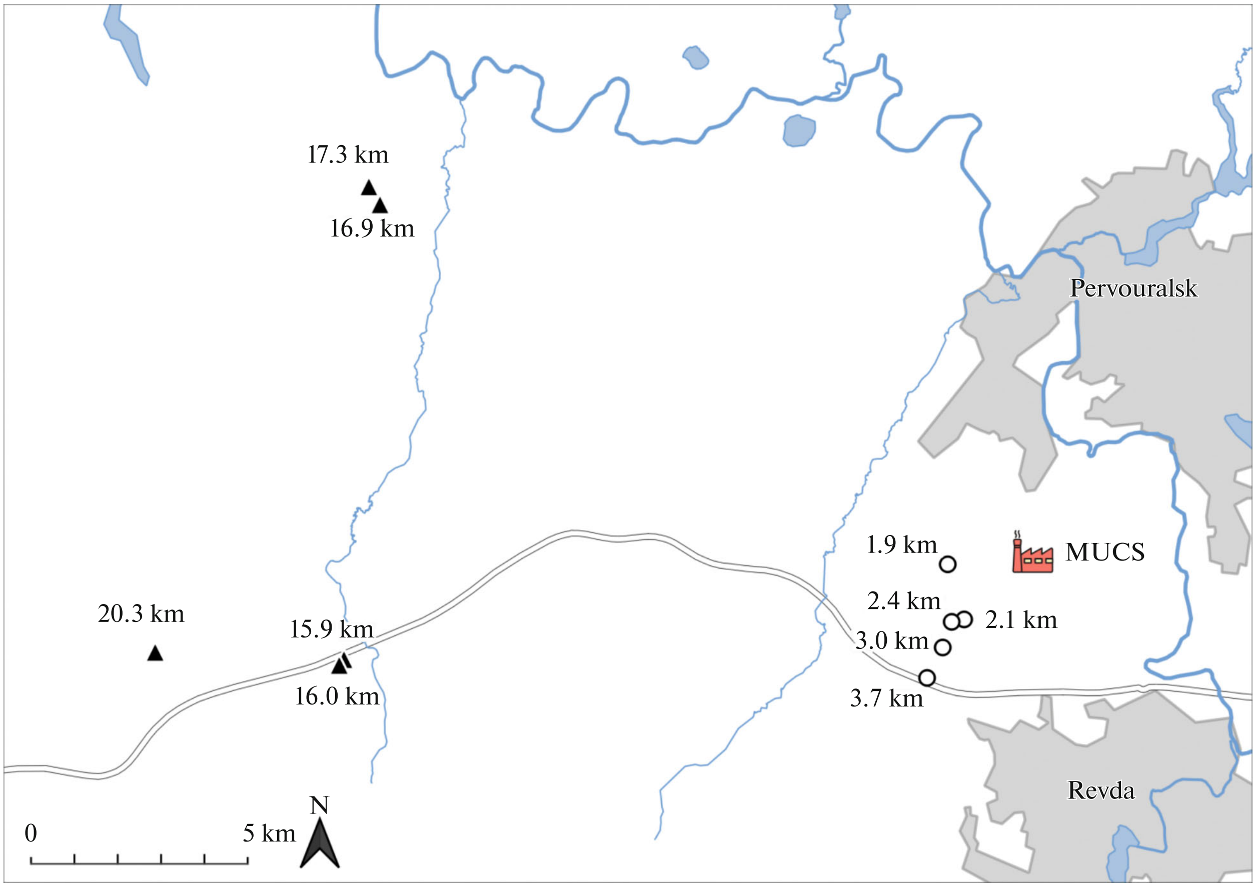


Fig. 1. Layout of sample plots relative to the source of pollution—the Middle Ural Copper Smelter (MUCS).

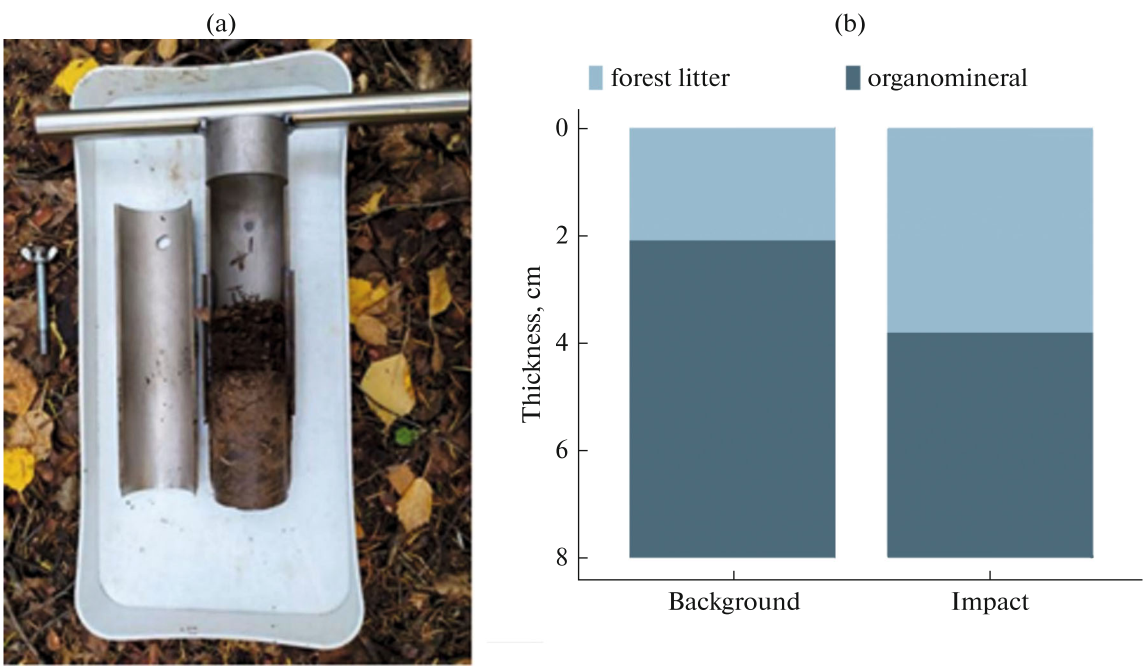


Fig. 2. General view of the soil core in a collapsible soil sampler (a) and a diagram of the relationship of soil core layers with the average thickness of the forest litter and organomineral horizon in birch forests of the background and impact pollution zones (b).

than the publication of the referenced source or missing from it were identified by comparison with the original description. The taxonomy has been brought into line with the World Register of Marine Species database [25]. For living, mature individuals, the species affiliation was established; immature individuals were in most cases identified to the genus level. The dead enchytraeids formed the group Enchytraeidae indet.

After identification and counting, all enchytraeids were preserved in a 95% ethanol solution. For each specimen, a scaled image was obtained using a binocular microscope and a digital camera. In the ImageJ program [26], the length (mm) and body area (mm^2) were measured on the images, on the basis of which the average body width of each specimen was calculated. Body volume (mm^3) was calculated using the standard cylinder formula, taking the width of the body as the diameter. The wet body mass (mg) was obtained by multiplying the volume by the average body density value for all enchytraeids (1.051 mg/mm^3). It has been shown that for different species the deviation of density from the average does not exceed 1% [27]. The method we have described allows us to determine the mass of each enchytraeid specimen, including those that were not determined to be of a particular species or those that have died. Alternative calculation methods use species-specific regression relationships of body length and body mass [28, 29] and are only applicable to a limited list of species. To obtain dry biomass values from the data we have provided, it is sufficient to apply a coefficient of 0.18, which is the standard ratio of dry to wet body mass for all enchytraeids [30].

Data Analysis

The data were analyzed in the R software environment [31]. For the density and biomass of enchytraeids, the mean ± error was calculated for the contamination zones for five SPs (for all 60 samples), as well as the occurrence of each species in the samples (for 20 samples in the background zone and 30 in the impact zone) and in the sample plots (for five SPs). Occurrence was estimated as the ratio of the number of samples (or SPs) in which the species was encountered to the total number of samples (or SPs). The litter thickness data obtained for each sample allowed us to estimate the distribution of enchytraeid density and biomass in the forest litter and the upper part of the organomineral horizon. Data layout and processing were performed using the dplyr package [32], and visualization was performed using the ggplot2 package [33], which are part of the tidyverse library set. The proportions of enchytraeid density in each soil layer and soil horizon relative to the total density were compared between contamination zones using the equality of proportions test with Yates's correction for continuity (the prop.test function in the base R package).

Size classes were calculated for logarithmic enchytraeid body mass data presented as a variation series. The boundaries of size classes, as well as weighted average class intervals, are given in the Appendix, Table S2. Diversity profiles (Hill numbers) were constructed for background and impact zones based on species lists with summarized density and biomass values (CommunityProfile function of the entropart package [34]). The profiles of potential diversity (rarefaction and extrapolation) for the background and impact zones, as well as the assessment of the completeness of the sample volumes in them, were calculated using the iNEXT function of the package of the same name [35]. The confidence interval boundaries were estimated using a permutation test (999 permutations).

The ordination of the density and biomass of enchytraeid species relative to the "pollution zone" factor was performed using the principal component method in the ggord package [36]; the model parameters (characteristic numbers and distances to centroids) were calculated using the pairwiseAdonis package [37]. Before performing the ordination, the density and biomass values for the species list in each sample were normalized using the Hellinger transformation in the labdsv package [38]. The effect size (log response ratio) for several arrangements of enchytraeid density, biomass, and species richness at the sample level was estimated using the LRRd function of the SingleCaseES package [39].

RESULTS

Enchytraeid Population Density

Enchytraeid population densities varied greatly between pollution zones. In the background zone at different SPs, the average density varied from 13666 to 44903 specimens/m^2 (Appendix, Table S3). The highest density (61% of the total) of enchytraeids was noted in the upper 2 cm of the soil core (Fig. 3a; Appendix, Table S4), although in the forest litter and organomineral horizon the density was comparable (Fig. 3b).

In the impact zone, the average density was reduced by 80 times compared to the background and ranged from 0 to 1188 specimens/m^2 at different SPs (Appendix, Table S3). The highest density (59% of the total) was recorded in the 2.1–4 cm layer, while in the 0–2 cm layer it was reduced (Fig. 3a; Appendix, Table S4). A comparison of the distribution of enchytraeids between the layers of the soil core (as a share of the total density) in the background and impact zones confirmed the differences between the 0–2 cm layers ($p = 0.0007$) and 2.1–4 cm ($p = 0.0002$) and no differences between layers 4.1–6 cm ($p = 0.8818$) and 6.1–8 cm ($p = 0.4941$). The proportion of enchytraeids in the total density in the forest litter increased in the impact zone compared to the background zone ($p =$

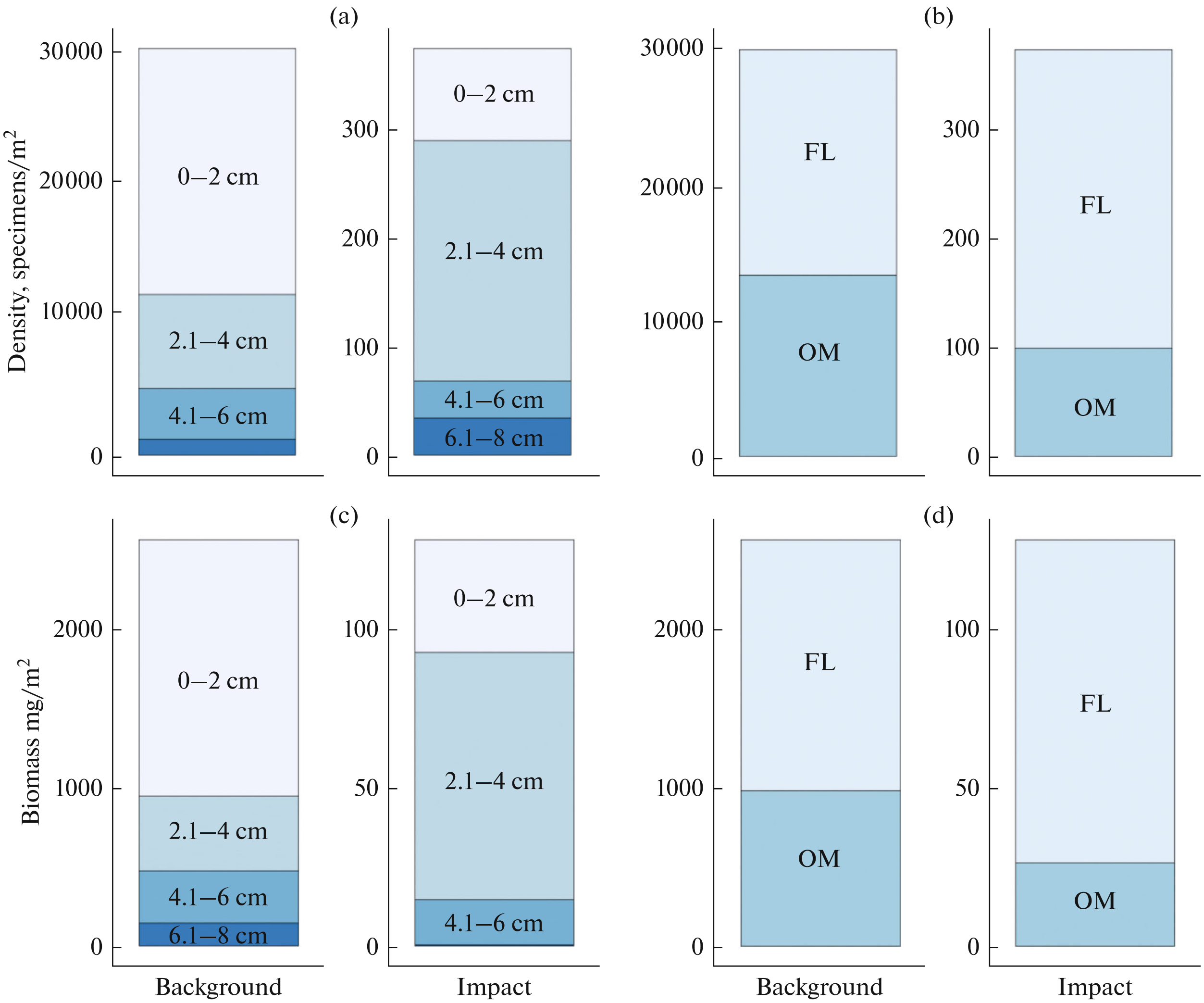


Fig. 3. Distribution of enchytraeids in soil core layers (a—population density, c—biomass) and soil horizons (b—density, d—biomass). Soil horizons: FL—forest litter, OM—organomineral. The sampling unit is the sample plot ($n = 5$).

0.0059, Fig. 3b); the proportion in the organomineral horizon in both zones was comparable ($p = 0.1413$).

A pronounced effect of the "contamination zone" factor on the density of enchytraeids was noted in all the examined soil horizons, soil core layers, and variants of combining layers (Fig. 4).

Enchytraeid Biomass

In the background zone, the average biomass of enchytraeids varied within the range of 1357–3699 mg/m^2 at different SPs (Appendix, Table S5). The highest biomass (as well as density) was noted in the 0–2 cm layer (63% of the total, Fig. 3c; Appendix, Table S6). The biomass in the forest litter was somewhat greater than in the organomineral horizon (Fig. 3g). In the impact zone, the average biomass was reduced by 20 times and varied from 4 to 424 mg/m^2 at different SP (Appendix, Table S5). The highest biomass was noted in the 2.1–4 cm layer (61% of the total, Fig. 3c; Appendix, Table S6). A comparison between the background and impact zones of the proportions of enchytraeid biomass in the soil core layers relative to the total biomass revealed significant differences in all combinations ($p < 0.0001$). A similar comparison for soil horizons revealed an increase in the proportion of enchytraeid biomass in the forest litter ($p < 0.0001$) and a decrease in the proportion of biomass in the organomineral horizon ($p < 0.0001$) in the impact zone compared to the background zone (Fig. 3g).

The effect of the "contamination zone" factor on biomass in all the considered soil horizons, soil core layers, and layer combination options was pronounced and comparable in magnitude to the effect for density (Fig. 4).

The distribution of enchytraeids by size classes was close to lognormal (see Appendix, Table S2; Fig. 5): the largest number of individuals were included in the

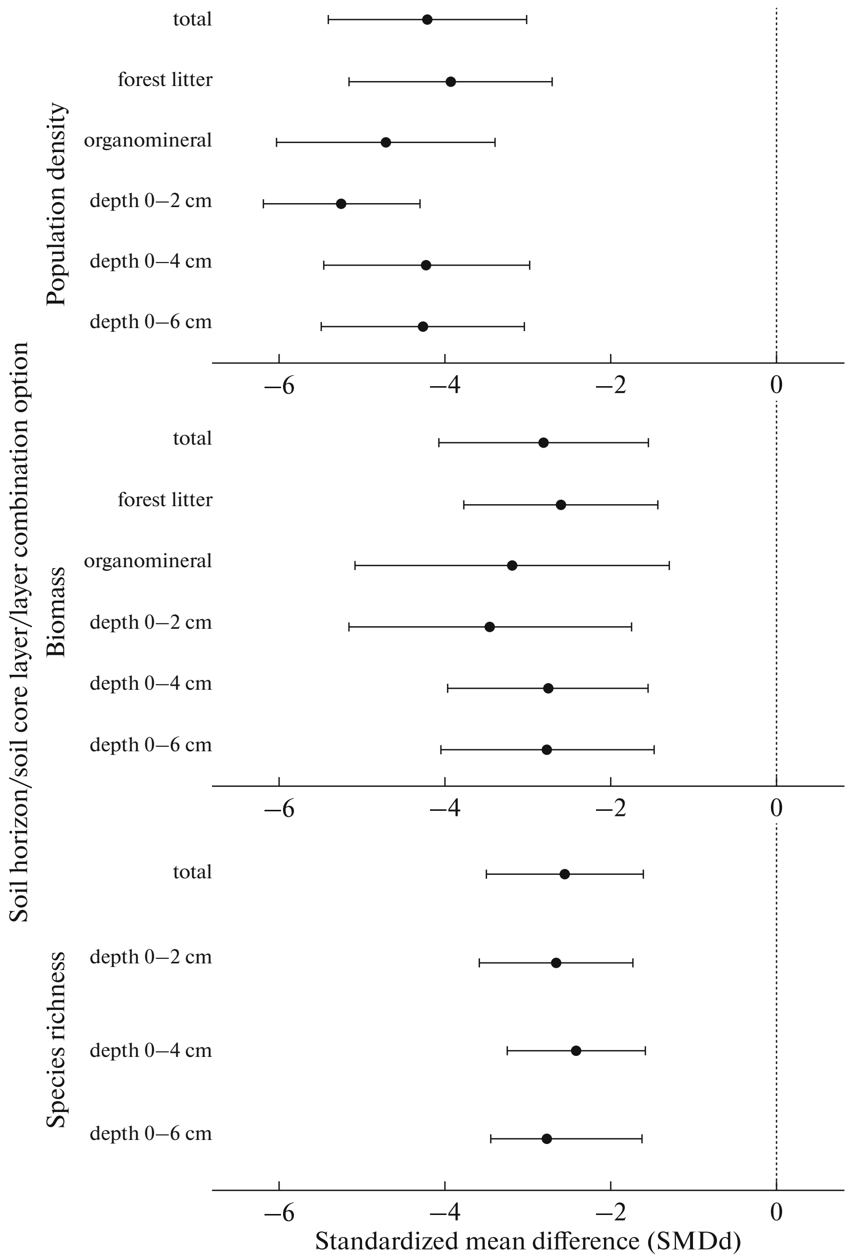


Fig. 4. The effect size of the “pollution zone” factor on the population density, biomass, and species richness of enchytraeids in different soil horizons, soil core layers, and layer combination options (depth 0–4 cm and 0–6 cm). The sampling unit is the sample plot ($n = 5$).

central classes (2, 3, 4), the smallest were included in the extreme ones (1, 6, 7). In the impact zone (Fig. 5b), despite the differences in the ratio of class shares, the range of changes in their values generally coincided with those described above (Fig. 5a).

Diversity of Enchytraeids

In total, 19 species of enchytraeids were identified in the study area (Table 1). In the background zone, 17 species were found (from 6 to 13 at different sites), among the dominant ones were: *Buchholzia appendiculata* (Buchholz, 1862), *Enchytronia parva* Nielsen & Christensen, 1959 s.l., and *Cognettia sphagnetorum* (Veydovsky, 1878) s.l. (see Appendix, Table S3). Seven species were identified in the impact zone, and their number at the SP increased with distance from the smelter: from absence at the nearest (1.9 km from the plant) to five species at the most distant (3.7 km). *Enchytronia parva* and *Fridericia ratzeli* (Eisen, 1872) were predominant (Appendix, Table S3).

For most species recorded in the impact zone, a decrease in occurrence was noted. Only *Enchytraeus buchholzi* Vejdovský, 1878 and *Fridericia ratzeli* showed an incidence similar to that of the background zone (see Table 1). In general, the effect of the "pollution zone" factor on species richness is clearly pronounced (see Fig. 4).

The structure of enchytraeid communities in the background and impact zones differs only at the level of species richness and abundance of rare species ($q <$ 0.9), while the "cores" of communities (i.e., the number of the most abundant species and the ratio of their numbers) are similar (Fig. 6a). The cumulation-extrapolation curve of potential species richness in the background zone is close to saturation in the region of 20 species (Fig. 6b). In the impact zone, assessing potential diversity is difficult due to the reduced abundance of enchytraeids. However, the values of the actual cumulative diversity in the background and impact zones are very close (solid lines in the diagram, see Fig. 6b).

The minimum sample size in the impact zone, sufficient to obtain a diversity of enchytraeids comparable to the background within the range of values, is about 50 samples (the beginning of the area of intersection of confidence intervals; Fig. 7). However, the potential sample size required for a complete study of the diversity of enchytraeids in the impact zone includes at least 150 samples (the area where the cumulative curve reaches a plateau; Fig. 7).

The results of the ordination of the species structure of enchytraeid communities indicate its similarity in the background and impact zones (Fig. 8). When analyzing both abundance and biomass, no species were identified that were clearly drawn to the conditions of the contaminated area. The average distance to the centroids increased with increasing proximity to the pollution source: both for abundance (in the background zone—0.551, in the impact zone—0.709) and for biomass (0.635 and 0.730, respectively).

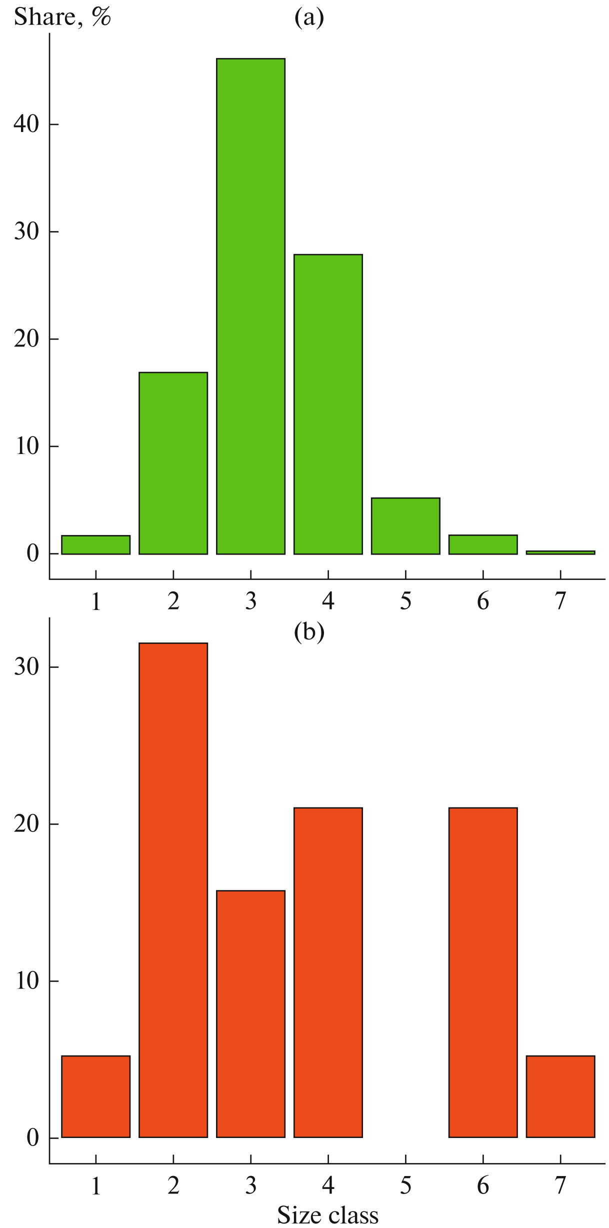


Fig. 5. Distribution of enchytraeids by size classes in the background (a) and impact (b) pollution zones.

DISCUSSION

Changes in Population Density, Biomass, and Diversity

During the study, 19 species of enchytraeids were identified. According to recent reports [40, 41], 9 of them were noted for the first time in the Sverdlovsk oblast, and another one was noted for the first time in Russia (*Fridericia auritoides* Schmelz, 2003). Of the 14 species previously noted for the Sverdlovsk oblast, we identified 10 (71%) [41].

The indicators of enchytraeid communities in the background zone are comparable with those given for unpolluted birch forests in central Europe (14 species, density 15000–30000 specimens/m^2, wet biomass 1200–1400 mg/m^2) [42]. When comparing the background zone with the coniferous forests of central and northern Europe, population density and species richness were comparable (from 6 to 21 species, 6000–134000 specimens/m^2), and the biomass was lower (in raw terms – 2170–29079 mg/m^2) [8, 13, 28, 43–45]. In the more southern broadleaf forests of England, Canada, and Japan, the average biomass of enchytraeids is also higher than that noted by us (in terms of wet body mass 10610–16680 mg/m^2; cited from [28]).

In the impact zone of MUCS, all registered species of enchytraeids have reduced density and biomass, and the majority of species also have reduced occurrence. This result correlates well with previous studies, which in most cases also described a decrease in the density

Table 1. Occurrence of enchytraeids in samples and in sample plots in different pollution zones

Species	In samples		On SPs	
	B	I	B	I
Buchholzia appendiculata (Buchholz, 1863)	0.7	0	1	0
Fridericia auritoides Schmelz, 2003	0.2	0	0.4	0
Fridericia bulboides Nielsen & Christensen, 1959	0.1	0	0.2	0
Fridericia callosa (Eisen, 1878)	0.2	0	0.2	0
Fridericia lacii Dózsa-Farkas, 2009	0.2	0	0.2	0
Fridericia paroniana Issel, 1904	0.2	0	0.4	0
Fridericia schmelzi Cech & Dózsa-Farkas, 2005	0.1	0	0.2	0
Fridericia sp.	0.6	0	1	0
Henlea ghilarovi Nurminen, 1980	0.1	0	0.2	0
Henlea perpusilla Friend, 1911	0.2	0	0.6	0
Henlea sp.	0.3	0	0.8	0
Marionina communis Nielsen & Christensen, 1959	0.4	0	0.8	0
Marionina vesiculata Nielsen & Christensen, 1959	0.1	0	0.2	0
Marionina sp.	0.2	0	0.8	0
Stercutus niveus Michaelsen, 1888	0.2	0	0.6	0
Cognettia sphagnetorum (Vejdovsky, 1878)	0.3	0.03	0.6	0.2
Enchytraeus buchholzi Vejdovsky, 1878	0.7	0.10	0.8	0.6
Enchytraeus sp.	0.1	0.03	0.4	0.2
Enchytronia parva Nielsen & Christensen, 1959	0.5	0.03	0.6	0.2
Fridericia ratzeli (Eisen, 1872) s.l.	0.3	0.13	0.6	0.4
Henlea nasuta (Eisen, 1878)	0.5	0.03	1	0.2
Bryodrilus sp.	0	0.03	0	0.2
Cognettia glandulosa (Michaelsen, 1888)	0	0.03	0	0.2

Pollution zones: B—background, I—impact. The occurrence of species in samples in the background zone is calculated for $n = 20$, in the impact zone for $n = 30$; occurrence in sample plots (SPs) in both zones for $n = 5$.

[7, 8, 10, 11, 13,] and diversity [8, 13] of enchytraeids. It is noteworthy that the impact of pollution sources on surrounding ecosystems differed from that of MUCS, the emissions of which led to a decrease in the pH of the soils of surrounding biocenoses [17]. Thus, the activities of a copper smelter in Sweden [13], zinc plants in Poland [12, 13], and the Netherlands [8] led to the neutralization of the initially acidic soils of coniferous forests. The copper-nickel plant in Finland did not have a significant impact on the acidity of surrounding soils [7, 9, 10]. Near the zinc plant in Poland, on the contrary, an increase in diversity in enchytraeid communities has been described [11]. The study's authors linked this to a decrease in the density of *C. sphagnetorum*, which dominates background sites and avoids sites contaminated by more neutral soils or high metal concentrations. Other species, on the contrary, moved into contaminated areas, which led to a decrease in dominance and an increase in the evenness of communities in the impacted area.

It is important to note that the diversity structure of enchytraeid communities in the background and impact zones is quite similar (see Fig. 6a). This may mean that the seemingly catastrophic decline in numbers in the impact zone has had little effect on the community structure itself. A decrease in occurrence was noted in the impact zone for almost all species (see Table 1), which suggests that less abundant enchytraeid species have simply not yet been taken into account. This assumption may be indirectly confirmed by the fact that almost all species represented in the impact zone have a high abundance in the background zone (see Appendix, Table S3).

It is also necessary to consider the distribution of enchytraeids by size classes, the range of values of which is comparable in both zones (see Fig. 5). In enchytraeids, body mass is closely related to length and correlates with species affiliation [30], so the presence of almost all size classes near the MUCS, despite a 20-fold decrease in biomass, suggests higher diversity than found in this study. More direct confirmation is the high similarity of the cumulation curves of the background and impact zones in the area of real cumulated diversity (see Fig. 6b), as well as the results

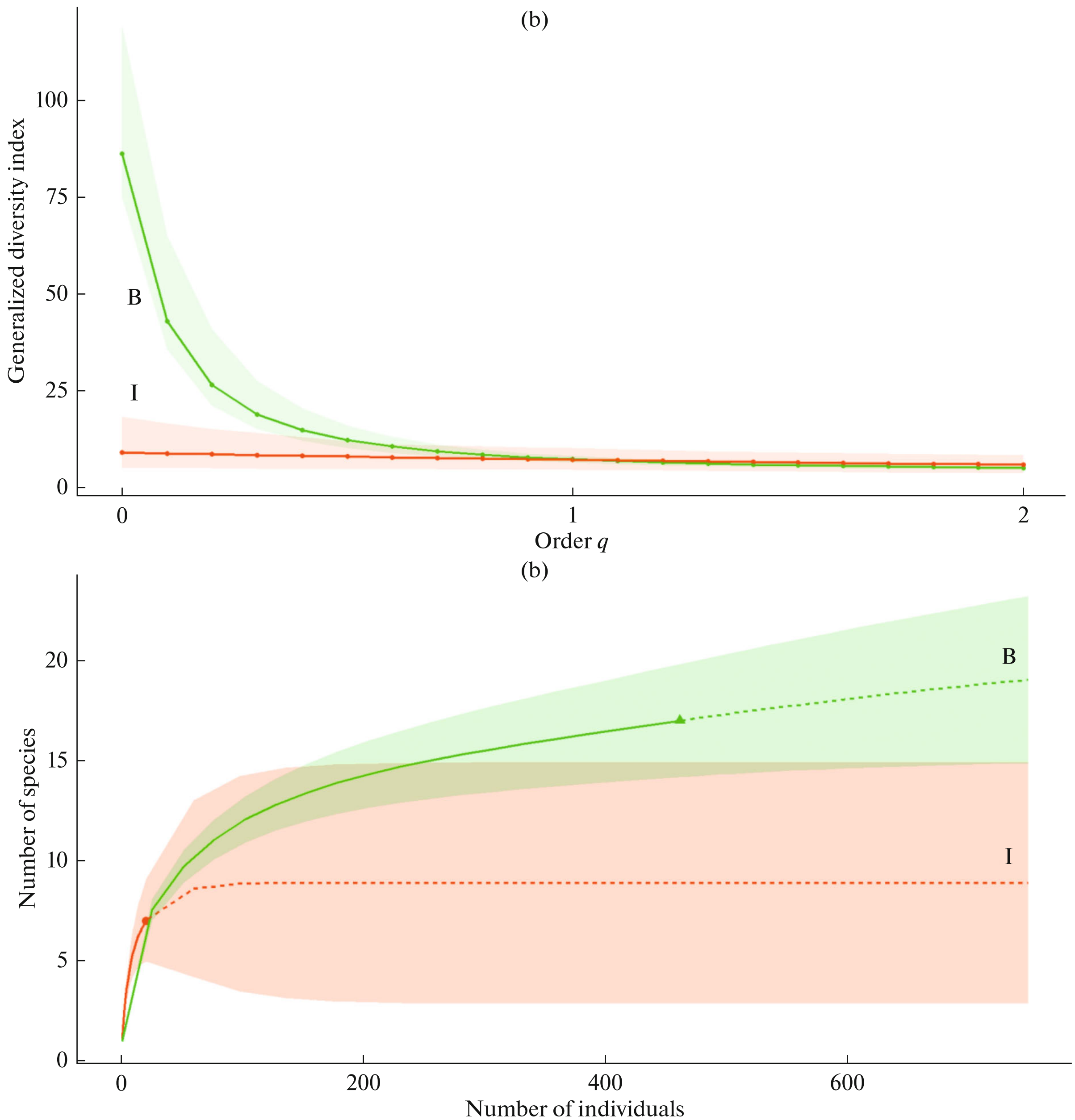


Fig. 6. Profiles of diversity indices (Hill numbers) (a) and extrapolation of potential diversity (b) of enchytraeids in contaminated zones (B—background, I—impact): dotted lines—extrapolated values; shaded areas—boundaries of the 95% confidence interval; accounting unit—sample plot (n = 5).

of the ordination of the structure of enchytraeid communities (Fig. 8). The state of communities in the impact zone of MUCS, is less stable (the distance to the centroids of the ordination diagrams is increased), and species that are attracted to the conditions of the contaminated area have not been identified at this stage of the study. However, to identify the actual diversity of the impact zone, significant additional efforts are required (the minimum sample is estimated at 50 samples, the optimal one is 150–200 samples, see Fig. 7). This conclusion can serve as a methodological guideline for organizing further studies in this direction.

Despite the arguments presented, a significant decrease in the density, species richness, and biomass of enchytraeids in the impact zone of MUCS allows us to state a pronounced negative effect in all soil horizons and variants of combining soil core layers (see Fig. 4). From a methodological point of view, this may mean that the general trend of the reaction of enchytraeid communities to pollution can be correctly determined by analyzing any of the given parameters

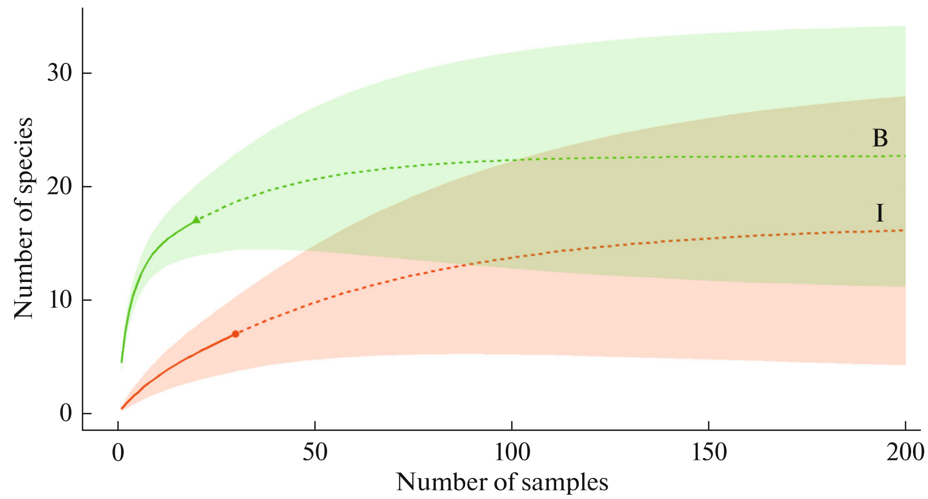


Fig. 7. Extrapolation of the potential sample size in the impact zone (I) required to achieve species richness comparable to the values of the background zone (B): dotted lines are extrapolated values; shaded areas are the boundaries of the 95% confidence interval; the sampling unit is the sample plot ($n = 5$).

and any of the layers considered, regardless of the depth of extraction. However, due to the low population density of enchytraeids in the impact zone, this conclusion needs to be confirmed by a study with a larger sample size.

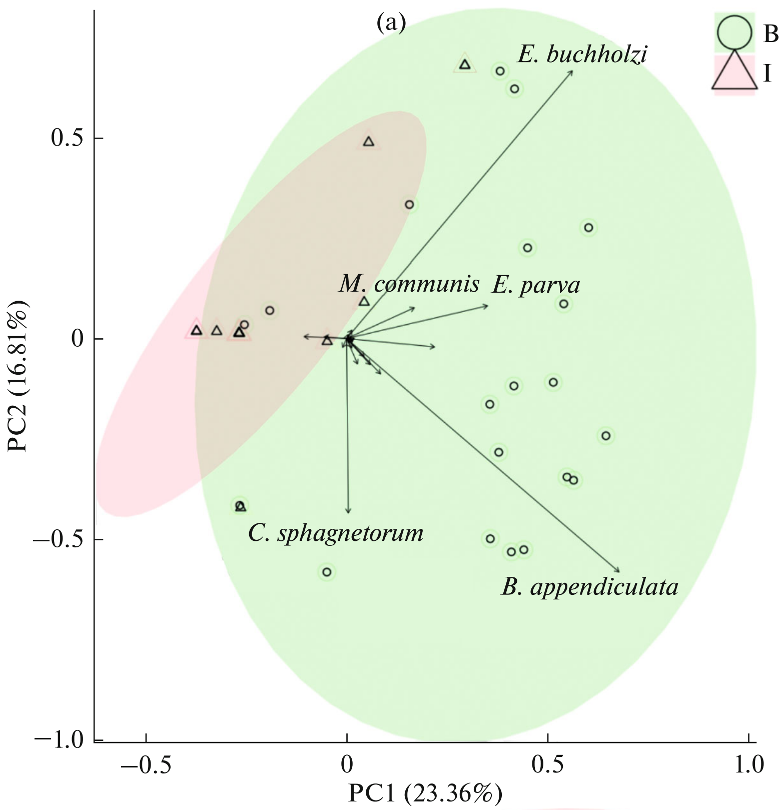
Changes in the Vertical Distribution

The total density and biomass of enchytraeids are vertically distributed unevenly and in the background zone are concentrated in the upper 2 cm of soil, which corresponds to the average thickness of the forest litter (see Figs. 2 and 3). A similar situation has been described for the soils of European spruce [14] and pine [16, 44] forests, where 47–70% of the total density and at least 54% of the total biomass [44] of enchytraeids were concentrated in the upper 2–3 cm of soil.

In the contaminated area, both parameters tend to shift deeper into the soil, being "redistributed" into the 2.1–4 cm layer. This corresponds to the situation near a copper smelter in Sweden, where 80% of the total abundance of enchytraeids was recorded in the upper layer (0–3 cm) of the background plots, while only 50% was recorded near the pollution source [13]. The shares of density and biomass in the forest litter in relation to the total values increased near MUCS (see Figs. 3b and 3d) and began to approximately correspond to the sums of the shares of density and biomass in the 0–2 cm and 2.1–4 cm layers of the background zone (see Figs. 3a and 3c). This phenomenon is undoubtedly associated with a twofold increase in the thickness of the litter (from 2.1 ± 0.3 cm to 3.8 ± 0.2 cm; see Fig. 2b and Appendix, Table S1) in birch forests of the impact zone due to the well-described inhibition of destructive processes [47]. In the background zone, the thickness of the litter on average corresponded to a layer of 0–2 cm, in the impact zone it corresponded to the sum of layers of 0–2 and 2.1–4 cm.

The displacement of enchytraeids to the lower part of the litter can hardly be associated with the avoidance of pollutants, since in the impact zone the maximum concentrations of all metals (except Fe) were noted precisely in the lower layer of the litter, at the border with the organomineral horizon [18]. Thus, the highest density of enchytraeids was recorded in the most toxic soil layer. Factors that can influence the vertical distribution of enchytraeids may be the quantity and quality of the food substrate [44, 47], namely detrital material 5–10 years old [48], the highest concentration of which is associated with the lower part of the litter. Another possible factor is insufficient moisture in the upper part of the litter, which can cause the migration of enchytraeids into the underlying layers [49, 50].

It can also be assumed that at least some species of enchytraeids may maintain a preferred depth regardless of the soil horizon in which it is located. In this case, thickened litter layer in the impact zone simply "captures" new, "depth-preferring" species. This version is consistent with the features of the vertical distribution of the species *Enchytronia parva*, which was the most numerous in the impact zone. A typical deep-layer species [15, 16], *E. parva,* is concentrated in the 2.1–4 cm layer in the background zone (33.3% of the total density in the layer), which belongs to the upper part of the organomineral horizon. In the impact zone, *E. parva* still predominates in the same 2.1–4 cm layer (38.5%), which at this level is already part of the forest litter (see Appendix, Table S3). The biomass of the species changes in a similar way (see Appendix, Table S6).



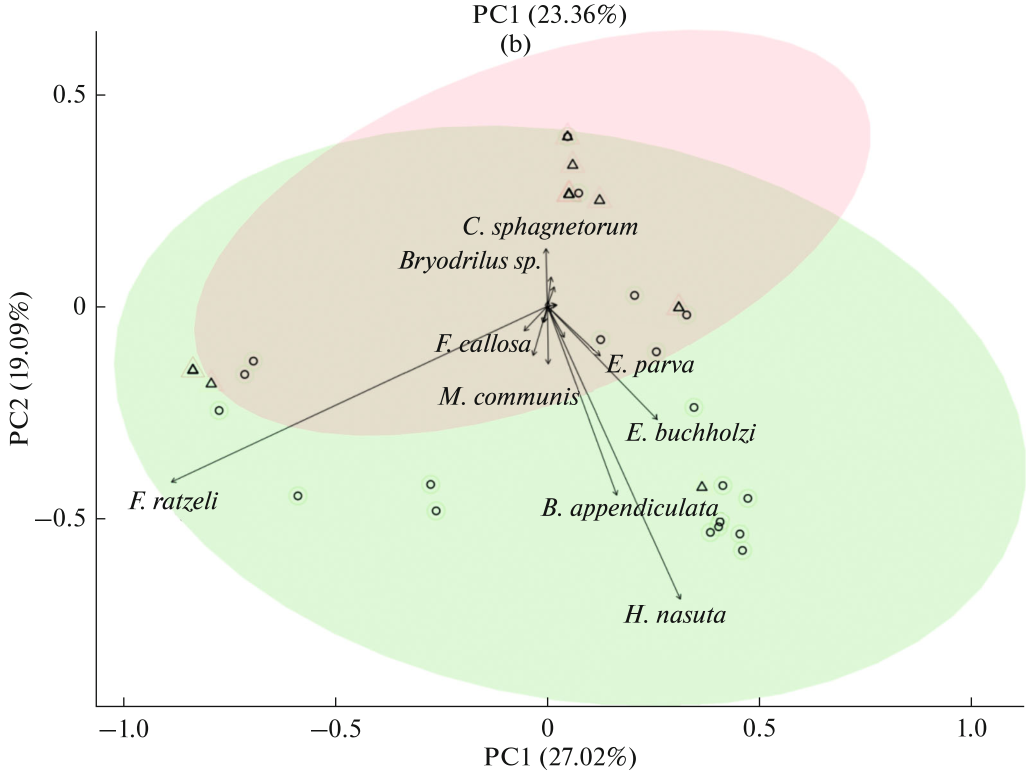


Fig. 8. Ordination of abundance (a) and biomass (b) in enchytraeid communities using the principal coordinate analysis (PCA) relative to the "pollution zone" factor; pollution zones: B—background, I—impact.

It should be noted that at this stage of the study, any discussion about the distribution patterns of enchytraeids in the impact zone are to some extent speculative due to the small number of collected worms and their uneven distribution.

CONCLUSIONS

The study showed that the population density, biomass, and species richness of enchytraeids were reduced in birch forests in the impact zone of MUCS. However, the diversity structure of enchytraeid communities has changed little. Diversity in the impact zone is potentially comparable to the background diversity, but its assessment is difficult due to the reduced frequency of species. The vertical distribution of the density and biomass of enchytraeids in the contaminated area shifted due to an increase in the proportion of the 2.1–4 cm layer, which in the background zone belongs to the organomineral soil horizon, and in the impact zone it belongs to the lower part of the forest litter.

Thus, the tested hypotheses regarding the decrease in density, biomass, and species richness, as well as about the change in the vertical distribution of enchytraeids in the contaminated area were confirmed. The hypothesis regarding changes in the structure of enchytraeid communities has not been confirmed at this time.

SUPPLEMENTARY INFORMATION

The online version contains supplementary material available at https://doi.org/10.1134/S1067413625600788.

ACKNOWLEDGMENTS

The authors are grateful to E.L. Vorobeychik (Institute of Plant and Animal Ecology, Ural Branch, Russian Academy of Sciences) for participating in the discussion of the article and suggestions for its improvement. The authors are sincerely appreciative to I.N. Korkina and E.L. Vorobeychik (Institute of Plant and Animal Ecology, Ural Branch, Russian Academy of Sciences) for providing data on pH and metal content in soils of sample plots; and M.R. Trubina (Institute of Plant and Animal Ecology, Ural Branch, Russian Academy of Sciences) for providing the vegetation characteristics.

FUNDING

The study was carried out as part of R&D project 122021000076-9 within the state assignment of the Institute of Plant and Animal Ecology, Ural Branch, Russian Academy of Sciences.

ETHICS APPROVAL AND CONSENT TO PARTICIPATE

According to the decision of the Bioethics Commission of the Institute of Plant and Animal Health of the Ural Branch of the Russian Academy of Sciences (protocol no. 14 of May 12, 2023), no special approval regarding compliance with ethical principles is required for research on invertebrates.

CONFLICT OF INTEREST

The authors of this work declare that they have no conflicts of interest.

REFERENCES

1. Didden, W.A.M., Ecology of terrestrial Enchytraeidae, *Pedobiologia*, 1993, vol. 37, no. 1, pp. 2–29.
2. van Vliet, P.C.J., Beare, M.H., and Coleman, D.C., Population dynamics and functional roles of Enchytraeidae (Oligochaeta) in hardwood forest and agricultural ecosystems, *Plant Soil,* 1995, vol. 170, no. 1, pp. 199–207.
3. Laakso, J. and Setälä, H., Sensitivity of primary production to changes in the architecture of below ground food webs, *Oikos*, 1999, vol. 87, no. 1, pp. 57–64.
4. Gajda, Ł., Gorgoń, S., and Urbisz, A.Z., Food preferences of enchytraeids, *Pedobiologia*, 2017, vol. 63, pp. 19–36.
5. Edwards, C.A., Subler, S., Chen, S.K., et al., Essential criteria for selecting bioindicator species, processes, or systems to assess the environmental impact of chemicals on soil ecosystems, in *Bioindicator Systems for Soil Pollution*, Netherlands: Kluwer Acad., 1996, pp. 67–84.
6. Didden, W. and Römbke J., Enchytraeids as indicator organisms for chemical stress in terrestrial ecosystems, *Ecotoxicol. Environ. Saf.*, 2001, vol. 50, no. 1, pp. 25–43.
7. Haimi, J. and Siira-Pietikäinen, A., Decomposer animal communities in forest soil along heavy metal pollution gradient, *Fresenius' J. Anal. Chem.*, 1996, vol. 354, nos. 5–6, pp. 672–675.
8. Noteboom, J., Relevance of laboratory toxicity data of Enchytraeus crypticus for single species assays and community changes in metal impacted field soils, in *Validation of Toxicity Data and Risk Limits for Soils: Final Report. Report 607505004*, Bilthoven: Natl. Inst. Public Health Environ., 1998, pp. 111–129.
9. Salminen, J. and Haimi, J., Horizontal distribution of copper, nickel and enchytraeid worms in polluted soil, *Environ. Pollut.*, 1999, vol. 104, no. 3, pp. 351–358.
10. Haimi, J. and Mätäsniemi, L., Soil decomposer animal community in heavy-metal contaminated coniferous forest with and without liming, *Eur. J. Soil Biol.*, 2002, vol. 38, no. 2, pp. 131–136.
11. Tosza, E., Dumnicka, E., Niklinska, M., et al., Enchytraeid and earthworm communities along a pollution gradient near Olkusz (southern Poland), *Eur. J. Soil Biol.*, 2010, vol. 46, nos. 3–4, pp. 218–224.
12. Kapusta, P., Szarek-Łukaszewska, G., and Stefanowicz, A.M., Direct and indirect effects of metal con-

tamination on soil biota in a Zn–Pb post-mining and smelting area (S. Poland), *Environ. Pollut.*, 2011, vol. 159, no. 6, pp. 1516–1522.

13. Kapusta, P. and Sobczyk, L., Effects of heavy metal pollution from mining and smelting on enchytraeid communities under different land management and soil conditions, *Sci. Total Environ.*, 2015, vol. 536, pp. 517–526.
14. Bengtsson, G. and Rundgren, S., Population-density and species number of enchytraeids in coniferous forest soils polluted by a brass mill, *Pedobiologia*, 1982, vol. 24, no. 4, pp. 211–218.
15. Abrahamsen, G., Ecological study of Enchytraeidae (Oligochaeta) in Norwegian coniferous forest soils, *Pedobiologia*, 1972, vol. 12, no. 1, pp. 26–82.
16. Dózsa-Farkas, K., Über die vertikale verbreitung der Enchytraeiden (Oligochaeta: Enchytraeidae) in einem hainbuchen-eichenwald Ungarns, *Opusc. Zool.*, 1992, vol. 25, pp. 61–74.
17. Pilipiuk, I., Enchytraeidae (Oligochaeta) of pine forests in Poland, *Fragmenta Faunistica*, 1993, vol. 36, no. 6, pp. 75–107.
18. Vorobeichik, E.L. and Kaigorodova, S.Yu., Long-term dynamics of heavy metals in the upper horizons of soils in the region of a copper smelter impacts during the period of reduced emission, *Eurasian Soil Sci.*, 2017, vol. 50, pp. 977–990.
19. Vorobeichik, E.L. and Korkina, I.N., A bizarre layer cake: Why soil animals recolonizing polluted areas shape atypical humus forms, *Sci. Total Environ.*, 2023, vol. 904, p. 166810.
20. Vopobeichik, E.L., Sadykov, O.F., and Farafontov, M.G., *Ekologicheskoe normirovanie tekhnogennykh zagryaznenii nazemnykh ekosistem (lokal'nyi uroven')* (Ecological Rating of Technogenic Pollutants in Terrestrial Ecosystems: Local Level), Ekaterinburg: Nauka, 1994.
21. Vorobeichik, E.L., Trubina, M.R., Khantemirova, E.V., et al., Long-term dynamic of forest vegetation after reduction of copper smelter emissions, *Russ. J. Ecol.*, 2014, vol. 45, no. 6, pp. 498–507
22. Vorobeichik, E.L., Ermakov, A.I., and Grebennikov, M.E., Initial stages of recovery of soil macrofauna communities after reduction of emissions from a copper smelter, *Russ. J. Ecol.*, 2019, vol. 50, no. 2, pp. 146–160
23. Graefe, U., Eine einfache methode der extraktion von Enchytraeiden aus bodenproben, in *Protokoll des Workshops zu Methoden der Mesofaunaerfassung*, Bremen: Universität Bremen, 1984, p. 1.
24. Kobetičová, K. and Schlaghamerský, J., On the efficiency of three schemes of enchytraeid wet funnel extraction, *Newsl. Enchytraeidae*, 2003, vol. 8, pp. 25–31.
25. Schmelz, R. and Collado, R., A guide to European terrestrial and freshwater species of Enchytraeidae (Oligochaeta), *Soil Organisms*, 2010, vol. 82, no. 1, pp. 1–176.
26. WoRMS. Enchytraeidae d'Udekem, 1855. 2025. https://www.marinespecies.org/aphia.php?p=taxdetails&id=2038.
27. Schneider, C.A., Rasband, W.S., and Eliceiri, K.W., NIH Image to ImageJ: 25 years of image analysis, *Nat. Methods*, 2012, vol. 9, pp. 671–675
28. Abrahamsen, G., Studies on body-volume, body-surface area, density and live weight of Enchytraeidae (Oligochaeta), *Pedobiologia*, 1973, vol. 13, pp. 6–15.
29. Makulec, G., Enchytraeidae (Oligochaeta) of forest ecosystems. I. Density, biomass and production, *Ekol. Polska*, 1983, vol. 31, no. 1, pp. 9–56.
30. Conti, E. and Mulder, C., Chemistry-driven Enchytraeidae assemblages acting as soil and ecosystem engineers in edaphic communities, *Ecol. Indic.*, 2022, vol. 144, p. 109529.
31. Berg, M., De Ruiter, P., Didden, W., et al., Community food web, decomposition and nitrogen mineralization in a stratified Scots pine forest soil, *Oikos*, 2001,vol. 94, no. 1, pp. 130–142.
32. R Core Team. R: A language and environment for statistical computing, 2025. http://www.R-project.org/.
33. Wickham, H., Francois, R., Henry, L., et al., dplyr: A grammar of data manipulation. R package version 1.1.3, 2023. https://CRAN.R-project.org/package=dplyr.
34. Wickham, H., *ggplot2: Elegant Graphics for Data Analysis*, New York: Springer-Verlag, 2016.
35. Marcon, E. and Hérault, B., entropart: An R package to measure and partition diversity, *J. Stat. Software*, 2015, vol. 67, no. 8, pp. 1–26.
36. Hsieh, T.C., Ma, K.H., and Chao, A., iNEXT: iNterpolation and EXTrapolation for species diversity. R package version 3.0.0, 2022. http://chao.stat.nthu.edu.tw/-wordpress/softwaredownload/.
37. Beck, M.W., ggord: Ordination plots with ggplot2. R package version 1.1.6, 2021. https://zenodo.org/badge/latestdoi/35334615.
38. Martinez Arbizu, P., pairwiseAdonis: Pairwise multilevel comparison using adonis. R package version 0.4, 2020. https://github.com/pmartinezarbizu/pairwiseAdonis.
39. Roberts, D.W., labdsv: Ordination and multivariate analysis for ecology. R package version 2.1-0, 2023. https://CRAN.R-project.org/package=labdsv.
40. Pustejovsky, J.E., Chen, M., and Swan, D.M., SingleCaseES: A calculator for single-case effect sizes. R package version 0.7.3, 2024. https://CRAN.R-project.org/package=SingleCaseES.
41. Degtyarev, M.I., Zaitsev, A.S., Danilova, M.A., et al., Population of soil-dwelling enchytraeids (Annelida, Clitellata, and Enchytraeidae) in the European part of Russia, *Russ. J. Ecol.*, 2024, vol. 55, pp. 145–153.
42. Degtyarev, M.I., Saifutdinov, R.A., Korobushkin, D.I., et al., Taxonomic diversity and abundance of enchytraeids (Annelida, Clitellata, Enchytraeida) in the Northern Palaearctic. 1. Asian part, *Biodiversity Data J.*, 2024, vol. 12, e114682.
43. Nowak, E., Enchytraeid communities in successional habitats (from meadow to forest), *Pedobiologia*, 2001, vol. 45, no. 6, pp. 497–508.
44. Nurminen, M., Ecology of enchytraeids (Oligochaeta) in Finnish coniferous forest soil, *Annales Zool. Fenn.*, 1967, vol. 4, no. 2, pp. 147–157.
45. Didden, W.A.M. and de Fluiter, R., Dynamics and stratification of Enchytraeidae in the organic layer of a

Scots pine forest, *Biol. Fertil. Soils*, 1998, vol. 26, no. 4, pp. 305–312.

46. Abrahamsen, G., Biomass and body-surface area of populations of Enchytraeidae and Lumbricidae (Oligochaeta) in Norwegian coniferous forest soils, *Pedobiologia*, 1973, vol. 13, no. 1, pp. 28–39.
47. Vorobeichik, E.L., Changes in the spatial structure of the destruction process under conditions of atmospheric pollution of forest ecosystems, *Izv. Ross. Akad. Nauk, Ser. Biol.*, 2002, no. 3, pp. 368–379.
48. Briones, M.J.I., Carrera, N., Huang, J., et al., Substrate quality and not dominant plant community determines the vertical distribution and C assimilation of enchytraeids in peatlands, *Funct. Ecol.*, 2020, vol. 34, no. 6, pp. 1280–1290.
49. Briones, M.J.I. and Ineson, P., Use of 14C carbon dating to determine feeding behaviour of enchytraeids, *Soil Biol. Biochem.*, 2002, vol. 34, no. 6, pp. 881–884.
50. Springett, J.A., Brittain, J.E., and Springett, B.P., Vertical movement of Enchytraeidae (Oligochaeta) in moorland soils, *Oikos*, 1970, vol. 21, no. 1, pp. 16–21.
51. Uhía, E. and Briones, M., Population dynamics and vertical distribution of enchytraeids and tardigrades in response to deforestation, *Acta Oecol.*, 2002, vol. 23, pp. 349–359.

Translated by V. Mittova