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Successive development of soil ecosystems at abandoned coal-ash landfills

Stanislav Pen-Mouratov · Nosir Shukurov · Jun Yu · Shakhnoza Rakhmonkulova · Obidjon Kodirov · Gineta Barness · Michael Kersten · Yosef Steinberger

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Abstract The main goal of the present study was to determine the effect of the native vegetation on the successive development of the soil ecosystem at abandoned coal-ash landfills of the Angren coal-fired power plant in Uzbekistan. Two different landfills (one not in use for 3 years, termed newer, and the other not in use for 10 years, termed older) with different degrees of vegetation cover were chosen to assess the time and vegetation effects on soil biota and habitat development. The soil biotic structure, including soil microorganisms and soil free-living nematode communities, was investigated both at open plots and under different native plants at the coal-ash landfill area. The observed soil microorganisms were found to be the most important component of the observed ecosystems. Total abundance, biomass, species, trophic and sexual diversity of soil free-living nematodes, along with fungi and organic-matter content, were found to be correlated with trace metals. The nematode trophic and species abundance and diversity increased from the newer toward the older coal-ash landfills. The sex ratio of the nematode communities was found to be dependent on the environmental conditions of the study area, with the males being the most sensitive nematode group. All applied ecological indices confirmed that open landfill plots distant from plants are the most unfavorable areas for soil biota. In that respect, the native plants *Alhagi maurorum Desv*. and *Tamarix sp*. were found to be important environmental components for the natural remediation of a soil ecosystem in the coal-ash landfill area.

Keywords Coal-ash landfill · Heavy metal · Microorganism · Nematode · Sex diversity · Industrial pollution

S. Pen-Mouratov (✉) · J. Yu · G. Barness · Y. Steinberger
The Mina and Everard Goodman Faculty of Life Sciences, Bar-Ilan University, 5290002 Ramat-Gan, Israel
e-mail: stanislav.pen-mouratov@biu.ac.il

N. Shukurov · S. Rakhmonkulova · O. Kodirov
Institute of Geology and Geophysics, Academy of Sciences, Tashkent, Uzbekistan 700041

J. Yu
State Key Laboratory of Forest and Soil Ecology, Institute of Applied Ecology, Chinese Academy of Sciences, Shenyang 110164, People's Republic of China

M. Kersten
Geosciences Institute, Gutenberg University, 55099 Mainz, Germany

Introduction

According to the United Nations Economic Commission for Europe (UNECE 2002), one of the most common environmental challenges for many countries is fly-ash utilization and the disposal of coal-based electricity byproducts. Large plots of land are affected by coal ash, occupying a large area for its deposition and leading to an increased hazard of environmental contamination (Adriano 1986). Significant amounts of fly ash are still being disposed in lagoons and landfills (Haynes 2009). According to Singh et al. (2010), the disposal of fly ash produced from 1 MW electricity may require up to 1 acre of land. Miller et al. (2002) and White and Claxton (2004) emphasized the lack of knowledge related to its toxicity and post-depositional reactivity, followed by improper disposal and poor management, which resulted in serious soil contamination problems at these sites. Coal-burning contamination, mainly consisting of SO_2 and fly ash (15–90 % of the total

residue), was generated during coal combustion and dependent on the coal's geological origin, combustion conditions, efficiency of particulate removal, and degree of weathering before final disposal (Adriano et al. 1980; Haynes 2009; Klein et al. 1975; Mathur et al. 2003).

Fly ash generally has a silt loam texture (Chang et al. 1977) and is mainly represented by SiO_2, Al_2O_3, Fe_2O_3, CaO, and numerous trace elements (Adriano et al. 1980; Snellings et al. 2012). Coal ash consists of over 40 elements and most of the trace elements are essential nutrients for plant growth (Li et al. 2003). The major elemental constituents of fly ash are Si, Al, Fe, Ca, C, Mg, K, Na, S, Ti, P, and Mn (el-Mogazi et al. 1988). Potentially toxic trace elements in coal ash, such as As, Be, Cd, Ba, Cr, Cu, Pb, Hg, Mo, Ni, Ra, Se, Th, U, V, Zn can have a negative impact on the activity and development of a soil biota association (Arthur et al. 1984).

One of the more promising solutions for fly-ash contamination might be the on-site usage of phytoremediation plants (Dmowska and Ilieva-Makulec 2006; Ghosh and Singh 2005). Moreover, native plants can be more promising for phytoremediation tasks (Newman et al. 1998; Singh et al. 2010; Barrutia et al. 2011). However, as of now, little is known about the effect of the native plants on soil biotic abundance and diversity in the fly-ash contamination area (Haynes 2009).

Numerous studies showed that soil microorganisms, along with soil free-living nematode communities, have been found to be among the best biological tools for assessing soil disturbances, including heavy-metal pollution, in terrestrial systems (Bongers et al. 2001; Brookes et al. 1984; Georgieva et al. 2002; Gupta and Yeates 1997; Neher 1999; Pen-Mouratov et al. 2010). The qCO_2 metabolic quotient, known as the ecophysiological index for the soil microbial community, can also be used as a specific activity parameter to assess environmental stress (Anderson and Domsch 1993). Furthermore, the soil biota was found to be useful bioindicators for assessing the process of remediation of a stressed environment in the power-plant coal-ash landfills (Dmowska 2005; Paoletti 1999). Previous studies conducted in the Angren industrial area that focused on both the direct and indirect (through soil property changes) effects of industrial pollution on soil biotic abundance and diversity, confirmed the expediency of using soil biota as biological indicators in the study area (Pen-Mouratov et al. 2010; Shukurov et al. 2009).

The Angren coal-fired power plant is one of the main sources of electricity in Uzbekistan, using an average of 100,000 tons of coal per year. The remaining coal-burning ash is deposited in an isolated, well-defined area outside the power plant. Increasing the industrial deposition space causes severe damage to natural ecosystems in Uzbekistan (UNECE 2000).

The main goal of the present study was to determine effect of the native vegetation on the successive development of the soil ecosystem at abandoned coal-ash landfills of the Angren coal-fired power plant in Uzbekistan. Two different landfills (one had been abandoned at the time of the study for 3 years, termed newer, and the other has been abandoned for 10 years, termed older) with different degrees of vegetation cover were chosen to assess the time and the vegetation effects on soil biota and habitat development. Based on the above, we hypothesized that abundance of the soil microorganisms and total number and diversity of the soil free-living nematodes, following vegetation development, will increase during the process of forming a new ecosystem at abandoned coal-ash landfills and will be higher at the vegetation area in the older coal-ash landfill.

Materials and methods

Study site

The study site is located in one of the largest coal-mining regions and power-production centers in Uzbekistan (Fig. 1). The climate is continental, with a maximal temperature of 40 °C in summer and −25 °C in winter. Multiannual yearly precipitation ranges between 300 and 500 mm, 96 % of which falls from autumn to spring season. Thermal inversions provide cyclic circulation of air masses and cause pendulum distributions of dust and gas-smoke emissions from the industrial enterprises.

The coal-ash landfills are located on the upper side of the Akhangaran River Valley (Fig. 1), near the city of Angren (41°01′N–70°09′E), where the coal-burning power plant (260 MW capacity) is one of the major sources of air and soil pollution in this area. The electrical power station is operated on a brown-coal basis from the nearby Angren coal mine, with a production capacity of 2.5 million tons per year, producing 11–35 % of its ash waste.

The waste produced by the electrical power center as a result of coal usage is accumulated at isolated sites. As result, units of coal-ash landfills of different ages can be found dispersed on the landscape. In each of these, the coal waste units undergo natural succession processes, where local plants of different species exert a significant effect on the soil ecosystem development.

Two coal-ash landfills of defined and different ages of abandonment, as well as one uncontaminated site, were used for the present study. One was an older landfill (OLF, ca 2 km^2) that had been abandoned at the time of the study for 10 years and the other was a newer landfill (NLF, ca 2 km^2) that had been abandoned for 3 years. The third site represented the control area, which had never been used as a coal-ash landfill (CS, ca 0.6 km^2) and was within a

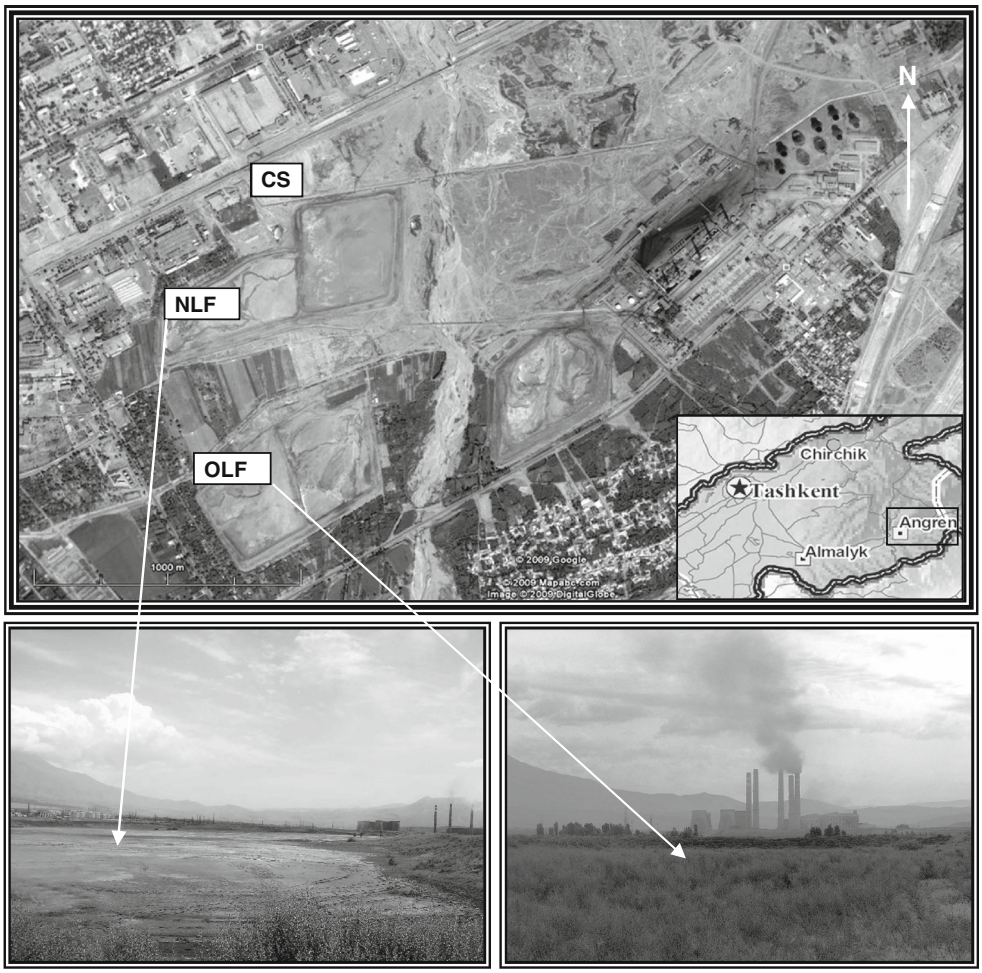

Fig. 1 Location of the study site. The coal-ash landfills were located on the upper side of the Akhangaran River Valley, near the city of Angren (41°01′N–70°09′E) in Uzbekistan. *NLF* new coal-ash landfill; *OLF* old coal-ash landfill; *CS* control area that was never used as a landfill. The lower *pictures* show a difference in vegetation cover between the older and newer landfills, where the older landfill (OLF) was characterized by more developed vegetative growth

distance of 0.5 km from the landfill basin. The coal-ash landfills were fenced by protective dams with a maximum height of about 35 m. The thickness of the waste material ranged from 10 to 20 m (Fig. 1).

The most common perennial-vegetation covers at the coal-ash landfill area were *Alhagi maurorum Desv.* and *Tamarix sp*., where *A. maurorum* (Camelthorn) is known to grow in soils with high carbonate levels (Kassas 1952). The older and the newer coal-ash landfills had similar soil types but differed in the degree of vegetation covering. The soil control area was represented by Calcisol (haplic) (FAO 2012) soil type. The vegetation cover throughout the study sites is dominated by annual and perennial plants, the most common being *Astragalus, Stipa*, *Medicago*, and *Artemisia*. These soils contain high levels of $CaCO_3$ and have low organic-matter content (Makhmudov and Khaitov 2000).

Sampling and sampling design

Non-grid soil sampling methods, used for observation of landscape with high soil homogeneity, were applied for soil sampling.

Samples were taken from the three above-mentioned sites, the new coal-ash landfill (NLF), the old coal-ash landfill (OLF), and the control site (CS), which had never been used as a coal-ash landfill and was located outside of the coal-ash landfill area and was used for comparison between the industrial area and natural ecosystems. Sampling points were located in the following order: completely open space at a distance of 20 m from the plants (COS); under *A. maurorum* (PA); one meter around *A. maurorum* (CPA); under *Tamarix sp*. (PT); and four meters from the stem of the *Tamarix sp*. (CPT). The CPA and CPT, bare plots were used to define the development of the industrial ecosystem outside of the vegetation area, whereas the COS was considered to be part of the observed industrial ecosystem that was entirely free from the influence of vegetated.

Moreover, in order to define the effect of vegetation on the environment, the soil samples were grouped as (1) PA+PT, the soil samples collected under plants (*A. maurorum and Tamarix sp*) at the landfill area; and (2) CPA+CPT, the soil samples collected at the open space of the landfill area close to the *A. maurorum* and *Tamarix sp*.

Soil samples (n = 4) were collected from the 0–10-cm upper soil layers at each of the study plots 1 week after the rainfall, in June 2011. Each soil sample, which consisted of five subsamples from the study area, was collected using the following approach: (1) a stainless steel core sampler and trowel were used to scarify a solid portion of collected soil; (2) soil subsamples untouched by the steel sampler and trowel were scooped up with a plastic spoon, mounted on a plastic sheet, and stirred into a homogeneous mixture (Alloway 1995; EPA 1995). An amount of 0.5 kg of each replicate was placed in an individual plastic bag and transported to the laboratory in an insulated box. At the laboratory, the replicates were kept in cold storage at 4 °C. Before sieving, an amount of 100 g from each replicate was used to determine the soil free-living nematode community. The remaining soil was sieved through a 2-mm mesh sieve before microbial, physical, and chemical analyses.

Sample analysis

Subsamples of each replicate were subjected to the following analyses:

a. *Soil moisture* (SM) was measured gravimetrically as percentage of dry mass by oven-drying to a constant weight (105 °C, 48 h).
b. *Organic matter* (OM) was determined by oxidization with dichromate in the presence of H_2SO_4 without application of external heat (Rowell 1994).
c. *Heavy-metal concentrations* were determined using both atomic absorption spectrometry (AAS) and X-ray fluorescence analysis (XRF, ED 2000Rh, Oxford Instruments, England). Powder pellets of subsamples were used for XRF analyses of the chemical elements. For atomic absorption analysis of the element concentration, subsamples were extracted overnight by digestion with a mixture of three parts concentrated HNO_3 and one part concentrated $HClO_3$.
d. *Soil microbial biomass (MB) and soil basal respiration (MR)* were determined by the MicroResp™ method (Anderson and Domsch 1978; Campbell et al. 2003b). This system consisted of a deep-well plate and a 96-well detection microtiter plate (Campbell et al. 2003a). CO_2 evolution and microbial biomass were measured by dye plates—a colorimetric reaction using absorbent alkali with the ability to measure carbon dioxide evolution. Water was added to whole soil samples in deep well plates covered by the dye plates in order to measure respiration. Glucose was added to determine microbial biomass according to the substrate-induced respiration method. CO_2 values were measured after 6 h of soil respiration. The respired CO_2 was absorbed by the detection plate and quantified using a spectrophotometer reading at 590 nm (Berg and Steinberger 2008).
e. *Colony-forming units (CFUs) of fungi and bacteria*. A plate count method was used to estimate the population of heterotrophic fungi and bacteria in each soil sample. Martin's Rose Bengal (MRB) agar and tryptic soy agar (TSA) were prepared for fungal and bacterial cultures, respectively. MRB and TSA media were separately inoculated with 100 µl 10- and 100-fold soil dilutions, 4 replicates for each soil suspension, and then incubated in the dark at 27 °C for 7 days (for fungi) or 5 days (for bacteria). CFUs on each plate were determined.
f. *The nematode population* was extracted from 100 g aliquots of the subsamples using the Baermann funnel procedure (Cairns 1960). The recovered organisms were counted and preserved in formaldehyde (Steinberger and Sarig 1993). The nematodes from each sample were collected and identified according to order, family, genus, and sex using a compound microscope. Nematodes were classified according to known feeding habitats and morphology (i.e., Goodey 1963; Yeates et al. 1993; Bongers 1994; Yeates and King 1997) into the following trophic groups: bacteria-feeding (BF), fungi-feeding (FF); plant-parasitic (PP) and omnivore-predator (OP) (Steinberger and Loboda 1991; Pen-Mouratov et al. 2003). The total number of nematodes (Tnem) was counted and adjusted to 100 g dry soil. Maximum body width (±1 µm) and 'length' (±5 µm) were also measured in order to determine the biomass of the extracted population using the Andrassy (1956) and Yeates (1972) methods. The nematode bodies were measured using normal optical magnification by 216×, and by 840×for the length and width, respectively.

The following formula was used to calculate the wet body weight (BW):

$$(\mathrm{BW}) = (\mathrm{W}^2 \times \mathrm{L})/(16 \times 100)\ \mu\mathrm{g}$$

where W (maximum body width) and L (body 'length') are in µm (Andrassy 1956).

Ecological indices: nematode community

The characteristics of the microbial and nematode communities were described using the following indices: (a) metabolic coefficient (qCO_2), calculated as the ratio between CO_2 production and microbial biomass (Anderson and Domsch 1990). The qCO_2 is a specific parameter for

evaluating the effects of environmental conditions on soil microbial biomass; (b) the nematode channel ratio, NCR = BF/(BF + FF) (Yeates 2003); (c) Shannon–Weaver index, $H' = -\sum P_i$ ($\ln P_i$) (Shannon and Weaver 1949);(d) evenness, EV = H'/Ln(S), where S is the number of taxa and H' is the Shannon index (Pielou 1977); (e) species richness, SR = (S − 1)/ln(N), where N is the number of individuals identified (Yeates and King 1997); (f) trophic diversity, $T = 1/\sum P_i^2$, where P_i is the proportion of the *i*-th trophic group (Heip et al. 1988); (g) Simpson's dominance index, $D = \sum P_i^2$ (Simpson 1949); (h) maturity index, $MI = \sum v_i f_i/n$, where v_i is the *c-p* value of the *i*-th genus in the nematode assigned by Bongers (1990), f_i is the frequency of family *i* in the sample, and *n* is the total number of individuals in a sample (Neher and Darby 2005); the *c-p* value is the nematode life strategy index, ranging from 1 (colonizers, tolerant to disturbance) to 5 (persisters, sensitive to disturbance) (Bongers and Bongers 1998; McSorley 1997; Neher et al. 1995; Wasilewska 2006; Yeates 1996); (i) maturity index modification (MMI), including plant-feeding nematodes (Yeates and Bird 1994).

Statistical analysis

All data were expressed on a dry-soil basis and subjected to statistical analysis of variance (ANOVA) using the SAS model (GLM) (SAS Institute 1988; Sokal and Rohlf 1969). Duncan's multiple range tests and Pearson correlation coefficient were used to evaluate significant differences and inter-relationships among separate means. A two-tailed probability index ($P < 0.05$) was considered to be statistically significant. The data were also tested by computing multivariate redundancy analysis (RDA) in order to provide more information by taking into account differences between planted and open spaces at the two, different-aged landfills (Program CANOCO, Version 4.54, October 2005—written by ter Braak (C) 1988–2005). The Monte Carlo permutation test (499 permutations were used for this study) was used to calculate the significance of a given factor and its relevance for the measured parameter (ter Braak 1995; ter Braak and Prentice 1996). The graphical output arrows, pointing roughly in the same direction, indicated a positive correlation, while arrows pointing in the opposite direction indicated a negative one. The length of the arrow indicated the relative strength of the relationship.

Results

Soil properties

SM levels were found to range from 5.3 to 18.3 % and were highest in the COS in the NLF (ANOVA, $P < 0.001$), and in the CPA at the OLF (ANOVA, $P < 0.005$) (Figs. 2, 3). The SM values showed no significant differences between the two observed coal-ash landfills, except for the soil samples around *A. maurorum*, which exhibited more moisture at the OLF (ANOVA, $P < 0.05$).

OM ranged from 1 to 2.6 % and was found to be higher in the soil samples under and around the PA and CPA, respectively and under the PT at the OLF, and showed no differences between the observed plots at the NLF (Figs. 2, 3). OM content showed a positive correlation with Cd, Pb, Zn, Mo, W, Bi, and Ni (r = 0.57, 0.54, 0.58, 0.59, 0.63, 0.52, and 0.51, respectively, $P < 0.05$) under plants of the observed area, and showed a negative correlation with Cd, Sb, Pb, Cu, Y, Rb, Bi, and Ni (r = −0.45, −0.53, −0.42, −0.45, −0.45, −0.59, −0.50 and −0.46, respectively, $P < 0.05$) in the open plots of the landfills. No correlation was found between OM and the observed trace-element concentration in the CS.

Out of a total of 24 elements analyzed, only As, Cd, Co, Cr, Cu, Mo, Ni, Pb, Rb, Sc, and Zn exhibited significantly (ANOVA, $P < 0.05$) different contents between different sampling sites of the study areas. The concentration of some trace elements was significantly higher in the barren plots of the landfills and in the control area (Table 1).

Basal microbial respiration and biomass

MR values fluctuated between 1.29 and 2.18 μg CO_2–C (g soil h)$^{-1}$ at the OLF and between 1.5 and 3.5 μg CO_2-C (g soil h)$^{-1}$ at the NLF, with no significant differences between the observed plots in the landfills and between the landfill area and the control site (Fig. 2). In addition, the MR showed no significant differences between the two landfills, except for the soil samples under the PT, where MR values of the NLF were two times higher than the OLF (ANOVA, $P < 0.05$) (Fig. 2). MB ranged between 90 and 197 μg C g^{-1} soil at the OLF and between 135 and 219 μg C g^{-1} soil at the NLF (Fig. 2). The MB, similar to the MR, showed no significant differences between the two landfills, except for the soil samples under the PT, where MB values of the NLF were almost two times higher than the OLF ($P < 0.05$) (Fig. 2).

Colony-forming units (CFUs) of fungi and bacteria

The amount of culturable bacteria ranged between 0 and 134 CFU × 10^4 per g dry soil at the OLF, and between 0 and 43 CFU × 10^4 per g dry soil at the NLF (Fig. 2). The amount of culturable bacteria between the landfills and between the landfills and the CS (Fig. 2) was not significant.

The amount of culturable fungi ranged between 2 and 71 CFU × 10^2 per g dry soil at the OLF, and between 6 and 104 CFU × 10^2 per g dry soil at the NLF (Fig. 2).

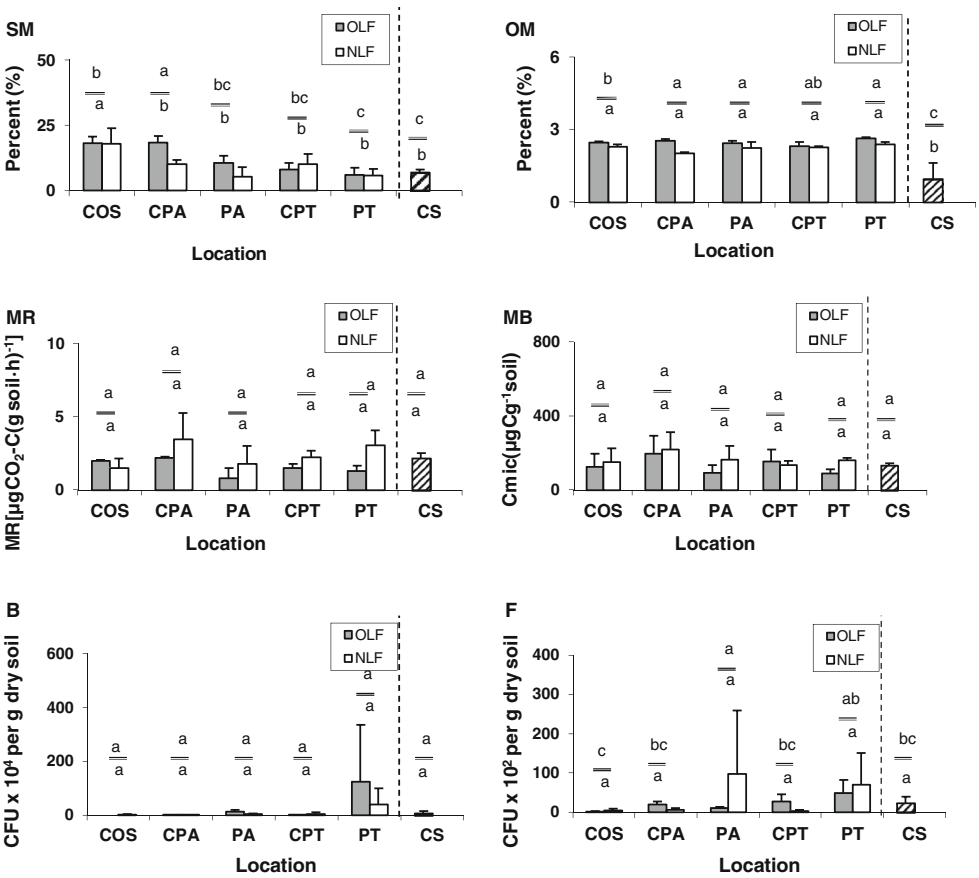

Fig. 2 Changes in soil moisture (SM), organic matter (OM), microbial biomass (MB), microbial respiration (MR), bacteria (B), and fungi (F), in the upper (0–10 cm) soil layer at new (NLF) and old (OLF) coal-ash landfills, and at the control area, which had never been used as a coal-ash landfill (CS) of the Angren study area. Different *letters* indicate significant differences ($P < 0.05$, n = 4, for each sampling plot). Sampling plots: *COS* completely open space at a distance from the plants; *PA* soil samples under *Alhagi maurorum*; *CPA* soil samples around *Alhagi maurorum*; *PT* soil samples under *Tamarix* sp., *CPT* soil samples around *Tamarix* sp. Different *letters* indicate statistical differences between sampling plots: *letters* of the *upper row* for the OLF, *letters* of the *lower row* for the NLF

Unlike bacteria, the amount of culturable fungi was found to be higher in the soil samples under the observed plants at the OLF (ANOVA, $P < 0.009$), but there was no difference between observed plots at the NLF (Fig. 2). The fungi showed no significant differences between the two landfills except for the soil samples around the CPA, where the amount of fungi at the OLF was more than three times higher than the amount of fungi at the NLF (ANOVA, $P < 0.05$) (Fig. 2). The total number of bacteria and fungi, along with microbial biomass and basal microbial respiration was found to correlate with soil properties and trace elements in the landfills (Table 2). Multivariate analysis of the microbial community (Fig. 3) showed clear discrimination between the open and the overgrown vegetation plots at the observed landfills, with prevalence of the bacteria and fungi at the vegetation plots.

Nematode community structure

Twenty-five nematode taxa were identified in the present study: 7 taxa belonged to the bacterivore trophic group (BF), 6 were fungivores (FF), 7 were plant-parasites (PP), and 5 were omnivore-predators (OP) (Table 3). Mean density of the soil free-living nematodes (Tnem) was significantly higher at the OLF than at the NLF both under plants and at the interspaces between the shrubs (ANOVA, $P < 0.05$) (Fig. 4; Table 3). In contrast to the OLF, soil free-living nematodes at the NLF were represented only by BF nematodes (Table 3). Moreover, at the NLF, Tnem exhibited the highest values under the PA, whereas at the OLF it exhibited the highest values under the PT. At the OLF, the BF trophic group was more numerous under plants than in the bare plots, while the density of the FF trophic group in the same landfill under plants was not significantly different from the bare plots (ANOVA, $P < 0.05$). The PP and OP nematode trophic groups were represented only by *Longidorella* and *Aporcelaimus*, respectively, under the PT at the OLF (Table 3).

In general, the obtained data indicated that soil free-living nematode density and total number of nematode genera were higher under the observed plants and when close to the vegetated plots, with the most favorable habitat being under the PT at the OLF (ANOVA, $P < 0.001$) and under the PA at the NLF (ANOVA, $P < 0.05$) (Fig. 4; Table 3). Among the observed soil free-living nematodes, *Aporcelaimus*, *Ditylenchus*, *Eucephalobus*, *Paraphelenchus*, and *Longidorella* were found only under plants, while *Cephalobus*, *Metateratocephalus*, and *Panagrolaimus* were found only in the open space of the landfill area (Table 3).

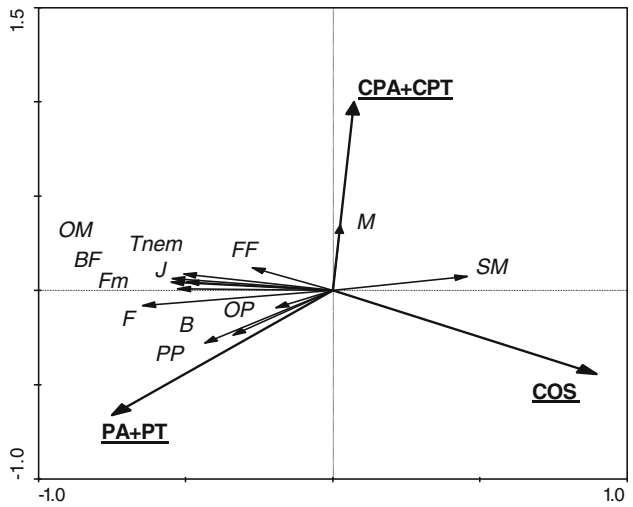

Fig. 3 Redundancy analysis (RDA) demonstrates that plots under the canopy of the observed plants and near the vegetated area are more favorable to the soil-biota than at a distance from the plant area. The length and angles of *arrows* indicate correlation between soil biota and soil properties and between soil biota and habitat. The first axis explains 23 % of the total variability in the observed data, with the sum of all canonical eigenvalues being 25 %. The significance of these variations was confirmed by the Monte Carlo permutation test (P value = 0.006; F-ratio = 7.85; number of permutations = 499). Sampling plots: PA+PT, soil samples collected under *Alhagi maurorum* and under *Tamarix* sp. at the two landfills; CPA+CPT, soil samples collected around *Alhagi maurorum* and *Tamarix* sp. at the two landfills; COS, soil samples collected at the completely open space at a distance from the plants. Environmental variation: *SM*,soil moisture; *OM* organic matter; *B* bacteria; *F* fungi abundance, respectively; *Tnem* total number of free-living nematodes. Trophic structure: OP, omnivore-predator; PP, plant-parasitic; FF, fungi-feeding; *BF* bacteria-feeding nematodes. Sex groups: *M* male; *Fm* female; *J* juvenile

Of all the observed nematode species, only a few specimens of BF and FF trophic groups were correlated with SM and OM. *Acrobeloides* was found to correlate with both SM and OM; *Chiloplacus* and *Aphelenchoides* showed a positive correlation with OM; *Ditylenchus*, *Nothotylenchus*, and *Paraphelenchus* species showed a positive correlation with SM (Table 4).

The Tnem, along with the BF and PP trophic groups, showed a positive correlation with the total number of bacteria ($P < 0.05$, r = 0.46, r = 0.42, r = 0.57, respectively) and fungi ($P < 0.05$, r = 0.46, r = 0.40, r = 0.42, respectively) at the OLF, while OP showed a positive correlation only with the total number of bacteria (r = 0.50, $P < 0.01$) at the same place. The Tnem and BF of the NLF correlated with the total number of fungi (r = 0.73, $P < 0.001$), but there was no correlation between the nematodes and bacterial density.

The nematode species were also positively correlated with the concentrations of Ba, Bi, Co, Cr, Mo, Nb, Ni, Pb, W, Zn, Y, and U, and negatively with Ga, Rb, and Sc at the landfill area (Table 4). The same was true for nematode taxa at the CS (but with negative correlation with Co and Cr and positive correlation with As and Th) (Table 4). Note that the trace elements Rb and Sc are commonly used as indicator elements for the clay contents in soils and sediments. Of all the observed soil nematodes, *Cephalobus*, *Metateratocephalus*, *Nothotylenchus*, *Tylencholaimus*, *Coslenchus*, *Filenchus, Psylenchus*, *Tylenchorhynchus*, and *Eudorylaimus* were found to be the least sensitive to the trace elements and showed no correlation with the observed chemicals (Table 4). The occurrence of negative correlation between the soil free-living nematodes and the observed trace elements was more frequent at the open plots of the landfill area in comparison to other plots of the study area (Table 4).

Similar to the total number and trophic groups of soil free-living nematodes, biomass of the observed nematodes was found to correlate with the observed chemical elements (Table 5). The total biomass of bacteria- and fungi-feeding nematodes showed a positive correlation with OM under plants (Table 5). The total biomass of free-living nematodes, along with the biomass of bacteria- and fungi-feeding trophic groups, showed a positive correlation with fungal density in the soil samples collected in the open spaces of the landfill area (CPA+CPT) in contrast to the plant (PA+PT) and control (CS) sampling sites. The total biomass showed a positive correlation with chemical elements under plants and a mixed (negative/positive) correlation with chemical elements in the bare plots and the CS (Table 5). The biomass of the trophic groups also exhibited a positive and negative correlation with the chemical elements (Table 5).

The nematode sex ratio was represented by juveniles and females as dominant groups and males as the smallest sex group (Fig. 4). Moreover, nematode males were found only at the CS and at the CPT at the OLF (Fig. 4). The numbers of juveniles together with females were higher under plants than in the bare plots around the plants (ANOVA, $P < 0.05$) (Fig. 4). The total number of juveniles was lower at the NLF than at the CS. The total number of juvenile and female nematodes was positively correlated with the total number of bacteria at the OLF (r = 0.38, $P < 0.05$ and 0.52 $P < 0.01$, respectively) and with the total number of fungi at the NLF (r = 0.50).

Ecological indices

The qCO_2 values were found to increase from 14.43 ± 9.1 and 16.9 ± 5 mg CO_2–C g C_{mic} h^{-1} in the soil samples collected in the NLF and the OLF, respectively, to 20.6 ± 6.8 mg CO_2–C g C_{mic} h^{-1} in the control area (CS), with no statistically significant differences between the landfill and control areas. The Duncan's multiple range

Table 1 Range of concentration of chemical elements at the upper soil layer (0–10 cm) of coal-ash landfills and control plots in the Angren industreial area

Element (mg kg^{-1})	OLF					NLF					Control	GLM data
	COS	CPA	PA	CPT	PT	COS	CPA	PA	CPT	PT	CS	F-test
As	**91.7^{a}**	**47.3^{ba}**	**48.3^{ba}**	**30.0^{b}**	**32.3^{b}**	**16.7^{b}**	**11.3^{b}**	**10.7^{b}**	**13.0^{b}**	**11.0^{b}**	**60.3^{ba}**	**2.26**
Cd	**11.6^{a}**	**4.3^{ba}**	**5.0^{ba}**	**4.2^{ba}**	**4.3^{ba}**	**0.2^{b}**	**1.0^{b}**	**0.3^{b}**	**0.4^{b}**	**0.6^{b}**	**7.3^{ba}**	**2.05**
Co	**10.3^{ba}**	**7.7^{bc}**	**6.3^{bc}**	**6.3^{bc}**	**5.7^{bc}**	**8.7^{bc}**	**6.7^{bc}**	**6.3^{bc}**	**7.0^{bc}**	**7.7^{bc}**	**12.3^{a}**	**2.37**
Cr	**44.7^{ba}**	**28.7^{b}**	**28.7^{b}**	**26.3^{b}**	**28.3^{b}**	**26.7^{b}**	**25.0^{b}**	**25.0^{b}**	**25.3^{b}**	**25.3^{b}**	**58.7^{a}**	**2.09**
Cs	28.7^{a}	32.3^{a}	29.0^{a}	29.3^{a}	26.3^{a}	31.7^{a}	27.3^{a}	29.7^{a}	30.0^{a}	32.0^{a}	17.7^{a}	0.78
Cu	**34.7^{a}**	**30.0^{bc}**	**29.0^{c}**	**29.0^{c}**	**30.0^{bc}**	**32.7^{ba}**	**28.7^{c}**	**27.7^{c}**	**31.7^{bac}**	**30.7^{bac}**	**35.0^{a}**	**2.47**
Mo	**22.0^{a}**	**22.0^{a}**	**22.8^{a}**	**19.8^{ba}**	**20.4^{ba}**	**3.2^{c}**	**4.9^{c}**	**4.3^{c}**	**4.2^{c}**	**4.1^{c}**	**9.8^{bc}**	**5.52**
Ni	**21.7^{a}**	**10.3^{b}**	**10.3^{b}**	**11.0^{b}**	**10.7^{b}**	**9.0^{b}**	**9.3^{b}**	**8.3^{b}**	**8.3^{b}**	**7.7^{b}**	**30.0^{a}**	**3.89**
Pb	**412^{a}**	**187^{ba}**	**183^{ba}**	**165^{b}**	**157^{b}**	**72.0^{b}**	**52.7^{b}**	**47.7^{b}**	**49.3^{b}**	**46.0^{b}**	**242^{ba}**	**9.47**
Rb	**99.7^{a}**	**81.7^{bdc}**	**81.3^{dc}**	**82.0^{bdc}**	**84.7^{bdc}**	**98.0^{a}**	**85.0^{bdc}**	**82.0^{bdc}**	**93.7^{ba}**	**91.3^{bac}**	**103^{a}**	**4.79**
Sb	13.0^{a}	5.3^{a}	4.0^{a}	3.3^{a}	3.0^{a}	1.7^{a}	1.0^{a}	1.3^{a}	1.0^{a}	1.3^{a}	6.0^{a}	1.33
Sc	**11.3^{ba}**	**10.7^{bc}**	**10.3^{bc}**	**10.3^{bc}**	**10.0^{bc}**	**10.3^{bc}**	**10.0^{bc}**	**9.7^{c}**	**10.0^{bc}**	**10.3^{bc}**	**12.3^{a}**	**3.76**
Th	21.8^{a}	24.0^{a}	23.2^{a}	23.6^{a}	24.4^{a}	27.3^{a}	25.4^{a}	24.8^{a}	26.0^{a}	26.2^{a}	17.4^{a}	1.79
U	10.5^{a}	13.0^{a}	13.1^{a}	12.4^{a}	12.8^{a}	10.5^{a}	12.2^{a}	11.1^{a}	10.8^{a}	11.0^{a}	6.9^{a}	1.44
V	86.0^{a}	82.7^{a}	80.7^{a}	76.7^{a}	77.3^{a}	91.3^{a}	77.3^{a}	82.3^{a}	88.7^{a}	90.7^{a}	99.3^{a}	1.75
W	18.3^{a}	7.0^{a}	6.0^{a}	15.7^{a}	9.0^{a}	12.3^{a}	17.0^{a}	13.3^{a}	12.7^{a}	14.0^{a}	39.7^{a}	0.73
Y	36.0^{a}	30.7^{a}	31.0^{a}	29.7^{a}	31.0^{a}	30.3^{a}	29.0^{a}	28.7^{a}	29.7^{a}	29.0^{a}	32.0^{a}	0.93
Zn	**1356^{a}**	**597^{ba}**	**591^{ba}**	**551^{ba}**	**519^{ba}**	**121^{b}**	**121^{b}**	**102^{b}**	**89.0^{b}**	**81.7^{b}**	**874^{ba}**	**2.11**
Tc					**6967**					**2715**	**1664**	

Bold indicates the significant different values

OLF old coal-ash landfill, *NLF* new coal-ash landfill, *CS* control site, *COS* completely open space at a distance from the plants, *PA* soil samples under *Alhagi maurorum, CPA* soil samples around *Alhagi maurorum, PT* soil samples under *Tamarix sp*., *CPT* soil samples around *Tamarix sp, Tc* total concentration of the observed chemicals

^{a,b,c,d} Different letters indicate significant differences between sampling locations, n = 44, $P < 0.05$

Table 2 Pearson correlation coefficients between microbial characteristics, soil properties and trace elements in the study area

	PA + PT		**CPA + CPT**	
	Old landfill	New landfill	Old landfill	New landfill
MR	**Sc$^{-0.71*}$**,V$^{0.72*}$	**SM$^{-0.99***}$**	SM$^{0.65*}$	**SM$^{-0.67*}$,OM$^{-0.93***}$,Cs$^{-0.67*}$**,U$^{0.72*}$
MB	**OM$^{-0.69*}$,Sc$^{-0.73*}$**,V$^{0.72*}$,Cs$^{0.69*}$	**Sb$^{-0.86*}$,Ga$^{-0.86*}$**		**V$^{-0.67*}$**,Cd$^{0.72*}$,As$^{0.56*}$,W$^{0.96**}$,Ba$^{0.86*}$
B	**Sr$^{-0.70*}$**	OM$^{0.85*}$	**Co$^{-0.57*}$,Rb$^{-0.58}$**	
F	Bi$^{0.73*}$, Cs$^{0.67*}$,Mo$^{0.83**}$,Pb$^{0.73*}$		**As$^{-0.57*}$,Co$^{-0.61*}$,Bi$^{-0.56}$, Sc$^{-0.75}$,Rb$^{-0.65*}$**,Ba$^{0.65*}$	

Bold numbers are the negative correlation

PA+PT soil samples collected under plants at the waste field, *CPA+CPT* soil samples collected between plants at the waste field, *MR* soil basal respiration, *MB* microbial biomass, *B* total number of bacteria, *F* total number of fungi, *SM* soil moisture, *OM* organic matter

* Values with $P < 0.05$; ** values with $P < 0.01$; *** values with $P < 0.001$

tests showed no difference in the qCO_2 values between sampling locations (Table 6). However, multivariate analysis indicated that qCO_2 values were higher in the open plots around the PA at the NLF and in the COS at the OLF (Fig. 5). The qCO_2 showed a negative correlation with the soil moisture in the bare plots and with the vanadium and cesium in the soil samples collected under plants at the landfill area (Table 7).

The NCR values ranged from 1.0 ± 0 and 0.84 ± 0.15 at the NLF and OLF, respectively, to 0.52 ± 0.17 at the CS, with statistically significant differences between the NLF, the OLF, and the CS (Table 6). The NCR was found

Table 3 Mean abundunce (per 100 g^{-1} dry soil) and standard deviation of nematode genera (classification Yeates and King 1997) at the landfill and control area

Station	Old landfill				New landfill						
	COS	CPA	PA	CPT	PT	COS	CPA	PA	CPT	PT	CS
Trophic groups/genus/family											
Tnem	0	112 ± 14	140 ± 136	189 ± 197	2324 ± 518	0	2.5 ± 2.2	28.8 ± 25	0	4.6 ± 6.2	61.6 ± 36
Tg	0	8	9	6	7	0	2	2	0	1	18
Bacteria-feeding											
Acrobeloides	0	47 ± 20	44 ± 38	48 ± 23	1105 ± 505	0	2.0 ± 1.8	25.1 ± 21	0	4.6 ± 6.2	3.3 ± 5.7
Cephalobus	0	3.5 ± 6.1	0	0	0	0	0	0	0	0	0
Cervidellus	0	19 ± 19	27 ± 41	8.3 ± 11	69 ± 60	0	0.5 ± 0.8	3.8 ± 4.5	0	0	3.4 ± 5.9
Chiloplacus	0	25 ± 22	8.6 ± 15	93 ± 117	326 ± 564	0	0	0	0	0	1.1 ± 2.0
Eucephalobus	0	0	2.1 ± 3.7	0	0	0	0	0	0	0	0.5 ± 1.0
Metateratocephalus	0	2.2 ± 3.8	0	0	0	0	0	0	0	0	0
Panagrolaimus	0	1.8 ± 3.0	0	2.1 ± 3.6	0	0	0	0	0	0	1.7 ± 1.7
Fungi-feeding											
Aphelenchoides	0	0	2.1 ± 3.7	14 ± 24	50 ± 87	0	0	0	0	0	2.8 ± 3.5
Aphelenchus	0	11 ± 8.6	41 ± 51	24 ± 34	57 ± 50	0	0	0	0	0	0
Ditylenchus	0	0	6.2 ± 6.2	0	0	0	0	0	0	0	0
Nothotylenchus	0	2.2 ± 3.8	4.1 ± 7.1	0	0	0	0	0	0	0	0
Paraphelenchus	0	0	4.1 ± 7.1	0	0	0	0	0	0	0	0
Tylencholaimus	0	0	0	0	0	0	0	0	0	0	5.0 ± 1.6
Plant-parasitic											
Coslenchus	0	0	0	0	0	0	0	0	0	0	2.2 ± 3.8
Filenchus	0	0	0	0	0	0	0	0	0	0	8.8 ± 10.0
Longidorella	0	0	0	0	622 ± 543	0	0	0	0	0	0
Merlinius	0	0	0	0	0	0	0	0	0	0	1.1 ± 2.0
Pratylenchus	0	0	0	0	0	0	0	0	0	0	1.1 ± 2.0
Psylenchus	0	0	0	0	0	0	0	0	0	0	1.1 ± 1.9
Tylenchorhynchus	0	0	0	0	0	0	0	0	0	0	12.1 ± 20.9
Omnivores-predators											
Aporcelaimus	0	0	0	0	95 ± 165	0	0	0	0	0	3.4 ± 3.4
Aporcelaimellus	0	0	0	0	0	0	0	0	0	0	0.5 ± 1.0
Dorilaimoides	0	0	0	0	0	0	0	0	0	0	3.4 ± 3.4
Eudorylaimus	0	0	0	0	0	0	0	0	0	0	8.9 ± 5.2
Microdorylaimus	0	0	0	0	0	0	0	0	0	0	1.1 ± 2.0

Values of $P < 0.05$ were considered significant

COS completely open space at a distance from the plants, *PA* soil samples under *Alhagi maurorum, CPA* soil samples around *Alhagi maurorum*, *PT* soil samples under *Tamarix sp*., *CPT* soil samples around *Tamarix sp*., *CS* soil samples of control area, *Tnem* number of total nematodes, *Tg* number of nematode genera

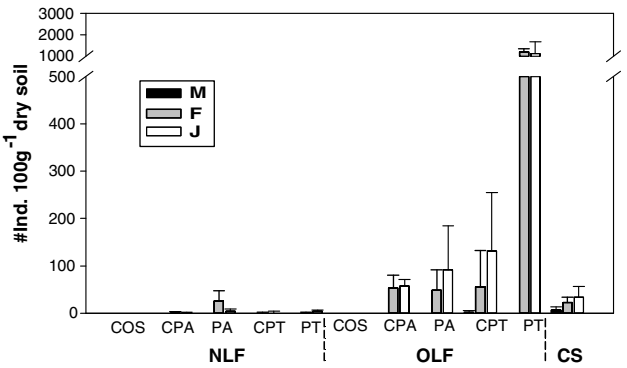

Fig. 4 Changes in the total number of soil free-living nematode sex groups ($P < 0.05$, n = 4, for each sampling plot in the upper (0–10 cm) soil layer at the Angren study area. Sex groups: *M* male; *F* female; *J* juvenile. Sampling plots: new (NLF) and old (OLF) coal-ash landfills; CS, control area that was never used as a landfill (the same site was used as a comparable control area for the two landfills); *COS* completely open space at a distance from the plants; *PA* soil samples under *Alhagi maurorum*; *CPA* soil samples around *Alhagi maurorum*; *PT* soil samples under *Tamarix* sp.; *CPT* soil samples around *Tamarix* sp

to be significantly different between the plant area of the landfills (PA + PT) and the CS (ANOVA, $P < 0.02$), and between the open spaces of the landfills (CPA + CPT) and the CS (ANOVA, $P < 0.0001$), with no differences between soil samples collected in the open spaces of the landfills (CPA + CPT) and under plants in the landfills (PA + PT). The NCR values were negatively correlated with trace elements in the landfill area under plants and in the bare plots, with no correlation in the soil control area (Table 7).

The Shannon index (H') ranged from 0.10 ± 0.2 and 0.87 ± 0.58 at the NLF and OLF, respectively, to 1.65 ± 0.53 at the CS, and showed statistically significant differences between the NLF, the OLF, and the CS (Table 6). The H' values showed no differences between the soil samples collected in the open space of the landfill area (CPA+CPT) and soil samples collected under plants in the landfills (PA+PT), but was found to be significantly different between the plant area in the landfills (PA+PT) and the CS (ANOVA, $P < 0.01$), and between the open spaces of the landfills (CPA+CPT) and the CS (ANOVA, $P < 0.003$). The H' index was correlated with trace elements in the landfill area under plants, with no correlation in the soil control area (Table 7).

The mean of evenness (EV) ranged from 0.68 ± 0.22 and 0.79 ± 0.02 at the NLF and OLF, respectively, to 0.75 ± 0.13 at the CS, with no statistically significant differences between the new and old landfills as well as between the landfills and the control area (Table 6). The EV was found to be significantly different in the soil samples collected in the open spaces of the landfills (CPA+CPT) compared to the soil samples collected under plants in the landfills (PA+PT) (ANOVA, $P < 0.03$). The EV values were positively correlated with trace elements in the landfill area under plants, in the bare plots, and in the soil control area (Table 7).

The SR values ranged from 0.18 ± 0.26 and 0.68 ± 0.3 at the NLF and OLF, respectively, to 1.99 ± 0.51 at the CS, with significant differences between the NLF, the OLF, and the CS (Table 6). The SR values showed no differences between the soil samples collected in the open spaces of the landfills (CPA+CPT) and the soil samples collected under plants in the landfills (PA + PT), but showed significant differences between the plant area in the landfills (PA + PT) and the CS (ANOVA, $P < 0.0001$), and between the open spaces of the landfills (CPA+CPT) and the CS (ANOVA, $P < 0.0001$). The species richness was both positively and negatively correlated with the observed trace elements in the landfills under plants and in the bare plots, as well as in the soil control area (Table 7).

The trophic diversity index (T) ranged from 1.0 ± 0 and 1.56 ± 0.43 at the NLF and OLF, respectively, to 2.56 ± 0.76 at the CS, and showed significant differences between the NLF, the OLF, and the CS (Table 6). The T index showed no differences between the soil samples collected in the open spaces of the landfills (CPA+CPT) and soil samples collected under plants in the landfills (PA+PT), but showed significant differences between the plant area in the landfills (PA + PT) and the CS (ANOVA, $P < 0.0001$), and between the open spaces of the landfills (CPA + CPT) and the CS (ANOVA, $P < 0.0001$). The T value showed a positive correlation with trace elements under plants but was not correlated with the trace elements in the bare areas and the soil control area (Table 7).

The mean Simpson's dominance index (D) ranged from 0.87 ± 0.18 and 0.36 ± 0.08 at the NLF and OLF, respectively, to 0.21 ± 0.09 at the CS (Table 6). The D showed differences between the new and the old landfills as well as between the NLF and the CS (Table 6). The D showed no significant differences between the soil samples collected in the open spaces of the landfills (CPA+CPT) and soil samples collected under plants in the landfills (PA+PT), but showed differences between the plants area in the landfills (PA+PT) and the CS, and between the open spaces of the landfills (CPA + CPT) and the CS (ANOVA, $P < 0.0001$). The D showed both a positive and negative correlation with trace elements in the observed area in the landfill area under plants and in the bare plots, as well as in the soil control area (Table 7).

The MI and MMI values were similar and ranged from 2.04 and 2.16 and from 3.13 and 3.39 at the OLF and CS, respectively with the same values (2.00) in the NLF (Table 6). These indices showed no differences between the soil samples collected in the open spaces of the landfills

Table 4 Correlation coefficients of the free-living nematodes with soil moisture, organic matter and trace elements in the study area

Trophic groups/ genus/family	PA + PT	CPA + CPT	CS
Bacteria-feeding			
Acrobeloides	$Cr^{0.67**}$, $Ni^{0.67**}$,$W^{0.55*}$,$Y^{0.60**}$, $Nb^{0.53*}$	$SM^{0.46*}$, $OM^{0.47*}$, $Mo^{0.43*}$, $W^{0.60**}$, **$Rb^{-0.53*}$**,**$Ga^{-0.54}$**	
Cephalobus			
Cervidellus	$Cr^{0.71**}$, $Nb^{0.58*}$,$Y^{0.58*}$,	**$Rb^{-0.43*}$; $Sc^{-0.44*}$**	$Bi^{0.57*}$,$Mo^{0.55*}$,$W^{0.57*}$,$Y^{0.52*}$,$Th^{0.47*}$,
- " -			$U^{0.52*}$,$Zn^{0.55*}$,$As^{0.59*}$,**$Co^{-0.55*}$; $Cr^{-0.60*}$**
Chiloplacus	$OM^{0.56*}$, $Ba^{0.53*}$	**$Rb^{-0.48*}$**	$As^{0.57*}$,$Bi^{0.52*}$,$Mo^{0.50*}$,$W^{0.55*}$,$Zn^{0.47*}$,
- " -			$Y^{0.55*}$,$Th^{0.53*}$,$U^{0.55*}$, **$Cr^{-0.57*}$**
Eucephalobus	$As^{0.53*}$, $Sb^{0.73**}$,$Cd^{0.51*}$,		
Metateratocephalus			
Panagrolaimus	$Cr^{0.52*}$		
Fungi-feeding			
Aphelenchoides	$OM^{0.57*}$, $Ba^{0.54*}$		$As^{0.60*}$,$Bi^{0.48*}$,$Mo^{0.55*}$,$W^{0.57*}$,$Zn^{0.52*}$,$Y^{0.58*}$,$Th^{0.55*}$
Aphelenchus	$Bi^{0.60*}$, $Mo^{0.58*}$, $Ni^{0.55*}$, $Pb^{0.53*}$,$W^{0.59}$,$Zn^{0.54*}$,$U^{0.57*}$	$W^{0.47*}$, **$Rb^{-0.43*}$**	
Ditylenchus	$SM^{0.52*}$, $Bi^{0.53*}$		
Nothotylenchus	$SM^{0.61*}$		
Paraphelenchus	$SM^{0.61*}$	**$Sc^{-0.46*}$**	
Tylencholaimus			
Plant-parasitic			
Coslenchus			
Filenchus			
Longidorella	$Ni^{0.56*}$,$W^{0.52*}$,$Y^{0.58*}$,$U^{0.56*}$	**$Sc^{-0.46*}$**	
Merlinius			$As^{0.55*}$,$Zn^{0.57*}$,
Pratylenchus			$Mo^{0.50*}$,$W^{0.54*}$,$Zn^{0.48*}$
Psylenchus			
Tylenchorhynchus			
Omnivores-predators			
Aporcelaimus	$Nb^{0.60*}$, $U^{0.55*}$,**$Co^{-0.60**}$**		
Aporcelaimellus			$Ba^{0.50}$, $Ga^{0.56}$, $Nb^{0.58}$
Discolaimus	$Co^{0.52*}$		
Dorilaimoides	$Co^{0.52*}$		$Ba^{0.56}$, $Ga^{0.48}$, $Nb^{0.50}$
Eudorylaimus			
Microdorylaimus		**$Sc^{-0.46*}$**	$As^{0.67**}$,$Bi^{0.52*}$,Mo^{60*},$Zn^{0.56*}$,$Th^{0.50}$,**$Cr^{-0.55}$**

Bold indicate negatively correlated values

PA+PT soil samples collected under plants at the landfills, *CPA+CPT* soil samples collected in outplant plots at the landfills, *SM* soil moisture, *OM* organic matter, *CS* control area, which never been used as landfills

* values with $P < 0.05$; ** values with $P < 0.01$

(CPA+CPT) and soil samples collected under plants in the landfills (PA+PT), but it did show differences between the plant area in the landfills (PA+PT) and the CS, and between the open spaces of the landfills (CPA+CPT) and the CS (ANOVA, $P < 0.0001$). In contrast to the MI, MMI was positively affected by W, Cr, and Ni under plants (Table 7).

Multivariate analysis indicated that in contrast to the qCO_2 metabolic coefficient, the applied nematode ecological indices were higher under and close to the observed plants at both landfills (Fig. 5).

Discussion

Industrial contamination can have an impact on the ecosystem as a whole and on individual relationships between

Table 5 Pearson correlation coefficients between biomass of nematodes, soil organic matter (OM) soil fungi (F), and trace-element contents in the study area

	PA+PT	CPA+CPT	CS
TB	$OM^{0.59*}$, $Mo^{0.55*}$, $W^{0.59*}$, $Ni^{0.61*}$, $Y^{0.56*}$	$F^{0.67**}$, **$Sc^{-0.46*}$**, **$Rb^{-0.49*}$**	$Cd^{0.52*}$, $Pb^{0.65**}$, $Mo^{0.53*}$, $W^{0.54*}$, $Th^{0.57*}$, $Cs^{0.53*}$, $Y^{0.56*}$, **$Ni^{-0.60*}$**, **$Cr^{-0.48*}$**
TBBF	$OM^{0.59*}$, $W^{0.55*}$, $Ni^{0.61*}$	$F^{0.70**}$, **$Cu^{-0.44*}$**, **$Rb^{-0.55*}$**	
TBFF	$OM^{0.53*}$	$F^{0.47*}$, **$Sc^{-0.48*}$**	
TBPP	$Y^{0.55*}$	**$Sc^{-0.46*}$**	
TBOP		**$Sc^{-0.46*}$**	$Sb^{0.55*}$, **$Co^{-0.57*}$**
BmM		$F^{0.52*}$	$As^{0.65**}$, $Cd^{0.54*}$, $Mo^{0.55*}$, $W^{0.57*}$, $Th^{0.55*}$, $Y^{0.60*}$, **$Co^{-0.55*}$**, **$Cr^{-0.52*}$**
BmF	$OM^{0.61**}$, $Mo^{0.55*}$, $W^{0.60*}$, $Ni^{0.61*}$, $Y^{0.54*}$	$F^{0.65**}$, **$Sc^{-0.47*}$**, **$Rb^{-0.46*}$**	$Sb^{0.67**}$
BmJ	$Mo^{0.53*}$, $W^{0.53*}$, $Ni^{0.57*}$, $Y^{0.60*}$, **$Co^{-0.55*}$**	$F^{0.49*}$, $W^{0.47}$, **$Sc^{-0.44*}$**, **$Rb^{-0.46}$**	

Bold indicate negatively correlated values

PA+PT soil samples collected under plants at the landfills, *CPA+CPT* soil samples collected in outplant plots at the landfills, *CS* soil samples of control area, *TB* biomass of total nematodes, *TBBF* biomass of bacterivores, *TBFF* biomass of fungivores, *TBPP* biomass of plant-parasites, *TBOP* biomass of omnivore-predators, *BmM* biomass of male, *BmF* biomass of female, *BmJ* biomass of juvenile

* values with $P < 0.05$; ** values with $P < 0.01$

the different components of the ecosystem, in particular. Moreover, as believed (Odum 1981), environmental contamination can act as a trigger for improving and developing stabilizing adaptive mechanisms for the ecosystem, which is periodically found in a state of stress.

The ecological investigations of the last decades came to the conclusion that industrial pollution in different parts of the world has both direct and indirect (through soil property changes), strong and negative effects on soil biota (Yeates et al. 1994; Georgieva et al. 2002; Pen-Mouratov et al., 2008). The main soil properties were found to be tightly interdependent on metal concentration, including relatively stable elements (Christiansen et al. 2002). In the present study, soil properties, along with soil biota, were found to be correlated with trace elements and location in the study area. Moreover, organic matter was found to be mainly negatively correlated with the trace elements at the open part of the landfill area, while under plant organic matter it exhibited mostly positive correlation with the chemicals.

Table 6 Comparative change in values of the ecological indices in the study area

Sampling location	qCO_2	NCR	H'	EV	SR	T	D	MI	MMI
NWF	14.4 ± 9.1 a	1.00*a	0.10 ± 0.20 c	0.68 ± 0.22 a	0.18 ± 0.26 c	1.00* c	0.87 ± 0.18 a	2.00* b	2.00* b
OWF	16.9 ± 5.0 a	0.84 ± 0.15 b	0.87 ± 0.58 b	0.79 ± 0.02 a	0.68 ± 0.30 b	1.56 ± 0.43 b	0.36 ± 0.08 b	2.04 ± 0.15 b	2.16 ± 0.36 b
CS	20.6 ± 6.7 a	0.52 ± 0.17 c	1.65 ± 0.53 a	0.75 ± 0.13 a	1.99 ± 0.51 a	2.56 ± 0.76 a	0.21 ± 0.09 b	3.39 ± 0.15 a	3.13 ± 0.38 a

Different letters indicate significant differences between sampling location, $P < 0.05$, n = 44

NWF new coal-ash landfill, *OWF* old coal-ash landfill, *CS* control area that was never used as a landfill, qCO_2, metabolic coefficient, *NCR* nematode channel ratio, *H'* Shannon–Weaver index *SR* species richness, *EV* Evenness, *T* trophic diversity, *D* Simpson's dominance index, *MI* maturity index, *MMI* maturity index modification

* The ecological index had the same value in all samples collected at the NWF

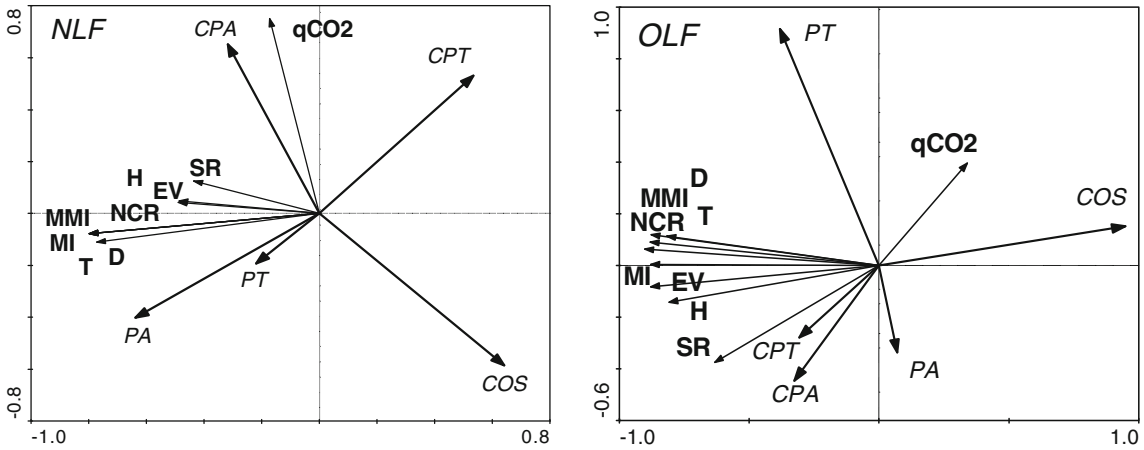

Fig. 5 Redundancy analysis (RDA) performed on values of ecological indices indicates correlation between the habitat variables and soil biotic community structure. The ecological indices showed significant differences between the plant areas and the completely open spaces at a distance from the plants. The length and angle of *arrows* indicate the strength and degree of correlation between the ecological indices and the environment. The first axis of the NLF figure explains 42 % of the total variability in the observed data, with the sum of all canonical eigenvalues amounting to 61 %. The significance of these variations was confirmed by the Monte Carlo permutation test (P value = 0.02; F-ratio = 4.0; number of permutations = 499). The first axis of the OLF figure explains 69 % of the total variability in the observed data, with the sum of all canonical eigenvalues amounting to 72 %. The significance of these variations was confirmed by the Monte Carlo permutation test (P value = 0.01; F-ratio = 20.1; number of permutations = 499). All four eigenvalues in the figures were found to be canonical and correspond to axes that are constrained by the environmental variables. Sampling plots: *NLF* new coal-ash landfill; *OLF* old coal-ash landfill; *COS* completely open space at a distance from the plants; *PA* soil samples under *Alhagi maurorum*; *CPA* soil samples around *Alhagi maurorum*; *PT* soil samples under *Tamarix* sp.; *CPT* soil samples around *Tamarix* sp. Ecological indices: qCO_2 metabolic coefficient; *NCR* nematode channel ratio; *H'* Shannon–Weaver index; *EV* evenness; *SR* species richness; *T* trophic diversity; *D* Simpson's dominance index; *MI* maturity index, *MMI* maturity index modification

Numerous researches have affirmed that trace elements can have a positive effect on soil organic matter (i.e., Li et al. 2005; Iorfa et al. 2011); however, in the current study, trace elements were found to have both a negative and positive effect on soil organic matter. The reason for this difference between our conclusions and those of other research studies is probably the different ways of interaction between trace elements and organic matter, microbial activity, and diversity in the soil of the open and planted areas. Soil microorganisms play a significant role in the environmental fate of toxic metals, affecting transformations between soluble and insoluble phases, exercising mobilization of metals or, conversely, transforming them into insoluble and chemically inert forms (Gadd 2001, 2004). They are also able to accumulate high amounts of different trace elements (Weyman-Kaczmarkowa and Pedziwilk 2000). Organic matter can affect the behavior of trace elements, forming simple and chelate complex compounds with ions of trace elements (Schnitzer and Kerndorff 1981; Krogstad 1983). Dissolved organic-matter molecules contain functional groups capable of complexing metal ions (Saar and Weber 1982) that decrease the sorption and increase the mobility of metals in soils (Ashworth and Alloway 2004). In turn, chemical-element contamination can suppress the activity and development of soil microbial communities, leading to the accumulation of nondecaying soil organic matter (Berg et al. 1991; Pen-Mouratov et al. 2010).

Based on previous research studies, it can be suggested that at the open part of the landfill area, the direct negative effect of trace elements on microbial activity led to deteriorating conditions conducive to the formation of organic-metal chelated complex compounds, reflected in the negative correlation between trace elements and organic matter. In contrast, completely different from the open part of the landfill area, the under-plant environment possibly provides more favorable conditions for microbial activity and—along with the increase of total organic-matter content—promotes the formation of the metal–organic-matter complex compounds as reflected in the positive correlation between the observed trace elements and organic matter. It should be emphasized that the mechanism of interaction between trace elements and organic matter and microorganisms in soil is far from complete and requires further development (Violante et al. 2008; Sessitsch et al. 2013).

The total number of bacteria and fungi, along with microbial biomass and basal microbial respiration, was found to correlate with the observed trace elements. Furthermore, the values of correlation between separate trace elements and microbial biomass, and the same elements and metabolic coefficient (qCO2), were found to be diametrically opposed to each other. Therefore, vanadium and

Table 7 Pearson correlation coefficients between ecological indices, soil properties, and trace-element contents

PA+PT	
qCO_2	**$V^{-0.51*}$, $Cs^{-0.66**}$**
NCR	**$As^{-0.64*}$, $Pb^{-0.55*}$, $Zn^{-0.55*}$, $Mo^{-0.56*}$**
H'	$As^{0.76**}$, $Cd^{0.78***}$, $Sb^{0.59*}$, $Pb^{0.83***}$, $Zn^{0.82***}$, $Mo^{0.80***}$, $W^{0.82***}$, $Cr^{0.73**}$, $Ni^{0.82***}$, $Y^{0.65**}$, **$V^{-0.56*}$**
EV	$Zn^{0.91**}$, $As^{0.81*}$, $Pb^{0.88**}$, $W^{0.93**}$, $Rb^{0.78*}$, $Cd^{0.9**}$, $Mo^{0.89**}$, $Bi^{0.75*}$
SR	$Zn^{0.83**}$, $As^{0.93***}$, $Pb^{0.86**}$, $W^{0.72*}$, $Sr^{0.72*}$, $Cd^{0.83**}$, $Sb^{0.64*}$, $Mo^{0.8**}$, $Cr^{0.67*}$, $Ni^{0.68*}$, $Bi^{0.77**}$
T	$As^{0.63*}$, $Cd^{0.59*}$, $Pb^{0.68**}$, $Zn^{0.68**}$, $Mo^{0.72**}$, $W^{0.77**}$, $Cr^{0.66**}$, $Ni^{0.68**}$
D	$V^{0.63*}$, **$As^{-0.91***}$, $Cd^{-0.89***}$, $Pb^{-0.94***}$, $Zn^{-0.95***}$, $Mo^{-0.95***}$, $W^{-0.96***}$, $Cr^{-0.84***}$, $Ni^{-0.92***}$, $Bi^{-0.80**}$, $Y^{-0.75**}$, $U^{-0.61*}$**
MMI	$W^{0.62*}$,$Cr^{0.58*}$,$Ni^{0.57*}$
CPA+CPT	
qCO_2	**$SM^{-0.51*}$**
NCR	**$Cd^{-0.65*}$, $Sb^{-0.60*}$, $Pb^{-0.62*}$, $Mo^{-0.60}$, $Bi^{-0.67}$**
H'	$OM^{0.50*}$, $Th^{0.46*}$, $Mo^{0.50*}$, $W^{0.67***}$, $Ba^{0.46*}$, $F^{0.87***}$, $W^{0.67***}$, **$Cu^{-0.48*}$, $Rb^{-0.67***}$**
EV	$Rb^{0.8*}$
SR	**$Th^{-0.76*}$**
D	$Th^{0.76**}$, **$As^{-0.62*}$, $Cd^{-0.66*}$, $Pb^{-0.75**}$, $Zn^{-0.76**}$, $Mo^{-0.72**}$, $W^{-0.71**}$, $F^{-0.73*}$**
CS	
EV	$Zr^{0.9*}$
SR	$Pb^{0.9*}$, $W^{0.9*}$, $Sr^{0.9*}$, $Cd^{0.9*}$, $Ga^{0.9*}$, $Y^{0.9*}$, $Nb^{0.9**}$, $Cs^{0.9**}$, $Th^{0.9*}$, $U^{0.9*}$, **$Cr^{-0.9*}$, $Ni^{-0.9*}$**
D	**$SM^{-0.99*}$**, $V^{0.99*}$

Bold indicate negatively correlated values. Different letters indicate significant differences between sampling location, $P < 0.05$, n = 44

PA+PT soil samples collected under plants at the landfills, *CPA+CPT* soil samples collected in outplant plots at the landfills, *CS* soil samples of control area, *qCO_2* metabolic coefficient, *NCR* nematode channel ratio, *H'* Shannon–Weaver index, *SR* species richness, *EV* Evenness, *T* trophic diversity, *D* Simpson's dominance index, *MI* maturity index, *MMI* maturity index modification

cesium were found to be positively correlated with microbial biomass and negatively correlated with the qCO_2, whose value is known to increase when environment stress rises (Anderson and Domsch 1993). The previous research studies showed that some microorganisms possess a bioaccumulation ability to absorb significant amounts of different trace elements, including vanadium and cesium, for chemical defense against predators, competitors, and so on (Avery 1995; Hernández et al. 1998; Odate and Pawlik 2007; Volesky and Holan 1995). Hence, it is logical to suggest that in the observed landfill area, the contribution of microorganisms possessing the ability to bioaccumulate the mentioned trace elements, is significant.

Nematode density was lowest in the NLF in comparison to both the OLF and outside the landfill area, which was considered a control area. No correlation was found between total number of potential prey (soil microorganisms) and predators (soil free-living nematodes) in the observed study area. However, the ecological indices such as species richness in the NLF ($r = 0.84$, $p < 0.01$) and evenness in the OLF ($r = 0.54$, $p < 0.01$) revealed a positive correlation between the microbial biomass and the biodiversity of soil nematodes in the industrial area. In contrast to the applied ecological indices, the Simpson's dominance index indicated that the predominance of separated species in the nematode community decreases with increase of the microbial biomass ($r = -0.91$, $p < 0.01$) in the NLF in contrast to the OLF. These data can also support the above-mentioned suggestion about the increase of local density of microorganisms possessing the ability to bioaccumulate the trace elements.

Moreover, those results indicate that predators (soil nematodes) in the newer successional ecosystem are more closely related to their prey (microorganisms) than in the older ecosystem, which is characterized by a more complex and branched degree of development of trophic relationships, and species diversity (Johnston and Odum 1956; Odum 1971).

Nematode density was found to increase under plants in the landfill area. Similar to nematode density, the amount of culturable fungi was found to increase under plants at the old landfill. These findings can be attributed to data presented by Ingham et al. (1985), who believed that vital nematode activity positively affects microorganism development. At the same time, microorganisms were found to be the important or even the main food resource of soil nematodes in the observed ecosystems. Thus, the females exhibited a positive correlation with bacteria at the older landfill and with fungi at the newer landfill, while the juvenile nematodes, along with a positive correlation with the total number of fungi at the NLF, showed a positive correlation with both the total number of bacteria and the total number of fungi at the OLF. Moreover, nematode biomass was found to be positively correlated with the fungal density in the open area, which suggests that the soil fungi are an important and available food source of the food web in this part of the landfill area.

Similar to Dmowska and Ilieva-Makulec (2006), who conducted their study at power-plant ash dumps near Warsaw, Poland, we found that in the Angren power-plant coal-ash landfill area, the contribution of K-strategist species at the older (abandoned for 10 years) landfill was greater than at the newer (abandoned for 3 years) landfill. However, in contrast to the above-mentioned study (Dmowska and Ilieva-Makulec 2006), our results showed that at the earlier stages of ecosystem succession (the new landfill), *Acrobeloides* and *Cervidellus* were the dominant bacterivore genera, as opposed to *Acrobeloides*, *Aphelenchoides*, and *Aphelenchus*, which were the dominant genera in the power-plant landfill in the Warsaw industrial

area. At the later stages of ecosystem succession (the old landfill), *Acrobeloides* bacterivores, followed by *Longidorella* plant-parasites, were the most numerous nematodes, with a density of about 40 and 25 %, respectively. The omnivore-predator nematodes belonging to K-strategists, in agreement with numerous publications [e.g., Wasilewska (1997); Georgieva et al. (2002)], were very scanty compared to the other trophic groups, amounting to 3 % of the observed nematode community, and they appeared to be concentrated mainly under the shelter of plants of the older landfill, avoiding the new landfill. Moreover, the open spaces distant from plants were the most unfavorable place for soil biota.

One explanation for the sex diversity is that the sex creates variability within a brood of progeny and thereby increases the probability of offspring with superior fitness in unpredictable environments (Ricklefs 1979; Williams 1975; Williams and Mitton 1973). Numerous investigations have shown that heavy metals can affect reproduction, sex ratio, survival, the development of juveniles, and trophic and species diversity (Anderson et al. 2001; Pen-Mouratov et al. 2008). The current study was no exception to the above-mentioned research studies and illustrated that total abundance, biomass, species, trophic and sex diversity of soil free-living nematodes (along with fungi) were affected by trace metals. The almost-complete absence of nematode males in the observed samples, except in the soil samples used as control, should be noted. In contrast to this finding, our previous study, which was conducted in the Almalyk mining and industrial area (Uzbekistan), discovered males of soil free-living nematodes in significant numbers, amounting to 30-50 % of the females (Pen-Mouratov et al. 2008). McSorley (2003) and Papadopoulou and Triantaphyllou (1982), in their study of root-knot nematodes, indicated that environmental change affects the nematode sex ratio, and a variety of stresses may lead to increased production of males. On the other hand, the abundance and development of females along with juveniles are dependent on favorable environmental conditions, including nutrient availability and suitable habitat. This fact suggests that at an early stage of ecosystem succession under such severe conditions, such as coal-ash landfill, the formation of genetic diversity involving nematode males is not so necessary for the nematode community's survival and development strategy. The main aim and strategy of the nematode community in the area is the occupation of free space. Under conditions of unlimited water and nutrients, females, together with juveniles, are more useful for this purpose. The inhospitableness of the coal-ash landfill area affects soil-nematode reproduction. One such effect is the production of mainly female individuals at an early stage of development in the industrial ecosystem (Kahel-Raifer and Glazer 2000).

The widely used ecological indices applied in the present research were sensitive to successive changes of the soil ecosystem at the abandoned coal-ash landfills. Maturity indices have been successfully used to distinguish between well-functioning and disturbed ecosystems (Neher 1999; Yeates and Bongers 1999), as well as to measure the ecological succession status of the soil community (Neher 2001) who showed that the coal-ash landfill area was a more unfavorable habitat for soil biota than the control area. The nematode channel ratio (NCR) [with variation between 1 (bacterial-feeding nematode dominance) and 0 (fungi-feeding nematode dominance) (Moore and Hunt 1988; Yeates 2003)], indicated that the bacterial-based decomposition process was dominant in the newer landfill. However, in the older landfill, and especially in the control area, the fungi-based decomposition process was found to play a more prominent role. The diversity indices, where the Shannon index is sensitive to rare taxa and the Simpson's index is used to measure common taxa (Nehe, 2001), indicated an increase of the contribution of rare species to the older and, hence, more developed ecosystem, whereas in the newer ecosystem, the common nematodes were the main contribution to the soil ecosystem. All applied ecological indices confirmed that open landfill plots distant from plants are the most unfavorable area for soil biota. In this detailed soil microbiological-ecology study, we showed that native plants, in particular *A. maurorum Desv.* and *Tamarix sp*., are the local ecological factor creating niches that moderate the environmental surroundings and participating in forming a new ecosystem in the observed coal-ash landfill area.

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Conflict of interest The authors declare that they have no conflict of interest.

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