

Soil biogeochemical properties of Angren industrial area, Uzbekistan

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Abstract

Background, aim, and scope The present study examined air pollution effects on soil health applying microbiological parameters. It was carried out near the Angren heavy industry complex in a semiarid region of Uzbekistan. This area was selected in order to establish a national monitoring program for assessing environmental condition of areas remote but downwind from greater emission sources. Moreover, little information exists about how air pollution affects microbiological functioning of soils in semiarid and arid regions of the world, and especially those of Central Asia.

Materials and methods Soil samples were collected in May 2005 along a 20-km NE–SW river valley transect downwind from the industrial complex. Soil chemical analyses included electrical conductivity, pH, water soluble Na, Ca, and K, total soluble nitrogen, and mineralizable nitrogen content upon a 1:2 digestion by deionized water. Major elements and heavy metal inventory in solids was measured by X-ray fluorescence and atomic absorption spectrometry.

Microbiological ecosystem properties were assessed by biological indicators such as basal respiration (R_B), microbial biomass related C and N contents, and microbial community functioning coefficients like the metabolic quotient qCO_2 .

Results There was a significant spatial dependence and differences for all soil chemical and microbiological parameters tested. The highest contents were found for the relatively volatile metals Zn ($\leq 1,136$ mg/kg) and Pb (≤ 373 mg/kg) in upper soil layers near the power station suggesting that the metal pollutants are derived from local stack emissions. Soil microflora was obviously affected by heavy metals. Significant positive correlations ($p \leq 0.001$) were found between the metal content, R_B , and qCO_2 , while a negative one was found for the mineralizable N and C_{mic}/C_{org} ratio. A high total number of nematodes was found only most distant from the industrial emission sources.

Discussion The results disclosed remarkable spatial dependence not only of the heavy metal impact onto the soil but also of microbiological soil properties in the study area. The latter suggests bioavailability of the anthropogenic metals in the soil affecting the soil microbial community. This is suggested by less biomass formation and higher qCO_2 values in heavy metal-contaminated compared to less-polluted soil plots.

Conclusions Knowledge of these spatial ecosystem functioning patterns and dependence could be very useful in determining and delineating specific land use and management programs that would be suited and feasible for the highly polluted area. Results of this study can be utilized to develop a monitoring program that may quantify harmful effects on the soil health and impact of any future remediation activities.

Recommendations and perspectives Studies on the relationship between soil biota and pollution levels have raised the

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question regarding the status of natural soil microbial health, stressing the importance of background data of environmental conditions, and elucidating the importance of this environmental monitoring approach even in semiarid and arid regions. Soil microbiological parameters, in particular the metabolic quotient qCO_2 as one of the most sensitive bioindicators identified for that region, should clearly become part of the national environmental monitoring program.

Keywords Bioindicators · Ecotoxicology · Heavy metals · Microbial biomass · Soil contamination

1 Background, aim, and scope

Ecological mine reclamation and soil restoration have become important parts of the sustainable environmental development strategy in many countries. Since the soil microbial communities are responsible for the mineralization of organic matter and nutrient cycling, increasing awareness concerning the health status of soil microorganisms in terrestrial ecosystems has emerged (Yao et al. 2003). Heavy metals are known to affect the growth, activity, and metabolism of microorganisms in soils (Giller et al. 1998; Vig et al. 2003; Hofman et al. 2003). The content of heavy metals in soil can easily reach levels that inhibit the normal development process of soil microorganisms even before causing functional disturbance in other ecosystem compartments. Microbial ecosystem functioning parameters are therefore well-accepted as indicators of overall soil environmental quality (Doran and Parkin 1996; Tate 2000). Soil microorganisms are usually monitored at the process or biomass levels (Baath 1989). The process level includes physiological activities of the soil microorganisms, especially respiration (Sparling 1997; Alon and Steinberger 1999; Sarig et al. 1999). At the biomass level, the entire microbial community is evaluated as a single mass of microbial matter, without specification of its structure. Soil microbial biomass has been found to be sensitive to increased heavy metal concentrations in soils (Valsecchi et al. 1995; Brookes 1995; Kandeler et al. 2000; Klumpp et al. 2003; Shukurov et al. 2005; Yao et al. 2003; Liao and Xie 2007). Carbon dioxide evolution, commonly known as soil respiration in aerobic catabolic processes in the carbon cycle, is also commonly measured. The metabolic quotient, i.e., the ratio of basal respiration to microbial biomass, is inversely related to the efficiency with which the microbial biomass uses the indigenous carbon substrates (Anderson and Domsch 1990). It can become a sensitive indicator for revealing heavy metal toxicity under natural conditions (Wardle and Ghani 1995). A number of other soil microbiological parameters, notably

microbial activity and community structure (Doelman et al. 1994; Kandeler et al. 1996), have been suggested as possible indicators of soil environmental quality affected by heavy metals. Kelly and Tate (1998) found that elevated metal spiking resulted in changes in the structure of the soil microbial communities, as indicated by changes in their metabolic profiles. A metabolic profile can be monitored by the carbon utilization pattern in a Biolog plates system, but changes in the average well color development were not found to be significantly associated with heavy metal loading (Knight et al. 1997; Baath 1989; Yao et al. 2003; Liao and Xie 2007). With more success, the free-living nematode population has been used as a bioindicator because of its ubiquity in soil, known response to chemical and physical perturbations, and consideration in regional and national monitoring programs (Gupta and Yeates 1997; Neher 2001; Shukurov et al. 2005). In the last decade, much attention has therefore been paid on the functional diversity of nematodes as described, e.g., by the maturity index (Bongers 1990; Zhang et al. 2006).

The aims of this study were to (1) determine concentrations of heavy metals in soils of an area severely affected by air pollution, (2) reveal relationships between heavy metal content and both geochemical and biological characteristics of the soil, and (3) evaluate their impact on soil microbial properties by biological indicators such as soil free living nematode abundance, basal respiration, microbial biomass related C and N contents, and microbial eco-physiological indices like the metabolic quotient qCO_2 . Parameter selection was made with respect to what might be useful for a future routine environmental monitoring program.

2 Materials and methods

2.1 Location and environmental situation of the study area

The research area represents a typical semiarid Central Asian mountain valley located along the Akhangaran River between 41°01' and 40°58' North, and 70°10' and 69°57' East, at a height of 900–950 m above sea level. The valley is spread between the mountain ranges of Qurama in the south and Chatkal in the north (foothills of the Tien-Shan), both of ridges enclosing the valley in the northeast. Thermal inversions provide cyclic circulation of air masses and cause pendulum distributions of dust and gas–smoke emissions from the industrial complex. The prevalent wind at the study site is in a western and southwestern direction. The climate is continental with temperature ranges between –30°C in January and +45°C in July. Annual rainfall ranges between 320 and 550 mm, with most of the precipitation in spring and winter. The vegetation cover along the study site

is dominated by annual and perennial plants, where the most common are of the *Astragalus*, *Stipa*, *Medicago*, and *Artemisa* genera. The grassland soils at the study area are classified as *Calcisols* (haplic) with high Ca-carbonate contents and a low content of organic matter with no fertilizer application: about 2 mg g^{-1} total carbon; $60 \text{ } \mu\text{g g}^{-1}$ total nitrogen; $20\text{--}30 \text{ } \mu\text{g g}^{-1}$ total phosphorus; $73\text{--}120 \text{ } \mu\text{g g}^{-1}$ potassium; $60 \text{ } \mu\text{g g}^{-1}$ magnesium, and a slightly alkaline $\text{pH}=7.8\text{--}8.5$ (Makhmudov and Khaitov 2000). The soil is relatively fine-grained with only 2.2% of particle size $2.0\text{--}0.2 \text{ mm}$, 54.5% of $0.2\text{--}0.02 \text{ mm}$, and 43.3% of $<0.02 \text{ mm}$, respectively (Egamberdiyeva and Hoflich 2003; Egamberdiyeva 2007).

The study area is extending into the southeast part of the Tashkent capital of the Republic of Uzbekistan. It hosts a heavy industry complex developed during and after World War II which includes a lignite brown coal open-cast working, a coal firing power station, a gold refinery, and a rubber factory located near the city of Angren along the river valley (Fig. 1). The power station produces 250 MW of electricity and works on the basis of brown coal from the nearby mine. This surface coal mine produces over 2.5 MT of coal every year with an average volatile matter and ash content of 40% and sulfur content around 1.5%. The coal mining and power generation industry uses old equipment that has not been upgraded since the 1990s. Air pollution control technology is therefore in a relatively poor condition. Dust generation from the mines along with fly

ash emissions from the power plant in the range of annually about 0.10 MT over the last half century have caused severe damage to natural ecosystems of the area. The impact of these and the high SO_2/NO_x emissions can already be observed in plants in the form of isolated damage symptoms in plant leaves as impaired growth and necroses (Talipov et al. 1996). Although the exact role played by their heavy metal and organic pollutant inventory in harming the biosphere has not yet been established, it is plain that their share of responsibility for ecotoxicological damage cannot be underestimated. Previous studies showed that surface- and groundwater in this area, as well as soil and vegetation, are highly contaminated with heavy metals (UNECE 2002; Sanitation Country Profile 2004). However, detailed studies on the effects of air pollutants on soil quality in the region are still lacking.

2.2 Soil sampling and soil analyses

Soil samples were collected in May 2005 along a 20-km NE–SW river valley transect downwind from the industrial complex. The soil samples were collected from the topmost 0–10 and 10–20 cm layers in eight sampling locations: (A) grassland at the border of the coal open pit mine (0 km, latitude/longitude: $41^\circ 01' \text{ N}/70^\circ 10' \text{ E}$), (B) east of the power station, industrial site, grassland (4 km, $41^\circ 00' \text{ N}/70^\circ 08' \text{ E}$), (C) west of the power station, industrial site, grassland (6 km, $41^\circ 00' \text{ N}/70^\circ 07' \text{ E}$), (D) nearby the coal-

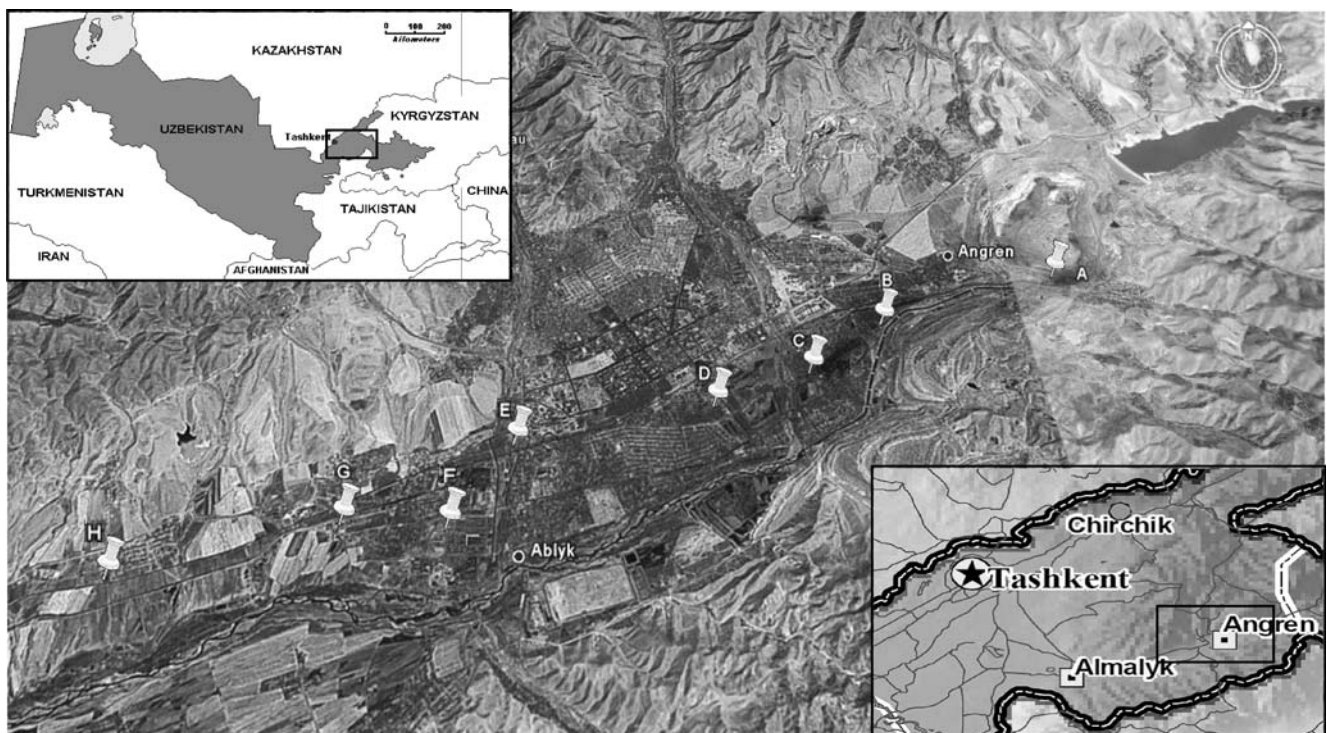


Fig. 1 Location of the study area (Google Earth map of the Angren Valley, with sampling spots pin-pointed)

ash depository, gardening area (8 km, 40°57' N/70°05' E), (E) summer camp, recreation area, grassland, and urban park (12 km, 40°58' N/70°03' E), (F) near the rubber factory, industrial site (14 km, 40°58' N/70°02' E), (G) near the gold refinery factory, arable land (16 km, 40°58' N/70°00' E), and (H) pasture site, grazing area (20 km, 40°58' N/69°57' E). Three random replicate samples were collected at each location from both soil layers, placed in individual plastic bags and transported to the laboratory. The soils were 2-mm mesh sieved for plant detritus and kept in cold storage at 4°C before biological and geochemical analyses.

Soil moisture (SM) was measured gravimetrically from drying subsamples in an oven to constant weight at 105°C. All data presented in this study relies on that dry weight basis. Total organic carbon (C_{org}) was determined using a modified method by Rowell (1994). The method is based on soil organic matter oxidation by K-dichromate, but does not oxidize coal dust. Extraction of water-soluble compounds was performed using distilled water with a soil to extract solution to a ratio of 1:2. These analyses included potentiometric pH and electrical conductivity (EC) measurements by electrodes, total soluble nitrogen (TSN) by an auto-analyser (Skalar Analytical; Houba et al. 1987), and water-soluble cations (Ca^{2+} , K^+ , Na^+) by a flame photometer (Ciba-Corning 410). Major element inventory in the solids was determined using X-ray fluorescence spectrometry (XRF, Philips). Oven-dried soil subsamples were ground in an agate mortar and homogenized before preparation of powder pellets for the XRF analysis. Trace metal concentrations were determined by atomic absorption spectrometry (AAS, Varian) upon sample digestion with conc. HNO_3 and conc. HClO_3 (3:1 ratio). Analytical quality assurance/quality control (QA/QC) was performed on basis of certified reference materials (Montana soil CRM's 2710 and 2711).

2.3 Nematodes and microbial community analyses

Nematode population was determined by extraction from 100 g soil subsamples using the Baermann funnel procedure (Cairns 1960). The recovered organisms were preserved in formalin and counted using a compound microscope (Steinberger and Sarig 1993). Soil microbial biomass carbon (C_{mic}) and the basal respiration (R_{B}) was determined using the classical chloroform fumigation incubation method (Jenkinson and Powlson 1976; Anderson and Domsch 1978; Heinemayer et al. 1989; Sparling and West 1990; Kaiser et al. 1992). Ten grams of soil samples were adjusted to 40% water-holding capacity and fumigated by a CHCl_3 -saturated atmosphere in a desiccator for 24 h. The fumigated and corresponding non-fumigated (control) samples were then transferred to closed 0.5-L glass vessels and incubated for 10 days at 25°C in the dark. Gas

chromatography (GC, Perkin-Elmer) was used to measure the CO_2 concentration in the head space of the glass vessels. The microbial biomass carbon was calculated as $C_{\text{mic}} = [(\text{CO}_2\text{-C from fumigated soil}) - (\text{CO}_2\text{-C from control sample})]/k_{\text{C}}$, given in units $\mu\text{g C g}^{-1}$ soil. A factor of $k_{\text{C}} = 0.41$ was used as proposed by Anderson and Domsch (1990). The samples that were used for measuring soil microbial biomass carbon C_{mic} were also used to measure soil microbial biomass nitrogen, N_{mic} , according to Sparling and Zhu (1993). The fumigated and non-fumigated subsamples were extracted with 25 mL of 0.01 M CaCl_2 solution by shaking for 1.5 h (Houba et al. 1987). Mineralizable nitrogen (E_{N} , $\mu\text{g g}^{-1}$) was estimated as the difference nitrogen in the fumigated soil samples minus the nitrogen in the non-fumigated samples. The amount of microbial biomass nitrogen was calculated as $N_{\text{mic}} = E_{\text{N}}/k_{\text{EN}}$, with a k_{EN} factor of 0.54 (Joergensen and Mueller 1996). The metabolic quotient ($q\text{CO}_2$) was calculated as the ratio between R_{B} and C_{mic} and given in units of $(\mu\text{g CO}_2\text{-C})/(\mu\text{g C}_{\text{mic}} \text{ day})^{-1}$ according to Anderson and Domsch (1990), or equal to day^{-1} . Note that this has to be divided by 24 to be equal to h^{-1} as an alternative unit for $q\text{CO}_2$ common in literature. Since the microbial biomass, C_{mic} , is strongly connected to the soil C_{org} contents, the microbial coefficient known as the ratio $C_{\text{mic}}/C_{\text{org}}$ was determined in order to evaluate substrate availability (Insam et al. 1989, 1996). All data were ultimately subjected to statistical univariate and multivariate analysis using the common SAS code (ANOVA, Duncan's multiple range tests, and Pearson correlation coefficients) to evaluate the spatial dependence between the soil properties and pollution status.

3 Results

3.1 Spatial heavy metal and C_{org} distributions

The analysis of spatial variance presented in Tables 1 and 2 indicates a highly significant dependence of organic carbon and heavy metal concentrations on transect direction, sampling location, and sampling depth. C_{org} exhibited the highest concentration of 2.4% in the upper soil layer at location A (coal mining site) and is decreasing along the transect reaching a mean value of 0.35%. The concentration of most metals (Cu, Zn, Ba, Pb, Th, and U) in soil samples collected along the transect shows a gradual decrease with increasing distance from the industrial pollution sources. The concentration of Zn, Pb, Ba, U, and Th were three to five times higher than their Clarkes values (calculated average values of metals in pristine soils and sediments) and local background levels, with mean value levels of 1,136, 2,430, 373, 12, and 29 mg kg^{-1} , respectively. The highest contents were found for Zn (up to 1,136 mg kg^{-1})

Table 1 Observed F and p values (ANOVA) for geochemical and microbiological properties of soils in the study area

Soil property	Transect location		Soil depth	
	F test	p Value	F test	p Value
Total nematodes abundance, T_{Nem}	21.6	≤ 0.0001	2.7	NS
C_{org}	45.9	≤ 0.0001	92.1	≤ 0.0001
Total soluble nitrogen, TSN	9.7	≤ 0.0001	13.2	≤ 0.0001
pH	1.6	NS	0.03	NS
Electrical conductivity, EC	3.5	0.006	2.2	NS
Ca^{2+}	17.3	≤ 0.0001	0.4	NS
Na^+	2.8	0.02	0.6	NS
K^+	21.9	≤ 0.0001	30.0	≤ 0.0001
Basal respiration, R_B	6.4	≤ 0.0001	44.4	≤ 0.0001
Microbial biomass, C_{mic}	22.4	≤ 0.0001	56.3	≤ 0.0001
Metabolic quotient, qCO_2	9.1	≤ 0.0001	3.2	NS
Microbial coefficient, C_{mic}/C_{org}	15.2	≤ 0.0001	5.1	0.0003
V	96.0	≤ 0.0001	11.9	0.001
Cr	174.6	≤ 0.0001	11.7	0.001
Co	31.9	≤ 0.0001	0.1	NS
Ni	117.8	≤ 0.0001	4.6	0.04
Cu	29.5	≤ 0.0001	24.2	≤ 0.0001
Zn	219.5	≤ 0.0001	72.9	≤ 0.0001
Ba	108.7	≤ 0.0001	16.2	0.0002
Pb	122.2	≤ 0.0001	94.5	≤ 0.0001
Th	137.1	≤ 0.0001	11.7	0.001
U	62.5	≤ 0.0001	18.7	≤ 0.0001

and Pb (up to 373 mg kg⁻¹) in upper soil layers near the power station (Fig. 2). Some other metals (V, Cr, Co, Ni) show an opposite distribution tendency with lower concentrations near the emission sources. Metals of this second

group were found at or below Clark values and local background levels. Significant differences ($p \leq 0.0001$, $N = 8$ locations \times 2 depths \times 3 replicates = 48) were observed between the upper (0–10 cm) and lower (10–20 cm) soil

Table 2 Correlation coefficients between soil microbiological and geochemical properties

	T_{Nem}	C_{org}	R_B	C_{mic}	qCO_2	C_{mic}/C_{org}
C_{org}	-0.02		0.40**	0.27**	0.12	0.19
TSN	0.09	0.44**	0.07	0.16	-0.28**	-0.14
pH	-0.05	0.18	0.20	0.30**	0.05	-0.12
EC	0.01	0.25**	0.02	-0.22**	0.12	0.04
Ca^{2+}	0.33**	-0.32**	-0.10	0.19	-0.20	0.43**
Na^+	-0.12	0.07	0.16	-0.04	0.21	0.14
K^+	0.01	0.46**	-0.05	-0.03	-0.12	-0.19
V	0.23*	-0.22**	0.01	0.16	-0.24*	0.35**
Cr	0.14	-0.24**	-0.16	0.12	-0.32**	0.24*
Co	0.19	-0.16	-0.16	0.11	-0.29**	0.19
Ni	0.13	-0.24**	-0.19	0.14	-0.36**	0.27*
Cu	-0.10	0.66**	0.35**	0.00	0.40**	-0.30**
Zn	0.94**	0.58**	0.32**	0.00	0.29**	-0.28**
Ba	-0.15	0.52**	0.30**	-0.09	0.49**	-0.32**
Pb	-0.11	0.61**	0.36**	0.00	0.33**	-0.28**
Th	-0.23*	0.43**	0.23**	-0.10	0.41**	-0.35**
U	-0.17	0.53**	0.34**	-0.08	0.45**	-0.30**

*, **Correlation coefficients significant at $p \leq 0.05$ and $p \leq 0.01$ (in bold), respectively

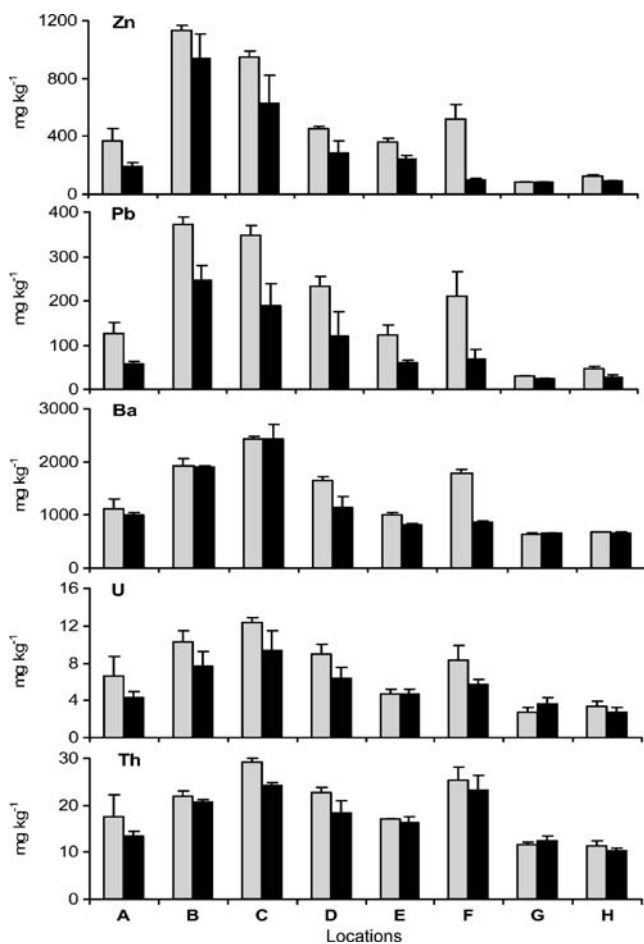


Fig. 2 Heavy metal distribution along the transect at two soil depths (gray 0–10 cm, black 10–20 cm)

layers for the first group of metals, with higher concentrations being found in the upper soil layer at all sampling sites (see Table 1).

3.2 Water soluble components

Soil extract samples are alkaline with pH levels in between 7.7 and 8.1. No significant differences between sampling locations and soil depth were found for the pH values along the 20-km downwind transect (see Table 1). Soil electrical conductivity (EC) reached highest levels at the first four sampling locations (A, B, C, D) near the emission sources, with values of 0.4–0.5 mS g⁻¹, while at locations E and F, a lower value of 0.33 mS g⁻¹ was found (see Table 1). A significant sampling location effect was also found for soluble Ca²⁺, K⁺ ($p \leq 0.0001$), and Na⁺ ($p \leq 0.02$) (see Table 1). Maximum TSN values were found in the upper soil layer near the Angren coal open pit, power station, and rubber factory (8.9, 6.0, and 6.8 mg kg⁻¹ at locations A, B, and F, respectively). Minimum values were found west of power station and gold refinery factory at locations C and G.

3.3 Nematode population

Nematode population densities, T_{Nem} , were found to change significantly ($p \leq 0.0001$) along the sampling gradient (see Table 1). The population size of nematodes in the soil was found to increase with distance away from the power station. Minimal numbers of nematodes (23–38 individuals per 100 g soil in 0–10 cm and 52–59 in 10–20 cm layers) were found at sampling locations B and C near the power station. Toward the end of the transect at location H (pasture area), the largest numbers of nematodes were counted with mean values of 1,154 in the upper, and 668 individuals per 100 g soil in the deeper soil layers (Fig. 3). Significant positive correlation was found between soluble Ca²⁺, Zn, and T_{Nem} (see Table 2).

3.4 Biomass carbon and nitrogen, basal respiration, metabolic coefficients

The soil microbial biomass, C_{mic} , reached a maximal value at location E of 990 and 531 $\mu\text{g C g}^{-1}$ soil in samples taken from the upper (0–10 cm) and deeper (10–20 cm) layers, respectively. Minimal values for C_{mic} were measured near the pollution sources (91–335 $\mu\text{g g}^{-1}$ in upper soil layer, and 35–96 $\mu\text{g g}^{-1}$ soil in deeper soil layer; see Fig. 3). Statistical analyses revealed that significant differences ($p \leq 0.0001$) were found along the downwind transect (see Table 1). C_{mic} show also a significant ($p \leq 0.01$) positive correlation with C_{org} and pH, and negative correlation ($p \leq 0.05$) with EC (see Table 2). Microbial biomass nitrogen, N_{mic} , was found to follow the C_{mic} pattern, with relatively lower values of 0.1–1.4 $\mu\text{g g}^{-1}$ at locations F and G near the pollution sources. Highest values were found at the sites E and H remote from the emission sources (5.2–5.7 $\mu\text{g g}^{-1}$ in upper and 3.4–3.9 $\mu\text{g g}^{-1}$ in deeper soil layers, see Fig. 3). Soil basal respiration, R_B , shows also significant differences between sampling locations and soil layers ($p \leq 0.0001$, see Table 1). Highest values for R_B were found near the power station at locations B and C, with mean values of 561–629 and 221–343 $\mu\text{g CO}_2\text{-C g}^{-1} \text{ day}^{-1}$ soil in the upper and deeper soil layers, respectively. At location E (recreation area), R_B shows vertical distribution pattern different from other locations, with a value measured in upper soil layer three times lower than the value measured in deeper soil layer (188 and 510 $\mu\text{g CO}_2\text{-C g}^{-1} \text{ day}^{-1}$, respectively, see Fig. 3). Statistical analysis showed a positive correlation between R_B , C_{org} , and the second group of metals ($p \leq 0.01$, see Table 2).

Values of the eco-physiological status, $q\text{CO}_2$, of the soil microbial community show significant ($p \leq 0.0001$) differences between sampling location, but significant differences with depth were not found (see Table 1). As a result of significant changes in C_{mic} , R_B , and N_{mic} , $q\text{CO}_2$ values

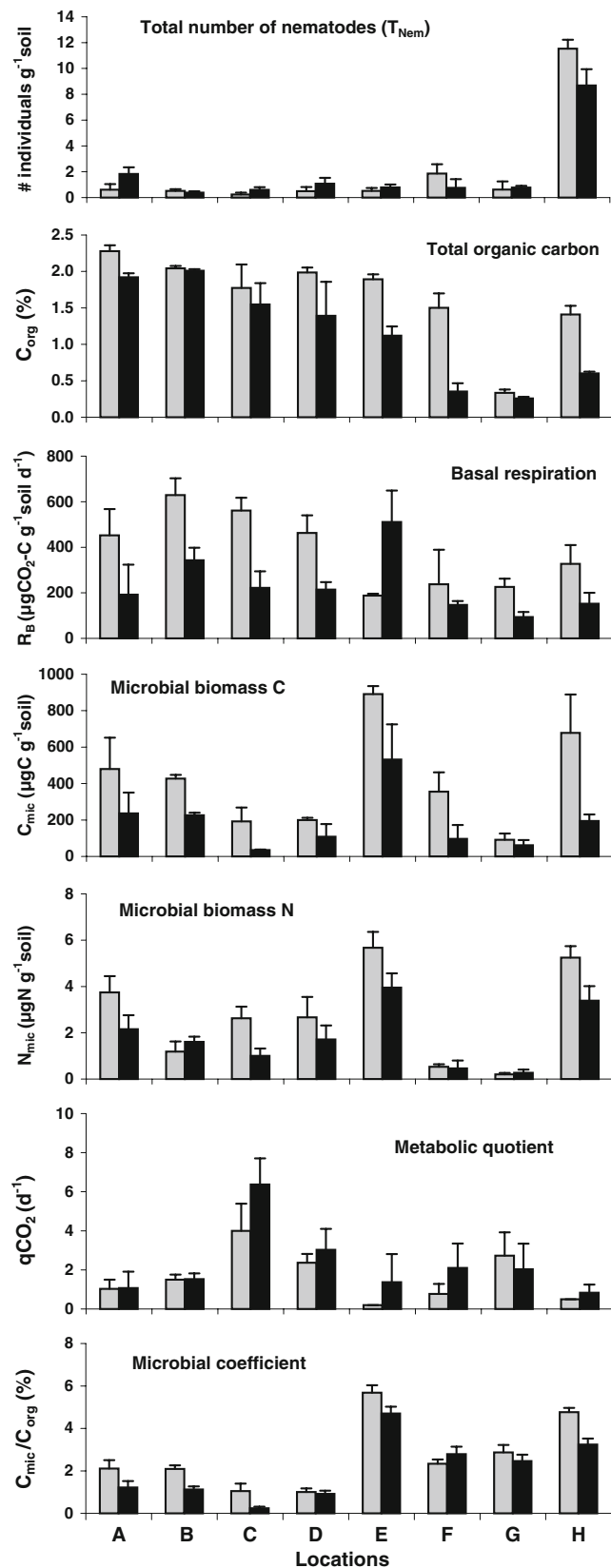


Fig. 3 Microbiological ecosystem functioning parameters along the transect at two soil depths (0–10 and 10–20 cm)

were found to decrease from a maximal value of 4.0–6.7 $\mu\text{g CO}_2\text{-C } \mu\text{g}^{-1} C_{\text{mic}} \text{ day}^{-1}$ at location C (near the power station) to 0.2–1.4 $\mu\text{g CO}_2\text{-C } \mu\text{g}^{-1} \text{ day}^{-1}$ at locations E and H (recreation and pasture areas, Fig. 3). A significant positive correlation was found between the $q\text{CO}_2$ values and the first group of metals, and a negative one with content of the second group of trace metals ($p \leq 0.01$, see Table 2).

The $C_{\text{mic}}/C_{\text{org}}$ ratio representing substrate availability was found to be relatively similar in the first two locations (A, B) with values ranging from 2.0% to 2.1% in upper to 1.1% to 1.2% in deeper soil layers. The lowest value of 0.2% was found at location C (near the power station) in deeper soil layer. At location D (ash depository), no significant variation was found between soil layers. On the other hand, the highest values of $C_{\text{mic}}/C_{\text{org}}$ ratio were found for locations E and H (recreation and pasture areas), with values of 5.7–4.8% for upper and 4.7–3.2% for deeper soil layers. Locations F and G (rubbery and gold refinery factories) showed intermediate levels (see Fig. 3). Statistical analyses show significant variation with locations ($p < 0.0001$) and depths ($p < 0.0003$; see Table 1). Significant ($p < 0.05$) negative correlation was found between $C_{\text{mic}}/C_{\text{org}}$ ratio values and concentrations of the first group of trace elements (Cu, Zn, Ga, Sr, Y, Zr, Nb, Ba, Pb, U, and Th; see Table 2).

4 Discussion

The concentrations of volatile trace metals in the ash produced by combustion of coal are significantly higher than those in the raw coal (Swaine and Goodarzi 1995). Heavy metals emitted from coal-fired power plants to atmosphere are in a highly mobilizable form as a part of flue gas (e.g., Hg) or condensed on the emitted particulates during cooling of the flue gas (e.g., Zn, Pb; Kersten and Förstner 1995). The inability of aged air pollution control equipment to capture the metal-laden fine-grained particulates may therefore cause environmental and human health problems. The location of the study sites were selected in relation to the main downwind direction. Earlier studies by Hutchinson and Whitby (1974), as well as by Kandeler et al. (1999) support our findings that trace elements released into the atmosphere from coal combustion accumulate mainly in the top soil layer. This is corroborated by a rapid and gradual decrease in concentration with increasing distance from the emission sources. Results disclosed remarkable spatial dependence not only of the metal impact onto the soil but also of soil organic matter and microbiological soil properties in the study area (see Figs. 2 and 3). The accumulation of organic matter in the most polluted area indicates reduced substrate utilization efficiency by the

microorganisms rather than a coal dust impact. Moreover, the concentration distribution pattern suggests that origins of all studied heavy metals were divided in two groups, (1) anthropogenic (airborne) source for Cu, Zn, Ba, Pb, Th, and U, and (2) lithogenic (geochemical) source for V, Cr, Co, and Ni. Contents of the first group of metals showed a clear gradient with increased concentrations toward the power station. The highest contents were found for the relatively volatile metals Zn and Pb in upper soil layers near the power station suggesting that the metal pollutants are derived from the stack emissions. Although a high level of carbonate content and relatively high pH values in the studied soils might reduce mobility of these pollutants, a deleterious effect to soil microbial properties is obvious on the basis of statistical analysis. Less volatile elements such as Th and U are retained in the solid combustion wastes. Although the concentration of these radioactive elements in solid combustion wastes are still approximately ten times the concentration in the original coal, uranium concentration of fly ashes ($10\text{--}30\text{ mg kg}^{-1}$) is still in the range found in granitic and phosphate rocks, and shales.

In our study, the studied biotic soil components and their indices showed a clear relationship with the pollutant concentration in soil. Such detrimental effects of heavy metals and other soil contaminants have repeatedly been reported in the literature (Hattori 1992; Bardgett et al. 1994; Kandeler et al. 1996; Wilcke et al. 1999; Klumpp et al. 2003). The soil free-living nematode population, e.g., exhibited a significant positive response to a decrease in soil heavy metal contamination because of its biological function as a well-known bioindicator (Wardle et al. 1995; Yeates and Williams 2001). On the other hand, the significant positive correlation between total number of nematodes, T_{Nem} , and Zn concentration can be explained with essentiality of this element for living organisms. Zhang et al. (2006) found a significant reduction in the T_{Nem} only to occur at the highest Zn ($400\text{--}800\text{ mg kg}^{-1}$) treatment. During their pot experiments, T_{Nem} increased at intermediate Zn ($100\text{--}200\text{ mg kg}^{-1}$) concentrations, compared with the control. Lower Zn concentrations apparently stimulate, while higher concentrations inhibit total number of nematodes and their community structure, i.e., the Zn effect on soil nematodes can be positive or negative depending on its concentration in soil. In our case, Zn show accordingly elevated concentration ($400\text{--}1000\text{ mg kg}^{-1}$) near the pollution sources in locations B, C, and D, and decreasing T_{Nem} , while other locations remote from the sources showing less Zn levels ($100\text{--}400$) and has a significant positive correlation with T_{Nem} (see Table 2, Fig. 3). Microbial biomass as well as CO_2 evolution patterns were found to increase gradually in response to the decrease in heavy metal concentration at the two soil layers, yielding in a clear inverse significant relationship

with the pollutant concentration in the soil. These results are also supported by similar studies (Kandeler et al. 1996, 1999). Moreover, the ecophysiological quotient $q\text{CO}_2$, which represents a specific physiological status evaluating the environmental effect on the soil microbial community, revealed a small difference between the two soil layers, but a significant change toward the end of the transect mainly affected by heavy metal distribution. Results from our study on microbial indices were found to be within the range of values reported by Hofman et al. (2003), working on a monitoring microbial biomass and respiration program aimed to determine the relationship between soil health properties and contamination. The microbial coefficient interpreted as substrate availability exhibited a significant increase at stations E and H complementary to the significant decrease in eco-physiological status.

Regression analysis yielded in significant positive correlations between first group of metals and soil organic carbon (C_{org}) and a negative correlation with $C_{\text{mic}}/C_{\text{org}}$ (see Table 2). The lowest values for the latter parameter in both soil depths were found at the most polluted sites A–D (see Fig. 3). Usually, there is a close positive relationship between both C_{mic} and C_{org} contents in uncontaminated soils, but this relationship is quite weak in the present soil samples. Therefore, the ratio of the biomass C to organic C can be used as an indicator of the effects of metals on the functioning of a soil ecosystem (McGrath et al. 1995). A low ratio value with higher heavy metal concentration indicates that more substrate is diverted toward catabolic processes at the expense of anabolic processes leading to reduced microbial biomass in the long run (Sparling 1997; Brookes 1995; Yao et al. 2003). Basal respiration (R_{B}) showed a less pronounced response to the pollution impact. This is mainly due to a cross-correlation with soil organic carbon. The decrease by distance from the emission sources of the metabolic quotient $q\text{CO}_2$, which revealed a significant positive correlation with metal load, suggests bioavailability of the anthropogenic metals in the soil affecting the microbial community. On the other hand, metals from the second lithogenic metal group showed negative correlation with $q\text{CO}_2$ and positive correlation with $C_{\text{mic}}/C_{\text{org}}$. Chander and Brookes (1991) reported less biomass formation from labeled substrate and higher $q\text{CO}_2$ values in metal-contaminated compared to uncontaminated soils. Note that although a plausible explanation, a high microbial $q\text{CO}_2$ value in metal-contaminated soil is in itself no proof of either a higher maintenance-energy requirement or lower substrate-utilization efficiency. Nonetheless, $q\text{CO}_2$ values can serve as an important indicator of soil health quality and can be closely related to pollution as also concluded recently by Liao and Xie (2007).

5 Conclusions and recommendations

This preliminary monitoring study applied a wide range of geochemical and microbiological parameters to characterize the air pollution impact on the soil health in the Angren industrial complex region. Variability of individual parameters differed widely and significantly among sampling sites and transect directions. Although detrimental effects and cross-correlation by persistent organic pollutants (POP's) cannot be ruled out on the basis of this study, similar effects of heavy metals on the soil microbiological parameters have repeatedly been reported in the literature as discussed in the introduction. Because of its well-known function as a bioindicator, the soil free-living nematode population exhibited a most sensitive indicator of unpolluted soil, and hence appear to be useful for an evaluation of any future efforts for the recovery of the polluted soil sites. The metabolic quotient qCO_2 revealed a small difference between the two soil layers, but a significant effect in the most air polluted sampling sites. Since the microbial coefficient, C_{mic}/C_{org} , exhibited a significant increase at stations E and H in an inverse manner to the significant decrease in the ecophysiological status, both parameters seem to elucidate complementarily the sensitivity of the microbial community to soil contamination. We conclude that both coefficients (or even a ratio thereof) are the most useful bioindicator of soil pollution in that region. Based on the results of the present study and on previous investigations in the same and similar regions (Talipov et al. 1996; Shukurov et al. 2005), an extensive environmental monitoring of the industrial regions of Angren (and Almalyk) is proposed. Such a program should include not only the determination of the soil inorganic and POP inventory, but also the aforementioned (minimum) three biological soil health parameters.

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References

- Alon A, Steinberger Y (1999) Response of the soil microbial biomass and nematode population to a wetting event in nitrogen-amended Negev desert plots. *Biol Fertil Soils* 30:147–152
- Anderson JPE, Domsch KH (1978) Physiological method for quantitative measurement of microbial biomass in soils. *Soil Biol Biochem* 10:215–221
- Anderson TH, Domsch KH (1990) Application of eco-physiological quotients (qCO_2 and qD) on microbial biomass from soils of different cropping histories. *Soil Biol Biochem* 22:251–255
- Baath E (1989) Effects of heavy-metals in soil on microbial processes and populations (a review). *Water Air Soil Pollut* 47:335–379
- Bardgett RD, Speir TW, Ross DJ, Yeates GW, Kettles HA (1994) Impact of pasture contamination by copper, chromium, and arsenic timber preservative on soil microbial properties and nematodes. *Biol Fertil Soils* 18:71–79
- Bongers T (1990) The maturity index: an ecological measure of environmental disturbance based on nematode species composition. *Oecologia* 83:14–19
- Brookes PC (1995) The use of microbial parameters in monitoring soil pollution by heavy metals. *Biol Fertil Soils* 19:269–279
- Cairns EJ (1960) Methods in nematology. In: Sasser JN, Jenkins WR (eds) *Nematology—fundamentals and recent advances with emphasis on plant parasitic and soil forms*. University of North Carolina Press, Chapel Hill, NC, pp 33–84
- Chander K, Brookes PC (1991) Effects of heavy-metals from past applications of sewage-sludge on microbial biomass and organic matter accumulation in a sandy loam and silty loam UK soil. *Soil Biol Biochem* 23:927–932
- Doelman P, Jansen E, Michels M, Van TM (1994) Effects of heavy metals in soil on microbial diversity and activity as shown by the sensitivity–resistance index, an ecologically relevant parameter. *Biol Fertil Soils* 17:177–184
- Doran JW, Parkin TB (1996) Quantitative indicators of soil quality: a minimum data set. In: Doran JW, Jones AJ (eds) *Methods for assessing soil quality*. *Soil Sci Soc Am Spec Publ No 49*, Madison, WI, pp 25–38
- Egamberdiyeva D (2007) The effect of plant growth promoting bacteria on growth and nutrient uptake of maize in two different soils. *Appl Soil Ecol* 36:184–189
- Egamberdiyeva D, Hoflich G (2003) Influence of growth-promoting bacteria on the growth of wheat in different soils and temperatures. *Soil Biol Biochem* 35:973–978
- Giller KE, Witter E, McGrath SP (1998) Toxicity of heavy metals to micro-organisms and microbial processes in agricultural soils: a review. *Soil Biol Biochem* 30:1389–1414
- Gupta VVSR, Yeates GW (1997) Soil microfauna as bioindicators of soil health. In: Pankhurst CE, Doube BM, Gupta VVSR, Grace PR (eds) *Soil biota management in sustainable farming systems*. CAB Intern, Oxon, UK, pp 201–233
- Hattori H (1992) Influence of heavy-metals on soil microbial activities. *J Soil Sci Plant Nutr* 38:93–100
- Heinemayer O, Insam H, Kaiser EA, Walenzik G (1989) Soil microbial biomass and respiration measurements: an automated technique based on infrared gas analysis. *Plant Soil* 116:191–195
- Hofman J, Bezchlebova J, Dusek L, Dolezal L, Holoubek I, Andel P, Ansorgova A, Maly S (2003) Novel approach to monitoring of the soil biological quality. *Environ Intern* 28:771–778
- Houba VJG, Novozamsky I, Vittenbogaard J, Van Der Lee JJ (1987) Automatic determination of total soluble nitrogen in soil extracts. *Landw Forsch* 40:295–302
- Hutchinson TC, Whitby LM (1974) Heavy-metal pollution in the Sudbury mining and smelting region of Canada. I. Soil and vegetation contamination by nickel, copper and other metals. *Environmental Conservation* 1:123–132
- Insam H, Parkinson D, Domsch KH (1989) Influence of macroclimate on soil microbial biomass. *Soil Biol Biochem* 21:211–221
- Insam H, Amor K, Renner M, Crepaz C (1996) Changes in functional abilities of the microbial community during composting of manure. *Microb Ecol* 31:77–87
- Jenkinson DS, Powlson DS (1976) The effects of biocidal treatments on metabolism in soil. V. A method for measuring soil biomass. *Soil Biol Biochem* 8:209–213
- Joergensen RG, Mueller T (1996) The fumigation–extraction method to estimate soil microbial biomass: calibration of the k_{EN} value. *Soil Biol Biochem* 28:33–37

- Kaiser EA, Mueller T, Joergensen RG, Insam H, Heinemeyer O (1992) Evaluation of methods to estimate the soil microbial biomass and the relationship with soil texture and organic matter. *Soil Biol Biochem* 24:675–683
- Kandeler E, Kampichler C, Horak O (1996) Influence of heavy metals on the functional diversity of soil microbial communities. *Biol Fertil Soils* 23:299–306
- Kandeler E, Tschermo D, Spiegel H (1999) Long-term monitoring of microbial biomass, N mineralisation and enzyme activities of a Chernozem under different tillage management. *Biol Fertil Soils* 28:343–351
- Kandeler E, Tschermo D, Bruce KD, Stemmer M, Hobbs PJ, Bardgett RD, Amelung W (2000) Structure and function of the soil microbial community in microhabitats of a heavy metal polluted soil. *Biol Fertil Soils* 32:390–400
- Kelly JJ, Tate RL (1998) Effects of heavy metal contamination and remediation on soil microbial communities in the vicinity of a zinc smelter. *J Environ Qual* 27:609–617
- Kersten M, Förstner U (1995) Speciation of trace metals in sediments and combustion waste. In: Ure A, Ch D (eds) *Chemical speciation in natural systems*. Chapman & Hall, London, pp 234–275
- Klumpp A, Hintemann T, Lima JS, Kandeler E (2003) Bioindication of air pollution effects near a copper smelter in Brazil using mango trees and soil microbiological properties. *Environ Pollut* 126:313–321
- Knight B, McGrath SP, Chaudri AM (1997) Biomass carbon measurements and substrate utilization patterns of microbial populations from soils amended with cadmium, copper, or zinc. *Appl Environ Microbiol* 63:39–43
- Liao M, Xie XM (2007) Effect of heavy metals on substrate utilization pattern, biomass, and activity of microbial communities in a reclaimed mining wasteland of red soil area. *Ecotox Environ Safety* 66:217–223
- Makhmudov MM, Khaitov R (2000) Effective use of steppe pastures and recommendations for raising productivity. *ICARDA* 26, Tashkent.
- McGrath SP, Chaudri AM, Giller KE (1995) Long-term effects of metals in sewage sludges on soils, microorganisms and plants. *J Ind Microbiol* 14:94–104
- Neher DA (2001) Role of nematodes in soil health and their use as indicators. *J Nematol* 33:161–168
- Rowell DL (1994) *Soil science: methods and applications*. Longmans Group UK, London
- Sarig S, Fliessbach A, Steinberger Y (1999) Soil microbial biomass under the canopy of coastal sand dune shrubs. *Arid Soil Res Rehab* 13:75–80
- Sanitation Country Profile (2004) Uzbekistan, <http://www.un.org/esa/agenda21/natlinfo/countr/uzbek/sanitatzb04f.pdf>
- Shukurov N, Pen-Mouratov S, Steinberger Y (2005) The impact of Almalyk industrial complex on soil chemical and biological properties. *Environ Pollut* 136:331–340
- Sparling GP (1997) Soil microbial biomass, activity and nutrient cycling as indicators of soil health. In: Pankhurst CE, Doube BM, Gupta VVSR (eds) *Biological indicators of soil health*. CAB International, Wallingford, pp 97–120
- Sparling GP, West AW (1990) A comparison of gas chromatography and differential respirometer methods to measure soil respiration and to estimate the soil microbial biomass. *Pedobiologia* 34:103–112
- Sparling GP, Zhu C (1993) Evaluation and calibration of biochemical methods to measure microbial biomass C and N in soils from Western Australia. *Soil Biol Biochem* 25:1793–1801
- Steinberger Y, Sarig S (1993) Responses by soil nematode populations in the soil microbial biomass to a rain episode in the hot, dry Negev Desert. *Biol Fertil Soils* 16:188–192
- Swaine DJ, Goodarzi F (1995) Environmental aspect of trace elements in coal. Kluwer, Dordrecht, p 312
- Talipov R, Shukurov N, Tverskaya K (1996) Geochemical study of soils, water and vegetation for estimation of anthropogenic pollution of Chirchik and Akhangaran rivers valleys. *Uzbek Geol J* 1:44–49
- Tate RLIII (2000) *Soil microbiology*, 2nd edn. Wiley, New York, p 508
- UNECE (United Nations Economic Commission for Europe) (2002) *Environ Perform Rev Ser 14: Uzbekistan*, 196 pp
- Valsecchi G, Gugliotti C, Farini A (1995) Microbial biomass, activity, and organic matter accumulation in soils contaminated with heavy metals. *Biol Fertil Soils* 20:253–259
- Vig K, Megharaj M, Sethunathan N, Naidu R (2003) Bioavailability and toxicity of cadmium to microorganisms and their activities in soil: a review. *Adv Environ Res* 8:121–135
- Wardle DA, Ghani A (1995) A critique of the microbial metabolic quotient (qCO₂) as a bioindicator of disturbance and ecosystem development. *Soil Biol Biochem* 27:1601–1610
- Wardle DD, Yeates GW, Watson RN, Nicholson KS (1995) The detritus food-web and the diversity of soil fauna as indicators of disturbance regimes in agroecosystems. *Plant Soil* 170:35–43
- Wilcke W, Guscher C, Kobza J, Zech W (1999) Heavy metal concentrations, partitioning, and storage in Slovak forest and arable soils along a deposition gradient. *J Plant Nutr Soil Sci* 162:223–229
- Yao H, Xu J, Huang C (2003) Substrate utilization pattern, biomass and activity of microbial communities in a sequence of heavy metal-polluted paddy soils. *Geoderma* 115:139–148
- Yeates GW, Williams PA (2001) Influence of three invasive weeds and site factors on soil microfauna in New Zealand. *Pedobiol* 45:367–383
- Zhang XK, Li Q, Wang SB, Jiang Y, Liang W (2006) Effect of zinc addition to soil on nematode community structure. *Bull Environ Contam Toxicol* 76:589–594