



Mobile forms and migration ability of Cu, Pb and Zn in forestry system in Poland

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Abstract The total concentrations of Cu, Pb and Zn and their bioavailability were studied in forest soil and vegetative communities (grass and wild strawberries) collected from two areas (A and B). Two single extraction methods were used for the evaluation of the availability of Cu, Pb and Zn in forest soils. The total metal concentrations ranged from 27.43 to 143.34 mg/kg for Zn, 49.82 to 95.84 mg/kg for Pb and 0.96 to 3.24 mg/kg for Cu in soil samples A and 43.55 to 89.65, 50.62 to 117.59 and 0.95 to 4.37 mg/kg, respectively, in soil samples B. The Pb and Zn concentrations in some soil samples were significantly elevated with respect to the background levels of Poland area. The mean concentrations of Zn, Pb and Cu in grass samples from area A were 71.10, 37.48 and 1.81 mg/kg, respectively, and they were higher than the corresponding values in grass from area B. The concentrations of Zn, Pb and Cu in fruits of wild strawberries, amounting to 59.89, 19.05 and 2.71 mg/kg, respectively, were at the similar level as the metal concentrations in grass. The highest level of grass contamination was found for Pb, and their concentrations were above the critical ranges. The mean EDTA-extractable Zn, Pb and Cu concentrations in soils samples A were 20.69, 17.30 and 0.50 mg/kg, respectively, and in soils samples B 14.10, 23.67 and 0.46 mg/kg. Correlation between the concentrations of heavy metals in grass and the chemical parameters of soil (pH and OM), the transfer factor values and the total and extractable metal concentrations in soil were calculated.

Keywords Available forms of metals · Soil · Grass · Wild strawberry

Introduction

The origins of trace metals in soil ecosystem include both natural and anthropogenic sources. For the natural sources, soil-forming parent material and processes have a strong impact on metal concentrations in soils. The influence of human activities (industry and economic) include mining and smelting operations for metallic ores (Chen et al. 2008), sewage irrigation, urban sludge application, waste incineration, construction wastes and airborne dusts.

Industry activities are more concentrated in cities, and urban areas have become the geographic focus of resource consumption and chemical emission, which may cause environmental problems. In comparison with agricultural soils, urban soils are usually more strongly influenced by anthropogenic activities, fuel combustion, waste disposal and traffic emissions (Wong et al. 2006; Morel and Heinrich 2008). Heavy metals are transported with gases and aerosols on long distances. It is known that air pollution with heavy metals leads to their accumulation in forest soil ecosystems. The increased acidity of forest soils can have an impact on the increase of their availability and their higher uptake rate by plants. The transfer of heavy metals from soil to plant is a complex and dynamic process influenced by soil and climatic conditions, and plant genotype (Kabata-Pendias 2004). Numerous studies have shown that elevated concentrations of some metals in environmental systems not only influence the quality of the surrounding environment, but also pose a potential hazard to biota, including humans. The bioavailability of trace metals in soil should be considered as a key factor in

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assessment of the risk to ecosystem and human health. The most common studied sites include industrial areas with emissions of pollutants (Bullock and Gregory 1991), roadside greenbelts in relation to traffic emissions (Chen et al. 2010), recreational areas (Shi et al. 2008) and national parks. Staszewski et al. (2012) present results of screening analysis of 23 Polish national parks contamination with Cu, Pb, Zn and Cd (metal concentration in needles of trees and metal concentrations in soils). The mobility of trace metals in soil, determined by solid-solution partitioning, could be assessed by a sequential leaching procedure or by single chemical extraction (Tessier et al. 1979; Quevauviller et al. 1994; Umoren et al. 2007; Rao et al. 2008; Anju and Banerjee 2011; Peters 1999). Among the studied metals, Pb has no essential biological function, but is taken up by plants from metal-enriched soils. It is a typical pollutant, passively taken up by plants both from the subsoil and from the polluted air with a dust fall. Zn and Cu are essential plant nutrients; however, they can cause toxicity at elevated concentrations (Kabata-Pendias and Pendias 1992). When any or several of these elements are present in soil above their respective background concentrations, remedial actions may be necessary (Valcho et al. 2008).

The results of the partitioning study reported by Umoren et al. (2007) showed that zinc prevailed in the more soluble fractions and was distributed between the acid-extractable (32.4 %) and the reducible (40.3 %) fractions, whereas Pb was distributed mainly in the reducible fraction. Copper was predominantly associated with the reducible and residual fractions. In a single chemical extraction, the choice of extracting agent depends on specific aims of a certain investigation. EDTA, DTPA and acetic acid extractions are often used for studies on physicochemical processes in soils like trace metal mobility (Peijnenburg et al. 2007). Gupta and Sinha (2007) used various single extractant to evaluate the bioavailability of heavy metals from tannery wastewater contaminated soil and translocation of metals to the plant of *Brassica juncea*. Environmental pollution with heavy metals has a harmful impact not only on the biological activity of soils but also on the quality and yield of crops and—in consequence—on the health of humans and animals (Ure 1996).

Natural metal content in soils are of geogenic origin and are weathering products, while anthropogenic metals are mainly deposited on the soil surface. Heavy metals of anthropogenic origin are usually expected to accumulate in the topsoil (Oleszek et al. 2003). The basic assumption of sequential extractions in relation to soil contamination is that metals from anthropogenic sources are more readily extractable than from lithogenic and pedogenic sources (Kaasalainen and Yli-Halla 2003). The pattern of metals accumulation in plants, regarding the type soil and plants, the nature of trace metal and metal concentrations is

discussed by many investigators (Xinghui et al. 2010; Grytsyuk 2006; Niesiobedzka 2012).

In this work, the total and the EDTA, H₂O_{distilled}-extractable metals (Cu, Pb and Zn) concentrations in soils and the transfer of these elements to grass and wild strawberries were investigated. These metals may enter the food chain posing an important risk to human health. In order to limit the accumulation of heavy metals in plants, a good understanding of the transfer characteristics of the heavy metals from soil to the plant is necessary and should always be investigated for the particular soil–plant system. The ground part of the grass, fruits of wild strawberry and soils were studied to evaluate the degree of metal contaminations of soil–plant ecosystem in relation to traffic emission from two roads. Correlation analysis was used to explore the relationships between measurements of the metal concentrations in plant tissues and the total and EDTA-extractable concentration of the metals in soils, TF values and soil properties (pH and OM).

Materials and methods

Study area and sampling

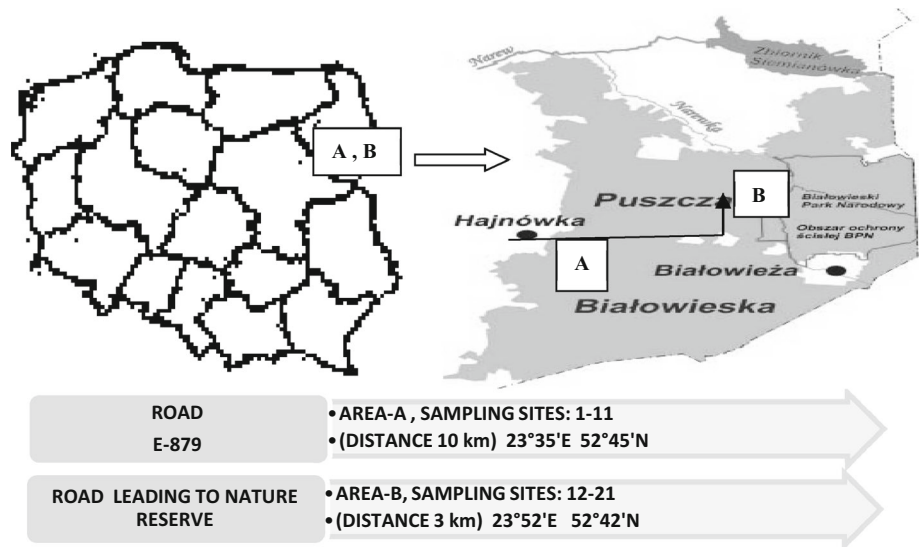
The Bialowieza National Park is situated in the northeast part of Poland. The Park covers the central part of Bialowieza's Forest. The Park covers the area of 10,517.27 ha, which constitutes 1/6 of the Polish part of Bialowieza's Forest. 5725.75 ha is under strict protection, 4438.20 ha is under active protection, and landscape protection covers the area of 353.32 ha. There has been a protection zone created around the Park which covers the state commercial forest having an area of 3224.26 ha. Bialowieza National Park protects the best-preserved fragment of Bialowieza's Forest—last natural forest at the European Lowland Area, having the primaeval character, identical with the one which covered the area of deciduous and coniferous forests years ago.

Soil and plant samples

The soil, grass and wild strawberries samples were taken from Bialowieza National Park in Poland in June 2012. Eleven sampling sites were located along the road 689 with high traffic intensity from Hajnowka to Bialowieza (area A, distance 3 km), and ten sampling sites were located along the forest road leading to the car park in front of the nature reserve (area B, distance 10 km) (Fig. 1).

Twenty-one samples of soil (from the depth 0–20 cm) and grass (aboveground part) were taken from two areas: 11 from area A and 10 from area B). Fruits of wild strawberries were sampled at six locations in area B (on

Fig. 1 Sampling sites of studied soils from two areas (sampling sites: A and B)



area A they did not grow). Soil samples were air-dried and screened through a 1-mm sieve. The grass and strawberries fruits samples were washed in distilled water and dried at 60 °C to constant weight. Then, they were milled into powder for measurement of metal concentrations.

Chemical analysis of studied samples

Chemical analysis of soils: pH and content of organic matter (OM) were carried out by standard techniques used in Poland (Ostrowska et al. 1990). In order to determine the total metal concentration, 1 g of dried samples (soils, grass and wild strawberries) was mineralized by a mixture (3 ml HNO₃ + 1 ml HClO₄) according to the method reported by Jones and Case (1990). The reagents composition and digestion conditions were chosen in order to achieve complete mineralization and the composition of the solid phase into the liquid phase. All the reagents used were of trace pure grade (from Merck). After digestion, the solutions were adjusted to 50 ml with deionized water. For analysis of available metal forms in soil, air-dried soil samples were extracted by 0.05 mol/l ethylene-diamine-tetra-acetic acid disodium (pH 7.0). One hundred milliliter of the EDTA solution was added to 10 g of soil sample placed in polypropylene tubes. The content of tubes was shaken on a rotating shaker (for 1 h). After shaking, the samples were centrifuged. The soluble form of heavy metals was examined by extraction of soil samples (10 g) with 100 ml distilled water. Metal concentrations in the supernatant liquid were measured with a flame atomic absorption spectrometry (FAAS). Three replications were conducted for each sample. Quality assurance and quality control (QA/QC) for metals in soil samples were estimated by determining metal concentrations in the standard solutions

(Merck, Darmstadt, Germans). Limits of detection (mg/kg dry matter) for Cu, Pb and Zn were 0.001, 0.003 and 0.001, respectively. The quality control of measurements was assured by test analyses of the TILL-3 reference material from Canada (Certificate of analysis 1995). The obtained results are summarized in Table 1.

The relative differences (in %) between measured by AAS and certified concentrations fluctuated within a range: for Cu: 94–98 %, for Pb: 98–104 % and for Zn: 97–103 %.

Transfer factor of heavy metals in soil–plant ecosystem (TF)

The soil-to-plant factor (transfer factor, TF) was defined as the ratio of metal concentration in plant and total metal concentration in the soil on which the plant has grown expressed in mg/kg of dry mass (Eq. 1):

$$TF = C_{\text{plant}}/C_{\text{soil}} \tag{1}$$

where C_{plant} is the element concentration in plant (mg/kg) and C_{soil} is the element concentration in the soil

Table 1 Measured and certified values of Cu, Pb and Zn in the TILL-3 reference material

Metal	Cv (mg/kg)	AAS (min and max) (mg/kg)	D (%)
Cu	22	20.7	94
		21.6	98
Pb	26	25.5	98
		27	104
Zn	56	54	97
		57.7	103

Cv certified value, AAS measured value, D the relative difference between measured by AAS and certified concentrations in %

compartment (mg/kg). The TF values are commonly used for quantifying elements uptake by plant (Sheppard and Evenden, 1990). TF is constant by definition and based on the assumption of a linear relationship between plant and soil concentration.

Statistical analysis

The basis statistics and the correlation significance were performed using Microsoft Office Excel 2007 for Windows (Correlation Data Analysis Tool). Pearson's correlation was used to explore the relationships between measurements of availability of the metals in soils and the concentrations of the metals in plant tissues. The hypothesis testing in the statistical analysis is based on a 0.05 significance level. The values of Pearson's correlation coefficient (*R*) can perhaps explain the plant metal uptake accounted by effective concentration of the metals in soil and resin-extractable concentrations of the metals.

Results and discussion

Chemical properties of soils

The results (Table 2) showed some differences in soil characteristics. The values of pH ranged from acid to mild

alkaline, and it was location dependent. In detail, 11 soil samples A and 10 soil samples B ranged from 6.58 to 7.78 and from 5.84 to 7.82, respectively. The content of organic matter in soil samples from two areas A and B varied 3.30–12.85 and 11.94–58.37 %, respectively.

The total metal concentrations ranged from 27.43 to 143.34 mg/kg for Zn, 49.82 to 95.84 mg/kg for Pb and 0.96 to 3.24 mg/kg for Cu in soil samples A and 43.55 to 89.65, 50.62 to 117.59 and 0.95 to 4.37 mg/kg, respectively, in soil samples B. The highest values of Zn concentration were detected in soil samples A area. In all soil samples, the metal concentrations were much higher than corresponding values reported by Staszewski et al. (2012). In the study carried out in 2004 in 23 Polish national parks, the mean concentrations of Cu, Pb and Zn in soils of Bialowieza National Park were 0.08, 0.09 and 3.89 mg/kg d.w. In comparison with data presented for Polish north-eastern soils reported by Niesiobedzka (2000), the concentrations of Cu, Pb and Zn in studied soils were much higher. In the study carried out in 2000, the concentrations of Cu, Pb and Zn in soils of Bialowieza National Park were 3.1, 18.0 and 37.0 mg/kg, respectively. The lowest concentrations in soils were observed for Cu at two studied areas. The highest concentrations of Pb and Zn in some soil samples were above the permissible ranges given by Kabata-Pendias and Pendias (1992), and they were significantly elevated with respect to the background levels of

Table 2 Chemical properties of soils (pH, OM) and heavy metal concentrations in soils

Sampling area	Statistical parameters	pH	OM (%)	Zn (mg/kg)	Pb (mg/kg)	Cu (mg/kg)
A	<i>n</i> = 11					
	Mean	7.44	7.65	75.90	69.22	1.94
	Range	6.58–7.78	3.30–12.85	27.43–143.34	49.82–95.84	0.96–3.24
	SD	0.43	4.74	38.05	17.95	0.86
B	<i>n</i> = 10					
	Mean	6.90	30.11	55.39	76.90	2.46
	Range	5.84–7.42	11.94–58.37	43.55–89.65	50.62–117.59	0.95–4.37
	SD	0.75	18.86	16.30	20.96	1.02

Mean mean value, *Range* range value, *SD* standard deviation

Table 3 Heavy metal concentrations (mg/kg) in grass and wild strawberries

Sampling area	Statistical parameters	Grass			Wild strawberries		
		Zn	Pb	Cu	Zn	Pb	Cu
A	<i>n</i> = 11						
	Range	37.86–108.41	18.41–116.94	1.00–2.89			
	SD	24.11	39.07	0.66			
B	<i>n</i> = 10				<i>n</i> = 6		
	Range	33.86–84.50	16.07–65.71	1.12–5.21	49.34–70.43	15.44–21.11	2.55–2.84
	SD	20.06	19.47	1.63	1.91	2.18	0.12

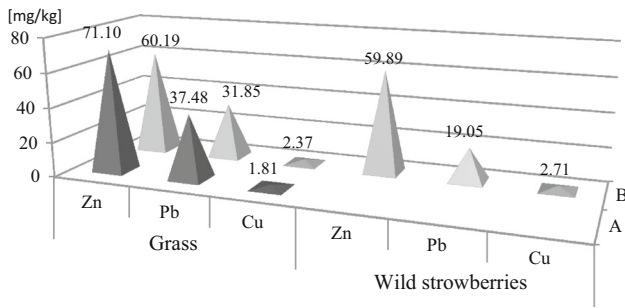


Fig. 2 Heavy metal concentrations (mg/kg) in grass and wild strawberries

Poland area. The critical soil total concentration is defined as the range of values above which toxicity is considered to be possible. High level of contamination of soils by Zn and Pb is probably due to the atmospheric emissions from the traffic that significantly increase the concentrations of these metals in the upper horizons of the neighboring soils. Comparing the effect of the traffic pollution on soil–plant system in area A to the effect on soil–plant ecosystem in area B, it shows the same trends. The values of average concentrations of studied metals allowed one to arrange them in order of occurrence, depending on their concentrations, in the following way: Zn > Pb > Cu.

Heavy metal concentrations in plants

The results of metal concentrations in grass and wild strawberries (range and SD) are presented in Table 3 and Fig. 2 (mean values).

The mean concentration of Zn, Pb and Cu in grass from area A was 71.10, 37.48 and 1.81 mg/kg, respectively, and they were higher than the corresponding values in grass samples from area B. The concentrations of Zn, Pb and Cu in wild strawberries, amounting to 59.89, 19.05 and 2.71 mg/kg, respectively, were at the similar level as the metal concentrations in grass. According to the research of Ciepala and Rycman (1996), the concentrations of Zn, Pb and Cu in leaf of wild strawberry fluctuated within a large

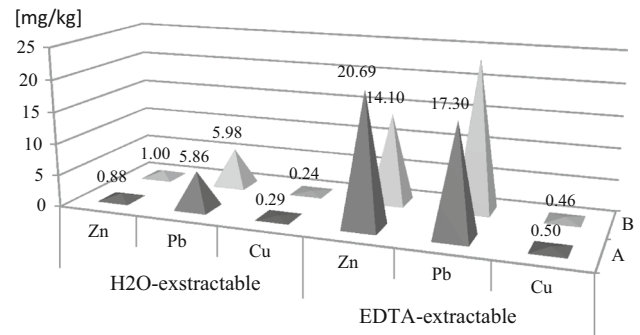


Fig. 3 Extractable metal concentrations (mg/kg) in soils

range 100–140, 31.2–145 and 5–15 g/g, respectively. The highest level of grass enrichment was found for Pb, and its concentrations were above the critical ranges, reported by Kabata-Pendias and Pendias (1992).

Concentrations of H₂O_{distilled} and EDTA-extractable metal concentrations in soils

The concentrations of extractable Zn, Pb and Cu with distilled water and EDTA (Table 4; Fig. 3) showed significant variations related most specifically with both areas and type of metal.

The EDTA-extractable forms’ concentrations of Zn, Pb and Cu were much higher than H₂O-extractable forms’ concentrations (for Zn: 14–20, for Pb: 3–4 and for Cu: 1.5–2 times). The mean EDTA-extractable Zn, Pb and Cu concentrations in soils samples A were 20.69, 17.30 and 0.50 mg/kg, respectively, whereas the distilled water-extractable Zn, Pb and Cu concentrations were in each case markedly low 0.88, 5.86 and 0.29 mg/kg, respectively. The mean EDTA-extractable Zn, Pb and Cu concentrations in soils samples B were 14.10, 23.67 and 0.46, respectively. The distilled water-extractable Zn, Pb and Cu concentrations were at the level 1.00, 5.98 and 0.24 mg/kg, respectively. Zeng et al. (2011) noted lowest values for EDTA-extractable metal concentrations in China (1.40–12.23 mg/

Table 4 Concentrations of extractable metals in soils (mg/kg)

Sampling area	Statistical parameters	Water-extractable metals			EDTA-extractable metals		
		Zn	Pb	Cu	Zn	Pb	Cu
A	<i>n</i> = 11						
	Range	0.54–1.41	4.75–6.27	0.22–0.39	7.54–52.84	10.67–26.04	0.15–0.79
	SD	0.31	0.56	0.06	17.25	6.83	0.21
B	<i>n</i> = 10						
	Range	0.47–2.34	5.10–6.62	0.15–0.44	5.88–28.41	13.02–50.64	0.31–0.74
	SD	0.79	0.58	0.12	9.54	15.61	0.18

Table 5 Proportion of extractable metals in the total concentrations in soils (%)

Sampling area	Statistical parameters	Water-extractable metals			EDTA-extractable metals		
		Zn	Pb	Cu	Zn	Pb	Cu
A	<i>n</i> = 11						
	Range	0.5–5.1	6.3–11.8	9.1–30.6	13.8–44.2	11.1–44.4	5.3–77.8
	SD	1.69	1.98	9.75	10.38	12.42	23.76
B	<i>n</i> = 10						
	Range	0.5–5.2	4.3–13.1	5.3–28.2	13.4–44.4	19.1–50	7.1–53.6
	SD	1.84	3.45	9.96	13.05	12.73	21.54

kg for Zn, 2.04–9.74 mg/kg for Pb and 1.38–5.66 mg/kg for Cu).

The proportion of H₂O_{distilled} and EDTA-extractable metals in total metal concentrations in soils

The proportion of each element extractable with distilled water and EDTA gives information about metal availability. The proportions of Zn, Pb and Cu extracted with EDTA from soils indicated (Table 5; Fig. 4), on average, 29.4, 28.4 and 40.1 % availability forms, respectively, in soil samples A and 24.8, 28.9 and 27.4 % availability forms, respectively, in soil samples B.

It has been found that for the soil samples A the order of metal availability was Cu > Zn > Pb. The availability for Zn, Pb and Cu in soil samples B generally formed the order

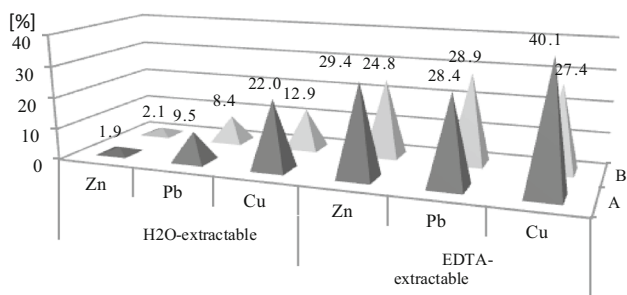


Fig. 4 Proportions of extractable metals in soils from two sampling areas

of Pb > Cu > Zn. In these soils, availability showed wide variations, which can be explained by the variability of the soil parameters (pH, OM) but also by the contamination route of these soils (soil solution, dust deposition, emissions from the traffic).

The mean values of TF (Table 6; Fig. 5) from soil (area A) to grass of Zn, Pb and Cu were higher than the corresponding values for samples from area B. Transfer factor coefficients from soil to wild strawberries fluctuated with the mean values for Zn, Pb and Cu were 1.23, 0.36 and 1.35, respectively. The TF values of studied metals from soil to wild strawberries were at the similar level to the TF values of metals from soil to grass.

To assess the potential mobility and bioavailability of Zn, Pb, and Cu in soils, the statistical correlations between the metal concentrations in grass and the chemical parameters of soil (pH and OM), the transfer factor values, the total metal and the extractable metal concentrations in soil were calculated (Table 7).

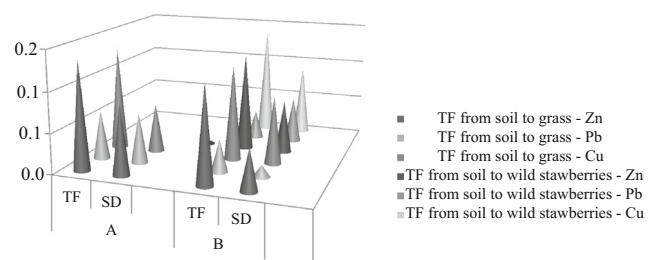


Fig. 5 Mean of TF values and standard deviation (SD)

Table 6 Transfer factor values (TF)

Sampling area	Statistical parameters	TF from soil to grass			TF from soil to wild strawberries		
		Zn	Pb	Cu	Zn	Pb	Cu
A	<i>n</i> = 11						
	Range	0.60–2.75	0.29–1.87	0.66–2.41			
	SD	0.88	0.63	0.61			
B	<i>n</i> = 10				<i>n</i> = 6		
	Range	0.38–1.66	0.16–0.65	0.37–2.41	0.44–1.41	0.18–0.50	0.42–2.36
	SD	0.52	0.15	0.85	0.66	0.58	0.88

Table 7 Correlation coefficient values between the metal concentrations in grass and the soil properties (pH and OM), the total and EDTA-extractable metal concentrations in soils and the TF values (correlation is significant at the 0.05 probability level)

Parameter	pH	OM (%)	Zn soil	Pb soil	Cu soil	TF(Zn)	TF(Pb)	TF(Cu)	Zn EDTA
Zn grass			0.363			0.331			0.408
Pb grass	-0.677			0.168			0.943		
Cu grass		-0.165			0.147			0.644	
Zn EDTA			0.923						
Pb EDTA				0.439					
Cu EDTA								0.991	

It was observed that pH and OM were not significantly correlated with Cu and Zn concentrations in grass. Only the Pb concentration in grass was significantly correlated with pH ($R = -0.677$). This result demonstrates that the soil pH is important parameter that influenced the toxicity in Pb-contaminated soils. Generally, heavy metals display the highest mobility at low pH values, although Cappuyns and Swennen (2008) studied different leaching tests were applied on soil samples in order to obtain information on the potential mobility of heavy metals. For example, Zn is only released at a pH below 6 from soil to soil solution. Cu is also released to some extent at alkaline pH values. This is due to complexation of these metals with DOC or inorganic ligands (e.g., OH⁻). For Cu, a higher leachability was even observed at pH 10 than at pH 2 in examined soils. The Cu concentration in grass showed linear negative correlation with OM ($R = -0.165$). No significant correlations were observed between the concentrations of Pb and Cu in grass and the total concentrations of metals in soils. The positively correlation coefficient was found for only Zn ($R = 0.363$). The amount of Cu, Pb and Zn extracted by EDTA and their total concentrations in soils showed linear positive correlation, which are statistically significant (R values for Cu, Pb and Zn being 0.991, 0.439 and 0.923, respectively, and P values being <0.005). The concentrations of Cu, Pb and Zn in grass were positively correlated with the TF values (R values: 0.644, 0.943 and 0.331, respectively). Among all the analyzed metals only in the case of Zn, a statistical relationship between the heavy metal concentration in grass and the EDTA-extractable Zn concentrations in soils was found ($R = 0.408$).

Conclusions

The high concentration of Pb and Zn in the soils may be explained by the fact that the pollutants are present in the soils as a result of the contact with the polluted atmosphere by emissions from the traffic that significantly increase the concentrations of these metals in the upper horizons of the soils.

The concentrations of metals in wild strawberries were at the similar level as the metal concentrations in grass. The

high concentrations of Pb and Zn in some grass and strawberries samples were above the critical ranges given by Kabata-Pendias and Pendias (1992). The highest level of grass contamination was found for Pb.

Transfer factor can be used to evaluate the bioavailability of heavy metals in soils, but it requires great caution. Among all the analyzed metals only in case of Zn, a statistical relationship between the heavy metal concentrations in grass and EDTA-extractable metal concentrations in soils was found. The result suggests that the single extraction (EDTA extractions) can be helpful to estimate the bioavailability of Zn in the studied soils, but tests must be conducted in a specific area and type of soil. No statistically significant dependency was found for Cu and Pb.

The results showed that heavy metals (Pb and Zn) in fruits strawberries could be potentially transferable to the human and could then contribute, to a considerable share, to the exposure of the population brought about through consumption of fruits from the area.

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