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Trace Element Accumulation in Selected Bioindicators Exposed to Emissions along the Industrial Facilities of Danube Lowland

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The scope of this study was to investigate the effects of the atmospheric emissions of two significant industrial centres of the Danube Lowland: the Schwechat and Slovnaft refineries. In this connection the environmental pollution of emission zones by heavy metals on the base of selected bioindicators (soil, lucerne, wheat, earthworms) should be evaluated. The phenomenon of the atmospheric impact of heavy metal aerosols on the food chain and substance transfer for the model studied (soil - plants - terrestrial invertebrates) was quantified by transfer (bioaccumulation) factors. Due to the orographic depression between the Eastern Alps and Western Carpathian Mountains and the characteristic atmospheric circulation with a predominant north-west wind direction, heavy metal aerosols from closed industrial activities are expected to influence the quality of the surrounding environment. However, most of the 112 top soil samples taken from arable fields in the Schwechat District (Austria) showed natural levels of heavy metals. Heavy metal uptake by wheat via the soil was also rather low. At the Slovakian monitoring site the heavy metal bioaccumulation within the lucerne, as a consequence of availability from the soil, was slightly increased and a considerably higher transfer factor was recorded in related earthworm tissues. Cadmium content in the earthworms exceeded the recommended limit for agricultural feed.

Key Words: heavy metals, soil, lucerne, wheat, earthworms, bioaccumulation

Introduction

Heavy metals may be important trace elements in the nutrition of plants, animals or humans (e.g. Zn, Cu, Mn, Cr, Ni, V), while others are not known to have positive nutritional effects (e.g. Pb, Cd, Hg). However, all of these may cause toxic effects (some of them at a very low content level) if they occur excessively. The bioaccumulation of heavy metals over large territories and long time periods, which may result in the gradual damage of living organisms, necessitates careful monitoring of the input, mobility and effects of these pollutants¹.

The bioaccumulation of trace elements in living organisms and biomagnification in them means or describes the processes and pathways of these (possible) pollutants from one trophic level to another, thus exhibiting the higher bioaccumulation ability in organisms related to the higher living status. Increasing

concentration through the foodchain is caused by a higher retention time of toxic substances than of the general food components in the organisms².

Heavy metals are emitted from a great number of sources which contribute to metal loads in terrestrial and aquatic food chains. These elements are released into the environment as a result of a wide range of industrial activities as well as from the combustion of fossil fuels.

Cadmium enters the environment via three main routes: refining and use of cadmium, copper and nickel smelting and fuel combustion.

Vanadium compounds are widely distributed in the earth's crust and are present at an average concentration of 100 mg.kg⁻¹ of vanadium - roughly equal to the concentration of zinc and nickel. Vanadium and nickel occur in fossil fuels where they exist as a porphyrin complex³.

The increases in heavy metals in the environment are assumed to result primarily from fly ash formed during burning of fossil fuels. Vegetation, airborne soil particles, volcanic aerosols and forest fires contribute considerably to natural emissions of trace elements. Current anthropogenic metal emissions have resulted in soil and water toxic element concentrations up to several orders of magnitude higher than estimated natural concentrations. High levels of lead contamination occur near highways and cities due to the combustion of leaded petrol⁴.

Inputs of heavy metals into soils have gradually increased over the past decades as a consequence of agricultural practices (fertilisers, pesticides), disposal of sewage sludges, landfilling and from atmospheric deposition. Unlike other environmental compartments (i.e. atmosphere, aquatic bodies) heavy metals in soil are characterised by long residence times⁵.

The objective of this study was to evaluate the effects of atmospheric emissions of two significant industrial centres of the Danube Lowland, Schwechat and Slovnaft refineries, to evaluate the phenomenon of atmospheric impact of airborne heavy metals on the food chain and substance transfer for the model: soil - plants - terrestrial invertebrates⁶⁻⁸. Some other possible contributions to heavy metal concentrations in the soil of the monitored south-east sites of the refineries were evaluated. For this purpose the results of two different studies carried out in Slovakia and Austria were evaluated and summarised.

Experimental

Study area and sampling strategy

Study areas for heavy metals monitoring at various food chain levels including soil substrates represented a south-east quadrant up to 5 km distance from the industrial refinery centres of Schwechat and Slovnaft.

The continental climate with a mild zone typical of this area is determined by the wind conditions of the Danube Lowland and the Low Carpathian Mountains. The orographic situation with its characteristic air circulation results in the prevailing north-west wind direction of mean velocity 5.2 m.s⁻¹. The mean annual temperature is 9.8°C; the warmest month is July and the coldest January. The long-term average annual precipitation in the area is 550 mm.

The Slovakian sampling zone may be characterised as an industrial-residential one, and the Austrian region as predominantly agricultural. With respect to soil classification, both soil types are rich in carbonates (e.g. Calcaric Fluvisol).

At the Slovakian sampling zone 10 localities for sampling were selected, i.e. situated according to Propositions for Sampling by Emissions Measurements STN 83 5511⁹. The sampling of top soil, vegetation

and worms was done manually over a period of several months in summer (1996–1998). Sample drawing in Schwechat was described in detail by Spiegel⁸.

In the Schwechat district (Austrian region) soil samples were taken from 112 sampling sites on arable land which were evenly distributed over the district with particular consideration of the soil types. Eighty-seven soil samples were taken from 0-25 cm soil depth and a further 25 samples were taken at 25-50 cm and 50-75 cm depth. At each sampling site 24 soil samples were taken; this represents an area of ca. 1.5 ha respectively.

On the base of the industrial-residential nature of the Slovakian sampling zone, the most common plant bioindicator - lucerne (*Medicago sativa* L.) - was collected about 2 cm over the soil horizon. At the Schwechat emission site plant samples were chosen from 10 localities, and the whole wheat plant (*Triticum aestivum*) just before flowering was taken according to the methodology of Bergmann¹⁰.

Lumbricid earthworms were collected in Sloznaft from the soil by the method of digging and hand-sorting and transferred in their native soil to the laboratory where they were rinsed clean from adhering soil particles with tap water and maintained under moist conditions for several days in PE flasks to allow the final emptying of consumed gut material¹¹.

Samples Pretreatment

Top soil samples were taken from both emission sites, dried in a laboratory drier at 40°C for about 20 h, screened across a 2 mm mesh sized synthetic sieve and homogenised in an agate mill.

Plants and biota samples were dried for about 24 h at 105°C in the laboratory dryer and ground into fine, less than 0.06 mm grain-sized fraction in an agate ball mill.

Analytical Procedures

Determination of pH and organic matter

The soil pH value in Sloznaft was determined in a slurry of air dried soil and distilled water. A sample of 20 g soil screened over a 1 mm mesh-sized sieve to remove aggregates larger than 1 mm was allowed to equilibrate with 50 mL distilled water for 5 min and to measure pH by continual mixing, using MV 870 Digital pH Messgerät (Precitronic, Germany) with a glass-calomel electrode. In Austria, soil (< 2mm) was mixed with 0.1 mol.L⁻¹CaCl₂-solution (1:2.5 ratio), left overnight and mixed well before measuring with a glass-calomel electrode.

The organic matter content of Sloznaft soils was determined by loss-on-ignition (1 g of soil heated in muffle oven at 600°C to a constant weight). The results were correlated with TOC (total organic carbon) determination by the IR method, measured on a TOC analyser (DC-190, Dohrmann, USA) as well as with soil composition estimated manometrically. In Austria the humus content was analysed with wet oxidation (oxidation of 0.5–10 g soil with 10 ml potassium-dichromate and 20 ml H₂SO₄ and the following titration with Fe(II)-sulphate).

Heavy metal extractions

Soil extraction procedures were accomplished using various extraction agents, e.g. 2 mol.L⁻¹HNO₃, 0.05 mol.L⁻¹EDTA, 0.5 mol.L⁻¹CH₃COOH, 0.1 mol.L⁻¹BaCl₂ and aqua regia^{12,8}.

HNO₃ extraction

By the above mentioned procedure a 10 g soil sample was shaken with 100 mL of HNO₃ extractant in a horizontal LT2 Shaker (200 cycles per minute) over 6 h in a polyethylene vessel at room temperature. After shaking, the sample was centrifuged (5 min, 3000 rpm) and the supernatant was analysed.

Aqua regia extraction

Three-gram soil samples were put together with 21 ml HCl and 7 ml HNO₃, left overnight at room temperature, boiled for 2 h at 200°C and poured into 100 ml vessels.

BaCl₂ extraction

Twenty grams of soil and 50 ml 0.1 mol.L⁻¹BaCl₂ extraction were shaken and filtered according to Blum et al.¹³

Table 1. Concentration range and precision of element determinations in environmental samples of Slovnaft site

Values		Samples (n=10)					
		Soil	Soil extractants in			Plant	Biota
			2 M HNO ₃	0.05 M EDTA	0.43 M CH ₃ COOH		
Element		Cr					
Conc.range (mg.kg ⁻¹)		0.17-0.44	0.14-0.33	0.10-0.21	0.09-0.20	0.13-0.34	2.34-7.20
RSD(%) of used methods	FAAS	1.98	n.d.	n.d.	n.d.	n.d.	n.d.
	ETAAS	2.66	4.40	12.16	13.77	n.d.	21.0
	ICP AES	n.d.	n.d.	n.d.	n.d.	8.96	n.d.
Element		Cr					
Conc.range (mg.kg ⁻¹)		54.90-67.28	5.0-8.30	0.72-1.84	0.47-1.08	0.82-1.52	9.18-17.71
RSD(%) of used methods	FAAS	5.10	n.d.	n.d.	n.d.	n.d.	n.d.
	ETAAS	6.72	11.73	12.08	13.97	2.03	10.69
Element		Ni					
Conc.range (mg.kg ⁻¹)		22.24-34.33	8.40-14.30	1.58-3.41	1.01-2.13	1.24-2.50	3.51-10.49
RSD(%) of used methods	FAAS	7.81	n.d.	6.48	7.24	4.43	n.d.
	ETAAS	11.27	2.02	5.42	9.48	5.17	3.63
	ICP AES	n.d.	3.07	n.d.	n.d.	n.d.	n.d.
Element		Pb					
Conc.range (mg.kg ⁻¹)		19.30-31.20	14.80-22.30	5.60-8.20	2.60-5.50	0.54-4.77	4.40-10.50
RSD(%) of used methods	FAAS	7.55	n.d.	6.30	4.74	n.d.	n.d.
	ETAAS	9.10	6.12	7.33	3.79	4.46	3.55
Element		V					
Conc.range (mg.kg ⁻¹)		59.51-90.48	40.40-58.30	23.33-28.42	21.0-26.20	0.55-2.49	21.11-34.02
RSD(%) of used methods	ETAAS	8.92	n.d.	10.21	8.87	5.39	12.10
	ICP AES	8.03	9.02	n.d.	n.d.	n.d.	n.d.

Acid decomposition of the lucerne samples was performed in a mixture of 10 mL 65% HNO₃ (suprapure) and 10 mL D.I. water in closed PTFE vessels for 50 min using a microwave oven (CEM, MSD-2000, USA) by 5 stages set and 80% power. Mineralised solutes were transferred into 100 mL volumetric flasks and filled up to the mark with D.I. water. Wheat samples (0.8 g) were decomposed in 8 ml acid mixture (HNO₃:HClO₄= 5:1) 4-5 h by heat impact.

The heavy metal concentrations were analysed using an ICP - AES 2070 (Baird, USA), an ETAAS Varian GTA-95 (Victoria, Australia) and a FAAS - Perkin Elmer 1100 equipped with deuterium arc background corrector (USA) as well as with an AAS with Zeemann background correction HITACHI Z-8000. The analytical accuracy of measurements was checked by means of certified reference materials (CRM).

Table 1 shows the concentration range and precision of element determinations in environmental samples using the above mentioned methods. By the applied atomic spectroscopy methods an acceptable detection limit of the studied elements was achieved and the reliability of analytical results was statistically satisfactory.

Results and Discussion

Most of the 112 top soil samples taken from the agricultural fields in the district of Schwechat (Lower Austria) show quite natural levels of heavy metals. However, it must be taken into consideration that normally there exists a considerable dilution of immitted heavy metals in arable soils due to ploughing. Measurable enrichment caused by widespread anthropogenic pollution, predominantly by atmospheric aerosols of heavy metals, was observed only in a few cases. One top sampling spot at the sampling raster was enriched with Pb and Cr concentrations respectively over the tolerable level¹⁴. The anthropogenic impact on metal accumulation in the topsoil horizon was confirmed by indications of Pb, Cd, Cu, Zn and Cr concentrations decrease with the soil depth⁸. The fixation/mobilisation potential of the soil with respect to heavy metals is dependent on the pH, organic matter and clay content of the soil. In Schwechat, these characteristics favour the immobilisation of (toxic) metals to a great extent. Table 2 presents characteristic data of the studied soils from both emission zones in Austria and Slovakia. The pH soil reactions were slightly alkaline. The Austrian part of the agricultural area has medium humus content, while the Slovakian one can be classified as deficient in organic matter with a humus content of less than 1%.

Table 2. Some characteristic data on the soils in the studied areas

Austrian Emission Zone		Slovakian Emission Zone	
pH	7.2	pH	7.4
CaCO ₃ (%)	15.1	humus (%)	0.3
Clays (%)	27	silica	
Sand (%)	24	clays	
Humus (%)	3.2	mica (%)	70
		limonite	
		dolomite (%)	27
		loss-on-ignition (%)	11.8
		TOC (mg.kg ⁻¹)	4534.5

The quantification of the total content of selected heavy metals in the soils by aqua regia digestion and by the applied extractants in soil eluates exhibit different levels, but the metal concentrations in 0.1

mol.L⁻¹ BaCl₂ extractant are mostly below the detection limit. In this context Table 3 shows a survey of the two investigated sites, and detailed statistical evaluations are given in Tables 4 and 5.

The transfer factors from soil to plant are quite low and the heavy metal uptake by wheat via the soil is also rather low. At the Slovakian monitoring site the heavy metal bioaccumulation within the lucerne, as a consequence of availability from the soil and atmospheric aerosols as well, showed a slightly increased tendency. A considerably higher, multiple bioaccumulation factor was recorded in related earthworm tissues¹⁵⁻¹⁶.

Table 3. Mean heavy metal contents in environmental samples from impacted south-eastern sites of Schwechat and Slovnaft refineries

Metal (mean content)	Austrian Region				Slovakian Region					
	top soil mg.kg ⁻¹	wheat mg.kg ⁻¹	soil eluate in BaCl ₂ mg.kg ⁻¹	K ₁	top soil mg.kg ⁻¹	lucerne mg.kg ⁻¹	earth- worms mg.kg ⁻¹	K ₁	K ₂	K ₃
Cd	0.27	0.04	< 0.01	0.16	0.39	0.22	5.30	0.56	24.09	13.58
Cr	38.70	0.23	< 0.05	0.01	58.10	1.09	14.10	0.02	12.93	0.24
Ni	28.0	0.20	< 0.10	0.01	29.60	1.63	7.20	0.05	4.42	0.24
Pb	31.50	0.95	< 0.20	0.03	28.30	1.60	7.0	0.06	4.37	0.25
V	-	-	-	-	81.40	1.25	28.80	0.02	23.04	0.35

K₁ = metal content in plant/metal content in soil, K₂ = metal content in earthworms/metal content in plant, K₃ = metal content in earthworms/metal content in soil

Table 4. Mean element contents incl. SD in soil, various soil extractants, plant and biota of Slovnaft site

Mean contents in mg.kg ⁻¹ (n = 10)						
Elements	Total dec. of soil sampl.	Soil extractants in			Lucerne	Earthworms
		2M HNO ₃	0.05M EDTA	0.43M CH ₃ COOH		
Cd	0.39 ± 0.18	0.21 ± 0.05	0.17 ± 0.03	0.15 ± 0.03	0.22 ± 0.06	5.30 ± 1.70
Cr	58.10 ± 3.20	5.60 ± 1.0	1.25 ± 0.39	0.60 ± 0.22	1.09 ± 0.25	14.10 ± 2.70
Ni	29.60 ± 3.40	11.30 ± 2.20	2.59 ± 0.60	1.62 ± 0.38	1.63 ± 0.41	7.20 ± 2.10
Pb	28.30 ± 3.02	17.30 ± 2.40	6.60 ± 0.80	4.30 ± 1.04	1.60 ± 1.20	7.0 ± 2.0
V	81.40 ± 9.93	52.0 ± 6.10	25.30 ± 1.70	22.90 ± 1.80	1.25 ± 0.6	28.80 ± 4.10

Table 5. Medians, means, concentration range and variation coefficients for heavy metals in soils (aqua regia) and plants of Schwechat

	top soil mg.kg ⁻¹ (n = 112)					wheat mg.kg ⁻¹ (n = 10)			
	Min.	Max.	Median	Mean	Var. Coef.	Min.	Max.	Mean	Var. Coef.
Cd	0.15	0.55	0.26	0.27	23.80	0.03	0.06	0.04	20.90
Cr	7.60	101.90	37.30	38.7	39.20	0.19	0.33	0.23	16.60
Ni	6.60	46.60	28.90	28.0	24.70	0.09	0.31	0.20	35.40
Pb	15.10	116.30	27.70	31.5	43.0	0.79	1.18	0.95	11.10

According to plant bioindicator (lucerne) the obtained results indicated concentrations below the total admitted amounts for agricultural feed in Slovakia; except for vanadium, the concentration in plant tissue of which slightly overpassed this limit (Table 3).

The amount of heavy metal contaminant that is assimilated by organisms is not well known¹⁷⁻¹⁸. Since the mobility of soil organisms may have significant implications for food chain transfer and the use

of an accumulation model in establishing environmental quality criteria, it was of importance to investigate the accessible heavy metal interactions of the soil matrix with lumbricids¹⁹. In this study, at the Slovakian industrial-residential area, average bioaccumulation factors for vanadium (23.04) and for cadmium (24.09) were recorded in related earthworms/plants; however, these were not confirmed in related soils/earthworms. These enhanced metal concentrations at the monitored area may result from numerous dumps and closed industrial activities²⁰. As mentioned above, vanadium and nickel emissions may result from Russian crude oil burning processes.

Lumbricid biomass is rich in nutrients and proteins and therefore may be used for the feeding of poultry, birds, fish and other animals. However, the cadmium concentration in the earthworm tissues exceeded the limit for agricultural feed and from this point of view may cause the gradual intoxication of higher species on the trophic chain²¹.

The correlation between individual heavy metals and selected soil parameters for the Schwechat district was statistically evaluated using linear regression analysis. The influence of close settlement, industrial emission sources and sewage sludge application were partially confirmed.

Lead and cadmium concentrations increased with organic matter, but not with increasing clay content in the soil. The opposite correlation was found for chromium and nickel. These two metals showed some positive correlation with pedogene Fe and Mn oxide content, respectively.

Generally summarised, the total decomposition of soils on the Slovnaft site may not indicate external (anthropogenic) contamination but geological metal content. In the Schwechat area, soil eluate data in BaCl₂ under the limit of detection correlated adequately to very low metal concentrations in the wheat.

While FAAS and ICP AES spectrometry were generally faster and operationally easier for the higher concentration range, ETAAS was more appropriate for the low metal concentration range.

The accuracy of the measured data was verified by a determination of the studied elements in CRM, whereas the analytical results were in good agreement with each other by all applied methods as well as with CRM.

Conclusions

The bioavailability, toxicity and transport of heavy metals in natural systems depend as presented above on the specification of heavy metals. The possible chemical forms of heavy metals in a natural system are hydrated metal ions, soluble metal complexes, metal precipitates, metal ions adsorbed to organic matter and bacterial residues and metal ions adsorbed to the surface and interstices of minerals. From this point of view various analytical and modelling approaches can be used to determine the specification of heavy metals in a natural system. Together with analytical methods, modelling approaches can provide an additional understanding of the environmental factors controlling metal distribution in natural systems including soil. For the same reasons, the extractability of heavy metals from soil can be better interpreted and predicted if more insight is available with respect to the metal binding properties of the soil particles.

This study presented a number of approaches and underlined the bioaccumulation factors of toxic heavy metals in higher living organisms. Nevertheless, this phenomenon may gradually endanger the entire trophic system.

List of Symbols

FAAS	- Flame atomic absorption spectrometry
ETAAS	- atomic absorption spectrometry with electrothermal atomization
ICP AES	- atomic emission spectrometry with inductively coupled plasma
IR	- infrared spectroscopy
PE	- Polyethylene
PTFE	- Teflon
n.d.	- Not determined
n	- Number of element determinations
SD	- Standard deviation
RSD	- Relative standard deviation
min.	- Minimum concentration
max.	- Maximum concentration
Var.coef.	- Variation coefficient

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