

ENVIRONMENTAL MONITORING USING LINDEN TREE LEAVES AS NATURAL TRAPS OF ATMOSPHERIC DEPOSITION: A PILOT STUDY IN TRANSILVANIA, ROMANIA

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Abstract

Atmospheric pollution caused by toxic elements is an emerging problem of concern. Tree leaves have been widely used as indicator of atmospheric pollutions and they are effective alternatives to the more usual biomonitoring methods. Tree leaves can be used as natural traps of atmospheric deposition. Elemental composition of dust deposited onto leaf surfaces can be used to characterize the urban environment. A pilot survey including 16 Romanian settlements was carried out in order to evaluate the characteristics and sources of air pollutants. Tree leaves (*Tilia tomentosa*, *Tilia cordata*, *Tilia platyphyllos*) were collected and used for the measurements. Elemental analyses were carried out by ICP-OES and ICP-MS. Principal component and discriminant analyses were used to characterizing and estimating the level of pollution. Settlements were grouped on the basis of discriminant function values. Multivariate comparison of chemical data ordered the settlements into 3 main groups, which showed a systematic geographic distribution.

Keywords: biomonitoring, air pollution, *Tilia*, ICP-OES, dust deposition

1. Introduction

Atmospheric pollution caused by toxic elements is an emerging problem of concern. Many countries have established programmes to control emissions. Industry (steel and chemical industries, power plants), agriculture, waste incineration, combustion of fossil fuels and road traffic are the most important local sources (Çelik et al. 2005). Long-range transport of atmospheric pollutants adds to the metal load and is the main source of heavy metals in remote natural areas (European Environmental Agency, 1995). Air pollutants can be identified by dispersion modelling and by field measurements of immission (Wolterbeek, 2002). However, immission measurements require long-term sampling at large numbers of sampling sites, and field measurements need expensive equipment, manpower, and associated with high costs.

The use of plant tissues has long been shown to be an indicator of atmospheric pollution. Most plants have the ability to accumulate heavy metals so that their metal contents are much higher than those in the air. Further, the observed result is

time-averaged and it could be more reliable than that obtained from direct measurements of pollutants in air over a short period (Lau – Luk, 2001).

Urban areas are exposed to numerous airborne contaminants emitted from anthropogenic sources in form of solid particles. The elemental composition of the atmospheric aerosol and flying, depositing dust can be used to characterize the urban environment (Ferguson – Kim, 1991). Particulate matter can be attributed to different sources according to its particle size. Fine particles derive from combustion processes and are mainly attributed to anthropogenic sources. Middle-sized particles are formed from elder particles during air transport or are newly generated in the atmosphere, and coarse particles derive from resuspension and mechanical wear (Goldish, 1991; De Miguel et al. 1997). The distribution of particulates vary within wide range. Toxic elements occurring in urban air are usually sorbed or incorporated in the particulate matter.

Tree leaves have been widely used as indicator of atmospheric pollutions (Kovács, 1992) and they are effective alternatives to the more usual monitoring methods, including mosses and lichens. Trees are long-lived organisms, which can take up trace elements from the soil, water, or air, and retain them for a long time (Majedón et al. 2006). Foliar analysis is helpful to detect elements present in the air in low concentrations or only temporary. The concentration of essential and nonessential elements in leaves may provide information on the incidence of each element in the environment (Alfani et al. 2000).

Airborne pollutants can deposit on leaf surfaces and some elements could enter leaf tissues via the stomata and accumulate (Maisto et al. 2007). Foliage of tree species from contaminated regions can be considered an accumulation monitor where significant amount of chemical elements are deposited on the surface or in the wax layer (Maňkiovská et al. 2004). The dust can accumulate on the leaves in various amounts depending on the degree of impact, meteorological, topographical factors, crown shape, density, leaf size, crown density (Komarnicki, 2005). The particle mass depends on air humidity, wind speed and wind direction, and rain.

Survey of air quality of large geographic regions is a rather difficult task, because it requires well coordinated sampling and time consuming laboratory operations (Houthuijs et al. 2001). We developed a procedure to collect and quantify and measure of elemental composition of dust deposited onto surface of tree leaves (Margitai – Braun, 2005a, Margitai et al. 2005). We improved a new kind of biological survey which provide a spatially representative picture of the environmental state of settlements or specific geographic regions (Margitai – Braun, 2005b). The aim of this work was to investigate and characterise the heavy metal pollution in Romanian settlements (which are different in size, industrialisation, traffic and land use) using dust deposited on linden tree (*Tilia sp.*) leaves as an indicator. Our re-

sults could be used as a preliminary baseline for trace element concentrations for future survey or biomonitoring.

2. Methods

2.1. Sampling area

Romania can be characterized by mismanaged industrialisation, associated with the lack of environmental protection from both industry and urban wastes (Amemia et al. 1996). The industry based mainly on the reprocessing of the raw materials and the power production largely based on coal and heavy fuel oil. Romania's energy sector has a strong impact on the country's environment in relation to oil, gas and coal production. Thermal power plants burning low caloric heat coal and high-sulphur content heavy fuel contribute to air pollution as well, and the low-quality coal that Romanian households burn for heat adds to poor air quality in urban centres. Coal mining regions are significant sources of air pollution. In the recent years Romania has drawn specific integration and EU directives implementation programs for main economy sectors. The overall result of EU standards implementation was a substantial decrease of gaseous pollutants (e.g. SO₂, NO_x and dust) (Cruceru et al. 2005).

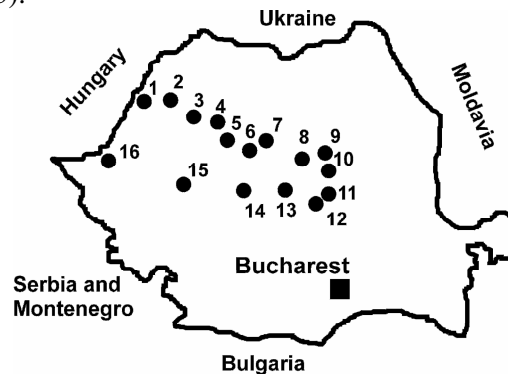


Fig. 1. Sampling locations

1. Nagyvárad (*Oradea*), 2. Élesd (*Aleșd*), 3. Bánfihunyad (*Huedin*), 4. Kolozsvár (*Cluj-Napoca*), 5. Torda (*Turda*), 6. Radnót (*Iernut*), 7. Marosvásárhely (*Târgu-Mureș*), 8. Székelyudvarhely (*Odorheiu Secuiesc*), 9. Csíkszereda (*Miercurea-Ciuc*), 10. Tusnádfürdő (*Băile Tușnad*), 11. Sepsiszentgyörgy (*Sfântu Gheorghe*), 12. Brassó (*Brașov*), 13. Fogaras (*Făgăraș*), 14. Nagyszeben (*Sibiu*), 15. Déva (*Deva*), 16. Arad (*Arad*)

2.2. Sample collection and preparation

Leaf samples were collected from 16 settlements in Transylvania, Romania (Fig. 1.) between 20 and 23 August 2004. We sampled linden leaves because these trees can be found in parks among streets in most of the settlements. Little leaf linden

(*Tilia cordata*), large-leaf linden (*Tilia plathyphyllos*) and silver linden (*Tilia tomentosa*) were selected as object for our survey for the following reasons:

- the morphology, canopy structure and the dust preserving epicuticular wax on the leaf surface make linden trees an ideal study object;
- *Tilia* species are widespread in Europe, allowing further geographic extension of investigations.
- it is easy to identify and distinguish from other species, making collection easy.
- linden leaves can be stored in refrigerators for long time (~30 days) without damage or moulding.

Ten individual trees were sampled from each settlement. They were chosen randomly from different parts of the towns (parks, roads with moderate and heavy traffic). The total leaf surface of each sampled tree represented 10-12 dm².

2.3. Apparatus and measurements

To ensure accurate comparisons, leaf area, as well as wet and dry weight were measured. The area of the leaves was determined using a flat bed scanner. The calibration was performed by using black images with known surfaces. Black and white images were taken by a resolution of 300 dpi. The number of black pixels were proportional to the area.

The dust deposited on the surface of leaves was removed by distilled water using a shaker machine and an ultrasonic bath. The water content of suspension was reduced by evaporation in a microwave oven. The suspension was transferred into 50 ml baker. After drying at 105°C the amount of dust was weighted.

Dust samples were digested with 5 ml of 65% (m/m) HNO₃ heated on hot plate to 80°C. After the evolution of NO₂ fumes had ceased, the mixture was evaporated almost to dryness and mixed with 2 ml of 30% (m/m) H₂O₂. The mixture was again evaporated to dryness. The samples were diluted to 10 ml of 0.1 mol/dm³ nitric acid.

The concentration of As, Ba, Be, Cd, Co, Cr, Cu, Mo, Ni, Pb, Sb, Sr, Tl and V were determined by inductively coupled plasma mass spectrometry (ICP-MS) using a VG-Elemental model PQ2 instrument from Thermo-Finnigan. The concentration of Al, Ca, Fe, K, Mg, Mn, S were analysed with inductively coupled plasma atomic emission spectrometry (ICP-AES) using a Thermo SPEC/WIN (IRIS) from Thermo Optek. Quantification of trace metal concentrations was based upon calibration curves involving standard solutions prepared from a commercial stock solution (Trace Metals 1 ICM-411H in 5% HNO₃, Radian International LLC). As internal standards In, Bi, Ru, Re were used to correct ionization suppression and

instrument drift (Nguyen et al. 2005). Blanks and certified reference materials (CRMs) were included in each digestion batch to verify the accuracy of the extraction method.

2.4. Statistical methods

Analytical results have been evaluated by using SPSS program. Principal component analysis (PCA) was applied to characterize the contribution of elements to the quality of the urban environment. Discriminant analysis was used to assign objects to established groups (Swan – Sandilands, 1995), and to estimate the level of pollution.

3. Results

3.1. Dust deposited on the leaf surface

Urban air usually contains significant amounts of dust (e.g. the rate of atmospheric deposition in Budapest $130 \text{ t km}^{-2} \text{ yr}^{-1}$) (Bernatzky, 1974). Trees play an important role in reducing the concentration of settling and flying particles by capturing them in the canopy. Foliage of a mature tree can lower the dust concentration of the air by 60-80%, making sensible difference in air quality between parks and treeless built-in environments. Dust filtration by the foliage has a direct effect on human health since the trapped particles contain toxic and allergenic compounds, heavy metals, organic pollutants, bacteria and pollen grains etc. The dust filtering capacity of trees and shrubs depends mainly on the taxonomic properties, leaf morphology and surface structure, the age-related size and habit of the canopy. The canopy lowers the velocity of air currents letting the dust settle on the leaf surface. A part of the captured dust is washed off by rainwater and delivered into the lower segment of the canopy, to the soil surface or to the drainage system. The leaf surface renewed by the rain become capable of further filtration. Large amounts of dust fixed by the foliage can be removed after litterfall thus precluding accumulation of toxic compounds in the soil (Kovács, 1985).

The exposition time in our study was c.a. 4 month. The deposition period started in May when the new leaves unfolded and ended at the date of sampling in August. Dust deposited on the leaf surface was expressed as $\mu\text{g}/\text{cm}^2$. Significantly high rate of deposition ($200\text{-}400 \text{ mg}/\text{dm}^2$ during 4 month) was observed for Kolozsvár, Torda, Marosvásárhely, Brassó, Fogaras and Szeben (Fig. 2).

If the environmental quality is to be characterized, data should be compared to reference values specified in legislations or scientific sources. Reference values of contaminants in soil, water, air, animal and plant tissues are available. In spite of

the simple collection of dust deposited on different surfaces in the in-door and out-door urban environment, the number of reported data is relatively small.

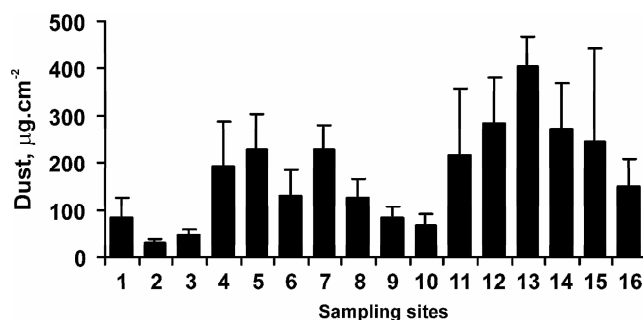


Fig. 2. Dust concentration on leaf surfaces

1. Nagyvárad (*Oradea*), 2. Élesd (*Aleșd*), 3. Bánfihunyad (*Huedin*), 4. Kolozsvár (*Cluj-Napoca*), 5. Torda (*Turda*), 6. Radnót (*Iernut*), 7. Marosvásárhely (*Târgu-Mureș*), 8. Székelyudvarhely (*Odorhei Secuiesc*), 9. Csíkszereda (*Miercurea-Ciuc*), 10. Tusnádfürdő (*Băile Tușnad*), 11. Sepsiszentgyörgy (*Sfântu Gheorghe*), 12. Brassó (*Brașov*), 13. Fogaras (*Făgăraș*), 14. Nagyszeben (*Sibiu*), 15. Déva (*Deva*), 16. Arad (*Arad*)

Primarily the settlements were characterised by average concentration of elements (Table 1). High concentrations of As, Cd, Cu, Pb and Zn indicate settlements with intense heavy industry (e.g. Fogaras, Szeben). In cities with high traffic (e.g. Nagyvárad) elevated concentrations of Pb and S were detected.

3.2. Enrichment factor analysis

Enrichment factors (EF) can be calculated to show the degree of enrichment of a given element compared to the relative abundance of that element in crustal material and used as a first step in attempting to evaluate the strength of the crustal and non-crustal sources. The enrichment factor is defined as

$$EF = \frac{(c_{i,j} / c_{i,Al})_{dust}}{(c_j / c_{Al})_{UC}}$$

where $(c_{i,j}/c_{i,Al})_{dust}$ and $(c_j/c_{Al})_{UC}$ refer to the ratio of concentration of element j to that of Al in the dust deposited on the leaf surface and in the average of the upper continental crust, respectively. Aluminium is the common reference element for EF calculations based on the crustal chemical composition given by Wedepohl (1995).

As Fig. 3. shown Cd and Pb exhibited the highest enrichment factors (>100). Other toxic elements such as Zn, Cu, As, Sb and Mo also showed high enrichment factors.

Table 1. Average concentration of elements in dust deposited on leaf surfaces

	Codes of settlements															
	(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)	(11)	(12)	(13)	(14)	(15)	(16)
	<i>Concentration of elements. (g/kg)</i>															
K	14.6	10.2	30.1	39.7	30.3	12.4	28.2	24.5	14.4	34.7	17.7	55.3	12.2	29.9	32.3	32.1
Ca	29.4	23.9	23.9	31.0	42.5	18.3	23.3	37.1	34.7	12.8	26.6	26.5	7.2	15.9	26.1	36.1
Fe	24.5	14.1	13.0	14.0	15.4	14.3	13.5	12.8	11.1	7.8	7.9	8.9	7.6	11.8	16.6	16.4
Al	8.63	6.94	5.94	4.98	6.70	8.57	8.45	7.07	7.33	3.94	4.42	4.15	3.50	6.46	8.07	6.65
Mg	3.67	3.15	3.27	5.33	5.08	2.94	3.27	3.32	7.23	1.60	3.24	4.03	1.93	4.94	8.20	10.0
P	1.31	3.81	1.18	2.64	4.59	4.22	4.28	3.13	2.01	2.45	2.32	4.02	4.30	4.41	4.92	2.36
S	4.07	2.38	2.97	2.51	3.96	2.56	2.89	2.41	1.45	1.83	2.29	2.33	2.29	3.31	5.48	3.02
Na	1.27	0.00	0.26	0.95	0.48	0.17	2.73	1.03	0.48	0.00	0.70	0.12	0.01	0.37	0.01	1.60
Mn	0.32	0.18	0.26	0.22	0.36	0.29	0.21	0.24	0.19	0.21	0.19	0.13	0.09	0.17	0.33	0.28
	<i>Concentration of elements. (mg/kg)</i>															
Zn	395	277	124	257	360	220	249	339	30	86	153	131	372	465	381	171
Ba	204	130	130	205	104	124	129	111	92	132	87	88	45	80	107	135
Pb	129	74	24	74	89	142	124	253	31	41	43	47	225	296	116	57
Cu	193	43	13	89	61	22	64	66	7	10	24	74	80	99	56	62
Sr	57.1	33.4	32.7	72.4	133.8	49.5	57.9	93.3	51.3	51.0	54.2	40.1	21.4	46.9	80.3	60.6
Cr	33.65	9.14	2.57	11.16	17.30	8.82	15.89	22.31	1.93	1.69	7.40	8.89	15.97	31.59	15.39	8.35
V	17.87	9.16	3.16	6.33	12.80	16.95	17.69	20.59	2.38	2.66	7.11	5.40	13.09	27.83	14.17	6.45
Ni	31.24	0.44	0.04	7.22	8.08	5.68	13.04	19.96	0.21	<0.1	6.98	1.86	3.14	10.63	2.70	7.35
As	7.63	5.73	1.33	3.55	6.28	3.57	3.77	6.99	0.81	1.11	1.71	1.57	6.57	7.06	6.29	2.78
Cd	<0.1	0.36	0.25	3.52	2.51	4.88	2.03	4.75	0.58	0.63	1.73	2.85	11.42	17.13	7.00	0.74
Co	5.58	2.21	0.85	1.93	2.89	3.27	3.34	5.38	0.56	0.66	2.11	1.67	4.62	7.75	3.09	1.74
Mo	4.04	0.71	0.41	3.99	3.44	0.25	3.65	2.53	0.05	0.03	0.83	1.80	4.61	6.13	1.43	1.51
Sb	2.11	1.57	0.64	1.11	0.67	0.27	1.51	1.36	0.12	0.41	0.21	0.87	3.57	2.84	1.59	0.50
Be	0.40	0.25	0.08	0.14	0.17	0.19	0.21	0.37	0.05	0.07	0.19	0.09	0.24	0.42	0.20	0.12
Tl	0.08	0.06	0.02	0.04	0.09	0.11	0.08	0.14	0.02	0.11	0.07	0.03	0.19	0.29	0.10	0.03

1. Nagyvárad (*Oradea*), 2. Élesd (*Aleșd*), 3. Bánfihunyad (*Huedin*), 4. Kolozsvár (*Cluj-Napoca*), 5. Torda (*Turda*), 6. Radnót (*Iernut*), 7. Marosvásárhely (*Târgu-Mureș*), 8. Székelyudvarhely (*Odorheiu Secuiesc*), 9. Csíkszereda (*Miercurea-Ciuc*), 10. Tusnádfürdő (*Băile Tușnad*), 11. Sepsiszentgyörgy (*Sfântu Gheorghe*), 12. Brassó (*Brașov*), 13. Fogaras (*Fagăraș*), 14. Nagyszeben (*Sibiu*), 15. Déva (*Deva*), 16. Arad (*Arad*)

The enrichment of these elements suggested that the dominant sources for these elements were non-crustal and a variety of pollution contributed to their appearance in the dust. Metal smelting and combustion of fuels are usually the sources of volatile metals (e.g. Cd, Cu, As). Leaded gasoline still in use in Romania thus Pb and Zn are traditional tracers of vehicle emissions (Salma et al. 2000). On the contrary Fe, Ca, Mg, Mn had rather high concentration but showed low enrichment factors (<10), thus suggesting less contribution of anthropogenic sources and mainly originating from soils and street dust.

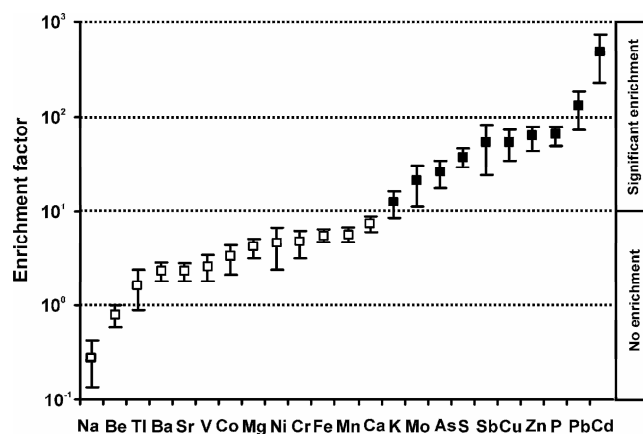


Fig.3. Average enrichment factor of elements in dust deposited on leaf surface

3.3. Principal component analysis of enrichment data

In a further attempt to assess the sources responsible for the observed pollution levels, factor analysis using principal component method with varimax rotation was applied to data set of enrichment factors. Principal components with eigenvalues larger than unity were retained. Total of 80.27% of the variance was explained by the first five factors that describe the group of elements with different sources (Table 2). The first principal component (PCA₁) explained about 44.27% of the total variance. The high loading for Co, Cr, As, Zn, Pb, Cd, Cu, Mo, Tl, Sb and Be in this factor indicate the contribution of anthropogenic sources. PCA₂ with high loading for Ca, Mg, Sr, K, S and PCA₃ with high loading for Fe, Mn, P and Ba represent the different sources of soil dusts. Na loaded in PCA₄ with small variance (4.49%).

The enrichment properties of elements were investigated in dust deposited on leaf surfaces were similar that of were measured in coarse aerosol particles by others in Central Europe (Salma et al. 2001).

3.4. Discriminant analysis

Local factors determining the chemical composition of dust on leaves were examined via discriminant analysis. Since enrichment properties of toxic elements were similar, calculations were based on these including As, Be, Cd, Co, Cr, Cu, Mo, Ni, Pb, Sb, Tl, V, Zn. Settlements were characterized by the values of the discriminant functions. Originally grouped cases were correctly classified (97.7%) indicating

that the chemical composition of dust deposited on leaf surface was considerably determined by local factors.

Table 2. Principal components of enrichment factor data

Element	PCA ₁	PCA ₂	PCA ₃	PCA ₄	PCA ₅
Co	0.972	-0.011	-0.024	-0.134	0.006
Cr	0.958	0.056	-0.134	-0.015	0.098
As	0.946	0.044	0.072	0.009	-0.051
V	0.935	-0.006	-0.107	-0.235	0.007
Be	0.935	0.025	-0.085	-0.101	0.023
Zn	0.910	0.057	0.146	0.057	-0.001
Pb	0.907	-0.112	0.222	-0.080	-0.093
Mo	0.887	0.056	0.109	0.100	0.142
Sb	0.807	-0.162	0.430	0.092	-0.012
Tl	0.786	0.007	0.231	-0.131	-0.196
Cd	0.782	-0.199	0.408	-0.046	-0.149
Cu	0.775	-0.032	-0.054	0.339	0.172
Ni	0.606	0.181	-0.602	-0.019	0.314
Sr	0.032	0.858	-0.010	0.191	-0.146
Mg	0.009	0.774	0.211	-0.112	0.163
Ca	-0.158	0.750	-0.146	0.257	0.020
K	-0.019	0.660	0.509	0.022	0.216
S	0.222	0.537	0.502	0.127	0.018
P	0.314	0.279	0.762	-0.067	-0.082
Fe	0.164	0.100	-0.037	0.849	0.114
Ba	-0.203	0.122	0.079	0.770	0.047
Mn	-0.130	0.508	-0.103	0.575	-0.329
Na	-0.024	0.055	-0.076	0.079	0.857
% Variance	44.27	15.98	9.06	6.47	4.49

Similarity of settlements was interpreted as Euclidean distance between group averages of discriminant scores (centroids). The settlements were compared by cluster analysis of the distance matrix using paired group method (Fig 4.).

The settlements were ordered to 4 clusters which showed clear non-random geographic segregation (Fig 4.). One of the two major clusters included five settlements located in the western part of Romania. These showed enhanced levels of certain heavy metals. The second major cluster of settlements comprised six towns and cities situated in the central part of Romania. Average concentrations of toxic elements were found to be comparatively lower. The most contaminated settlements (Radnót – Iernut, Fogaras – Fagâraş, Nagyszeben – Sibiu) were classified into a separate group. There the concentrations of As, Cd, Pb and Zn were considerably high. Two settlements close to the western frontier of Romania (Nagyvárad – Oradea, Élesd – Aleşd) were markedly different from all other clusters owing to very high levels of Co, Cu, Mo, S, and Zn. This emphasizes the role of road lorry transport. Observed pattern based on the multivariate interpretation of dust composition data may reflect local and regional differences in geological features and anthropogenic activities.

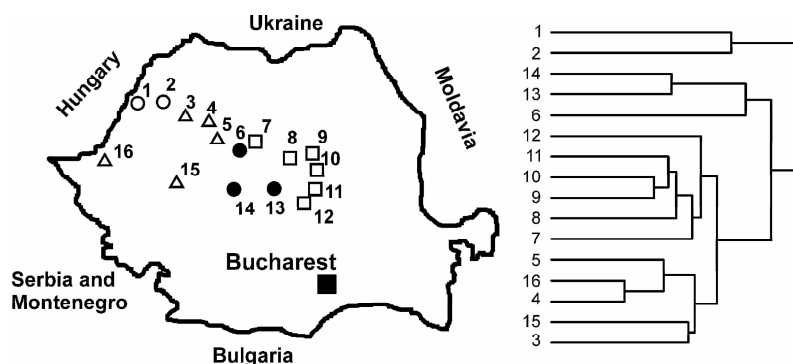


Fig. 5. Grouping of settlements by cluster analysis of discriminant centroids
 1. Nagyvárad (*Oradea*), 2. Élesd (*Aleșd*), 3. Bánfihunyad (*Huedin*), 4. Kolozsvár (*Cluj-Napoca*), 5. Torda (*Turda*), 6. Radnót (*Iernut*), 7. Marosvásárhely (*Târgu-Mureș*), 8. Székelyudvarhely (*Odorheiu Secuiesc*), 9. Csíkszereda (*Miercurea-Ciuc*), 10. Tusnádfürdő (*Băile Tușnad*), 11. Sepsiszentgyörgy (*Sfântu Gheorghe*), 12. Brassó (*Brașov*), 13. Fogaras (*Fagăraș*), 14. Nagyszeben (*Sibiu*), 15. Déva (*Deva*), 16. Arad (*Arad*)

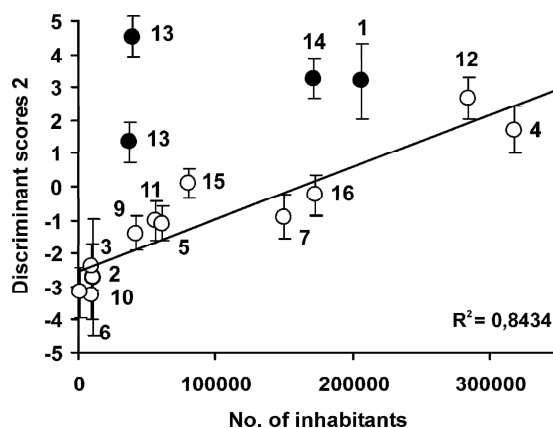


Fig. 6. Correlation between the number of inhabitants and discriminant scores
 (Settlement codes are above)

Most aspects of the environmental state can be related to the number of inhabitants of a settlement. In towns or cities with larger population environmental loads associated with traffic, domestic pollution, industry and agriculture are expected to be high. Therefore we tested if any correlation exists between discriminant functions and the number of inhabitants. We found significant positive regression only for the second function ($R^2=0.3563$). After the exclusion of samples from Nagyvárad (*Oradea*), Székelyudvarhely (*Odorheiu Secuiesc*), Fogaras (*Fagăraș*) and Nagyszeben (*Sibiu*), the regression became stronger (Fig. 6). Because this discriminant function explains a large proportion (14.9%) of the total variance for the selected elements, the number of inhabitants can be regarded as an important background factor. The pollution status of the four settlements scattered off the regression line is larger than is expected on the basis of their population sizes.

4. Conclusions

Enrichment factors of toxic elements were considerably high in the dust deposited on linden leaf surface. Principal component analysis of enrichment factors arranged the elements into groups. All the toxic elements (Cd, Pb, Zn, Cu, As, Sb) were loaded into the first principal component indicating that the enrichment of these elements is independent of the others and their origin is mainly anthropogenic. Elements reflecting local geological and pedological sources (e.g. Ca, Mg, Fe, Mn) of dust components were loaded into separated principal components.

Discriminant analysis of heavy metal concentration data revealed that the composition of dust is characteristic of the settlements and also suggested some relevant pollution factors. We found correlation between the size of the settlements (characterized by the number of inhabitants) and the chemical composition of dust. The relatively simple methodology of sampling, chemical analysis and statistical evaluation would allow to extend the geographical range of the focal settlements. By the inclusion of further background (e.g. meteorologic, economic, epidemiologic) variables our model can be improved to a more realistic description of the observed differences.

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