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Dandelion Plants as a Biomonitor of Urban Area Contamination by Heavy Metals

Kleckerová, A. and Dočekalová, H.*

Department of Chemistry and Biochemistry, Faculty of Agronomy, Mendel University in Brno, Zemědělská 1/1665, CZ-61300 Brno Brno, Czech Republic

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ABSTRACT: To examine metal content of dandelion plants in relation to environmental metal levels, concentrations of Cd, Hg and Pb were analyzed in plant parts (leaves and roots) and soil samples from five sites in the city of Brno (Czech Republic), differentially impacted by pollution. Soils and plants were collected mid-April 2011. Atomic absorption spectrometry was used to determine concentrations of the studied elements. The amount of metals measured in soils and plants corresponded with the contamination load of the sampling place. The highest values of metals were found in the soil and plants sampled at Opuštěná Street, a heavily polluted locality with high traffic density situated in the city centre. Significant correlations were found between the amount of Cd in the soil and in the dandelion roots (r= 0.863) and between the amount of Pb (r= 0.870) and Hg (r= 0.828) in the soil and in the dandelion leaves. Higher Cd content was found in underground part of the plants, indicating soil contamination. The higher Hg and Pb content in leaves rather than in roots in all locations illustrated a contribution of significant atmospheric deposition. Washing the leaves before the analysis significantly reduced the measured metal concentrations. This indicated that substantial amount of metals was on the leaves surface as dry aerosol particles. Inter-metal correlations between soil, leaves and roots samples showed that the sources of Cd, Pb and Hg pollution in Brno urban areas were mainly of anthropogenic origin. The content of heavy metals both in the dandelion plant tissue and in the soil should be seen as a good indicator of natural urban environmental pollution.

Key words: Environmental pollution, Urban soil, Determination of metals, Inter-metal correlations, Plant monitoring

INTRODUCTION

Air and soil pollution resulting dominantly from the emission of fumes and smoke from industrial plants and automobile exhaust fumes have increased appreciable the level of heavy metals in urban areas. Following wet or dry deposition, these metals remain in the soil and may retard growth of plants and of soil microorganisms, may be transferred from contaminated soils into the plant tissue and via food chain endanger the human health. Searching to find reliable, low cost methods of assessing the degree of metal contamination within a locality and the exposure risk to the biota is therefore highly advisable. Positive relations have been found between atmospheric heavy metal deposition and heavy metal concentration in plants (Ugulu *et al.*, 2012). The contribution of heavy metal accumulation has also been evidenced for forest and other types of vegetation (Berthelsen *et al.*, 1995). Terrestrial and water plant species are thus increasingly used as biological monitors of trace elements (Wittig, 1993; Loppi *et al.*, 1997; Monaci *et al.*, 2000; Divis *et al.*, 2012). A good biological monitor should be widely spread species easy to identify, characterized by a relatively high tolerance to environmental pollutants. Metals accumulated in monitor body should reflect those in the environment (Bargagli, 1998; Kadukova *et al.*, 2008). Dandelion *(Taraxacum officinale, Weber)* is a ubiquitous plant that can be found in various ecosystems and it has been mentioned frequently in environmental studies (Djingova and Kuleff, 1993; Simon *et al.*, 1996; Czarnowska and Milewska, 2000). Its uses include the evaluation of environmental pollution with SO_2, polycyclic aromatic hydrocarbons and heavy metals (Królak, 2003). The concentration of many metals in the leaves of *Taraxacum officinale* has been correlated with local environmental pollution in previous studies (Djingova and Kuleff, 1993; Kabata-Pendias and Dudka, 1991; Marr *et al.*, 1999; Keane *et al.*, 2001). However, the relationships between metal concentrations in plant tissues and surrounding environment have not been clearly explained to date, due to different and even opposing results.

*Corresponding author E-mail: hana.docekalova@mendelu.cz

The purpose of this study was to investigate the application of the common dandelion in a biomonitoring study of metal pollution (Cd, Hg and Pb) in Brno city (Czech Republic). The correlation between metal concentrations in plant tissues (leaves, roots) and in soil was tested and the ability of dandelion plant as metal biomonitor in urban areas was observed.

MATERIALS & METHODS

Brno is the second largest city of the Czech Republic with a wide range of industrial activities, including smelting operations (heating plants, incineration plants, cement and lime works, crematory) and automotive exhaust, which produced emission, and consequent deposits on soil and vegetation surface. The soil and plant samples were collected from the topsoil of five sampling sites in the city of Brno. In fact, as the metal content of leaves could vary seasonally (Djingova and Kuleff, 1994), sampling was performed in the middle of April 2011, when first flowers appeared. In this phenophases the plant could be quite sensitive to environmental conditions. The sampling sites (Fig. 1) were selected according to long-term atmospheric deposition and traffic density measured by The Brno Regional Office of the Czech Hydrometeorological Institute (CHMI) and Brno Traffic research centre (CDV) as quoted by Mikuska *et al.* 2011. The sampling site at Opuštěná Street represents an industrial heavily polluted location with high traffic density, situated in the city centre. Sampling sites at Vídeňská and Podstránská Streets belong to medium polluted locations that are situated close to busy roads. Relatively clean locations are represented by Musorgského and Šrámkova Streets residential areas, which are situated in peripheral city district with lower contamination loading. Soil samples were collected in the immediate vicinity of the dandelion plant from a depth horizon of 0-10 cm, ten samples at every sampling place, which represented an area of 10sq.m.

Ten plants of the common dandelion grown on the sampling place were carefully extracted from the soil, placed in plastic bags and transported to the laboratory. There dandelion leaves and roots were separated and washed with distilled water and air-dried. One-half of leaves were not washed to assess aerial contamination. Two dry weeks before sampling without any rainfall caused sedimentation of dry aerosol particles containing metals on the leaves surface. Soil physicochemical properties were determined on the air-dried fraction with particle size of less than 2 mm in diameter obtained by sieving of the air-dried raw sample, from which large components were separated (stones, plant parts). pH/KCl and C_{ox} were determined using conventional methods.

Single leaching procedure commonly used in Czech Republic for measuring of quasi-total metal fraction

Fig. 1. Location of sampling sites: A – Opuštěná, B – Vídeňská, C – Podstránská, D – Musorgského, E – Šrámkova

(Zbíral, 1996) was used for Cd and Pb determinations in soils samples. A portion of 7 g dried and sieved soil was shaken in an extraction bottle at the ambient temperature (25ºC) with 70 mL of 2 mol/L nitric acid for 16 hours. The extract was immediately filtered and the filtrate was collected in a polyethylene bottle. 0.2 g of dried and homogenized plant samples was decomposed in the microwave oven (Milestone, MLS 1200, Italy) using 6 mL of concentrated HNO_3 and 1 mL of 30% H_2O_2.

Total mercury content in soil and dried plant samples was measured using one purpose atomic absorption spectrometer Advanced Mercury Analyser, AMA254 (Altec, Czech Republic) based on combustion of the sample in oxygen atmosphere and amalgamation preconcentration. Cd and Pb concentrations were determined in both the HNO_3 leachates and plant digests.Electrothermal atomic-absorption spectrometer (AAS ZEEnit 60, Analytik Jena, Germany equipped with Zeeman ackground correction) was used under the recommended conditions specified by the manufacturer.

The reference materials (CRM 7001- Light Sandy Soil with normal analyte levels and 7003 – Silty Clay Loam with normal analyte levels, Analytika, Czech Republic and IAEA-V-10 – Hay powder) were used for method validation. Trace metals translocation from soil to plant parts expressed by bioconcetration factor (BCF = metal concentration in plant parts / metal concentration in soil (μg/g)) and transportation index (Ti = metal concentration in leaf / metal concentration in root (μg/g)) were according to Díaz *et al.*, 2011. Normal distribution of measured data was tested by Kolgomorov-Smirnov test.

Statistical analyses of metal content in soil and plant parts were made using one-way analysis of variance (ANOVA) and statistical significance was declared when p value was equal to or less than 0.05. The significance of differences between the average values of the individual localities was tested at the level of significance ? = 0.05 with the Tukey's HSD (Honestly Significant Difference) test. The t-test analysis was carried out with the washed and unwashed samples of plants; analyzed metals showing differences were identified. The results are presented as means ± standard deviation (SD). Relationships between parameters (plant parts) were expressed by correlation coefficients after Pearson. All results were evaluated by the statistical software STATISTICA.cz (version 10).

RESULTS & DISCUSSION

All analytical methods used during this study passed the quality control tests. The precision and accuracy of the methods have been verified by analysis of certified reference materials (CRM 7001- Light Sandy Soil with normal analyte levels and 7003 – Silty Clay Loam with normal analyte levels, Analytika, Czech Republic and IAEA-V-10 – Hay powder). Results are summarized in Table 1. Detection limits were adequate for the metal analysis of selected samples.
The soils were classified as a neutral soil with lack of humus. Basic characteristic of soil samples are summarized in Table 2.

Table 1. Certified and found values of elements in CRM 7001, 7003 (μg/gdry weight) and IAEA-V-Hay Powder (mg/kg dry weight), n = 5

CRM	7001	7001	7003	7003	IAEA-V-10	IAEA-V-10
	certified	Found	certified	found	certified	found
Element	Mean±Unc	Mean±Unc	Mean±Unc	Mean±Unc	95% C.I.	Mean±Unc
Cd	0.18 ± 0.02**	0.16 ± 0.03**	0.23 ± 0.01**	0.21 ± 0.02**	0.02 – 0.05	0.03 ± 0.01
Hg	0.087 ± 0.006*	0.090 ± 0.010*	0.096 ± 0.014*	0. 100 ± 0.018*	0.009 – 0.016	0.013 ± 0.003
Pb	20.7 ±0.6**	22.2 ± 1.0**	19.3 ± 0.4**	18.9 ± 0.70**	0.8 – 1.9	0.14 ± 0.06

* Total element content
** Content of element, using 2 mol/l HNO_3

Table 2. Basic characteristic of soil samples

Sampling site	Opuštěná Street	Vídeňská Street	Podstránská Street	Musorgského Street	Šrámkova Street
pH	7.10	7.27	7.43	7.23	7.09
* C_{ox} [%]	1.5	1.2	1.7	1.5	2.1

* C_{ox} [%] - the percentage of the organic matter content in homogenized samples

Djingova and Kuleff, (1993); Kabata-Pendias and Dudka, (1991) and Keane *et al.*, (2001) presented that dandelion plants are able to absorb metals from the atmosphere as well as from the soil. Uptake of heavy metals through leaves is considered as passive intake and is faster and more efficient than uptake by roots. Therefore the content of metals was measured in both dandelion leaves and roots.

The concentration of investigated metals in the soil was found, as expected, significantly higher in the samples collected in the city centre than those collected in clean areas situated in the peripheral city district. This fact was confirmed by one-way variance analysis ANOVA. Significant variation ($p<0.05$) for metal concentrations in soil, washed and unwashed plants from studied localities was found (see the Table 3-5).

The mean concentrations of cadmium found in soil, unwashed and washed leaves and washed roots of *T. officinale* sampled in studied localities are presented in Table.3. Mean Cd concentrations in Opuštěná Street are statistically significantly higher ($p<0.05$) than those in other localities. Significantly lower amounts of Cd were detected at Šrámkova Street, the clean city locality.

Washing the leaves significantly reduced the measured Cd concentrations (confirmed by t-test). In our study the washing procedure removed ca 20% of Cd independent of the sampling site. Keane *et al.* (2001) claimed even 40% washed metals in industrial areas. This indicated that substantial amount of metals was on the leaves surface as dry aerosol particles. The metals bound in solid particles are quickly absorbed by plant leaves and contribute a significant portion of the leaves metals content.

Compared to Pb and Hg, higher content of Cd was found in dandelion roots sampled at all studied localities. Higher amount of cadmium in roots than in leaves was also found in Klórak's study (2003), where the mean concentration in dandelion roots was found to be 2.9 mg/kg in highly loaded industrial site.

The highest content of Pb in the soil was found at the sampling site Opuštěná Street (Table 4). The maximum tolerable level defined by the Czech legislation - Regulation No. 13/1994 of the Czech Ministry of Environment (1994) (the limit of lead content in soil is 70 mg/kg) was exceeded only in one case. Approximately half the levels of Pb in soil were detected at the sampling locations Vídeňská, Podstránská and Musorgského Streets, these results were confirmed by Tukey's test. Statistically significantly lower Pb concentrations were measured in soil and plant samples collected at Šrámkova Street. The content of Pb in dandelion leaves and roots corresponded to the contamination load, the highest lead content was found at Opuštěná Street with heavy traffic density. Although the unleaded petrol is used at present, dust particles containing Pb from leaded petrol in the past are still in the environment (Koeppe, 1981; Asgari, 2011). It is generally known that the amount of Pb in soil had significant relation with traffic (Rahmani *et al.,* 2001; Pydtt, 1999; Celik and Aslihan, 2004).

The same as for Hg, a higher content of Pb was found in dandelion leaves than in roots. Similar results were found by Królak (2001), Kabata-Pendias and Krakowiak (1997) and Ziedler (2005) who found a higher content of Pb in dandelion leaves in both clean and polluted localities. The studies focused on washed and unwashed plant parts had shown that source of heavy metals in all sites directly or indirectly related to human activities and were classified as "pollution with human source". T-test confirmed that the washing of leaves had an effect on analytical results. Statistical highly significant difference between washed and unwashed plants was found. 16 - 35% Pb was removed depending on the contamination loading of the locality. The similar results (reduction of 14 - 48%) were found in Aksoy *et al.* study (2012) where washing the leaves significantly reduced the Pb, Cd, Cu a Zn concentrations in *P. judaica* plant.

The highest Hg content in soil was found at sampling site Opuštěná Street. The concentrations of Hg in soil collected from other three sampling sites were much lower. These results were confirmed by Tukey's test. The low contamination of soil as well as of plants by Hg was surprisingly found also at Vídeňská Street situated in the city centre. Contents of Hg in the dandelion roots and leaves corresponded with the Hg contents in soil. Results are summarized in Table 5. Plants may absorb elemental mercury directly from the air and therefore they can be used for monitoring airborne mercury pollution. Most Hg is absorbed through the leaves but a small part is translocated to the roots. The mercury forms which can be adsorbed by leaves include Hg^0, Hg^{2+}, CH_3Hg^+ and $C_2H_5Hg^+$ (Lodenius, 1994). It has been suggested that Hg is mainly retained through the direct uptake of elemental Hg via stomata (Lindberg *et al.*, 1991). The washing procedure reduced the content of Hg by a range of 20 – 28%. Statistically significant difference between washed and unwashed leaves was confirmed by the t-test. In Xiao *et al.* study (1998), where they used artificially planted rye grass in open ground as a biological monitor for air-borne mercury (Hg) in Sweden, unwashed samples generally gave >10% higher values than washed ones. Lindberg *et al.* (1994) have reported that dry deposition of Hg aerosol was about 10% with a range of 4–23%.

Table 3. Concentration of cadmium in soil and in plant parts (mg/kg dry weight)

Sampling sites	Cd (mg/kg)			
	soil	leaves^A	leaves^B	roots
Opuštěná	0.666±0.058^a	0.212±0.049^a	0.171±0.041^a	0.240±0.041^a
Vídeňská	0.450±0.099^b	0.169±0.018$^{b\,**}$	0.137±0.021^b	0.166±0.024^b
Podstránská	0.332±0.049^c	0.153±0.021$^{b\,**}$	0.125±0.025^b	0.170±0.009^b
Musorgského	0.289±0.107^c	0.069±0.018^c	0.056±0.014^c	0.096±0.017^c
Šrámkova	0.104±0.070^d	0.058±0.012$^{c\,*}$	0.047±0.011^c	0.092±0.013^c

Values are means of 10 samples ± standard deviation

^{A} Unwashed; ^{B} Washed

Different letters in the same column indicate significant differences at p<0.05 (ANOVA).

Significance of differences between washed and unwashed plants from paired t-test is indicated beside letters in the columns ($^*p<0.05$, $^{**}p<0.01$, $^{***}p<0.001$).

Table 4. Concentration of lead in soil and in plant parts (mg/kg dry weight)

Sampling sites	Pb (mg/kg)			
	soil	leaves^A	leaves^B	roots
Opuštěná	63.1±14.7^a	10.9±3.04^a	9.06±0.96^a	4.02±1.75^a
Vídeňská	29.4±5.68^b	6.10±1.19$^{b\,***}$	3.94±0.75^b	1.67±0.49^b
Podstránská	25.9±2.06^{b,c}	5.68±0.83$^{b\,***}$	4.09±0.59^b	1.44±0.23^b
Musorgského	18.7±3.79^{c,d}	4.69±0.71$^{b,c\,**}$	3.61±0.61^b	1.40±0.26^b
Šrámkova	10.6±3.91^d	3.10±0.47$^{c\,***}$	2.13±0.31^c	1.49±0.31^b

Values are means of 10 samples ± standard deviations

^{A}Unwashed; ^{B} Washed

Different letters in same column indicate significant differences at p<0.05 (ANOVA).

Significance of differences between washed and unwashed plants from paired t-test are indicated beside letters in the columns ($^*p<0.05$, $^{**}p<0.01$, $^{***}p<0.001$).

Table 5. Concentration of mercury in soil and in plant parts (mg/kg dry weight)

Sampling sites	Hg (mg/kg)			
	soil	leaves^A	leaves^B	Rous
Opuštěná	0.628±0.207^a	0.086±0.029$^{a\,*}$	0.062±0.013^a	0.049±0.008^a
Vídeňská	0.063±0.015^b	0.048±0.006$^{a\,**}$	0.038±0.008^b	0.012±0.003^b
Podstránská	0.215±0.086^c	0.070±0.012$^{b\,***}$	0.051±0.008^c	0.029±0.004^c
Musorgského	0.047±0.016^c	0.029±0.005$^{c\,**}$	0.022±0.004^d	0.014±0.002^c
Šrámkova	0.050±0.040^c	0.028±0.004$^{c\,***}$	0.020±0.002^d	0.009±0.001^c

Values are means of 10 samples ± standard deviation

^{A} Unwashed; ^{B} Washed

Different letters in same column indicate significant differences at p<0.05 (ANOVA).

Significance of differences between washed and unwashed plants from paired t-test are indicated beside letters in the columns ($^*p<0.05$, $^{**}p<0.01$, $^{***}p<0.001$).

The highest values of Pearson's correlation coefficients (Table 6) and thus significant positive correlation was found between the amount of Cd in soil and in dandelion roots (r=0.863) and between the amount of Cd in leaves and roots (r = 0.836). Statistically highly significant and positive correlation was found between Pb in dandelion leaves and soil (r = 0.870). Similarly strong positive correlation was found between Hg in leaves and soil (r = 0.828). When dandelion plant would be used as biomonitor for Cd pollution, analysis of dandelion roots is recommended, whereas for Hg and Pb pollution, the dandelion leaves are the choice.

Inter-element correlation gave information about heavy metal sources. Table 7 represented the correlations between each analyzed trace metal, expressed by the Pearson's correlation coefficients. There were high significant correlation coefficients at $p<0.001$ between metals in plant parts and soil. These results suggested that the sources of Cd, Pb and Hg pollution in Brno urban areas were mainly anthropogenic origin.

The translocation of metals from soil to plant parts expressed as bioconcentration factor (BCF) was calculated to determine the relative uptake of metals by the plants with respect to soil. The values of BCF for samples collected at all sampling sites are summarized in Table 8. Higher BCF of Cd than those of Hg and Pb confirmed higher phyto accessabilty of Cd. Higher metals intake by the plants growing in clean areas corresponded with the observation of Królak (2003), who found three times higher BCF in clean locality compared to an industrial contaminated locality in Poland. Pb and Hg are relatively immobile in soil and therefore less available for plant uptake. The similar results were found in Kabata – Pendias and Pendias's study (1999). Calculated BCF leaves/soil for Hg and Pb had higher value than BCF roots/soil. Both these metals showed, contrary to Cd, the accumulation predominantly in leaves.

The transportation index (*Ti*) gives the ratio of leave/root metal concentration and describes the availability of the plant to translocate the metal species between roots and above-ground part of the plant (Zayed *et al.,* 1998). The results Obtained results confirmed the accumulation of Cd in roots and of Hg and Pb in leaves. Transportation indexes of studied metals are shown in Table 9.

Table 6. Correlation analysis among concentration of metals in soil and plant parts

Cd	soil	leaves	roots	**Pb**	soil	leaves	Roots	**Hg**	Soil	leaves	roots
soil	1			Soil	1			soil	1		
leaves	0.786^{***}	1		leaves	0.870^{***}	1		leaves	0.828^{***}	1	
roots	0.863^{***}	0.836^{***}	1	roots	0.815^{***}	0.788^{***}	1	roots	0.752^{***}	0.712^{***}	1

n = 50 Values listed are Pearson correlation coefficients. $^{**}p<0.01$, $^{***}p<0.001$

Table 7. Correlation between metals in soil, leaves and roots samples

soil	Cd	Pb	Hg	**leaves**	Cd	Pb	Hg	**roots**	Cd	Pb	Hg
Cd	1			Cd	1			Cd	1		
Pb	0.863^{***}	1		Pb	0.842^{***}	1		Pb	0.709^{***}	1	
Hg	0.727^{***}	0.854^{***}	1	Hg	0.779^{***}	0.755^{***}	1	Hg	0.783^{***}	0.658^{***}	1

Values listed are Pearson correlation coefficients. $^{**}p<0.01$, $^{***}p<0.001$

Table 8. The bioconcentration factors (BCF) for metals in relation: leaves /soil, roots /soil at the studied sampling sites

BCF Sampling sites	**Cd**		**Pb**		**Hg**	
	leaves/soil	roots/soil	leaves/soil	roots/soil	leaves/soil	roots/soil
Opuštěná	0.32	0.45	0.18	0.08	0.14	0.11
Vídeňská	0.39	0.48	0.21	0.06	0.78	0.37
Podstránská	0.47	0.63	0.22	0.08	0.35	0.21
Musorgského	0.27	0.47	0.27	0.11	0.64	0.45
Šrámkova	0.71	1.45	0.31	0.18	0.66	0.29

Table 9. The transportation index (T_i) for metals between dandelion roots and leaves

T_i leaves/roots	Cd	Pb	Hg
Opuštěná	0.70	2.54	1.56
Vídeňská	0.81	2.61	2.34
Podstránská	0.76	2.98	1.84
Musorgského	0.57	2.58	1.60
Šrámkova	0.48	1.76	2.61

CONCLUSION

Results obtained in this study have demonstrated that Brno urban soils, contrary to industrial activities and vehicular traffic, are not highly contaminated. Mean concentrations of Cd and Hg in soils samples were below those fixed by the Czech Ministry of Environment (REMC, 1994). The average content of Pb in sampling site Opuštěná was 63.1±14.7 mg/kg, only the value of one sample (76.0 mg/kg) exceeded the maximum tolerable levels defined by the Czech legislation (70.0 mg/kg). The amount of metals measured in soils and plants of the common dandelion corresponded with the contamination load of the sampling place. Inter-metal correlations showed that the sources of Cd, Pb and Hg pollution in Brno urban areas were mainly anthropogenic origin. The differentiation of the airborne contamination was assessed by washing the plant leaves. There was substantial aerial deposition on the leaves at all five sampling sites, which was removed by washing procedure. Elimination of the metals by washing was confirmed by statistically significant results.

Significant correlations were found between the amount of Cd in the soil and in the dandelion roots (r= 0.863) and between the amount of Pb (r= 0.870) and Hg (r= 0.828) in the soil and in the dandelion leaves. Analysis of dandelion roots and leaves could be used for differentiating between atmospheric pollutants and soil contaminants. Because internal transport of Hg and Pb from roots to leaves of vascular plants is limited, the higher content of Pb and Hg in dandelion leaves compared with roots indicated a preferential atmospheric receipt of these metals. Cd was preferentially accumulated in dandelion roots and identified soil contamination. The correlation analysis supported this conclusion and confirmed dandelion roots as a good biomonitor for Cd and dandelion leaves for Hg and Pb pollution. Nevertheless, analyses of both dandelion leaves and roots together with soils are recommended for study of pollution because the chemical status of plant is influenced not only by metals content in the environment but there are also many other biotic, abiotic and seasonal factors that affect metal accumulation by plant.

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