Bulgarian Journal of Agricultural Science, 20 (No 2) 2014, 325-329
Agricultural Academy

PLANTAGO LANCEOLATA L. AS A BIOMONITOR OF TRACE ELEMENTS IN AN URBAN AREA

S. PETROVA[1], I. VELCHEVA[1], L.YURUKOVA[2] and M.BEROVA[3]
[1]*University of Plovdiv "Paisii Hilendarski", Faculty of Biology, Department of Ecology and Environmental Conservation, BG – 4000 Plovdiv, Bulgaria*
[2]*Bulgarian Academy of Sciences, Institute of Biodiversity and Ecosystem Research, BG – 1113 Sofia, Bulgaria*
[3]*Agricultural University of Plovdiv, BG - 4000 Plovdiv, Bulgaria*

Abstract

PETROVA, S., I. VELCHEVA, L.YURUKOVA and M.BEROVA, 2014. *Plantago lanceolata* L. as a biomonitor of trace elements in an urban area. *Bulg. J. Agric. Sci*., 20: 325-329

To get real information on environmental pollution with trace elements (Al, Cd, Cr, Cu, Pb and Zn) and their mobility in urban ecosystems, *Plantago lanceolata* L. plants and soil samples were collected in May 2010. Sampling sites were selected in urban areas with different anthropogenic impact (Plovdiv, Bulgaria). Significantly, elevated concentrations of Cd, Cr and Al were registered in all samples. Differences between trace element concentrations in sampled phytomass led to evaluation of the urban gradient theory in the context of the city of Plovdiv. The results of the investigation revealed the main problems and unfavorable factors, which affected urban vegetation. Automobile transport was found to be the major source of contamination, followed by the industry. The contribution of some additional factors as canyon-street effect, wind rose, climate and topography of the region towards forming the environmental state was discussed. The approach used in our study could be successfully applied as a model to explore possibilities of using other herbaceous plant species for biomonitoring of the urban environmental status.

Key words: trace elements, urban environment, biomonitoring, herbaceous plants

Introduction

Urban conditions are specific, where increased amounts of trace elements are found in air and soil as a mark of environmental load. This situation is related to those elements' accumulation in plant tissues, disturbing the phenology and physiology cycles and worsening the health conditions of plants. Vegetation, especially herbs, can act as biomonitors of air pollution and habitat quality because of their wide distribution and accessibility (Malawska and Wilkomirski, 2001; Kardel et al., 2010; Gjorgieva et al., 2011). The simultaneous collection of leaf and root samples provides an additional possibility of distinguishing the possible sources of trace elements for plant uptake, particularly of the metallic pollutants (Kabata-Pendias and Tarlowski, 1981; Kabata-Pendias et al., 1989; Punz and Sieghardt, 1993).

In our approach, we quantified the content of Al, Cd, Cr, Cu, Pb and Zn in *Plantago lanceolata* L. plants, sampled from different areas of the city of Plovdiv, and the content of same elements in the soils, in which these plants were growing, respectively. For better understanding of the mechanisms of trace element accumulation, mobility and transport, the aboveground and underground phytomass samples were analyzed separately. Our work was based on the hypothesis that values obtained can be used to trace the source of pollution; to distinguish areas on the urban landscape with specific types of pollution, as well as to provide an adequate assessment of the urban environment quality. As this species is a natural remedy in traditional medicine and is the raw material for many herbal mixtures, it was also important to check whether the ribwort plantain can accumulate toxic elements at higher concentrations than those accepted by World Health Organization (WHO, 2007).

Materials and Methods

Study area and sampling sites

This study was performed in the city of Plovdiv (N 42°9' E 24°45'), Bulgaria, one of the most densely populated cities in the country (over 340 000 inhabitants on 102 km^2). The selection of the sampling sites was made following the urban gradient hypothesis (McDonell and Hans, 2008). Seven sampling sites with different type of anthropogenic activity were chosen on the urban landscape (using GPS Garmin eTrex Vista HCx), as follows: Site 1 - NE district, green area, near an industrial area; Site 2– East district, suburb, near a city park; Site 3– SE district, suburb, near Rail station; Site 4 (green belt in the middle of busy street) and Site 5 (city park) – urban area, the real center of Plovdiv; Site 6– SW district, suburb, near a city park; Site 7 - West district, suburb, near city park.

Methodology of sampling

The sampling period was 25-26 May 2010. Ten whole plants at flowering stage, growing at 5-10 m away from intensive traffic, were collected from all sampling sites. Exception was Site 4, where plants were collected from the green belt of the interim alley, up to 1 m from the roadway. The aboveground and underground plant parts were carefully separated and the representative samples were prepared for analyses. Soil samples were taken immediately surrounding the roots of the collected plants, at a depth of 0-40 cm. All the samples were stored in clean, labeled, polyethylene bags, closed tightly to avoid contamination during transportation. In laboratory conditions, the samples were air-dried, ground to a powder and homogenized.

Chemical analysis

The chemical analyses of plant samples were carried out in the Faculty of Chemistry, University of Plovdiv. About 1 g ground plant material was treated with 5 ml 65% nitric acid (Merck) for 24 h at room temperature and then for 5 min at 600 W (Microwave Digestion System CEM MDS 81D) in closed vessels. After cooling (1 h), vessels were opened and 2 ml nitric acid and 3 ml 30% hydrogen peroxide were added and were left to react for another 1 hour. For full digestion of the organic matter, samples were treated for 10 min again at 600 W. The content of Al, Cd, Cr, Cu, Pb and Zn was determined by inductively coupled plasma mass spectrometry(ICP-MS) using instrument Agilent 7700 ICP-MS (2009), DF 1000. All samples, blanks and standards were calibrated with international standards. Quality control was performed using the standard reference plant material (NCS DC73348).The chemical analyses of soil samples were carried out in Accredited Laboratory Complex, Agricultural university-Plovdiv, by ICP-MS (ISO 11466:95 and ISO 11047:95).

Statistical analysis

For evaluation of determined values, the descriptive statistical analysis was applied. The means of type sample and area location were evaluated by one-way ANOVA ($p<0.05$). For grouping the studied elements the cluster analysis was used, and the relationships between the contents of individual elements in collected samples were studied using Pearson correlation coefficients. All statistical analyses were done with the STATISTICA 7.0 statistical package (StatSoft Inc., 2004).

Results and Discussion

The content of Cd, Cr, Cu, Pb and Zn in the soil samples, collected immediately surrounding the roots of analyzed plants, was presented in Table 1. The descending row of the studied trace elements was found as follows: Zn >Pb> Cu > Cr> Cd ($p<0.05$). Data were compared with the Bulgarian permissible levels of contaminants in urban soils (Regulation norm 3, 2008). According to this legal document, our results were significantly lower than the maximal legal limit and we could not speak about any soil contamination in the area of Plovdiv, in general.

Concentrations of Al, Cd, Cr, Cu, Pb and Zn in the aboveground and underground phytomass sampled from the selected areas were given in Table 1. The descending row of the analyzed elements in both plant parts was found as follows: Al > Zn > Cu >Pb> Cr > Cd ($p<0.05$). Data were compared with the maximal permissible content of the elements Cd (0.3 $mg.kg^{-1}$) and Pb (10 $mg.kg^{1}$) in herbal medicines, given by WHO (2007). According to this criterion, all root samples were above the limit for Cd, while in the leaves its permissible level was exceeded only in Site 6 and Site 4. Increased lead content was measured only in the leaf samples from the intense traffic area – Site 4.

***Aluminum*:** Normal concentrations of Al in vascular plant species vary up to 200 $mg.kg^{-1}$ dry weights (Kabata-Pendias and Pendias, 1992). The mean values for Al concentrations in plantain leaves and roots, found in this study, were 224.43±6.4 $mg.kg^{-1}$and 649.57±18.9 $mg.kg^{-1}$, respectively. Maximal concentrations were detected in samples from Site 3, but almost all analyzed samples revealed an increased Al content in comparison with mentioned norm, especially in roots. Assuming the shoot-to-root ratio - lowest from all examined trace elements (0.345), it could be supposed that this element was soil derived, entered into the plants mainly through the roots and *P. lanceolata* showed a tendency to accumulate it.

***Cadmium*:** Generally, it is accepted that the normal Cd concentrations in plants are 0.2-0.8 $mg.kg^{-1}$and in soil the critical level is between 3-5 $mg.kg^{-1}$ (Kabata-Pendias and Pendias, 1992). Based on our results the area of Plovdiv could be assessed as not highly polluted by Cd according to these criteria. Plants take up this element primarily from the soil

Table 1
Concentrations of Al, Cd, Cr, Cu, Pb and Zn (mean±SD) in collected leaf, root and soil samples from the selected areas in Plovdiv (mg.kg^{-1})

Element	Sample	Site 1	Site 2	Site 3	Site 4	Site 5	Site 6	Site 7
Al	Leaves	284±8.2	224±5.6	370±11.5	171±5.5	128±3.3	252±6.6	142±3.8
	Roots	620±18.0	602±16.9	1266±43.0	597±15.5	377±8.3	555±13.3	530±17.0
	Soil	-	-	-	-	-	-	-
Cd	Leaves	0.14±0.02	0.25±0.03	0.21±0.02	0.58±0.03	0.18±0.01	0.45±0.02	0.33±0.02
	Roots	0.25±0.02	0.54±0.02	0.48±0.02	0.80±0.04	0.49±0.03	1.05±0.04	0.55±0.03
	Soil	0.38±0.02	0.2±0.01	<0.1	0.72±0.03	0.26±0.01	<0.1	<0.1
Cr	Leaves	1.32±0.11	1.26±0.12	1.08±0.1	2.82±0.13	0.49±0.08	1.5±0.07	0.25±0.04
	Roots	2.33±0.16	2.55±0.15	5.78±0.23	2.0±0.14	1.15±0.05	2.31±0.09	1.74±0.12
	Soil	23.2±1.1	32.5±1.5	38.8±1.8	24.4±1.1	15.75±0.73	30.3±1.4	49.1±2.3
Cu	Leaves	8±0.4	11±0.33	11±0.3	9±0.49	10±0.24	10±0.48	10±0.27
	Roots	17±0.63	18±0.86	29±0.96	19±0.46	19±0.48	23±0.55	20±0.54
	Soil	43.8±1.3	27.6±0.8	26.3±0.8	58.04±1.7	30.2±0.9	27.3±0.8	37.4±1.1
Pb	Leaves	2.2±0.09	3.6±0.18	3.0±0.14	10±0.36	1.3±0.06	2.6±0.09	2.0±0.09
	Roots	2.7±0.17	5.5±0.28	4.2±0.19	4.0±0.15	3.9±0.15	4.9±0.19	3.3±0.15
	Soil	44.25±1.86	36.6±1.53	39.6±1.66	150±6.29	39.3±1.7	29.7±1.25	48.4±2.03
Zn	Leaves	31±1.55	44±2.55	55±1.27	43±0.95	38±0.99	53±2.49	44±1.1
	Roots	95±4.37	185±8.7	134±4.42	72±1.87	111±2.44	149±3.73	123±3.2
	Soil	140.1±4.1	94.3±2.7	102.3±3.0	215.8±6.3	100.4±3.0	99.5±2.9	110.1±3.0

through the roots and from the air through the leaves. The ratio between leaf and root Cd content, especially in Site 4 (0.725), Site 7 (0.600) and Site 1 (0.560), illustrated a significant percentage of its aerial origin. The statistical analysis revealed that a strong positive correlation ($r=0.83$, $p<0.05$) existed between Cd concentrations in leaves and in roots,confirming that in the studied area this element was air derived.

***Chromium*:** An excess of chromium was observed in aboveground phytomass from Site 4 (shoot to root ratio 1.41), followed by Site 6 (0.649) and Site 1 (0.567), which was in agreement with the fact that it was air derived and the main emitter was transport (because maximums were found in areas with highest traffic activity - Site 3 and Site 4).

***Copper*:** Although copper is an essential enzymatic element for normal plant growth and development, it can be toxic at concentrations above 25 mg.kg^{-1}(Allen, 1989). The most common sources for Cu in the environment are pesticides, fertilizers, industries and sewage sludge. The mean values for Cu concentrations in plantain leaves and roots reached up to 9.86±0.36 mg.kg^{-1}and to 20.71±0.64 mg.kg^{-1}, respectively. Maximal values were obtained from Site 2 and Site 3, and from root samples in Site 6. Shoot-to-root ratio was highest in samples from Site 2 (0.611), followed by Site 5 (0.526). Obviously, *P. lanceolata* had an ability to bioaccumulate Cu, as a negative correlation ($r=-0.75$, $p<0.05$) between soil and root concentration of copper was found. This fact was confirmed by the descending order of the studied elements where Cu level was higher than the lead one in the plant tissues, although in the soil it was contrary wise.

***Lead*:** Normal concentrations in plants are 0.1-10 mg.kg^{-1} according to Kabata-Pendias and Pendias (1992). In our study, the high lead concentrations in aerial parts of plantain from Site 4 (shoot-to-root ratio is 2.5) was due to the lead coming from the exhaust emissions of vehicles, and its presence in the roots (4±0.18 mg.kg^{-1}) and in the soil (150±6.29 mg.kg^{-1}) due to pollution with waste water from different sources and wet deposition.

***Zinc*:** Zn is not considered highly phytotoxic and its toxicity limit varied between 300 and 400 mg.kg^{-1} (Kabata-Pendias and Pendias, 1992). A plant foliar concentration of 100 mg.kg^{-1} has been quoted by Allen (1989) as a critical indicator of whether the environment is polluted with Zn. In our study, the average Zn concentration was 44±1.56 mg.kg^{-1}in leaves and 124±4.1 mg.kg^{-1}in roots. According to Allen's criterion, it could be concluded that the city of Plovdiv was contaminated to some extent with this metal. The dominant zinc content in the roots, rather than the aboveground plant organs, was evidence that it was soil derived and *P. lanceolata* showed a tendency for bioaccumulation (correlation between soil and root content was $r=-0.78$, $p<0.05$).

Based on trace elements contents in *Plantago lanceolata*, the studied sampling sites were divided into two major clusters (Figure 1). The first one consisted of the pair Site 6 – Site 7 (both situated in the West suburban area) and the pair Site 2 (East suburban area) – Site 5 (Central urban area). The second included the pair Site 1 (North industrial area) – Site 4 (Central urban area) and Site 3 (Southeast suburban area).

Root content of each one of six studied trace elements in all sampling sites was from 22% (Pb, Site 1) to 696% (Cr, Site 7) higher than that in the aboveground phytomass. Exception was established only for chromium and lead in plantain leaves from Site 4. This site was situated in the real center of Plovdiv, along one of the major traffic arteries, and characterized by the presence of tall buildings, high trees, some other barriers, and in this case, we could talk about the impact of the canyon-street effect (Vardoulakis et al., 2003; Tong and Leung, 2012). The ranking of the studied zones by cluster analysis (Figure 1) confirmed the urban gradient hypothesis only to some extent. Explanations of the observed deviations could be found in the topography of the studied region, the presence of the canyon-street effect, the character of the wind rose, climate conditions and some other factors.

We have compared our results with data from a previous biomonitoring research in the region of Plovdiv using same plant species (Dimitrova and Yurukova, 2005). It could be noticed that in the North part of the town, the contamination with Cd, Cu, Pb and Zn was from 1.5 to 10 times lower. When regarding the Central city part, we found a slight decrease (1.5-3 times) in the concentrations of the mentioned elements.

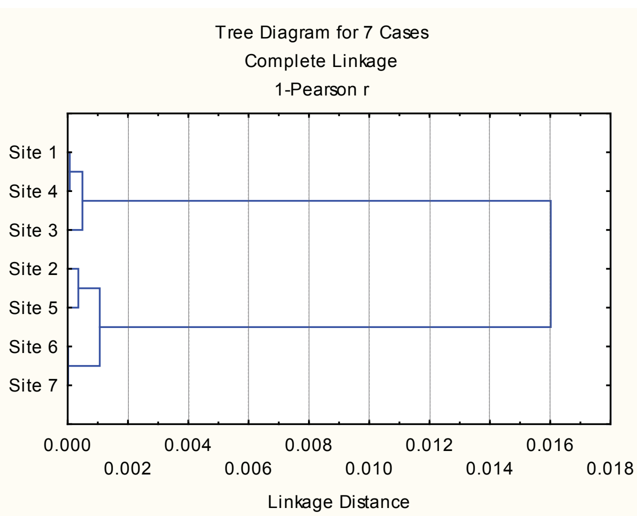

Fig. 1. Cluster analysis of the selected sampling sites

Conclusion

The results revealed that the distribution of the studied trace elements in analyzed plant organs was not homogenous and probably was dependent on the physicochemical characteristics of the element and its origin. Cd, Cr, Cu and Pb were defined mainly as air-derived pollutants in the studied urban environment, while aluminum and zinc - as soil derived. Most of the analyzed trace elements showed a tendency to accumulate primarily in the roots rather than in the leaves, but the mechanism was related to wet and dry atmospheric deposition and the elements' mobility in the soil and plant tissues.

Differences between trace element concentrations in sampled phytomass led to evaluation of the urban gradient theory in the context of the city of Plovdiv. Automobile transport was found to be the major source of contamination, followed by the industry. The contribution of some additional factors as the canyon-street effect, the character of the wind rose, climate conditions and the topography of the studied region towards forming the environmental state was established too.

The approach used in our study can be successfully applied as a model to explore possibilities of other herbaceous plant species being used for biomonitoring of the urban environmental quality.

References

Allen, S. E., 1989. Chemical analyses of ecological material.2nd Edition. *Blackwell Scientific*, London, 380 pp.

Dimitrova, I. and L. Yurukova, 2005. Biondication of anthropogenic pollution with *Plantagolanceolata* (*Plantaginaceae*): metal accumulation, morphological and stomatal leaf characteristics. *PhytologiaBalcanica*, **11** (1): 89-96.

Gjorgieva, D., T. Kadifkova-Panovska, K. Baĉeva and T. Stafilov, 2011. Assessment of heavy metal pollution in Republic of Macedonia using a plant assay. *Archives of Environmental Contamination and Toxicology*, **60:** 233-240.

Kabata-Pendias, A. and P. Tarlowski, 1981. Impact of a copper smelter on agricultural environment. Part II. Contamination of herbage. *Roczn. Glebozn*, **32:** 215-221.

Kabata-Pendias, A., B. Galczynska and S. Dudka, 1989. Baseline zinc content of soils and plants in Poland. *Environmental Geochemistry and Health*, **11**: 19-24

Kabata-Pendias, A. and H. Pendias, 1992. Trace elements in soils and plants. Boca Raton, FL, *CRC Press*, 486 pp.

Kardel, F., K. Wuyts, M. Babanezhad, U.W.A. Vitaharana, T. Wuytack, G. Potters and R. Samson, 2010. Assessing urban habitat quality based on specific leaf area and stomatal characteristics of *Plantago lanceolata* L. *Environmental Pollution*,

158: 788-794.

Malawska, M. and B. Wilkomirski, 2001. An analysis of soil and plant (*Taraxacumofficinale*) contamination with heavy metals and polycyclic aromatic hydrocarbons (PAHs) in the area of the railway junction Ilawa Glowna, Poland. *Water, Air and Soil Pollution*, **127:** 339-349.

McDonnell, M. J. and A. K. Hahs, 2008. The use of gradient analysis studies in advancing our understanding of the ecology of urbanizing landscapes: current status and future directions. *Landscape Ecology*, **23**: 1143-1155.

Punz, W. F. and H. Sieghardt, 1993. The response of roots of herbaceous plant to heavy metals.*Environmental and Experimental Botany*, **33**: 85-98.

Regulation Norm 3, 2008. About permitted level of contaminants in soils. eea.government.bg/bg/legislation/soil/normipochvi.rtf. (Accessed on 13.11.2012).

StatSoft Inc. 2004. STATISTICA (Data analysis software system), Vers. 7. Computer software. [htpp//www.statsoft.com].

Tong, N. Y. O. and D. Y. C. Leung, 2012.Effects of building aspect ratio, diurnal heating scenario, and wind speed on reactive pollutant dispersion in urban street canyons. *Journal of Environmental Sciences*, **24** (12): 2091-2103.

Vardoulakis, S., B. E. A. Fisher, K. Pericleous and N. Gonzalez-Flesca, 2003. Modelling air quality in street canyons: a review. *Atmospheric Environment*, **37**: 155-182.

WHO Guidelines for assessing quality of herbal medicines with reference to contaminants and residues, 2007. *WHO Press,* World Health Organization, Geneva, Switzerland.

Received August, 2, 2013; accepted for printing February, 2, 2014.