



## Possibilities of using deciduous tree species in trace element biomonitoring in an urban area (Plovdiv, Bulgaria)

Slaveya Petrova<sup>1</sup>, Lilyana Yurukova<sup>2</sup>, Iliana Velcheva<sup>1</sup>

<sup>1</sup> University of Plovdiv "Paisii Hilendarski", Faculty of Biology, Department of Ecology and Environmental Conservation, Plovdiv 4000, Tzar Assen Str. 24, Bulgaria

<sup>2</sup> Bulgarian Academy of Sciences, Institute of Biodiversity and Ecosystem Research, Sofia 1113, Acad. G. Bonchev Str., Bl. 23, Bulgaria

### ABSTRACT

Leaves of *Acer platanoides* L., *Aesculus hippocastanum* L. and *Betula pendula* Roth. were collected from urban sites with different anthropogenic impact in the city of Plovdiv (Bulgaria). Concentrations of Cd, Cr, Cu, Fe, Pb and Zn in foliage samples were analyzed by ICP–MS. Three studied deciduous trees clearly showed variations in element concentrations depending on the sampling site (Pb, Fe, Cr) and the plant species (Zn). *Betula pendula* was found to be a better bioaccumulator for the elements Cr, Cd, and especially for Zn. Our approach could be successfully applied in the monitoring of air pollution due to trace elements in urban areas, regardless of their size and topography.

**Keywords:** *Acer platanoides*, *Aesculus hippocastanum*, *Betula pendula*, urban environment, air pollution



**Corresponding Author:**

**Slaveya Petrova**

☎ : +359-32-261519

☎ : +359-32-261566

✉ : slaveya\_petrova@uni-plovdiv.bg

### Article History:

Received: 13 August 2013

Revised: 09 December 2013

Accepted: 09 December 2013

doi: 10.5094/APR.2014.024

### 1. Introduction

Most of trace metals in terrestrial ecosystems originate from atmospheric wet and dry deposition. Studies of their transport and mobilization have attracted attention for many years, and have shown that trace metals are persistent, widely dispersed in the environment, and interacting with different natural components, cause threat to human health and environment (Alcamo, 1992; Pacyna et al., 2007; Takuchev, 2011a; Takuchev, 2011b).

Green system in the cities is a typical example of ecosystem that influence and is influenced by the urban environment and industrial activities. Town conditions are specific, where increased amount of trace elements are found in air and soil as markers of environmental load. Increasing or decreasing of the element levels in plant organs depend on either abundance or deficiency in the soil and air. This situation has been related to the element accumulation increasing in leaves, disturbing of phenology and physiology cycles and worsened healthy conditions of woody plants (Supuka et al., 2008).

In severe cases, pollutants can cause visible plant injuries and, in some extreme cases, even plant death. Changes in foliar element concentrations, however, can take place long before pollution-mediated plant injuries, and hence foliar element composition could be used as biomonitor to investigate the spatial distribution of atmospheric pollution. Plants have a very large surface area and their leaves function as an efficient pollutant-trapping device, therefore, their assimilative organs are directly

affected by air pollution. Contaminants deposited onto the leaf surface can further penetrate into inner tissues (Cook et al., 1994). Leaves with different shapes and sizes have different particle retention capabilities (Smith, 1981), and pubescent leaves have been shown to have greater scavenging efficiency than hairless ones both for inorganic and organic contaminants (Little and Wiffen, 1977; Howsam et al., 2000). In addition, the roughness and integrity of the cuticle affect particle adhesion on the leaf surface (Neinhuis and Barthlott, 1997), and cuticular and epicuticular waxes play an important role in the sorption of lipophilic compounds in leaves (Simonich and Hites, 1994). This feature of foliage's capability to accumulate contaminants has been used for years to study pollution effects in forests (Rautio and Huttunen, 2003) as well as in urban areas (Wyttenbach et al., 1985; Alfani et al., 1995; Aksoy and Ozturk, 1997; Bargagli, 1998; Salemaa et al., 2001).

Main topic of the urban ecology is to investigate the relationships between the spatial pattern of urbanization and the ecological processes (Loucks, 1994; Breuste et al., 1998; Sukopp, 1998; Sukopp, 2006). Most of the studies on the human impact on the natural habitats search for any correlations between the changes in natural systems and the level of urbanization (population density, percentage of built-up areas, etc.). The intensity of these disturbances is expected to decline predictably with distance from the core of the city in order: urban – suburban – cultivated – managed – natural areas. The theories considering the urban gradient were based mainly on some American megacities such as Chicago, San Francisco, Boston (McDonnell and Pickett, 1990;

McDonnell and Hahs, 2008) and studies of smaller urban areas are few (Dimitrova and Yurukova, 2005; Yilmaz et al., 2006).

The use of plants as passive biomonitors to complete the information on trace elements deposition from fully or semiautomatic gauges, commonly used in current pollution monitoring programs, has gained increasing attention. This reliable, versatile and inexpensive method can assist us on the subject of health and environmental protection against potentially hazardous trace elements. Providing a high density of sampling points, the biomonitors are very effective for tracing maps of airborne metal contamination in the urban environments (Baycu et al., 2006; Klumpp et al., 2009). An advantage of plants as biomonitors is that they are effective collectors which reflect the accumulated effect of environmental pollution and accumulation of toxicants from atmospheric pollution (deposition, binding and solubility of metals on the leaf surface).

Different biomonitors have been used for evaluation of the distribution of heavy metal pollution: mosses and lichens (Steinnes, 1993; Gonzalez et al., 1996; Culicov and Yurukova, 2006; Anicic et al., 2009; Kularatne and de Freitas, 2013), grasses (Klumpp et al., 2009), many trees as chestnut (Yilmaz et al., 2006), maple, linden, willow, birch (Piczak et al., 2003), poplar (Djingova et al., 1995), oak (Monaci et al., 2000). Leaves of deciduous trees are recognized as useful air pollution biomonitors for trace elements from almost 20 years (Markert, 1993; Bargagli, 1998), but still there are some difficulties to compare data among different phytomonitoring studies. The problems are not only due to the use of various plant species, but also from the application of diverse experimental approaches (Tomasevic et al., 2011).

Biomonitoring studies with tree species in urban habitats on Balkans are scarce (Yilmaz et al., 2006; Sucur et al., 2010; Tomasevic et al., 2011). Yilmaz et al. (2006) have studied Pb, Cd, Zn and Cu content in *A. hippocastanum* leaf samples from urban, suburban and roadsides in the Thrace region of Turkey (Istanbul, Edirne, Tekirdag, Corlu). Baycu et al. (2006) measured concentrations of Cd, Ni, Pb and Zn in foliar samples of *A. platanoides*, *A. hippocastanum* and *B. pendula*, collected from the urban area of Istanbul (Turkey).

There are only limited studies considering the accumulation of trace elements in urban plant species from Bulgaria. Doncheva–Boneva (2000) studied oak leaves from different sites in the region of Sofia. Dimitrova and Yurukova (2005) published some data about *Plantago lanceolata* plants from an industrial and a central part of Plovdiv.

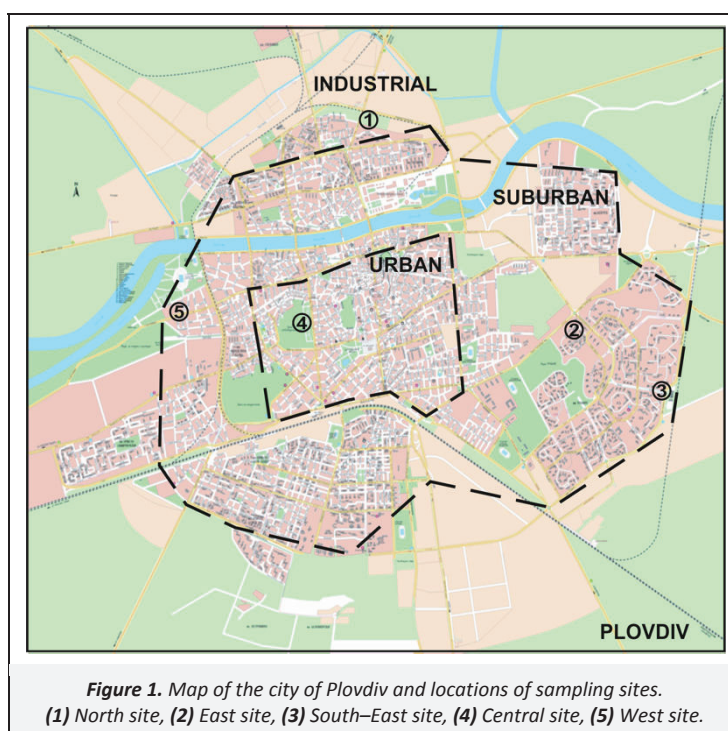
Our study was conducted in the city of Plovdiv which was pointed as one of the most polluted cities in Europe in 2008 by National Institute of Statistics of Italy (ISTAT, 2011). We aimed (i) to make an impact monitoring considering the broad leaved tree species as biomonitors of trace elemental air pollution (Cd, Cr, Cu, Fe, Pb and Zn), and (ii) to evaluate some deciduous tree species which could be applied for the air pollution phytomonitoring in urban environment.

## 2. Materials and Methods

### 2.1. Study area and sampling sites

Plovdiv (42°9' N, 24°45' E) is the second-largest city of Bulgaria after the capital Sofia with a population of over 338 000 inhabitants (NSI, 2011). Plovdiv is situated in south Bulgaria on the two banks of the Maritsa River. The climate is temperate with mild influence from the Mediterranean Sea and a large temperature range between summers and winters. The average annual temperature is 12.3 °C with maximum in July (32.3 °C) and minimum in January (6.5 °C). The average relative humidity is 73%. It is highest in December (86%) and lowest in August (62%). The total precipitation is 540 mm – the wettest months of the year are May and June with an average precipitation of 66.2 mm, while the driest is August with an average of 31 mm. Gentle winds with speed of up to 1 m s<sup>-1</sup> represent 95% of all winds during the year. The prevailing wind direction is from west, rarely from east.

For the purposes of our study, five sites were selected on the basis of typology of urban green they represent and the type of anthropogenic impact. According to the urban gradient scheme, city's area was divided to urban, suburban and industrial sectors (Figure 1). The selection of sites was based on the identification of common features in trees like age, size, canopy density, light, and temperature conditions.



The North site (N 42°10'32.3", E 24°45'53.4") was chosen in the city outskirts, close to an industrial area (North industrial zone in the city of Plovdiv) with a fossil fuel-fired power plant, cosmetic production, glass production and other small-point sources of contaminants. Examined trees were growing in a large green area, located at a distance of about 1 km to the East of the industrial zone.

The East site (N 42°08'24.3", E 24°47'06.2") was situated in the "Trakiya" suburb, and was characterized by high traffic and moderate household pressure. Selected trees were growing in small green patches, 5–10 m away from tall buildings.

The South–East site (N 42°07'42.2", E 24°47'40.5") was also chosen in the "Trakiya" suburb, but the trees were located in small green patches near (50–100 m) a Railway station.

The West site (N 42°07'54.4", E 24°42'18.0") was chosen in the "Smirnenski" suburb, subjected to a moderate anthropogenic impact. Examined trees were located in small green patches around the buildings (5–10 m distance) similarly to the other sites.

The Central site (N 42°08'22.8", E 24°44'24.1") was situated in the center of the town of Plovdiv, along one of the major streets (Ruski Boulevard) with intensive daily traffic. The examined trees were growing in a big city park (Natural monument "Bunardjik"), on the East slope of the Bunardjik Hill, 10–20 m away from the boulevard.

## 2.2. Sampling and sample preparation

In this study we studied leaves from three woody plants – *Acer platanoides* L., *Aesculus hippocastanum* L. and *Betula pendula* Roth. These deciduous trees were extensively used for greenery in Plovdiv and in a number of European cities and are commonly met at rural and natural areas. At each sampling site, at least three individuals per species were sampled in the end of June 2010. Small branches located 2.5–3 m above the ground, from the outer part of the canopies and from the four sides of the tree (N, S, E, W) were cut by Expert Tree Pruner with telescopic handle 1.5–2.5 m (Draper Tools, UK) (De Nicola et al., 2008). In order to obtain a homogeneous sample, a large number of one-year-old leaves, comparable in size and shape, were taken by hand from the branches, taking care to minimize contact with the leaf surface. Usually 80–100 fully expanded leaves per tree were collected and a composite sample was prepared for analyses. All the samples were stored in clean, labeled, polyethylene bags, closed tightly to avoid

contamination during transport. The collected material for chemical analyses was air dried at room temperature for two weeks, ground to a powder and homogenized.

## 2.3. Chemical analysis

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 Cd, Cr, Cu, Fe and Zn was determined by inductively coupled plasma–mass spectrometry (ICP–MS) (Agilent 7700 ICP–MS). ICP–MS system was calibrated with international standards. Quality control was performed using the standard reference plant material (NCS DC73348): the percent recovery ranged from 91 to 99%, depending on each analyzed element.

## 2.4. Data processing

All data presented in the present study were an average of triplicate analysis of three separate subsamples. The concentrations were expressed as arithmetic means and standard deviations (mean±SD). For the statistical evaluation of the data obtained, the raw values of the three subsamples per species per site were used.

Multi ANOVA (MANOVA) and Student/Fisher test were used for testing the differences of elemental concentrations, both between the three plant species in one site and also between the five studied sampling sites ( $p<0.05$ ). A cluster analysis was used for grouping the studied sampling sites on the basis of trace elemental contents of leaf samples. Relationships between the contents of individual elements in collected leaf samples were tested using Pearson correlation coefficients ( $p<0.05$ ). The data were also processed with Principal Components Analysis (PCA). All statistical analyses were made with the STATISTICA 7.0 statistical package (StatSoft, 2004).

## 3. Results

The element concentrations in leaves of *A. platanoides*, *A. hippocastanum* and *B. pendula* from all studied sites were shown in Table 1. Statistically, it was found that the analyzed trace elements in our leaf samples could be ordered as follows: Fe>Zn>Cu>Pb>Cr>Cd.

**Table 1.** Element concentrations (mg kg<sup>-1</sup> dry weight) in collected leaf samples from the urban sites with different anthropogenic impacts in the city of Plovdiv

| Element | Species                 | North Site | East Site  | South–East Site | Central Site | West Site  |
|---------|-------------------------|------------|------------|-----------------|--------------|------------|
| Cd      | <i>A.platanoides</i>    | 0.10±0.001 | 0.20±0.01  | 0.50±0.02       | 0.22±0.007   | 0.10±0.001 |
|         | <i>A. hippocastanum</i> | 0.17±0.009 | 0.14±0.009 | 0.23±0.011      | 0.17±0.009   | 0.10±0.005 |
|         | <i>B. pendula</i>       | 0.17±0.006 | 0.36±0.008 | 0.34±0.006      | 0.25±0.014   | 0.18±0.006 |
| Cr      | <i>A.platanoides</i>    | 0.41±0.04  | 0.39±0.04  | 0.32±0.04       | 0.65±0.04    | 0.49±0.06  |
|         | <i>A. hippocastanum</i> | 0.11±0.02  | <0.1       | 0.3±0.044       | 0.71±0.088   | <0.1       |
|         | <i>B. pendula</i>       | 0.31±0.02  | 0.44±0.01  | 0.43±0.017      | 1.2±0.03     | 0.27±0.017 |
| Cu      | <i>A.platanoides</i>    | 6±0.29     | 12.1±1.17  | 6.4±0.16        | 5.6±0.31     | 9.4±0.36   |
|         | <i>A. hippocastanum</i> | 8±0.22     | 7.1±0.39   | 12.4±0.41       | 9.3±0.44     | 4.8±0.23   |
|         | <i>B. pendula</i>       | 4.5±0.21   | 5.1±0.21   | 4.9±0.19        | 5.3±0.45     | 3.4±0.40   |
| Fe      | <i>A.platanoides</i>    | 113±3.05   | 95±3.04    | 100±3.9         | 252±5.8      | 90±2.7     |
|         | <i>A. hippocastanum</i> | 88±2.55    | 111±4.66   | 99±2.84         | 135±3.51     | 77±2.54    |
|         | <i>B. pendula</i>       | 89.3±1.61  | 97.9±1.96  | 103.4±1.86      | 249±6.23     | 77.7±1.48  |
| Pb      | <i>A.platanoides</i>    | 1.9±0.07   | 2.4±0.07   | 5±0.14          | 1.83±0.007   | 2.2±0.06   |
|         | <i>A. hippocastanum</i> | 1.7±0.07   | 2.3±0.07   | 4.2±0.12        | 2.18±0.02    | 2.2±0.06   |
|         | <i>B. pendula</i>       | 1.17±0.009 | 3.34±0.023 | 3.27±0.023      | 2.66±0.029   | 1.36±0.021 |
| Zn      | <i>A.platanoides</i>    | 13.3±0.47  | 24±0.74    | 22.7±0.64       | 22.3±0.22    | 20.4±0.73  |
|         | <i>A. hippocastanum</i> | 11.6±0.33  | 14.2±0.36  | 21.8±0.7        | 24.6±0.27    | 15.3±0.52  |
|         | <i>B. pendula</i>       | 104±0.94   | 102±1.02   | 142±1.14        | 140±1.26     | 122±1.1    |

Statistical evaluation with MANOVA revealed strong significant differences on trace element concentrations depending of the plant species ( $p < 0.001$ ). From the three studied plant species, *B. pendula* was found to be a better bioaccumulator for the elements Cr, Cd, and especially for Zn (5–10 fold higher levels in comparison with *A. platanoides* and *A. hippocastanum*). *A. platanoides*, followed by *B. pendula*, demonstrated superior ability to retain Fe. *A. hippocastanum* showed a preference to Cu, and all three species had a similar attitude to Pb. Our results on the capacity of these tree species to accumulate heavy metals and toxic elements were in agreement with data from Gorelova et al. (2011).

We could point the Central area of Plovdiv as more polluted with Fe (2–2.5 times), Zn (up to 1.5 times) and Cr (2–3 times) in comparison with other sites. In South–East area, followed by East and Central area, we measured higher concentrations of Cd (1.5–3 times) and Pb (2–2.5 times). Maximal values for the elements Cu (up to 1.5 times greater) were obtained from East area and West area, respectively. The results from MANOVA revealed that there was significant differences in trace elemental contents between sampling sites (for Cr and Pb,  $p < 0.05$ ; for rest studied elements,  $p < 0.001$ ). Cluster analysis showed greater similarity between North and East areas, followed by South–East and West areas, and the distance of Central area was bigger (Figure 2).

Significance of the factors “plant species” and “area location” was also studied on the basis of trace elemental leaf content. Zinc concentration was found to be much more influenced by plant species used for biomonitoring, than to sampling areas. The content of elements Cr, Fe and to some extent Pb in foliage samples was mainly due to the sampling site location. Data for cadmium and copper showed no clear relationship to plant species or studied areas (Figure 3).

#### 4. Discussion

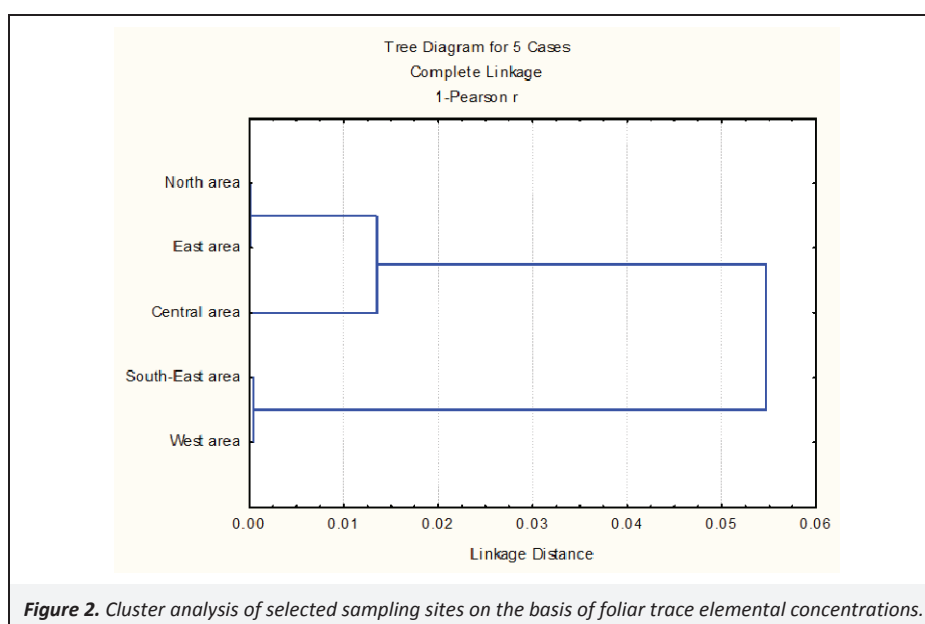
The concentrations of Cr, Cu, Fe and Pb, found in our study, were significantly lower than values reported from Belgrade, Serbia (Tomasevic et al., 2011) in the same plant species which can be explained with the different factors affecting the both cities. We also found an elevated content (2–4 times) of Cd in all three studied species. According to Kabata–Pendias and Pendias (2001), Cd is effectively absorbed by both the root and leaf systems, and it shows a clear tendency to be accumulated in root tissues. The increased cadmium concentration in our leaf samples could not be

a result of soil uptake because we did not found any statistical significant relationships between soil and foliar content of this element in our preceding studies with the same tree species (unpublished data).

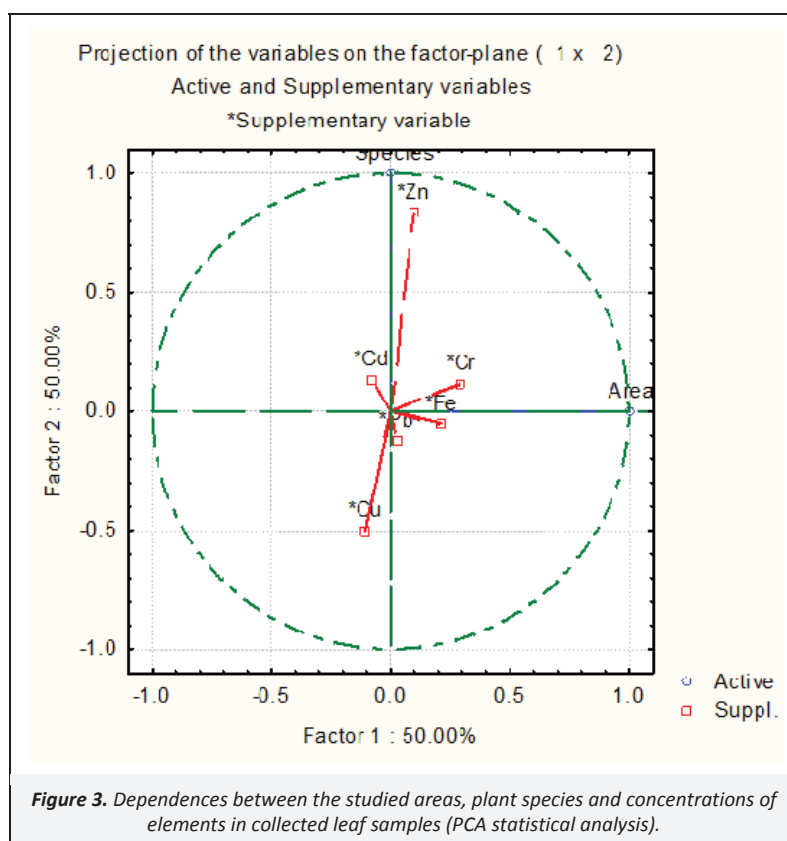
Thus, the mechanism of Cd uptake was mainly air–related and probably the elevated Cd levels in the air could be due to two major point sources – the non–ferrous smelter “KCM–Plovdiv” (located at the 14 km from Plovdiv, South direction) and power plant (located in the North industrial zone) (MOEV, 2010).

From the three studied deciduous tree species, we only found a higher Zn concentration in *B. pendula* foliar samples in comparison with data from Belgrade, Serbia (Tomasevic et al., 2011). PCA analyses confirmed that this element accumulation was influenced to a great extent by plant species used for biomonitoring. Similarly to the Cd, we considered that Zn in plants was air derived. The zinc emissions in the city of Plovdiv could be related mainly to the vehicle exhausts and other waste products from the transport as abrasion of brake linings and tires (Adachi and Tainosho, 2004; Dongarra et al., 2009), and to a smaller degree with the activity of the non–ferrous smelter “KCM–Plovdiv” (MOEV, 2010). Accumulation by the foliar plant parts of the elements Cd, Fe, Pb and Zn, originated from anthropogenic pollution, was also reported by the Zalud et al. (2012) after an experiment with leafy vegetables simulating the atmospheric deposition.

The content of Cu and Cd in *A. hippocastanum* and *B. pendula* leaves from Plovdiv was very similar to that measured from Pizcak et al. (2003) in samples from Wroclaw, Poland, but the levels of Pb in our study were triple in *A. hippocastanum* and twice higher in *B. pendula*. Izrael (1998) showed that the amount of lead in leaves of deciduous and coniferous vegetation in Europe varies between 1.5 and 2.1  $\text{mg kg}^{-1}$ . According this criterion, it could be concluded that the air in Plovdiv, assessed by foliar uptake, is polluted with Pb, as the highest value obtained was  $5.0 \pm 0.1 \text{ mg kg}^{-1}$  (South–East area), i.e. 2.5 fold higher than the average value for Europe. Previous data for atmospheric lead concentrations in Plovdiv (Tarnovska et al., 2003) measured with instrumental methods were quite higher than those, obtained in our study. This might be a consequence of diminished usage of leaded gasoline in favor of the unleaded one, similarly of the concentration trends, reported for other European cities: Belgrade – Tomasevic and Anicic (2010); Warsaw – Dmuchowski and Bytnerowicz (2009); Rome – Gratani et al. (2008).







Central site of Plovdiv was found clearly distinguished by the other four examined sites. Three sites in suburbs – West, South-East and East areas – were found to be not as similar in their pollution level as it was presumed by the urban gradient hypothesis. Higher proximity was found between East and North sites, situated close to an industrial zone. According to the mentioned theory, the pollution level must decrease predictably from the center to the periphery of the town (McDonnell and Hahs, 2008). Our results correspond only to some extent (Table 1). Most likely this is due to the specific characteristics and topography of Plovdiv: the presence of large lawns and abundant planting trees on main roads, tall buildings and other obstacles (like the six hills) which prevent dispersion of emitted pollutants in this area and contribute to the retention of pollutants in the surface air layer, their recirculation and deposition (canyon–street effect). The highest concentrations of traffic–related contaminants Fe, Cr and Zn were measured in this study in the foliar samples from the Central area (Table 1). Their dependence of sampling site location (Figure 3) matched the statements given by Atanasov et al. (2006), that only about 5% of the air pollution in Plovdiv was due to industrial sector, while the remaining 95% was emitted from a variety of sources and mainly by the transportation.

## 5. Conclusions

Our study clearly demonstrated the possibilities of using the leaves of selected deciduous trees to give an adequate assessment of trace elemental pollution and their ability to reflect any microhabitat differences. This approach could be successfully integrated in the regular ecological monitoring in urban areas regardless of their size and topography. Moreover, the surveys on atmospheric contamination have frequently been limited by high cost of instrumental monitoring methods, and difficulties in carrying out extensive sampling in time and space. On the other hand, the instrumental monitoring techniques lack information on impact of atmospheric pollutants on the living systems. The use of biomonitors, which are represented by large numbers of sites all over the monitoring areas, have a wide geographical range and are easy to sample, could be a preferable method for an impact

monitoring of heterogeneous landscapes. The only prerequisite is the sufficient number of sites and samples to obtain statistically reliable data and also using standardized methods for sampling and analyses.

## References

- Adachi, K., Tainosho, Y., 2004. Characterization of heavy metal particles embedded in tire dust. *Environment International* 30, 1009–1017.
- Aksoy, A., Ozturk, M.A., 1997. *Nerium oleander* L. as a biomonitor of lead and other heavy metal pollution in Mediterranean environments. *Science of the Total Environment* 205, 145–150.
- Alcamo, J., 1992. Transboundary air pollution in Central Europe and Eastern Europe, in *Coping with Crisis in Eastern Europe's Environment*, edited by Alcamo, J., IIASA, Parthenon Publishing Group, Carnforth.
- Alfani, A., Bartoli, G., Andolfi, G., 1995. Amount and elemental composition of dry deposition to leaf surface of *Quercus ilex* in the urban area of Naples. *Agricoltura Mediterranea* 194–199 (special volume).
- Anicic, M., Tasic, M., Frontasyeva, M.V., Tomasevic, M., Rajsic, S., Mijic, Z., Popovic, A., 2009. Active moss biomonitoring of trace elements with *Sphagnum girgensohnii* moss bags in relation to atmospheric bulk deposition in Belgrade, Serbia. *Environmental Pollution* 157, 673–679.
- Atanasov, D., Spassova, S., Grancharova, D., Krastev, S., Yankova, T., Nikolov, L., Chakarova, M., Krasteva, P., Genov, N., Stamenov, J., Dimitrov, E., 2006. Air pollution monitoring and modelling system of the town of Plovdiv (phase I). *Journal of Environmental Protection and Ecology* 7, 260–268.
- Bargagli, R., 1998. *Trace Elements in Terrestrial Plants: An Ecophysiological Approach to Biomonitoring and Biorecovery*, Springer–Verlag, Berlin, Heidelberg, NY.
- Baycu, G., Tolunay, D., Ozden, H., Gunebakan, S., 2006. Ecophysiological and seasonal variations in Cd, Pb, Zn, and Ni concentrations in the leaves of urban deciduous trees in Istanbul. *Environmental Pollution* 143, 545–554.

- Breuste, J., Feldmann, H., Uhlmann, O., 1998. Urban ecology, edited by Breuste, J., Feldmann, H., Uhlmann, O., Springer-Verlag, Berlin, Heidelberg, NY, pp. 644–647.
- Cook, C.M., Sgardelis, S.P., Pantis, J.D., Lanaras, T., 1994. Concentrations of Pb, Zn, and Cu in *Taraxacum* spp. in relation to urban pollution. *Bulletin of Environmental Contamination and Toxicology* 53, 204–210.
- Culicov, O.A., Yurukova, L., 2006. Comparison of element accumulation of different moss- and lichen-bags, exposed in the city of Sofia (Bulgaria). *Journal of Atmospheric Chemistry* 55, 1–12.
- De Nicola, F., Maisto, G., Prati, M.V., Alfani, A., 2008. Leaf accumulation of trace elements and polycyclic aromatic hydrocarbons (PAHs) in *Quercus ilex* L. *Environmental Pollution* 153, 376–383.
- Dimitrova, I., Yurukova, L., 2005. Bioindication of anthropogenic pollution with *Plantago lanceolata* (Plantaginaceae): Metal accumulation, morphological and stomatal leaf characteristics. *Phytologia Balkanica* 11, 89–96.
- Djingova, R., Wagner, G., Peshev, D., 1995. Heavy-metal distribution in Bulgaria using *Populus nigra* 'Italica' as a biomonitor. *Science of the Total Environment* 172, 151–158.
- Dmchowski, W., Bytnerowicz, A., 2009. Long-term (1992–2004) record of lead, cadmium, and zinc air contamination in Warsaw, Poland: Determination by chemical analysis of moss bags and leaves of Crimean linden. *Environmental Pollution* 157, 3413–3421.
- Doncheva-Boneva, M., 2000. Effect of the vehicle emissions on some environmental components. *Proceedings of International Conference 75 Years of Dendrological Education in Bulgaria, Section Ecology and Environmental Conservation*, 2000, Sofia, pp. 535–543 (in Bulgarian).
- Dongarra, G., Manno, E., Varrica, D., 2009. Possible markers of traffic-related emissions. *Environmental Monitoring and Assessment* 154, 117–125.
- Gonzalez, C.M., Casanovas, S.S., Pignata, M.L., 1996. Biomonitoring of air pollutants from traffic and industries employing *Ramalina ecklonii* (Spreng) Mey and Flot in Cordoba, Argentina. *Environmental Pollution* 91, 269–277.
- Gorelova, S.V., Frontasyeva, M.V., Yurukova, L., Coskun, M., Pantelica, A., Saitanis, C.J., Tomasevic, M., Anicic, M., 2011. Revitalization of urban ecosystems through vascular plants: Preliminary results from the BSEC-PDF project. *Agrochimica* 55, 65–84.
- Gratani, L., Crescente, M.F., Varone, L., 2008. Long-term monitoring of metal pollution by urban trees. *Atmospheric Environment* 42, 8273–8277.
- Howsam, M., Jones, K.C., Ineson, P., 2000. PAHs associated with the leaves of three deciduous tree species. I – Concentrations and profiles. *Environmental Pollution* 108, 413–424.
- ISTAT, 2011. <http://www.en.istat.it>, accessed in September 2013.
- Izrael, Y., Nazarov, I., Pressman, A., Rovinski, F., Ryaboshapko, A., Filippova, L., 1989. *Kislотноye dozhdі*, (2<sup>nd</sup> ed.), Gidrometeoizdat, Leningrad, 269 pp. (In Russian).
- Kabata-Pendias, A., Pendias, H., 2001. *Trace Elements in Soils and Plants*, 3<sup>rd</sup> edition, CRC Press, Boca Raton, London, New York, Washington.
- Klumpp, A., Ansel, W., Klump, G., Breuer, J., Vergne, P., Sanz, M.J., Rasmussen, S., Ro-Poulsen, H., Artola, A.R., Penuelas, J., He, S., Garrec, J.P., Calatayud, V., 2009. Airborne trace element pollution in 11 European cities assessed by exposure of standardised ryegrass cultures. *Atmospheric Environment* 43, 329–339.
- Kularatne, K.I.A., de Freitas, C.R., 2013. Epiphytic lichens as biomonitors of airborne heavy metal pollution. *Environmental and Experimental Botany* 88, 24–32.
- Little, P., Wiffen, R.D., 1977. Emissions and deposition of petrol engine exhaust Pb–I. Deposition of exhaust Pb to plant and soil surface. *Atmospheric Environment* 11, 437–447.
- Loucks, O.L., 1994. Sustainability in urban ecosystems: Beyond an object of Study, in *The Ecological City*, edited by Platt, R.H., Rowntree, R.A., Muick, P.C., University of Massachusetts Press, Amherst.
- Markert, B., 1993. Instrumental analysis of plants, in *Plants as Biomonitors. Indicators for heavy metals in terrestrial environment*, edited by Markert, B., VCH, Weinheim.
- McDonnell, M.J., Hahs, A.K., 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.
- McDonnell, M.J., Pickett, S.T.A., 1990. Ecosystem structure and function along urban rural gradients – an unexploited opportunity for ecology. *Ecology* 71, 1232–1237.
- MOEV, 2010. National Report on the Environment Status, <http://eea.government.bg/bg/output/index.html>, accessed in September 2013.
- Monaci, F., Moni, F., Lanciotti, E., Grechi, D., Bargagli, R., 2000. Biomonitoring of airborne metals in urban environments: New tracers of vehicle emission, in place of lead. *Environmental Pollution* 107, 321–327.
- Neinhuis, C., Barthlott, W., 1997. Characterization and distribution of water-repellent, self-cleaning plant surfaces. *Annals of Botany* 79, 667–677.
- NSI, 2011. <http://www.nsi.bg>, accessed in September 2013.
- Pacyna, E.G., Pacyna, J.M., Fudala, J., Strzelecka-Jastrzab, E., Hlawiczka, S., Panasiuk, D., Nitter, S., Pregger, T., Pfeiffer, H., Friedrich, R., 2007. Current and future emissions of selected heavy metals to the atmosphere from anthropogenic sources in Europe. *Atmospheric Environment* 41, 8557–8566.
- Piczak, K., Lesniewicz, A., Zyrnicki, A., 2003. Metal concentrations in deciduous tree leaves from urban areas in Poland. *Environmental Monitoring and Assessment* 86, 273–287.
- Rautio, P., Huttunen, S., 2003. Total vs. Internal element concentrations in Scots pine needles along a sulphur and metal pollution gradient. *Environmental Pollution* 122, 273–289.
- Salemaa, M., Vanha-Majamaa, I., Derome, J., 2001. Understorey vegetation along a heavy-metal pollution gradient in SW Finland. *Environmental Pollution* 112, 339–350.
- Simonich, S.L., Hites, R.A., 1994. Vegetation-atmosphere partitioning of polycyclic aromatic-hydrocarbons. *Environmental Science & Technology* 28, 939–943.
- Smith, W.H., 1981. *Air pollution and forests*. Springer-Verlag, Inc. New York, NY, 379 pages.
- StatSoft Inc., 2004. STATISTICA (data analysis software system), version 7.
- Steinnes, E., 1993. Some aspects of biomonitoring of air pollutants using mosses as illustrated by a Norwegian survey, in *Plants as Biomonitors*, edited by Markert, B., Weinheim, VCH, pp. 381–394.
- Sucur, K.M., Anicic, M.P., Tomasevic, M.N., Antanasijevic, D.Z., Peric-Grujic, A.A., Ristic, M.D.J., 2010. Urban deciduous tree leaves as biomonitors of trace element (As, V and Cd) atmospheric pollution in Belgrade, Serbia. *Journal of the Serbian Chemical Society* 75, 1453–1461.
- Sukopp, H., 2006. Urban ecology and its application in Europe, in *Urban Ecology: Plants and Plant Communities in Urban Environments*, edited by Sukopp, H., Hejny, S., Kowarik, I., SPB Academic publishing B.V., The Hague, the Netherlands.
- Sukopp, H., 1998. Urban ecology – scientific and practical aspects, in *Urban Ecology*, edited by Breuste, J., Feldmann, H., Uhlmann, O., Springer, Berlin.
- Supuka, J., Feriancova, L., Bihunova, M., 2008. Leaf impact trends of silver birch (*Betula pendula* Roth.) by allochthonous elements at Nitra town urban vegetation. *Thaiszia Journal of Botany* 18, 37–49.
- Takuchev, N., 2011a. Morbidity from non-cancer deceases, associated with air pollution by carbon oxide of Stara Zagora, Bulgaria. *Proceedings of the International Conference "Food science, Engineering and Technologies 2011"*, October 14–15, 2011, University of Food Technologies – Plovdiv, Bulgaria, Vol. LVIII (2): 223–226.
- Takuchev, N., 2011b. Morbidity of the population of Stara Zagora region of non-oncological diseases related to air emissions of nitrogen dioxide from large sources in the region *Proceedings of the International*

- Conference "Food science, Engineering and Technologies 2011", October 14–15, 2011, University of Food Technologies – Plovdiv, Bulgaria, Vol. LVIII (2): 227–232.
- Tarnovska, H., Kuzmanov, N., Kostadinova, P., 2003. Air pollution characteristics in the town of Plovdiv during the period of 2000–2002. *Proceedings of the Sixth Scientific Practical Conference "Ecological problems of Agriculture"*, Agroeco 2003, Agricultural University – Plovdiv, Vol. XLVIII: 345–350 (in Bulgarian).
- Tomasevic, M., Anicic, M., 2010. Trace element content in urban tree leaves and SEM–EDAX characterization of deposited particles. *Facta Universitatis, Series: Physics, Chemistry and Technology* 8, 1–13.
- Tomasevic, M., Anicic, M., Jovanovic, L., Peric–Grujic, A., Ristic, M., 2011. Deciduous tree leaves in trace elements biomonitoring: A contribution to methodology. *Ecological Indicators* 11, 1689–1695.
- Wyttenbach, A., Bajo, S., Tobler, L., 1985. Major and trace–element concentrations in needles of *Picea abies*: Levels, distribution functions, correlations and environmental influences. *Plant and Soil* 85, 313–325.
- Yilmaz, R., Sakcali, S., Yarci, C., Aksoy, A., Ozturk, M., 2006. Use of *Aesculus hippocastanum* L. as a biomonitor of heavy metal pollution. *Pakistan Journal of Botany* 38, 1519–1527.
- Zalud, P., Szakova, J., Sysalova, J., Tlustos, P., 2012. Factors influencing uptake of contaminated particulate matter in leafy vegetables. *Central European Journal of Biology* 7, 519–530.