Journal of the Serbian Chemical Society
JSCS@tmf.bg.ac.rs • www.shd.org.rs/JSCS
J. Serb. Chem. Soc. 82 (6) S272–S276 (2017)
Supplementary material

SUPPLEMENTARY MATERIAL TO

Possibilities of assessing trace metal pollution using *Betula pendula* Roth. leaf and bark – Experience in Serbia

DRAGANA PAVLOVIĆ[1]*, MARIJA PAVLOVIĆ[1], MILICA MARKOVIĆ[1], BRANKO KARADŽIĆ[1], OLGA KOSTIĆ[1], SNEŽANA JARIĆ[1], MIROSLAVA MITROVIĆ[1], IVAN GRŽETIĆ[2] and PAVLE PAVLOVIĆ[1]

[1]*Department of Ecology, Institute for Biological Research "Siniša Stanković", University of Belgrade, Bulevar despota Stefana 142, 11000 Belgrade, Serbia and* [2]*Faculty of Chemistry, University of Belgrade, Studentski trg 12–16, 11000 Belgrade, Serbia*

J. Serb. Chem. Soc. 82 (6) (2017) 723–737

Study area

Pančevo (44°54'00" north, 20°40'00" east, Hs 77 m) is an industrial city located in Vojvodina, the northern province of the Republic of Serbia with over 76 000 inhabitants. It lies at the confluence of the Tamiš and the Danube rivers (Fig. S-1a). The sampling site in Pančevo was the National Garden the main city park located in the southeast part of the city. Major sources of pollution are the oil refinery, nitric fertilizers factory and petrochemical industry in Pančevo.

Smederevo (40°39'00" north, 20°57'00" east, H_s 73 m) is city located in the north-east of the Republic of Serbia. It lies on the right bank of the Danube River (Fig. S-1b). The urban zone covers about 38 km^2 with over 62 000 inhabitants. The sampling site in Smederevo was the city park located in the central zone of the city. The main source of pollution in Smederevo is the steel manufacturing conglomerate "Železara Smederevo" located in the industrial zone 7 km south-east of the city center.

Obrenovac (44°39'00" north, 20°12'00" east, H_s 76 m) is one of the 17 municipalities of Belgrade located at the confluence of rivers Kolubara and Sava with about 25 000 inhabitants (Fig. S-1d). The largest thermoelectric power plant in Serbia, "Nikola Tesla A" (TENT-A), is located in the municipality of Obrenovac and produces the largest amount of fly ash (around 3.6 Mt a year) in Serbia.[1] The sampling site in Obrenovac was the main park located 4 km from the source of pollution (the thermoelectric power plant and fly ash disposal site).

* Corresponding author. E-mail: dragana.pavlovic@ibiss.bg.ac.rs

Belgrade (44°49'14" north, 20°27'44" east, H_s 117 m) is the capital and largest city in Serbia, and has a population of around 1.7 million. It is situated in southeastern Europe on the Balkan Peninsula, at the confluence of the Sava and Danube rivers (Fig. S-1c). Two sites with regard to source of pollution were selected in Belgrade: the "Pioneer" park in the central zone of the city exposed to car exhaust pollution (polluted site), as one of the most polluted areas of the city[2] and the control site – the *Arboretum* of the Faculty of Forestry which is a protected natural area and a valuable archive of domestic and foreign tree species in Belgrade located in an area without a direct source of pollution; it is situated 10 km far from the city center within the zone of mixed *Quercus frainetto* and *Quercus cerris* forest.

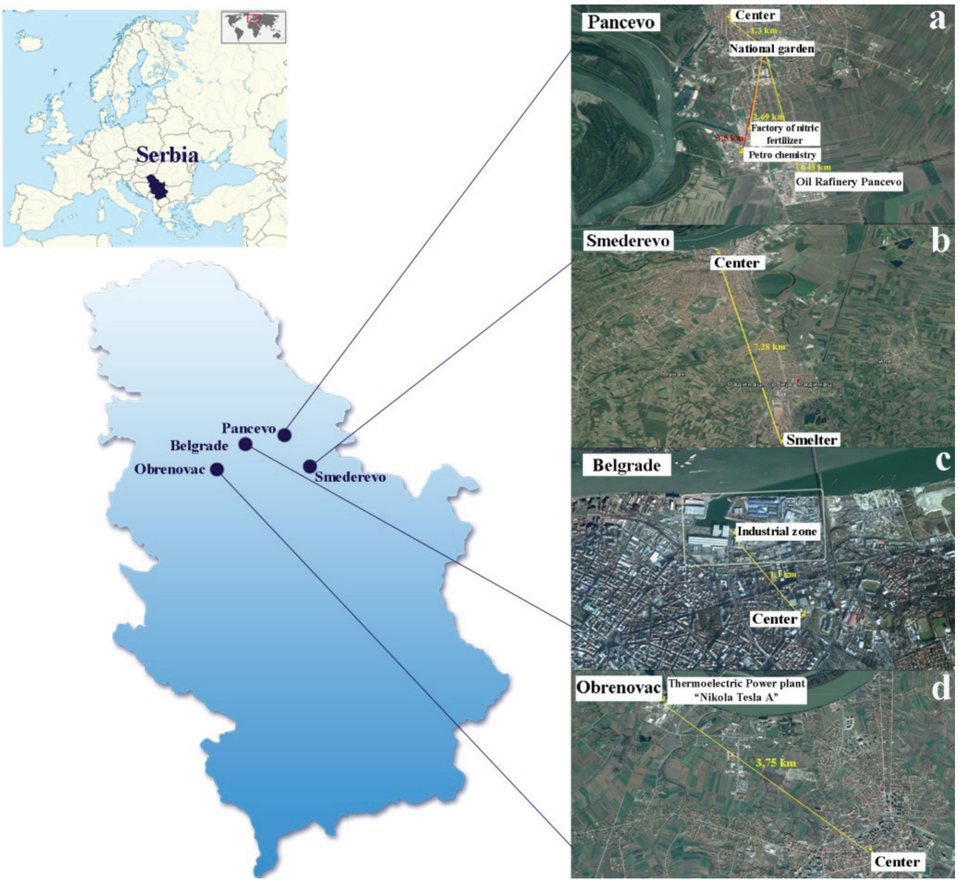

Fig. S-1. Urban sampling sites in Serbia: a) Pančevo, b) Smederevo, c) Belgrade and d) Obrenovac, and distance from source of pollution.

Species description

Silver birch (*Betula pendula* Roth.) belongs to the genus *Betula* L. which belongs to the family Betulaceae in the order Fagales. The genus *Betula* contains some 60 known taxa that are distributed throughout the southern temperate region. Silver birch is medium-sized deciduous tree, with golden-brown bark that later turns to white as a result of a papery tissue developing on the surface and peeling off in flakes. The bark remains smooth until the tree gets quite large, but in older trees, the bark thickens, becoming irregular, dark and rugged. The leaves are roughly triangular with broad, untoothed, wedge-shaped bases, slender pointed tips and coarsely double-toothed, serrated margins and, therefore, have the potential to be efficient particulate scavengers. Birch belongs to the Euro-Siberian floristic geographic element and grows naturally in most of Europe. It is widely planted in urban parks and gardens as a ruderal ornamental tree.[3]

Sampling

The research was carried out at three monthly intervals (June, August, October) during the year 2012. It should be mentioned that it was the warmest and the least rainy year (in relation to the 1981–2010 reference period), which surpassed the number of tropical days and nights since the beginning of their measurement in Serbia.[4]

Determination of heavy metal content and biochemical analyses were conducted on the leaves and bark of *B. pendula* collected from three to five randomly chosen trees at each sampling site. Trees were 20–30-years old. Leaves were taken uniformly from the lower foliage and from different quarters of the tree crowns with stainless-steel scissors using polyethylene gloves, with an initial quantity of about 30 g of each sample. Flakes of the bark layer of about 4–5 mm thickness and maximum dimensions about 1×3 cm^2 were carefully cut with a stainless-steel knife at 1.2–1.5 m above ground level (in all directions around a tree).

Composite topsoil samples at 0-10 cm of depth were taken at 5 points around each tree using a stainless-steel shovel. Foreign objects and stones were removed by hand, and samples were packed in clean plastic bags.

TABLE S-I. Texture, pH and average trace element concentrations (mg kg^{-1} d.w.) in soils sampled from five urban sites. The values are mean, with standard deviation in parentheses (n = 5)

Site	Soil					
	Soil texture	pH	B	Cu	Sr	Zn
Pančevo	Sandy – clay loam	8.44	128.63	11.93	18.32	61.96
		(0.02)	(3.66)	(0.22)	(0.49)	(3.22)
Smederevo	Sandy – clay loam	8.51	137.89	30.66	47.66	119.56
		(0.02)	(2.30)	(0.50)	(1.41)	(1.57)

TABLE S-I. Continued

Site	Soil					
	Soil texture	pH	B	Cu	Sr	Zn
Obrenovac	Clay loam	8.44	155.96	15.11	85.53	45.68
		(0.03)	(3.18)	(0.27)	(3.88)	(4.88)
Belgrade	Sandy – clay loam	8.47	152.03	23.53	50.13	134.53
		(0.02)	(2.09)	(0.11)	(0.87)	(0.67)
Control	Sandy – clay loam	8.30	166.28	12.61	28.16	54.84
		(0.02)	(2.09)	(0.16)	(1.66)	(0.35)

TABLE S-II. Trace element concentrations of soil and plants (mg kg^{-1} d.w.) from the literature[5,6]

Element	Soil		Plant		
	Normal	Average	Deficit	Normal	Toxic
B	1-134	22-40	3-30	10-100	50-200
Cu	1-100	13-23	2-5	5-30	20-100
Sr	5-1000	87-210	–	1-10	>30[6]
Zn	3.5-770	45-60	10-20	27-150	100-400

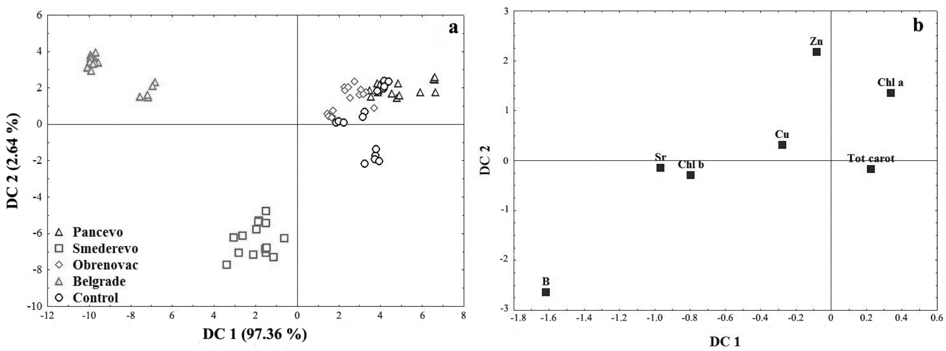

Fig. S-2. Discriminant analysis (DA) for five sampling sites based on metal concentration and chlorophyll content in leaves of *B. pendula.*

REFERENCES

1. O. Kostić, M. Mitrović, M. Knežević, S. Jarić, G. Gajić, L. Djurdjević, P. Pavlović, *Arch. Biol. Sci.* **64** (2012) 145
2. Institute of Public Health of the Republic of Serbia, *Report on Air Pollution Caused by Substances from Exhaust Emissions*, ISO Health Related Particle Sampling Conventions IS 7708, Belgrade ISO. 1994, 2004 (in Serbian)
3. B. Jovanović, *Dendrology*, Faculty of Forestry University of Belgrade, Belgrade, 1985 (in Serbian)
4. J. Smailagić, A. Savović, D. Nešić, M. Milenković, S. Zdravković, *Climatological Analysis of Summer 2012. for Serbia*, *Seasonal Bulletin*, Republic Hydrometeorological Service of Serbia, Belgrade, Republic of Serbia, 2012 (in Serbian)

5. A. Kabata-Pendias, H. Pendias, *Trace elements in soil and plants*, CRC Press LLC, Boca Raton, FL, 2001
6. H. T. Shacklette, J. A. Erdman, T. F. Harms, in: *Toxicity of Heavy Metals in the Environment*, Part I, F. W. Oehme, Ed., Marcel Dekker, New York, 1978, p. 25.