ИНСТИТУТ ЗА ФИЗИКУ

ПРИМЉЕНО: 22-12-2016			
Рад.јед.	број	Арх.шифра	Прилог
0801	2218/1		

ЗАХТЕВ

Молим Научно веће Института за физику у Београду да покрене мој избор у звање **истраживач сарадник**, а на основу приложене документације:

1. Мишљење руководиоца пројекта на коме је подносилац захтева запослен и предлог чланова комисије за избор у звање.
2. Стручна биографија;
3. Списак и копије објављених радова и саопштења до момента подношења захтева;
4. Подврда да је кандидат уписан на докторским студијама
5. Диплома о научном степену дипломирани хемичар и мастер-хемичар (копије);
6. Кратког прегледа научних активности кандидата

У Београду,
22.12.2016. год.

Подносилац захтева

Тијана Милићевић

Тијана Милићевић

Научно веће
Институт за физику у Београд

Датум:
Београд, 22. децембар 2016. год.

Предмет:
Мишљење руководиоца пројекта за избор Тијане Милићевић у звање истраживач сарадник

Тијана Милићевић је запослена у Лабораторији за физику животне средине Института за физику у Београду од 1. јуна 2015. године. Колегиница је ангажована на пројекту интегралних интердисциплинарних истраживања Министарства просвете, науке и технолошког развоја ИИИ43007 под називом „Истраживање климатских промена и њиховог утицаја на животну средину – праћење утицаја, адаптација и ублажавање”, потпројекат 3 „Интегрална истраживања квалитета ваздуха у урбаној средини”.

Докторске студије на Хемијском факултету Универзитета у Београду је уписала 2014. године.

Резултати досадашњег истраживачког рада колегинице Милићевић су публиковани у два водећа међународна часописа, као и неколико радова штампаних у зборницима међународних и домаћих конференција.

До сада постигнути резултати колегинице Милићевић показују да поседује квалитете неопходне за бављење научно-истраживачким радом и израду докторске дисертације. С обзиром да колегиница испуњава све критеријуме прописане Правилником о поступку и начину вредновања, и квантитативном исказивању научних резултата Министарства просвете, науке и технолошког развоја сагласан сам са покретањем поступка за избор Тијане Милићевић у звање истраживач сарадник.

За састав Комисије за избор Тијане Милићевић у звање истраживач сарадник предлажем колеге:

1. др Мира Аничић Урошевић, виши научни сарадник, Институт за физику у Београду;
2. др Гордана Вуковић, научни сарадник, Институт за физику у Београду
3. др Александар Поповић, редовни професор, Хемијски факултет, Универзитет у Београду

Руководилац потпројекта

Зоран Мијић

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РАДНО ИСКУСТВО:

1. јун 2015. до сада: истраживач приправник у Лабораторији за физику животне средине Института за физику у Београду

ОБРАЗОВАЊЕ

2014. до сада: Докторске студије: Хемијски факултет Универзитета у Београду; Хемија (300 ЕСПБ)

2013/2014: Специјалистичке студије: Правни факултет Универзитета у Београду Студије за иновацију (специјализацију) знања из области еколошко право Специјалистичка теза - *Правни аспекти заштите шума и њихов утицај на промену климе у Србији*

2012/2013: Мастер студије: Хемијски факултет Универзитета у Београду Мастер хемичар (60 ЕСПБ)

Мастер теза - *Поређење различитих типова екстракционих средстава за изоловање елемената који су лакодоступни биљци*

2008–2012: Основне студије: Хемијски факултет Универзитета у Београду; Дипломирани хемичар, Хемија животне средине (240 ЕСПБ)

Дипломска теза - *Биоиндикатори загађења животне средине са простора Србије - могући лабораторијски огледи*

НАУЧНИ ПРОЈЕКТИ

2015. до сада: Национални пројекат Министарства просвете, науке и технолошког развоја ИИИ43007 - *Истраживање климатских промена и њихов утицај на животну средину - праћење утицаја, адаптација и ублажавање*

2014: *Биомониторинг тешких метала у ваздуху дуж главних саобраћајница града Београда* - подржан је од компаније а.д. НИС и Града Београда

ТЕХНИЧКЕ ВЕШТИНЕ

MS Office, Project Manager, SPSS, Statistica, Internet

ЈЕЗИЦИ

Енглески језик - активно

ОСТАЛО

Добитник ERASMUS + гранта; Члан Српског хемијског друштва; Члан European Geosciences Union; Радионица: JEOL SEM and TEM microscopes - радионица у организацији *SCAN d.o.o.*, Крањ, Словениа и *JEOL S.A.S. (Europe)*

Списак научних радова и саопштења

Научни радови:

1. **T. Milićević**, D. Relić, S. Škrivanj, Ž. Tešić, A. Popović, *Assessment of major and trace element bioavailability in vineyard soil applying different single extraction procedures and pseudo-total digestion*, Chemosphere, 2016, *Accepted for publication*; DOI: http://dx.doi.org/10.1016/j.chemosphere.2016.12.090.

   M21, IF: 3.698

2. G. Vuković, M. Aničić Urošević, S. Škrivanj, **T. Milićević**, D. Dimitrijević, M. Tomašević, A. Popović, *Moss bag biomonitoring of airborne toxic element decrease on a small scale: a street study in Belgrade, Serbia*, Science of the Total Environment, 2015, 542, 394 – 403.

   M21a, IF: 4.099

Међународне конференције:

1. **T. Milićević**, D. Relić, G. Vuković, M. Perišić, D. Majstorović, M. Aničić Urošević and A. Popović, *Survey of potentially toxic element pollution of the vineyard soil* (poster presentation), 13th International Conference on Fundamental and Applied Aspects of Physical Chemistry, Physical Chemistry 2016, September 26–30, 2016, Belgrade, Serbia, Proceedings, pp. 739 – 742

   M33

2. **T. Milićević**, D. Relić, G. Vuković, S. Škrivanj, A. Popović, M. Aničić Urošević: *Grapevine accumulation of potentially toxic elements from vineyard soil* (poster presentation), 18th International Conference on Heavy Metals in the Environment, 12-15 September 2016, Ghent, Belgium, Book of abstracts, pp. 415.

   M34

3. M. Aničić Urošević, G. Vuković, **T. Milićević**, K. Vergel, M. Frontasyeva, M. Tomašević and A. Popović, *Moss bag biomonitoring of airborne toxic element decrease on a small scale: crossroad and two - and one - lane street study,* 29th ICP Vegetation Task Force Meeting, February 29 – March 4, 2016, Dubna, Russian Federation, Book of abstract, pp. 40.
   http://icpvegetation.ceh.ac.uk/publications/documents/Bookofabstracts_000.pdf

   M34

4. G. Vuković, M. Aničić Urošević, S. Škrivanj, **T. Milićević**, D. Dimitrijević, M. Tomašević and A. Popović, *Upgrade of micro-scale siting of airborne toxic elements by*

moss bag technique: crossroad, two- and one-lane street study (poster presentation), The 5th International WeBIOPATR Workshop & Conference, October 14–16, 2015, Belgrade, Serbia, Proceedings, pp. 48 – 53.
https://www.vin.bg.ac.rs/webiopatr/#Workshop/Abstracts-and-papers

M33

5. **T. Milićević**, D. Relić, A. Popović, *Correlation between macro- and microelements isolated from the vineyard soil by different extractant methods* (poster presentation), 7th Symposium Chemistry and Environmental Protection EnviroChem 2015, 12 – 15 June, Palić, Serbia, Book of Abstracts, pp. 229 – 230.
http://www.envirochem.rs/res/EC2015_Book_of_Abstracts.pdf

M64

6. **T. Milicević**, D. Relic, A. Popović, *Assesment of CH_3COOH, Na_2EDTA, $CaCl_2$, NH_4NO_3 and distilled water extraction procedures and microwave digestion for leaching of macro- and microelements from vineyard soil* (poster presentation), 7th Symposium Chemistry and Environmental Protection EnviroChem 2015, 12 – 15 June, Palic, Serbia, Book of Abstracts, pp. 252 – 253.
http://www.envirochem.rs/res/EC2015_Book_of_Abstracts.pdf

M64

7. G. Vuković, M. Aničić, **T. Milicević**, M. Tomašević, S. Škrivanj, A. Popović, *Moss-bag biomonitoring of small-scale decline in toxic element content: crossroad, two- and one-lane street study* (poster presentation), 7th Symposium Chemistry and Environmental Protection EnviroChem 2015, 12 – 15 June, Palić, Serbia, Book of Abstracts, pp. 192 – 193. http://www.envirochem.rs/res/EC2015_Book_of_Abstracts.pdf

M64

8. **T. Milićević**, D. Relić, A. Popović, *Determination of bioavailable macro- and microelements from agricultural soil using different extractants* (poster presentation), Book of Abstracts, European Geoscience Union General Assembly, 12 – 17 April, 2015, Vienna, Geophysical Research Abstracts, Vol. 17, EGU2015 – 1138. Austria
http://meetingorganizer.copernicus.org/EGU2015/EGU2015 - 1138.pdf

M34

Chemosphere xxx (2016) xxx-xxx

Contents lists available at ScienceDirect

Chemosphere

journal homepage: www.elsevier.com

Assessment of major and trace element bioavailability in vineyard soil applying different single extraction procedures and pseudo-total digestion

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ARTICLE INFO

Article history:
Received 13 July 2016
Received in revised form 9 December 2016
Accepted 19 December 2016
Available online xxx

Handling Editor: Martine Leermakers

Keywords:
Vineyard soil
Grapevine
Single extraction
Pseudo-total digestion
Ecological risk assessment

ABSTRACT

A different single extraction procedures (CH_3COOH, Na_2EDTA, $CaCl_2$, NH_4NO_3, deionized water), and pseudo-total digestion (aqua regia) were applied to determine major (Al, Fe, K, Mn, Na, P, S, and Si) and trace (Cd, Co, Cr, Cu, Mo, Ni, Pb, V, and Zn) element bioavailability in a topsoil from the experimental vineyard (Radmilovac, Belgrade, Serbia). For the first time, the extraction with deionized water during 16 h was tested as an alternative method for isolating bioavailable major and trace elements from the soil. Concentrations of the elements were determined by inductively coupled plasma – optical emission spectroscopy (ICP-OES). The extraction of Cu and S from the soil by deionized water during 16 h extracting, NH_4NO_3, and $CaCl_2$ indicated that these elements could originate from the anthropogenic sources, such as fungicide. In addition according to the soil – plant experiment, performed as a preliminary experiment for future studies in vineyards, deionized water was recommended for isolation of bioavailable elements from grape seed and grape pulp; CH_3COOH, Na_2EDTA, $CaCl_2$ and NH_4NO_3 for grape skin, while for assessment of leaf bioavailable elements from soil fraction, aqua regia was recommended. In addition, identification of similarities between the plant parts and the plant species were performed. Applying environmental risk assessment formulas, the most polluted vineyard parcel in the vineyard region "Radmilovac" was determined. The leaves of some grapevine species showed the high ability for accumulation some of the potentially toxic trace elements from the soil.

1. Introduction

Soil could be contaminated by pollutants originating from different natural or anthropogenic sources, such as toxic elements released from wastewater, sewage sludge, chemical fertilizers, pesticides, solid waste disposal, vehicular exhaust, etc. (Khan et al., 2008; Islam et al., 2015). In the vineyard soils, the major impact of the soil pollution could be caused by the potentially toxic elements (especially Cu and Zn) from the chemical fertilizers and pesticides (Kabata – Pendias and Mukherjee, 2007; Tiecher et al., 2016). The presence in increase concentration of trace elements in soils can cause a potential risk to human health because of their subsequent involvement into the food chain by plant uptake (Radha et al., 1997; Alonso Castillo et al., 2011; Sakan et al., 2011; Islam et al., 2015). Translocation of major and trace elements from soil to plant has a major impact on the plant growing and quality of its products. Human activities can increase the content of pollutants, up to phytotoxic. Trace and other elements in soils may affect human health through the inhalation of dust, ingestion of soil, or by dermal contact (Morel, 1997; Kabata - Pendias and Kabata, 2001; Hazrat et al., 2013; Sylvain et al., 2016).

Research on element bioavailability of contaminated agricultural soil, monitoring and management attracted attention at international level and it has been ongoing for more than 20 years (Ruby et al., 1996; Pelfrêne et al., 2012). With an aim of determination of element bioavailability to plant and metal availability in the environment from the 1980s to now, there are many publications based on developing and applying procedures of single and sequential extraction procedures (Quevauviller, 2002). Nowadays many procedures are included in national and international regulations or they have been considered in the framework of normalization bodies as CEN or ISO (Quevauviller et al., 1996). In German protocol, 0.1 mol L^{-1} NH_4NO_3 is the standard used to estimate mobile and potentially hazardous forms of trace elements (DIN 19730) (Hall et al., 1998). In Dutch legislation, 0.01 mol L^{-1} $CaCl_2$ is the standard used for the assessment of nutrients and heavy metals in soils (Pueyo et al., 2004).

Many single extraction procedures are recommended for isolation of major and trace elements from agricultural soils. For example, aqua regia and Na_2EDTA procedures were useful for determination environmental risk assessment while 0.05 mol L^{-1} Na_2EDTA procedure was usually useful for agricultural and environmental studies. The weak extractants such as 0.01 mol L^{-1} $CaCl_2$ and 0.1 mol L^{-1} NH_4NO_3 or deionized H_2O were usually applied for soil – plant uptake studies. In addition, aqua regia and 0.01 mol L^{-1} $CaCl_2$ were used to determine soil fertility (McGrath, 1996; Quevauviller et al., 1996; Pueyo et al., 2004). These single extractions are a simple and cost – effective way to extract elements, and they give detailed information on the assessment of the labile elements in soils (Meers et al., 2007; Rao et al., 2008; Santos et al., 2010).

Because of its stringent safety precautions, usually used HF procedure is not recommended for routine analyses (Sastre et al., 2002; Sandroni et al., 2003). In addition, the hardly available soil fraction of the elements is unavailable to plants. Moreover, HF could react with Ca, and it could form Ca—F complexes which could trap trace

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http://dx.doi.org/10.1016/j.chemosphere.2016.12.090
0045-6535/

elements (Marina et al., 2008). An alternative to this procedure is pseudo-total digestion by aqua regia (US EPA, 1996). The European Community Bureau of Reference has certified several soil and sediment samples based on it (Quevauviller et al., 1993; Chen and Ma, 2001; Relić et al., 2011).

According to the available literature, there is no a comprehensive study comparing performances of these five above – mentioned extraction procedures. For the first time, the extraction using deionized water during 16 h was performing as an alternative single extraction procedure. In this study, five single extractions (CH_3COOH, Na_2EDTA, $CaCl_2$, NH_4NO_3, deionized water) and pseudo-total digestion procedures were used for extraction of major and trace elements from the vineyard soil. In addition, for the first time, it was tested which extraction procedure should be the best option for determining bioavailability elements soil fractions which have an influence on different grapevine parts. The aims of the study were to assess:

1) a suitability of using deionized water for 16 h as an alternative procedure to the other single extraction procedures;
2) which single extraction procedure is the best for extraction of major and trace elements from the vineyard soil;
3) whether concentrations of major and trace elements in the soil are in relationship with their content in the grapevine (leaf, seed, pulp, and skin); and
4) environmental risk assessment of major and trace elements in the agricultural area such as a vineyard.

2. Materials and methods

2.1. Study area

In this study, soil samples were collected from the vineyard "Radmilovac", Serbia during the grapevine season of 2013. "Radmilovac" is a suburban settlement of Belgrade, (44° 45′ 24″ N; 20° 34′ 54″ E), the capital of Serbia. The experimental agricultural area of "Radmilovac" is the original core of the neighborhood and it covers an area of 88 ha. The vineyard parcels are located between the Institute of nuclear research "Vinča", the hazardous waste landfills and the main road. The institute landfill is oriented towards the vineyard parcels of agricultural area "Radmilovac" (Fig. 1).

2.2. Sampling

Surface soil layer (0–30 cm), was collected from nine different vineyard parcels, marked as tables T1, T2, T3, T4, T5, T6, T10, P and K. In each parcel, the soil samples were taken as the composite samples of 10 subsamples sampled along the diagonal. The samples T2 and T4 were collected from the central part of vineyard area. The P table is situated close to the local stream. Tables T1, T5, and K are without grapevine plants. Table K was used as a control sample and to determination of the initial values of the measured elements (Fig. 1).

In the vineyard, seven different grapevines (*Vitis vinifera* L.) species are planted. They were located by the order: T2 – *Riesling Rain* and *Burgundac*, T3 – *Cabernet Sauvignon* and *Riesling Italian*, T4 – *Prokupac* and *Cabernet Sauvignon*, T6 – *Cabernet Franc*, T10 – *Cabernet Franc* and *Merlot*, P – *Cabernet Franc*. For need of the pilot study, the grape leaf and grape were sampled from each table (parcel).

2.3. Chemical analyses

The experiment was designed to determine behavior of the content of 17 major and trace elements in the vineyard soil, grapevine (seed, pulp, and skin) and grape leaves samples. All chemicals used in the experiment were of analytical grade and produced by Sigma – Aldrich.

2.3.1. Soil sample preparation

Soil samples were stored at the room temperature in the laboratory. Each soil sample was air-dried, sieved through 2 mm stainless sieve and ground to a fine powder with a pestle in an agate mortar with a pestle. The samples were packed in polyethylene bags and kept under stable laboratory conditions until the chemical analysis.

The hygroscopic moisture of each soil sample was determined at 105 °C until the dry weight. In addition, the pH of soil samples was determined in a mixture (1:5) of soil-distilled H_2O and soil – 1 M KCl. Acidity (pH) of the soil samples was determined using a glass electrode.

Different extraction solutions were used (Table 1) for the assessment of element bioavailability from the soil: $0.11\ \text{mol L}^{-1}$

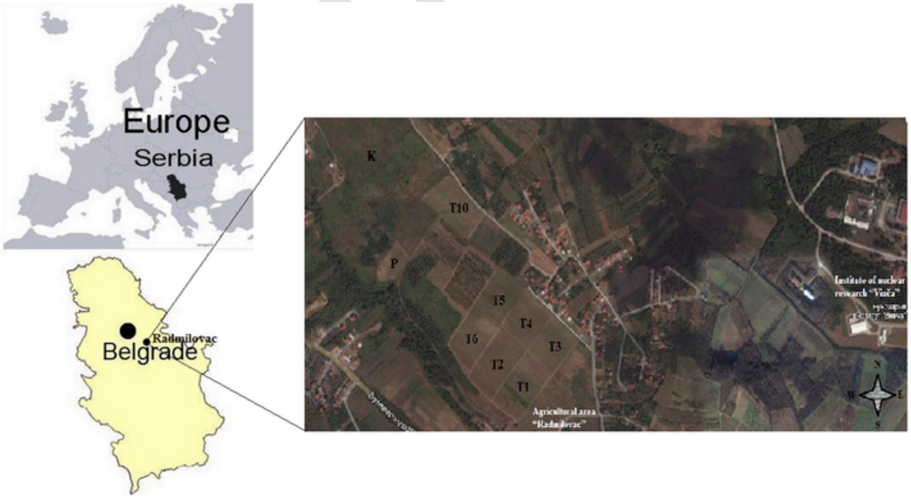

Fig. 1. Location of vineyard "Radmilovac".

Table 1
Procedures for extraction of major and trace element from the soil samples.

Extractant	Procedure	References
0.11 mol L^{-1} CH_3COOH	1 g of each soil sample was measured and 40 mL of extractant was added. The extraction was performed for 16 h on a rotary shaker.	Pueyo et al., 2004; Quevauviller, 1998.
Distilled water	2 g of each soil sample was measured and 20 mL of distilled water was added. The extraction was performed for 16 h on a rotary shaker.	a
Distilled water	2 g of each soil sample was measured and 20 mL of distilled water was added. The extraction was performed for 2 h on a rotary shaker.	Pueyo et al., 2004; Quevauviller, 1998.
0.05 mol L^{-1} Na_2EDTA	2 g of each soil sample was measured and 20 mL of extractant was added. The extraction was performed for 1 h on a rotary shaker.	Pueyo et al., 2004; Quevauviller, 1998.
0.01 mol L^{-1} $CaCl_2$	2 g of each soil sample was measured and 20 mL of extractant was added. The extraction was performed for 3 h on a rotary shaker.	Pueyo et al., 2004; Quevauviller, 1998.
0.1 mol $L^{-1}NH_4NO_3$	4 g of each soil sample was measured and 10 mL of extractant was added. The extraction was performed for 2 h on a rotary shaker.	Quevauviller, 1998.
Aqua regia (HNO_3: HCl)	0.5 g of each soil sample was digested using 9 mL HCl and 3 mL HNO_3	US EPA 3050b Method

a The major and trace element extraction from soil by distilled water during 16 h was performed for comparison with soil extraction by distilled water during 2 h and other one step extraction procedures.

CH_3COOH, 0.05 mol L^{-1} Na_2EDTA, 0.01 mol L^{-1} $CaCl_2$, 0.1 mol L^{-1} NH_4NO_3 and deionized water extraction during 2 h and during 16 h as an alternative extraction procedure.

After extraction, all samples were centrifuged at 3000 rpm for 10 min. The supernatants were separated from precipitate through a filter. The pseudo-total digestions were performed by aqua regia solution in a microwave oven (ETHOS 1, Advanced Microwave Digestion System, Milestone, Italy) in sealed PTFE vessels using 9 mL of HCl and 3 mL of HNO_3 for 0.5 g per each soil sample (US EPA 3050b Method).

2.3.2. Grapevine and grape leave samples preparation

The plant – soil experiment was performed as a previous experiment (pilot study) to future studies. The leaf samples were dried in an oven at 45 °C during 24 h, powdered in an agate mortar. Fresh grape samples were separated into seed, pulp, and skin; all portioned samples of grape were blended. Approximately 0.5 g of each leaf samples (6 samples) and 1 g of each part of grape sample (6 seed samples, 2 pulp samples and 7 skin samples) were digested for 45 min in a microwave digester (ETHOS 1, Advanced Microwave Digestion System, Milestone, Italy) at 200 °C with 1 mL of 30% H_2O_2 (Sigma-Aldrich, puriss. p.a.) and 7 mL of 65% HNO_3 (Sigma-Aldrich, puriss. p.a., distilled by the apparatus for acid distillation – BERGHOF, Products + Instruments GmbH, Germany). The digested leaf samples were diluted with double-distilled water to a total volume of 50 mL and digested grape parts samples were diluted with double-distilled water to a total volume of 25 mL.

2.3.3. Sample analysis

Concentrations of 17 bioavailable major (Al, Fe, K, Mn, Na, P, S and Si), and trace (Cd, Co, Cr, Cu, Mo, Ni, Pb, V and Zn) elements were determined in all samples using inductively coupled plasma optical emission spectroscopy, ICP-OES (Thermo Scientific iCAP 6500 Duo, Thermo Scientific, UK). For calibration, a Multi – Element Plasma Standard Solution 4, Specpure (Alfa Aesar GmbH & Co KG, Germany) was used. To determine the quality of the element measurements, the analytical blanks and certified reference material were analyzed once every 10 samples. Certified reference material BCR 483 (Sewage sludge amended soil) and ERM – CC135a (Contaminated Brickworks Soil) was used to validate the single extractions ($CaCl_2$, NH_4NO_3, and Na_2EDTA) and pseudo-total digestion protocols, respectively. The recovery of elements using BCR 483 ranged from 72% to 130%. The recoveries for Cr and Cu extracted with $CaCl_2$ were 139%, and 140%, respectively. Using ERM – CC135a the recovery ranged from 82% to 106%, with exception of Pb (76%), and V (51%). For validation, the plant sample protocols, the moss *Pleurozium schreberi*, *M2,* and *M3* were used as the certified reference materials (Steinnes et al., 1997). The recovery of elements from the reference material *M2* ranged from 71% to 106%. However, recovery of Cr was 60%. For the reference material *M3* recoveries are ranged from 73% to 117%. Recovery of Cr was 42%.

2.4. Data processing

For data analysis, SPSS software version 21 for Windows and Statistic8 (StatSoft Inc., Tulsa, OK, USA) were used. The normality ($p < 0.05$) of the data was tested by the Kolmogorov – Smirnov test. Chemometrics of the obtained results were based on correlation, principal component and cluster analyses. Principal component analysis (PCA) and cluster analysis (CA) were used to identify a correlation between the elements concentrations in the soil samples and used extractants. Varimax normalized was used as the rotation method in the analysis following standardization of the data (Kaiser, 1958).

For the pilot part of the study, PCA, and CA analyses were used aiming to assume a connection between the element content in different grapevine parts, and also of major and trace elements extracted from soil by various extractants. For identification, the similarities among plant parts, plant species, and soil samples, the Q mode of PCA was applied.

2.4.1. Ecological risk assessment

Contamination factor (CF), pollution load index (PLI), geo-accumulation index (I_{geo}), enrichment factor (EF) and bioaccumulation factor (BF) among plant parts, species and soil samples were calculated for each sample to investigate the level of pollution of the vineyard soil by from the potentially pollution sources around the study area "Radmilovac". To investigate the level of contamination of soils by the trace elements in the study area, contamination factor was calculated according to the following equation.

$$CF = C_n/B_n$$

where C_n is a pollutant's (element's) concentration and B_n is the initial concentration (K parcel) of that metal in the soil.

To assess quality of the soil, an integrated approach of PLI of trace elements was calculated. The PLI is defined as the nth root of the multiplications of CF of trace elements.

$$PLI = (CF1 \text{ x } CF2 \text{ x } CF3 \ldots \text{ x } CF_n)^{1/n}$$

where n is the number of pollutant (element) assessed and PLI is the single factor pollution index of each element (Likuku et al., 2013).

A geo-accumulation index (I_{geo}) was applied to determine and define metal contamination in soils by comparing measured metal concentrations with local background levels. I_{geo} was calculated for all determined major and trace elements in soil samples.

$$I_{geo} = \log_2 [(C_n/1.5 \times B_n)]$$

where, C_n is the measured concentration of the metal n, B_n is the local background concentration of metal n. The constant factor 1.5 is the background value correlation factor (Likuku et al., 2013). I_{geo} was classified into seven grades: $I_{geo} \leq 0$ (grade 0), unpolluted; $0 < I_{geo} \leq 1$ (grade 1), slightly polluted; $1 < I_{geo} \leq 2$ (grade 2), moderately polluted; $2 < I_{geo} \leq 3$ (grade 3), moderately severely polluted; $3 < I_{geo} \leq 4$ (grade 4), severely polluted; $4 < I_{geo} \leq 5$ (grade 5), severely extremely polluted; $I_{geo} > 5$ (grade 6), extremely polluted (; Yaquin et al., 2008).

Enrichment factor (EF) was applied to distinguish among elements originating from human activities and those from natural ancestry, and to assess the degree of anthropogenic influence (Lu et al., 2014; Chen et al., 2015).

$$EF = (Cn/C_{ref})_{sample}/(Cn/C_{ref})_{background}$$

where Cn is the concentration of a metal element in soil (mg/kg) and C_{ref} is the concentration of reference element (mg/kg). Usually used elements for background are Al, Fe (Sakan et al., 2014) and Mn (Tasdemir and Kural, 2005; Chen et al., 2015). According to EF soils could be classified: deficiency to minimal enrichment ($EF < 2$), moderate enrichment ($2 \leq EF < 5$), significant enrichment ($5 \leq EF < 20$), very high enrichment ($20 \leq EF < 40$) or extremely high enrichment ($EF \geq 40$). In addition, the EF values among 0.05 and 1.5 indicate that the metal originates from the crustal materials or some natural source, and for EF values higher than 1.5 originate from anthropogenic sources (Zhang and Liu, 2000; Chen et al., 2015).

The ratios of concentrations between the plant parts and soil were estimated applying calculation of bioaccumulation factor (BF):

$$BF = C_p/C_s$$

where Cp is the major or trace element concentration in different grapevine parts and Cs is the concentration of same element in the soil sample collected from the same parcel as the grapevine samples. If the values $BF > 1$ then the plants could be accumulators; when $BF = 1$ there are no influences of the soil and if the $BF < 1$ means that the plant can be an excluder (Radulescu et al., 2013).

3. Results and discussion

The acidity of the soil samples was determined in range pH H_2O (4.84–8.05) and pH KCl (3.85–7.07). Based on this soil acidity result, it could be concluded that the vineyard's soil samples varied from weak acidic to weak alkaline. The obtained pH values influence the bioavailability of the elements from the soil and play an important role in their toxicity for plants (Kabata – Pendias and Mukherjee, 2007; Bravo et al., 2015).

3.1. Single extraction procedures and pseudo-total digestion for assessment of element bioavailable fractions from vineyard's soil

Aqua regia has been shown as the most aggressive agent for extraction of the major and trace elements from the soil samples (Fig. S1). In addition, CH_3COOH and Na_2EDTA were also proven to be aggressive extractants, which could be applied for isolation of higher concentrations of the major and trace elements from the soil rather than deionized water, $CaCl_2$ and NH_4NO_3 (Table 2, Fig. S1). The acidity of CH_3COOH could enhance destruction of carbonates associated with trace elements, and thus, the extraction of bioavailable element fraction.

For determination of the total extractable S, pseudo-total digestion with aqua regia was not proven as the best isolating method, probably due to conditions of high pressure and acidity, which may influence the loss of organic S. The S concentrations (12–18 $\mu g\ g^{-1}$) extracted by $CaCl_2$ in this study are comparable with data obtained in the other studies (e.g., Hu et al., 2005). As not aggressive extractant $CaCl_2$ isolated 42% S comparing to concentration obtained by pseudo-total digestion, which proves this weak salt to be suitable for extracting S.

Based on the results of this study (Fig. S1), the similarity was observed of the major elements by $CaCl_2$, and deionized H_2O (during 16 h and 2 h), in comparison with the concentrations of these elements isolated by the pseudo-total digestion procedure. In addition, there was a similarity in capacity to extract the trace element by deionized water extraction during 16 h and 2 h. However, there is no unique extractant appropriate for the extraction the most available fraction for all studied elements from the soil (Fig. S1).

The highest concentrations of all major and trace elements in soil (except S) were isolated by the pseudo-total digestion with aqua regia. The concentrations of Cu (93–118 $\mu g\ g^{-1}$) and Cd (3.6–4.3 $\mu g\ g^{-1}$) from this study were comparable with data obtained for similarly treated soils by fertilizers and pesticides (Pueyo et al., 2004; Meers et al., 2007; Kelepertzis et al., 2015). The pseudo-total concentrations of Cu isolated from the soil samples were higher than maximum prescribed values (100 $\mu g\ g^{-1}$) by the national and international regulative (National Regulation, Republic of Serbia 88/2010; EU Council Directive 86/278/EEC) (Table 2). In addition, the concentration of Pb at the parcel T6 was higher than the concentrations prescribed in the national and international regulative. Higher concentrations of Cu could originate from Cu – based fungicides used for the treating grapevine whereas a high concentration of Pb could be explained by the proximity of the main road (Fig. 1).

Among the single extractants, the lowest element concentrations were obtained applying NH_4NO_3, and $CaCl_2$, in comparison to the pseudo-total digestion (Table 2, Fig. S1). The concentrations of Ni (0.02–0.27 $\mu g\ g^{-1}$) and Pb (0.7–9.4 $ng\ g^{-1}$) isolated by NH_4NO_3 are comparable with the results reported by Pinto et al. (2015) (Table 2). Higher concentrations of K and Mn were isolated by NH_4NO_3 than by $CaCl_2$ as also reported in some other studies (Pueyo et al., 2004). These two unbuffered salt solutions simulate natural soil solutions, and therefore they are included in regulations and are useful for evaluation the ecological relevance of elements (Kabata – Pendias and Kabata, 2001). The concentrations of Ni (0.02–0.27 $\mu g\ g^{-1}$) extracted by $CaCl_2$ are comparable with the corresponding results reported by Bakircioglu et al. (2011). Thus, NH_4NO_3 and $CaCl_2$, as weak diluted salt solutions, could only be used as extractants for the elements presented in the exchangeable phase as well as in water – soluble phase (Pinto et al., 2015). There were also some other values with similar research results. There were similarities between the concentrations of Cu (0.9–1.8 $\mu g\ g^{-1}$, 4.1–27 $\mu g\ g^{-1}$) extracted with deionized water during 2 h mixing (Table 2) reported in a study by Bakircioglu et al. (2011). In addition, the concentrations of Zn (0.2–0.9 $\mu g\ g^{-1}$) extracted with deionized water during 2 h from soil samples (Table 2) were comparable with results in the study reported by Niesiobedzka (2016).

Table 2
Descriptive statistics (mean, standard deviation, minimum and maximum) of the element concentrations (μgg^{-1}, *ngg^{-1}) extracted from soil samples.

		Al	Fe	K	Mn	Na	P	S	Si	Cd	Co	Cr	Cu	Mo	Ni	Pb	V	Zn
Single extraction																		
CH_3COOH	M	45	16	66	24	41	77	36	220	0.06	0.05	0.08	1.9	0.1	0.8	51.2*	0.02	1.0
	SD	11	14	15	5	7.2	70	19	28	0.02	0.04	0.02	1.2	0.2	0.3	0.6*	0.02	1.2
	MIN	27	5.6	49	20	30	14	14	170	0.04	0.01	0.06	0.4	0.0	0.5	49.9*	0.01	0.0
	MAX	59	49	88	35	50	200	74	260	0.08	0.11	0.11	4.3	0.4	1.2	51.6*	0.05	3.7
16 h H_2O	M	25	22	7.7	0.3	13	3.8	27	58	3.9*	4.0*	0.03	1.1	16*	0.1	0.1	0.06	0.6
	SD	5.8	5.3	2	0.2	4.5	1.8	8.4	12	1.0*	2.4*	0.02	0.9	7.9*	0.2	0.2	0.02	0.5
	MIN	15	14	5.9	0.2	9.2	2.1	20	37	2.3*	0.9*	0.01	0.3	3.5*	0.0	0.0	0.04	0.0
	MAX	33	31	11	0.6	22	7.2	46	73	5.6*	7.7*	0.04	2.6	26*	0.4	0.4	0.07	1.6
2 h H_2O	M	35	20	7.14	0.06	15	5.1	10	88	0.253*	6.7*	0.04	1.3	17*	0.3	0.04	0.10	0.4
	SD	20	12	2.6	0.03	4	1.9	5.5	46	0.003*	4.5*	0.03	0.3	6.8*	0.1	0.03	0.06	0.3
	MIN	16	6.2	3.5	0.02	11	3.2	2.5	17	0.249*	1.7*	0.02	0.9	3.6*	0.2	0.02	0.06	0.2
	MAX	79	44	11	0.1	23	8.9	19	140	0.256*	15*	0.11	1.8	29*	0.6	0.09	0.22	0.9
Na_2EDTA	M	16	100	89	78	a	20	4.3	167	0.08	0.6	0.04	16	19*	1.3	2.6	0.1	2.0
	SD	12	54	25	55	a	14	1.7	128	0.04	0.6	0.04	8.0	9.9*	0.8	1.1	0.2	1.5
	MIN	2.3	39	52	14	a	1.9	1.4	44	0.03	0.0	0.00	4.1	2.6*	0.3	1.3	0.0	0.8
	MAX	42	190	132	190	a	36	7.4	463	0.16	1.8	0.09	27	37*	3.0	4.4	0.4	5.3
$CaCl_2$	M	1.1	1.1	13	0.02	7.2	0.4	15	21	0.7*	1.4*	4.4*	0.15	0.02	0.13	6.8*	3.4*	0.0
	SD	1.6	1	5.2	0.05	1.2	0.3	2.1	22	0.5*	0.8*	3.6*	0.03	0.02	0.07	8.1*	1.5*	0.2
	MIN	0.1	0.2	8.6	0	5.7	0	12	8.7	0.2*	0.4*	0.3*	0.10	0.00	0.02	2.6*	0.9*	0.0
	MAX	5.2	3.6	23	0.13	8.8	0.8	18	77	1.3*	2.6*	12*	0.20	0.06	0.27	28*	5.7*	0.3
NH_4NO_3	M	0.6	0.7	57	0.1	11	0.6	2.8	21	1.6*	1.1*	4.8*	0.13	0.513*	0.04	5.2*	3.8*	0.00
	SD	0.4	0.4	13	0.3	1.7	0.2	0.9	22	1.1*	0.6*	1.2*	0.04	0.003*	0.02	2.6*	1.4*	0.02
	MIN	0.2	0.3	44	0	7.2	0.3	1.7	8.7	1.0*	0.3*	2.9*	0.06	0.511*	0.00	0.7*	1.2*	0.00
	MAX	1.3	1.3	83	0.6	12	0.8	4.3	77	4.3*	2.3*	6.3*	0.19	0.517*	0.06	9.4*	6.0*	0.03
Pseudo-total																		
Aqua regia	M	4160	4380	2920	66	220	190	35	295	4.0	13	11	106	1.2	17	29	19	8.2
	SD	686	777	376	12	92	60	19	200	0.3	0.9	0.9	8.2	0.2	5.5	74	1.2	1.4
	MIN	3090	3500	2400	53	92	87	14	15.2	3.6	12	9.0	93	1.0	8.8	0.9	17	6.6
	MAX	5020	5720	3520	82	380	270	68	541	4.3	14	12	118	1.4	23	230	20.54	11

[a] The values were not taken into consideration because of the type of used extractant Na_2EDTA.

Considering other single extraction procedures, the highest concentrations of Al, Mo, Na, P, S and Si were extracted by CH_3COOH, as an aggressive extractant which could isolate elements from the rhizosphere of many plants. Chelating agent Na_2EDTA isolated the highest concentrations of potentially mobile soil fraction (Cd, Co, Cu, Fe, K, Mn, Ni, Pb, V, Zn). The concentrations of Cd (0.03–0.16 μg g^{-1}), Cr (0.00–0.09 μg g^{-1}), Ni (0.3–3.0 μg g^{-1}), Pb (1.3–4.4 μg g^{-1}) and Zn (0.8–5.3 μg g^{-1}) extracted by Na_2EDTA were comparable with the data obtained in study of McGrath (1996). In addition, similar ranges of Pb concentrations (0.02–0.09 μg g^{-1}, 1.3–4.4 μg g^{-1}) were extracted with Na_2EDTA in the study of Bakircioglu et al. (2011) (Table 2, Fig. S1).

3.1.1. PCA and CA analysis for assessment of element mobile/bioavailable phases in vineyard's soil

Multiple correlations obtained among elements by PCA could indicate their common agro-chemical origin, but also similar plant uptake mechanism from the soil (Kabata – Pendias and Kabata, 2001; Buccolier et al., 2010; Vystavna et al., 2015). The most significant correlations ($p < 0.05$) were determined between Cu, S and Zn concentrations extracted using deionized water during 16 h (Fig. 2C). Using Na_2EDTA, and CH_3COOH as a single extraction procedure the significant correlation ($p < 0.05$) between Cu and Zn were obtained (Fig. 2A, F). A significant correlation ($p < 0.05$) between concentrations of Cu and S, extracted with deionized water during 16 h, NH_4NO_3, and $CaCl_2$, were established (Fig. 2C–E). These elements could have entered only through the soil surface layer, while grapevines were primarily treated with foliar by fungicide, copper (II)-sulphate. In addition, the concentration of S, extracted with deionized water during 16 h, was correlated with the concentrations of Mn and Na. It can be assumed that the correlation between these elements points to their origin from the fertilizers or pesticides used in agriculture production (Kabata – Pendias and Kabata, 2001). Contrary, no correlation was observed between Cu and the other element concentrations after 2 h water extraction. The 2 h extraction was not long enough for obtaining the balance between solubility and sorption on the substrate matrix.

3.2. Plant – soil experiment: selection of best procedure for extraction of plant bioavailable major and trace elements from agricultural soil

Considering the analyzed grapevine parts (leaf, skin, pulp, and seed), the highest concentrations of Cd, Cr, Cu, Ni, Pb, and Zn were determined in the leaf samples. Potassium, as one of the most important nutrients essential for plant growth, was determined in the highest concentrations in all grapevine parts. The highest concentration of Ni (52 μg g^{-1}) was determined in the skin of *Prokupac* species, planted on the parcel T4 located 1.5 km away from the Institute of Nuclear Science "Vinča". Contrary, much lower concentration of Ni (0.12 μg g^{-1}) was determined in the skin samples of species *Cabernet Sauvignon* planted at the same parcel. The concentrations of Ni in all grapevine parts and species varied from 0.41 to 1.44 μg g^{-1} with the exception of the concentration in *Prokupac* skin (52 μg g^{-1}). In all these samples, Ni concentrations were below the range of excessive or toxic amounts, which in the most plant species vary from 10 to 1000 μg g^{-1} (Kabata – Pendias and Mukherjee, 2007). The high concentration of Cu was determined in the leaf samples of all grapevine species (29–170 μg g^{-1}). Therefore, pH value (3.85–7.07) plays an important role in bioavailability of Cu in soils. Concentrations of Pb in the grapevine parts were low. Low concentrations were especially determined in the leaf (0.02 μg g^{-1}) and the seed (0.02 μg g^{-1}) of *Cabernet Franc,* planted on the parcel T6 (the obtained concentration in soil was 226 μg g^{-1}). Lead is only passively adsorbed by the plant roots (Kabata – Pendias and Mukherjee, 2007), and thus, plant uptake of Pb from soil is generally very low (Table 3).

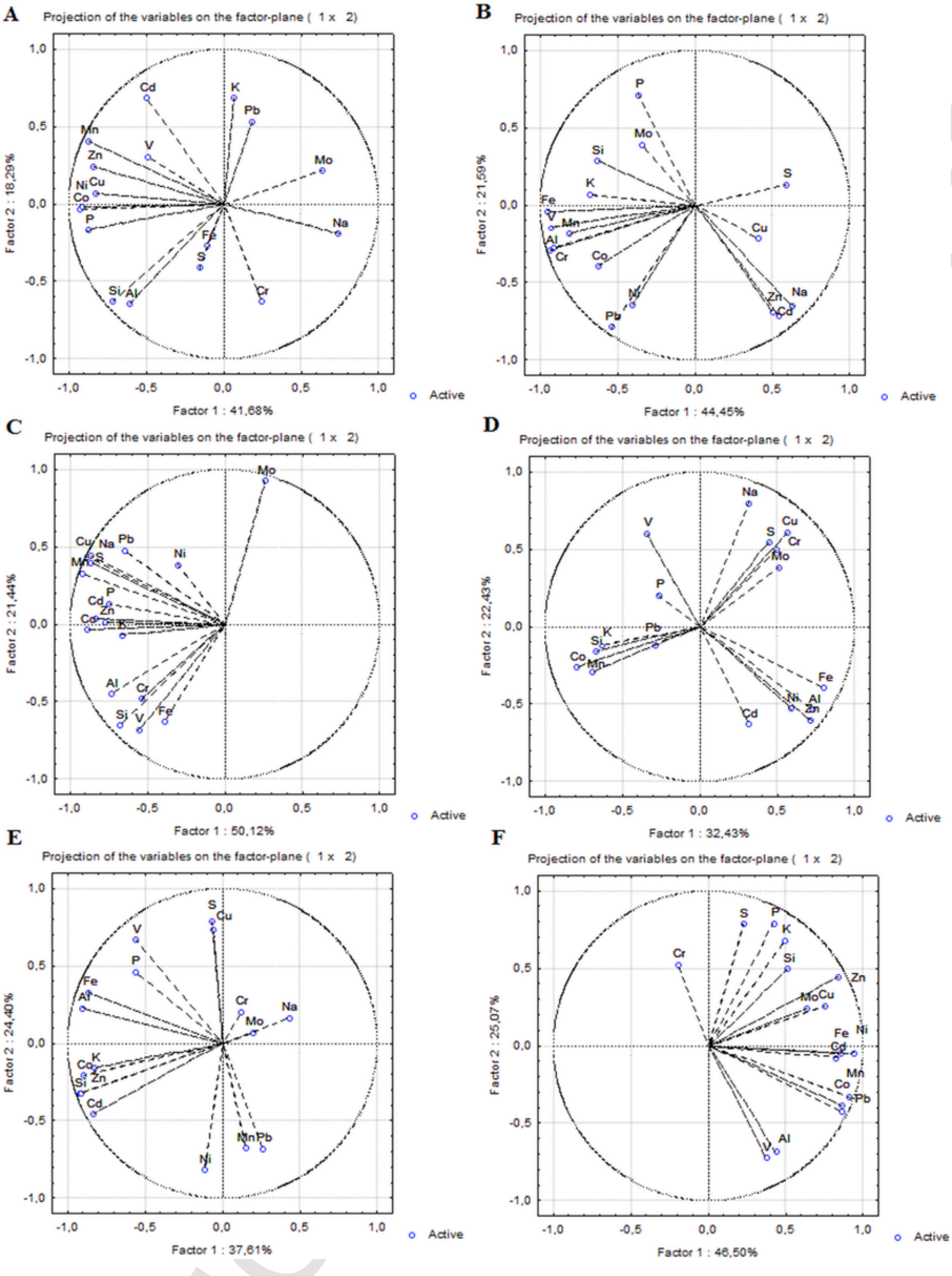

Fig. 2. The overview of PCA for all obtained major and trace elements extracted applying CH_3COOH (A), 2 h H_2O (B), 16 h H_2O (C), $CaCl_2$ (D), NH_4NO_3 (E) and Na_2EDTA (F) single-extraction procedures.

Similarities among the element concentrations determined in the skin, seed and pulp samples were observed by PCA. In addition, the leaf samples of all species were grouped (Fig. 3). Plot scores 1 and 2 were isolated. The exception was the *Prokupac* species. The skin and leaf samples of *Prokupac* species were distanced from the other skin and leaf samples which indicate the different behavior of *Prokupac* from other plant species (Fig. 3).

Different extractant took place in the extraction process of either major or trace elements. Bioavailable soil fraction of the major elements (Al, Fe, and K) extracted applying CH_3COOH, $CaCl_2$, NH_4NO_3, Na_2EDTA, 2 h and 16 h deionized water procedures were grouped as the separated cluster (Fig. 4a). However, according to the obtained dendrogram (Fig. 4b), the aqua regia was efficiently in the extraction of trace elements from the soil which were bioavailable for the grapevine leaves. Deionized water procedure enabled extraction of bioavailable trace elements which were in relation with trace elements isolated from the grape seed and the grape pulp. Finally, the weak salts $CaCl_2$ and NH_4NO_3 as well as Na_2EDTA and CH_3COOH

Table 3
Major and trace element concentrations isolated from grape (leaf, seed, pulp, and skin) (μg g^{-1}, ^{a}ng g^{-1}) (n = 3).

Major and trace elements in plants		Al	Cd^{a}	Cr	Cu	Fe	K	Ni	Pb	Zn
Riesling Rain seed (*RRSE)	M	< DL	6	0.034	3.6	9.6	1403	0.15	0.032	4.8
	SD	/	0.1	0.001	0.6	1.0	320	0.01	0.002	0.1
Cabernet Franc seed (*CSSE)	M	0.9	6	0.16	4.5	11.2	2099	0.080	0.023	5.7
	SD	0.1	0.2	0.02	0.6	1.2	430	0.001	0.002	0.2
Merlot seed (*MSE)	M	0.1	3	0.023	4.1	10.2	1885	0.041	< DL	4.9
	SD	0.0	0.1	0.002	0.3	0.8	470	0.001	/	0.2
Prokupac seed (*PSE)	M	0.3	4	0.027	4.7	9.3	1108	0.050	0.042	3.8
	SD	0.0	0.1	0.001	0.9	1.1	320	0.001	0.006	0.1
Cabernet Sauvignon seed (*CSSE)	M	0.4	3	0.038	4.6	11.4	1585	0.119	0.060	4.7
	SD	0.0	0.1	0.001	0.8	1.2	110	0.002	0.008	0.3
Burgundy seed (*BSE)	M	0.6	5	0.083	5.1	14.3	1444	0.102	0.021	5.7
	SD	0.0	0.1	0.001	1.1	1.2	260	0.006	0.002	0.2
Cabernet Sauvignon pulp (*CSP)	M	0.6	6	0.420	1.4	5.2	1956	0.075	0.11	0.6
	SD	0.1	0.2	0.008	0.1	0.3	480	0.008	0.01	0.0
Burgundy pulp (*BP)	M	0.5	4	0.14	1.1	5.4	1796	0.050	0.169	0.4
	SD	0.1	0.1	0.01	0.5	0.6	420	0.004	0.009	0.0
Riesling Rain skin (*RRS)	M	0.8	5	0.028	1.5	4.8	1237	0.099	0.049	1.3
	SD	0.1	0.1	0.001	0.6	0.6	320	0.007	0.007	0.1
Cabernet Franc skin (*CFS)	M	1.7	4	0.007	1.7	5.9	1604	0.050	< DL	1.2
	SD	0.1	0.1	0.001	0.2	0.8	280	0.003	/	0.1
Merlot skin (*MS)	M	0.8	4	0.005	1.2	4.1	1212	0.045	< DL	0.6
	SD	0.1	0.1	0.001	0.1	0.2	630	0.007	/	0.0
Prokupac skin (*PS)	M	4	6	0.045	1.2	7.8	755	52	0.24	1.1
	SD	0.3	0.2	0.001	0.1	0.2	140	3	0.03	0.0
Cabernet Sauvignon skin (*CSS)	M	1.1	2	0.22	1.2	5.2	1124	0.12	< DL	0.7
	SD	0.1	0.1	0.01	0.1	0.3	380	0.01	/	0.0
Burgundy skin (*BS)	M	2.3	4	0.093	1.8	6.8	1032	0.09	0.062	1.1
	SD	0.6	0.1	0.003	0.1	0.2	510	0.01	0.008	0.1
Riesling Rain skin (*RRS)	M	70	20	0.28	140	100	2585	0.872	0.19	14
	SD	0.8	1.2	0.02	8	8	380	0.006	0.01	0.1
Cabernet Franc leaf (*CFL)	M	52	33	0.26	63	72	1913	0.44	0.023	12.
	SD	5	1.6	0.02	10	10	380	0.01	0.003	2.3
Merlot leaf (*ML)	M	79	20	0.31	29	120	1973	0.72	0.0043	7.8
	SD	11	2.6	0.04	3	20	520	0.06	0.0001	0.8
Prokupac leaf (*PL)	M	72	20	0.30	90	110	1080	0.72	0.365	7.3
	SD	12	2.1	0.01	10	10	180	0.08	0.008	0.8
Cabernet Sauvignon leaf (*CSL)	M	60	20	0.29	82	100	2035	0.44	0.072	9

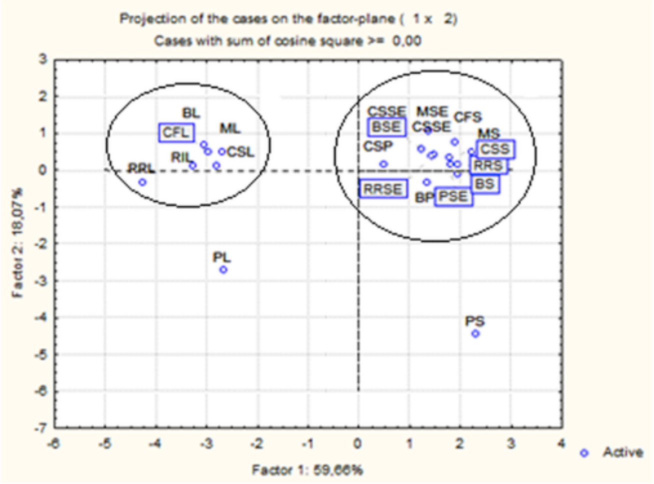

Fig. 3. The observation of similarities between grapevine plant parts and species applying PCA.

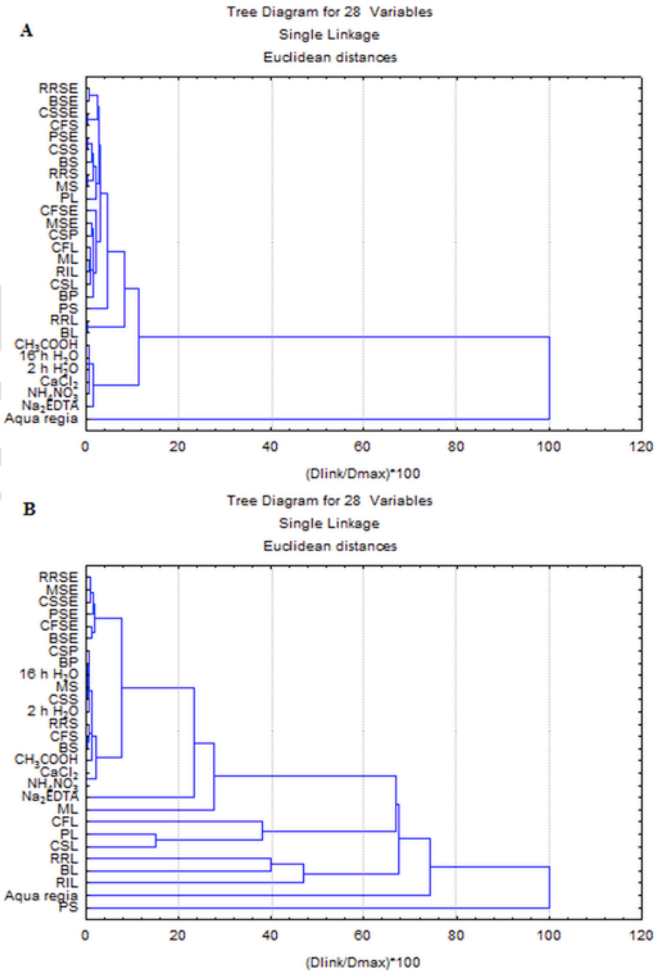

Fig. 4. The clusters of different extracting bioavailable major (A) and trace (B) elements from soil and plant (*Table 3).

enabled extraction of trace elements from the soil, which were in relation with trace elements isolated from the grape skin. Thus, bioavailable soil fractions have an important influence on the trace elements content in the grape seed and pulp, but not in leaves because in the leaves trace elements could originate from either air or soil.

3.3. Environmental risk assessment

3.3.1. Soil samples

Calculated values of CF showed that the concentrations of Zn (Table S1) in the soil samples from parcels T4 and T5 were higher than 1 (refers to moderate contamination), and for T6 parcel only CF for Pb (27.3) was higher than 1. Calculated CF higher than 6 indicates very high contamination (Matong et al., 2016). The higher CF could be explained by the pollution influence of traffic from the nearby main road. In addition increased values of CF (Table S1) calculated for the soil samples collected from parcels T4 (Cd, Co, Cu, Zn) and T5 (Cd, Co, Cr, Cu, Mo, V, Zn), could be a result of the influence of the pollution originating from the waste disposal area of Institute of nuclear research "Vinča". Finally, the PLI values higher than 1 were only calculated for the parcel T6 (1.16).

The calculated I_{geo} for soil samples in parcel T6 for Pb was in grade 5 (4.2), which indicate that the T6 parcel is severely extremely polluted. The calculations for EF (background Al) shown the most values among range $1 < EF < 2$. Base on the calculation it could be noticed that for the most parcels enrichment was minimal to moderate, expect of EF for Pb in the T6 parcel (Table S2). According to EF values, it could be noticed that there were the anthropogenic influences of Pb, and Ni in parcel T6, and Zn in parcel T4 (Table S2).

All the above environmental calculations indicate that just for the parcel T6 there were high environmental implications of the pollution by the studied elements, and for parcels T4, T5, T10, and P there were some moderate environmental implications (Tables S1–S2).

3.3.2. Grapevine samples

The bioaccumulation factors (BF) of Al, Cd, Cr, Cu, Fe, K, Ni, Pb and Zn for all grapevine parts were calculated (Table S3). Observing the results, it could be noticed that the leaves of *Riesling Rain* (1.77), *Riesling Italian* (1.66), *Cabernet Sauvignon* (1.36), and *Cabernet Franc* (1.38; 1.82) could be considered as accumulators of Zn from the soil. In addition, the leaves of *Riesling Rain* (1.19), *Burgundy* (1.02), and *Riesling Italian* (1.66) could be an accumulator of Cu from the soil. The skin of species *Prokupac* (4.89) has only shown as a markedly plant accumulator of Ni from the soil (Table S3).

4. Conclusion

Obtaining the results of applied single extraction procedures, and pseudo-total digestion on the vineyard soil samples and grapevine parts, it could be noticed that grapevine leaves and different parts of grapevine barriers could contain major and trace elements originating from the soil and from the agricultural activities in the vineyards. Concentrations of Cu and Pb (at location T6) obtained by pseudo-total digestion procedure were higher than the concentrations prescribed in the national and international .

The single extraction procedures, such as $CaCl_2$, NH_4NO_3, and deionized water extracting during 16 h could be recommended as suitable single extractions for obtaining the concentrations of Cu and S which could originate from the same source. In addition, single extractions of grapevine soil with Na_2EDTA, CH_3COOH and deionized water extracting during 16 h could be appropriate procedures for determining correlated concentrations of Cu and Zn. Contrary to 2 h, a longer water extraction time (16 h) enable to take out the soluble elements which could be associated with complexing compounds presented in soil.

Bioavailable trace elements soil fraction for the grape seed and grape pulp could be extracted by deionized water. Trace elements from the soil which were bioavailable to the grape skin could be extracted by CH_3COOH, Na_2EDTA, $CaCl_2$ and NH_4NO_3. However, due to a different way of element uptake into grapevine leaves, bioavailable soil fractions have an influence on trace elements content in the seed and the pulp, but not in the leaves, probably because of the additional influence. Aqua regia could be recommended for extraction of the leaf bioavailable fraction of the elements. All plant species except *Prokupac* have shown similarities between each other.

Environmental risk formulas (CF, PLI, I_{geo}, and EF) enable to identify moderate (T4, and T5) to extremely (T6) polluted parcels of the vineyard influenced by surrounding sources of pollution. According to BF, the leaves of *Riesling Rain*, *Riesling Italian*, *Cabernet Sauvignon* and *Cabernet Franc* could be good accumulators of Zn from vineyard soils and the leaves of species *Riesling Rain*, *Burgundy* and *Riesling Italian* could be accumulators of mobile Cu from vineyard soil.

Acknowledgements

The authors are grateful to the Ministry of Education and Science of the Republic of Serbia (Project No. III43007 and 172001).

Appendix A. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.chemosphere.2016.12.090.

References

Alonso Castillo, M.L., Vereda Alonso, E., Siles Cordero, M.T., Cano Pavón, J.M., García de Torres, A., 2011. Fractionation of heavy metals in sediment by using microwave assisted sequential extraction procedure and determination by inductively coupled plasma mass spectrometry. Microchem. J. 98, 234–239.

Bakircioglu, D., Bakircioglu Kurtulus, Y., Ibar, H., 2011. Comparison of extraction procedures for assessing soil metal bioavailability of to wheat grains. Clean. - Soil Air Water 39, 728–734.

Bravo, S., Amorós, J.A., Pérez-de-los-Reyes, C., García, F.J., Moreno, M.M., Sánchez-Ormeño, M., Higueras, P., 2015. Influence of the soil pH in the uptake and bioaccumulation of heavy metals (Fe, Zn, Cu, Pb and Mn) and other elements (Ca, K, Al, Sr and Ba) in vine leaves, Castilla-La Mancha (Spain). J. Geochem. Explor. 0.1016/j.gexplo.2015.12.012.

Buccolier, I.A., Buccolieri, G., Dell'Atti, A., Strisciullo, G., Gagliano-Candela, R., 2010. Monitoring of total and bioavailable heavy metals concentration in agricultural soils. Environ. Monit. Assess. 168, 547–560.

Chen, M., Ma, L.Q., 2001. Comparison of three aqua regia digestion methods for twenty Florida soils. Soil Sci. Soc. Am. J. 65, 491–499.

Chen, H., Teng, Y., Lu, S., Wang, Y., Wang, J., 2015. Contamination features and health risk of soil heavy metals in China. Sci. Total Environ. 512 – 513, 143–153.

EU Council Directive 86/278/EEC.

Hall, G.E.M., MacLaurin, A.I., Garrett, R.G., 1998. Assessment of the 1 M NH_4NO_3 extraction protocol to identify mobile forms of Cd in soils. J. Geochem. Explor 64, 153–159.

Hazrat, A., Khan, E., Anwar, Sajad M., 2013. Phytoremediation of heavy metal concepts and applications. Chemosphere 91–7, 869–881.

Hu, Z.Y., Zhao, F.J., McGrath, S.P., 2005. Sulphur fractionation in calcareous soils and bioavailability to plants. Plant Soil 268, 103–109.

Islam, S., Ahmed, K., Al-Mamun, H., 2015. Distribution of trace elements in different soils and risk assessment: a case study for the urbanized area in Bangladesh. J. Geochem. Explor 158, 212–222.

Kabata – Pendias, A., Kabata, H., 2001. Trace Elements in Soils and Plants. CRC Press, USA.

Kabata – Pendias, A., Mukherjee, A., 2007. Trace Elements from Soil to Human. Springer-Verlag Berlin Heidelberg, Germany.

Kaiser, H.F., 1958. The Varimax criterion for analytic rotation in factor analysis. Psychometrika 23, 187–200.

Kelepertzis, E., Paraskevopoulou, V., Argyraki, A., Fligos, G., Chalkiadaki, O., 2015. Evaluation of single extraction procedures for the assessment of heavy metal extractability in citrus agricultural soil of a typical Mediterranean environment (Argolida, Greece). J. Soils Sediments 15, 2265–2275.

Khan, S., Cao, Q., Zheng, Y.M., Huang, Y.Z., Zhu, Y.G., 2008. Health risks of heavy metals in contaminated soils and food crops irrigated with wastewater in Beijing, China. Environ. Pollut. 152, 686–692.

Likuku, A.S., Mmolawa, K.B., Gaboutloeloe, G.K., 2013. Assessment of heavy metal enrichment and degree of contamination around the copper - nickel mine in the Selebi Phikwe region, Estern Botswana. Environ. Ecol. Res. 2, 32–40.

Lu, X.W., Zhang, X.L., Li, L.Y., Chen, H., 2014. Assessment of metals pollution and health risk in dust from nursery schools in Xi'an, China. Environ. Res. 128, 27–34.

Marina, B., Chopina, E.I.B., Jupinet, B., Gauthierb, D., 2008. Comparison of microwave-assisted digestion procedures for total trace element content determination in calcareous soils. Talanta 77, 282–288.

Matong, J.M., Nyaba, L., Nomngongo, P.N., 2016. Fractionation of trace elements in agricultural soils using ultrasound assisted sequential extraction prior to inductively coupled plasma mass spectrometric determination. Chemosphere 154, 249–257.

McGrath, D., 1996. Application of single and sequential extraction procedures to polluted and unpolluted soils. Sci. Total Environ. 178, 37–44.

Meers, E., Samson, R., Tack, F.M.G., Ruttens, A., Vandegehuchte, M., Vangronsveld, J., Verloo, M.G., 2007. Phytoavailability assessment of heavy metals in soils by single extractions and accumulation by Phaseolus vulgaris. Environ. Exp. Bot. 60, 385–396.

Morel, J.L., 1997. Bioavailability of trace elements to terrestrial plants. In: Tarradellas, J., Bitton, D. (Eds.), Soil Ecotoxicology 6. CRC. Lewis Publishers, Boca Raton, pp. 141–175.

National Regulation, Republic of Serbia 88/2010.

Niesiobedzka, K., 2016. Mobile forms and migration ability of Cu, Pb and Zn in forestry system in Poland. Environ. Earth Sci. 75, 122.

Pelfrêne, A., Waterlot, C., Mazzuca, M., Nisse, C., Cuny, D., Richard, A., Denys, S., Heyman, C., Roussel, H., Bidar, G., Douay, F., 2012. Bioaccessibility of trace elements as affected by soil parameters in smelter-contaminated agricultural soils: a statistical modeling approach. Environ. Pollut. 160, 130–138.

Pinto, E., Almeida, A.A., Ferreira, I.M.P.L.V.O., 2015. Assessment of metal(loid)s phytoavailability in intensive agricultural soils by the application of single extractions to rhizosphere soil. Ecotox. Environ. Safe 113, 418–424.

Pueyo, M., López-Sánchez, J.F., Rauret, G., 2004. Assessment of $CaCl_2$, $NaNO_3$ and NH_4NO_3 extraction procedures for the study of Cd, Cu, Pb and Zn extractability in contaminated soils. Anal. Chim. Acta 504, 217–226.

Quevauviller, Ph, 1998. Operationally defined extraction procedures for soil and sediment analysis, II. Certified reference materials. Trends Anal. Chem. 17, 632–642.

Quevauviller, Ph, 2002. Operationally-defined extraction procedures for soil and sediment analysis. Part 3: new CRMs for trace element extractable contents. Trends Anal. Chem. 21, 774–785.

Quevauviller, P., Imbert, J.L., Olle, M., 1993. Evaluation of the use of microwave oven systems for the digestion of environmental samples. Microchim. Acta 112, 147–154.

Quevauviller, Ph, Lachica, M., Barahona, E., Rauret, G., Ure, A., Gomez, V., Muntau, H., 1996. Interlaboratory comparison of EDTA and DTPA procedures prior to certification of extractable trace elements in calcareous soil. Sci. Total Environ. 178, 127–132.

Radha, R., Tripathi, R.M., Vinod, K.A., Sathe, A.P., Khandekar, R.N., Nambi, K.S.V., 1997. Assessment of Pb, Cd, Cu and Zn exposures of 10 year old children in Mumbai. Environ. Res. 80, 215–221.

Radulescu, C., Stihi, C., Popescu, I.V., Dulama, I.D., Chelarescu, E.D., Chilian, A., 2013. Heavy metal accumulation and translocation in different parts of Brassica oleracea L.. Rom. Journ. Phys. 58, 1337–1354.

Rao, C.R.M., Sahuquillo, A., Lopez Sanchez, J.F., 2008. A Review of the different methods applied in environmental geochemistry for single and sequential extraction of trace elements in soils and related materials. Water Air Soil Pollut. 189, 291–333.

Relić, D., Đorđević, D., Sakan, S., Anđelković, I., Miletić, S., Đuričić, J., 2011. Aqua regia extracted metals in sediments from the industrial area and surroundings of Pančevo, Serbia. J. Hazard. Mater. 186, 1893–1901.

Ruby, M.V., Davis, A., Schoof, R., Eberle, S., Sellstone, C.M., 1996. Estimation of lead and arsenic bioavailability using a physiologically based extraction test. Environ. Sci. Technol. 30, 422–430.

Sakan, S., Đorđević, D., Dević, G., Relić, D., Anđelković, I., Đuričić, J., 2011. A study of trace element contamination in river sediments in Serbia using microwave-assisted aqua regia digestion and multivariate statistical analysis. Microchem. J. 99, 492–502.

Sakan, S., Dević, G., Relić, D., Anđelković, I., Sakan, N., Đorđević, D., 2014. Environmental assessment of heavy metal pollution in freshwater sediment, Serbia. Clean. Soil Air Water 42, 1–8.

Sandroni, V., Smith, C.M.M., Donovan, A., 2003. Microwave digestion of sediment, soils and urban particulate matter for trace metal analysis. Talanta 60, 715–723.

Santos, S., Costa, C.A.E., Duarte, A.C., Scherer, H.W., Schneider, R.J., Esteves, V.I., Santos, E.B.H., 2010. Influence of different organic amendments on the potential availability of metals from soil: a study on metal fractionation and extraction kinetics by EDTA. Chemosphere 78, 389–396.

Sastre, J., Sahuquillo, A., Vidal, M., Rauret, G., 2002. Determination of Cd, Cu, Pb and Zn in environmental samples: microwave-assisted total digestion versus aqau regia and nitric acid extraction. Anal. Chim. Acta 462, 59–72.

Sylvain, B., Mikael, M.H., Florie, M., Emmanuel, J., Marilyne, S., Sylvain, B., Domenico, M., 2016. Phytostabilization of As, Sb and Pb by two willow species (S. viminalis and S. purpurea) on former mine technosols. Catena 136, 44–52.

Tasdemir, Y., Kural, C., 2005. Atmospheric dry deposition fluxes of trace elements measured in Bursa Turkey. Environ. Pollut. 138, 462–472.

Tiecher, T.L., Ceretta, C.A., Tiecher, T., Ferreira, A.A., Nicoloso, F.T., Soriani, H.H., Rossato, L.V., Mimmo, T., Cesco, S., Lourenzi, C.R., Giachini, A.J., Brunetto, G., 2016. Effects of zinc addition to a copper-contaminated vineyard soil on sorption of Zn by soil and plant physiological responses. Ecotoxicol. Environ. Saf. 129, 109–119.

US EPA 3050b, 1996. Method, Acid Digestion of Sediments, Sledges, and Soils. USEPA, Washington, DC, USA.

Vystavna, Y., Rätsep, R., Klymenko, N., Drozd, O., Pidlisnyuk, V., Klymenko, M., 2015. Comparison of soil-to-root transfer and translocation coefficients of trace elements in vines of Chardonnay and Muscat white grown in the same vineyard. Sci. Hortic. - Amst. 192, 89–96.

Zhang, J., Liu, C.L., 2000. Riverine composition and estuarine geochemistry of particulate metals in China-weathering features, anthropogenic impact and chemical fluxes. Estuar. Coast. Shelf Sci. 54, 1051–1070.

Science of the Total Environment 542 (2016) 394–403

Contents lists available at ScienceDirect

Science of the Total Environment

journal homepage: www.elsevier.com/locate/scitotenv

Moss bag biomonitoring of airborne toxic element decrease on a small scale: A street study in Belgrade, Serbia

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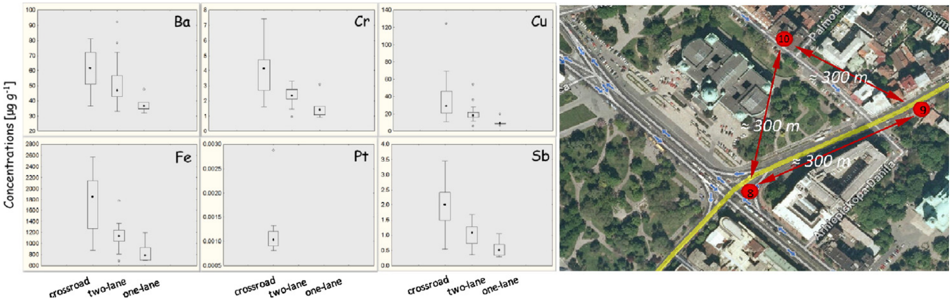
HIGHLIGHTS

- The moss element decline was observed along crossroads and two- and one-lane streets.
- The moss element content was correlated with the counted traffic flows ($r > 0.65$).
- Pedestrian zones could not be assumed as urban pollution background.
- Sb, Cu and Cr have been marked as reliable tracers of traffic pollutant emissions.

GRAPHICAL ABSTRACT



ARTICLE INFO

Article history:
Received 16 July 2015
Received in revised form 16 October 2015
Accepted 19 October 2015
Available online xxxx

Editor: J. P. Bennett

Keywords:
Urban area
Traffic-related air pollution
S. girgensohnii
H. cupressiforme
Devitalizing pretreatment

ABSTRACT

A database of potentially hazardous substances, necessary for estimating the exposure of humans to air pollutants, may be deficient because of a limited number of regulatory monitoring stations. This study was inspired by undeniably harmful effects of human long-term exposure to intense traffic emissions in urban area. Moss bag biomonitors were used to characterize spatial variation of airborne toxic elements near crossroads and two- and one-lane streets. The *Sphagnum girgensohnii* and *Hypnum cupressiforme* moss bags were exposed for 10 weeks to 48 sampling sites across Belgrade (Serbia) during the summer of 2014. In addition, oven-drying pretreatment of the moss bags was tested. During the experimental period, traffic flows were estimated at each site by counting the number of vehicles during the rush hours. The concentrations of 39 elements were determined in the moss samples. There was no significant difference between the results obtained for nontreated and oven-dried moss bags. For the majority of elements, the moss bags identified a common pattern of decrease in the concentration from crossroads to two- and one-lane streets. The exposed moss bags were enriched with Sb, Cu and Cr. The correlation coefficients ($r = 0.65–0.70$) between the moss concentrations of Cr, Cu, Fe and Sb and the site-counted traffic flows also confirmed a dependence of the airborne element content on traffic emissions. A strong correlation with traffic flows makes Sb, Cu and Cr reliable traffic tracers.

1. Introduction

The transport sector represents the dominant source of air pollution in urban areas. Traffic-related air pollutants, including particulate matter (PM) associated with toxic elements, are widespread. These pollutants are often found in higher concentrations in areas near urban

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http://dx.doi.org/10.1016/j.scitotenv.2015.10.091

micro-environments such as major roads, crossroads, junctions and bus stations. Increase of air pollutants beyond the limit values has been associated with many hazardous health effects (European Environment Agency, 2013; United States Environmental Protection Agency, 2013).

Epidemiological studies commonly use data from one or few stations of regulatory monitoring network to estimate personal exposure to PM (Brauer, 2010). However, the results of recent studies have indicated that monitoring stations may not accurately characterize complexities of the spatial dispersion of PMs in an urban area (Wilson et al., 2005; Brauer, 2010). Concentrations of traffic-related air pollutants show dramatic temporal and spatial variations from on-road to near-road micro-environments. For example, particles with size <2.5 μm (<PM2.5), volatile organic compounds (VOCs), NO and polycyclic aromatic hydrocarbons (PAHs) have demonstrated steep gradients in concentrations that attain elevated levels near- and on-road, and a return to background level at distances of approximately 150–200 m (Hagler et al., 2009; Karner et al., 2010).

Regulatory network of air quality monitoring has been typically designed to assess regional and temporal variations of air pollution. For micro-scale setting, sampling sites should be placed at a distance of at least 25 m from the edge of crossroads and not on the carriageways of roads (Directive 2004/104/EC; Directive 2008/50/EC). Because of this, examination of long-term on-road exposure of pedestrians, commuters, cyclists and workers (e.g. drivers and policemen) to traffic-related toxic elements requires alternative approaches (Brauer, 2010).

Biomonitoring represents a simple and cost-effective alternative to regulatory monitoring of air quality. Moss is the most efficient biomonitor available to entrap air pollutants. It lacks a developed root system and thus obtains nutrients from air. Moreover, the high cation exchange capacity of moss increases its adsorbent efficiency (Brown and Bates, 1990). 'Moss bag technique' was introduced by Goodman and Roberts (1971), and it has been developing over the last several decades as an efficient method of active moss biomonitoring of hazardous air pollutants such as heavy metals, non-metals and PAHs (Vasconcelos and Tavares, 1998; Adamo et al., 2003; Ares et al., 2009; Cao et al., 2009; Salo and Mäkinen, 2014; Calabrese et al., 2015; Kosior et al., 2015). This method has been particularly useful for conducting a detailed survey of diversely polluted micro-environments within urban areas, where native mosses are usually absent because of predominantly paved and landscaped surfaces. In contrast to instrumental measurements, moss bags do not require power supply and maintenance. Thus, the moss bag technique could be applied to obtain a satisfactory spatial resolution of monitoring sampling sites, and consequently a detailed database. Another important difference between the moss bag technique and instrumental measurements is sampling time. Instrumental measurements are usually restricted to short time periods, providing daily concentration of pollutants, whereas moss bags act as a long-term integrator of air pollutants providing average concentration. Long-term sampling is prerequisite for the assessment of cumulative exposure to a certain pollutant that has hazardous effect on human health.

The purpose of this study was to assess the level of airborne toxic elements near highly traffic-burdened crossroads and two- and one-lane streets, using the moss bag technique. It is hypothesized that distance decline of toxic element concentrations exists from crossroads to two- and one-lane streets depending on the traffic burden. The aim was also to assess if the moss bags could reflect this phenomenon. The specific objectives were related to the methodological aspects of the application of moss bag technique. The main reason for selecting a particular moss species is its abundance in the study region. Thus, we studied whether (1) two moss species could be interchangeably used for biomonitoring purpose and (2) recommended oven-drying pretreatment of moss bags (Giordano et al., 2009; Ares et al., 2012) is necessary before moss exposure in active biomonitoring studies.

2. Materials and methods

2.1. Study area

The study was conducted in the urban area of Belgrade (44°50′–44°44′N and 20°22′–20°32′ E at 70–250 m altitude). The city is the capital of Serbia, situated at the confluence of the rivers Sava and Danube, with approximately 1.7 million inhabitants.

The biomonitoring survey was conducted from June 15 to August 15, 2014. During the experimental period, the average daily air temperature was 22.5 °C, the prevailing wind direction was WNW, the average daily wind speed was 2 m s^{-1} and the average relative air humidity and total rainfall were 68% and 225 mm, respectively (Republic Hydrometeorological Service of Serbia, 2014).

2.2. Traffic flow estimation

During the experimental period, traffic is considered to be the major source of air pollution, particularly toxic elements, because of the absence of heating systems. To estimate the contribution of traffic intensity to the toxic element concentrations in air, traffic flows were counted. For each study site, traffic flows were recorded by video cameras for later off-site counting. The vehicle fleet was classified into categories of passenger cars, buses, trams, trolleys, motorcycles and light- and heavy-duty vehicles. Traffic flows of each vehicle category were counted for 15 min during the rush hours (7:00–9:00 am and 4:00–6:00 pm) on Wednesdays and Sundays of June and July. The average traffic flows during the experimental period were estimated from these counts (Table 1), using a procedure recommended by the Secretariat for Transport of Belgrade (personal communication).

2.3. Moss bags — sampling, bag preparation and pre- and post-exposure treatments

Two moss species were chosen for moss bag biomonitoring: the most recommended biomonitor species obtained from abroad and the other one, common for the study area. Thus, at the end of May 2014, the moss *Sphagnum girgensohnii* Russow (*S.g.*) was collected from a pristine wetland area located near Dubna, Russia. This area is considered to be an appropriate background site in the previous research (Aničić et al., 2009a, 2009b). The moss *Hypnum cupressiforme* Hedw. (*H.c.*) was obtained from the protected area 'Vršačke planine', Serbia, which is also selected in the previous study (Vuković et al., 2015a). In the laboratory, the green apical parts of the mosses were separated from the brown tissue and manually cleaned of soil particles, plant remains and epiphytes with care. Subsequently, moss material was rinsed thrice with double-distilled water (~10 L of water per 100 g of moss dry weight and 10 min of shaking). Such prepared mosses were air-dried and gently hand-mixed to obtain a homogeneous material. Approximately 1.5 g of the homogeneous moss was packed loosely in 7 × 7 cm nylon net bags with mesh size of 2 mm. The net was previously washed in 0.1 M HNO_3 to eliminate any contamination. The bag dimensions were selected to obtain a moss weight/surface area ratio of approximately 30 mg cm^{-2} for each square centimetre of bag surface, which was suggested by Ares et al. (2012). Simultaneously, the researchers indicated that a higher moss entrapment capacity for particles could be reached if the ratio of moss weight to surface area of the bag is lower (Ares et al., 2014).

To minimize the influence of possible moss growth on element uptake during the experimental period, Giordano et al. (2009) and Ares et al. (2012) recommended an oven-drying devitalizing pretreatment of moss before its exposure in the field. However, in the previous survey (Aničić et al., 2009b), a poor vitality of moss was evident after exposure, because of dry continental climate conditions in the study area. To test the recommended devitalizing moss pretreatment, one half of the prepared moss bags were oven-dried (dry moss, $DM_{S.g.}$ and $DM_{H.c.}$) at 120

°C for 24 h, whereas no devitalizing treatment was applied to another half of the bags (live moss, $LM_{S.g.}$ and $LM_{H.c.}$).

The entire process of the moss sampling and bag preparation was conducted wearing polyethylene powder-free gloves to avoid contamination of the moss material. Several moss bags of both species were stored in the laboratory conditions, at room temperature, as control samples for determination of the initial moss element concentrations.

Although it is recommended to expose three moss bags per sampling site, for the majority of the elements, it is sufficient to expose one moss bag to achieve an error of <10% (Ares et al., 2012). Because of the practical constraints, it was decided to expose only one moss bag of each type ($DM_{S.g.}$, $DM_{H.c.}$, $LM_{S.g.}$ and $LM_{H.c.}$) per sampling site. Particularly, four bags were exposed for 10 weeks using plasticized aluminium holders, specially designed for this purpose. The holders were mounted perpendicular to the lamp posts, at open space, at a representative height of 3–4 m (Vuković et al., 2013). The holders with moss bags were positioned over the Belgrade urban area at 48 sampling sites burdened with different traffic flows – crossroads and two- and one-lane streets (Fig. 1). Three pedestrian zones were chosen to represent urban background sites, which were selected from three different parts of the city: urban-central zone, peri-urban zone and within an urban forest.

After the exposure period, the moss bags from one sampling site were missing, which is indicated by an asterisk in Table 1.

2.4. Chemical analyses

The moss samples were air-dried and homogenized. Approximately 0.3 g of each moss sample (three subsamples per exposure site) was digested for 45 min in a microwave digester (ETHOS 1, Advanced Microwave Digestion System, Milestone, Italy) at 200 °C with 1 mL of 30% H_2O_2 (Sigma-Aldrich, puriss. p.a.) and 7 mL of 65% HNO_3 (Sigma-Aldrich, puriss. p.a., distilled by the apparatus for acid distillation — BERGHOF, Products + Instruments GmbH, Germany). The digested samples were diluted with double-distilled water to a total volume of 50 mL.

Table 1
Characteristics of the biomonitoring sampling sites and traffic flows by vehicle categories (vehicles h^{-1}); LDV — light-duty vehicles; HDV — heavy-duty vehicles.

Sampling site	Type of sampling site	Geographic coordinates	Car	Bus	Tram	Trolley	LDV	HDV	Motorcycle	Total traffic flow [vehicles h^{-1}]
1	Crossroad	44°48′30″ N 20°27′22″ E	1716	260	44		124	76	24	2244
2[a]	Two-lane street	44°48′23″ N 20°27′18″ E	/	/	/	/	/	/	/	/
3	Two-lane street	44°48′29″ N 20°27′30″ E	536	32	/	/	16	/	8	592
4	Two-lane street	44°48′42″ N 20°27′33″ E	708	20	/	/	16	/	12	756
5	Crossroad	44°48′30″ N 20°27′49″ E	3604	108	/	64	124	4	60	3964
6	Two-lane street	44°48′37″ N 20°27′44″ E	1128	28	/	32	24	/	36	1248
7	Two-lane street	44°48′20″ N 20°27′47″ E	504	8	20	/	28	/	4	564
8	Crossroad	44°48′38″ N 20°27′59″ E	4456	128	/	24	148	/	64	4820
9	Two-lane street	44°48′42″ N 20°28′09″ E	2264	28	/	28	112	/	8	2440
10	One-lane street	44°48′45″ N 20°27′58″ E	300	0	/	/	8	/	/	308
11	Two-lane street	44°48′50″ N 20°27′06″ E	588	12	24	/	68	24	8	724
12	Two-lane street	44°48′51″ N 20°27′18″ E	32	68	/	/	/	/	/	100
13	One-lane street	44°48′46″ N 20°27′04″ E	32	0	/	/	/	/	/	32
14	Crossroad	44°48′52″ N 20°27′28″ E	3596	120	/	/	68	/	64	3848
15	Crossroad	44°48′52″ N 20°27′46″ E	3012	88	/	/	88	4	68	3260
16	One-lane street	44°48′54″ N 20°27′50″ E	596	20	/	/	28	/	32	676
17	Two-lane street	44°48′45″ N 20°27′49″ E	1592	32	/	/	44	/	48	1716
18	*Pedestrian zone*	44°49′06″ N 20°27′21″ E	/	/	/	/	/	/	/	/
19	Two-lane street	44°49′00″ N 20°28′12″ E	1032	120	/	/	60	12	16	1240
20	Crossroad	44°49′02″ N 20°28′37″ E	2668	152	/	/	152	4	48	3024
21	Crossroad	44°49′03″ N 20°30′06″ E	1760	68	/	/	80	20	32	1960
22	Two-lane street	44°48′51″ N 20°30′25″ E	124	0	/	/	4	/	4	132
23	One-lane street	44°48′48″ N 20°30′26″ E	296	60	/	/	20	/	12	388
24	Crossroad	44°48′18″ N 20°28′37″ E	2568	28	32	/	52	4	60	2744
25	Two-lane street	44°48′06″ N 20°20′06″ E	2020	0	28	/	32	/	44	2124
26	One-lane street	44°48′08″ N 20°28′43″ E	416	16	/	/	16	/	12	460
27	Crossroad	44°46′60″ N 20°30′05″ E	3140	112	/	28	116	28	20	3444
28	Two-lane street	44°46′59″ N 20°30′16″ E	1124	92	/	4	40	4	8	1272
29	Two-lane street	44°47′24″ N 20°29′54″ E	1672	0	/	28	72	8	20	1800
30	Crossroad	44°47′31″ N 20°28′00″ E	5788	184	/	/	176	/	52	6200
31	One-lane street	44°47′11″ N 20°28′13″ E	360	16	/	/	20	/	/	396
32	Crossroad	44°47′50″ N 20°27′08″ E	1024	60	/	/	20	24	4	1132
33	Crossroad	44°47′58″ N 20°27′53″ E	1104	84	/	/	64	12	12	1276
34	Crossroad	44°47′21″ N 20°26′40″ E	348	4	/	/	4	/	20	376
35	*Pedestrian zone*	44°47′20″ N 20°26′55″ E	/	/	/	/	/	/	/	/
36	Crossroad	44°46′55″ N 20°25′05″ E	1232	144	16	/	60	8	16	1476
37	Two-lane street	44°46′48″ N 20°24′60″ E	1488	144	12	/	52	/	8	1704
38	One-lane street	44°46′46″ N 20°24′53″ E	140	/	/	/	/	/	/	140
39	Crossroad	44°48′53″ N 20°26′06″ E	4476	144	20	/	176	/	48	4864
40	Two-lane street	44°49′00″ N 20°25′48″ E	2496	120		/	88	4	28	2736
41	Crossroad	44°48′12″ N 20°23′17″ E	4472	80	40	/	224	12	20	4848
42	Two-lane street	44°48′08″ N 20°22′59″ E	3616	72	40	/	144	/	16	3888
43	Crossroad	44°49′36″ N 20°23′52″ E	2596	84	/	/	112	/	16	2808
44	Two-lane street	44°49′32″ N 20°24′01″ E	872	68	/	/	40	/	/	980
45	Crossroad	44°50′26″ N 20°24′52″ E	1392	160	/	/	32	/	32	1616
46	Two-lane street	44°50′34″ N 20°24′45″ E	1036	112	/	/	16	4	16	1184
47	*Pedestrian zone*	44°50′41″ N 20°24′48″ E	/	/	/	/	/	/	/	/
48	One-lane street	44°50′55″ N 20°24′06″ E	88	16	/	/	/	/	/	104

[a] The moss bags were missing after the exposure period.

The concentrations of Al, B, Ba, Cu, Fe and Zn were determined using inductively coupled plasma-optical emission spectroscopy (ICP-OES, Thermo Scientific iCAP 6500 Duo, Thermo Scientific, UK). For calibration, a Multi-Element Plasma Standard Solution 4, Specpure (Alfa Aesar GmbH & Co KG, Germany) was used. The concentrations of As, Cd, Ce, Co, Cr, Dy, Er, Eu, Ga, Gd, Ho, La, Li, Lu, Nd, Ni, Pb, Pr, Pt, Rb, Sb, Sc, Sm, Sn, Sr, Tb, Th, Ti, Tl, Tm, V, Y and Yb were determined using inductively coupled plasma-mass spectrometry (ICP-MS, Thermo Scientific iCAP Q, Thermo Scientific, UK). A low-level Elements Calibration Stock, EPA Method Standard (VHG Labs, Manchester) was used for calibration.

To determine the quality of the element analysis, the control samples, certified reference materials and analytical blanks were analyzed once every 10 samples. The moss *Pleurozium schreberi*, M2 and M3 (Steinnes et al., 1997) were used as the certified reference materials.

2.5. Data processing

To assess the element accumulation of the studied moss species, relative accumulation factor (RAF) was calculated as on the moss content of each element after exposure (C_{exposed}) subtracted and then divided by the element content before exposure (C_{initial}):

$$\text{RAF} = (C_{\text{exposed}} - C_{\text{initial}})/C_{\text{initial}}.$$

The abundances of the rare earth elements were graphically represented by normalizing the concentrations of the elements observed in the moss samples to those in different standards such as North American Shale Composite (NASC) and Post-Archean Australian Shales (PAAS) (Dołęgowska and Migaszewski, 2013).

The data were processed using STATISTICA 8.0 (StatSoft Inc., Tulsa, OK, USA) and the free software R (R Development Core Team, 2014). The normality of the data was tested by Shapiro–Wilk test ($p < 0.05$). Wilcoxon signed-rank test ($p < 0.05$) was used to determine significant differences in the elemental concentrations between DM and LM of both species. Spearman's rank correlation was used ($p < 0.05$) to determine the correlation between moss elemental concentrations and traffic flows.

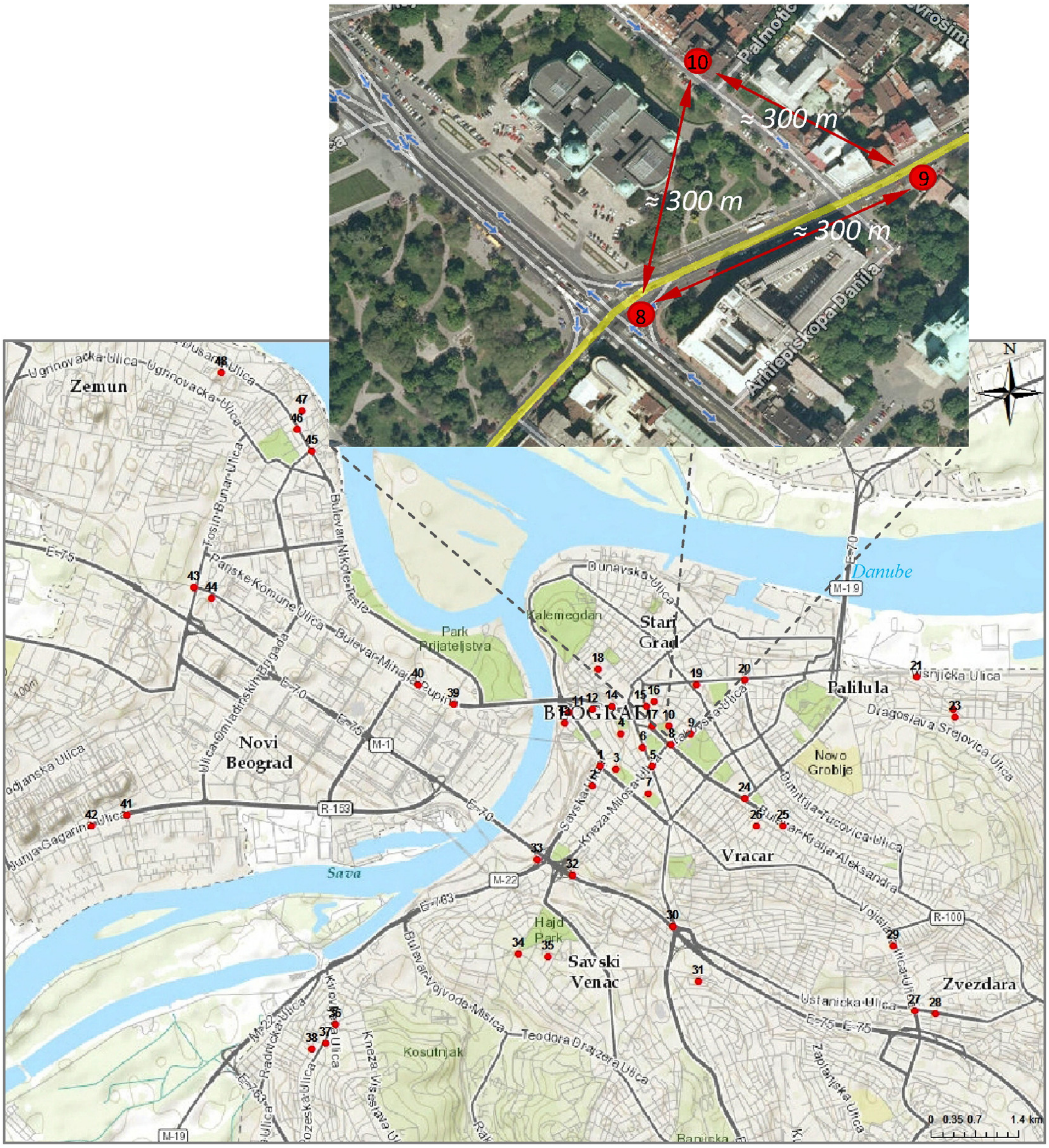

Fig. 1. Map of the Belgrade urban area (Serbia) with 48 sampling sites; the zoom view: an example of experimental set-up of three sampling sites — crossroad and two- and one-lane street.

For comparing the element concentrations between two mosses, a type II regression model was applied by calculating the regression as a standardized major axis regression (Legendre, 2014).

3. Results

3.1. Quality control

The recovery of elements from the reference material M2 ranged from 75% to 111% for the majority of the determined elements; however, recoveries of Ni and Ce were 126% and 56%, respectively. For the reference material M3, the recovery of elements ranged from 80% to 112%, whereas those of Sm, Th, Sc, Cu and Ce were 76%, 69%, 67%, 63% and 57%, respectively. It is important to note that if the element concentrations differed by >20% of the reference value, the results should be considered only as indicative.

The analytical limit of quantification (LOQ) of elements determined in the moss samples by ICP-OES varied from 0.04 to 1.30 $\mu g\ g^{-1}$ for Ba to Al, respectively. For the elements determined by ICP-MS, the LOQ values varied from 0.0002 to 0.1518 $\mu g\ g^{-1}$ for Tb to Li, respectively. All the data obtained were higher than the corresponding LOQ values, except the concentrations of Li and Pt. However, in 20% of the sampling sites, at the highly traffic-burdened crossroads, the concentrations of Pt were higher than the LOQ values, showing their significant enrichment in the moss samples.

3.2. On-road concentrations of airborne elements

The RAF values of 39 elements determined in the *S.g.* and *H.c.* moss samples are shown in Table 2. The concentrations of most elements in the exposed moss bags of both species were higher than the initial element concentrations in the mosses. For all elements, significantly higher ($p < 0.05$) RAF values were obtained for *S.g.* than *H.c.*

Fig. 2 represents a spatial decline of element concentrations in the *S.g.* and *H.c.* moss samples. The *S.g.* and *H.c.* concentrations of As, Ba, Co, Cr, Fe, Ni, Sb and V decreased from the crossroad to the two- and one-lane streets, except for Pb and Zn. The shale-normalized concentrations of the rare earth elements showed a common pattern of decline across the sampling sites (Fig. 3).

In the *S.g.* moss samples, the Spearman's rank correlation coefficients ($p < 0.05$) among the elements were >0.80 for the following pairs of the elements: As–Cu, As–Sb, As–V, Cr–Cu, Cr–Ni, Cr–V, Cu–Fe, Cu–Sb and Ni–V, and >0.90 for the following pairs: As–Cr, As–Fe, Cr–Fe, Cr–Sb and Fe–Sb (Table 1, Supplementary material). In the *H.c.* moss samples, the correlation coefficients were >0.80 between As–Cr, Ba–Co, Ba–Cr, Ba–Cu, Ba–Fe, Ba–Sb, Cr–Cu, Cr–Sb, Cr–V and Fe–V, and >0.90 between As–Co, As–Fe, Co–Cr and Co–Fe (Table 2, Supplementary material). For both moss species, the correlation coefficients between concentrations of Cr, Cu, Fe and Sb and the total traffic flows were significant and ranged from 0.60 to 0.70.

Table 2
Relative accumulation factor (RAF) in the *S.g.* and *H.c.* moss bags exposed for 10 weeks at sites with different traffic intensity: crossroad and two- and one-lane street.

RAF	Element
Sphagnum girgensohnii	
<0	Rb
0–5	Al, B, Ba, Co, Dy, Mg, Sr, Tl, Y
5–10	As, Cd, Ce, Er, Eu, Fe, Ga, Gd, Ho, La, Lu, Nd, Ni, Pb, Pr, Pt[a], Sc, Tb, Ti, Tm, Yb
10–20	Th, V
20–30	Cr, Zn
30–40	Cu
>40	Sb, Sm, Sn
Hypnum cupressiforme	
< 0	Rb
0–5	Al, As, Ba, Fe, Cd, Ce, Co, Dy, Er, Eu, Ga, Gd, Ho, La, Lu, Nd, Ni, Pb, Pr, Sc, Sm, Tm, Ti, Tl, V, Y, Yb
5–10	B, Cr, Cu, Pt[a], Th
10–20	/
20–30	Zn
30–40	/
>40	Sb, Sn

[a] Pt was only detected at the crossroads, thus RAF values were calculated only for the data from crossroads.

The results of type II linear regression model are presented for the elements among which significant correlation ($r > 0.80$) was observed (Fig. 4). The obtained regressions were significant ($p < 0.05$) for As, Ba, Cr, Cu, Fe, Sb and V, but the highest determination coefficients were observed for Sb, followed by Cu and Cr.

The element concentrations in the moss bags exposed in the pedestrian zones were significantly lower than those in the moss bags exposed at the nearest traffic-burdened sampling sites (Table 3). However, the pedestrian zone in the very central area of the city was characterized with markedly increased level of the measured elements, especially Cu. On the contrary, the low content of all elements was obtained in the moss bags exposed at the urban forest sampling site.

Comparison of the element concentrations between $DM_{S.g.}$ and $LM_{S.g.}$, and $DM_{H.c.}$ and $LM_{H.c.}$ indicated no significant difference ($p < 0.05$). Similar patterns of abundance and spatial decline of the element concentrations were observed for DM and LM of both species. Thus, the results obtained for both $DM_{S.g.}$ and $DM_{H.c.}$ were not considered in the further discussion.

4. Discussion

4.1. Spatial variability of the moss element content: crossroads and two- and one-lane streets

Moss bag technique enables a longer sampling period of airborne pollutants, such as toxic elements, and better spatio-temporal resolution of sampling sites. These advantages provide accurate characterization of long-term human exposure to toxic elements, which is more important for the assessment of chronic health effects than daily instrumental measurements. In this study, markedly high RAF values of Sb, Sn, Sm, Cu, Cr and Zn (Table 2) in the moss samples of both study species, *S.g.* and *H.c.*, imply significantly elevated concentrations of these elements in on-road micro-environments. Moreover, at the crossroads, Pt, which is naturally present in very low concentrations in mosses, was markedly enriched. Thus far, concerning the moss bag technique application, Pt has only been observed in the moss bags exposed within a road tunnel (Zechmeister et al., 2006a) and very scarcely in outdoor air in the proximity of bus lines (Rivera et al., 2011).

Both moss species indicated spatial variability for the majority of determined element concentrations within sets of three sampling sites with different traffic flows. Particularly, concentration gradients of As, Ba, Co, Cu, Cr, Ni, Sb and V in the moss samples (Fig. 2) clearly indicate crossroads as pollution hotspots where pedestrians and commuters are exposed to increased levels of toxic elements. At crossroads, a pattern of braking and accelerating driving mode, the so-called 'stop-and-go' mode, is increased. In comparison with constant free flowing traffic mode, which characterized two- and one-lane streets, 'stop-and-go' mode leads to increased emission of pollutants from both exhaust and non-exhaust traffic sources (Tsai et al., 2003).

The decrease in the moss shale-normalized concentration of the rare earth elements (Fig. 3) confirms spatial dependence of these elements on anthropogenic activities, that is, vehicle movements. Both the moss species, *S.g.* and *H.c.*, identified similar order of abundance of the rare earth elements, in agreement with the results of the previous study (Vuković et al., 2015b), which confirms consistent signature of rare earth elements in the urban area under study.

Because of practical constraints, an interchangeable use of the studied moss species was examined. *S.g.* is a species of the most recommended biomonitoring moss genera, whereas *H.c.* is a common moss in Serbia. Although an interchangeable use of the studied moss species

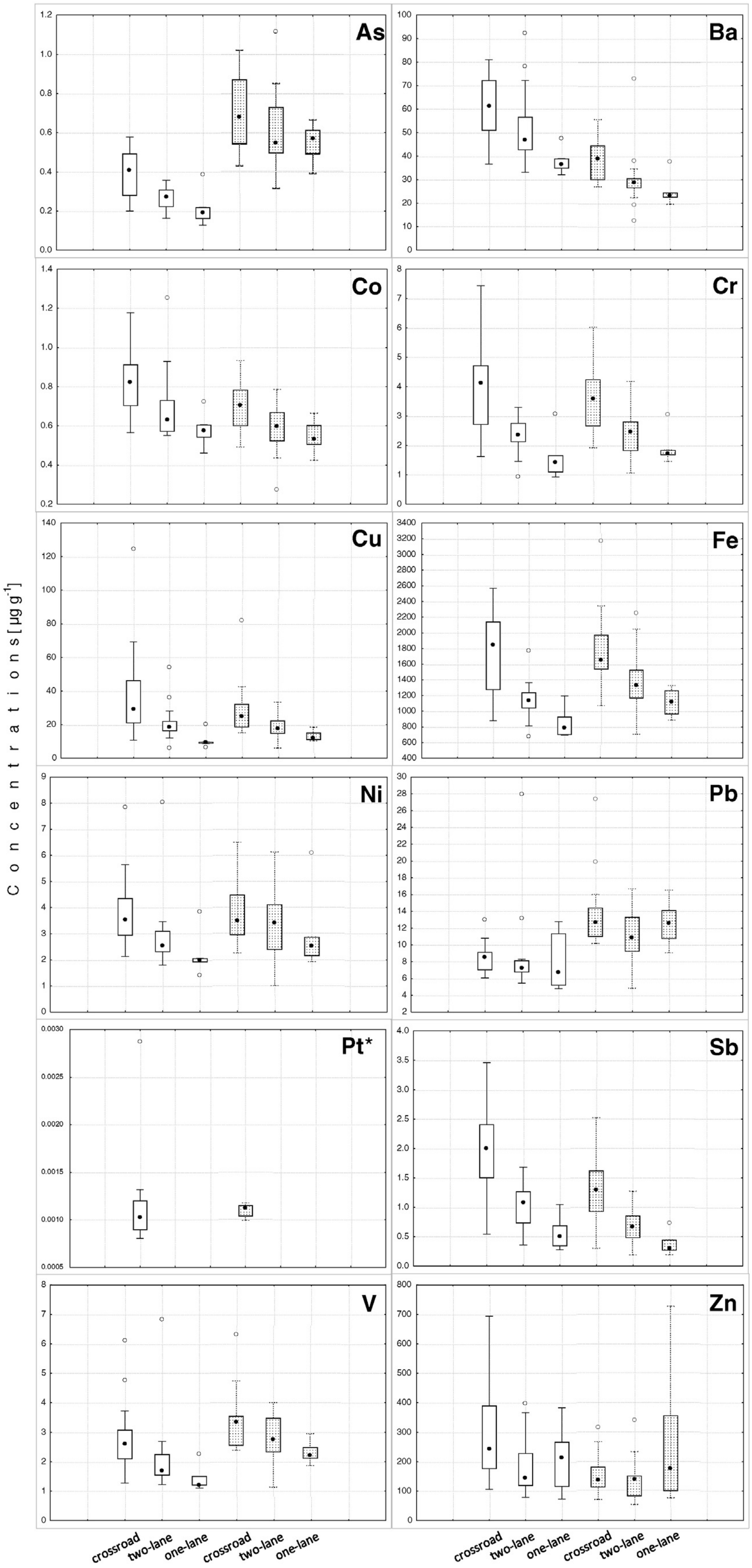

Fig. 2. Box plot of the element (As, Ba, Co, Cr, Cu, Fe, Ni, Pb, Pt, Sb, V and Zn) concentrations (µg g^{-1}) in the *S.g.* (blank box) and *H.c.* (dotted box) moss samples; median, 25th and 75th percentiles are shown in the box; whiskers indicate minimum and maximum values; * platinum was only detected at the crossroads.

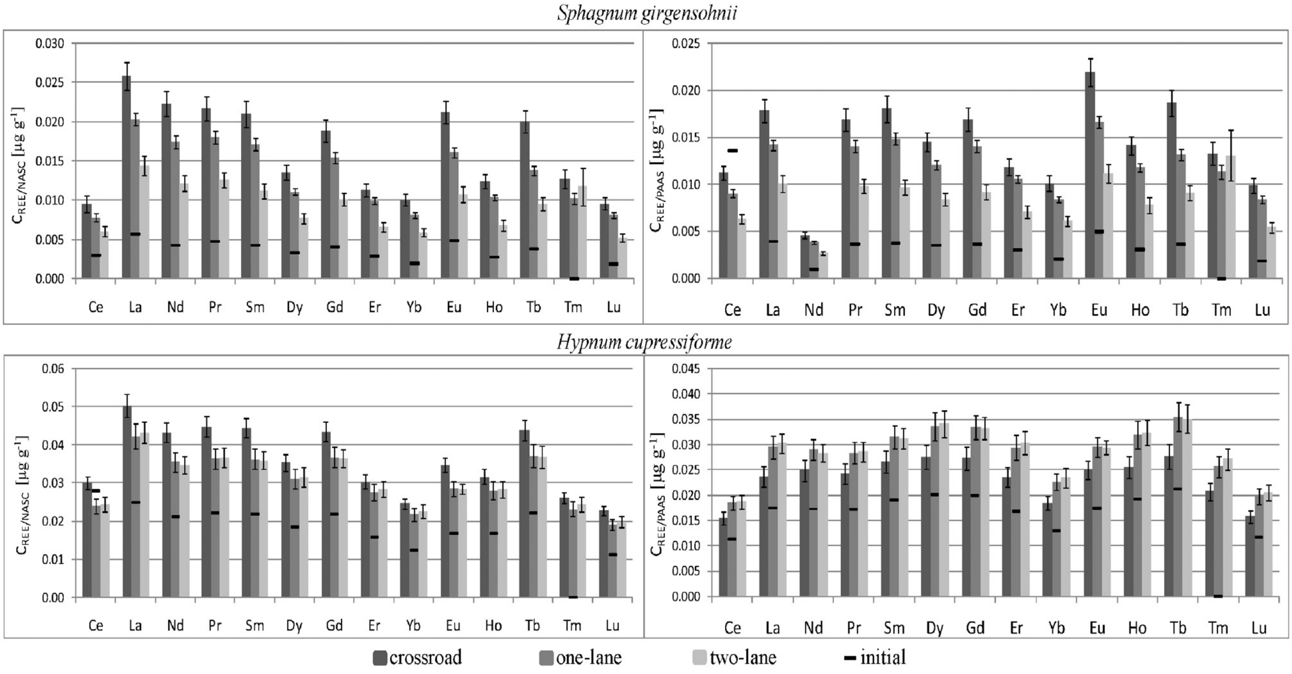

Fig. 3. The median concentrations (µg g^{-1}) of the rare earth elements normalized to NASC and PAAS in the *S.g.* and *H.c.* moss bags exposed at sites with different traffic flows: crossroad and two- and one-lane street; initial — element concentrations in the unexposed moss; the REE order on the *x*-axis is chosen according to their abundances in the NASC and PAAS.

could be helpful, only few studies of bag-based active moss biomonitoring have elaborated the comparison of *S.g.* and *H.c.* (Culicov and Yurukova, 2006; Vuković et al., 2015a). In this study, the moss bags were exposed close to the traffic emission sources. High determination coefficients ($r^2 \geq 0.65$, Fig. 4) obtained for Sb, Cu and Cr suggest the interchangeable use of *S.g.* and *H.c.* in biomonitoring of these elements. In the previous study (Vuković et al., 2015a), the interchangeable use of *S. g.* and *H.c.* was recommended for Cu only based on the determination

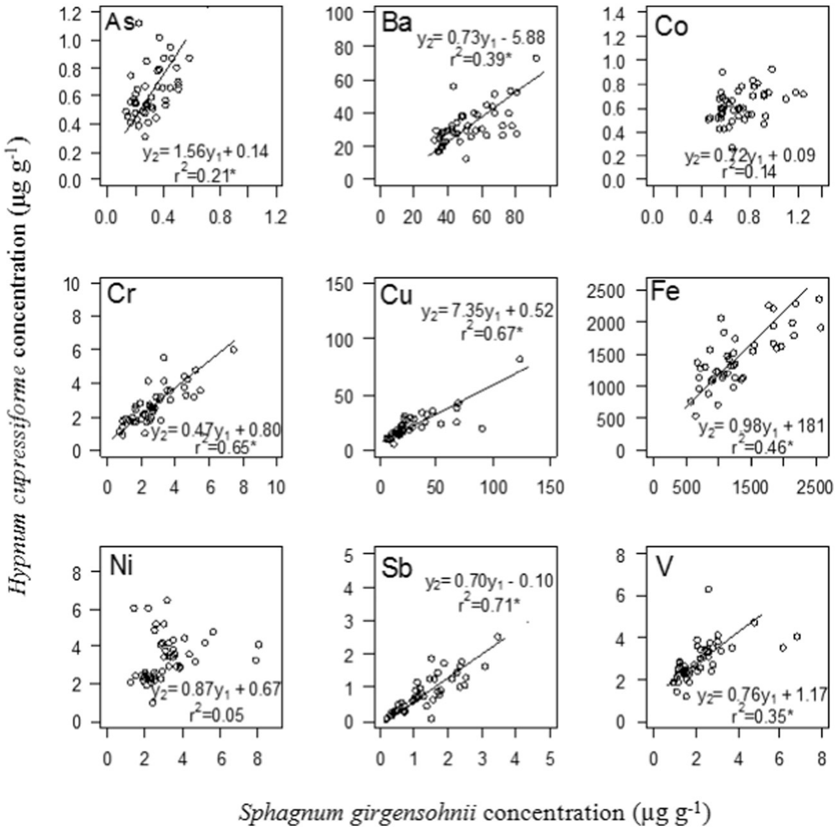

Fig. 4. Type II regression lines for As, Ba, Co, Cr, Cu, Fe, Ni, Sb and V concentrations (µg g^{-1}) in the *S.g.* and *H.c.* moss samples; the black line is obtained using the standardized major axis regression method; * coefficients of determination that are significant ($p < 0.05$).

Table 3
Average concentrations (μg g^{-1}, $n = 3$) of pronounced traffic-related elements ($r = 0.65–0.70$) in the *S.g.* and *H.c.* moss bags exposed in the pedestrian zones and the nearest on-road sampling sites.

Sampling site	*Sphagnum girgensohnii*			
	Cr	Cu	Fe	Sb
Crossroad	4.56 ± 0.85	22.8 ± 3.7	1852 ± 384	1.672 ± 0.094
Central pedestrian zone	2.55 ± 0.46	90.6 ± 11.2	1374 ± 157	0.531 ± 0.095
Crossroad	2.44 ± 0.43	16.0 ± 1.9	998 ± 109	1.497 ± 0.101
Peri-urban pedestrian zone	0.90 ± 0.04	5.6 ± 0.7	645 ± 75	1.512 ± 0.130
Crossroad	2.01 ± 0.13	13.5 ± 0.9	910 ± 93	0.540 ± 0.074
Urban forest	0.73 ± 0.11	5.8 ± 0.8	584 ± 88	0.201 ± 0.049
Sampling site	*Hypnum cupressiforme*			
	Cr	Cu	Fe	Sb
Crossroad	3.76 ± 0.78	27.1 ± 7.1	1672 ± 342	1.420 ± 0.106
Central pedestrian zone	2.01 ± 0.53	19.9 ± 3.3	1129 ± 126	0.285 ± 0.014
Crossroad	2.65 ± 0.76	15.7 ± 2.4	1214 ± 253	1.862 ± 0.131
Peri-urban pedestrian zone	0.90 ± 0.08	9.6 ± 1.1	528 ± 89	0.072 ± 0.005
Crossroad	1.92 ± 0.19	15.2 ± 2.1	1072 ± 238	0.297 ± 0.019
Urban forest	1.10 ± 0.11	9.1 ± 0.9	756 ± 98	0.075 ± 0.007

coefficient ($r^2 = 0.50$). However, in that study, sampling sites were chosen to be representative for the wider areas and not only the on-road micro-environment. Finally, the results of this study indicate that the *S.g.* and *H.c.* moss bags could not be interchangeably used for studies related to on-road micro-environments, except for the biomonitoring of Sb, Cu and Cr.

These results draw attention for better spatial resolution of regulatory monitoring sites from which data are used in epidemiological studies. Moreover, it is important to note that the permitted levels of emission of Cu, Sb, Cr, Pt and rare earth elements, such as Sm, in the air have not been regulated yet by legislation (Directive 2004/107/EC), although these elements are recognized as toxic by the World Health Organization (World Health Organisation, 2013).

4.2. Correlation between the moss element concentrations and traffic flows

A relationship among the element concentrations in the samples of *S.g.* and *H.c.* can be used as a possible indicator of the traffic-related air pollutants. On the basis of the high correlation coefficients ($r > 0.80$) among Ba, Cr, Cu, Fe, Ni, Sb and V (Tables 1 and 2, Supplementary material) in the moss samples of both species, the common sources of these elements can be assumed. The vehicle movements cause: (1) exhaust emissions from the tailpipe; (2) non-exhaust emissions due to wear and tear of vehicle parts, such as brake, tyre and clutch; and (3) resuspension of road dust. Non-exhaust emissions mainly contribute to increased concentrations of Ba, Fe, Cu and Sb (Thorpe and Harrison, 2008; Gietl et al., 2010), whereas fuel combustion is the major source of Cr, Ni, V and Cu emissions (Song and Gao, 2011; EMEP/EEA, 2013).

Given the moderate, but significant correlation coefficient ($r = 0.65–0.70$, $p < 0.05$) (Tables 1 and 2, Supplementary material) between the total traffic flows and the concentrations of Cr, Cu, Fe and Sb in the *S. g.* and *H.c.* samples, these elements could be considered as reliable tracers of traffic-related emissions. The results of this study are in good agreement with those of the previous road traffic studies using both terrestrial mosses and moss bag biomonitors (Zechmeister et al., 2005; Zechmeister et al., 2006b), in which Cr, Cu, Fe and Sb have been identified to be traffic-originated. Regarding the categories of vehicles, the significant correlation coefficient ($r > 0.60$, $p < 0.05$) implies a close relation between the flows of passenger cars and light-duty vehicles and the moss element concentrations of Cr, Cu, Fe and Sb (Tables 1 and 2, Supplementary material), which is in agreement with the previous results obtained for the correlation between the flow of light-duty vehicles and concentrations of Cu and Fe in the moss samples (Adamo et al., 2011).

Zinc has often been reported as one of the tracers for non-exhaust traffic emissions, for example, tyre-wear emissions of Zn are shown to be approximately 1000 and 500 times higher than those of Pb and Cu, respectively (Napier et al., 2008). However, in this study, very low correlation coefficients ($r < 0.20$) were obtained between the moss concentrations of Zn and traffic flows (Tables 1 and 2, Supplementary material). A distinct spatial variability, which is dependent on traffic flow, was not observed (Fig. 2). In addition, no regularity was observed in the Zn spatial distribution related to traffic flow at the study sites in the previous studies (Vuković et al., 2013; Vuković et al., 2015a). The high RAF values obtained for Zn in this study, together with the absence of any other pollution source except traffic is assumed to be due to traffic-related origin of Zn. However, Zn could not be considered as a reliable tracer of traffic flow variability within urban area.

4.3. Could pedestrian zones be considered as an urban background?

It was hypothesized that significantly lower moss element content would be obtained at the control sites in comparison with the remaining on-road sampling sites. However, the markedly elevated concentrations of the majority of elements were determined in the moss bags exposed in the central pedestrian zone (Table 3). Particularly, a higher concentration of Cu was found in this pedestrian zone, which is in accordance with the results of the previous studies (Vuković et al., 2015a; Aničić et al., 2009c). Elevated Cu concentrations were also obtained for the same pedestrian zone in the previous studies related to the use of tree leaf biomonitors (Aničić et al., 2011) and bulk deposition collectors (Mijić et al., 2010). Thus, the enrichment of Cu in the moss could not be considered as an accidental case.

The moss element content indicated that the second pedestrian zone, situated in peri-urban area, was less polluted than the central pedestrian zone. Finally, the third control site was within the biggest urban forest zone. Because the lowest element content was obtained in the moss bags exposed there, it proved to be the cleanest ambient within the city (Table 3).

According to the results of this study, control sites should be carefully chosen in experimental set-up in urban areas with regard to its position. The sampling sites situated in green urban niches could be recommended as appropriate for the assessment of the urban background levels of air pollutants.

5. Conclusion

The *S.g.* and *H.c.* moss bags have indicated distinct spatial decline in the airborne toxic element content along crossroads and two- and one-lane streets. This result necessitates a better characterization of microscale setting for monitoring toxic element distribution. Moss bag technique represents a suitable approach to achieve this goal, because of its simplicity and cost-effectiveness.

According to the results of linear regression analysis, it is concluded that the studied moss species could only be interchangeably used for the assessment of Cr, Cu and Sb within on-road micro-environments. In addition, high moss RAF values were also obtained for these elements: >20 and >5 for *S.g.* and *H.c.*, respectively. Both these moss species indicated Sb, Cu and Cr as reliable traffic tracers regarding their high correlation coefficients ($r > 0.65$) between the moss element concentrations and counted traffic flows. Because moss bags provide information about long-term pollutant concentrations, the results could be indicative for the assessment of exposure of pedestrians, commuters, cyclists and workers to traffic-related Cr, Cu and Sb emissions.

In this study, oven-drying devitalizing pretreatment of moss bags before exposure, one of the recommended methodological steps, was also examined for standardizing the moss bag technique. The live and dry moss bags of *S.g.* and *H.c.* showed no significant differences in abundance and spatial pattern of the majority of 39 determined elements. Under the conditions of the continental climate zone, which is characteristic of the study area, the entrapment of elements is mainly via passive uptake. Thus, concerning the study period from June to August, that is, the summer season, devitalizing pretreatment should not be considered as a necessary step for further application of moss bag biomonitoring in areas characterized by high air temperatures and low relative air humidity. However, devitalized moss bags should be appropriate if the objective is to compare results from biomonitoring surveys performed in regions with diverse climate conditions.

Acknowledgements

The authors are grateful to the City of Belgrade, the company NIS (Serbia) and the Ministry of Education and Science of the Republic of Serbia (Project No. III43007) for financial support, and to the bilateral cooperation with Joint Institute for Nuclear Research, Dubna, Russia.

Appendix A. Supplementary data

Supplementary data to this article can be found online at http://dx.doi.org/10.1016/j.scitotenv.2015.10.091.

References

Adamo, P., Giordano, S., Vingiani, S., Cobianchi, R.C., Violante, P., 2003. Trace element accumulation by moss and lichen exposed in bags in the city of Naples (Italy). Environ. Pollut. 122, 91–103.

Adamo, P., Giordano, S., Sforza, A., Bargagli, R., 2011. Implementation of airborne trace element monitoring with devitalised transplants of *Hypnum cupressiforme* Hedw.: assessment of temporal trends and element contribution by vehicular traffic in Naples city. Environ. Pollut. 159, 1620–1628.

Aničić, M., Tasić, M., Frontasyeva, M.V., Tomašević, M., Rajšić, S., Strelkova, L.P., Popović, A., Steinnes, E., 2009b. Active biomonitoring with wet and dry moss: a case study in an urban area. Environ. Chem. Lett. 7, 55–60.

Aničić, M., Tomašević, M., Tasić, M., Rajšić, S., Popović, A., Frontasyeva, M.V., Lierhagen, S., Steinnes, E., 2009c. Monitoring of trace element atmospheric deposition using dry and wet moss bags: accumulation capacity versus exposure time. J. Hazard. Mater. 171, 182–188.

Aničić, M.,. Tasić, M, Frontasyeva, M.V., Tomašević, M., Rajšić, S., Mijić, Z., Popović, A., 2009a. Active moss biomonitoring of trace elements with *Sphagnum girgensohnii* moss bags in relation to atmospheric bulk deposition in Belgrade, Serbia. Environ. Pollut. 157, 673–679.

Aničić, M., Spasić, T., Tomašević, M., Rajšić, S., Tasić, M., 2011. Trace elements accumulation and temporal trends in leaves of urban deciduous trees (*Aesculus hippocastanum* & *Tilia* spp.). Ecol. Ind. 11, 824–830.

Ares, Á., Aboal, J.R., Fernández, J.A., Real, C., Carballeira, A., 2009. Use of the terrestrial moss *Pseudoscleropodium purum* to detect sources of small scale contamination by PAHs. Atmos. Environ. 43, 5501–5509.

Ares, Á., Aboal, J.R., Carballeira, A., Giordano, S., Adamo, P., Fernández, J.A., 2012. Moss bag biomonitoring: a methodological review. Sci. Total Environ. 432, 143–158.

Ares, Á., Fernández, J.A., Carballeira, A., Aboal, J.R., 2014. Towards the methodological optimization of the moss bag technique in terms of contaminants concentrations and replicability values. Atmos. Environ. 94, 496–507.

Brauer, M., 2010. How much, how long, what, and where, air pollution exposure assessment for epidemiologic studies of respiratory disease. Proc. Am. Thorac. Soc. 7, 111–115.

Brown, D.H., Bates, J.W., 1990. Bryophytes and nutrient cycling. J. Linn. Soc. Bot. 104, 129–147.

Calabrese, S., D'Alessandro, W., Bellomo, S., Brusca, L., Martin, R.S., Saiano, F., Parello, F., 2015. Characterization of the Etna volcanic emissions through an active biomonitoring technique (moss-bags): part 1 — major and trace element composition. Chemosphere 119, 1447–1455.

Cao, T., Wang, M., An, L., Yu, Y., Lou, Y., Guo, S., Zuo, B., 2009. Air quality for metals and sulfur in Shanghai, China, determined with moss bags. Environ. Pollut. 157, 1270–1278.

Culicov, O.A., Yurukova, L., 2006. Comparison of element accumulation of different moss and lichen-bags, exposed in the city of Sofia (Bulgaria). J. Atmos. Chem. 55, 1–12.

Directive 2004/107/EC of the European parliament and of the council of 15 December 2004 relating to arsenic, cadmium, mercury, nickel and polycyclic aromatic hydrocarbons in ambient air, Official Journal of the European Union, 23, 1–16, (26/01/2005).

Directive 2008/50/EC of the European parliament and of the council of 21 May 2008 on ambient air and cleaner air for Europe, Official Journal of the European Union, 1–44, (11/06/2008).

Dołęgowska, S., Migaszewski, Z.M., 2013. Anomalous concentrations of rare earth elements in the moss–soil system from south-central Poland. Environ. Pollut. 178, 33–40.

EMEP/EEA air pollutant emission inventory guidebook, 2013. Passenger cars, light commercial trucks, heavy-duty vehicles including buses and motor cycles 2013, update September, 2014 http://www.eea.europa.eu/publications/emep-eea-guidebook-2013/.

European Environment Agency, 2013. The Contribution of the Transport Sector to Total Emissions of the Main Air Pollutants in 2009 (EEA-32). Retrieved March 16, 2013, from. http://www.eea.europa.eu/data-and-maps/figures/thecontribution-of-the-transport-1.

Gietl, K.J., Lawrence, R., Thorpe, J.A., Harrison, M.J., 2010. Identification of brake wear particles and derivation of a quantitative tracer for brake dust at a major road. Atmos. Environ. 44, 141–146.

Giordano, S., Adamo, P., Monaci, F., Pittao, E., Tretiach, M., Bargagli, R., 2009. Bags with oven-dried moss for the active monitoring of airborne trace elements in urban areas. Environ. Pollut. 157, 2798–2805.

Goodman, T.G., Roberts, T.M., 1971. Plants and soils as indicators of metals in the air. Nature 231, 287–292.

Hagler, G.S.W., Baldauf, R.W., Thoma, E.D., Long, T.R., Snow, R.F., Kinsey, J.S., Oudejans, L., Gullett, B.K., 2009. Ultrafine particles near a major roadway in Raleigh, north Carolina: downwind attenuation and correlation with traffic-related pollutants. Atmos. Environ. 43, 1229–1234.

Karner, A.A., Eisinger, D.S., Niemeier, D.A., 2010. Near-roadway air quality: synthesizing the findings from real-world data. Environ. Sci. Technol. 44, 5334–5344.

Kosior, G., Ciężka, M., Górka, M., Samecka-Cymerman, A., Kolon, K., Kempers, A.J., Jędrysek, M.-O., 2015. δ34S values and S concentrations in native and transplanted *Pleurozium schreberi* in a heavily industrialised area. Ecotoxicol. Environ. Saf. 118, 112–117.

Legendre P. Package "lmodel 2", R package version 1.7–2, 2014, http://cran.rproject.org/web/packages/lmodel2/lmodel2.pdf.

Mijić, Z., Stojić, A., Perišić, M., Rajšić, S., Tasić, M., Radenković, M., Joksić, J., 2010. Seasonal variability and source apportionment of metals in the atmospheric deposition in Belgrade. Atmos. Environ. 44, 3630–3637.

Napier, F., D'Arcy, B., Jefferies, C., 2008. A review of vehicle related metals and polycyclic aromatic hydrocarbons in the UK environment. Desalination 226, 143–150.

Republic Hydrometeorological Service of Serbia (RHMSS), 2014 http://www.hidmet.gov.rs/index_eng.php.

Rivera, M., Zechmeister, H., Medina-Ramón, M., Basagaña, X., Foraster, M., Bouso, L., Moreno, T., 2011. Monitoring of heavy metal concentrations in home outdoor air using moss bags. Environ. Pollut. 159 (4), 954–962.

Salo, H., Mäkinen, J., 2014. Magnetic biomonitoring by moss bags for industry-derived air pollution in SW Finland. Atmos. Environ. 97, 19–27.

Song, F., Gao, Y., 2011. Size distributions of trace elements associated with ambient particular matter in the affinity of a major highway in the New Jersey New York metropolitan area. Atmos. Environ. 45 (37), 6714–6723.

Steinnes, E., Rühling, A., Lippo, H., Mäkinen, A., 1997. Reference materials for large scale metal deposition surveys. Accred. Qual. Assur. 2, 243–249.

Thorpe, A., Harrison, R., 2008. Sources and properties of non-exhaust particulate matter from road traffic: a review. Sci. Total Environ. 400, 270–282.

Tsai, J.H., Chiang, H.L., Hsu, Y.C., Weng, H.C., Yang, C.Y., 2003. The speciation of volatile organic compounds (VOCs) from motorcycle engine exhaust at different driving modes. Atmos. Environ. 37, 2485–2496.

United States Environmental Protection Agency, 2013. National Summary of Nitrogen Oxides emissions, NEI 2008. Retrieved April 1 6, 2013, from. http://www.epa.gov/cgi-bin/broker?polchoice¼.0NOX&_debug¼0&_service¼data&_program¼dataprog.

Vasconcelos, M.T.S.D., Tavares, H.M.F., 1998. Atmospheric metal pollution (Cr, Cu, Fe, Mn, Ni, Pb and Zn) in Oporto city derived from results for low-volume aerosol samplers and for the moss *Sphagnum auriculatum* bioindicator. Sci. Total Environ. 212, 11–20.

Vuković, G., Aničić Urošević, M., Razumenić, I., Goryainova, Z., Frontasyeva, M., Tomašević, M., Popović, A., 2013. Active moss biomonitoring of small-scale spatial distribution of airborne major and trace elements in the Belgrade urban area. Environ. Pollut. Res. 20, 5461–5470.

Vuković, G., Aničić Urošević, M., Goryainova, Z., Pergal, M., Škrivanj, S., Samson, R., Popović, A., 2015a. Active moss biomonitoring for extensive screening of urban air pollution: magnetic and chemical analyses. Sci. Total Environ. 521–522, 200–210.

Vuković, G., Aničić Urošević, M., Pergal, M., Janković, M., Goryainova, Z., Tomašević, M., Popović, A., 2015b. Residential heating contribution to persistent air pollutants (PAHs, trace and rare earth elements): a moss bag case study. Environ. Sci. Pollut. Res. http://dx.doi.org/10.1007/s11356-015-5096-0.

World Health Organisation, 2013. Review of Evidence on Health Aspects of Air Pollution REVIHAAP Project. Technical Report. World Health Organization, Regional Office for Europe, Copenhagen, Denmark.

Wilson, J.G., Kingham, S., Pearce, J., Sturman, A.P., 2005. A review of intraurban variations in particulate air pollution: implications for epidemiological research. Atmos. Environ. 39, 6444–6462.

Zechmeister, H.G., Hohenwallner, D., Riss, A., Hanus-Illnar, A., 2005. Estimation of element deposition derived from road traffic sources by using mosses. Environ. Pollut. 138, 238–249.

Zechmeister, H.G., Hagendorfer, H., Hohenwallner, D., Hanus-Illnar, A., Riss, A., 2006a. Analyses of platinum group elements in mosses as indicators of road traffic emissions in Austria. Atmos. Environ. 40, 7720–7732.

Zechmeister, H.G., Dullinger, S., Hohenwallner, D., Riss, A., Hanus-Illnar, A., Scharf, S., 2006b. Pilot study on road traffic emissions (PAHs, heavy metals) measured by using mosses in a tunnel experiment in Vienna, Austria. Environ. Sci. Pollut. Res. 13 (6), 398–405.

PHYSICAL CHEMISTRY 2016

13th International Conference on
Fundamental and Applied Aspects of
Physical Chemistry

Proceedings
Volume II

BELGRADE
September 26-30, 2016

ISBN 978-86-82475-33-0
Title: Physical Chemistry 2016 (Proceedings)
Editors: Željko Čupić and Slobodan Anić
Published by: Society of Physical Chemists of Serbia, Studentski trg 12-16,
11158, Belgrade, Serbia.
Publisher: Society of Physical Chemists of Serbia
For Publisher: Slobodan Anić, President of Society of Physical Chemists of Serbia
Printed by: "Jovan", Printing and Publishing Company; 200 Copies
Number of pages: 3+434; Format B5; printing finished in September 2016

Text and Layout: "Jovan"

200 - *Copy printing*

CONTENT

Volume II

SURVEY OF POTENTIALLY TOXIC ELEMENT POLLUTION OF THE VINEYARD SOIL

T. Milićević[1], D. Relić[2], G. Vuković[1], M. Perišić[1], D. Majstorović[3],
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ABSTRACT

In this paper we presented assessment of the potentially toxic elements in the vineyard topsoil (0–5 cm) near the main route in region “Oplenačko vinogorje”. We presented concentrations of potentially toxic elements (Cd, Co, Cr, Cu, Ni, Pb, Sb, V and Zn) and pH values determined in deionized H_2O, 1 M KCl and 0.01 M $CaCl_2$. Concentrations of Cr (89–280 µg g^{-1}) and Ni (59–220 µg g^{-1}) were determined in values higher than maximum prescribed concentration by regulative. In addition, the high concentration of other elements (Co, Cu, Sb and Zn)points to the necessity of performing more frequently monitoring of potentially toxic elements in the vineyard soil. Significant correlations were obtained between concentrations of Cr–Ni–Sb, but also significant dependence of Zn concentration on pH values was determined.

INTRODUCTION

Vineyard soil has influences on human health via agricultural production. Foodborne diseases have a significant impact on human health [1]. The increasing release of agrochemicals into the environment has led to growing public concern over the potential accumulation of pollutants (e.g. heavy metals) within agricultural soil.

One of the most important steps in monitoring content of potentially toxic elements in soil is to determine whether the total contents of potentially toxic elements are within the range or over the concentration prescribed by national and international legislation. Therefore, there is a clear interest for monitoring of major and trace elements in agricultural soil, especially those recognized as toxic or carcinogenic.

Soil acidity (key phisico-chemical parameter for determine soil bioavailability and mobility fractions) was measured to determine potentially toxic element concentration relation to soil acidity.

The aims of this study were to determine concentrations of potentially toxic elements, their positive correlations and the topsoil acidity (pH).

EXPERIMENTAL

The “Oplenac” Wine Route is well known region for vine growing and production, 80 km away from Belgrade, a capital of Serbia. The topsoil (0–5 cm) samples were collected during grapevine season of 2015. Twenty-three sampling sites from the vineyard parcels along main route (1st parcel x 5 sites, 2nd parcel x 2 sites, 3rd parcel x 2 sites, 4th parcel x 5 sites, 5th parcel x 4 sites, 6th parcel x 4 sites), and one control sample on the meadow in the vicinity of vineyard parcels.

The soil samples were air dried, grinded in a mortar with a pestleand, and sieved through 2 mm stainless sieve. The hygroscopic moisture of each soil sample was determined at 105°C.The aqua regia digestions were performed in microwave oven (ETHOS 1, Start E Microwave Extraction System, Milestone, Italy) in sealed PTFE vessels using 9 mL HCl and 3 mL HNO_3 for 0.5 g of each soil sample [2,3].

Using inductively coupled plasma optical emission spectrometry (ICP–OES), 25 elements in the soil samples were determined. In this paper we only pay attention to potentially toxic elements (Cd, Co, Cr, Cu, Ni, Pb, Sb, V and Zn) which could have negative consequences for humans and environment.

The pH of soil samples was determined in mixture of (1:5) soil–distilled H_2O, soil–1M KCl, and soil–0.01M $CaCl_2$. Acidity (pH) of soil samples were determined using glass electrode.

For data analysis SPSS software version 21 for Windows and Statistic8 (Stat Soft Inc., Tulsa, OK, USA) were used. Correlation analysis andprincipal component analysis (PCA) were used to identify specific correlations.Normality of the data was tested by the Kolmogorov–Smirnov test for each element test. Distributions of all determined elements were normal.

RESULTS AND DISCUSSION

In the vineyard soil samples, determined concentrations of Cr and Ni (Table 1)were above the maximum allowed concentrations proscribed by national Serbian regulative (100 $\mu g\ g^{-1}$, 50 $\mu g\ g^{-1}$, respectively) [4,5].The highest correlation coefficients were determining between Cr, Ni and Sb (Cr–Ni: $R=0.951$, $p<0.01$; Cr–Sb: $R=0.935$, $p<0.01$; Ni–Sb: $R=0.887$, $p<0.01$). This indicates that these toxic heavy metals could originate from same source,probably the nearness traffic route. These elements in the found concentrations could contaminate grapevine and human health in this environment.

Table 1. Descriptive statistic for the element content (μg g^{-1}) and pH in soil samples (N=23):

	Mean	Min.	Max.	SD
Cd	2.1	1.9	2.3	0.2
Co	26	21	34	3.4
Cr	150	89	280	40
Cu	51	36	110	15
Ni	110	59	220	37
Pb	21	17	27	3.0
Sb	3.3	2.4	4.7	0.6
V	64	58	71	2.9
Zn	93	69	157	17
pH H_2O	7.3	6.7	7.9	0.4
pH KCl	6.5	5.2	6.9	0.4
pH $CaCl_2$	6.7	5.5	7.1	0.4

Mobility and bioavailability of the elements in soil largely depends on soil acidity. The pH values measured in the soil sample was 5.48–7.06 in 1M KCl, 6.7–7.9 in H_2O, and 5.5–7.1 in $CaCl_2$. According to pH values, these soil samples were moderately acid to neutral, in the soil solution (the most bioavailable fraction) they were neutral to alkaline, and in mineral fraction sampleswere moderately acid to neutral (Fig 1).

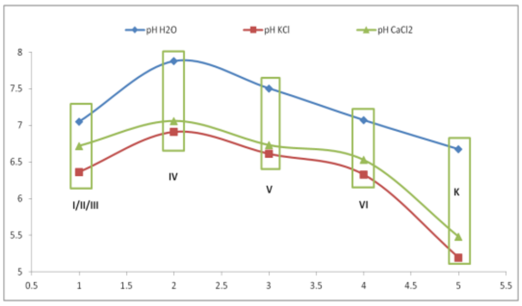

Figure 1. Determined soil pH values in 6 parcels and control location

According to PCA analysis the most correlated concentrations of Cr–Ni–Sb weregrouped in the same quadrant. In addition, determined concentrations of Zn were in significant relationship with determined pH values (Fig 2).

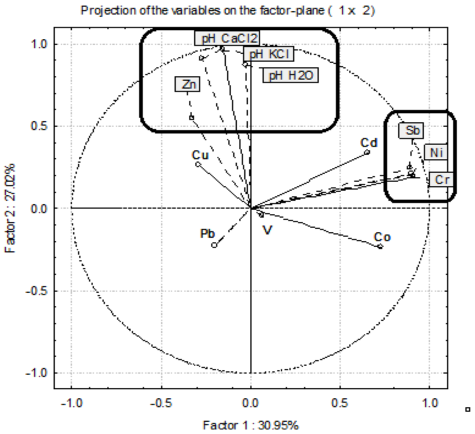

Figure 2. PCA of the determined elements, and pH values

CONCLUSION

In addition, according to national regulations and proscribed maximum allow concentrations in soils it could be concluded that beside concentrations of Cr and Ni, monitoring of Co, Cu, Sb and Zn concentrations should be implemented frequently in this vineyard region. In addition, it could be concluded that determined concentration of Zn in these vineyard soil samples widely depends of pH value.

Acknowledgement

The authors are grateful to the Ministry of Education and Science of the Republic of Serbia (Project No. III 43007, 172001) and to winery "Aleksandrović", Vinča, Topola.

REFERENCES

[1] World Health Organization (WHO), http://www.who.int/topics/food_safety/en/, accessed on 25th May, 2016.

[2] H. Ronkkomaki, R. Poykio, H. Nurmesniemi, K. Popov, E. Merisalu, T. Tuomi, I. Valimaki, Int. J. Environ. Sci. Technol., 2008, **5,** 485 - 494.

[3] US EPA 3050b Method, Acid digestion of sediments, sledges, and soils, USEPA, Washington, DC, USA, 1996.

[4] National Regulation, Republic of Serbia 88/2010.

[5] EU Council Directive 86/278/EEC.

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GRAPEVINE ACCUMULATION OF POTENTIALLY TOXIC ELEMENTS FROM VINEYARD SOIL

Tijana Milićević[1], D. Relić[2], G. Vuković[1], S. Škrivanj[2], A. Popović[2], M. Aničić Urošević[1]

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Keywords: bioavailability, grape seed, grape pulp, grape skin, grape leaf

Introduction

Nowadays, foodborne diseases have a major impact on human health. Xenobiotics are a significant cause of foodborne illness, although effects are often difficult to link with a particular food or agricultural area. Contaminants in agricultural soil and plants usually include fertilizers and pesticides (WHO). One of the most important steps in preventing contamination of agricultural products is monitoring of major and trace element content, which present in excess could be toxic. Variations of the physico-chemical conditions of major and trace elements as well as soil can influence the release of these elements (Filgueiras et al., 2002). Trace metals and other elements in soils and plants may affect human health through the inhalation of dust, ingestion of soil, dermal contact, or consuming products from agricultural field contaminated by trace element (Morel et al., 1997; Kabata - Pendias and Pendias, 2001). This experiment was performed to investigate bioavailability fractions from vineyard soil, and accumulation in different parts of grapevine (seed, skin, pulp and leaf).

Methods

The "Oplenac" Wine Route is well known region for vine growing, in the vicinity of "Topola" city, 80 kilometers away from Belgrade, the capital of Serbia. The topsoil (0-30 cm), grape leaves and grapevine samples were collected from 26 sampling sites in the vineyard, during grapevine season of 2015. Bioavailable elements from the soil samples were extracted with four different single extraction procedures: 0.11 mol L^{-1} CH_3COOH during 16 h extraction, 0.01 mol L^{-1} $CaCl_2$ during 3 h extraction, 1 mol L^{-1} NH_4NO_3 during 2 h extraction and distilled water during 2 h (Quevauviller, 1998) and 16 h. Distilled water extraction of 16 h was tested as an alternative single extraction process for toxic elements from vineyard soils. Leaf and grape samples were digested in a microwave digester, with 1 mL of 30 % H_2O_2 and 7 mL of 65 % HNO_3. Eight potentially toxic elements (Al, B, Cu, Fe, Mn, Ni, Sr and Zn) were determined using inductively coupled plasma optical emission spectrometry, ICP-OES.

Results

For all extractants the most significant correlations between concentrations of Fe and Al, were established. These two elements originate from soil clay minerals, i.e., Fe and Al oxides. The most significant correlations were determined between concentrations of Ni and Mn extracted with $CaCl_2$ ($R=0.965$, $p<0.01$)

and NH_4NO_3 (R=0.964, $p<0.01$). Significant correlation between concentration of Sr and Mn (R=0.896, $p<0.01$) were determined during extraction with CH_3COOH. However, the most pairs of significant correlations between the pairs of elements, e.g., Zn-Mn (R=0.776, $p<0.01$) or Zn-Ni (R=0.0826, $p<0.01$) were investigated during extraction procedure with NH_4NO_3. This weak salt extracted many pairs of element, which could be connected with metal containing agro-chemicals used in a conventional vineyard treatment. Considering all positive correlations, significantly positive correlations could be pointed out between concentrations of Cu-Mn, Cu-Ni and Cu-Zn just in case of the soil extraction with distilled water during 16 h. Thus, this single procedure could be a good alternative for investigation of Cu content in vineyard soils.

A unique extractant could not be best solution for determination of all bioavailable elements, and for determination of major and trace element transport in soil-plant. However, judging by correlation and PCA analysis, concentrations of Mn and Sr extracted from soil samples with $CaCl_2$ and NH_4NO_3 showed significant correlation with the same element concentrations isolated from the grape leaves (Mn_{CaCl2}-Mn_{leaf}: R=0.663, $p<0.01$; Sr_{CaCl2}-Sr_{leaf}: R=0.798, $p<0.01$; Mn_{NH4NO3}-Mn_{leaf}: R=0.664, $p<0.01$; Sr_{NH4NO3}-Sr_{leaf}: R=0.642, $p<0.01$). Furthermore, concentration of Sr extracted from the soil with $CaCl_2$, NH_4NO_3 and distilled water during 2 h were in correlation with concentration of Sr isolated from the grape skin (Sr_{CaCl2}-Sr_{skin}: R=0.863, $p<0.01$; Sr_{NH4NO3}-Sr_{skin}: R=0.646, $p<0.01$; $Sr_{2h\ H2O}$-Sr_{skin}: R=0.544, $p<0.01$). The highest significant correlation was determined between concentration of Sr extracted with CH_3COOH and Sr isolated from the grape pulp (R=0.940, $p<0.01$), also CH_3COOH extracted concentration of Zn which was in correlation with Zn isolated from the grape skin (R=0.528, $p<0.01$). On the other hand, concentrations of Cu, Fe and Sr isolated from the grape skin were in correlation with Cu, Fe and Sr isolated from the grape leaves.

Conclusion

It could be concluded that NH_4NO_3 extractant was shown ability to isolate a pairs of potentially toxic elements, which could originate from the fertilizers or pesticides used in agriculture production. Extraction with distilled water during 16 h could be investigated as an alternative procedure for extracting some potentially toxic elements from vineyard soils. There was not unique extractant which could be best solution for determination all bioavailable elements, and for determination of accumulation of major and trace elements.

References

World Health Organization (WHO), http://www.who.int/topics/food_safety/en/, accessed on 10th April, 2016.

Filgueiras, A. V.; Lavilla,I ., Bendicho, C. (2002). Chemical sequential extraction for metal partitioning in environmental solid samples. *J. Environ. Monit.*, *4*, 823 - 857.

Quevauviller, Ph. (1998). Operationally defined extraction procedures for soil and sediment analysis, II. Certified reference materials. *Trends Anal. Chem.*, *17*, 632-642.

Kabata-Pendias A.; Kabata H. (2001). Trace elements in Soils and Plants, *CRC Press*, USA.

Morel, J.L. (1997). Bioavailability of Trace Elements to Terrestrial Plants. In: Tarradellas, J.; Bitton, D. (Eds.) Soil Ecotoxicology. CRC Press, USA

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MOSS BAG BIOMONITORING OF AIRBORNE TOXIC ELEMENT DECREASE ON A SMALL SCALE: CROSSROAD AND TWO- AND ONE-LANE STREET STUDY

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The use of "moss bag technique" has been developing in last several decades as a method of active monitoring of air pollutants. This method has been particularly useful for conducting a detailed survey of diversely polluted microenvironments within urban areas, where native mosses are usually absent because of predominantly paved and landscaped surfaces. In this study, *Sphagnum girgensohnii* moss bags were exposed at 48 crossroads, two- and one-lane streets across the city of Belgrade (Serbia) for 10 weeks during the summer of 2014. During the experimental period, traffic flows were estimated at each site by vehicles counting during the rush hours. The concentrations of 39 elements were determined in the moss samples by inductively coupled plasma – mass spectrometry (ICP-MS). For the majority of elements, the moss bags identified a common pattern of decrease in the element concentration from the crossroad to the one-lane street, especially for the most enriched elements – Sb, Cu, and Cr (Figure). A significant correlation (r = 0.65 – 0.70) between the moss concentration of Sb, Cu, and Cr, and traffic flows makes these elements reliable traffic tracers [1].

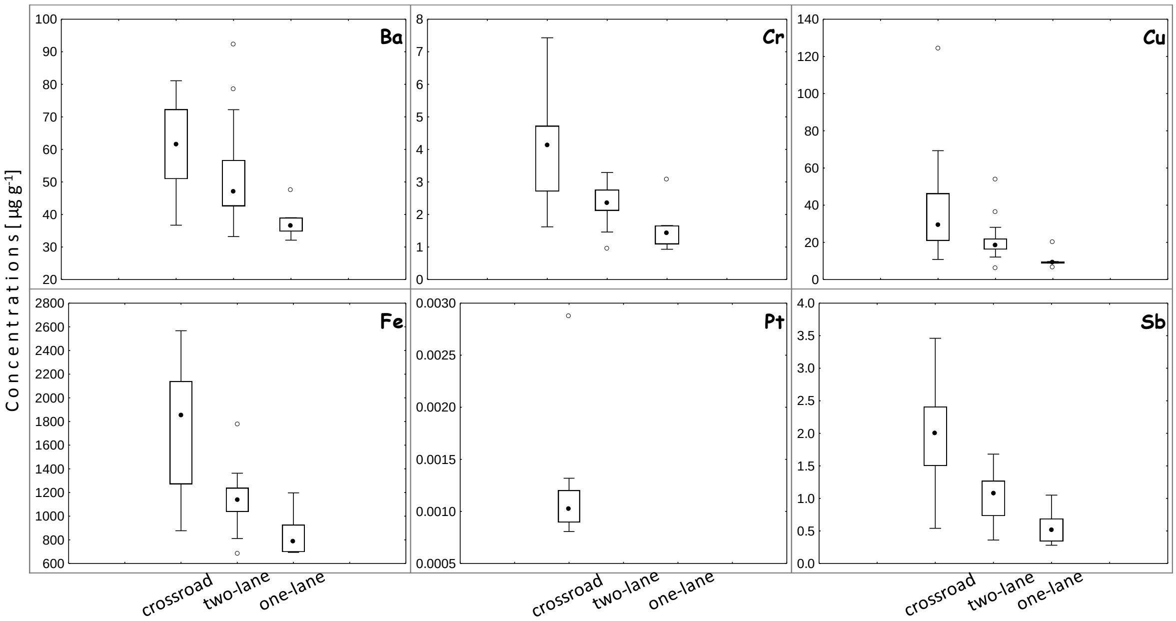
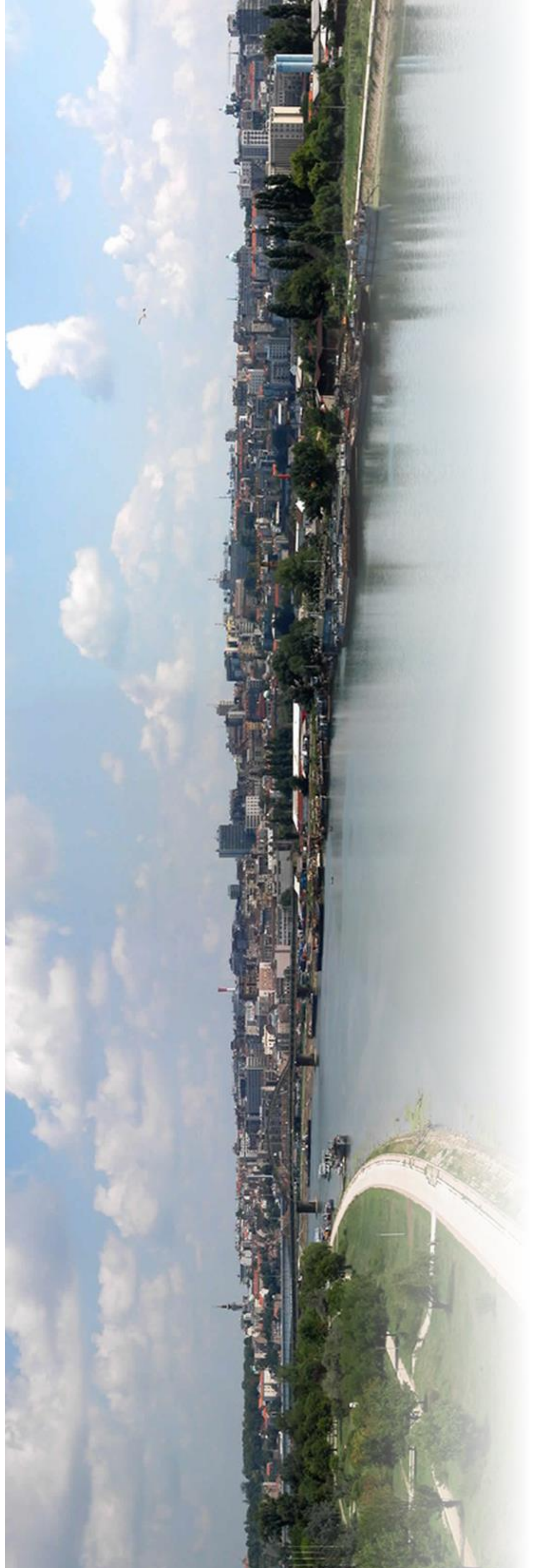


Figure. Element concentrations (µg g^{-1}) in the *S. girgensohnii* moss bags;
* Pt was only detected at the crossroads

1. Vuković G., Aničić Urošević M., Škrivanj S., Milićević T., Dimitrijević D., Tomašević M. and Popović A.: Moss bag biomonitoring of airborne toxic element decrease on a small scale: A street study in Belgrade, Serbia, Science of the Total Environment 542 (2016) 394–403.



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PARTICULATE MATTER AND REGULATORY ISSUES	• *issues related to monitoring of particulate matter* • *legislative aspects* • *abatement strategies*

2.8 UPGRADE OF MICRO-SCALE SITING OF AIRBORNE TOXIC ELEMENTS BY MOSS BAG TECHNIQUE: CROSSROAD, TWO- AND ONE-LANE STREET STUDY

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ABSTRACT

Database of potentially hazardous substances, necessary for estimation of human exposure to air pollutants, is principally deficient because of a limited number of regulatory monitoring stations. In this study, the moss bags were exposed at 48 crossroads, two- and one-lane streets across the city of Belgrade (Serbia) for 10 weeks during the summer of 2014. During the experimental period, traffic flows were estimated at each site by vehicles counting during the rush hours. The concentrations of 39 elements were determined in the moss samples. The most enriched elements were Sb, Cu, and Cr. For the majority of elements, the moss bags identified a common pattern of decrease in the concentration from the crossroad to the one-lane street. A significant correlation (*r* = 0.65 – 0.70) between the moss concentration of Sb, Cu, and Cr, and traffic flows makes these elements reliable traffic tracers.

INTRODUCTION

Transport sector represents dominant source of air pollution in urban areas. Traffic-related air pollutants, that include particulate matter (PM) associated with toxic elements, are widespread. These pollutants can often be locally-elevated close to urban microenvironments such as major roads, crossroads, junctions, and bus stations. Exceedances in the limit values of the air pollutants have been associated with many hazardous health effects (e.g., EEA, 2013). Epidemiological studies commonly utilise data from one or few stations of regulatory monitoring network to estimate personal exposure to PM air pollution (Brauer, 2010). However, recent research has indicated that monitoring stations may not accurately characterize complexities of the PM spatial dispersion across an urban area (Brauer, 2010). Regulatory network of air quality monitoring have been typically designed to assess regional and temporal air pollution variability. Specifically, for micro-scale siting, sampling sites should be placed at least 25 m from the edge of crossroads and not on the carriageways of roads (Directive 2004/104/EC, Directive 2008/50/EC). Thus, testing of long-term on-road exposure of pedestrians, commuters, cyclists, and workers (e.g., drivers, policemen) to traffic-related toxic elements requires alternative approaches (Brauer, 2010).

Over the past several decades, biomonitoring has been developed as a valuable tool for assessing environmental pollution. Biomonitoring represents usage of living organisms (plants, animals) as a measure of environmental quality. Among the terrestrial organisms employed as plant biomonitors, mosses have been proven as reliable biomonitors of air quality due to their morpho-physiological characteristic. Moss lacks a developed root system and thus obtains nutrients from air. In addition, a high cation-exchange capacity of moss increases adsorbent efficiency (Brown and Bates, 1990). Between the two types of moss biomonitoring, passive (using naturally growing mosses in a certain area) and active (using moss transplants), the active approach has been applied for studies in anthropogenically devastated areas, such as industrial and urban, where naturally growing mosses are scarce or even absent. Since the early work of Goodman and Roberts (1971), the “moss bag technique” was introduced as a method of active moss biomonitoring. The technique involves exposure of anthropogenically unpolluted moss material within mesh bags for monitoring of the presence of pollutants in ambient air, and has been tested in different environments (Aničić et al, 2009a; Ares et al, 2012; Vuković et al, 2013). The moss bag technique gives a reliable picture of pollution patterns in a much more cost-effective way than regulatory monitoring devices, which could not provide dense monitoring network, and also require power supply and maintenance. It also integrates air pollutant levels over longer period than most of instrumental measurements do. Long-period sampling is prerequisite for assessment of cumulative exposure to a certain pollutant, which has hazardous impact on human health.

The purpose of this study was to assess a level of airborne toxic elements close to highly traffic-burdened crossroads, two- and one-lane streets using the moss bag technique. We hypothesized that distance-decline of the toxic element concentrations exists from crossroads to two- and one-lane streets depending on the traffic burden. The aim was also to assess if the moss bags could reflect this phenomenon.

METHODOLOGY

Moss Bag Preparation

The moss *Sphagnum girgensohnii* Russow was collected from a pristine wetland area located near Dubna, Russia. This area is pronounced to be an appropriate background site in our previous research (Aničić et al, 2009a; 2009b). In the laboratory, the green apical parts of the moss was separated from the rest brown tissue and carefully manually cleaned of soil particles, plant remains, and epiphytes. Subsequently, it was rinsed three times with double-distilled water. Such prepared moss was air-dried and gently hand-mixed to obtain homogeneous material. Approximately 1.5 g of the moss was packed loosely in 7 × 7 cm nylon net bags with 2-mm mesh size. For determination of the moss element content before the bag exposure (initial concentrations), several moss bags were stored at room temperature in the laboratory conditions.

Experimental Set-up and Traffic Flow Estimation

The moss bags were exposed for 10 weeks using plasticized aluminium holders, specifically designed for this purpose. The holders were mounted perpendicular to the lampposts, at open space, at the representative height of 3 – 4 m (Vuković et al, 2013). The holders with moss bags were exposed at 48 sampling sites across the city of Belgrade urban area (44^0 50'- $44^0$44'N, and $20^0$22' - $20^0$32'E at 70 - 250 m altitude) from June 15th to August 15th, 2014. The sets of 3 sampling sites – crossroad, two- and one-lane street, away from each other approximately 300 m, were selected. Three pedestrian zones were chosen to represent urban background sites. The sites were selected in different parts of the city: urban-central and peri-urban zone, and within an urban forest. In addition, for each study site, traffic flows were recorded by video cameras for later off-site counting. The vehicle fleet were classified into categories of passenger cars, buses, trams, trolleys, motorcycles, light and heavy duty vehicles. Traffic flows of each vehicle category were counted for 15 minutes during the rush hours (7:00 – 9:00 am and 4:00 – 6:00 pm) on Wednesday and Sunday in June and July. The average traffic flows during the experimental period were estimated from these counts, using a recommended procedure by the Secretariat for Transport of Belgrade (*personal communication*).

Chemical Analyses

After exposure period, the moss samples were air-dried and homogenised. Approximately 0.3 g of each moss sample (3 subsamples per exposure site) was digested for 45 min in a microwave digester (ETHOS 1, Advanced Microwave Digestion System, Milestone, Italy), at 200°C, with 1 mL of 30% H_2O_2 (Sigma Aldrich, puriss. p.a.) and 7 mL of 65% HNO_3 (Sigma Aldrich, puriss. p.a., distilled by the apparatus for acid distillation – BERGHOF, Products+ Instruments GmbH, Germany). Digested samples were diluted with double-distilled water to a total volume of 50 mL. The concentrations of Al, B, Ba, Cu, Fe, and Zn were determined using inductively coupled plasma-optical emission spectroscopy, ICP-OES (Thermo Scientific iCAP 6500 Duo, Thermo Scientific, UK). The concentrations of As, Cd, Ce, Co, Cr, Dy, Er, Eu, Ga, Gd, Ho, La, Li, Lu, Nd, Ni, Pb, Pr, Pt, Rb, Sb, Sc, Sm, Sn, Sr, Tb, Th, Ti, Tl, Tm, V, Y, and Yb were determined using inductively coupled plasma-mass spectrometry, ICP-MS (Thermo Scientific iCAP Q, Thermo Scientific, UK). To control the quality of the element analysis, the control samples, certified reference materials and analytical blanks, were analysed once every 10 samples. As the certified reference materials, the moss *Pleurozium schreberi*, M2 and M3 (Steinnes et al, 1997) was used.

Data Processing

To assess the element enrichment of the studied moss species, relative accumulation factor (RAF) was calculated based on the moss content of each element after exposure ($C_{exposed}$) reduced and then divided by the element content before moss exposure ($C_{initial}$):

$$RAF = (C_{exposed} - C_{initial})/C_{initial}$$

The data were processed using STATISTICA 8.0 (StatSoft, Inc., Tulsa, OK, USA). The normality of the data was tested by Shapiro-Wilk test ($p < 0.05$). Spearman's correlation was applied ($p < 0.05$) to determine the correlation between the moss elemental concentrations and the traffic flows. To represent graphically abundances of the rare earth elements (REE), concentrations of the REE observed in the moss samples were normalised to those in different standards North American Shale Composite (NASC) and Post-Archean Australian Shales (PAAS) (Dołęgowska and Migaszewski, 2013).

RESULTS AND DISCUSSION

Spatial Gradient of Moss Element Content: Crossroad, Two- and One-Lane Street

Moss bag technique enables a longer sampling period of airborne pollutants such as toxic elements and better spatio-temporal resolution of sampling sites. These advantages could provide characterization of long-term human exposure to toxic elements, which is more accurate and relevant in the assessment of chronic health effects than daily instrumental measurements. In this study, the concentrations of the most elements in the exposed moss bags of *S. girgensohnii* were above the initial element concentrations in the moss. Markedly high RAF values of Sb, Sn, Sm, Cu, Cr, and Zn (Table 1) in the moss samples imply significantly elevated concentrations of these elements within on-road microenvironments. Moreover, at the crossroads, Pt, which naturally occurs in very low concentrations in mosses, was markedly enriched. Thus far, concerning the moss bag technique application, Pt has only been observed in the moss bags exposed within a road tunnel (Zechmeister et al, 2006) and very scarcely in ambient air in the proximity of bus lines (Rivera et al, 2011).

Table 1. *RAF values in the S. girgensohnii moss bags exposed at sites with different traffic intensity: crossroad, two- and one-lane street, for 10 weeks*

RAF	Element
0 - 5	Al, B, Ba, Co, Dy, Mg, Sr, Tl, Y
5 - 10	As, Cd, Ce, Er, Eu, Fe, Ga, Gd, Ho, La, Lu, Nd, Ni, Pb, Pr, Pt, Sc, Tb, Ti, Tm, Yb
10 - 30	Th, V, Cr, Zn
> 30	Cu, Sb, Sm, Sn

The *S. girgensohnii* moss bags indicated spatial variability in the concentrations of the majority of determined elements. Particularly, concentration gradient of As, Ba, Co, Cu, Cr, Ni, Sb, and V in the moss samples (Fig 1) clearly emphasizes the crossroads as pollution hotspots where pedestrians and commuters are likely to be exposed to highly increased level of toxic elements. At crossroads, a pattern of braking and accelerating driving mode, so called “stop and go”, is especially conspicuous. In comparison to constant free-flowing traffic mode, which characterised two- and one-lane streets, “stop and go” mode leads to elevated pollutant emissions from both, exhaust and non-exhaust traffic source (Tsai et al, 2003).

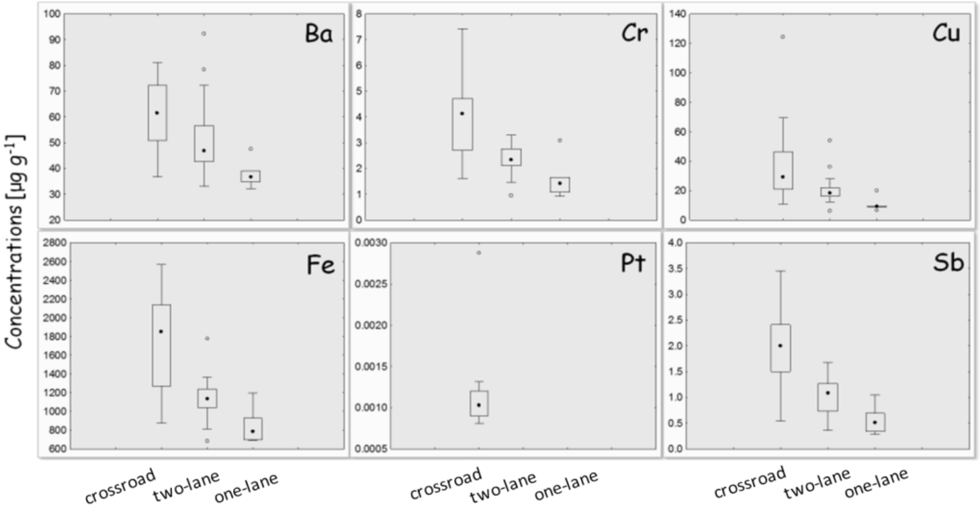

Figure 1. *Concentrations (µg g^{-1}) of Ba, Cr, Cu, Fe, Pt, and Sb in the S. girgensohnii moss bags; * Pt was only detected at the crossroads*

The decrease in the moss shale-normalized concentration of the REE (Figure 2) confirms spatial dependence of these elements on anthropogenic activities, i.e., vehicle movements. Similar order of abundance of the REE was identified, just as it has been pointed out in our previous study (Vuković et al, 2015b). This confirms consistent signature of REE in urban area.

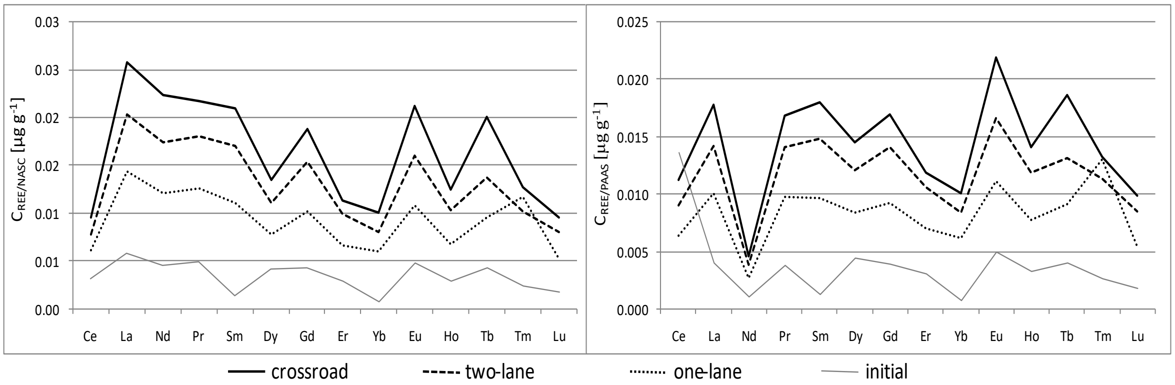

Figure 2. *The median concentrations (µg g^{-1}) of the REE normalized to NASC and PAAS in the S. girgensohnii moss bags exposed at sites with different traffic flows: crossroad, two- and one-lane street; initial – element concentrations in the unexposed moss; the REE order on the x-axis is chosen according to their abundances in the NASC and PAAS.*

The results draw attention for better spatial resolution of regulatory monitoring sites from which data are usually used in epidemiological studies. Note that permitted levels of emission of Cu, Sb, Cr, Pt and rare earth elements such as Sm, in the air has not been regulated yet by legislation (Directive 2004/107/EC), although these elements are recognized as toxic by WHO (2013).

Correlation between Moss Element Concentration and Traffic Flow

Based on the high correlation coefficients ($r > 0.80$) among Ba, Cr, Cu, Fe, Ni, Sb, and V in the moss samples, the common sources of these elements can be assumed. The vehicle movements cause: (i) exhaust emissions from the tailpipe; (ii) non-exhaust emissions due to wear and tear of vehicle parts such as brake, tyre and clutch; and (iii) re-suspension of road dust. Non-exhaust emissions mainly contribute to increased concentrations of Ba, Fe, Cu, and Sb (Gietl et al, 2010) while fuel combustion is the major source of Cr, Ni, V, and Cu (EMEP/EEA, 2013). Given the moderate, but significant correlation coefficient ($r = 0.65 – 0.70$, $p < 0.05$) between the total traffic flows and the concentrations of Cr, Cu, Fe, and Sb in the moss bag samples, these elements could be considered as reliable tracers of traffic-related emissions. Our results support previous findings from the road traffic studies using both terrestrial mosses and moss bags biomonitors (Zechmeister et al, 2005) in which Cr, Cu, Fe, and Sb have been identified to be traffic-originated. Regarding the categories of vehicles, the significant correlation coefficient ($r > 0.60$, $p < 0.05$) implies tight relation between the flows of passenger cars and light duty vehicles, and the moss element concentrations of Cr, Cu, Fe, and Sb. This is in agreement with the results previously pronounced for the correlation between the flows of light duty vehicles and concentrations of Cu and Fe in the moss samples (Adamo et al, 2011). Zinc has often been reported as one of the tracers for non-exhaust traffic emissions, e.g., tire-wear emissions of Zn are shown to be approximately 1000 and 500 times higher than those of Pb and Cu, respectively (Napier et al, 2008). Nevertheless, in this study, very low correlation coefficients ($r < 0.20$) were obtained between the moss concentrations of Zn and traffic flows. A distinct spatial variability, which is dependent on traffic flow, was not observed. In addition, in our previous studies (Vuković et al, 2013; Vuković et al, 2015a), there was no regularity in the Zn spatial distribution, related to traffic flow at the study sites. Considering the high RAF values obtained for Zn in this study along with the absence of any other pollution source except traffic, the Zn origin could be assumed as traffic-related. However, Zn could not be considered as reliable tracer of traffic flow variability within urban area.

Could Pedestrian Zones Be Considered as an Urban Background?

It was hypothesized that significantly lower moss element content would be obtained at the control sites in comparison with the remaining on-road sampling sites. However, the markedly elevated concentrations of the majority of elements were determined in the moss bags exposed in the central pedestrian zone. Specifically, markedly high concentration of Cu was found in this pedestrian zone, which is in accordance with the results of our previous studies (Vuković et al, 2015a; Aničić et al, 2009c). Elevated Cu concentrations was also previously pronounced for the same pedestrian zone in the research related to the usage of the tree leaf biomonitors (Aničić et al, 2011) and the bulk deposition collectors (Mijić et al, 2010). Thus, the marked Cu moss enrichment in this study could not be considered as an accidental case. The moss element content indicated that the second pedestrian zone, situated in peri-urban area, was less polluted than the central pedestrian zone. Finally, the third control site was in the biggest urban forest zone, and since the lowest element content was obtained in the moss bags exposed there, it proved to be the cleanest ambient within the city. According to our findings, control sites should be chosen with caution in experimental set-up in urban areas in regards with its position. The sampling sites situated in green urban niches could be recommended as appropriate for the assessment of the urban background levels of air pollutants.

CONCLUSIONS

The *S. girgensohnii* moss bag biomonitoring has outlined distinct spatial decline in the airborne toxic element content along crossroads, two- and one-lane streets. This result points to a need for monitoring and better characterization of micro-scale siting of toxic element distribution. Moss bag technique represents a suitable approach to achieve this goal given by simplicity and cost-effectiveness of its performing. Combined use of moss bag technique and regulatory instrumental measurements could enable dense spatial coverage and consequently, larger database of air pollutant concentrations. In addition, the results marked Sb, Cu, and Cr as reliable traffic tracers regarding the high correlation coefficients ($r > 0.65$) between the moss element concentrations and the counted traffic flows. Since moss bags provide information about long-term pollutant concentrations, the results could be indicative for assessment of the pedestrians, commuters, cyclists and workers exposure to traffic-related Cr, Cu, and Sb emissions.

ACKNOWLEDGEMENT

We are grateful to the City of Belgrade, the Company NIS (Serbia) and the Ministry of Education and Science of the Republic of Serbia (project No III43007) for support and to the bilateral cooperation with Joint Institute for Nuclear Research, Dubna, Russia.

REFERENCES

1. Adamo, P., Giordano, S., Sforza, A., Bargagli, R. 2011. Implementation of airborne trace element monitoring with devitalised transplants of *Hypnum cupressiforme* Hedw.: Assessment of temporal trends and element contribution by vehicular traffic in Naples city. Environ. Pollut. 159, 1620-1628.
2. Aničić, M., Tasić, M., Frontasyeva, M.V., Tomašević, M., Rajšić, S., Strelkova, L.P., Popović, A., Steinnes, E. 2009a. Active biomonitoring with wet and dry moss: A case study in an urban area. Environ. Chem. Lett. 7, 55-60.
3. Aničić, M., Tomašević, M., Tasić, M., Rajšić, S., Popović, A., Frontasyeva, M.V., Lierhagen, S., Steinnes, E. 2009b. Monitoring of trace element atmospheric deposition using dry and wet moss bags: Accumulation capacity versus exposure time. J. Hazard. Mater. 171, 182-188.
4. Aničić, M.,. Tasić, M, Frontasyeva, M.V., Tomašević, M., Rajšić, S., Mijić, Z., Popović, A. 2009c. Active moss biomonitoring of trace elements with *Sphagnum girgensohnii* moss bags in relation to atmospheric bulk deposition in Belgrade, Serbia. Environ. Pollut. 157, 673-679.
5. Aničić, M., Spasić, T., Tomašević, M., Rajšić, S., Tasić, M. 2011. Trace elements accumulation and temporal trends in leaves of urban deciduous trees (*Aesculus hippocastanum* & *Tilia* spp.). Ecol. Ind. 11, 824-830.
6. Ares, Á., Aboal, J.R., Carballeira, A., Giordano, S., Adamo, P., Fernández, J.A. 2014. Moss bag biomonitoring: A methodological review. Sci. Total Environ. 432, 143-158.
7. Brauer, M. 2010. How Much, How Long, What, and Where, Air Pollution Exposure Assessment for Epidemiologic Studies of Respiratory Disease. Proc. Am. Thorac. Soc. 7, 111-115.
8. Brown, D.H., Bates, J.W. 1990. Bryophytes and nutrient cycling. J Linn Soc Bot 104, 129-147.
9. Directive 2004/107/EC of the Europen parliament and of the council of 15 December 2004 relating to arsenic, cadmium, mercury, nickel and polycyclic aromatic hydrocarbons in ambient air, Official Journal of the European Union, 23, 1-16, (26/01/2005).
10. Directive 2008/50/EC of the Europen parliament and of the council of 21 May 2008 on ambient air and cleaner air for Europe, Officional Journal of the European Union, 1-44, (11/06/2008).
11. Dołęgowska, S., Migaszewski, Z.M. 2013. Anomalous concentrations of rare earth elements in the moss–soil system from south-central Poland. Environ. Pollut. 178, 33-40.

12. EMEP/EEA air pollutant emission inventory guidebook. Passenger cars, light commercial trucks, heavy-duty vehicles including buses and motor cycles 2013, update Septembar, 2014 http://www.eea.europa.eu/publications/emep-eea-guidebook-2013/
13. European Environment Agency, 2013. The Contribution of the Transport Sector to Total Emissions of the Main Air Pollutants in 2009 (EEA-32). Retrieved March 16, 2013, from. http://www.eea.europa.eu/data-and-maps/figures/thecontribution-of-the-transport-1.
14. Gietl, K.J., Lawrence, R., Thorpe, J.A., Harrison, M.J. 2010. Identification of brake wear particles and derivation of a quantitative tracer for brake dust at a major road. Atmos. Environ. 44, 141-146.
15. Goodman, T.G., Roberts, T.M. 1971. Plants and soils as indicators of metals in the air. Nature 231, 287-292.
16. Mijić, Z., Stojić, A., Perišić, M., Rajšić, S., Tasić, M., Radenković, M., Joksić, J. 2010. Seasonal variability and source apportionment of metals in the atmospheric deposition in Belgrade. Atmos. Environ. 44, 3630-3637.
17. Napier, F., D'Arcy, B., Jefferies, C. 2008. A review of vehicle related metals and polycyclic aromatic hydrocarbons in the UK environment. Desalination 226, 143-150.
18. Rivera, M., Zechmeister, H., Medina-Ramón, M., Basagaña, X., Foraster, M., Bouso, L., Moreno, T., 2011. Monitoring of Heavy Metal Concentrations in Home Outdoor Air Using Moss Bags. Environ. Pollut. 159 (4), 954-62.
19. Steinnes, E., Rühling, A., Lippo, H., Mäkinen, A. 1997. Reference materials for large scale metal deposition surveys. Accred Qual Assur 2, 243-249.
20. Tsai, J.H., Chiang, H.L., Hsu, Y.C., Weng, H.C., Yang, C.Y. 2003. The speciation of volatile organic compounds (VOCs) from motorcycle engine exhaust at different driving modes, Atmos. Environ. 37, 2485-2496.
21. Vuković, G., Aničić Urošević, M., Razumenić, I., Goryainova, Z., Frontasyeva, M.,Tomašević, M., Popović, A. 2013. Active Moss Biomonitoring of Small-Scale Spatial Distribution of Airborne Major and Trace Elements in the Belgrade Urban Area. Environ. Sci. Pollut. Res. 20, 5461-70.
22. Vuković, G., Aničić Urošević, M., Goryainova, Z., Pergal, M., Škrivanj, S., Samson, R., Popović, A. 2015a. Active moss biomonitoring for extensive screening of urban air pollution: Magnetic and chemical analyses. Sci. Total Environ. 521-522, 200-210.
23. Vuković, G., Aničić Urošević, M., Pergal, M., Janković, M., Goryainova, Z., Tomašević, M., Popović, A. 2015b. Residential heating contribution to persistent air pollutants (PAHs, trace and rare earth elements): a moss bag case study. Environ. Sci. Pollut. Res. 23, 18956-18966.
24. WHO, 2013. Review of Evidence on Health Aspects of Air Pollution REVIHAAP Project. Technical Report. World Health Organization, Regional Office for Europe, Copenhagen, Denmark.
25. Zechmeister, H.G., Hohenwallner, D., Riss, A., Hanus-Illnar, A. 2005. Estimation of element deposition derived from road traffic sources by using mosses. Environ. Pollut. 138, 238-249.
26. Zechmeister, H.G., Hagendorfer, H., Hohenwallner, D., Hanus-Illnar, A., Riss, A., 2006. Analyses of platinum group elements in mosses as indicators of road traffic emissions in Austria. Atmos. Environ. 40, 7720-32.

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502.3:502.175(048)
613.15(048)
66.071.9(048)

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Proceedings Abstracts of Keynote Invited Lectures and Contributed Papers
/ #The #Third International WeBIOPATR Workshop & Conference
Particulate Matter: Research and Management-WeBIOPATR2015
[Belgrade, October 14th-16th, 2015.] ;
editors Alena Bartonova, Milena Jovašević-Stojanović ;
[organisers Vinča Institute of Nuclear Sciences, Serbia
[and] Public Health Institute of Belgrade, Serbia
[and] Norwegian Institute for Air Research, Norway [and] with contribution by
World Health Organization, Regional Office Europe]
- Belgrade : Vinča Institute of Nuclear Sciences, 2016
(Belgrade : Printing Office of the Vinča Institute of Nuclear Science)
- 236 str. ; 30 cm. -

Kor. stv. nasl.: Proceedings. -
Tiraž 200. –
Str. 5: Preface / Editors. -
Bibliografija uz svaki rad. - Registar.

ISBN 978-86-xxxxx-xx-x

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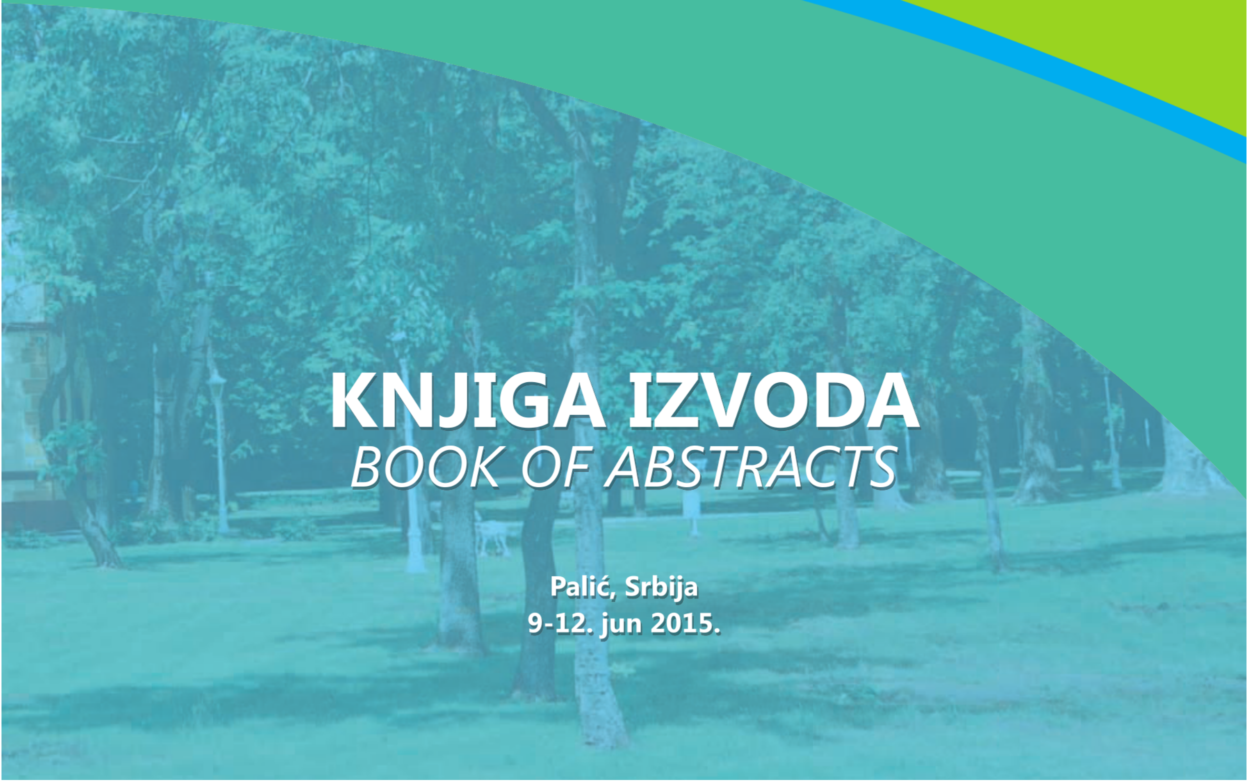

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Naslov	**KNJIGA IZVODA** **7. simpozijum Hemija i zaštita životne sredine**
Title	*BOOK OF ABSTRACTS* *7th Symposium Chemistry and Environmental Protection*
Izdavač	**Srpsko hemijsko društvo** **Karnegijeva 4/III, Beograd, Srbija**
Publisher	*The Serbian chemical society* *Karnegijeva 4/III, Belgrade, Serbia*
Za izdavača *For the publisher*	**Živoslav Tešić, predsednik Društva** *Živoslav Tešić, president of the Society*
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Tehnički urednik *Technical assistance*	**Maja Milanović**
Prelom i priprema *Design and prepress*	**Ivan Pinćjer**
Štampa *Printed by*	**FTN - Grafički centar GRID, Trg D. Obradovića 6, Novi Sad** *FTN - Graphic centre GRID, Trg D. Obradovića 6, Novi Sad*
Tiraž *Circulation*	**200 primeraka** *200 copies*
ISBN	**978-86-7132-058-0**

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CIP - Каталогизација у публикацији
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54(048)
502/504(048)
577.1(048)
66(048)

СИМПОЗИЈУМ Хемија и заштита животне средине са међународним учешћем (7 ; 2015 ; Палић)

Knjiga izvoda = Book of Abstracts / 7. simpozijum Hemija i zaštita životne sredine sa međunarodnim učešćem = 7th Symposium Chemistry and Environmental Protection with International Participation, Palić, Srbija, 09-12. jun 2015. ; [urednici, editors Branimir Jovančićević ... et al.]. - Beograd : Srpsko hemijsko društvo = Belgrade : Serbian Chemical Society, 2015 (Beograd : Grafički centar Grid). - 405 str. : ilustr. ; 25 cm

Tiraž 200. - Tekst na srp. i engl. jeziku. - Bibliografija uz svaki apstrakt. - Registar.

ISBN 978-86-7132-058-0

a) Хемија - Апстракти b) Животна средина - Заштита - Апстракти
c) Биохемија - Апстракти

COBISS.SR-ID 215457804

Prethodni skupovi iz oblasti hemije i zaštite životne sredine
Previous symposia on chemistry and environmental protection

1985 - I Jugoslovenski simpozijum, Beograd
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2008 - V Simpozijum, planina Tara
2013 - VI Simpozijum, Vršac

ISBN 978-86-7132-058-0

Moss-bag biomonitoring of small-scale decline in toxic element content: crossroad, two- and one-lane street study

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Exceeding in air pollutant limit values has been associated with many adverse health effects [1]. Transport sector represents one of the most important air pollution sources in urban areas. Traffic-related air pollutants, including particulate matter (PM) associated with toxic elements, are widespread, and can be often locally-elevated near major roads, crossroads and junctions, etc. For example, steep gradient in concentrations of fine particles (< PM2.5) was demonstrated attaining elevated levels near and on roads, and a return to background levels at distances of roughly 150 to 200 m [2].

In most urban areas, regulatory monitoring networks are designed to routinely provide continuous information on the concentrations of a small number of pollutants at limited number of locations. Thus, characterization of small-scale air pollution differences is lacked. However, biomonitoring as an easy-performed and cost-effective approach for air quality assessment could be useful for detailed surveys in urban microenvironments diversified by air pollution. Namely, the use of moss-bag technique has been developing in last several decades as a method of active monitoring of toxic air pollutants (e.g. heavy metals, PAHs) and has been tested in different environments [3, 4, 5, 6].

The goal of this study was to test possibility of *Sphagnum girgensohnii* moss bags to detect small-distance decline of toxic element concentrations, dependent on traffic flow. In the summer of 2014, the moss bags were exposed at 48 sampling sites in total. The sets of 3 sampling sites – crossroad, two- and one-lane road, away from each other approximately 300 m, were selected. Three pedestrian zones were chosen as urban background areas. In addition, traffic flows, total and per vehicle category, were counted at the sampling sites.

After 10-week exposure period, concentration of 41 elements was determined in the moss samples by inductively coupled plasma mass spectrometry (ICP-MS). Special attention was paid to Ba, Cd, Co, Cr, Cu, Fe, Ni, Pb, Pt, Sb, V and Zn which are recognized as toxic [1]. To assess the moss element enrichment, the moss concentration of each element after exposure was reduced and divided by the element baseline concentration before exposure.

The most of measured element concentrations were significantly increased in the exposed moss in comparison to their baseline level in the moss tissue. This indicates that the studied sites are polluted by the elements in question. The highest enrichment of the moss was obtained for the elements: Sb > Cu > Cr > Zn which is probably related to traffic emissions. Moreover, significant moss enrichment with some rare earth elements, such as Pt, were observed at heavy traffic sites which implies the elements as a possible new tracer of traffic-related air pollution.

In particular, at each pair of the sampling sites, a noticeable decline of the most element concentrations was observed from the crossroad to the one-lane road. In addition, high correlation coefficients ($R > 0.80$) were obtained between the traffic flow and concentration of certain elements such as Ba, Cr, Cu, Fe and Sb. All of this indicates the traffic-related origin of the elements in the exposed moss.

Finally, the results indicate that *S. girgensohnii* moss bag is sensitive to small-scale variations of air pollutant concentrations. This implies its usage for extensive screening of highly diversified urban air pollution. Moreover, combined use of moss-bag technique and regulatory measurements could enable detailed spatial coverage and consequently, larger database for air pollutants.

References

1. WHO, 2013. REVIHAAP Project. Technical Report, Copenhagen, Denmark.
2. Karner, A.A., Eisinger, D.S., Niemeier, D.A., *Environ. Sci. Technol.* 44 (2010), 5334-5344.
3. Aničić, M., Tasić, M., Frontasyeva, M.V., Tomašević, M., Rajšić, S., Mijić, Z., Popović, A., *Environ. Pollut.* 157 (2009) 673-679.
4. Ares, A., Fernández, J.A., Carballeira, A., Aboal, J.R., *Atmos. Environ.* 94 (2014) 496-507.
5. Vuković, G., Aničić Urošević, M., Razumenić, I., Goryainova, Z., Frontasyeva, M.,Tomašević, M., Popović, A., *Environ. Sci. Pollut. Res.* 20 (2013) 5461-70.
6. Vuković, G., Aničić Urošević, M., Razumenić, I., Kuzmanoski, M., Pergal, M., Škrivanj, S., Popović, A., *Atmos. Environ.* 85 (2014) 31-40.

Assesment of CH_3COOH, Na_2EDTA, $CaCl_2$, NH_4NO_3 and distilled water extraction procedures and microwave digestion for leaching of macro- and microelements from vineyard soil

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The bioavailability, mobility and distribution of macro- and microelements in soil depend on their total concentrations, as well as on the characteristics of the particle soil surface, kind of the bonds they are attached to the substrate and on the properties of the soil solution that is in contact with the solid material. Variations of the chemical or physical conditions can influence the release of these elements [1]. Translocation of elements from soil has a major impact on the growing plants and on their quality in any agricultural area.

In this study, soil samples were collected from Radmilovac vineyard (Serbia) during grapevine season in 2013. The samples were air-dried and homogenized in the laboratory conditions. Bioavailability of elements were tested by five different extractants: 0.11 mol L^{-1} CH_3COOH during 16 h extraction [2], 0.05 mol L^{-1} Na_2EDTA during 1 h extraction [2], 0.01 mol L^{-1} $CaCl_2$ during 3 h extraction [2], 1 mol L^{-1} NH_4NO_3 during 2 h extraction [2] and distilled water during 2 h [3] and 16 h extractions [2]. Microwave digestion of the soil samples with aqua regia (HNO_3: HCl in 3:1) was also performed. The concentrations of extracted macroelements: Al, Ca, Fe, K, Mg, Mn, Na, P, S, Si and microelements: B, Be, Cd, Co, Cr, Cu, Mo, Ni, Pb, Sb, V, Zn were determined by, inductively coupled plasma-optical emission spectroscopy (ICP-OES).

The highest concentrations of Al, B, Be, Mg, Mo, Si and Zn have been extracted by CH_3COOH. Extraction with Na_2EDTA yielded the highest concentrations of Ca, Cd, Co, Cu, Fe, K, Mn, Ni, P, Pb and V. Distilled water has shown to be good extraction medium for Na and S. Acetic acid has been proven to be an aggressive extractant and it can be used for isolation of higher concentrations of elements, rather than distilled water, $CaCl_2$ and NH_4NO_3. The acidity of CH_3COOH enhances the extraction of bioavailable fraction of microelements from various substrates and destruction of carbonates as well.

Aqua regia, as a mixture of concentrated acids, reacts very aggressively on the matrix and destroys it. After microwave digestion of soil samples by aqua regia, only silicate fraction remains partially non-destroyed. Determined element concentrations (macroelements: Al, Ca, Fe, K, Mg, Mn, Na, P, S, Si and microelements: B, Be, Cd, Co, Cr, Cu, Mo, Ni, Pb, Sb, V, Zn) isolated by aqua regia were considerably higher than determined element concentrations that have been isolated by the other extractants.

Copper (106 µg g^{-1}) and Cd (3.98 µg g^{-1}) concentrations digested by aqua regia, were higher than the concentration limits prescribed by the legislation for these elements which amount to 0.8 µg g^{-1} for Cd and 36 µg g^{-1} for Cu. However, the concentration of these two elements did not exceed the remediation values prescribed by the legislation of the Republic of Serbia (12 µg g^{-1} for Cd and 190 µg g^{-1} for Cu).

The concentrations of Co (13 µg g^{-1}) and Sb (3.68 µg g^{-1}) were also slightly elevated, i.e., above the limit values prescribed by the legislation (9 µg g^{-1} and 3 µg g^{-1}, respectively). However, the concentrations of these elements were significantly lower than the remediation values prescribed by the legislation (240 µg g^{-1} for Co and 15 µg g^{-1} for Sb).

References

1. Filgueiras, A. V., Lavilla,I ., Bendicho, C., *J. Environ. Monit.*, 4 (2002), 823 - 857
2. Quevauviller, Ph., *Trends in analytical chemistry*, 17 (5) (1998), 289 - 297
3. Zakari, H., Adams, D., Shimbayev, M., Nyam, P., *Jorind* , 9(2) (2011), 114 - 118

Correlation between macro- and microelements isolated from the vineyard soil by different extractant methods

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It is well known that distribution, mobility and bioavailability of macro- and microelements in the soils depend not only on their total concentration but also on the association form in the solid phase to which they are bound. Even slight changes of the chemical or physical conditions (pH, temperature, redox potential, organic matter composition, leaching and ion-exchange processes and microbial activity) in the soil can accelerate or prevent the release of these elements. Therefore, these changes have a major impact on the growing plants and on their quality in every agricultural area [1]. In this study, soil samples collected from Radmilovac vineyard (Serbia) during grapevine season of 2013. were investigated.

Bioavailability of elements were tested by five different extractants: 0.11 mol L^{-1} CH_3COOH during 16 h extraction [2], 0.05 mol L^{-1} Na_2EDTA during 1 h extraction [2], 0.01 mol L^{-1} $CaCl_2$ during 3 h extraction [2], 1 mol L^{-1} NH_4NO_3 during 2 h extraction [2] and distilled water during 2 h [3] and 16 h extractions [2]. Concentrations of bioavailable macroelements: Al, Ca, Fe, K, Mg, Mn, Na, P, S, Si and microelements: B, Be, Cd, Co, Cr, Cu, Mo, Ni, Pb, Sb, V, Zn were determined by ICP-OES. The (Pearson's) correlation was applied in order to test the correlation between the element concentrations isolated by different extractants. The positive correlations were obtained between:

- The pairs of element concentrations isolated by 0.11 mol L^{-1} CH_3COOH: Cd-B, Co-Be, Cu-B, Cu-Be, Cu-Co, Fe-Al, Mg-Ca, Mn-B, Mn-Co, Mn-Cu, Mo-Ca, Mo-Mg, Ni-Co, Ni-Mn¸ P-Be, P-Co, P-Cu, P-Mn, P-Ni, Si-Al, Si-Be, Si-P, V-Cd, Zn-B, Zn-Be, Zn-Co, Zn-Cu, Zn-Mn, Zn-N and Zn-P.
- The pairs of element concentrations isolated by distilled water during 16 h extraction: Cd-B, Co-Al, Co-Cd, Cr-Al, Cu-B, Cu-Cd, Cu-Co, Fe-Al, Fe-Be, Fe-Cr, K-Cr, Mg-Fe, Mn-B, Mn-Cd, Mn-Co, Mn-Cu, Na-B, Na-Cd, Na-Co, Na-Cu, Na-Mn, P-B, P-Cr, P-Cu, P-K, Pb-Co, Pb-Cu, Pb-Mn, Pb-Mn, Pb-Na, S-B¸ S-Cd, S-Co, S-Cu, S-Mn, S-Na, S-Pb, Si-Al, Si-Co, Si-Cr, Si-Fe, V-Al, V-Co, V-Si, Zn-B, Zn-Cd, Zn-Cu, Zn-Mn, Zn-Na, Zn-P and Zn-S.
- The pairs of element concentrations isolated by distilled water during 2 h extraction: Cr-Al, Fe- Al, Fe-Cr, Mg-Ca, Mn-Al, Mn-Be, Mn-Cr, Mn-Fe, Na-Cd, Na-Cu, P-B, Pb-Al, Pb-Cr, Pb-Mn, Pb-Ni, Sb-Cu, Si-Fe, V-Al, V-Cr, V-Fe, V-Mn, Zn-Cd.
- The pairs of element concentrations isolated by 0.05 mol L^{-1} Na_2EDTA: Co-Cd, Fe-Co, K-Be, Mn-Cd, Mn-Co, Mn-Fe, Ni-Cd, Ni-Co, Ni-Fe, Ni-Mn, P-Be, P-K, Pb-Al, Pb-Cd, Pb-Co, Pb-Fe, Pb-Mn, Pb-Ni, Si-S, V-Al, Zn-Be, Zn-Cd, Zn-Co, Zn-Fe, Zn-Mn and Zn-Ni.
- The pairs of element concentrations isolated by 0.01 mol L^{-1} $CaCl_2$: Fe-Al, Mn-Co, Mn-Mg, Mo-Cr, Na-Cu, Ni-Al, Ni-Fe, P-K, Sb-Na, Si-K, Si-Mn, V-Be, V-Sb, Zn-Al, Zn-Fe and Zn-Ni.
- The pairs of element concentrations isolated by 1 mol L^{-1} NH_4NO_3: Be-Al, Cd-Al, Cd-Be, Co-Be, Co-Cd, Fe-Al, Fe-Cd, Fe-Co, K-Be, K-Cd, K-Co, K-Fe, Mn-Be, Mn-Cd, Mn-Co, Mn-K, Na-Ca, Sb-Ca, Sb-Cr, Sb-Na, Si-Al, Si-Be, Si-Cd, Si-Co, Si-Fe, Si-K and Si-Mn.

It can be assumed that the pairs of element concentrations, which show positive correlation, in a certain percentage, originate from the same source in the vineyard soil.

It is interesting to note significant correlations of concentrations of Ca and Mg extracted from the soil samples using CH_3COOH (R = 0.8856, p = 0.0015) and distilled water during 2 h (R = 0.7425, p = 0.0219), but also that concentrations of neither of these macroelements is significantly correlated with the concentration of microelements isolated with the same extractant.

The concentration of S is in positive correlation with the concentrations of Cu, Mn, P and Na, probably witnessing their common source from the agents for treating vineyards. The concentrations of Cu and S, isolated from the soil samples by distilled water during 16 h, have significant correlation (R = 0.9294, p = 0.0003). These elements could have entered to environment while grapevines were treated by fungicide $CuSO_4$ [4].

Finally, it can be concluded that there is no uniform method and unique extractant for isolation of the most bioavailable fraction for all elements from the vineyard soil. The most common concentrations of macroelements K and Mn, are in correlation with concentrations of microelements Cd, Co, Ni and Zn. This indicates that the most of their concentrations in soils are followed by microelements, whose concentrations are much lower than concentrations of macroelements. However, the pairs of macro- and microelements have the same origin in soil and can be isolated by the same extractant.

References

1. Filgueiras, A. V., Lavilla,I ., Bendicho, C., *J. Environ. Monit.*, 4 (2002), 823 - 857
2. Quevauviller, Ph., *Trends in analytical chemistry*, 17 (5) (1998), 289 - 297
3. Zakari, H., Adams, D., Shimbayev, M., Nyam, P., *Jorind* , 9(2) (2011), 114 - 118
4. Kabata - Pendias, A., Pendias, H., *Trace Elements in Soils and Plants, Third Edition,* (2001)

Geophysical Research Abstracts
Vol. 17, EGU2015-1138, 2015
EGU General Assembly 2015

Determination of bioavailable macro- and microelements from agricultural soil using different extractants

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Translocation of elements from soil to plant has a major impact on the growing plants and on their quality in any agricultural field. In this study, soil samples were collected from agricultural area Radmilovac, Serbia during grapevine season in 2013. Bioavailable elements from soil to plant (grapevine) were isolated by five different extractants: 0.11 mol L^{-1} CH_3COOH, 0.05 mol L^{-1} Na-EDTA, 0.01 mol L^{-1} $CaCl_2$, 1 mol L^{-1} NH_4NO_3 and distilled water during 2 and 16 h. Concentrations of 22 bioavailable macroelements: Al, Ca, Fe, K, Mg, Mn, Na, P, S, Si and microelements: B, Be, Cd, Co, Cr, Cu, Mo, Ni, Pb, Sb, V, Zn were determined by ICP-OES. The best extractant for Al, B, Be, Mg, Mo, Si and Zn was CH_3COOH, Na-EDTA for Ca, Cd, Co, Cu, Fe, K, Mn, Ni, P, Pb, V, and distilled water for Na and S. Acetic acid has been proven to be an aggressive extractant and it can be used for isolation of higher concentrations of plant bioavailable elements from soil, rather than distilled water, $CaCl_2$ and NH_4NO_3. The acidity of CH_3COOH enhances the extraction of bioavailable fraction of microelements from various substrates and destruction of carbonates as well. However, it can be concluded that there is no unique extractant for isolation of the most bioavailable fraction for all elements from the soil. It can be noticed that the most common concentrations of macroelements, K and Mn, are in correlation with concentrations of microelements, Cd, Co, Ni and Zn. This indicates that the most of their concentrations in soils are followed by microelements, whose concentrations are much lower than concentrations of macroelements. However, as these correlations are the most common, it can be concluded that the pairs of macro- and microelements (e.g. Mn-Cd, Mn-Co, Ni-Cd, Ni-Co, Ni-Mn, Zn-Cd, Zn-Co, Zn-Mn, Zn-Ni) have the same source in soil and can be isolated by the same extractant. It is interesting to note that the concentrations of Ca and Mg extracted from soil using CH_3COOH are in correlation and that neither of these macroelements is in correlation with the concentration of microelements isolated with the same extractant. The concentrations of Cu and S extracted from soil by distilled water during 16 h are in correlation. These elements could have entered only through the soil surface layer while grapevines were primarily treated by fungicide copper(II)-sulphate. In addition, the concentration of S is correlated with the concentrations of Mn, P and Na. It can be assumed that the correlation between these elements points to their origin from the pesticides used in agriculture production.

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У Београду, 25. 10. 2016. године

На основу члана 161. Закона о општем управном поступку и службене евиденције издаје се

УВЕРЕЊЕ

Тијана (Драгомир) Милићевић, рођена **23. 1. 1989.** године у месту **Крушевац**, општина **Крушевац**, **Република Србија**, број индекса **ДХ01/2014**, уписана је школске **2016/2017.** године на студијски програм докторских академских студија **Хемија**, као **студент који се финансира из буџета**.

Студент је први пут уписан на наведени студијски програм школске **2014/2015.** године. По статуту високошколске установе студије трају **6 семестара**, односно **3 године**. Рок за завршетак студија јесте двоструко трајање студија.

Уверење се издаје на лични захтев студента.

Овлашћено лице високошколске установе

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Дозволу за рад број 612-00-02666/2010-04 од 10. децембра 2010.
године је издало Министарство просвете и науке Републике Србије

Диплома

Тијана, Драгомир, Милићевић

рођена 23. јануара 1989. године у Крушевцу, Република Србија, уписана школске 2012/2013. године, а дана 8. октобра 2013. године завршила је мастер академске студије, другог степена, на студијском програму Хемија, обима 60 (шездесет) бодова ЕСПБ са просечном оценом 9,75 (девет и 75/100).

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Дозволу за рад број 612-00-02666/2010-04 од 10. децембра 2010.
године је издало Министарство просвете и науке Републике Србије

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рођена 23. јануара 1989. године у Крушевцу, Република Србија, уписана школске 2008/2009. године, а дана 24. септембра 2012. године завршила је основне академске студије, првог степена, на студијском програму Хемичар за животну средину, обима 240 (двеста четрдесет) бодова ЕСПБ са просечном оценом 7,81 (седам и 81/100).

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Кратак преглед научних активности кандидата

Тијана Милићевић, истраживач приправник у Лабораторији за физику животне средине Института за физику у Београду, је од 1. јуна 2015. године укључена у активности на националном пројекту Министарства просвете, науке и технолошког развоја ИИИ43007 – „Истраживање климатских промена и њихов утицај на животну средину – праћење утицаја, адаптација и ублажавање Истраживање климатских промена и њиховог утицаја на животну средину – праћење утицаја, адаптација и ублажавање”, потпројекат 3 „Интегрална истраживања квалитета ваздуха у урбаној средини”.

У оквиру потпројекта кандидат се бави научно-истраживачким радом за потребе израде докторске тезе. Циљеви научно-истраживачких активности кандидата су проучавање биодоступности загађујућих супстанци и њихов транспорт у систему земљиште–биљка–ваздух. Земљиште представља главни медијум у ком се депонују загађујуће супстанце из ваздуха, а истовремено је и извор емисија истих у ваздух ресуспензијом. Истраживања кандидата се заснивају на испитивању транспорта загађујућих супстанци из земљишта у биљку, али и њихово усвајање из ваздуха. Усвајање загађујућих супстанци пропорционално њиховом присуству у околном амбијенту је својство које неку биљку опредељује као потенцијалну биомониторску врсту. Последњих деценија биомониторинг је препознат као комплементарна метода конвенционалним инструменталним мерењима загађења ваздуха односно земљишта.

Активности кандидата у периоду од 1. јуна 2015. до сада:

1. Прикупљање различитих типова узорака (земљиште, лист, плод) у пољопривредној области (комерцијални виногради, огледна добра и органски виноград).

Узорци земљишта (површински и дубински), лист и плод (кожица, пулпа, семе) винове лозе су прикупљени током вегетационе сезоне 2015. и 2016. године.

2. Активни биомониторинг загађења ваздуха коришћењем маховине као активног биомонитора у пољопривредној области.

Експерименталне поставке су спроведене у различитим периодима (2, 4 и 6 месеци излагања маховине) у различитим виноградима (комерцијални виногради, огледна добра и органски виноград), а узорци су анализирани на садржај одабраних токсичних елемената.

3. Тестови биодоступности загађујућих супстанци из земљишта

Извршено је тестирање шест различитих екстракционих метода за изоловање биодоступних фракција макро- и микроелемената из земљишта. Одређиван је укупни и псеудо-укупни садржај елемената у узорцима земљишта, као и листа и плода (семена, пулпе, кожице) винове лозе.

4. Анализа добијених података, процена еколошког ризика и ризика по људско здравље. Публиковање радова у мђународним часописима.

Извршено је процењивање утицаја токсичних елемената на квалитет животне средине, процена ризика изложености радника у виноградима токсичним елементима из земљишта, као и процена утицаја токсичних елемената из плода винове лозе на здравље људи (процена дневне дозе). У току је израда и публиковање научних радова.