DANGEROUS HEAVY METALS IN SOIL AND SMALL FOREST FRUIT AS A RESULT OF OLD ENVIRONMENTAL LOADS

VOLLMANNOVA A.[1], ZUPKA S.[1], BAJCAN D.[1], MEDVECKY M.[2] and DANIEL J.[2]

[1] Slovak University of Agriculture in Nitra, Faculty of Biotechnology and Food Sciences, Dept. of Chemistry, Tr. A. Hlinku 2, 94976 Nitra, Slovak Republic, [2] National Agricultural and Food Centre, Grassland and Mountain Agricultural Research Institute, Research Station in Kriva na Orave, 027 55 Kriva na Orave, Slovak Republic
E-mail: alena.vollmannova@uniag.sk

ABSTRACT

Middle Spis is one of risky regions of the Slovakia with soils affected by acid pollutants as well as by heavy metals. Localities Rudnany, Krompachy and Spisska Nova Ves were the determining sources of the environment contamination during several decades. Despite of weakening of mining the negative consequences of the metallurgical production in acid soils of this region still persist.

The aim of this study was to assess the degree of risk for Pb, Cd, Cr, Cu, Zn, Ni, Co and Hg input from soil into small forest fruit species grown in the loaded region of Middle Spis.

The samples of soil and fruit were collected from 12 exactly fixed sampling points using GPS. In soil the agrochemical characteristics (pH/KCl, content of K, Mg, P, Ca and humus) were determined. Pseudototal (total) content of heavy metals including all metal forms except of residual fraction was determined in soil extract by *aqua regia* and the content of bioavailable forms in soil extract by NH_4NO_3 (c = 1mol/dm^3). Used analytical method was flame AAS (AAS Varian AA Spectr DUO 240 FS/240Z/UltrAA). The contents of risky metals in plant samples (fresh fruits and dry leaves of blackberries, rose hips, sloes, hawthorn and raspberries) were determined by AAS method, too. The content of Hg was determined using AMA 254 Advanced Mercury Analyzer. Obtained results were evaluated according to legislation valid in the Slovak Republic.

In all sampling sites the determined total soil Cd content (1.76-5.86 mg/kg) as well as content of mobile Pb forms (0.11 – 1.16 mg/kg) were higher than limit values. Contents of Cu with intervals of increased values in mg/kg (60.8 – 215.3), Zn (150 – 355), Co (15.1 – 23.7) and Hg (0.52 – 20.78) exceeded limit values in 3, 5 and in 10 soil samples, respectively. In 4 sampling points also content of mobile Cd and in one site content of mobile Zn exceeded critical values given by the legislation.

Only in 2 fruit samples (blackberries and rose hips) the determined Ni and in 1 sample (hawthorn) content of Cd were higher than hygienic limit values. On the other hand, in dry leaves of investigated fruit species the enhanced Hg, Ni and Cd contents were determined (in 7, 8 and all 12 samples, respectively). The intervals of determined values of Hg, Pb and Cd content (mg/kg) in leaves were: 0.051 – 0.188, 0.37 – 1.19 and 0.25 – 0.75.

Our results suggest the casual connection among enhanced contents of dangerous heavy metals, especially Hg, in soils as well as plants grown in the vicinity of former emission sources and the previous metallurgical activity in this area

Keywords: forest fruit, old environmental load, heavy metals, contamination

1. Introduction

The old mines, scrap-heaps, landfills, industrial and municipal wastes are important sources of the environment contamination by heavy metals. The region of middle Spiš is, according to environmental regionalization, classified as an environmentally loaded and unhealthy (Angelovicova and

Fazekasova, 2014). There are two main centres of pollution, Krompachy village and its surrounding (with numerous abandoned copper mines and still ongoing copper processing plant) and Rudnany village and its surrounding, where mercury was long time mined and processed (Angelovicova *et al*., 2014). Iron-ores mines Rudnany became the biggest ore enterprise in former Czechoslovak Republic and the biggest importer of mercury. Gradually the storages of siderite, chalkopyrite, tetraedrite with the mixture of mercury, silver and antimony had been running out. After 1989 mining activities declined. Before the attenuation of production, 4 t of mercury per year were emitted into the air (Hanculak *et al*., 2006). In the surrounding area of Krompachy there was copper and iron ore mined and processed during the 700 years (Krokusova *et al*., 2013**).** The present smelter was launched in 1951 for processing ore concentrates and recycled copper materials, The production was ceased in 1999 due to falling world copper prices. The smelter was reopened in September 2000 by the newly established joint-stock company Kovohuty (Barecz, 2000).

The aim of this study was to assess the degree of risk for Pb, Cd, Cr, Cu, Zn, Ni, Co and Hg input from soil into small forest fruit species grown in the loaded region of Middle Spis.

2. Material and methods

2.1. Sampling sites

The 12 sampling sites were determined using GPS, from each one the samples of soil and fruits plants were collected and analysed.

2.2. Determination of dangerous metals content in soil samples

Pseudototal content of Pb, Cd, Cr, Cu, Zn, Ni and Co including all metal forms except of residual metal fraction was assessed in soil extract by *aqua regia* (HCl: CentralChem, Slovakia, HNO_3: Merck, Germany) and content of mobile forms of heavy metals in soil extract by NH_4NO_3 (c=1mol/dm^3, Merck, Germany). Contents of heavy metals were determined by AAS method (AAS Varian AA Spectr DUO 240FS/240Z/UltrAA, Varian, Australia). The total Hg content was determined using AMA 254 Advanced Mercury Analyzer. Gained results of heavy metal content expressed as mg/kg were evaluated according to Law No. 220/2004 (valid in Slovakia).

2.3. Determination of dangerous metals content in fruit samples

Homogenized fruit samples (4 g) were mineralized in a closed system of microwave digestion using Mars X-Press 5 (CEM Corp., USA) in a mixture of 5 mL HNO_3 (Suprapur, Merc, Germany) and 5 mL deionized water (0.054 μS/cm) from Simplicity185 (Millipore, UK). Metal determinations were performed in a Varian AA240Z (Varian, Australia) atomic absorption spectrometer with Zeeman background correction. The obtained results were expressed as mg/kg FM (fresh matter) - fruits and as mg/kg DM (dry matter) - leaves. Gained values were compared to maximal allowed amounts given by the Food Codex of the Slovakia.

2.4. Statistical analysis

Statistical processing of the results was carried out using software Statgraphics Centurion XVI.I. One-way analysis of variance ($\alpha = 0.05$) was used. Mean comparisons between investigated parameters were done by the LSD test.

3. Results and discussion

3.1. Sampling sites

The samples of soil and small forest fruit (4 samples of blackberry, 3 samples of rose hip, 3 samples of sloe, one sample of hawthorn and one sample of raspberry) were collected from 12 exactly fixed sampling points using GPS. The humus content was in interval 1.65 – 16.35 % (low – very high humous soil). The interval of exchangeable soil reaction values was 3.54 (extremely acid) – 7.2 (neutral). Negative relationships between pH values and exchangeable heavy metals were recorded by many authors (e.g. Li *et al*., 2005; Ogundiran and Osibanjo, 2009; Berbecea *et al*., 2010). The determined contents of K, Mg and P in soil from sampling sites are 88.1 – 634.2; 84.3 – 608.6 and 11.3 – 296 mg/kg, respectively. Results presented by Zhu *et al*. (2015) demonstrated detrimental effects of excess N on soil inorganic nutrients (mainly inorganic P and exchangeable base cation nutrients) of different forest soil types.

3.2. Determination of dangerous metals content in soil samples

Pseudototal content of risk metals was assessed in soil extract by *aqua regia* (see Table 1).

Table 1: Pseudototal content of heavy metals in soil extract by *aqua regia* (Hg total content) in mg/kg

No.	Fruit species	Heavy metals in soil extract by ***aqua regia*** **(Hg total content) in mg/kg**							
		Pb	**Cd**	**Cr**	**Cu**	**Zn**	**Ni**	**Co**	**Hg**
1	Blackberry I.	23.60^{a}	2.69^{e}	24.60^{i}	60.80^{h}	74.10^{b}	40.30^{g}	14.60^{e}	13.89^{h}
2	Rose hip I.	40.80^{cd}	3.88^{j}	22.60^{g}	24.80^{c}	190.00^{h}	39.10^{f}	16.50^{g}	20.76^{i}
3	Sloe I.	40.80^{cd}	3.88^{j}	22.60^{g}	24.80^{c}	190.00^{h}	39.10^{f}	16.50^{g}	20.76^{i}
4	Rose hip II.	200.00^{f}	2.99^{f}	27.10^{j}	14.90^{h}	145.80^{e}	32.40^{e}	10.70^{c}	0.52^{b}
5	Blackberry II.	42.50^{d}	3.51^{i}	23.10^{h}	30.60^{f}	182.40^{g}	49.70^{i}	16.30^{g}	0.18^{h}
6	Sloe II.	37.00^{c}	5.53^{k}	10.60^{a}	51.10^{g}	79.00^{c}	25.20^{c}	10.30^{b}	0.17^{a}
7	Hawthorn	107.00^{e}	3.17^{g}	17.10^{d}	20.20^{b}	150.00^{f}	17.30^{a}	5.10^{a}	2.02^{d}
8	Blackberry III.	28.30^{ab}	1.76^{a}	13.70^{b}	27.00^{d}	63.70^{a}	22.60^{b}	11.60^{d}	3.44^{f}
9	Blackberry IV.	27.20^{ab}	2.47^{d}	20.60^{f}	28.40^{e}	101.60^{d}	46.50^{h}	15.30^{f}	1.34^{c}
10	Rose hip III.	25.40^{ab}	3.41^{h}	30.20^{k}	150.80^{i}	81.10^{c}	54.00^{j}	23.70^{h}	3.67^{g}
11	Sloe III.	24.90^{ab}	2.08^{b}	15.20^{c}	215.30^{j}	61.50^{a}	24.90^{c}	11.70^{d}	2.06^{d}
12	Raspberry	29.40^{b}	2.21^{c}	17.60^{e}	51.20^{g}	235.00^{i}	28.70^{d}	11.50^{d}	3.06^{e}
	Limit value*	**115.00**	**1.00**	**90.00**	**70.00**	**200.00**	**60.00**	**20.00**	**0.75**

** Limit value given by the Law No. 220/2004*
Values marked with the same letter are not significantly different (P-value < 0.05)

Table 2: Content of heavy metals in soil extract by NH_4NO_3 (c=1mol/L) in mg/kg

No.	Fruit species	**Content of heavy metals in soil extract by NH_4NO_3 (c=1mol/L) in mg/kg**						
		Pb	**Cd**	**Cr**	**Cu**	**Zn**	**Ni**	**Co**
1	Blackberry I.	0.11^{a}	0.04^{a}	0.03^{a}	0.08^{cd}	0.08^{e}	0.12^{a}	0.05^{a}
2	Rose hip I.	0.18^{b}	0.66^{e}	0.08^{cd}	0.53^{g}	0.04^{ab}	0.15^{b}	0.09^{b}
3	Sloe I.	0.18^{b}	0.66^{e}	0.08^{cd}	0.53^{g}	0.04^{ab}	0.15^{b}	0.09^{b}
4	Rose hip II.	1.16^{f}	0.29^{d}	0.17^{e}	0.07^{bc}	4.12^{k}	0.53^{f}	0.48^{d}
5	Blackberry II.	0.23^{cd}	0.08^{c}	0.09^{d}	0.06^{b}	0.12^{f}	0.19^{c}	0.12^{c}
6	Sloe II.	0.20^{bc}	0.09^{c}	0.05^{b}	0.17^{e}	0.05^{bc}	0.20^{cd}	0.12^{c}
7	Hawthorn	0.18^{b}	0.08^{c}	0.03^{a}	0.19^{f}	0.03^{a}	0.15^{b}	0.09^{b}
8	Blackberry III.	0.14^{a}	0.06^{b}	0.04^{ab}	0.09^{d}	1.49^{j}	0.26^{e}	0.06^{a}
9	Blackberry IV.	0.11^{a}	0.06^{b}	0.05^{b}	0.01^{a}	0.34^{h}	0.19^{c}	0.10^{b}
10	Rose hip III.	0.26^{d}	0.09^{c}	0.07^{c}	0.57^{h}	0.06^{c}	0.21^{d}	0.13^{c}
11	Sloe III.	0.41^{e}	0.04^{a}	0.07^{c}	0.00^{a}	0.47^{i}	0.19^{c}	0.05^{a}
12	Raspberry	0.14^{a}	0.05^{ab}	0.03^{a}	0.16^{e}	0.26^{j}	0.16^{b}	0.09^{b}
	Limit value*	**0.10**	**0.10**	**-**	**1.00**	**2.00**	**1.50**	**-**

** Limit value given by the Law No. 220/2004*
Values marked with the same letter are not significantly different (P-value < 0.05)

In all soil samples the determined pseudototal Cd content was higher than legislative limit value even more than 5-fold; with exception of 3 samples also Hg content exceeded the hygienic limit; in 3 sample sites extremely high Hg content was determined (even more than 27-fold compared with the limit value). In 2 soil samples also Cu content and in 1 sample Co and Zn contents were higher than the limit values.

The content of bioavailable forms of heavy metals assessed in soil extract by NH_4NO_3 (c=1mol/L) is presented in Table 2.

In all samples content of Pb, in 3 samples also Cd and in 1 sample Zn mobile forms were higher than legislative limit values. The high heavy metals concentration in soils is usually reflected by higher metals content in plants bodies (Buszewski *et al*., 2000). Soil properties are important factors modifying metal bioavailability and toxicity and should be considered during the ecological risk assessment of metals in contaminated soils (Bradham *et al*., 2006).

3.3. Determination of dangerous metals content in fruit samples

Plant samples (fruits and leaves) were collected from the same sampling points as soil samples. Hazardous metal contents were determined in dry leaves (often used as components in tea mixtures) as well as in fresh fruits. The obtained results were compared to maximal allowed amounts in foodstuffs given by the Food Codex of the Slovakia. Only in 2 fruit samples the determined Cd (blackberry I. and hawthorn) and Ni (rose hip I. and blackberry IV.) contents exceeded the hygienic limits given by the legislation (see Table 3). Our values are lower (blackberries) or similar (raspberries) compared to those documented by Balaban *et al*. (2012). Wieczorek *et al*., (2010) similarly reported that the highest acceptable concentrations based upon Polish standards for Pb and Cd were not exceeded in forest fruit.

Table 3: Content of heavy metals in fruit in mg/kg of fresh matter

No.	**Fruit species**	**Content of heavy metals in fruit in mg/kg of fresh mater**							
		Pb	**Cd**	**Cr**	**Cu**	**Zn**	**Ni**	**Co**	**Hg**
1	Blackberry I.	0.03^{b}	0.05^{g}	0.16^{f}	0.93^{d}	3.13^{h}	0.12^{e}	0.00^{a}	0.002^{ab}
2	Rose hip I.	0.00^{a}	0.04^{e}	0.15^{e}	0.96^{e}	2.94^{e}	0.92^{l}	0.04^{d}	0.002^{ab}
3	Sloe I.	0.00^{a}	0.00^{a}	0.31^{i}	1.26^{i}	3.13^{i}	0.00^{a}	0.03^{c}	0.002^{ab}
4	Rose hip II.	0.00^{a}	0.00^{a}	0.33^{k}	1.75^{k}	5.19^{k}	0.09^{c}	0.59^{i}	0.003^{b}
5	Blackberry II.	0.00^{a}	0.03^{c}	0.14^{d}	0.99^{f}	2.56^{b}	0.11^{d}	0.03^{b}	0.002^{ab}
6	Sloe II.	0.00^{a}	0.00^{a}	0.30^{h}	2.57^{l}	4.21^{j}	0.08^{b}	0.03^{b}	0.002^{ab}
7	Hawthorn	0.00^{a}	0.08^{h}	0.25^{g}	1.05^{g}	2.72^{d}	0.44^{i}	0.18^{h}	0.001^{a}
8	Blackberry III.	0.00^{a}	0.05^{f}	0.03^{b}	0.48^{b}	2.08^{a}	0.38^{h}	0.06^{f}	0.002^{ab}
9	Blackberry IV.	0.00^{a}	0.03^{d}	0.03^{a}	0.72^{c}	2.59^{c}	0.78^{k}	0.05^{e}	0.001^{a}
10	Rose hip III.	0.00^{a}	0.00^{a}	0.35^{l}	1.47^{i}	7.71^{l}	0.49^{j}	0.11^{g}	0.005^{c}
11	Sloe III.	0.00^{a}	0.00^{a}	0.32^{j}	1.16^{h}	2.95^{f}	0.16^{g}	0.00^{a}	0.001^{a}
12	Raspberry	0.00^{a}	0.02^{b}	0.11^{c}	0.41^{a}	3.11^{g}	0.14^{f}	0.00^{a}	0.006^{c}
	Limit value**	**0.20**	**0.05**	**4.00**	**10.00**	**-**	**0.5**	**-**	**0.03**

*** Limit value given by the Food Codex*

Values marked with the same letter are not significantly different (P-value < 0.05)

The degree of heavy metal bioavailability and consequently plant uptake is influenced by many factors such as soil reaction, temperature, redox potential, cation exchange capacity of solid phase, competition with other metal ions etc. (Trangmar *et al*., 1985; Wopereis *et al*., 1988). Our results confirmed that fresh forest fruits could be considered safe from the aspect of heavy metals content.

On the other hand the determined Cd, Ni and Hg contents in dry leaves of almost all samples were higher than limit values (see Table 4). The maximal Cd amount determined in fruit leaves was 7.5–fold (rose hip II), Ni more than 3.5-fold (sloe I.) and Hg 3.8-fold (blackberry I. and rose hip I.) higher than limits. According Hazlett *et al*. (1983) bioavailability of Ni is influenced by physical factors (texture, temperature and water content), chemical factors (pH, organic substances, redox potential) and biological factors (plant species variability, microbial activity).

Our results indicate that use of dry leaves of forest fruits as components of tea mixtures could be risky for the human health from the aspect of hazardous metals content (Cd, Ni and Hg).

Table 4: Content of heavy metals in leaves of fruit species in mg/kg of dry matter

No.	Fruit species	Content of heavy metals in leaves of fruit species in mg/kg of dry matter							
		Pb	**Cd**	**Cr**	**Cu**	**Zn**	**Ni**	**Co**	**Hg**
1	Blackberry I.	0.30^{d}	0.18^{b}	1.00^{f}	6.50^{e}	30.70^{l}	0.90^{b}	0.40^{b}	0.19^{f}
2	Rose hip I.	0.40^{f}	0.25^{d}	0.90^{a}	7.20^{f}	16.40^{e}	6.80^{k}	0.60^{c}	0.19^{f}
3	Sloe I.	0.59^{g}	0.19^{b}	1.40^{e}	8.00^{g}	16.20^{d}	7.10^{l}	0.30^{a}	0.16^{e}
4	Rose hip II.	1.19^{h}	0.75^{i}	1.20^{d}	4.70^{a}	12.50^{b}	3.10^{h}	1.70^{f}	0.03^{a}
5	Blackberry II.	0.37^{e}	0.42^{h}	1.50^{f}	5.50^{b}	16.10^{c}	1.70^{d}	0.70^{d}	0.02^{a}
6	Sloe II.	0.30^{d}	0.16^{a}	1.20^{d}	9.60^{i}	19.60^{g}	1.10^{c}	0.60^{c}	0.03^{a}
7	Hawthorn	0.59^{g}	0.42^{h}	1.20^{d}	6.40^{d}	26.40^{j}	3.30^{i}	0.70^{d}	0.09^{d}
8	Blackberry III.	0.00^{a}	0.30^{f}	1.10^{c}	6.00^{c}	29.50^{k}	2.60^{g}	0.40^{b}	0.03^{a}
9	Blackberry IV.	0.37^{e}	0.23^{c}	1.40^{e}	5.50^{b}	25.70^{i}	2.30^{e}	0.60^{c}	0.08^{c}
10	Rose hip III.	0.20^{c}	0.27^{e}	1.50^{f}	8.40^{h}	19.40^{f}	2.50f	0.90^{e}	0.08^{cd}
11	Sloe III.	0.09^{b}	0.34^{g}	1.40^{e}	6.00^{c}	12.30^{a}	3.80^{j}	0.70^{d}	0.05^{b}
12	Raspberry	0.20^{c}	0.29^{f}	1.40^{e}	5.50^{b}	21.00^{h}	0.70^{a}	0.70^{d}	0.05^{b}
	Limit value**	**1.00**	**0.1**	**4.00**	**25.00**	**-**	**2.00**	**-**	**0.05**

*** Limit value given by the Food Codex*

Values marked with the same letter are not significantly different (P-value < 0.05)

4. Conclusions

Our results confirmed that the consumption of fresh forest fruit from the environmentally burden region Middle Spis could be considered safe from the aspect of heavy metals content. On the other hand our results indicate that use of dry leaves of forest fruits as components of tea mixtures could be risky for the human health from the aspect of hazardous heavy metals content (Cd, Ni, Hg). Our results also suggest causal relationship among enhanced contents of risk heavy metals, especially Hg, in soils and plants collected in the vicinity of former emission source in Rudnany and the previous metallurgical activity in this area. Residual metallic load of soil and following possible contamination of the food chain in Middle Spis presents a serious potential risk for the human health.

ACKNOWLEDGEMENTS

This work was co-funded by European Community under project No. 26220220180: Building Research Centre “AgroBioTech” and also supported by project: VEGA 1/0308/14.

REFERENCES

1. Angelovicova L., Fazekasova D. (2014) Contamination of the soil and water environment by heavy metals in the former mining area of Rudňany (Slovakia), Soil & Water Res., **9**, 18-24.
2. Angelovicova L., Lodenius M., Tulisalo E. and Fazekasova D. (2014) Effect of Heavy Metals on Soil Enzyme Activity at Different Field Conditions in Middle Spis Mining Area (Slovakia). Bull Environ Contam Toxicol DOI 10.1007/s00128-014-1397-0
3. Balaban, Z.M., Grujic S., Jasic M. and Vujadinovic D. (2012) Testing of chemical composition of wild berries. Third International Scientific Symposium "Agrosym Jahorina.2012". URL: http://www.agrosym.rs.ba/agrosym/agrosym_2012/dokumenti/2_biljna_proizvodnja/13_PP_Zeljka_Marjanovic_Balaban.pdf (accessed 27/01/2015)

4. Barecz P. (2000) Copper giant hits recovery parh. The Slovak Spectator (6th Nov. 2000). URL: http://spectator.sme.sk/c/20007603/copper-giant-hits-recovery-path.html. (accessed 23/01/2015) In: Kozlov M.V., Zvereva E. and Zverev V. (2009). Impacts of Point Polluters on Terrestrial Biota: Comparative analysis of 18 contaminated areas, Springer Science & Business Media, **2009**, pp. 483.
5. Berbecea A., Radulov I., Sala F. and Crista F. (2010) Trace elements and metal bioavailability in soils treated with industrial residues, Research Journal of Agricultural Science, **42**, 25-30.
6. Bradham K.D., Dayton E.A., Basta N.T., Schroder J., Payton M. and Lanno R.P. (2006) Effect of soil properties on lead bioavailability and toxicity to earthworms. Environ Toxicol Chem, **25**, 769-775.
7. Buszewski B., Jastrzębska A., Kowalkowski T. and Górna-Binkul A. (2000) Monitoring of Selected Heavy Metals Uptake by Plants and Soils in the Area of Toruń, Poland. Polish Journal of Environmental Studies, **9**, 511-515.
8. Hanculak J., Bobro M., Sestinova O., Brehuv J. and Slanco P. (2006). Mercury in the environment of old minig areas of Rudnany and Mernik. Acta Montanistica Slovaca, **11**, 295-299.
9. Hazlett P.W., Rutherford G.K. and Van Loon G.W. (1983) Metal contaminants in surface soils and vegetation as a result of nickel/copper smelting at Coniston, Ontario, Canada. Reclamat. Reveg. Res., **2**, 123-137.
10. Krokusova J., Cech V., Kunakova L. and Blahut M. (2013) Impact Of Air Pollution On The Environment In The Krompachy Town (Slovakia). 13th SGEM GeoConference on Energy And Clean Technologies, SGEM2013 Conference Proceedings, June 16-22, 2013, 651-658.
11. Li J., Xie Z., Zhu Y. and Ravi N. (2005) Risk assessment of heavy metal contaminated soil in the vicinity of a lead/zinc mine, J Environ Sci-China, **17**, 881-885.
12. Ogundiran M.B. and Osibanjo O. (2009) Mobility and speciation of heavy metals in soils impacted by hazardous waste, Chem Spec Bioavailab, **21**, 59-69.
13. Trangmar B.B., Yost R.S. and Uehara G. (1985). Application of Geostatistics to Spatial Studies of Soil Properties, Adv Agron, **38**, 45-94.
14. Wieczorek J., Petrzak M., Osowski A. and Wieczorek Z. (2010) Determination of lead, cadmium, and persistent organic pollutants in wild and orchard-farm-grown fruit in northeastern Poland, J Toxicol Environ Health A, **73**, 1236-1243.
15. Wopereis M.C., Gascuel-Odoux C., Bourrie G. and Soignet G. (1988) Spatial variability of heavy metals in soil on a one-hectare scale, Soil Sci, **146**, 113-118.
16. Zhu F., Lu X., Liu L and Mo J. (2015). Phosphate addition enhanced soil inorganic nutrients to a large extent in three tropical forests, Scientific Reports, **5**, doi:10.1038/srep07923