BIOACCUMULATION OF CADMIUM IN EDIBLE WILD MUSHROOM (*SUILLUS LUTEUS*), BULGARIA

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Abstract. The study was conducted on Cambisols soils and wild edible mushroom (*Suillus Luteus*) from the Batak Mountain, Bulgaria. The total cadmium content in the soils was measured after their decomposition with HCl and HNO_3 acids. The mushroom samples were prepared by dry ashing and subsequent dissolution in HCl. The cadmium levels were determined by PerkinElmer AAnalyst 800 model atomic absorption spectrometer (PerkinElmer Instruments, Shelton, CT, USA) equipped with transversely heated graphite atomization (THGA), and a longitudinal Zeeman effect background corrector was also used. A correlation/regression analysis was carried out to reveal possible associations between pH, humus content, and total cadmium content of the soils and the concentration of this element in the mushroom samples. It was calculated BF = 0.38. BF < 1 indicates that the fungus is a metal ion bioreactor.

Keywords: cadmium; *Suillus Luteus*; Bulgaria

Introduction

The wild edible mushrooms are appreciated for their numerous culinary features like taste, aroma, texture, and flavor. Generally, mushrooms are considered a balanced food as they contain considerable amounts of nutrients, such as carbohydrates, proteins (all essential amino acids) and water-soluble vitamins, especially from the B-group, few fatty acids, energy, and lipids (Witkowska et al., 2011; Dewanjee et al., 2013; Kalač, 2013; Petkovšek & Pokorny, 2013; Dospatliev et al., 2018a). Mushrooms are also good sources of macro- and microelements (Dospatliev & Ivanova, 2016; 2017a; 2017b; 2017c). But one must be aware that they are able to accumulate considerable amounts of toxic metals such as lead, cadmium, or mercury. Therefore, regular consumers of mushrooms are characterized by higher cadmium dietary exposure than from other foods[1,2] (Kalač, 2010; Fang et al., 2014; Dua et al., 2015; Wang et al., 2015; Dospatliev & Ivanova, 2017a; Dospatliev et al., 2018b).

Lead and cadmium are metals with a tendency to cumulate in living organisms. Lead pollution of soils is observed mainly in industrial areas. Exposure to lead is related to a wide range of health effects, including reduced intellectual development in children and increased blood pressure and cardiovascular diseases in adults.[1,2] (Dospatliev & Ivanova, 2017e). Cadmium is a natural constituent of earth crust. As a result of human activity, the amounts of cadmium in the soil increase. The main source of Cd exposure for non-smokers is food (90%), particularly cereals and vegetables. Exceeding cadmium can negatively affect kidney function and cause skeletal and reproductive disorders[1,2] (Dospatliev & Ivanova, 2017e). Generally, mushrooms picked in uncontaminated areas are characterized by lower contents of heavy metals, when compared to mushrooms growing in contaminated soils. Heavy metal contents in mushrooms are largely dependent on their trophic pattern, physiology of mushroom species, area of sample collection, mushroom accumulation of other metals, and the distance from the pollution sources. Moreover, the age of mycelium and lag between fructification seem to be further factors affecting metals content. Some mushroom species like Boletus edulis tend to accumulate metals due to the presence of Cd-binding proteins (Guerra et al., 2011; Sarikurkcu et al., 2011; Chew et al., 2016; Dospatliev and Ivanova, 2017e).

The aims of this study were to determine Cd contents of wild mushroom *Suillus luteus* growing in the Batak mountain, and thus to Calculation of Bioaccumulation factor (BF).

Materials and methods

The study was conducted on Cambisols soils with wild edible mushrooms. Fifteen Cambisols soil samples were taken from a depth of 0 – 20 cm. The following Cambisols soil characteristics were determined: pH in water, humus according to Turin (Tanov et al., 1978), total content of cadmium through decomposition by HCl and HNO_3 acids, following ISO (International Organization for Standardization) 11464 standard.[3]

Mushroom samples were collected in 2018 from the Batak mountain. Samples of mushroom and Cambisols soil was taken from the same place. All samples were washed so as to remove any adhering soil particles and rinsed with distilled water, after which they were dried at 105°C for 24 h and ground.

The preparation of mushroom samples was made by means of dry ashing and dissolution in HCl acids. The cadmium levels were determined by PerkinElmer AAnalyst 800 model atomic absorption spectrometer (PerkinElmer Instruments, Shelton, CT, USA) equipped with transversely heated graphite atomization (THGA), and a longitudinal Zeeman effect background corrector was also used. Lumina lamp, electrodeless discharge lamp (EDL) and hollow cathode lamps were employed as radiation sources. The operating parameters for working elements by graphite furnace were set as recommended by the application note. Atomic absorption spectrophotometer (AAS) graphite furnace operating parameters and temperature programme are given in Table 1, respectively.

Table 1. AAS parameters used

Parameters	Cd
Lamp settings wavelength (nm)	228.8
Spectral band width (nm)	0.7
Lamp current (mA)	6
Furnace temperature (°C)	
Drying 1 (ramp 1 s, hold 5 s)	110
Drying 2 (ramp 5 s, hold 15 s)	130
Ashing (ramp 10 s, hold 30 s)	350
Atomising (ramp 0 s, hold 5 s)	1500
Clean-up (ramp 1 s, hold 3 s)	2450

Samples were injected into the graphite tubes using a PerkinElmer AS-800 Autosampler. The atomic absorption signal was determined in peak height mode against a calibration curve.

Bioaccumulation factor (BF)

Bioaccumulation factor (BF): Bioaccumulation factor of heavy metals in mushroom fruit body was calculated as:

$$BF = \frac{C_{mushroom}}{C_{soil}} \quad (1)$$

where C represents the metal concentrations in fruit and soil.

Statistical processing

SPSS (Statistical Package for Social Science) program for Windows was used for statistical data processing.

Results and discussion

Soils

The total cadmium content in Cambisols soil ranges from 0.84 to 1.58 mg kg^{-1}. According to the requirements of the Bulgarian standards for allowable cadmium content in the Cambisols soil depending on the active soil reaction (pH) e below the limit concentration (0.8 – 1.5 mg kg^{-1}). The arithmetic mean is $\bar{X} = 1.17$ mg kg^{-1}, as its value is greater than the median (Me = 1.14), therefore there is a right-tail distribution. This is proved by positive coefficient of skewness (Sk = 0.37). The coefficient of kurtosis is negative (Kr = - 1.25), i.e. observed low peak height of the distribution. Not greater dispersion of the values of Cd, the mean value cadmium to no higher values of standard deviation (σ = 0.26) and coefficient of variation, which reaches 22.22% (Table 2).

Table 2. Soil properties, content of Cd in Cambisols soil and mushrooms (n = 15)

Statistical index	pH	Humus	Total content of Cd in soil, mg kg^{-1}	Content of Cd in Suillus Luteus, mg kg^{-1}	BF
Mean	5.76	2.60	1.17	0.43	0.38
Standard Error	0.17	0.14	0.07	0.03	0.02
Median	5.85	2.68	1.14	0.44	0.36
Mode	5.45	3.15	0.96	0.44	N/A
Standard Deviation	0.65	0.56	0.26	0.10	0.09
Sample Variance	0.42	0.31	0.07	0.01	0.01
Kurtosis	-1.52	-1.38	-1.25	-1.35	-0.42
Skewness	-0.15	-0.16	0.37	0.14	-0.02
Range	1.84	1.70	0.74	1.28	0.32
Minimum	4.81	1.75	0.84	0.30	0.20
Maximum	6.65	3.45	1.58	0.58	0.52
Sum	86.41	39.06	17.57	6.51	5.71
Count	15.00	15.00	15.00	15.00	15.00
Con. Level (95.0%)	0.36	0.31	0.14	0.58	0.52
CV, %	11.28	21.54	22.22	23.25	23.68

Mushrooms

The levels of cadmium found in the samples varied depending on the species of mushroom, the sample collection site and the cadmium content of the soil at the collection site. At least four factors could affect cadmium concentrations of the edible mushrooms: species, ecology (saprophyte, wood decaying and mycorrhizal), morphological parts and physical properties of the soil. These factors influence metal concentrations and thus Bioaccumulation factor (BF). BF measures the relationship between the metal concentration in mushrooms and the metal concentration in the underlying soil where mushrooms grow; a BF > 1 indicates an accumulation of metal ions by the mushrooms while a BF < 1 shows that the mushrooms are bioexclusors of the metal ions (Sun et al., 2017) (Table 2).

The total cadmium content in the samples of the mushrooms species varied in the range from 0.30 to 0.58 mg kg^{-1}. These values are several times lower than those specified in The Annex to Regulation (EC) No 1881 (2006) having in mind that the average water content in our samples was 90%. These results comply with the data published by other authors (Brzezicha-Cirocka et al., 2016; Stefanović et al., 2016; Mleczek et al., 2016; Melgar et al., 2016; Dospatliev & Ivanova, 2017a; Gąsecka et al., 2017).

Cadmium contents in the most of edible mushroom species growing in unpolluted areas are below 2 mg kg^{-1} dry matter. However, levels in *Boletus aestivalis*, *Leccinum scabrum*, *Calocybe gambosa*, *Armillaria mellea* and *Russula cyanoxan-*

tha can be up to 5 mg kg^{-1} dry matter and in the genus Agaricus up to 50 mg kg^{-1} dry matter, mainly in Agaricus species (Kalač, 2010).

Heavy metal contents in mushrooms are largely dependent on their trophic pattern, hysiology of mushroom species, area of sample collection, mushroom accumulation of other metals, and the distance from the pollution sources. Moreover, the age of mycelium and lag between fructification seem to be further factors affecting metals content (Peña-Fernández et al., 2014).

Cadmium is accumulated mainly in kidneys, spleen and liver and its blood serum level increases considerably following mushroom consumption (Dospatliev & Ivanova, 2017b). Thus, cadmium seems to be the most deleterious among heavy metals in the mushrooms. It accumulates in bones and can take in the role of calcium. The average Cd present in the studied wild mushrooms was 0.43 mg kg^{-1}, which was far below the 2 mg kg^{-1} for dry matter limit set.[2)]

Correlation dependencies among pH, humus,and total quantities of Cd in cambisols soil and in the mushrooms.

Correlation coefficients among soil parameters and concentration of cadmium in mushroom are summarized in Figs. 1 – 3.

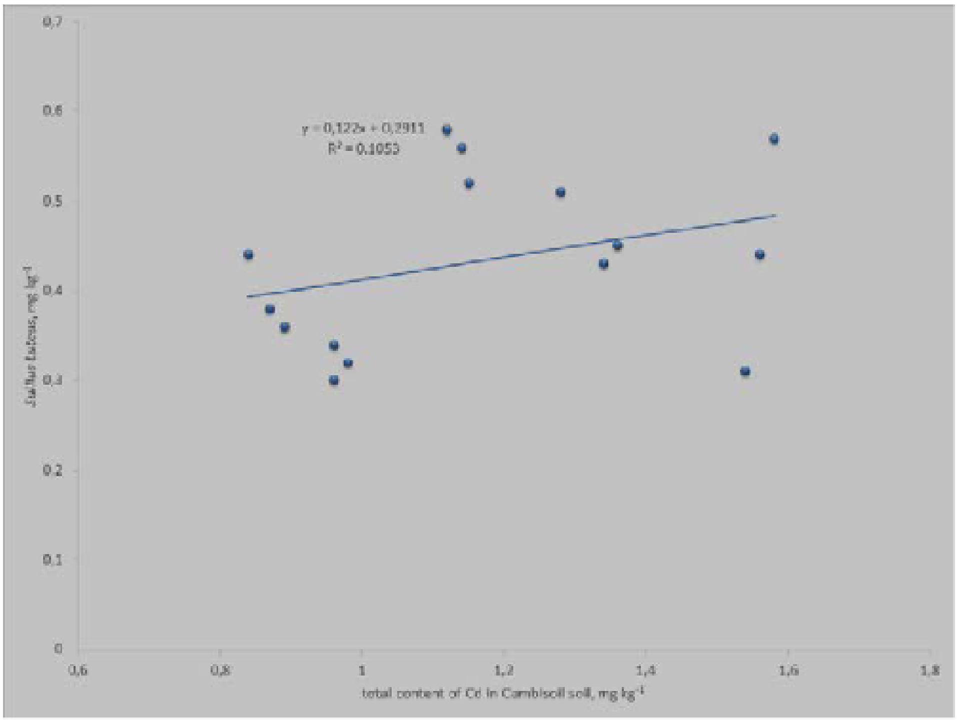

Figure 1. Correlation between the total Cd content in Cambisols soil and Cd content in mushrooms

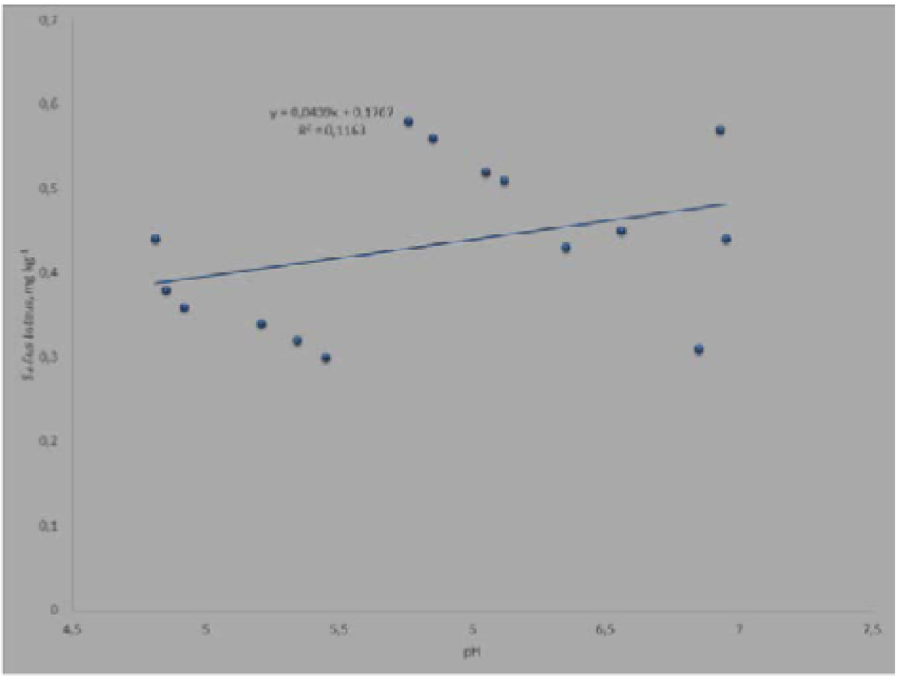

Figure 2. Correlation between the Cambisols soil pH and Cd content in mushrooms

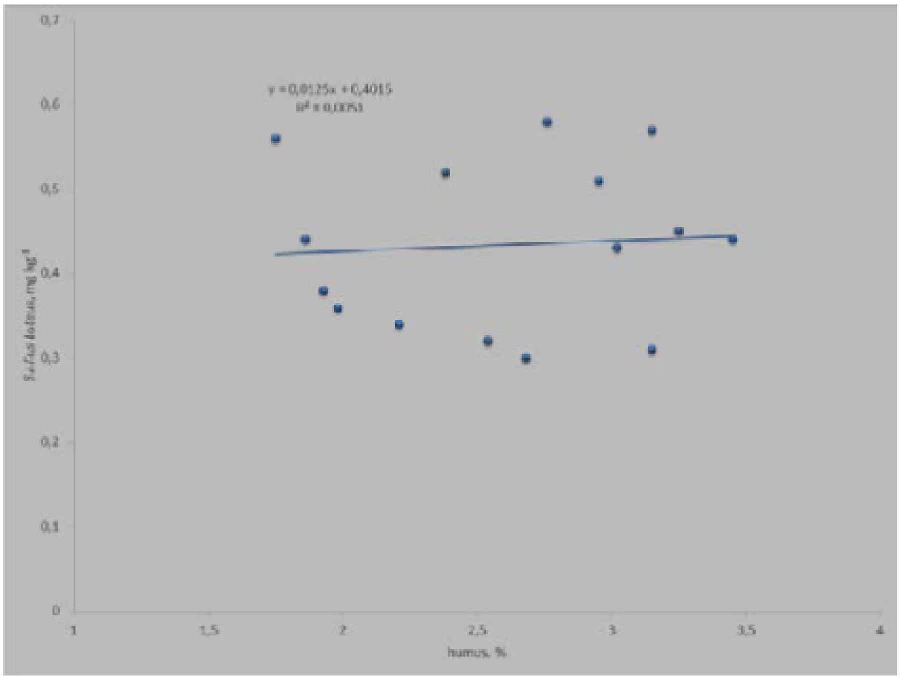

Figure 3. Correlation between the Cambisols soil humus and Cd content in mushrooms

The results of the conducted correlation/regression analysis show that there are very few statistically significant dependencies determined between the total forms and Cambisols soil pH and cadmium concentration in the mushrooms.

Cadmium concentration in mushrooms increases linearly with the increase of the total element content in the Cambisols soil. Determination coefficient show that nearly 11% of the cadmium concentration in mushrooms depend on the total cadmium content in the Cambisols soils.

Conclusions

The results of the conducted correlation/regression analysis show that there are very few statistically significant dependencies determined between the total forms and Cambisols soil pH and cadmium concentration in the mushrooms.

It was calculated BF = 0.38. BF < 1 indicates that the fungus is a metal ion bioreactor.

From the obtained concentrations of heavy metals one can say that the locality Batak mauntain is ecologically clean area and very suitable for collecting wild edible mushrooms that we can use in our daily menu.

NOTES

1. https://ec.europa.eu/food/safety/chemical_safety/contaminants/catalogue/cadmium_en
2. https://www.who.int/foodsafety/areas_work/chemical-risks/jecfa/en/
3. https://www.iso.org/standard/37718.html

REFERENCES

Brzezicha-Cirocka, J., Mędyk, M., Falandysz, J. & Szefer, P. (2016). Bio- and toxic elements in edible wild mushrooms from two regions of potentially different environmental conditions in eastern Poland. *Environ. Sci. Pollut Res.*, *23*, 21517 – 21522.

Chew, G., Sim, L., Ng, S., Ding, Y., Shin, R. & Lee, T. (2016). Development of a mushroom pwder certified rference material for calcium, arsenic, cadmium and lead measurements. *Food Chem.*, *190*, 293 – 299.

Dewanjee, S., Gangopadhyay, M., Sahu, R. & Karmakar, S. (2013). Cadmium induced pathophysiology: prophylactic role of edible jute (Corchorus olitorius) leaves with special emphasis on oxidative stress and mitochondrial involvement. *Food Chem. Toxicol.*, *60*, 188 – 198.

Dospatliev, L. & Ivanova, M. (2016). ICP determination of Pb, Mn, Cu, Zn, Fe, K, Na, P, Mg and Ca in edible wild mushroom (Boletus pinophilus). *Int. J. Sci. & Res. Publ.*, *6*, 266 – 269.

Dospatliev, L. & Ivanova, M. (2017a). Determination of heavy metals in mushroom samples by atomic absorption spectrometry. *Bulg. Chem. Commun.*, *49*, 5 – 9.
Dospatliev, L. & Ivanova, M. (2017b). ICP determination of Pb, Mn, Cu, Zn, Fe, K, Na, P, Mg and Ca in edible wild mushroom Cantharellus Cibarius. *Appl. Res. Technics Technologies & Education*, *5*, 66 – 71.
Dospatliev, L. & Ivanova, M. (2017c). Spectrophotometric investigations on liquid-liquid extraction systems containing cobalt and tetrazolium salts: application of the developed method for analysis of the cobalt content of biological samples (mushrooms and tobaccos). *Bulg. Chem. Commun.*, *49*, 787 – 791.
Dospatliev, L. & Ivanova, M. (2017d). Concentrations and risk assessment of lead and cadmiumin wild edible mushrooms from the Batak mountain, Bulgaria. *Oxid. Commun.*, *40*, 993 – 1001.
Dospatliev, L. & Ivanova, M. (2017e). Boletus pinophilus growing in Bulgaria: chemical profile and hazard index. *C. R. Acad. Bulg. Sci.*, *70*, 795 – 802.
Dospatliev, L., Ivanova, M., Petkova, Z., Angelova-Romov, M. & Antova, G. (2018a). Fatty acid composition in medicinal mushrooms (Craterellus tubaeformis). *Appl. Res. Technics Technologies & Education*, *6*, 35 – 39.
Dospatliev, L., Ivanova, M. & Gavazov, K. (2018b). Correlation between cambisols soil characteristics and lead content in wild edible mushrooms (Cantharellus cibarius, Tricholoma equestre, Craterellus cornucopioides). *Bulg. Chem. Commun.*, *50*, 32 – 37.
Dua, T., Dewanjee, S., Khanra, R., Bhattacharya, N., Bhaskar, B., Zia-Ul-Haq, M. & De Feo, V. (2015). The effects of two common edible herbs, Ipomoea aquatic and Enhydra fluctuans, on cadmium-induced pathophysiology: a focus on oxidative defence and anti-apoptotic mechanism. *J. Transl. Med.*, *13*, 245 – 264.
Fang, Y., Sun, X., Yang, W., Ma, N., Xin, Z., Fu, J., Liu, M., Mariga, A.M., Zhu, X. & Hu, Q. (2014). Concentration and health risks of lead, cadmium, arsenic and mercury in rice and edible mushroom in China. *Food Chem.*, *147*, 147 – 151.
Gąsecka, M., Rzymski, P., Mleczek, M., Siwulski, M., Budzyńska, S., Magdziak, Z., Niedzielski, P. & Sobieralski, K. (2017). The relationship between metal composition, phenolic acid and flavonoid content in Imleria badia from non-polluted and polluted areas. *J. Environ. Sci. Health Part B*, *52*, 171 – 177.
Guerra, F., Trevizam, A.R., Muraoka, T., Marcante, N.C. & Canniatti-Brazaca, S.G. (2011). Heavy metals in vegetables and potential risk for

Kalač, P. (2010). Trace element contents in European species of wild growing edible mushrooms: a review for the period 2000-2009. *Food Chem.*, *122*, 2 – 15.

Kalač, P. (2013). A review of chemical composition and nutritional value of wild-growing and cultivated mushrooms. *J. Sci. Food Agric.*, *93*, 209 – 218.

Melgar, M.J., Alonso, J. & García, M.A. (2016). Cadmium in edible mushrooms from NW Spain: bioconcentration factors and consumer health implications. *Food Chem. Toxicol.*, *88*, 13 – 20.

Mleczek, M., Niedzielski, P., Kalač, P., Budka, M., Siwulski, M., Gąsecka, M., Rzymski, P., Magdziak, Z. & Sobieralski, K. (2016). Multielemental analysis of 20 mushroom species growing near a heavily trafficked road in Poland. *Environ. Sci. Poll. Res.*, *23*, 16280 – 16295.

Peña-Fernández, A., González-Muñoz, M.J. & Lobo-Bedmar, M.C. (2014). Establishsing the importance of human health risk assessment for metals and metalloids in urban environments. *Environ. Int.*, *72*, 176 – 185.

Petkovšek, A.S. & Pokorny, B. (2013). Lead and cadmium in mushrooms from the vicinity of two large emission sources in Slovenia. *Sci. Total Environ.*, *443*, 944 – 954.

Sarikurkcu, C., Copur, M., Yildiz, D. & Akata, I. (2011). Metal concentration of wild edible mushrooms in Soguksu National Park in Turkey. *Food Chem.*, *128*, 731 – 734.

Stefanović, V., Trifković, J., Mutić, J. & Tešić, Ž. (2016). Metal accumulation capacity of parasol mushroom (Macrolepiota procera) from Rasina region (Serbia). *Environ. Sci. Poll. Res.*, *23*, 13178 – 13190.

Sun, L., Chang, W., Bao, C. & Zhuang, Y. (2017). Metal contents, bioaccumulation, and health tisk assessment in wild edible Boletaceae mushrooms. *J. Food Sci.*, *82*, 1500 – 1508.

Tanov, E., Lukanov, K., Miljanchev, I., Penchev, P., Andonov, A. & Konarev, A. (1978). *District-division, concentration and specialisation of tobacco-cultivation and tobacco-processing in Bulgaria*. Plovdiv: Hristo G. Danov.

Wang, X.-M., Zhang, J., Li, T., Wang, Y.-Z. & Liu, H.-G. (2015). Content and bioaccumulation of nine mineral elements in ten mushroom species of the Genus Boletus. *J. Anal. Methods Chem.*, *2015,* art. no. 165412.

Witkowska, A.M., Zujko, M.E. & Mirończukchodakowska, I. (2011). Comparative study of wild edible mushroms as sources of antioxidants. *Int. J. Med. Mushrooms*, *13*, 335 – 341.