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BALANCING PHYTOREMEDIATION AND FOOD SAFETY: ASSESSING HEAVY METAL ACCUMULATION IN SELECTED PLANTS FROM TWO LOCALITIES IN SERBIA

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ABSTRACT. This study explored the relationship between soil properties and heavy metal concentrations in soil and selected plants from Kopaonik National Park and the Aleksandrovac municipality in Serbia. Soil samples were analyzed for texture, basic chemical properties, and the plant-available fraction of various metals, while plant material was assessed for macro- and microelement content. Significant differences were observed between the localities, both in soil properties and plant metal accumulation. *Prunus domestica* L. exhibited strong bioaccumulation efficiency for most microelements, making it a significant candidate for phytoremediation. Notable accumulation of sodium, potassium, iron, nickel, copper, zinc, and chromium was observed in *Capsicum annuum* L., while *Juniperus communis* L. efficiently accumulated magnesium, calcium, manganese, molybdenum, and cobalt. Due to their capacity to bioaccumulate heavy metals, especially cadmium, these species highlight a dual role in phytoremediation efforts and underscore the need for careful agricultural management to mitigate consumer health risks in contaminated areas.

Keywords: phytoremediation, heavy metals, food safety, bioaccumulation potential, soil contamination.

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INTRODUCTION

In recent decades, numerous sources of pollution, including heavy metals, pesticides, and radioactive particles, have significantly increased their impact on the environment worldwide. These pollutants primarily originate from human activities such as mining, industrial processes, fossil fuel combustion, agricultural practices, vehicle emissions, and military operations (REICHMAN, 2002; JAISHANKAR *et al.*, 2014). These pollutants pose major health risks to both humans and wildlife because they enter the environment, contaminate the soil, water, and air, and then infiltrate the food chain.

Heavy metals are of particular concern due to their non-biodegradable nature and tendency to accumulate in living organisms. These metals, including lead (Pb), nickel (Ni), chromium (Cr), cadmium (Cd), mercury (Hg), zinc (Zn), copper (Cu), and molybdenum (Mo), have a high atomic weight and specific density, making them potentially toxic at elevated concentrations (HAWKES, 1997; TIMOTHY and WILLIAMS, 2019). Although some heavy metals, like zinc, copper, and molybdenum, are essential in trace amounts for plant metabolism and enzyme function, excessive concentrations can disrupt crucial physiological and biochemical processes, leading to oxidative stress, inhibition of photosynthesis, and disruptions in nutrient uptake in plants. This results in reduced plant growth, decreased crop yield, and lower quality (NAGAJYOTI *et al.*, 2010; DOĞANLAR and ATMACA, 2011).

As an essential component of terrestrial ecosystems, soil plays a central role in supporting plant growth, regulating water cycles, and sustaining agricultural productivity. However, the continuous degradation of soil quality due to pollution and other factors poses a significant threat to food security and environmental sustainability (KLJAJIĆ *et al.*, 2012). Key soil properties, such as humus content, calcium carbonate levels, pH, and texture, critically influence its fertility by affecting nutrient availability and retention. Pollutants, especially heavy metals, can alter soil composition, reduce its microbial diversity, and impair nutrient cycling, leading to decreased plant health and diminished ecosystem functioning. Over time, such changes can disrupt agricultural productivity, reduce crop yields, and lead to the loss of biodiversity, further threatening ecosystem resilience and the ability to provide essential ecosystem services.

Some plant species, referred to as hyperaccumulators, have developed unique defenses that enable them to withstand and accumulate elevated levels of heavy metals in their tissues without suffering harmful consequences. The potential application of these plants in phytoremediation—a sustainable and economical method of cleaning up contaminated soils—has attracted a lot of attention. Utilizing plants, phytoremediation eliminates, stabilizes, or breaks down environmental pollutants, including inorganic pollutants like heavy metals and organic pollutants like pesticides (MCGRATH *et al.*, 2002). Furthermore, according to MALIZIA *et al.* (2012), these plants function as bioindicators of soil pollution, offering important information about contamination levels and assisting in determining the effects of pollutants on the environment. Hyperaccumulators are essential parts of ecological restoration and pollution control initiatives because of their dual function in environmental monitoring and remediation.

This study aims to comprehensively assess the impact of metal pollution on soil and plant health in the municipality of Aleksandrovac and the Kopaonik National Park. Specifically, we examined the mechanical composition and chemical properties of the soil, analyzed the total and plant-available macro- and microelement content in the soil, and evaluated the macro- and microelement content as well as the bioaccumulation coefficient in plant material (stem and leaf) of four edible plant species: *Prunus domestica* L., *Rubus fruticosus* L., *Capsicum annuum* L., and *Juniperus communis* L. The findings of this study will contribute to a better understanding of metal pollution in the environment and highlight strategies for sustainable soil and plant management, emphasizing the importance of implementing practices that balance phytoremediation with food safety to reduce heavy metal pollution.

MATERIALS AND METHODS

The research method used to obtain the results presented in this paper involved field and laboratory work. The fieldwork required the collection of soil and plant material. Sampling and analysis of the material was carried out in the period from October 2022 to June 2023.

Investigated localities

Parčin is a village located in the municipality of Aleksandrovac, Rasina District, at an altitude of 541 meters (43°25'13"N, 21°05'13"E). Soil samples were collected from two sites in the village: Parčin 1 (P1) (Fig. 1A) (43°24'56"N, 21°05'06"E) and Parčin 2 (P2) (Fig. 1B) (43°25'09"N, 21°04'00"E). At P1, samples of *Prunus domestica* L. (stem and leaf) were collected, while at P2, *Rubus fruticosus* L. (stem and leaf) samples were gathered. These samples were used to evaluate the metal concentration and soil characteristics as well as how they affected the bioaccumulation of metals in plant tissues at each location.



Figure 1. Localities of sampled soil and plant materials in the village of Parčin: A) Parčin 1 (P1) and B) Parčin 2 (P2)

Kopaonik Mountain is the largest mountain massif in southern Serbia, stretching from northwest to southeast over approximately 75 km. Soil and plant material were sampled from two locations within Kopaonik National Park. At the first site, located in the village of Rudnice (Fig. 2A) (43°14'20"N, 20°40'56"E), referred to as Kopaonik 1 (K1), soil samples and the plant *Capsicum annuum* L. (stem and leaf) were collected. At the second site on Kopaonik Mountain (Fig. 2B), marked as Kopaonik 2 (K2), located at 43°16'28"N, 20°47'08"E, soil, stem, as well as leaf samples of *Juniperus communis* L. were gathered. These samples were used to evaluate the metal content and bioaccumulation potential of the selected plant species at both locations.



Figure 2. Localities of sampled soil and plant materials sampled in Kopaonik National Park: A) Kopaonik 1 (Rudnica village) (K1) and B) Kopaonik 2 (K2)

Investigated plant species

In this study, four plant species were examined for the presence and concentrations of macro- and microelements in their tissues (Fig. 3). The results obtained were compared and statistically analyzed. The species investigated in this research are: *Prunus domestica* L. (plum), *Rubus fruticosus* L. (blackberry), *Capsicum annuum* L. (pepper), and *Juniperus communis* L. (juniper).

In this analysis, the E variation of the pyrophosphate method was used to prepare the sample, and the pipette method was used to determine the soil mechanical fractions of different sizes (BOŠNJAK *et al.*, 1997). By precisely separating soil particles according to size, the pipette method makes it possible to evaluate soil texture in great detail, which is essential for comprehending the physical characteristics and fertility of the soil.



Figure 3. Investigated plant species: A) *Prunus domestica* L. (plum), B) *Rubus fruticosus* L. (blackberry), C) *Capsicum annuum* L. (pepper), and D) *Juniperus communis* L. (juniper) (https://sr.wikipedia.org)

Soil texture analysis

Analysis of basic soil chemical properties

The chemical properties of soil determine its ability to supply plants with available nutrients and affect its physical properties and the health of its living population (SPOSITO, 1984). The main soil chemical characteristics were analyzed using the following methods: soil organic carbon (SOC) by the dichromate method, humus content was calculated = SOC x 1.72 (ROWELL, 1997); soil pH was measured potentiometrically in a suspension (soil/water and soil/1 M KCl, 1/2.5) (VAN REEUWIJK, 2002); total acidity (H) was determined by Kappen's method with 1 M Na-acetate solution (MINEEV *et al.*, 2001); calcium carbonate (CaCO₃) content was measured by the volumetric method with a calcimeter (NELSON, 1982); cation exchange capacity (CEC) and exchangeable bases (S) were determined by extraction with 1 M ammonium acetate (AAc) buffered at pH 7 (ALLEN *et al.*, 1974); base saturation (V) was calculated from S and CEC.

Determination of plant-available and total macro and microelements in the soil

To determine the plant-available fraction of various elements, ammonium lactateammonium acetate (AL) was used for phosphorus (P), while ammonium acetate (AAc) was utilized for magnesium (Mg), calcium (Ca), potassium (K), and sodium (Na) (ALLEN *et al.*, 1974). A buffered solution of diethylenetriaminepentaacetic acid (DTPA) and triethanolamine (TEA) at pH 7.3 was employed for other metals, including molybdenum (Mo), manganese (Mn), iron (Fe), nickel (Ni), copper (Cu), zinc (Zn), chromium (Cr), lead (Pb), cadmium (Cd), and cobalt (Co). Pseudototal concentrations of Mo, Mn, Fe, Ni, Cu, Zn, Cr, Pb, Cd and Co 6

were extracted by microwave digestion with concentrated HNO₃ (LINDSAY and NORVELL, 1978). The concentrations of chemical elements in the fine soil samples subjected to different extraction procedures were determined by inductively coupled plasma optical emission spectroscopy (ICP-OES, SpectroGenesis EOP II, Spectro Analytical Instruments GmbH, Kleve, Germany). However, phosphorus concentrations were specifically determined by the colorimetric molybdenum blue method at a wavelength of 580 nm.

Determination of macro- and microelements in plant material

The total content of mineral elements in the plant material was determined by microwave digestion of 0.2 g of homogenized dry sample with the addition of 3 mL of concentrated HNO₃ (LINDSAY and NORVELL, 1978) and 2 mL of 30% H_2O_2 . The concentration of mineral elements was determined by ICP-OES.

Bioaccumulation factor

The bioaccumulation factor (BCF) is an index that reflects a plant's ability to accumulate a specific metal relative to its concentration in the substrate. It is calculated as the ratio of the metal concentration in the plant (or its parts) to the concentration in the soil (GHOSH and SINGH, 2005). BCF values greater than 1 indicate that the metal concentration in the organism exceeds that in the substrate, suggesting effective bioaccumulation capacity of the tested organism (https://www.epa.gov/). The equation for calculating the bioaccumulation factor is:

BCF = element concentration in plant / element concentration in soil

Statistical data processing

The statistical analysis of the data included the calculation of the mean value (M) and standard deviation (SD) for each analyzed parameter in the soil and the examined plant species at different localities. Analysis of variance (ANOVA) and Scheffé's post-hoc test (p < 0.05) were used to assess variability and identify statistically significant differences in the parameters related to soil chemical characteristics, total content of chemical elements, and the available concentrations of micro- and macroelements in the rhizosphere soil, as well as in the stems and leaves of the tested plant species. Statistical data analysis was performed using Statistica version 10 (StatSoft Inc., 2011).

RESULTS AND DISCUSSION

Texture of investigated soils

The mechanical composition of the soil, encompassing the relative proportions of sand, silt, and clay particles, is a fundamental aspect of soil characterization. These fractions have a major influence on soil properties such as texture, structure, porosity, water-holding capacity, and nutrient availability (BRADY and WEIL, 2008; UPADHYAY and RAGHUBANSHI, 2020). Understanding mechanical composition is essential for various applications, including agriculture, engineering, and environmental management. Based on the mechanical composition, soil is categorized into specific textural classes (ĐORĐEVIĆ and RADMANOVIĆ, 2018). In the study discussed, the mechanical composition was analyzed for soil samples from different localities, and distinctive characteristics were identified.

The results of the analysis of the content of mechanical fractions (Table 1) in the tested samples are expressed as percentages. The percentages of various fractions differed

significantly between the localities, indicating variations in soil texture and structure. The results of this study showed that the soil samples from the locality K1 contain the fraction of fine sand and coarse silt in the highest percentage. The soil samples from the K2 locality contained skeleton and coarse sand in the highest percentage. The highest percentage of fine silt, clay and physical clay was determined in the soil sampled at locality P1.

	Content of mechanical fractions in %								
ole	Skeleton	Coarse	Fine	Coarse	Fine	Clay	Physical		
lui		sand	sand	silt	silt		clay		
Sa	>2	2-0.2	0.2-0.05	0.05-0.02	0.02-0.002	< 0.002	<0.02 (mm)		
	(mm)	(mm)	(mm)	(mm)	(mm)	(mm)			
K1	ND	27.91	30.89	17.81	15.36	8.04	23.40		
<i>K2</i>	34.1	43.02	19.64	10.82	16.68	9.84	26.52		
P1	ND	8.74	8.29	14.31	35.31	33.35	68.67		
<i>P2</i>	ND	8.89	11.19	16.52	33.36	30.04	63.40		

Table 1. Mechanical composition of the examined soils

ND – not detected

The results indicate that the soil samples from localities K1, P1, and P2 can be classified as non-skeletal soils, whereas the K2 soil is categorized as highly skeletal, following Gračanin's classification (GRAČANIN, 1945). Based on the texture classification for fine soils (NATURAL RESOURCES CONSERVATION SERVICE, 2004) and the analyzed data, it can be concluded that the soils from localities K1 and K2 are sandy loam, while P1 is classified as silty clay loam, and P2 as clay loam.

Soil texture influences water infiltration, drainage, and rooting, and affects plant growth and agricultural practices (SPOSITO, 2013). Sandy soils have good drainage but lower water retention and are therefore suitable for crops that prefer drier conditions. Clay soils, with their high water retention capacity but poor drainage, are suitable for different crops. Additionally, the presence or absence of skeleton material, as indicated in the analysis, is crucial. Skeleton-free soils, such as those at localities K1, P1, and P2, have different properties compared to highly skeletal soils such as that at locality K2. Skeleton material affects soil structure and stability, influencing erosion and nutrient availability (BLUME *et al.*, 2010).

Furthermore, the analysis provides insights into the clay content, which is essential for understanding soil fertility and cation exchange capacity. Soils with a higher clay content generally have a greater nutrient-holding capacity, but too high clay content can lead to compaction and poor root development (BRADY and WEIL, 2008). In conclusion, the mechanical composition of the soil is a critical factor in determining soil properties and suitability for various uses. A detailed analysis, such as one conducted in this study, provides valuable information for land management decisions, agricultural practices, and environmental conservation efforts.

Basic chemical characteristics of the investigated soil

The chemical properties, including reaction (pH), content of carbonates, hydrolytic acidity, and adsorptive complex characteristics, play a crucial role in determining soil fertility, nutrient availability, and overall soil health (SPOSITO, 1984; LOEPPERT and SUAREZ, 1996). The results of the analysis of the chemical characteristics of the investigated soil are presented in Table 2.

Soil reaction, expressed in pH values, indicates the acidity or alkalinity of the soil, which influences nutrient availability to plants. The study revealed that the soil at locality K1 was slightly alkaline, while that at locality P1 was acidic. These findings have implications for crop selection and the need for soil amendments to optimize pH for plant growth (DJORDJEVIĆ *et al.*, 2022).

Carbonates, which are important for buffering the pH value and nutrient availability in the soil, were absent in the soils of localities K2 and P1, classifying these soils as non-calcareous. In contrast, the soils at localities K1 and P2 were classified as slightly calcareous (PELIŠEK, 1964), suggesting differences in soil chemistry and potential impacts on nutrient availability and soil structure (LOEPPERT and SUAREZ, 1996). Hydrolytic acidity, a measure of soil acidity, was highest at locality P1, suggesting that lime needs to be applied to neutralize acidity and improve soil fertility (KORUNOVIĆ, STOJANOVIĆ, 1989). This finding underlines the importance of understanding soil acidity levels for effective soil management practices (DJORDJEVIĆ *et al.*, 2022).

All analyzed soils were well-supplied with humus, with the K1 soil exhibiting a particularly high humus content, classifying it as very rich. In contrast, the soils from the other localities showed moderate humus levels.

Profile	K1	K2	P1	P2
Hummus (%)	7.70 (0.25) a ^{***} b ^{***} c ^{***}	4.11 (0.10) d ^{ns} e ^{ns}	4.24 (0.11) f*	3.59 (0.08)
рН (H2O)	$7.58 (0.50) a^{***} b^{***} c^{ns}$	5.56 (0.40) d ^{ns}	5.00 (0.44)	7.99 (0.56) e ^{***} f ^{***}
pH (KCl)	$\begin{array}{c} 7.11 \ (0.40) \\ a^{***} b^{***} c^{ns} \end{array}$	5.00 (0.35) d ^{ns}	4.62 (0.27)	7.16 (0.41) e ^{***} f ^{***}
CaCO3 (%)	2.06 (0.19)	ND	ND	3.20 (0.11) c***
H (cmol/kg)	ND	6.36 (0.10)	8.99 (0.14) d ^{***}	ND
S (cmol/kg)	29.60 (2.10) a***	9.19 (0.76)	37.66 (3.12) b ^{***} d ^{***}	60.00 (4.16) c ^{***} e ^{***} f ^{***}
CEC (cmol/kg)	30.60 (2.73) a ^{****}	15.60 (1.85)	46.65 (2.99) b ^{***} d ^{***}	60.00 (3.17) c ^{***} e ^{***} f ^{***}
V (%)	100.00	59.10	80.70	100.00

Table 2. Chemical characteristics of the soils investigated at different localities

Analysis of variances (ANOVA) and Scheffé's post-hoc test; results are presented as mean and standard deviation -M (SD), n=5; (a) K1 – K2, (b) K1 – P1; (c) K1 –P2; (d) K2 – P1; (e) K2 – P2; (f) P1 – P2;

* p<0.05, *** p<0.001, ns – no statistically significant differences, ND – not detected.

The adsorptive complex of the soil, characterized by parameters such as exchangeable bases (S), cation exchange capacity (CEC), and base saturation (V), influences nutrient retention and availability. Soils from localities P1 and P2 had higher S and CEC values, indicating greater nutrient retention capacity, while the degree of soil saturation with adsorbed base cations (V) was highest at localities K1 and P2, indicating higher nutrient availability (BRADY and WEIL, 2008). In order to improve soil fertility and increase crop productivity, a thorough examination of soil chemical properties provides essential information for efficient soil management techniques, such as optimal nutrient management, suitable liming techniques, and well-informed crop selection.

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Differences in the mechanical and chemical composition indicate significant geological and ecological variations within the Kopaonik region, which may influence the potential use of the soil in forestry and agriculture. These findings align with results reported by other researchers in studies conducted within the Kopaonik National Park (KADOVIĆ *et al.*, 2009; KOŠANIN *et al.*, 2013; MIŠLJENOVIĆ *et al.*, 2020). The soils analyzed in Aleksandrovac by GULAN *et al.* (2021) are characterized by a higher sand content and lower silt content, while the clay content is comparable to that of the soils examined in this study. Additionally, the soils in GULAN *et al.* (2021) research exhibited lower levels of humus and carbonates compared to those in the present study. The pH values reported by GULAN *et al.* (2021) ranged from 4.79 to 6.44, indicating a narrower variation compared to the pH range observed in this study.

Content of bioavailable macro- and microelements in the soil

Understanding soil fertility and how it can promote plant growth requires analyzing the bioavailable macro- and microelements in the soil. Plant development and metabolism depend on both macro- and microelements, which make up the nutrients required for plant life. Table 3 presents the analysis of plant-available macro- and microelements in soil samples from various localities. Soil from locality K1 exhibited the highest concentrations of Na, K, Zn, Pb, and Cd, suggesting potentially elevated nutrient availability. Locality K2 had the highest levels of Fe and Cr (with P1 also showing the highest Cr content), while soil from locality P2 recorded the highest concentrations of K, Mg, Ca, P, Mn, Ni, Cu, and Co.

The fertility of the soil and agricultural productivity are significantly impacted by these variations in nutrient content. For example, crops that need K, Mg, and Ca may do better in soils with high concentrations of these nutrients, such as those from locality P2. On the other hand, high Pb and Cd soils, like those from locality K1, may be dangerous for the safety of plants and crops and call for remediation or soil management techniques (PRIYA *et al., 2022*). The lack of plant-available molybdenum (Mo) in every locality that was sampled highlights the importance of integrated nutrient management that takes into consideration possible deficiencies in essential elements. All things considered, the information offers vital insights into enhancing soil fertility, optimizing nutrient management techniques, and guaranteeing sustainable agricultural productivity. RILEY *et al.* (1992) and NJDEP (https://dep.nj.gov/srp/guidance/scc/) have reported soil concentration ranges and regulatory guidelines for some heavy metals.

Total metal content in soil

The analysis of total metal content in the soils across the studied localities (Table 4) reveals significant variation in metal concentrations. Locality K1 had the highest levels of Zn, Pb, and Cd, indicating potential contamination concerns. Locality K2 contained the highest concentrations of Fe and Cr, a pattern also observed in locality P1 for Cr. In contrast, locality P2 had the highest levels of Mn, Ni, Cu, and Co, suggesting a distinct metal profile. Notably, no detectable levels of molybdenum (Mo) were found in the soils from any of the sampled sites.

These findings align with prior research indicating that soil metal concentrations are influenced by both natural factors and anthropogenic activities (ASSEMAVE and ANHWANGE, 2012; HONG *et al.*, 2014; NANGIA, 1991; ADRIANO, 2001). Local agricultural practices or industrial operations may be responsible for the high levels of Zn, Pb, and Cd in the soil of locality K1, which resulted in metal accumulation (ASSEMAVE and ANHWANGE, 2012). The presence of Fe and Cr in the soils of localities K2 and P1 can also be linked to industrial sources or natural geological formations (ASSEMAVE and ANHWANGE, 2012). The significantly higher Mo, Mn, Fe, and Co concentrations at locality P2 (p<0.001) likely reflect

variations in soil composition, land use, or underlying geological factors (HONG *et al.*, 2014). In parallel, the markedly elevated Zn, Pb, and Cd levels at locality K1 compared to the other locations (p<0.001) suggest localized contamination sources (ASSEMAVE and ANHWANGE, 2012).

Profile	K1	K2	P1	P2
Na	54.70 (2.10) b***	ND	1.10 (0.10)	47.90(3.12) $c^{ns}e^{***}f^{***}$
K	1152.80 (22.20) a ^{***} b ^{***}	140.20 (12.50)	858.50 (20.10) d ^{***} e ^{***}	1152.80 (32.00) c ^{ns} f ^{***}
Mg	705.90 (32.14) a***	145.80 (10.12)	1342.20 (29.64) b ^{***} d ^{***}	1722.20 (21.95) c ^{***} e ^{***} f ^{***}
Ca	4113.10 (30.10)	1523.10 (28.60) a***	4853.10 (32.20) b ^{***} d ^{***}	8493.10 (56.22) c*** e*** f***
Р	432.60 (21.40) a ^{****}	29.80 (2.21)	358.60 (19.22) b ^{***} d ^{***}	501.90 (21.70) c ^{***} e ^{***} f ^{***}
Мо	ND	ND	ND	ND
Mn	55.00 (3.32) a ^{***} b ^{***}	34.58 (2.01) d ^{****}	20.08 (1.50)	116.12 (5.64) c ^{***} e ^{***} f ^{***}
Fe	49.00 (3.11) b***	$\frac{120.40}{a^{***}} (7.74)$	20.50 (3.11)	$113.30 (9.12) \\ c^{***} f^{***}$
Ni	7.17 (1.14) a*** b***	1.18 (0.10)	4.06 (0.65) d ^{****}	21.70 (1.54) c*** e*** f***
Cu	$\frac{10.26 (1.65)}{a^{ns} b^{ns} c^{ns}}$	10.94 (0.92) d ^{ns} e ^{ns}	12.40 (1.52) f ^{ns}	12.88 (1.32)
Zn	154.30 (10.11) a ^{***} b ^{***} c ^{***}	25.20 (1.44) d ^{***} e ^{***}	3.59 (0.50) f*	2.73 (0.38)
Cr	ND	0.068 (0.001) d ^{ns} e ^{***}	0.068 (0.001) f ^{***}	0.032 (0.007)
Pb	58.90 (4.06) a ^{***} b ^{***} c ^{***}	7.92 (1.11) d*** e***	2.00 (0.45)	2.88 (0.11) f*
Cd	3.44 (0.94) a ^{***} b ^{***} c ^{***}	$\begin{array}{c} 0.13 (0.002) \\ d^{***} e^{***} \end{array}$	0.06 (0.001)	0.10 (0.004) f ^{***}
Со	0.18 (0.006) a ^{***} b ^{***}	0.17 (0.006) d ^{***}	0.08 (0.001)	0.35 (0.005) c ^{***} e ^{***} f ^{***}

Table 3. Content of macro- and microelements available to plants in the tested soil (mg/kg)

Analysis of variances (ANOVA) and Scheffé's post-hoc test; results are presented as mean and standard deviation -M (SD), n=5; (a) K1 – K2, (b) K1 – P1; (c) K1 – P2; (d) K2 – P1; (e) K2 – P2; (f) P1 – P2; * p<0.05, *** p<0.001, ns – no statistically significant differences, ND – not detected.

The contents of Cd, Pb and Zn in the soil of locality K1 were higher than the maximum allowed concentrations, limit and remediation values prescribed by the regulations of the Republic of Serbia (ANONYMOUS, 1994) as well as the limit values of these metals prescribed by the regulations of the European Union (EU DIRECTIVE 86/278/EEC). Also, the contents of Cd, Cu, Pb and Zn in the soil of locality K2 were higher than the limit values prescribed by the regulations of the Republic of Serbia (ANONYMOUS, 1994). The concentration of Cd in the soil of locality P2, as well as the concentration of Cu in all investigated localities, is higher than the limit values of these metals according to the mentioned regulations. The soils of all investigated localities (except K2) contain more Ni

than the limit values. The soil of locality P2 contains Ni more than the prescribed maximum allowed concentrations in the soil, as well as more than the remediation value and limit values of this metal prescribed by the European Union regulations.

Profile	K1	K2	P1	P2
Мо	ND	ND	ND	ND
Mn	550.0 (25.00) a ^{***} b ^{***}	345.8 (11.10) d ^{****}	200.8 (19.22)	1161.2 (17.12) c ^{***} e ^{***} f ^{***}
Fe	490.0 (28.32) b ^{***}	1204.0 (11.19) a ^{***} d ^{***}	205.0 (19.21)	1133.0 (22.92) c ^{***} e ^{***} f ^{***}
Ni	71.7 (15.20) a ^{***} b ^{***}	11.8 (1.29)	40.6 (3.33) d***	217.0 (14.12) c ^{***} e ^{***} f ^{***}
Cu	102.6 (4.98) a ^{ns}	109.4 (11.18)	124.0 (10.11) b [*] d [*] f ^{ns}	128.8 (11.14) c [*] e [*]
Zn	1543.0 (21.20) a ^{***} b ^{***} c ^{***}	252.0 (18.11) d ^{***} e ^{***}	35.9 (5.51) f ^{ns}	27.3 (4.11)
Cr	ND	0.7 (0.02) d ^{ns} e ^{***}	0.7 (0.01) f ^{***}	0.3 (0.01)
Pb	647.9 (15.12) a ^{***} b ^{***} c ^{***}	87.1 (4.12) d ^{***} e ^{***}	22.0 (2.15)	31.7 (2.11) f***
Cd	34.4 (3.51) a ^{***} b ^{***} c ^{***}	$1.3 (0.2) d^{***} e^{ns}$	0.6 (0.01)	1.0 (0.1) f***
Со	1.8 (0.20) a ^{ns} b ^{***}	1.7 (0.13) d ^{***}	0.8 (0.01)	3.5 (0.50) c ^{***} e ^{***}

Table 4. Total content of trace elements in the tested soil at K1, K2, P1, and P2 localities (mg/kg)

Analysis of variances (ANOVA) and Scheffé's post-hoc test; results are presented as mean and standard deviation -M (SD), n=5; (a) K1 -K2, (b) K1 -P1; (c) K1 -P2; (d) K2 -P1; (e) K2 -P2; (f) P1 -P2; * p<0.05, *** p<0.001, ns - no statistically significant differences, ND - not detected.

The analysis of total metal content in soil offers critical insights into soil quality and the environmental risks associated with metal contamination. These findings underscore the need for targeted management practices and ongoing monitoring to mitigate the adverse effects of metal pollution on both soil and environmental health. By comparing the obtained results with established literature values of metal concentrations in soils from various regions (ANONYMOUS, 2019; HONG *et al.*, 2014; NANGIA, 1991; ASSEMAVE and ANHWANGE, 2012; ADRIANO, 2001), our study emphasizes the significance of localized remediation efforts by highlighting areas where contamination may present unique risks.

The content of macroelements in plants

The presence and content of macroelements in plants is essential for their growth and development, as these elements play vital roles in numerous metabolic processes (RYAN *et al.*, 2001). The analysis of macroelements in the examined plant materials (Table 5) reveals significant differences in the concentrations of Na, K, Mg, and Ca between *C. annuum* and *J. communis* at the K1 and K2 localities. In addition, our results showed that the content of these macroelements also depended on the plant species, as well as the specific plant parts analyzed (stem or leaf).

In *C. annuum*, the stem exhibited elevated concentrations of Na, K, Mg, and Ca compared to the leaf, suggesting a distinct pattern of macroelement allocation within the plant. Conversely, *J. communis* demonstrated notable differences in macroelement distribution between its stem and leaf, with the leaf containing higher concentrations of Mg, Na, and Ca. The absence of Mg in the stem of *J. communis* indicates a specific and possibly adaptive distribution strategy for this species, emphasizing its unique physiological traits.

Profile	K	1	K2		
	<i>C. annuum -</i> stem	<i>C. annuum</i> - leaf	<i>J. communis</i> - stem	<i>J. communis -</i> leaf	
Na	11900 (102)	ND	ND	6800 (325)	
K	27800 (129)***	9400 (232)	ND	ND	
Mg	25700 (113)***	6000 (423)	ND	25900 (945)	
Ca	119800 (923)***	36400 (621)	9600 (756)	147600 (1034)***	
Profile	Р	1	P2		
	P. domestica -	P. domestica -	R. fruticosus -	R. fruticosus -	
	stem	leaf	stem	leaf	
Na	31600 (540)	6600 (770)***	3000 (120)***	500 (50)	
K	8500 (230)	10300 (450)***	ND	3200 (120)	
Mg	41600 (410)***	22400 (560)	15500 (720)***	1200 (85)	
Ca	129600 (990)***	90600 (840)	78900 (550)***	40300 (960)	

Table 5. Content of macroelements in the investigated plant material at the K1, K2, P1 and P2 localities (mg/kg)

Analysis of variances (ANOVA) and Scheffé's post-hoc test; results are presented as mean and standard deviation - M (SD), n=5; *** p<0.001, ND - not detected.

The results reveal notable differences in macroelement content between the two plant species. Specifically, the leaf of *J. communis* exhibited significantly higher levels of Ca compared to *C. annuum*, while the stem of *C. annuum* had elevated concentrations of Na and K. Additionally, the macroelement content in the plant materials of *P. domestica* and *R. fruticosus* from the P1 and P2 localities displayed significant variations. The stem of *P. domestica* contained the highest concentrations of Na, Mg, and Ca, whereas its leaf had the highest concentration of K. In contrast, *R. fruticosus* showed lower overall macroelement concentrations compared to *P. domestica*, with its stem exhibiting higher levels of Na, Mg, and Ca than the leaf.

The average content of Ca, Mg, K and Na in the dry matter of plants is: 0.5; 0.2; 1.0 and 0.01 to 2%, respectively (KASTORI, 1998; UBAVIĆ and BOGDANOVIĆ, 2001). The results of our research indicate the presence of a high content of macroelements in the organs of the studied plants. This indicates an increased need of plants for these elements and implies their high mobility and good absorption through the root system, as well as their transport to the above-ground parts of plants. The following are absorbed from the soil in ionic form: Ca²⁺, Mg²⁺, K⁺ and Na⁺, whereby mechanisms of competition and antagonism between different ions come into play, which affects their uptake and content in the plant (BOGDANOVIĆ *et al.*, 1993; MIHALJEV *et al.*, 2015). It has been shown that the contents of Ca, Mg, K and Na in our soils vary, as do their contents in different plant species (MIHALJEV *et al.*, 2015) as indicated by the results of this study.

Content of microelements in plants

Micronutrients, although required in smaller quantities, play crucial roles in various physiological processes in plants (RYAN *et al.*, 2001). The analysis of trace elements in the plant material of *C. annuum* and *J. communis* at the K1 and K2 localities revealed significant variations in their content (Table 6). For instance, the stem of *C. annuum* exhibited elevated concentrations of Fe, Ni, Cu, Zn, and Cr compared to the leaf, indicating distinct accumulation patterns within the plant. In contrast, *J. communis* demonstrated the highest levels of Mo and Co in its stem, alongside the highest Mn content in its leaf, suggesting specific mechanisms for nutrient uptake and allocation that may be influenced by the plant's physiological needs and environmental factors.

Profile	K	1	K2		
	<i>C. annuum</i> - stem	<i>C. annuum -</i> leaf	<i>J. communis</i> - stem	<i>J. communis</i> - leaf	
Мо	54 (5)	ND	86 (6)***	5 (0.5)	
Mn	68 (7)***	27 (3)	185 (15)	458 (32)***	
Fe	1373 (55)***	429 (32)	484 (22)	ND	
Ni	549 (45)	ND	ND	ND	
Си	162 (9)	ND	ND	102 (15)	
Zn	422 (21)	ND	ND	ND	
Cr	810 (50)	ND	ND	ND	
Pb	ND	ND	748 (49)	ND	
Cd	ND	ND	ND	ND	
Со	8.49 (2)	ND	43.7 (4.2)	ND	

Table 6. Content of trace elements in the examined plant material at the K1 and K2 localities (mg/kg)

Analysis of variances (ANOVA) and Scheffé's post-hoc test; results are presented as mean and standard deviation – M (SD), n=5; *** p<0.001, ND – not detected.

The differences in micronutrient content between the stem and leaf of *C. annuum* were statistically significant, with higher concentrations of Mn and Fe in the stem compared to the leaf (p<0.001). In contrast, at locality K2, the stem of *J. communis* exhibited significantly higher Mo content compared to the leaf (p<0.001), while Mn concentrations were greater in the leaf than in the stem (p<0.001). Additionally, several trace elements were not detected in the leaves or stems of either species at specific localities, reflecting variability in micronutrient accumulation and distribution.

Similarly, different accumulation patterns were found when microelements in *P. domestica* and *R. fruticosus* were analyzed at the P1 and P2 localities (Table 7). While *R. fruticosus* showed the highest Mo content in the leaf, *P. domestica* stem showed higher concentrations of Mn, Fe, Cu, Zn, Cr, Pb, and Co than the leaf. Interestingly, no samples of the plants under investigation contained either Ni or Cd.

The differences in microelement content between the stem and leaf of *P. domestica* were statistically significant, with higher concentrations of Mo, Mn, Fe, Cu, Cr, and Co in the stem compared to the leaf (p<0.001, at locality P1). Similarly, at locality P2, the stem of *R. fruticosus* had significantly higher Mn, Fe, and Cu content compared to the leaf (p<0.001), while Co content was higher in the leaf than in the stem (p<0.001). Additionally, several

microelements were not detected in either the stem or leaf of *R. fruticosus* at specific localities.

The contents of Cu, Zn, Ni, Fe and Pb in the examined organs of the investigated plants (except for the stem of the *R. fruticosus* species) where they were detected were higher than the range of normal concentrations, tolerant and toxic concentrations of these metals in plants (KABATA-PENDIAS, 2011).

Table 7. Content of microelements in the examined plant material at the P1 and P2 localities (mg/kg)

Profile	I	21	P2			
	<i>P. domestica</i> - stem	<i>P. domestica</i> - leaf	R. fruticosus - stem	R. fruticosus - leaf		
Мо	123 (17)***	66 (12)	178 (7.4)	184 (8.5)		
Mn	1481 (27.21)***	55 (5)	122 (15)***	31 (4.5)		
Fe	50020 (125)***	1459 (45.5)	2520 (25.5)***	627 (17.2)		
Ni	ND	ND	ND	ND		
Си	2358 (165)***	153 (12.12)	897 (27)***	168 (12.4)		
Zn	1109 (95)	ND	ND	ND		
Cr	242 (24)***	163 (12.25)	70 (5)	ND		
Pb	501 (50)	ND	251 (20)	ND		
Cd	ND	ND	ND	ND		
Со	56.69 (9.9)***	2.39 (0.84)	7.30 (0.9)	18.2 (2.2)***		

Analysis of variances (ANOVA) and Scheffé's post-hoc test; results are presented as mean and standard deviation - M (SD), n=5; *** p<0.001, ND - not detected.

These results point to species-specific variations in the distribution and accumulation of macro- and microelements in plants, which may be impacted by variables like soil nutrient availability, environmental factors, and genetic structure. Optimizing plant growth and productivity in ecological and agricultural contexts requires an understanding of these patterns.

Bioaccumulation factor

The bioaccumulation factor (BCF) is a crucial metric for evaluating a plant's efficiency in accumulating metals. It is defined as the ratio of metal concentration in plant material to its concentration in the soil (MALIK *et al.*, 2010) or the total metal concentration in the plant's stem and leaves relative to that in the soil (SARASWAT and RAI, 2009). BCF values indicate accumulation efficiency: BCF > 1 signifies strong accumulation, BCF = 1-0.1 indicates medium accumulation, BCF = 0.1-0.01 reflects weak accumulation, and BCF = 0.01-0.001 suggests no accumulation (KABATA-PENDIAS and DUDKA, 1991). Plants with BCF > 1 in their shoots are considered potential candidates for phytoextraction (ZACCHINI *et al.*, 2009).

The bioaccumulation factors (BCF) for microelements in the analyzed plant species are presented in Table 8. The data reveal distinct accumulation patterns among the different species and plant organs. Notably, *P. domestica* exhibited the highest BCF values across multiple microelements, particularly for Zn (30.89), Cr (345.71), and Co (70.86) in its stem, indicating its strong potential for phytoextraction. The elevated BCF values for these elements suggest that *P. domestica* effectively absorbs and concentrates these metals from the soil, making it a candidate for remediation strategies in contaminated sites.

The comparison of bioaccumulation factors (BCF) between leaves and stems across the studied species indicates notable differences in metal accumulation efficiency. For *C. annuum*, the stem exhibited a BCF of 7.66 for Ni and 2.80 for Fe, while the leaf had a lower BCF of 0.05 for Mn and 0.88 for Fe, reflecting a higher accumulation capacity in the stem. In *J. communis*, the stem's BCF values were 0.40 for Fe and 0.53 for Mn, while the leaf had a higher BCF of 1.32 for Mn, demonstrating that this species accumulates certain elements more effectively in its leaves. *P. domestica* showed the highest BCF values overall, with a remarkable 30.89 for Zn and 19.02 for Cu in the stem, while the leaf had a BCF of 7.12 for Fe. Conversely, *R. fruticosus* had much lower BCF values, with 0.11 for Mn and 6.96 for Cu in the stem, indicating less overall accumulation capacity.

Species/organ			MIC	ROELE	MENTS	/ BCF		
	Mn	Fe	Ni	Cu	Zn	Cr	Pb	Со
R. fruticosus - stem	0.11	2.22	/	6.96	/	233.33	7.92	2.09
R. fruticosus - leaf	0.03	0.55	/	1.30	/	/	/	5.20
<i>J. communis</i> - stem	0.53	0.40	/	/	/	/	8.59	25.71
<i>J. communis</i> - leaf	1.32	/	/	0.93	/	/	/	/
<i>C. annuum</i> - stem	0.12	2.80	7.66	1.58	0.27	/	/	4.72
<i>C. annuum</i> - leaf	0.05	0.88	/	/	/	/	/	/
<i>P. domestica</i> - stem	7.38	2.44	/	19.02	30.89	345.71	22.77	70.86
<i>P. domestica</i> - leaf	0.27	7.12	/	1.23	/	232.86	/	2.99

Table 8. Bioaccumulation factors (BCF) for microelements in investigated plants

The bioaccumulation factor (BCF) values indicate that *P. domestica* demonstrates the highest bioaccumulation potential, positioning it as a strong candidate for the phytoextraction of metals such as Mn, Na, Fe, Cu, Zn, Cr, Pb, and Co, with BCF values exceeding 1, which suggests significant accumulation capability (GHOSH and SINGH, 2005). Additionally, *J. communis* shows potential for phytoextraction of Mg, Ca, Mn, Pb, and Co, while *R. fruticosus* can accumulate Fe, Cu, Cr, Pb, and Co, and *C. annuum* is effective for K, Fe, Ni, Cu, and Co. Furthermore, the analysis of soil properties across various localities revealed significant variations in chemical composition and texture that impact the phytoaccumulation capabilities of these species. For instance, localities K1, P1, and P2 displayed skeletonless soils with sandy loams and clay loams, whereas locality K2 consisted of strongly skeletal soils. The distinct variations in macro- and microelement availability, particularly the high concentrations of Na, K, Zn, Pb, and Cd at locality K1 and elevated Fe and Cr levels at K2, create a diverse nutrient landscape that influences how different species adapt and accumulate elements, ultimately affecting their effectiveness in phytoremediation efforts.

The findings of the research highlight the complex function of the investigated plant species, which have significant potential for phytoremediation even though they are fit for human consumption. Because of their capacity to bioaccumulate heavy metals, including those that are harmful to humans, agricultural practices must be closely monitored and

managed. There is a higher chance of human exposure through the food chain because these species could absorb hazardous pollutants from contaminated soils. Establishing thorough guidelines for their cultivation is therefore crucial, especially in regions where soil contamination is known to exist. This study emphasizes the necessity of establishing a balance between the ecological advantages of using these plants to decontaminate soil and the necessity of safeguarding the general public's health from potential hazards of heavy metal accumulation in edible tissues.

CONCLUSION

The study highlights the critical issue of metal contamination in soils and plants within Aleksandrovac municipality and Kopaonik National Park. Variations in metal concentrations across locations highlight the importance of closely monitoring soil health and plant bioaccumulation potential. The ability of *Prunus domestica* to accumulate multiple heavy metals has made it a promising candidate for phytoremediation efforts. However, the significant presence of cadmium (Cd) in the soil at Kopaonik 1 site (K1) raises considerable concerns regarding agricultural practices and food safety. Given the toxic nature of cadmium, it is imperative to exercise caution when considering the agricultural use of plants grown in this area, as they may pose health risks to consumers. Developing integrated management practices that combine phytoremediation with sustainable agricultural techniques is essential for mitigating heavy metal pollution and enhancing food security.

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