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Field experiment on the uptake of lead, strontium, cobalt, and nickel in the wood and bark of spruce (*Picea abies* L.) and Douglas-fir (*Pseudotsuga menziesii* Mirb.)

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Abstract: Human activities have significantly altered the availability and circulation of pollutants, impacting their concentrations in the environment. This pollution notably affects trees. In this study, we conducted two separate experiments (I and II) to investigate the uptake of lead, strontium, cobalt, and nickel in spruce (*Picea abies* L.) and Douglas-fir (*Pseudotsuga menziesii* Mirb.) seedlings. These seedlings were exposed to elevated levels of these metals by adding them to the soil. Our field experiments provide insights into metal accumulation in natural environments. We measured concentrations of these elements, along with manganese and zinc, in the soil, wood, and bark using Inductively Coupled Plasma-Optical Emission Spectrometry (ICP-OES). The results showed increased levels of the added metals in the wood and bark of both tree species. Notably, there was a significant increase in lead and nickel concentrations in Douglas-fir wood. The lead concentration in Douglas-fir wood was 7 and 4 times higher in experiments I and II, respectively, compared to the control group of seedlings, while the nickel concentration was 18 and 10 times higher. These findings suggest that Douglas-fir wood has potential for phytostabilization of lead and nickel based on trace element concentrations and transfer factors.

Keywords: accumulation; trace elements; tree seedlings; phytostabilization; transfer factors.

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INTRODUCTION

Modern development and urbanization have led to the production of various pollutants, including trace elements, organic and inorganic compounds, and pesticides, which can persist in the soil and harm the environment and human health. Trace elements are particularly concerning due to their toxicity and resistance to natural degradation, accumulating in plants, animals, and the environment over time.[1] Vascular plants serve as valuable indicators of environmental pollution. Different tree parts, such as leaves/needles, seeds, bark, and tree-rings, are used to monitor trace element contamination.[2–6] Trees, with their high biomass production, long vegetation periods, strong root systems, and ability to tolerate and remediate pollutants, are effective in extracting trace elements.[7] This process, known as phytoextraction, helps prevent these elements from leaching into deeper soil layers and groundwater.

The bioavailability of metals to plants is influenced by their micronutrient demand and their ability to exude and eliminate toxic elements. Metal accumulation in plants is affected by the distribution of elements in different tissues, the presence of elements with similar physiochemical properties in the soil, the availability of elements in the soil, site conditions, and tree species.[1] Metals with similar properties, such as ion size and charge, compete for binding sites in plants, affecting their uptake, translocation, and accumulation.[8] Tree species also influence soil pH, impacting trace element availability. The release of organic acids and hydrogen ions from tree roots increases metal ion solubility and uptake.[8] Plants absorb trace elements through their roots and above-ground parts, like leaves and bark, with root uptake being the primary pathway for metals to enter trees.

Metal uptake generally increases as the concentration of metal ions in the external solution increases.[9] However, excessive concentrations of trace element in the soil can trigger protective mechanisms in plants that inhibit the absorption.[10] Therefore, the migration and accumulation of each metal within a tree involve individual and sophisticated biochemical processes and various transportation systems.[11] While the biological function of elements like Pb and Sr in higher plants is unknown and likely toxic, Ni is a part of the enzyme urease.[12] and Co is essential for several enzymes and coenzymes in higher plant systems.[13] The accumulation of trace elements in wood and bark has not been extensively studied, especially through field experiments. Donnelly et al. (1990) investigated lead mobility in red spruce seedlings, focusing on whether Pb ions remained in the xylem during uptake.[14] Numerous studies have focused on the uptake and translocation of radionuclides in plants, with a majority of them have focused on the bark and foliar surfaces.[15,16] Metal mobility in plants depends on their metabolic function, background metal levels, and the dosage applied to foliar surfaces. Some studies have examined the toxic effects of trace element salts on tree growth and their

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potential for phytoextraction and phytostabilization.[10,17–20] However, there is a significant research gap in studying trace element accumulation in wood and bark through field experiments. Furthermore, diverse trace elements accumulate at varying degrees within distinct plant species and parts, emphasizing the significance of understanding the levels of trace elements in plants. The investigation of Douglas-fir plant species in this regard remains insufficient. Studies that are based on collecting data about the accumulation of specific trace element in plants and their distribution among plant parts must be continued and diversified.[21]

The study aimed to investigate how elevated levels of Pb, Sr, Co, and Ni in soil affect their accumulation in spruce and Douglas-fir trees under natural conditions. This field experiment focused on trace element accumulation in wood and bark, with soil metal concentrations as the only variable. The research sought to understand how these metals are absorbed by the trees and to compare the sensitivity of spruce and Douglas-fir to Pb, Sr, Co, and Ni, evaluating their potential for trace element accumulation.

EXPERIMENTAL

Study site and experimental design

The field experiment was carried out in Kaluđerica, Belgrade, Serbia, on Livada 1 Street as depicted in Figure 1 (a) and (b). Five-year-old spruce (*Picea abies* L.) and Douglas-fir (*Pseudotsuga menziesii* Mirb.) seedlings were obtained from the Institute of Forestry, Belgrade. In May 2017, 24 seedlings were planted across 48 m², divided into six groups, with four seedlings per group (Figure 1c). To prevent metal transfer, seedlings were spaced 1 meter apart. After rooting until January 2018, they were watered five times between January and May 2018 with Pb, Sr, Co, and Ni in two experiments, with the I experiment having double the concentrations of the II experiment. The third group of seedlings served as the control group. In May 2019, the seedlings were harvested.

Amount of added metals

In experiment I, spruce and Douglas-fir seedlings were watered monthly with a solution containing 2 g L^{-1} of Pb and Sr, as well as 0.5 g L^{-1} of Co and Ni. In experiment II, the solution contained half of these concentrations. Seedlings were watered with tap water during the experiment. Metal solutions were prepared monthly using nitrate salts (all purchased from Merck: lead(II) nitrate $Pb(NO_3)_2$; strontium(II) nitrate $Sr(NO_3)_2$; cobalt(II) nitrate hexahydrate $Co(NO_3)_2{\cdot}6H_2O$ and nickel(II) nitrate hexahydrate $Ni(NO_3)_2{\cdot}6H_2O$) dissolved in tap water. Seedlings were watered for five months with solutions achieving concentrations of 2 g L^{-1} Pb and Sr, and 0.5 g L^{-1} Co and Ni for the first experiment, and 1 g L^{-1} Pb and Sr, and 0.25 g L^{-1} Co and Ni for the second experiment. Total metal added was 10 mg g^{-1} of Pb and Sr and 2.5 mg g^{-1} of Co and Ni for experiment I, and half of that for experiment II, and those concentrations are total amounts to which plants were exposed in this experiment. All trees survived with normal growth and no signs of metal toxicity.

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Sample preparation and experiment

To simplify the results, spruce and Douglas-fir trees from the first experiment are labeled 'SI' and 'DI,' and from the second experiment, 'SII' and 'DII.' Control groups are labeled 'SC' and 'DC.'

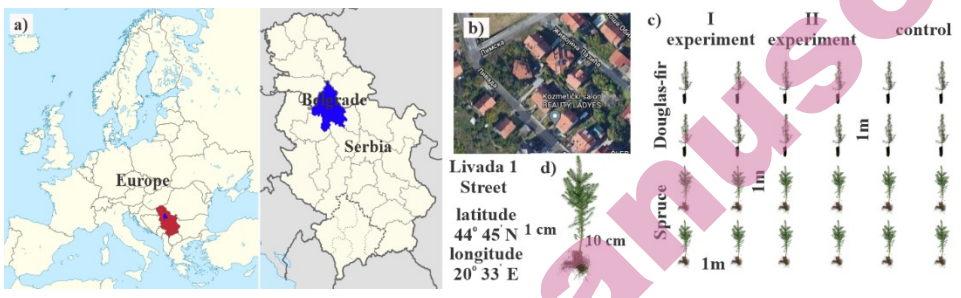

Figure 1. a) Sampling location of the performed field experiment, b) Google map of the location with the red arrow showing the planted area, c) display of the planted seedlings with two parallel field experiments and control and d) display of the wood sampling.

Four seedlings of each species were cut, and 1 cm stem disks were sampled from 10 cm above the base (Figure 1 (d)), along with bark and soil samples taken from 0-20 cm depth. Most of the fine roots of trees are found in the surface soil layer at this depth.[22] Soil before plantation was also collected to obtain the amount of the metal content in the soil before the field experiment. Tree wood, bark, and soil were digested using an Advanced Microwave Digestion System (ETHOS 1, Milestone, Italy). Sample digestion in the Advanced Microwave Digestion System (ETHOS 1) followed standard manufacturer-recommended programs, with the official software optimizing chemical volumes, temperature, and pressure based on sample type and mass. About 0.5 g of powdered oven-dried samples, of spruce and Douglas-fir, were precisely weighed and mixed in the clean vessel with a mixture of 3 mL 30 % H_2O_2 (Suprapur®, Germany) and 5 mL 65 % HNO_3 (Suprapur®, Germany) and then heated with microwave energy, (with parameters T= 200 °C, KW= 1800 W, t= 15 min and p= 90 bar). The soil was first dried at room temperature and then in order to ensure the homogeneity of soil samples, each of the samples was divided into six equal parts from which the same amount of soil was taken. This amount of soil was sieved through a plastic sieve, and then ground in a mortar to a powdery particle size. The sample was then dried in an oven to constant weight at a temperature of 60 °C. Soil samples (0.5 g) from each part of the experiment were thoroughly mixed before analysis to ensure a homogenous concentration, accurately measured, and placed in clean microwave preparation vessels with 8 mL 65 % HNO_3 (Suprapur®, Germany) and 2 mL 36 % HCl (Suprapur®, Germany). The parameters of the soil preparation program were the same as for the preparation of plant tissues. The temperature was controlled with a predetermined power program. After cooling and without filtration, the solution was diluted to a fixed volume of 25 mL. Quality control was assured by the use of procedural blanks. Precision and accuracy were confirmed by repeated analysis of NIST pine needles (1575a) as standard reference material.

The content of elements (Pb, Sr, Co, Ni, Mn and Zn) in each sample, prepared as diluted aqueous solutions, was quantified by inductively coupled plasma optical emission spectrometry-ICP-OES using a Thermo Scientific iCAP 7400 duo analyzer (Thermo Fisher Scientific Inc., USA). The calibration standard solutions were prepared from a Multi-element ICP IV standard stock solution (Merck). For each element determined, calibration curves were

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constructed and used to determine the analyte concentrations. The data acquisition and processing were performed by the Thermo Scientific Qtegra platform software. All measurements were carried out in triplicates. The pH of the soil was determined using a glass electrode (1741, La Motte Tracer-PockeTester) in a 1:5 (volume fraction) suspension of soil in deionized water. 2 g was precisely weighted, and 10 mL of deionized water was added. The suspension was stirred on a stirrer for 30 min and left for another 30 min to stand before pH measuring. The chemical analysis results were evaluated using One-Way ANOVA to compare the three groups (I experiment, II experiment, and control) for both tree species, followed by Tukey's test to identify specific group differences. Differences were considered statistically significant at the 0.05 level.

Transfer factor

The capacity of trees to extract trace elements from soil and their translocation to aboveground tissues can be evaluated by calculating the transfer factor, TF. Different calculations for transfer factors can be seen in the literature and most of them divide the average trace element concentration in the plant part by the concentration in soil.[23,24] Most of them showed only elevated concentrations of specific metals compared to soil or biogeochemical comparison of metals in different media (plant and soil) that occur under the same circumstances.[25] It does not show how the metal transfer from soil to plant changed on the treated site compared to the control site.[25] Including the control site (where natural processes affect metal transfer from soil to plant) in the calculations, we can get information about changes in transfer processes.[25] To investigate the transfer of externally added metals-to-soil (TF_{soil}); soil-to-wood transfer (TF_{wood}) and soil-to-bark transfer (TF_{bark}), for this experiment, TF was calculated (in %) by the following equations:

$$TF_{soil} = (I\text{-}C)_{soil}/(EA)\ x100\ (\%) \quad (1)$$

$$TF_{wood} = (I\text{-}C)_{wood}/(I\text{-}C)_{soil}\ x100\ (\%) \quad (2)$$

$$TF_{bark} = (I\text{-}C)_{bark}/(I\text{-}C)_{soil}\ x100\ (\%) \quad (3)$$

where I and C represent the mean Pb, Sr, Co, and Ni concentrations obtained in the first experiment and control for two examined coniferous species (S and D), while EA is externally added concentration of metals examined. The same equations were used for the II experiment.

RESULTS AND DISCUSSION

Concentration of Pb, Sr, Co, Ni, Mn, and Zn in soil, wood, and bark of spruce and Douglas-fir seedlings

Mean Pb, Sr, Co, and Ni concentrations in soil, wood, and bark of spruce and Douglas-fir seedlings were presented in Table I which were externally added in this field experiment. Also, mean Mn and Zn concentrations in soil, wood, and bark and the measured pH of the spruce and Douglas-fir corresponding soils can be seen in Table I. Soil concentrations measured before plantation and performed field experiment were 21±1 μg g^{-1} for Pb, 78±4 μg g^{-1} for Sr, 22±1 μg g^{-1} for Co, 87±5 μg g^{-1} for Ni, 822±42 μg g^{-1} for Mn, and 118±5 μg g^{-1} for Zn. Maximally allowed concentrations in the soil for Pb, Co, Ni, and Zn in the Republic of Serbia are 85 μg g^{-1}, 9 μg g^{-1}, 35 μg g^{-1}, and 140 μg g^{-1}.[26] Soil concentrations of Co and Ni, both before planting and in the control samples, exceed the maximum levels permitted by the Regulation, likely reflecting the

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geochemical conditions of the experimental area. Additionally, air pollution cannot be ruled out, given the moderate traffic, residential heating, and the proximity to the largest waste dump in Vinča, which is the only landfill in the Belgrade city area.

Table I Mean Pb, Sr, Co, Ni, Mn and Zn (n=4) concentrations (μg g^{-1}) in soil, wood, and bark seedlings of spruce first experiment-SI; second experiment-SII; control-SC; Douglas-fir first experiment-DI; second experiment-DII and control-DC, as well as corresponding soil pH.

	Pb			Sr		
	soil	wood	bark	soil	wood	bark
SI	827±56	0.41±0.05	4.73±0.55	680±51	101±7	256±13
SII	505±30	<0.15	1.87±0.09	325±19	54.39±3.45	246±13
SC	21±1	<0.15	1.67±0.10	75±4	31.53±1.89	81±5
DI	634±30	1.06±0,10	2.44±0.13	371±21	46.37±3.68	197±13
DII	369±19	0.61±0,08	1.28±0.06	301±15	44.32±2.72	154±9
DC	23±1	<0.15	0.68±0.05	77±4	15.85±1.07	26±2
	Co			Ni		
	soil	wood	bark	soil	wood	bark
SI	97±6	<0.04	0.44±0.04	278±16	0.34±0.03	2.71±0.16
SII	73±4	<0.04	0.32±0.02	199±9	0.29±0.04	1.78±0.09
SC	20±1	<0.04	0.12±0.02	88±4	0.24±0.03	1.09±0.09
DI	84±6	<0.04	0.27±0.04	226±11	0.73±0.06	1.13±0.06
DII	63±3	<0.04	0.23±0.04	182±9	0.40±0.03	1.03±0.06
DC	21±2	<0.04	<0.04	89±4	<0.04	0.36±0.04
	Mn			Zn		
	soil	wood	bark	soil	wood	bark
SI	950±43	8.23±0.46	18.7±1.4	128±6	18.0±0.9	67.9±3.9
SII	923±42	8.41±0.49	23.4±1.9	125±5	21.5±1.0	73.4±4.4
SC	823±42	9.22±0.57	28.9±1.9	118±7	28.4±1.6	100±5
DI	951±43	3.38±0.23	13.7±0.9	131±7	5.36±0.37	22.0±1.9
DII	954±43	2.63±0.20	12.0±0.6	122±5	3.52±0.19	25.0±1.4
DC	852±46	5.00±0.27	8.71±0.44	113±5	7.66±0.38	30.4±1.6
	SI	SII	SC	DI	DII	DC
Soil pH	6.7±0.05	6.6±0.05	6.8±0.05	6.9±0.05	6.7±0.05	7.0±0.05

Soil concentrations before plantation were as control concentrations (SC and DC) measured after metal addition in experiments I and II (Table I). Thus, levels of the metals of interest in the soil before additional metal watering were as in control, excluding the possibility of metal transfer from one experiment to another. Elevated concentrations in the soil (Table I) externally added did not affect plant growth during this 2-year experiment. Higher mean Pb, Sr, Co, and Ni concentrations for bark can be seen for both examined tree species, than in their wood compartments (Table I).

Mean differences of Pb, Sr, Co, and Ni for soil and bark of both examined species were significant, at the 0.05 level, between parallel experiments and

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control (One-Way ANOVA), except for the Pb concentration in SII for bark. In the case of wood, means differences were significant for Pb, Sr, and Ni, except between Ni SII concentration and control. For some soil, wood, and bark mean differences between the I and II experiments weren't significant (Tukey test). For the pH, mean differences were significant, at the 0.05 level, between the II experiment and control for both tree species. Means differences of Mn and Zn for soil, wood and bark of both examined species were significant, at the 0.05 level, between parallel experiments and control (One-Way ANOVA, Tukey test), except for the Douglas-fir DII and DC and spruce SI and SII soil Zn concentration. In some cases, for soil, wood and bark the mean differences weren't significant between the I and II experiment.

Pb and Sr concentration

From Figure 2 (a), it can be seen, for spruce, that the Pb content in the soil increases, and the increase is about 40 (SI) and 24 (SII) times, compared to the control soil sample. This implies an elevation of Pb concentration in the SI wood and barks for SI and SII, compared to the control (Table I). The increase was 2.8 times for SI wood and 2.8 and 1.1 times for SI and SII in the bark, respectively (Figure 2 (a)). Comparing the concentrations of SI and SII (Figure 2 (b)) for Pb in soil, wood, and bark we obtain an increased concentration in the SI experiment of about 1.6, 2.8, and 2.5 times, respectively. Increased Sr content in the soil from 75 μg g^{-1} to SI-680 μg g^{-1} and SII-325 μg g^{-1} (9 and 4 times higher compared to control) directly influences the increase of Sr concentration in the spruce wood and bark (Table I; Figure 2 (a)). The concentration of Sr in the soil was about twice as high if we observe the ratio between the I and II experiment (Figure 2 (b)). Also, the concentration of Sr in wood is almost twice as high, ie. about 1.9 times (86 %). In the bark, this increase between the I and II experiment is about 4 % (Figure 2 (b)).

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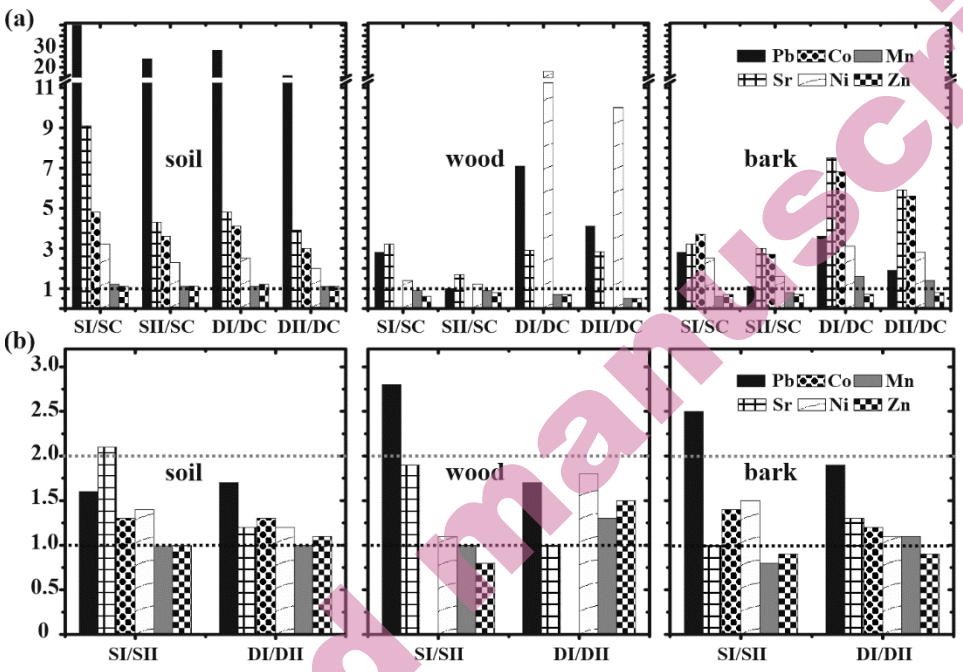

Fig. 2. Concentration ratios in soil, wood and bark a) between the I and II experiment compared to the control and b) between the first-I and second-II experiment, for spruce-S and Douglas-fir-D. (Black line indicates a ratio of 1, representing no change in concentration, gray line represents a ratio of 2, suggesting that accumulation aligns with the two times higher externally added concentration for the I experiment; Concentration ratios for cases where the detection limit was measured in the control-C were calculated using the value of the detection limit.).

In the case of Douglas-fir, it was noticed that with the increase of Pb concentration in the soil of 28 (DI) and 16 (DII) times, compared to control, the content in wood (7 and 4 times) and bark (4 and 2 times) also increases (Figure 2(a), Table I). An increase in the soil Pb content of 72 % between parallel experiments contributes to the increase of Pb in the wood of about 74 %, and the bark of about 91 %. The higher content of Sr in the soil (Table I) parallel experiments (5 and 4 times compared to control), as well as in the case of spruce, implies the higher content in Douglas-fir wood (3 times for both experiments) and bark (8 and 6 times). An increase of the Sr concentration in the soil for the I experiment of about 23 % (1.2 times) compared to the II experiment indicates that Sr content in wood is increased by about 5 % and in the bark by 28 %.

Co and Ni concentration

Elevated Co content in soil samples (5 and 4 times higher compared to control in the case of spruce, and 4 and 3 times for Douglas-fir) had an effect on increasing the concentration in the bark (4 and 3 times compared to control for spruce, and 7

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and 6 times in the case of Douglas-fir) but not on wood where Co concentration was below the detection limit (Figure 2 (a), Table I). Therefore, based on this experiment, the wood of spruce and Douglas-fir are not a good choice for Co. Increased Ni concentration in soil samples (3- SI and 2- SII times; 2.5- DI and 2- DII times compared to control) also have a direct effect on increasing the concentration (Figure 2(a), Table I) in both wood (1.4- SI and 1.2- SII times; 18- DI and 10- DII times compared to control) and bark (2.5- SI and 2- SII times; 3- DI and 3-DII times compared to control). About 40 % higher content of Ni in the soil was detected and about 14 % higher concentration for wood and about 52 % for the bark compared to the II experiment in the case of spruce. For Douglas-fir, it was 24 % for soil, 83 % for wood and 10 % for bark.

Mn and Zn concentration

Mn and Zn were examined because they are, among other elements, essential for higher plants and have several functions in plants.[13] As these elements weren't added in this experiment, differences compared to control are minimal, with most ratios near 1 (Figure 2(a)). Some phenomena can be seen in this experiment for examined tree species. Their concentrations in wood and bark compared to control (Table I; Figure 2(a)) which slightly decreased (except for the Mn concentration in Douglas-fir bark, which increased), despite slightly higher soil concentrations (Table I).

Slight soil concentration increases for Mn (15 %, 12 %) and Zn (9 %, 6 %) in spruce and Mn (12 %, 12 %) and Zn (15 %, 7 %) in Douglas-fir soil in I and II experiments compared to control that was observed could be the consequence of the soil pH change.[2] Other factors, such as the impact of specific tree species on soil pH and the addition of heavy metals to the soil, can also influence the adsorption dynamics of essential elements by competing with or altering the total organic content (TOC) during root uptake. Externally added heavy metals can interact with TOC, affecting the behavior and availability of Mn and Zn. The effect of TOC on Mn and Zn uptake may differ between tree species due to variations in root systems, uptake mechanisms, and tolerance to nutrient imbalances. However, these aspects were not measured in this study. In SI and SII the pH decrease was 0.1 and 0.2; and in DI and DII it was 0.1 and 0.3, respectively, compared to the control pH value. The decrease of Mn for spruce in parallel experiments (SI and SII) expressed as a percentage compared to control was 11 %, 9 % for wood, and 35 %, 19 % for bark, respectively. In the case of Zn, it was 37 %, 24 %, and 32 %, 27 %, respectively. For Douglas-fir wood, the decrease of Mn concentration was 32 %, 48 %, and the decrease of Zn concentration was 30 %, 54 % for wood and 27 %, 18 % for bark, respectively.

Studies indicate that trace elements move differently between tree organs accross different tree species,[5,20,27–29] and plants retain trace elements at varying levels.[6] Traffic is a significant source of Pb, Ni, and Zn pollution, [21,30] with Ni

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entering the atmosphere through fuel combustion, mining, and urban waste burning, while Co is mainly used in rechargeable batteries for electronics.[31] The elevated Ni levels at the control site in our experiment may be due to these factors. Pb concentration in the control soil were below regulatory limits in Serbia and similar to road dust levels in areas with moderate traffic and residential heating contributing to pollution.[30] Thus, observed elevated Pb and Ni in wood and bark from both species are direct outcomes of our experiments suggesting that bark and wood, especially of Douglas-fir, can collect and remediate these metals. The wood samples examined came from seedlings (with stem disks of 1 cm height and a volume of about 3.14 cm^3). If we scale this to mature trees (with 10 times the volume), we estimate that about 4.1 and 3.4 $\mu g\ g^{-1}$ of Pb and Ni could be collected by spruce and 10.6 and 7.3 $\mu g\ g^{-1}$ by Douglas-fir, with higher values possible when considering the full trunk. Several studies have shown elevated levels of trace elements in plants and soil in areas affected by air pollution,[5,6,31] but there is limited information on the trace element accumulation potential of many plant species. Research on Scots pines in Finland showed that Ni accumulates in wood,[29] while most elements are stored in roots.[17,18,20] Although roots were not examined, trace elements clearly moved from soil to wood and bark. Given the toxicity of Pb, Ni, and Co, and their detrimental health effects with prolonged exposure,[31] it is crucial to extract these elements from the environment. Spruce and Douglas-fir could be used to phytostabilize trace elements in soil, reducing their mobility and leaching. An indication is that the addition of other metals to the soil and their accumulation in the body of wood and bark influence essential elements, and lead to a decrease in their plant parts. Higher Mn concentrations in background trees compared to those grown on sludge were found in tree seedlings.[32] Similarly, in beech roots a decrease in mineral cations (K, Ca, Mg, and Mn) was observed with increasing Pb and Cd in soil.[33] Prolonged exposure to elevated heavy metal levels, as suggested by our findings, could further reduce nutrient levels in plants and potentially lead to plant death over time. However, this conclusion requires further research beyond this experiment. The observed decrease in Mn and Zn may also result from competition between metals during root uptake, as Ni^{2+} and Zn^{2+} have similar physical and chemical properties,[12] leading to reduced Zn levels in wood and bark due to elevated Ni in the soil.

The impact of tree species on soil pH is important, as it influences trace elements availability to plants. The effect of different tree species on soil pH is most significant in the first ten centimeters of the topsoil.[34] Topsoil pH was lower under *P. abies*, while *P. menziesii* appeared to be intermediate.[34] In this study, spruce (*P. abies*) had lower pH values than Douglas-fir (*P. menziesi*) in both control and parallel experiments (Table I). As a result, higher soil metal concentrations were observed for spruce (Figure 2 (a)). However, this did not

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translate into higher metal accumulation in wood and bark, as Douglas-fir showed greater accumulation, particularly for Ni and Pb (Figure 2 (a)).

Transfer factors

The capacity of trees to extract trace elements from soil and their translocation to aboveground tissues can be evaluated by calculating the transfer factor, TF. Three calculated ratios were presented in Figure 3.

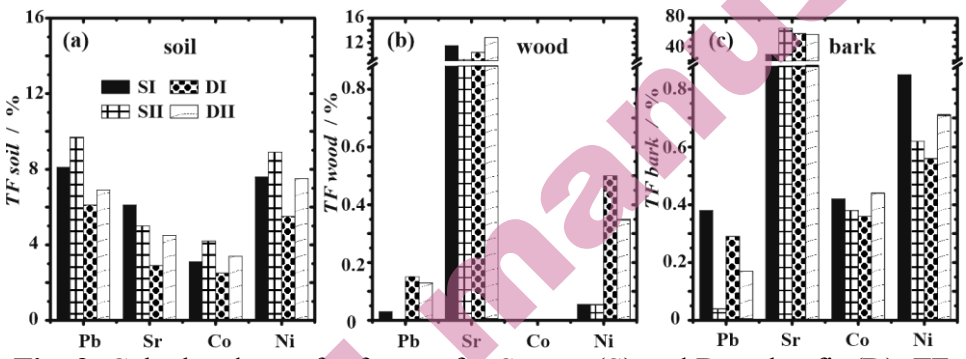

Fig. 3. Calculated transfer factors for Spruce (S) and Douglas-fir (D), TF, a) for soil- TF_{soil}, b) wood- TF_{wood} and c) bark- TF_{bark}, expressed as a percentage.

Only a small amount (less than 10 %) of the externally added elements (EA) in the soil were in a form that is available and can be absorbed by roots and translocated throughout the plant (Figure 3 (a)) in these experiments. The smallest percentage of TF_{soil} was for DI Co concentration and the highest was in the case of SII Pb concentration (Figure 3 (a)). Generally looking, the second experiment had greater TF_{soil} probably due to the greater change in soil pH for both species. Lower pH increases the metal availability due to competition between hydrogen ions and metal ions at the uptake sites in the roots.[9] In wood and bark (Figure 3 (b) and (c)), TF's are less than 1 % (except for Sr) and the highest TFs are for Sr, in both species. The Sr is chemically related to Ca and plant roots normally do not discriminate between absorption of Ca^{2+} and Sr^{2+} from nutrient solutions.[23] This could be the reason for elevated TF in wood and bark for Sr (Figure 3 (b) and (c)). Higher available Sr concentration in the soil in these experiments probably competes with the Ca concentration (a very important macronutrient for higher plants). Calculated TF_{wood} are greater in Douglas-fir than in spruce wood for examined elements and Ni and Pb in Douglas-fir stand out. Nickel is attributed to mobile elements,[25,32] and Ni uptake in our experiments confirm its high mobility. Pine, birch and black alder most efficiently took up Zn and Ni.[24] Although available soil concentrations of Pb compared to control (Figure 2(a)) are higher than for Ni, transfer factors in wood for Ni are greater. In the case of bark, transfer factors are more pronounced than in wood. The highest TF_{bark} is for Sr, then Ni, Co and Pb. Also, small differences

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between spruce and Douglas-fir transfer factors for bark can be seen (Figure 3 (c)). The bark is intensively used as a bioindicator of atmospheric pollution but uptake of Pb, Sr, Co and Ni by tree root in these experiments indicate to their translocation from the soil to the bark. This pathway has to be taken into consideration in the highly polluted areas like the one where the elements are incorporated into the bark, especially in the case of Co which couldn't be traced in spruce and Douglas-fir wood.

These experiments showed that metals added to natural soils were absorbed by tree roots and transferred to wood and bark within two years under normal conditions. Despite the soil concentration in the first experiment being double that of the second, the increase in metal content in soil, wood, and bark was not proportional. The second experiment had higher soil transfer factors (TF_{soil}) than the first, leading to a ratio of less than 2 between the experiments. Such studies provide valuable insights into heavy metal accumulation and distribution in tree species. Since plants can significantly reduce air pollution, expanding green spaces is a highly effective solution.

CONCLUSIONS

All spruce and Douglas-fir trees survived and grew normally during the two-year experiment. The general response of the two coniferous species was an increase of elements in wood and bark compared to controls, with only a slight decrease in Mn and Zn. Sr and Ni were absorbed most efficiently. While uptake wasn't directly proportional to soil metal concentrations, both species responded to elevated levels, indicating environmental pollution. Bark was also influenced by the added concentrations in the soil which has to be taken into consideration in highly polluted areas as the significant pathway. Bark, especially for Co, also acted as a useful indicator, unlike wood where Co was not detected. Thus, Douglas-fir wood can serve as a better bioindicator of Pb and Ni than spruce. Despite higher soil metal levels in spruce, Douglas-fir accumulated more Ni (18 and 10 times higher) and Pb (7 and 4 times higher), making it a better bioindicator for these metals. Expanding green spaces is a highly effective way to reduce air pollution, as plants play a significant role.

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И З В О Д

ТЕРЕНСКИ ЕКСПЕРИМЕНТ О УСВАЈАЊУ ОЛОВА, СТРОНЦИЈУМА, КОБАЛТА И НИКЛА У ДРВЕТУ И КОРИ СМРЧЕ (*Picea abies* L.) И ДУГЛАЗИЈЕ (*Pseudotsuga menziesii* Mirb.)

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Људске активности су значајно промениле доступност и циркулацију загађујућих материја, утичући на њихове концентрације у животној средини. Ово загађење посебно утиче на дрвеће. У овој студији, спровели смо два одвојена експеримента (I и II) да бисмо испитали акумулацију олова, стронцијума, кобалта и никла у садницама смрче (*Picea abies* L.) и дуглазије (*Pseudotsuga menziesii* Mirb.). Ове саднице су биле изложене повишеном нивоу ових метала додавањем у земљу. Наши теренски експерименти пружају увид у акумулацију метала у природном окружењу. Мерили смо концентрације ових елемената, заједно са манганом и цинком, у земљишту, дрвету и кори користећи индуктивно спрегнуту плазма-оптичку емисиону спектрометрију (ИСП-ОЕС). Резултати су показали повећане нивое додатих метала у дрвету и кори обе врсте дрвећа. Приметно је да је дошло до значајног повећања концентрације олова и никла у дрвету дуглазије. Концентрација олова у дрвету дуглазије је била 7 и 4 пута већа у огледима I и II у односу на контролу, док је концентрација никла била 18 и 10 пута већа. Ови резултати, на основу концентрација елемената у траговима и трансфер фактора, сугеришу да дрво дуглазије има потенцијал ка фитостабилизацији олова и никла.

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