



University of Belgrade, Technical Faculty in Bor



ECOENTER

**30th International Conference Ecological Truth
& Environmental Research
2023**

Proceedings

**Editor
Prof. Dr Snežana Šerbula**





University of Belgrade, Technical Faculty in Bor



ECOTRIP

30th International Conference Ecological Truth
& Environmental Research
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PREFACE

The 30th international conference Ecological Truth & Environmental Research – EcoTER'23 kept three areas in focus: ecology, environmental protection and sustainable development. The conference will be held on Mt Stara Planina in hotel Stara Planina, Serbia, 20–23 June 2023. The monograph is published on the occasion of the 30th anniversary of the conference. On behalf of the scientific and organizing committee, it is a great honor and pleasure to wish all the participants a warm welcome to the conference.

The monograph is published on the occasion of the 30th anniversary of the conference.

We hope to convey the message of the conference, which is that a transformation of attitudes and behavior would bring the necessary changes. This is also an opportunity for the participants who are experts in this field to exchange their experiences, expertise and ideas, and also to consider the possibilities for their collaborative research.

The 30th international conference Ecological Truth & Environmental Research – EcoTER'23 is organized by the University of Belgrade, Technical Faculty in Bor, and co-organized by the University of Banja Luka, Faculty of Technology, the University of Montenegro, Faculty of Metallurgy and Technology – Podgorica, the University of Zagreb, Faculty of Metallurgy – Sisak, the University of Pristina, Faculty of Technical Sciences – Kosovska Mitrovica and the Association of Young Researchers, Bor.

These Proceedings 103 papers from the authors coming from the universities, research institutes and industries in 11 countries: Australia, USA, Brazil, Spain, Portugal, Libya, Italy, Bulgaria, Bosnia and Herzegovina, North Macedonia, and Serbia.

As a part of this year's conference, the 5th Student Session – EcoTERS'23 is being held. We appreciate the contribution of the students and their mentors who have also participated in the conference.

The support of the Gold donor and their willingness and ability to cooperate has been of great importance for the success of the EcoTER'23. The organizing committee would like to extend their appreciation and gratitude to the Gold donor of the conference for their donation and support.

We appreciate the effort of all the authors who have contributed to these Proceedings. We would also like to express our gratitude to the members of the scientific and organizing committees, reviewers, speakers, chairpersons and all the conference participants for their support to the EcoTER'23. Sincere thanks go to all the people who have contributed to the successful organization of the EcoTER'23.

Prof. Snežana Šerbula,

President of the scientific and organizing committee

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MULTICRITERIA EFFICIENCY ASSESSMENT OF THE PINE TREE POTENTIAL FOR THE PHYTOREMEDIATION OF COPPER

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Abstract

Special emphasis in this paper was given to the multicriteria efficiency assessment of the phytoaccumulation and exclusion properties of the pine tree, as well as the Cu uptake from the soil by the root system, and further translocation to the needles, in the anthropogenically multi-medium polluted areas. Pine tree did not show the properties of the Cu hyperaccumulator plant under the environmental conditions of the study area, so pine could not be used in phytoextraction of copper from the soil. Pine tree from the sampling sites with the lowest pollution level had the properties of plant excluder of Cu. In the conditions of elevated Cu contents (>1500 mg/kg, the U1 and R2 sampling sites) in the soil, pine showed the ability to efficiently translocate Cu to the above-ground parts, which could be the tolerance mechanism to high Cu concentrations in the environment, and especially in the soil. Such fate of Cu through the soil-plant interface should be further tested in the specific physico-chemical conditions of the soil and Cu soil contents higher than the specified value.

Keywords: pine, copper, phytoremediation, translocation, environmental pollution.

INTRODUCTION

Environmental studies in the anthropogenically multi-medium polluted areas [1,2], are of the great importance from the aspects of human health protection [3] and preservation of natural resources. From the scientific point of view, the surroundings of open pit mines, overburden dumps, flotation tailing ponds and copper smelters, represent the “open air laboratory”, for multidisciplinary examinations of fate of polluting substances in the environment. Soil in these areas as the crucial environmental component, is a major sink of heavy metals released from the pollution sources. Change in the chemical and biological characteristics of the polluted soil [4] has huge consequences, and also affects the physico-chemical and metabolic functions of plants. Because of that, polluted soil and plants that exist on it, are the most significant elements in the studies of the possibilities of phytoremediation. Due to the adaption and tolerance to different environmental stresses, naturally abundant species have priority in this method [5]. Different species of *Pinus spp.* had good adaptability in polluted environment, and could be used in the phytoremediation and restoration of polluted soil [6–8].

The aim of this study was the assessment of the pine tree ability to absorb Cu from the soil and translocate it through the plant tissues, upon different soil pollution level. The obtained results were also used in the evaluation of possibilities of using pine tree in phytoremediation.

MATERIALS AND METHODS

Study area and analyses of samples

Sampling of soil and plant material was conducted at ten sampling sites: UI-urban-industrial; U-urban; SU-suburban; I1 and I2 (industrial zone); T1-Brestovac spa and T2-Bor lake (tourist zone); R1-village Oštrej and R2-village Slatina (rural zone); and B-Gornjane (background, unpolluted zone). It should be noted that impacts of pollution sources at most of the sampling sites are overlapping, because of the close vicinity of the mining and smelting facilities.

Soil from the surroundings of roots, as well as roots and needles of pine were sampled and prepared for chemical analyses according to the methodology described in Kalinovic *et al.* [9]. All chemical analyses were conducted by accredited chemical laboratory, at the Institute of Mining and Metallurgy Bor (Serbia). Samples of soil and plant material were digested according to the U.S. EPA method [10], in a microwave oven. Concentrations of Cu in extracts were determined by simultaneous dual view inductively coupled plasma atomic emission spectrometer (ICP-AES).

Data analyses

Soil and plant Cu concentrations were analysed using SPSS Statistics software. Possibilities of using the pine tree in phytoremediation were defined based on the bioaccumulation factor for roots and washed needles (BAF_{roots} , eqn. (1); BAF_{needles} eqn. (2)), translocation factor (TF, eqn. (3)) [8,11], as well as, by comparing the Cu concentrations in the washed needles with the concentrations which foliar parts of plants hyperaccumulators or excluders should contain [12].

$$BAF_{\text{roots}} = \frac{C_{\text{roots}}}{C_{\text{soil}}} \quad (1)$$

$$BAF_{\text{needles}} = \frac{C_{\text{needles}}}{C_{\text{soil}}} \quad (2)$$

$$TF = \frac{C_{\text{needles}}}{C_{\text{roots}}} \quad (3)$$

In the specified equations, C_{soil} , C_{roots} and C_{needles} represented Cu concentrations in the samples of soil, roots and needles, respectively.

If at the same time the following criteria are met: the aboveground plant part contain the Cu concentrations which are characteristic for plants hyperaccumulators (≥ 300 mg/kg Cu) [13]; and $BAF_{\text{roots}} > 1$, $BAF_{\text{needles}} > 1$ and $TF > 1$, then the plant was considered a hyperaccumulator, i.e. the plant had the abilities for being used in phytoextraction [5].

If the values of BAF_{roots} , BAF_{needles} and TF were < 1 , then the plant had the properties of excluder [5]. However, it should not be omitted that excluder plants certainly uptake necessary quantities of essential elements, which enable their primary metabolic functions [14]. It was suggested that Cu concentrations in excluder plants should not exceed values of

40 mg/kg [12]. Excluder plants can accumulate highest amounts of a certain element in their roots, compared to other parts, but translocation to the aboveground parts is limited. This property is suitable for the use of plants in phytostabilization.

If $BAF_{\text{roots}} < 1$, $BAF_{\text{needles}} < 1$ and $TF > 1$, only efficiency of Cu translocation from the roots to needles was considered. If Cu translocation from the roots to needles is efficient, then the roots compared to the washed needles should contain lower Cu concentrations.

The ranges of Cu concentrations in the soil from the sampling sites where the values of TF were < 1 and the range of Cu concentrations in the soil from the sampling sites where the values of TF were > 1 , were compared. In the soil Cu concentration zone of overlapping these values ($TF < 1$ and $TF > 1$), it was considered that Cu soil concentrations did not have dominant effect on the Cu translocation from the roots to needles. This method of analyses of the results indicated the Cu translocation efficiency in the range of lower and in the range of higher Cu concentrations in soil. That was one of the ways to define a strategy that pine tree uses to adapt to the increased Cu concentrations in the soil.

Based on the values of BAF, intensity of absorption of some elements from soil into roots and needles were defined. Depending on the values of the bioaccumulation factors, absorption was classified as follows: 10–100 (intensive absorption), 1–10 (strong absorption), 0.1–1 (average absorption), 0.01–0.1 (weak absorption) and 0.001–0.01 (very weak absorption) [6].

RESULTS AND DISCUSSION

Cu concentrations in the samples of soil and roots and needles from 10 sampling sites, were presented in Figure 1. It should be mentioned that Cu contents of the soil from all the sampling sites exceeded the limit value (LV) for soil of 36 mg/kg, while the remediation value (RV) (190 mg/kg) was not exceeded only in the soil from the B sampling site [15]. These exceedances indicated that the fundamental functions of the soil in the studied area were seriously disturbed and required remediation, recovery and other measures. This is of great importance for the soil in the rural zone, where the agricultural activities of cultivation of food for human and animal consumption took place.

The minimum Cu concentration of 67.89 mg/kg was determined in the soil from the B-unpolluted sampling site. Cu concentration that was marked with asterisk represented extreme Cu content determined in the soil from the UI sampling site that was three times higher than the values which were fell within 75% of the presented Cu soil concentrations.

The contents of Cu in the pine roots ranged from 3.36 $\mu\text{g/g}$ at the UI sampling site, to 44.42 $\mu\text{g/g}$ at the B sampling site, which was not expected due to the environmental and especially soil pollution in the study area (Figure 1b). Neither of the detected Cu concentrations in the roots was statistically characterized as atypical or extreme. Cu concentrations in the needles were in the range of 4.83–164.13 $\mu\text{g/g}$, where the minimum was detected in the needles from the B sampling site, while maximum was detected in the needles from the UI sampling site. Concentrations of Cu in needles sampled from the T2, R2, U, SU and UI sampling sites were higher than the defined critical for foliar parts of higher plants [16]. There was one atypical Cu concentration detected in the needles from the R2 sampling

site, which was more than 1.5 times greater than the 75% data values. Cu concentration in the needles from the UI sampling site represented extreme value that was three times higher than the 75% data, which was in accordance with the level of the pollution of the specified sites.

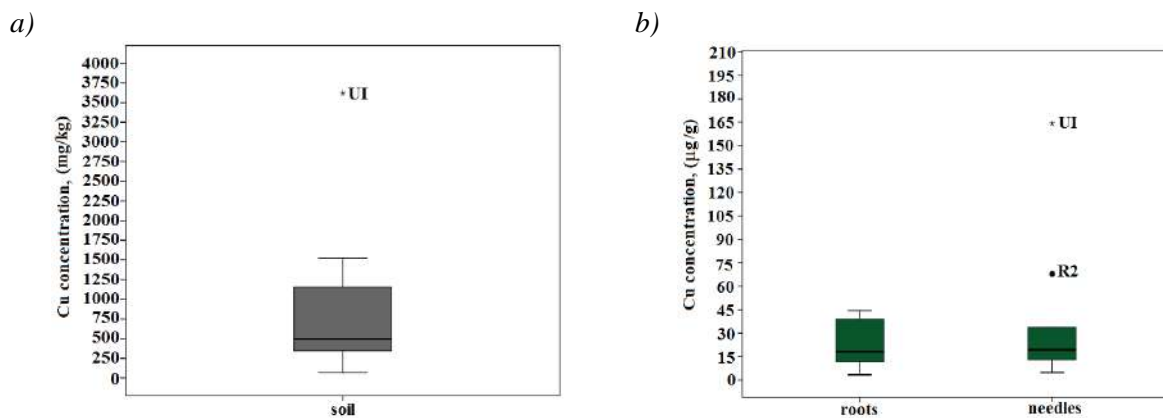


Figure 1 Box plots of Cu concentrations in the samples of a) soil; b) roots and needles of pine

Pine sampled from the study area could not be specified as Cu hyperaccumulator plant, because Cu content in the washed needles did not exceed 300 µg/g. The BAF_{roots} and BAF_{needles} at all the sampling sites were <1 (Table 1), which also indicated that pine cannot be used in the phytoextraction of copper, under the physico-chemical soil and air conditions at the studied sampling sites. Because of that, the results were considered from the aspect of the efficiency of the roots and needles Cu uptake from soil, but also the efficiency of their translocation from the roots to needles.

Table 1 BAF, and TF for Cu in pine tree, depending on the sampling site

	UI	SU	U	R1	T1	I1	R2	T2	I2	B
BAF_{roots}	0.001	0.034	0.082	0.012	0.059	0.033	0.009	0.034	0.075	0.654
BAF_{needles}	0.045	0.089	0.052	0.030	0.020	0.038	0.045	0.022	0.029	0.071
TF	48.833	2.601	0.629	2.462	0.334	1.139	4.835	0.642	0.388	0.109

Absorption of Cu in the roots from the soil at all the sampling sites was weak, except at the UI and R2 sampling sites where the absorption was very weak. Concentrations of Cu in the soil from these sampling sites compared to the rest of the sampling sites were the highest (3620.19 mg/kg and 1524.25 µg/g, respectively). The absorption efficiency of Cu in the needles from the roots was weak at all the sampling sites.

At the U, T1, T2, I2 and B sampling sites, values of TF were <1 . The needles from the specified sampling sites had Cu content lower than 40 µg/g, which was defined as one of the requirements which excluder plants s should fulfil [12]. However, for the samples from the U, T1 and T2 sampling sites, the condition that roots should contain higher Cu concentrations compared to the needles, was not met [17]. Higher Cu contents in the roots than in the needles, were only at the sampling sites I2 and B [17], where Cu in the needles (8.66 µg/g and 4.83 µg/g, respectively) did not exceeded the critical value of 20 µg/g [16] for the needles. Because of that, environmental conditions at the I2 and B sampling sites where pine was

sampled favour the pine Cu excluder properties. The Cu concentrations in the soil samples from these sampling sites (67.89 and 298.67 mg/kg, respectively), were lower compared to the concentrations of Cu in the soil from the other sampling sites. Cu concentrations in the pine roots from the U, T1 and T2 sampling sites, where the values of TF were <1 , were lower than Cu contents in the washed and unwashed needles.

The values of TF were >1 for the samples from 5 (UI, SU, R1, I1 and R2) out of 10 sampling sites. That means that at these sampling sites, and especially at the UI and R2 (the highest Cu soil contents were detected), Cu was efficiently translocated from the roots to the needles. Concentrations of Cu in the needles from the UI and R2 sampling sites (164.13 $\mu\text{g/g}$ and 67.94 $\mu\text{g/g}$, respectively), which were higher than the critical, also indicated that pine tree had the ability to efficiently translocate Cu from the soil with the highest Cu contents to the above-ground parts, but the possibility of the Cu absorption into the needles from the atmospheric deposition must not be ignored. These results indicated that the pine trees that were growing at the UI, SU, R1, I1 and R2 sampling sites, possess specific tolerance mechanisms to high Cu concentrations in the environment, and especially in the soil.

The range of Cu concentrations in the pine soil where the values of TF were <1 , was 67–1156.33 mg/kg, while the values of TF were >1 at the sampling sites where Cu concentrations were in the range of 345.54–3620.19 mg/kg. These two ranges of the Cu soil concentrations at the sampling sites where $\text{TF}<1$ and $\text{TF}>1$, overlap for some of the Cu soil concentrations. Based on that, it was supposed that in the zone of overlapping, Cu concentrations in soil did not have dominant influence on the translocation of Cu from the roots into the aboveground pine parts, at the environmental conditions that prevailed at the specified sampling sites. According to the obtained results, it could be said that with increasing concentrations of Cu in the pine soil over about 1500 mg/kg, which represented Cu concentration higher than concentrations in zone of overlapping, pine uses mechanism of avoiding Cu toxicity, by translocation of Cu from the roots to the needles. This was the case at the sampling sites R2 and UI, which are the two most polluted areas of the Bor city and the surroundings.

CONCLUSION

The obtained results indicated that soil sampled from all the sampling sites (except the B sampling site) at different distances from the pollution sources such as copper smelter, flotation tailings, tailing ponds, open pit mines, was polluted with Cu, required the remediation, recovery and other measures. The maximum Cu content in the soil and washed needles was determined in the samples from the urban-industrial sampling site, which was previously proven to be the most endangered area in the Bor region. Unlike that, the unexpected lowest Cu content had the roots from the UI sampling site, and the highest Cu content was detected in the roots from the B sampling site. The sampled pine tree did not show the properties of the hyperaccumulator plant, so pine could not be used in phytoextraction of copper, under the environmental conditions of the study area. Pine sampled from the I2 and B sampling sites had the properties of plant excluder. It was noticed that pine tree had the ability to efficiently translocate elevated Cu contents from the soil to the above-ground parts, that could be the specific tolerance mechanism to high Cu concentrations in the

environment, and especially in the soil. Such specific Cu fate in the soil-needles interface was determined for the concentrations of Cu in the pine soil over about 1500 mg/kg (UI and R2 sampling sites). This specific behaviour should be further tested in the conditions of Cu soil contents higher than the specified value.

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