

Impact of biochar on the mobility of heavy metals in reclaimed sites from mining activities in the Medet mine area

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Abstract

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Three reclaimed soils (~ 20 years old) formed on embankments in the Medet mine from the Assarel-Medet mining and processing complex were studied. An investigation was conducted on the influence of biochar (BC) in different doses 1%, 5%, 10% and 20% on the properties and immobilization of heavy metals in technogenic soils in a laboratory incubation experiment, for a period of six months. Mobile forms of the studied metals were determined after extraction with 0.01 M CaCl₂ and 1M NH₄NO₃.

An increase in pH values of the studied soils was noted, both with increasing rates of biochar and over the incubation time. Biochar was successful in decreasing the mobile (0.01M CaCl₂) and bioavailable (1M NH₄NO₃) forms of Cu, Co and Pb, however for the other studied metals, Fe, Mn and Zn there is no clear trend of the effect of biochar in decreasing their mobility and availability.

The results of this study confirm our previous findings that Cu and to some extent Pb in the soils of the area were behaving as anthropogenic pollutants, which can be immobilized by biochar, while the mobility and bioavailability of Co, Fe, Mn and Zn were less influenced by the biochar added.

Keywords: reclaimed soils; heavy metals; biochar; immobilization

Introduction

Biochar produced by pyrolysis of biomass (wood, agricultural and forestry residues) is a porous solid carbon material that has attracted increasing attention for its applications in environmental remediation (removal of pollutants). It is characterized by a large specific surface area (SSA), high cation exchange capacity (CEC), is rich in functional groups, and has high chemical and biological stability, which can effectively reduce the migration and bioavailability of heavy metals in soil (Gao et al., 2022; Atanassova et al., 2024;

Stoykova et al., 2024). In addition, biochar can further increase the adsorption effect on heavy metals by modifying or creating composites (Nosratabad et al., 2024). It also reduces the inhibition of root growth by heavy metals and reduces their phytotoxicity by 55.8. Biochar can provide favorable conditions for plant growth and soil microbial activity (Gao et al., 2022). The application of biochar to heavy metal contaminated soil can lead to metal immobilization, by increasing the cation exchange capacity, organic matter content and pH, (Medyńska-Juraszek and Ćwielałag-Piasecka, 2020; Ibrahim et al., 2022; Atanassova et al., 2024; Stoykova et al.,

2024). Alaboudi et al. (2019) in their study found that biochar has the potential to stabilize Pb and Cd and transform them from the labile fraction into less accessible forms. It significantly reduced the solubility of Pb and Cd in terms of water-soluble and exchangeable forms (extracted with ammonium nitrate). The largest reductions for Pb and Cd were 28.68% and 85.14%, respectively.

The purpose of the present investigation was to assess the influence of biochar application on the mobility of Cu, Pb, Zn, Co, Fe and Mn in reclaimed sites from mining activities in the Medet mine area.

Materials and Methods

Assarel-Medet mining and processing complex, situated near the town of Panagyurishte (Central Bulgaria) is the first and largest mining enterprise in Bulgaria for open-pit mining and processing of copper ores. The studied soils (pooled samples from eight points) were taken from: The “Great Southern” embankment – T1, T2, T3 and T4 materials from the overburden of the Medet mine with biological reclamation carried out in the period 1998–2006 with pine and birch plantations. Soil samples – T5, T6, T7 and T8 were taken from a tailings pond from the open-pit mining of the enterprise, stored a few kilometers from the Medet mine to “Stena Zlatishko Kale”, biological reclamation was carried out from 2001 to 2007 with afforestation of pine (*Pinus silvestris* and *Pinus nigra*) and birch (*Betula*). After a preliminary baseline study of the soil samples from the Medet site (Atanassova et al., 2023), soils T1, T4 and T7 were selected, based on their highest concentration of heavy metals exceeding the maximum permissible concentrations regulated in Bulgarian legislation (Regulation 3, 2008), and prepared for further investigation. An incubation experiment was conducted in a Climatic Chamber with Phytotron System KK 350 FIT DS–POL-EKO, 2022 for a period of six months. Before the incubation experiments, the soils were air-dried, then crushed and sieved through a 2 mm sieve. The experimental scheme includes one control variant without the addition of biochar (BC) and four variants with increasing biochar rates (1, 5, 10, 20% w/w) in three replications of the pooled sample. The highest dose of BC 20% in the present investigation approximate 200 t.ha⁻¹ of BC applied in 0–10 cm soil depth. This dose, is higher than usual, used in agricultural practice, but in specific conditions when contaminated soils are studied, such norms are also reported. In our greenhouse experiment (unpublished data) a positive effect was recorded after 20% of BC application on the immobilization of heavy metals and Alfalfa (*Medicago sativa*) biomass production. The measured amount of biochar was also sieved through 2 mm and

homogenized with the soil according to the variants, with distilled water added, in order to reach 75% of the WHC (Wraith and Or, 2001). All containers were weighed before placing them in the thermal chamber: T 20 °C, air humidity 65%, light level 30 and fan level 3, parameters selected to simulate climatic conditions during the spring-summer months. The biochar used was from deciduous woody vegetation (birch, sycamore, ash and maple), pyrolyzed at 400–420 °C (Nikimol OOD, Asenovgrad).

Changes in pH, electrical conductivity, and redox potential of the studied soils were monitored monthly for a period of 6 months. The concentrations of the mobile forms of HM (Cu, Pb, Zn, Co, Fe and Mn) in the soil (0.01M CaCl₂), were analyzed. After a period of three months of composting with the introduced ameliorant (from the third to the sixth stage of the experiment), the dynamics of the bioavailable forms of the elements (Cu, Pb, Zn, Co, Fe and Mn) in the soil (1M NH₄NO₃), were study.

Extraction of heavy metals from soil, using non-buffered salt solutions, e.g. 0.01 M CaCl₂, 1M MgCl₂ or 1M NH₄NO₃, which mainly dissolve soluble and cation-exchange fractions are popular methods used for the evaluation of soil contamination and the availability of heavy metals to plants (Atanassova et al., 2024; Atanassova et al., 2020; Atanassova et al., 2019; Nenova et al., 2018).

At the end of the experiment, at the 6th month of incubation, changes in the physicochemical properties of the soil with the introduction of BC-20% were monitored. The following analytical methods were used: total organic carbon (TOC) by the Tyurin method (oxidation with K₂Cr₂O₇/H₂SO₄) and the dry combustion method at 1200 °C (Primacs SNC100 – SKALAR). The physicochemical characteristics of the cation exchange capacity (CEC), measured by saturation with K malate at pH 8.2, strongly acidic and weakly acidic positions of the soil adsorbent, exchangeable forms of Ca, Mg and Al, degree of saturation with bases V% were determined by the method of Ganev and Arsova (1980); electrical conductivity (EC) in the soil: water ratio (1:5) was according to ISO 11265:2002 and pH/Eh of the soil were measured in a soil: water suspension 1:2.5.

Heavy metals in soils (were determined by the following methods: 0.01 M CaCl₂ extractable forms at a soil: solution ratio (1:5) and shaking for 2 hours (van Ranst et al., 1999). The extraction of 1M NH₄NO₃ was performed at a soil: solution ratio of 1:2.5, shaking for 1 h, filtration through a blue ribbon filter (Zeien and Bruemmer, 1989), and the elements were determined by ICP-OES (Agilent ICP-OES 5800). Statistical processing of the data for mobile forms of the elements in the extracts with 1M NH₄NO₃ and 0.01M CaCl₂ was performed using one-way ANOVA. Data were averaged

for the period III-VI month of the experiment and The Statgraphics Centurion XVIII package was used.

Results and Discussion

In Table 1 the concentration of total forms of metals and metalloids in the biochar are presented. After water extraction of the biochar the main cations and anions in solution were measured (Table 2). From the results obtained it could be stated that biochar is not a source of additional contamination in the studied soils. The biochar used has 10.4 cmol.kg⁻¹ CEC, due to the presence of carboxylic and phenolic acid groups, as a result of its production at a lower temperature (400–420 °C) (Atanassova et al., 2024). The total organic carbon (TOC) content determined by oxidation with K₂Cr₂O₇ is 19.80%, and by dry combustion in oxygen environment, 64.25% C and 0.027% N (Table 3).

The pH values of the soils (Fig. 1) showed an increase, both in the incubation process over time and with the addition of biochar. With the addition of 20% BC, the increase in pH is the largest in T1 – 1.2 units. In T4, the increase is the lowest with 0.8 compared to the control.

The addition of biochar with an alkaline reaction (pH

8.1) increases the pH of the soil-biochar mixture, due to adsorption of protons and/or desorption of OH⁻, caused by acid-base interactions and complexation processes occurring on the soil adsorbent, through the exchange of organic ligands mobilized by biochar with covalently bound OH- or COOH- groups. The oxidation-reduction potential (Fig. 2) in mV increases after the second stage, and by the end of the study, at the 6th stage indicating the occurrence of oxidation processes in the studied soils, both with an increase in the biochar rates and over time. It is known that metal mobility is governed by both complexes with soil organic matter (SOM) and physicochemical properties of the soil, such as pH, redox potential, soluble organic matter (DOC), speciation, and environmental factors (Atanassova et al., 2020).

Perfanova et al. (2025) found that after the introduction of biochar into the reclaimed soil, microbiological processes occur with varying intensity, depending on the percentage of BC added and the period of study. The biochar has a positive effect on ammonifying microorganisms and bacteria that absorb mineral nitrogen, but a period of time is needed for their adaptation and activation. At the 6th month after introducing biochar into the soil, ammonification and mineraliza-

Table 1. Content of total concentrations of metals in biochar

	Cd	Co	Cr	Cu	Fe	Mn	Mo	Ni	Pb	Zn	As	Hg	Se
	mg/kg												
BC	0.28	0.44	1.20	5.81	923.50	212.00	1.45	2.24	13.16	186.95	0.03	0.02	0.23

Source: Authors' own elaboration

Table 2. Main cations and anions in H₂O extraction of biochar

	NO ₃ ⁻	SO ₄ ²⁻	PO ₄ ³⁻	Cl ⁻	DOC	Cd	Cu	Cr	Ni	Pb	Hg	Zn	Fe	Mn	Mg	Ca	Na	K	As
	mg/L																		
BC	3	4.65	3.50	4.25	16.80	n.d	0.02	0.002	n.d	0.007	n.d	0.012	0.079	0.14	1.40	5.245	0.38	3.75	n.d

Source: Authors' own elaboration

Table 3. Physico-chemical characteristics of biochar and soils after the sixth month of incubation. Determination of total organic carbon and nitrogen by the dry combustion method on Primacs SNC100 – SKALAR and determination of total carbon by combustion in 1200 °C; Total organic carbon (TOC) by the Tyurin method (oxidation with K₂Cr₂O₇/H₂SO₄)

	pH/ H ₂ O	CEC	CEC _{SA}	CEC _{WA}	H _{8,2}	Al	Ca	Mg	Base satur.	TOC	TOC Dry combustion
	cmol. kg ⁻¹									%	
BC	8.1	10.4	–	–	0.0	0.0	7.2	3.3	100.00	19.80	64.25
T1 Control	4.6	17.2	12.2	5.0	7.0	1.8	7.5	2.5	59.30	0.86	
T1-20% BC	5.8	17.7	14.1	3.6	4.2	0.6	10.9	2.5	76.27	4.01	
T4 Control	4.9	19.5	13.0	6.5	8.1	1.5	9.0	2.3	58.46	1.08	
T4-20% BC	5.7	20.0	17.0	3.0	4.0	0.8	13.7	2.3	83.33	4.90	
T7 Control	4.9	19.5	13.8	5.7	7.1	1.4	10.0	2.2	63.59	1.24	
T7-20% BC	6.0	20.3	16.1	4.2	3.9	0.0	14.0	2.3	80.79	4.32	

Source: Authors' own elaboration

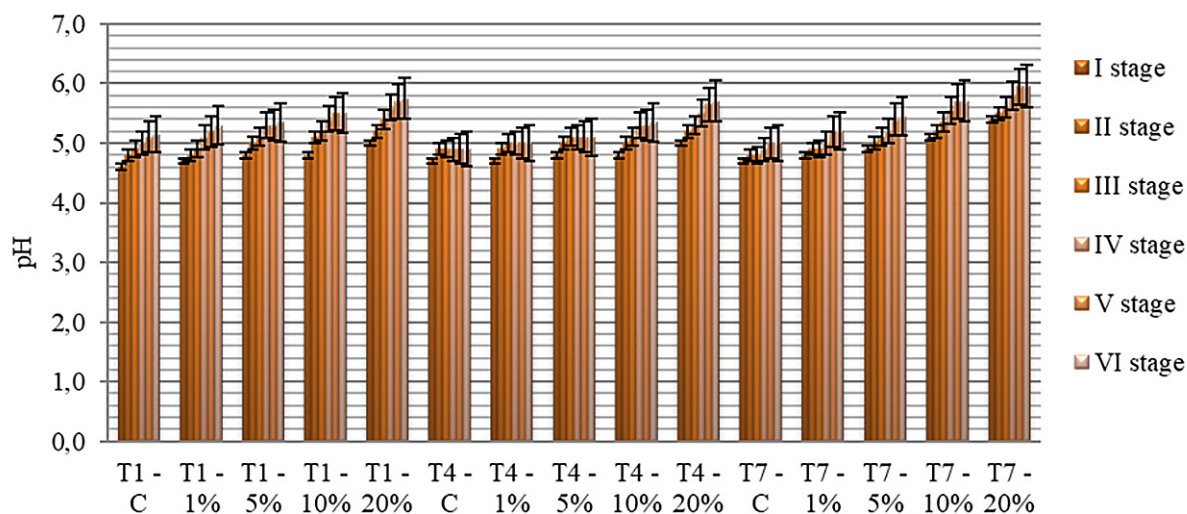


Fig. 1. Changes of pH values of soils from the Medet site

Source: Authors' own elaboration

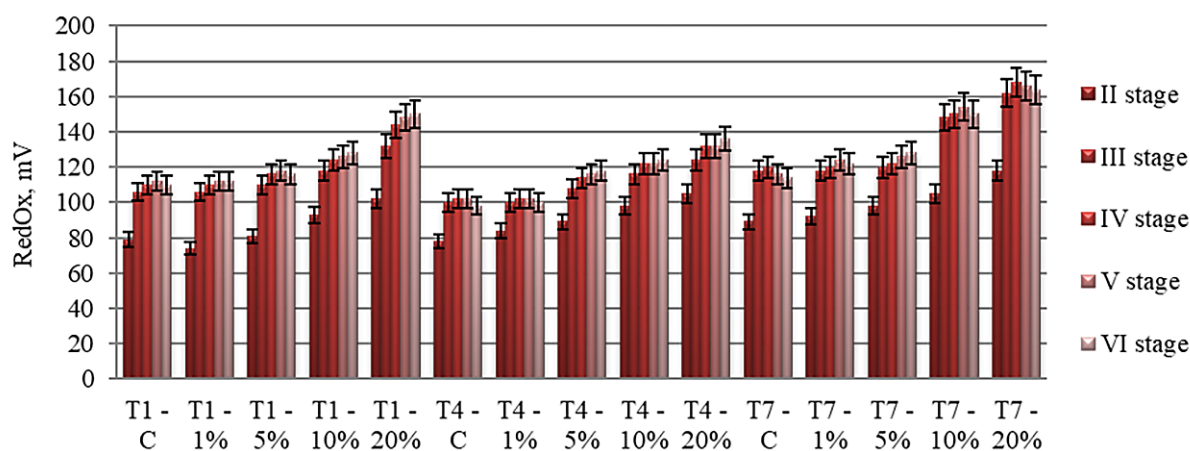


Fig. 2. Changes of the oxidation-reduction potential (RedOx) of the soils from the Medet site

Source: Authors' own elaboration

tion processes are actively occurring. Actinomycetes have a greater prevalence at the third month compared to the sixth, but even during this period they remain in larger quantities compared to the control variants. Biochar ensures favorable development of microscopic fungi and during both stages of the study they show a large increase.

In another study of the reclaimed soils from the area, Atanassova et al. (2023) found predominantly acidic reaction and higher than the maximum permissible concentration (MPC) regulated by national legislation for Cu content in three of the sites. The statistical analysis shows that Cu belongs to a different source in the reclaimed soils, while

Fe, Zn, Co and Mn are from heterogeneous sources, e.g., anthropogenic, and biopedogenic sources. Lead belongs to an external anthropogenic source introduced to the reclaimed soils, most likely through the process of waste transportation during the excavation of the ore mass and subsequent flotation.

The influence of biochar on the mobility of Cu, Pb, Fe, Zn, Co and Mn and their behavior after the 3rd month of composting was monitored. Copper is the main pollutant in the reclaimed soils from the area of the Medet copper mine. In the extract with 1M NH_4NO_3 and 0,01M CaCl_2 , the trends of decreasing copper concentrations with BC application are

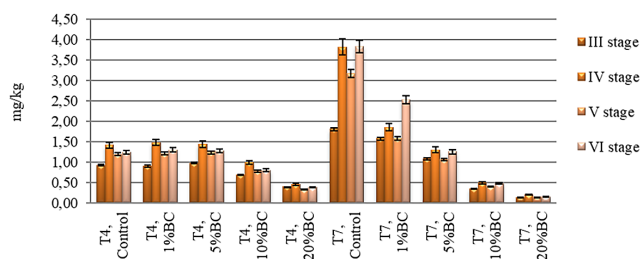


Fig. 3a. Copper extracted with 1M NH_4NO_3 , by stages and by variants in T1 – „Great Southern Embankment“ (SE 2–5%)

Source: Authors' own elaboration

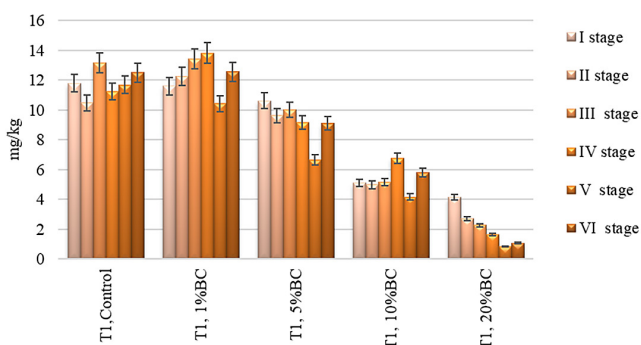


Fig. 3b. Copper extracted with 1M NH_4NO_3 , by stages and by variants in T4 – “Great South Embankment” and T7 – Tailings Reservoir next to “Zlatishko Kale Wall” (SE 2–5%)

Source: Authors' own elaboration

very well outlined (Fig. 3a, b; Fig. 4a, b), but no decrease in mobile forms is observed with the incubation time.

As a result of adding biochar, a 2 – 19-fold decrease in copper concentration was found when 1–20% BC was introduced.

Table 4 describes statistical processing of the data on the concentration of mobile forms of copper in the extracts with 1M NH_4NO_3 and 0,01M CaCl_2 using the One-way ANOVA method.

In all the three studied soils, T1, T4 and T7, the introduction of BC leads to a significant decrease in copper concentrations ($p \leq 0.001$) (Table 4). In soil T1, which is the most heavily contaminated, the average copper concentrations decrease from 23.0 mg.kg^{-1} in the control variant to 5.0 mg.kg^{-1} in variant T1 20% BC, with significant differences being established between the control and the two highest doses of biochar, 10 and 20%. In the soil from site T4, the trend is the same, variants T4 – 10% BC and T4 – 20% BC fall into separate homogeneous classes. In soil T7, the concentrations of

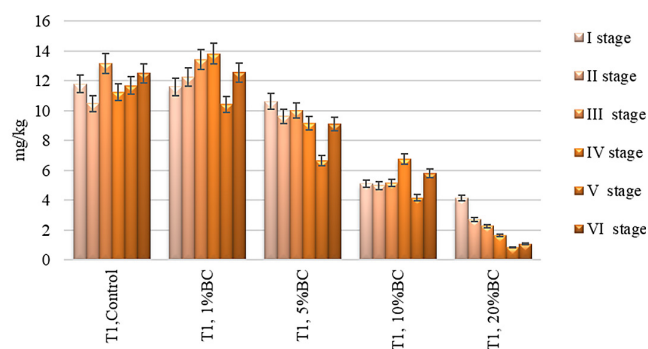


Fig. 4a. Copper extracted with 0.01M CaCl_2 by stages and by variants in T1 – „Great Southern Embankment“ (SE 2–5%)

Source: Authors' own elaboration

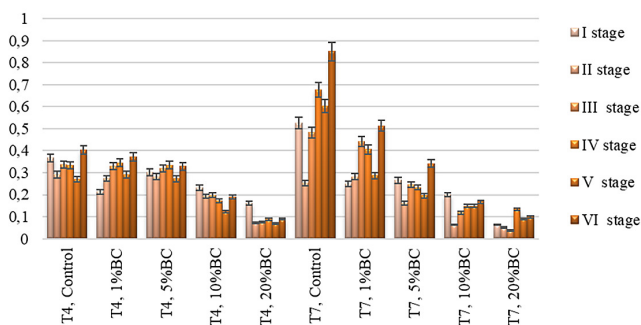


Fig. 4b. Copper extracted with 0.01M CaCl_2 , by stages and by variants in T4, “Great South Embankment” and T7, Tailings Reservoir next to “Zlatishko Kale Wall” (SE 2–5%)

Source: Authors' own elaboration

mobile forms of Cu are significantly reduced even with the introduction of the lowest dose of BC – 1%, and in variant T7 – 20% BC the average copper content is reduced 19 folds compared to the control. In this soil the effect of BC application is very pronounced, which is also seen in the complete neutralization of exchangeable acidity (exch. Al), recorded at the end of the experiment with the addition of 20% BC (Table 3). The trend of decreasing mobile forms of Cu in the extract with 0.01M CaCl_2 is the same, as it extracts lower concentrations compared to 1M NH_4NO_3 .

The change in the Pb values in the ammonium nitrate extract shows that the highest values are for the mobile forms of lead in site T4 (Fig. 5). In this soil, the influence of the addition of biochar to immobilize lead in the soil is also well manifested. It can be seen that the values of the control vary from 0.10 to 0.15 mg.kg^{-1} , while at the highest value of the addition of BC-20% in variant T4.20%, the values range

Table 4. Statistical processing of data using the One-way ANOVA method of mobile forms of Cu mg kg⁻¹, extracted with 1M NH₄NO₃ and 0.01M CaCl₂

Variants	Cu, mg.kg ⁻¹ – T1 Soil		Cu, mg.kg ⁻¹ – T4 Soil		Cu, mg.kg ⁻¹ – T7 Soil	
	1M NH ₄ NO ₃	0.01M CaCl ₂	1M NH ₄ NO ₃	0.01M CaCl ₂	1M NH ₄ NO ₃	0.01M CaCl ₂
Control	23.0±6.2 ^{c**}	11.8±0.9 ^d	1.19±0.2 ^c	0.33±0.05 ^c	3.15±0.9 ^c	0.57±0.20 ^d
1% BC	27.5±8.4 ^e	12.3±1.2 ^d	1.22±0.2 ^c	0.30±0.06 ^c	1.88±0.5 ^b	0.36±0.10 ^c
5% BC	21.4±5.1 ^{bc}	9.2±1.4 ^c	1.23±0.2 ^c	0.31±0.03 ^c	1.17±0.1 ^b	0.24±0.06 ^{bc}
10% BC	14.2±4.2 ^b	5.3±0.9 ^b	0.81±0.1 ^b	0.18±0.04 ^b	0.43±0.07 ^a	0.14±0.04 ^{ab}
20% BC	5.0±1.6 ^a	2.1±1.2 ^a	0.39±0.04 ^a	0.09±0.03 ^a	0.16±0.03 ^a	0.08±0.03 ^a
P-Value	0.0004	0.0000	0.0000	0.0000	0.0000	0.0000
LSD (95%)*	8.4	1.4	0.27	0.05	0.71	0.13

*Confidence level 95%

**Lowercase Latin letters indicate homogeneous groups, while variants that do not share a common letter are significantly different from each other.

Source: Authors' own elaboration

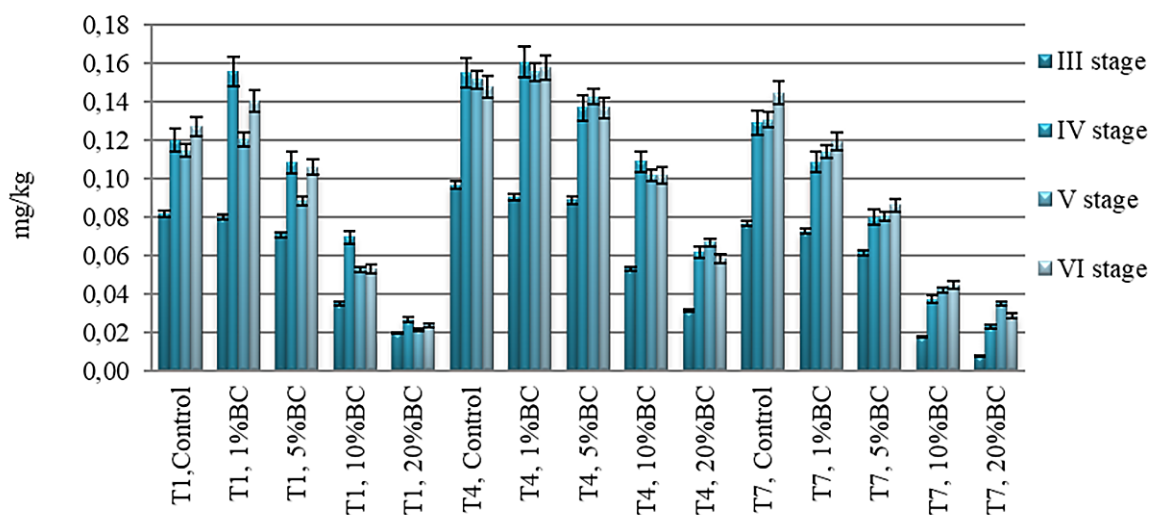
Table 5. Statistical processing of data using the One-way ANOVA method of mobile forms of Pb mg kg⁻¹, extracted with 1M NH₄NO₃ and 0,01M CaCl₂

Variants	Pb, mg.kg ⁻¹ – T1 Soil		Pb, mg.kg ⁻¹ – T4 Soil		Pb, mg.kg ⁻¹ – T7 Soil	
	1M NH ₄ NO ₃	0,01M CaCl ₂	1M NH ₄ NO ₃	0,01M CaCl ₂	1M NH ₄ NO ₃	0,01M CaCl ₂
Control	0.11±0.02 ^{b**}	0.04±0.03 ^b	0.14±0.03 ^c	0.04±0.02 ^a	0.12±0.03 ^c	0.03±0.02 ^a
1% BC	0.13±0.03 ^b	0.03±0.01 ^{ab}	0.14±0.04 ^c	0.03±0.02 ^a	0.11±0.02 ^{bc}	0.03±0.01 ^a
5% BC	0.10±0.02 ^b	0.03±0.01 ^{ab}	0.13±0.03 ^{bc}	0.03±0.02 ^a	0.08±0.01 ^b	0.03±0.01 ^a
10% BC	0.05±0.02 ^a	0.03±0.01 ^{ab}	0.09±0.03 ^{ab}	0.03±0.01 ^a	0.04±0.01 ^a	0.02±0.02 ^a
20% BC	0.02±0.01 ^a	0.02±0.01 ^a	0.06±0.02 ^a	0.03±0.01 ^a	0.03±0.01 ^a	0.02±0.02 ^a
P-Value	0.0000	0.3587	0.0012	0.7496	0.0000	0.9175
LSD (95%)*	0.03	0.02	0.04	0.02	0.03	0.02

*Confidence level 95%

**Lowercase Latin letters indicate homogeneous groups, while variants that do not share a common letter are significantly different from each other.

Source: Authors' own elaboration

**Fig. 5. Lead mobility over time and as a result of increasing BC rate (SE 2–5%), extracted with 1M NH₄NO₃**

Source: Authors' own elaboration

from 0.03 to 0.07 mg.kg⁻¹. In the other two studied soils, the lead concentrations are lower (0.08–0.13 in T1; 0.08–0.14 in T7), but here, a pronounced tendency in decreasing of mobile forms of lead is also clearly visible (at 20% BC 0.02–0.03 in T1; 0.01–0.04 in T4). For all the three sites T1, T4 and T7, the soils are Brown Forest, shallow, slightly eroded, slightly sandy-clayey with a dark brown humus horizon and relatively poorly stocked with organic carbon (TOC T1-1.01%; T4-1.36%; T7-0.91%). After adding biochar, the organic carbon values at the 6th stage increase in all three soils (Table 5), and

the mobility of lead decreases (Fig. 5). The immobilization of lead is due to its adsorption by soil organic matter Gustafsson et al. (2011). In a study by Atanassova et al. (2019), when investigating the mobility of heavy metals in contaminated, hydrophobic soils, the exchangeable forms of metals, including lead, are also well correlated to the amount of the total organic carbon (TOC), CEC, and % sand in the soil.

Statistical processing of the data for mobile forms of lead in two extractants 1M NH₄NO₃ and 0.01M CaCl₂ (Table 5) shows that in 1M NH₄NO₃ extraction significant differences

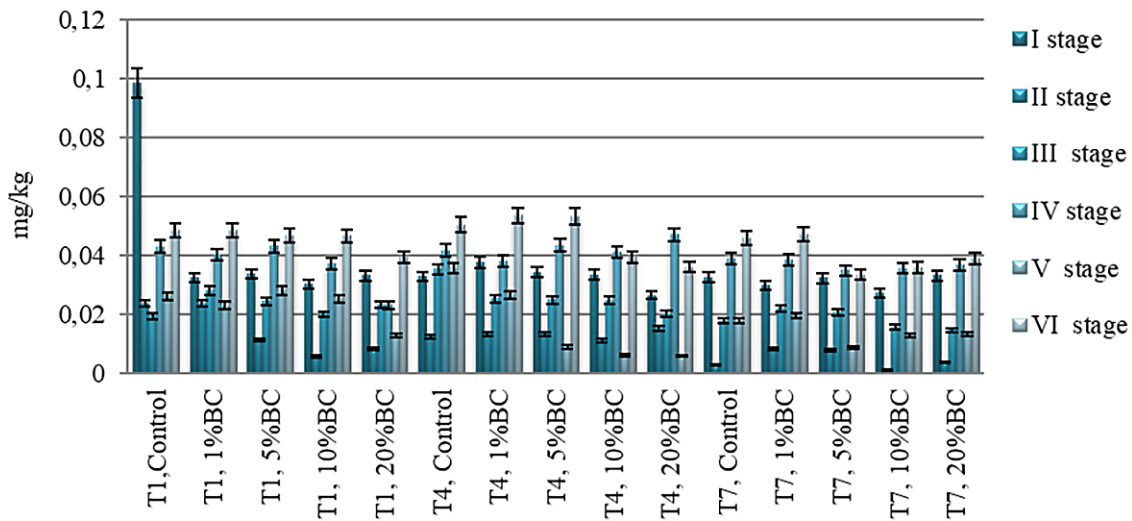


Fig. 6. Lead mobility over time and as a result of increasing BC rate (SE 2-5%), extracted with 0.01M CaCl₂, Source: Authors' own elaboration

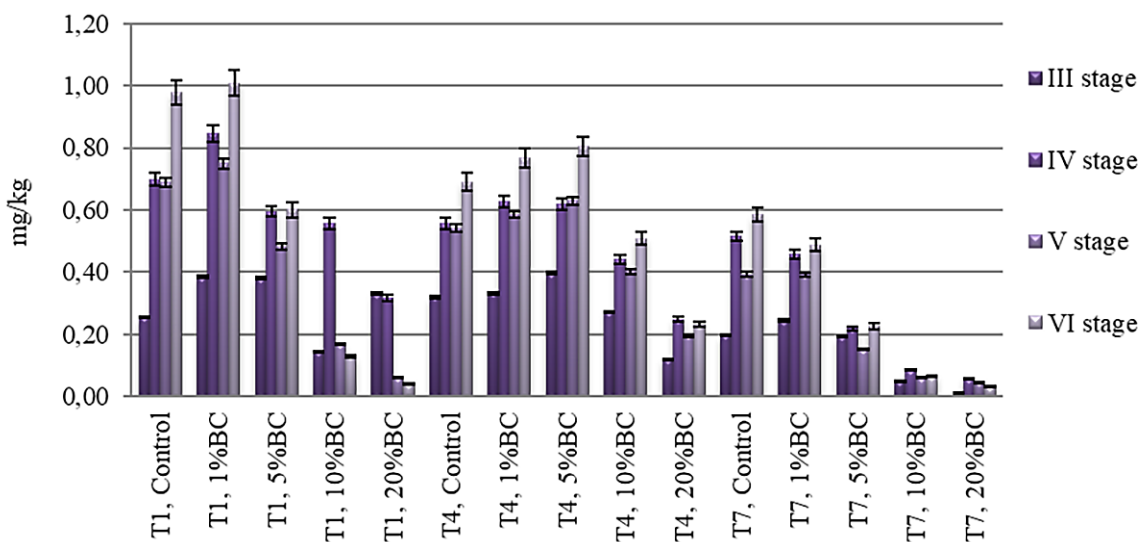


Fig. 7. Cobalt mobility over time and as a result of increasing BC rate (SE 2-5%), extraction with 1M NH₄NO₃, Source: Authors' own elaboration

by experimental variants ($p \leq 0.001$) were established. In all the three studied soils, the application of BC in the high doses of 10 and 20% leads to a significant decrease in the mobility of Pb. In the soil from site T7, the introduction of lower concentrations of the used ameliorant at a dose of 5% BC leads to a significant decrease in the mobility of the studied element. When extracting Pb with 0.01M CaCl_2 , very low values were obtained (in the range of 0.04–0.02 mg.kg^{-1}) and no significant differences were observed between variants (Fig. 6).

Similar behavior is observed for cobalt, concentrations in T7 decrease relative to the percentage increase in BC.

The processing of the data for mobile forms of Co mg.kg^{-1} after adding different doses of BC shows that there are sig-

nificant differences by variants in the experiment (Table 6). The highest Co concentrations are in the extraction with NH_4NO_3 in variant T1 control, an average of 0.66 mg.kg^{-1} , which decrease to 0.19 mg.kg^{-1} when adding 20% BC (Fig. 7). Significant differences in the studied soils are found when adding doses of 10 and 20% BC, and in soil T7, the addition of 5% BC also leads to a significant decrease in mobile forms of Co (Fig. 8).

In the experiment conducted, the trend across variants in the experiment shows that with increasing rates of added BC, mobile forms of Zn decrease only in T7, while in T1 and T4 they increase. Over the incubation time from stage III to VI of the experiment, an increase in the concentrations of mobile forms is observed, which could be explained by the

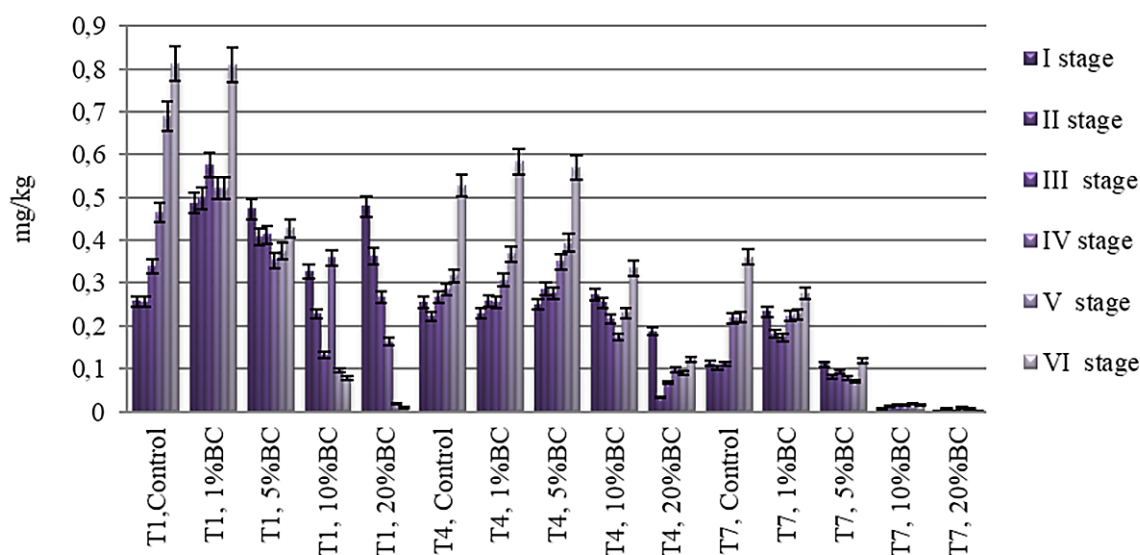


Fig. 8. Cobalt mobility over time and as a result of increasing BC rate (SE 2–5%), extraction with 0.01M CaCl_2
Source: Authors' own elaboration

Table 6. Statistical processing of data using the One-way ANOVA method of mobile forms of Co mg.kg^{-1} , extracted with 1M NH_4NO_3 and 0.01M CaCl_2

Variants	Co, mg.kg^{-1} – T1 Soil		Co, mg.kg^{-1} – T4 Soil		Co, mg.kg^{-1} – T7 Soil	
	1M NH_4NO_3	0,01M CaCl_2	1M NH_4NO_3	0,01M CaCl_2	1M NH_4NO_3	0,01M CaCl_2
Control	0.66±0.3 ^b	0.47±0.23 ^b	0.53±0.15 ^b	0.31±0.11 ^b	0.42±0.17 ^c	0.19±0.10 ^c
1% BC	0.75±0.3 ^b	0.59±0.12 ^b	0.58±0.18 ^b	0.33±0.13 ^b	0.40±0.11 ^c	0.22±0.04 ^c
5% BC	0.51±0.1 ^{ab}	0.41±0.04 ^b	0.61±0.16 ^b	0.36±0.12 ^b	0.20±0.06 ^b	0.09±0.02 ^b
10% BC	0.25±0.2 ^a	0.21±0.12 ^a	0.41±0.10 ^{ab}	0.25±0.05 ^b	0.07±0.01 ^{ab}	0.02±0.01 ^a
20% BC	0.19±0.1 ^a	0.22±0.19 ^a	0.20±0.06 ^a	0.10±0.05 ^a	0.04±0.02 ^a	0.01±0.01 ^a
P-Value	0.0092	0.0011	0.0052	0.0000	0.0000	0.0000
LSD (95%)*	0.33	0.18	0.21	0.12	0.14	0.06

*Confidence level 95%

**Lowercase Latin letters indicate homogeneous groups, while variants that do not share a common letter are significantly different from each other.

Source: Authors' own elaboration

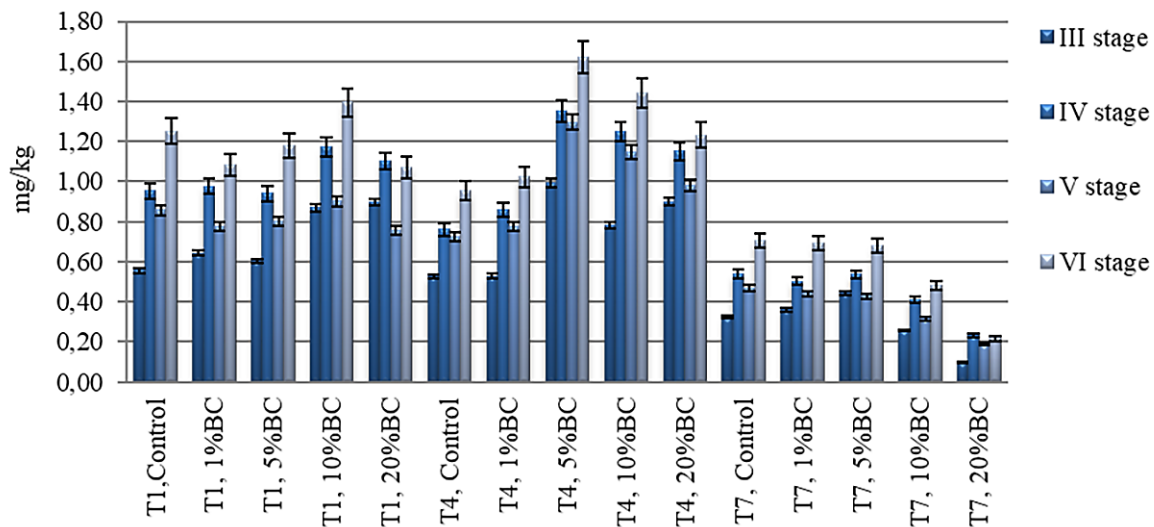


Fig. 9. Zinc mobility over time and as a result of increasing BC rate (SE 2–5%), extraction with 1M NH₄NO₃
Source: Authors' own elaboration

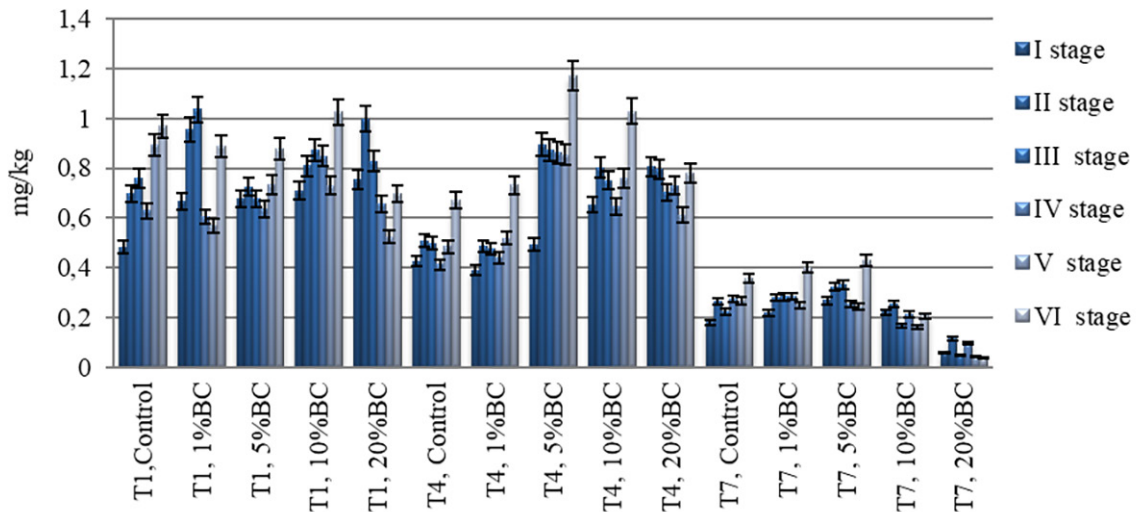


Fig. 10. Zinc mobility over time and as a result of increasing BC rate (SE 2–5%), extraction with 0.01M CaCl₂
Source: Authors' own elaboration

reduction in the effectiveness of biochar (Cui et al., 2024), due to the loss of adsorption sites and/or desorption of organic compounds.

In the soil from site T1, there are no statistically significant differences between the concentrations of mobile forms of Zn after adding BC at different doses (Table 7). In the soil from site T4, there is a tendency for zinc mobility to increase when BC is added at doses of 5, 10 and 20%. Only

in soil T7 does biochar significantly reduce Zn mobility. The values in this soil in the NH₄NO₃ extract range from 0.51 mg kg⁻¹ in the control variant to 0.19 mg kg⁻¹ in the variant with the maximum dose of BC T7-20%, and in the extract with 0,01M CaCl₂, the trend is the same (Fig. 9, 10).

In the soils T1 and T4, no statistically significant differences were observed by variants in terms of mobile forms of Fe in the soil, determined in the two extracts (Table 8). In

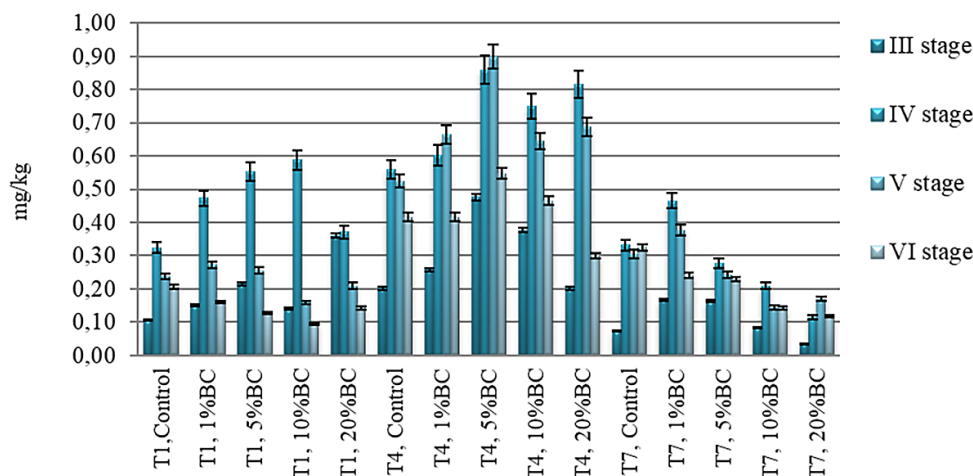
Table 7. Statistical processing of data using the One-way ANOVA method of mobile forms of Zn mg.kg⁻¹, extracted with 1M NH₄NO₃ and 0,01M CaCl₂

Variants	Zn, mg.kg ⁻¹ – T1 Soil		Zn, mg.kg ⁻¹ – T4 Soil		Zn, mg.kg ⁻¹ – T7 Soil	
	1M NH ₄ NO ₃	0,01M CaCl ₂	1M NH ₄ NO ₃	0,01M CaCl ₂	1M NH ₄ NO ₃	0,01M CaCl ₂
Control	0.91±0.28 ^a	0.79±0.20 ^a	0.74±0.17 ^a	0.50±0.09 ^a	0.51±0.16 ^b	0.26±0.06 ^{bc}
1% BC	0.87±0.19 ^a	0.83±0.12 ^a	0.80±0.20 ^a	0.51±0.12 ^a	0.50±0.14 ^b	0.29±0.06 ^c
5% BC	0.88±0.24 ^a	0.74±0.16 ^a	1.32±0.26 ^b	0.86±0.22 ^b	0.52±0.12 ^b	0.31±0.07 ^c
10% BC	1.08±0.24 ^a	0.72±0.09 ^a	1.16±0.28 ^b	0.77±0.14 ^b	0.37±0.10 ^{ab}	0.20±0.04 ^b
20% BC	0.95±0.16 ^a	0.74±0.18 ^a	1.07±0.15 ^{ab}	0.74±0.07 ^b	0.19±0.06 ^a	0.07±0.03 ^a
P-Value	0.9807	0.7177	0.0097	0.0002	0.0054	0.0000
LSD (95%)*	0.35	0.18	0.33	0.16	0.18	0.06

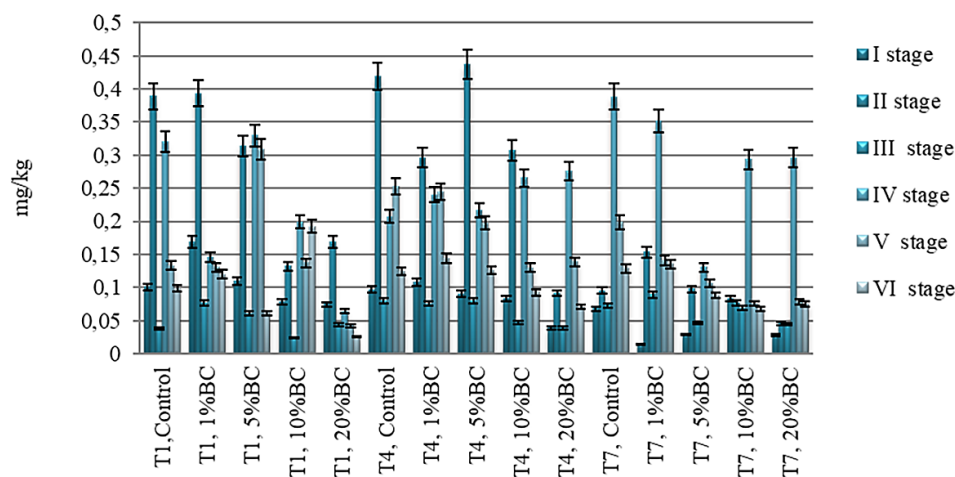
*Confidence level 95%

**Lowercase Latin letters indicate homogeneous groups, while variants that do not share a common letter are significantly different from each other.

Source: Authors' own elaboration

**Fig. 11. Iron mobility over time and as a result of increasing BC rate (SE 2-5%), extraction with 1M NH₄NO₃**

Source: Authors' own elaboration

**Fig. 12. Iron mobility over time and as a result of increasing BC rate (SE 2-5%), extraction with 0.01M CaCl₂**

Source: Authors' own elaboration

Table 8. Statistical processing of data using the One-way ANOVA method of mobile forms of Fe mg kg⁻¹, extracted with 1M NH₄NO₃ and 0,01M CaCl₂

Variants	Fe, mg.kg ⁻¹ – T1 Soil		Fe, mg.kg ⁻¹ – T4 Soil		Fe, mg.kg ⁻¹ – T7 Soil	
	1M NH ₄ NO ₃	0,01M CaCl ₂	1M NH ₄ NO ₃	0,01M CaCl ₂	1M NH ₄ NO ₃	0,01M CaCl ₂
Control	0.22±0.09 ^a	0.18±0.14 ^{ab}	0.43±0.16 ^a	0.20±0.13 ^a	0.26±0.12 ^{bc}	0.16±0.12 ^a
1% BC	0.26±0.15 ^a	0.17±0.11 ^{ab}	0.49±0.18 ^a	0.19±0.09 ^a	0.31±0.13 ^c	0.15±0.11 ^a
5% BC	0.28±0.18 ^a	0.20±0.13 ^b	0.69±0.21 ^a	0.19±0.13 ^a	0.23±0.05 ^{abc}	0.09±0.04 ^a
10% BC	0.25±0.23 ^a	0.13±0.07 ^{ab}	0.56±0.17 ^a	0.16±0.11 ^a	0.14±0.05 ^{ab}	0.11±0.09 ^a
20% BC	0.27±0.11 ^a	0.07±0.05 ^a	0.50±0.29 ^a	0.11±0.09 ^a	0.11±0.05 ^a	0.10±0.10 ^a
P-Value	0.9777	0.2540	0.4632	0.6350	0.0377	0.6416
LSD (95%)*	0.24	0.13	0.32	0.13	0.14	0.13

*Confidence level 95%

**Lowercase Latin letters indicate homogeneous groups, while variants that do not share a common letter are significantly different from each other.

Source: Authors' own elaboration

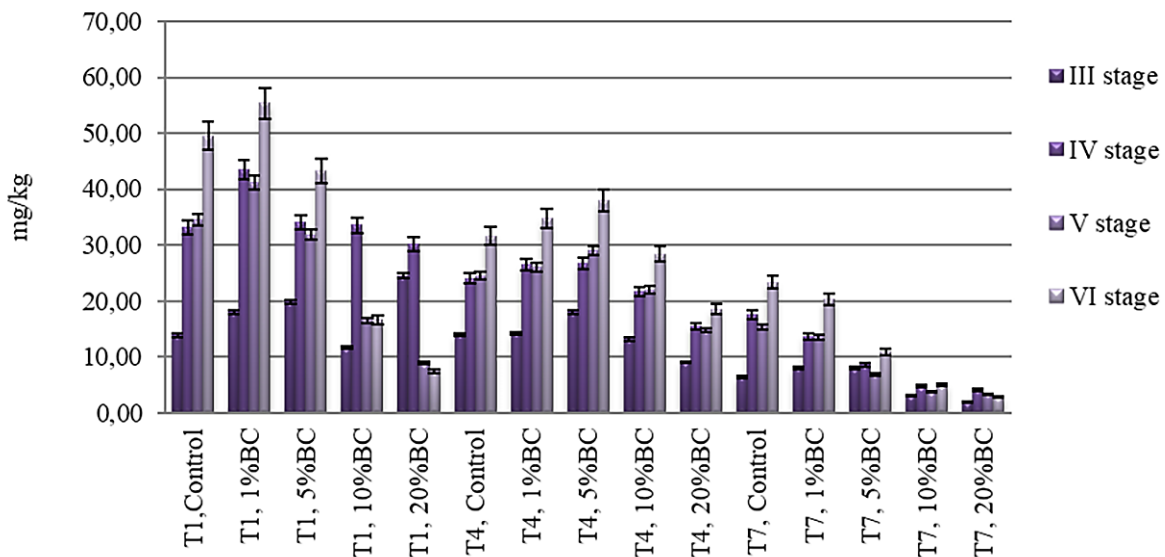
Table 9. Statistical processing of data using the One-way ANOVA method of mobile forms of Mn mg kg⁻¹, extracted with 1M NH₄NO₃ and 0,01M CaCl₂

Variants	Mn, mg.kg ⁻¹ – T1 Soil		Mn, mg.kg ⁻¹ – T4 Soil		Mn, mg.kg ⁻¹ – T7 Soil	
	1M NH ₄ NO ₃	0,01M CaCl ₂	1M NH ₄ NO ₃	0,01M CaCl ₂	1M NH ₄ NO ₃	0,01M CaCl ₂
Control	32.7±14.6 ^{ab}	22.63±12.6 ^{ab}	23.5±7.3 ^a	13.51±5.9 ^{ab}	15.6±7.0 ^c	6.75±5.1 ^c
1% BC	39.5±15.6 ^b	26.54±9.8 ^b	25.4±8.5 ^b	14.10±7.2 ^{ab}	13.8±5.0 ^{bc}	6.89±3.2 ^c
5% BC	32.2± 9.6 ^{ab}	21.90±5.3 ^{ab}	27.9±8.2 ^b	15.86±6.7 ^b	8.5±1.6 ^{ab}	3.98±1.6 ^{bc}
10% BC	19.5± 9.5 ^a	14.24±4.1 ^a	21.3±6.2 ^{ab}	12.57±3.9 ^{ab}	4.1±0.9 ^a	1.22±0.6 ^{ab}
20% BC	17.7±11.3 ^a	14.49±8.3 ^a	14.4±3.9 ^a	8.07±2.8 ^a	3.0±0.9 ^a	0.50±0.4 ^a
P-Value	0.1104	0.0787	0.1279	0.1952	0.0011	0.0007
LSD (95%)*	18.7	10.2	10.6	6.6	5.98	3.3

*Confidence level 95%

**Lowercase Latin letters indicate homogeneous groups, while variants that do not share a common letter are significantly different from each other.

Source: Authors' own elaboration

**Fig. 13. Manganese mobility over time and as a result of increasing BC rate (SE 2–5%), extraction with 1M NH₄NO₃**

Source: Authors' own elaboration

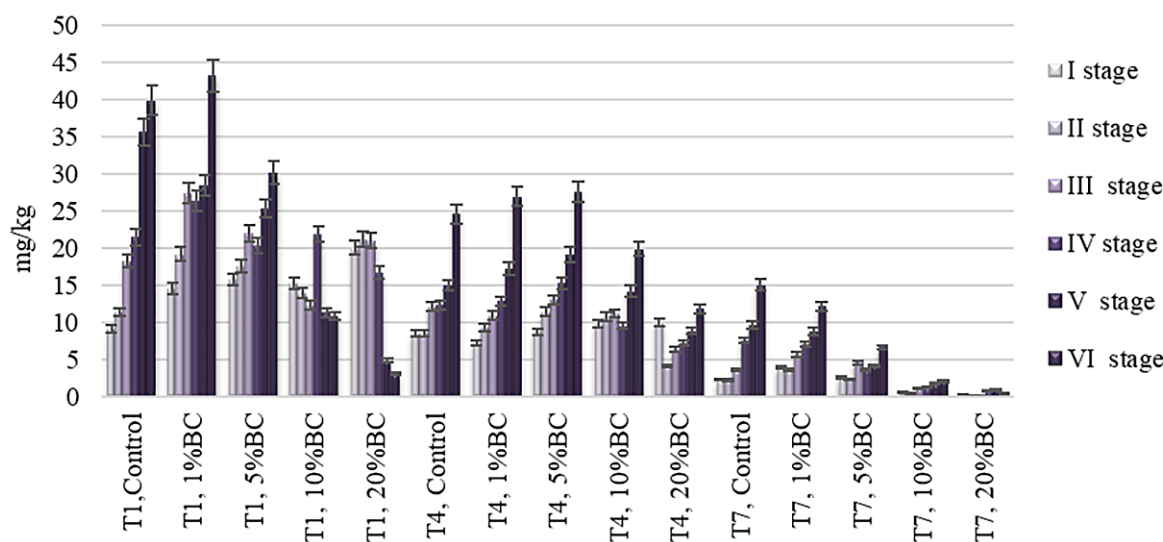


Fig. 14. Manganese mobility over time and as a result of increasing BC rate (SE 2–5%), extraction with 0.01M CaCl₂
 Source: Authors' own elaboration

the soil from site T7, mobile forms of iron determined in 1M NH₄NO₃ were significantly reduced at the highest rate of application of biochar, 20% BC (Fig. 11). The observed trend was also confirmed in the extraction with 0,01M CaCl₂, but the differences by variant were not significant (Fig. 12).

For manganese, there are no significant differences in the mobile forms of the variants with low BC application doses compared to the control, especially in T1 and T4 soils. Only in the soil T7 there is a significant decrease in the mobility of manganese after applying 5% and higher BC rates (Table 9).

In the extract with 1M NH₄NO₃, the concentrations of Mn were statistically affected by the addition of 10 and 20% BC (Fig. 13). The same trend was observed in the extract with 0.01M CaCl₂ (Fig. 14). An increase in Mn concentrations was observed in both extracts over the course of the incubation time.

Conclusions

The results of this study confirm our previous findings that Cu and to some extent Pb in the soils of the area were behaving as an anthropogenic pollutants, which can be immobilized by biochar, while the mobility and bioavailability of Co, Fe, Mn and Zn which might be from heterogeneous sources, e.g., anthropogenic, biopedogenic and were less influenced by the ameliorant added. The immobilization of lead is probably due to its adsorption by soil organic matter.

The effect of biochar on the immobilization of the elements Cu, Pb and Mn is significant in the soil taken from

the tailings pond of the mine (T7) even when introduced in lower doses, 1% and 5%, while in the other two studied soils only higher doses – 10% and 20% have a statistically significant impact.

The results obtained show that in the reclaimed mine soils, BC might be a useful adsorbent of mobile and bioavailable forms of Cu and Pb (bioavailable), however for other metals – nutrients, e.g. Fe, Mn and Zn there is no clear trend of the effect of biochar.

Acknowledgements

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Conflict of interest

This work is not a subject of conflict of interest.

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