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ORIGINAL ARTICLE

Biochar application modifies soil properties of a former mine technosol: SEM/EDS study to investigate Pb and As speciation

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Abstract

Metal (loid) soil pollution is an important issue worldwide that requires the use of remediation, using plants for instance. However, for plants to grow and be efficient, soil fertility must be improved and metal(loid) toxicity reduced. In such an assisted remediation context, biochar has been shown to improve soil conditions and thus plant growth. However, few field studies over long time periods have been conducted to assess biochar impact on metal(loid)s in soil. The objectives of this study were to evaluate the efficiency of a biochar amendment, under field conditions, to improve soil physico-chemical properties and immobilize metal(loid)s and to study the possible mechanisms of such metal(loid) immobilization. For this, biochar was applied to a former mine technosol highly contaminated with As and Pb. Soil samples were taken prior to biochar amendment, on the day of biochar application and 6 months after biochar incorporation. The analyses showed that biochar increased soil pH, organic matter content and nutrient availability, and immobilized Pb. Such results were observed directly after biochar application and were still observed after 6 months, whereas As was first mobilized then immobilized. Finally, the analysis of the samples using scanning electron microscopy coupled with energy dispersive spectroscopy showed that Pb was immobilized, at least partly on the biochar surface, as $PbCO_3$.

Keywords Arsenic · Biochar · Field experiment · Lead · Scanning electron microscopy

1 Introduction

In Europe, soil contamination is a very important issue, with approximately 2.5 million contaminated sites [1]. Among these, 35% are contaminated with elevated concentrations of metal(loid)s, especially As, Cu, Cd, Pb, Cr, Ni, Hg, and Zn, which are frequently encountered on polluted soils [2]. This is of concern because metal(loid)s, unlike some organic compounds, cannot be degraded by soil flora and thus accumulate and persist for a long period of time [2, 3]. The pollution of soils by metal(loid)s is a health issue as most metal(loid)s are toxic and carcinogenic [2].

Different techniques exist and have been used to remediate such contaminated soils. One method is the use of plants to restore contaminated sites and improve the biological activity and the physical structure and fertility of the soil [4]. In addition to improvement of soil conditions, the growth of plants, particularly trees, will permit the production of biomass that can be used for energy production. Therefore, this type of marginal soil can be valorized in terms of energy production, together with its remediation. However, such a process relies on the establishment and good growth of plant cover. In addition, the industrial and mining activities can reduce soil fertility and the presence of metal(loid)s can be disruptive for soil (micro)organisms, altering nutrient cycling. For this, the use of amendments is recommended and necessary to minimize the effect of contamination on plants, thus enhancing plant growth and biomass production [5–7].

One soil amendment gathering attention over the last decade is biochar. The International Biochar Initiative defines biochar as "a solid product obtained by the thermochemical conversion of biomass under anoxic conditions" [8]. Biochar

Highlights
- A field experiment was conducted on a former mine technosol.
- Biochar was applied and its effect evaluated after 6 months.
- Biochar ameliorated soil fertility and immobilized metal(loid)s.
- Pb mobilization was partly due to its sorption on the biochar surface.

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is characterized by an alkaline pH, a high specific surface area, a high stability and high amounts of oxygen-containing functional groups on its surface [9]. As a soil amendment, biochar can sequester carbon to reduce greenhouse gas emissions, form complexes with metal(loid)s and enhance soil productivity [5, 8]. Moreover, the production of biochar through pyrolysis brings economic benefits. Firstly, biochar can be produced using a large range of biomasses, including residues, which is a way of valorizing materials that are normally thrown away [10, 11]. Secondly, in addition to biochar, the pyrolysis process produces liquids and gases that can be used for different applications, including energy and heat production, as an alternative to fossil fuels [12, 13].

Previous studies demonstrated the potential of biochar to ameliorate soil physico-chemical properties, immobilize metal(loid)s and thus improve plant growth [14–16]. However, the mechanisms of metal(loid) immobilization require additional study as most research on the topic has been conducted under laboratory conditions in short term experiments.

Therefore, the first objective of the present study was to evaluate, under field conditions, the effect of a biochar amendment on contaminated soil physico-chemical properties and metal(loid) mobility and availability after a period of 6 months post-application. The second objective was to assess the potential mechanisms of metal(loid) immobilization via scanning electron microscopy (SEM) analysis of the unamended soil and the biochar before and after the amendment.

2 Materials and methods

2.1 Studied soil

This study was conducted on a former silver-lead extraction mine site of the Pontgibaud district (Auvergne-Rhone-Alpes, France), one of the largest mining and metallurgical districts in Europe during the nineteenth century. This important extraction activity has resulted in huge amounts of mine tailings deposits of silver-bearing ore to the soil, still containing toxic elements, mainly arsenic (As) and lead (Pb), at average concentrations of 539 mg kg^{-1} and 11,453 mg kg^{-1}, respectively [16–18]. The soil at the site is also characterized by an acidic pH, a sandy texture and a low organic matter content [15].

The contaminated area covers four hectares. This study was performed in field, at the second settling pond (Lambert II coordinates: *X* 638147.54 and *Y* 2087962.79), located 700 m above sea level and subject to snowy winters and hot summers with an average annual rainfall of 770 mm [19].

2.2 Biochar

The biochar used was provided by La Carbonerie (Crissey, France). It was created through the slow pyrolysis of hardwood biomass (oak, hornbeam, beech) at 500 °C with a residence time of 3 h and a heating rate of 2.5 °C per min. Following pyrolysis, the biochar was sieved to obtain a particle size between 0.5 and 1 mm.

Biochar was incorporated into the soil at a rate corresponding to 2% (*w*/w) on a 9.6 × 4.5 m area to a maximum depth of 25 cm, using a tiller.

2.3 Soil sampling

Soil samples were taken before biochar application (unamended soil), within 24 h after biochar application and 6 months after biochar application, with these final samples collected in October 2017. Sampling was done using a 2 × 2 m grid, within the top 25 cm of soil depth. On the entire plot, ten random sampling points were selected and a final soil mass of 1 kg was sampled and air-dried for analysis.

2.4 Analysis of the soil, biochar, and soil-biochar mixture physico-chemical properties

The unamended soil, and both the biochar and soil 6 months after amendment were analyzed for pH, organic matter and nitrogen content, cation exchange capacity (CEC), and metal(loid) total concentration and repartition.

pH was measured after mixing and shaking the samples in distilled water (1:10 solid:liquid ratio), using a pH meter (Metler-Toledo, Seveur Excellence). The organic carbon and total nitrogen contents were determined using a Thermo Flash 2000, after crushing the samples to 125 µm particle size. Organic matter content was calculated by multiplying the organic carbon content by the Wolf factor (1.724). The CEC was determined based on standard method NF X31–130, using cobaltihexamine chloride.

Samples were subjected to *aqua regia* digestion, as well as NH_4NO_3 digestion, as described in Nandillon et al. [16], followed by ICP-AES (Inductively Coupled Plasma Atomic Emission Spectroscopy) (ULTIMA, HORIBA, Labcompare, San Francisco, USA) analysis to determine the pseudo-total concentrations, and the phytoavailable concentrations of the pollutants (As and Pb), respectively. Moreover, to examine the distribution of As and Pb in the different soil fractions, sequential extractions were performed using the BCR (Community Bureau of Reference) protocol, as described in Pascaud et al. [20].

Finally, X-ray diffraction analysis was conducted, only on the unamended soil sample, using an INEL CPS120 coupled to a Co anode (Co Kα = 1.78897 Å), with the following parameters: angle 0–90 °C (2θ) and frequency 0.3° 20θ cm^{-1}.

Table 1 Physico-chemical characteristics (organic carbon content (C_{org}) (%), CEC (cmol kg^{-1}), organic matter content (OM) (%), pH and NH_4NO_3 extractable P, K, As, Pb, and Fe concentrations (mg kg^{-1})) of the Pontgibaud technosol and the biochar amendment. *ND*, not determined

	Pontgibaud soil	Biochar amendment
C_{org} (%)	0.27 ± 0.01	77.4±1.6
CEC (cmol kg^{-1})	0.7 ± 0.0	0.5±0.0
OM (%)	0.47 ± 0.04	ND
pH	4.8 ± 0.0	9.0±0.0
NH_4NO_3 extractable concentrations (mg kg^{-1})		
[P]	1.1 ± 0.1	8.4±0.7
[K]	15.8 ± 0.2	752±28
[As]	1.93 ± 0.15	0.9±0.1
[Pb]	3586 ± 164	1.6±0.1
[Fe]	12 ± 6	18±5

2.5 Microscopy

The initial, unamended technosol (PG) was subjected to a scanning electron microscopy (SEM) study coupled with energy dispersive spectroscopy (EDS). Subsequently, the biochar before its incorporation as well as following retrieval from the soil after 6 months were analyzed using SEM and EDS (Jeol FEG-SEM type SU70 with an EDAX EDS analysis system).

2.6 Statistical analysis

Data were analyzed using R software version 3.6.1 (R [21]). Shapiro and Bartlett tests were used to assess normality and homoscedasticity of the data, respectively. The means were subsequently compared using ANOVA or Kruskal tests, for parametric and non-parametric data, respectively, followed by a post hoc test. Differences were considered significant at $p < 0.05$.

3 Results

3.1 Initial soil and biochar characteristics

The unamended Pontgibaud technosol (PG) was characterized by low levels of organic carbon, organic matter, a low CEC and an acidic pH (4.8) (Table 1). Available P and K concentrations were also low (Table 1), below the amounts required for plants (150–250 mg kg^{-1} K and 10–20 mg kg^{-1} P). This demonstrates the poor fertility of the soil. In addition, the concentrations of available As and Pb were high, 1.93 and 3586 mg kg^{-1}, respectively (Table 1), confirming the contamination of PG by As and Pb.

Based on X-ray diffraction, the mineralogical background of the soil was mainly quartz, muscovite and albite, the last two being phyllosilicates (Fig. 1). The presence of anglesite and beudantite was also detected.

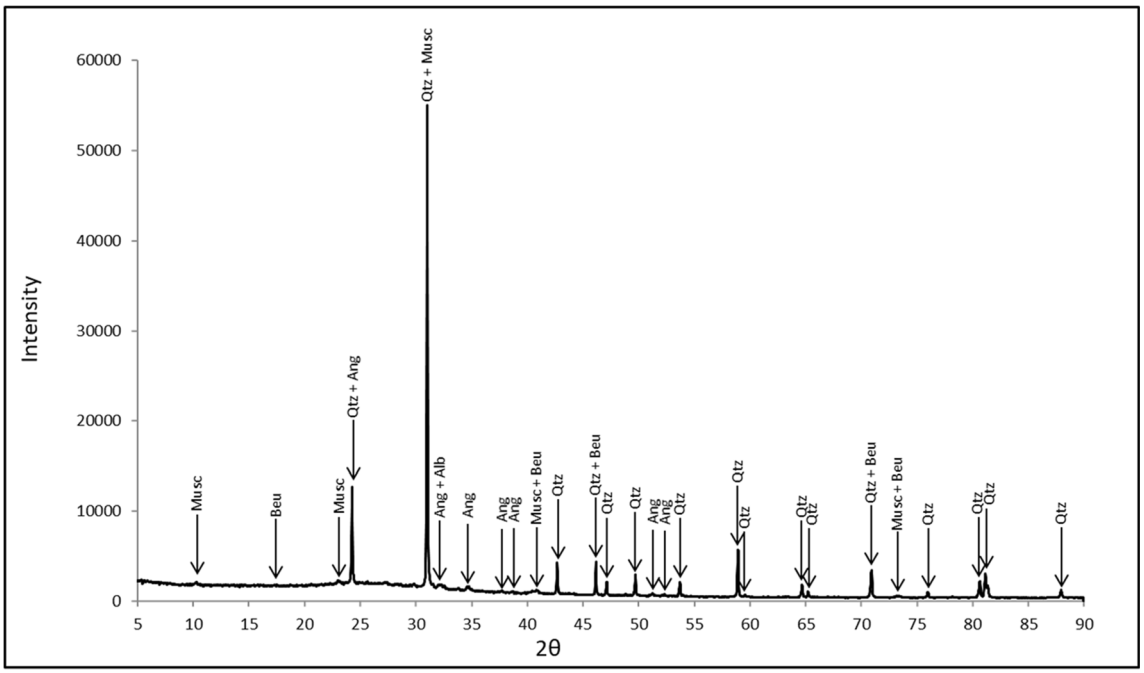

Fig. 1 X-ray diffraction analysis of the Pontgibaud technosol. Musc, muscovite; Alb, albite; Qtz, quartz; Fds, feldspar; Beu, beudantite; Ang, anglesite

Table 2 Soil physico-chemical characteristics (pH, CEC (cmol kg^{-1}), C_{org} content (%), P and K phytoavailable concentrations (mg kg^{-1})) of Pontgibaud technosol before biochar application (PG), immediately after biochar application (PB T0) and 6 months after biochar application (PB T6). Letters indicate significant differences between treatments ($p < 0.05$) ($n = 3 \pm SE$)

Treatments	pH	CEC (cmol kg^{-1})	C_{org} %*	MO %*	[P] (mg kg^{-1})	[K] (mg kg^{-1})
PG	4.8±0.0 a	0.7±0.0 a	0.3	0.5	1.1±0.1 a	15.8±0.2 a
PB T0	5.7±0.1 b	0.6±0.0 a	3.7	6.4	3.9±0.4 b	42.5±2.4 b
PB T6	5.7±0.2 b	0.9±0.0 b	2.8	4.8	1.7±0.1 a	41.4±0.3 b

*Results of one measurement

The biochar had a high organic carbon level (77.4%) and an alkaline pH (pH 9) (Table 1). The biochar CEC was low, as were the available As and Pb concentrations, 0.9 mg kg^{-1} and 1.6 mg kg^{-1}, respectively (Table 1). Finally, the biochar concentrations of available P and K were high, 8.4 mg kg^{-1} and 752 mg kg^{-1}, respectively.

3.2 Effect of biochar on the soil physico-chemical properties

The pH of PG was acidic (pH 4.8) and it increased by 0.9 units after biochar was added to the soil (PBT0) (Table 2). This increase was observed immediately after biochar application and remained stable over time (i.e., no significant difference in pH after 6 months compared to PBT0).

The soil CEC had increased by 30% 6 months after biochar was added (Table 2).

Organic carbon content was very low in PG, at 0.3%. Adding biochar to the soil immediately raised the organic carbon content more than tenfold; then, a slight decrease (24%) was observed after 6 months (Table 2). Organic matter content followed the same trend (Table 2).

The phytoavailable P concentration was low in PG (1.1 mg kg^{-1}) and increased 3.5-times immediately after biochar addition (Table 2). However, the concentration decreased after 6 months, to the same level as the initial unamended PG (Table 2).

Finally, phytoavailable K content was almost tripled immediately after biochar addition and remained constant after 6 months (Table 2).

3.3 Effect of biochar on the metal(loid) concentrations in different soil fractions

Both pseudo-total and phytoavailable metal(loid) concentrations (NH_4NO_3 extraction) were determined.

Pseudo-total As and Pb concentrations were high and not affected by biochar amendment, even after 6 months (Table 3).

The NH_4NO_3-extractable (phytoavailable) As concentration was 1.93 mg kg^{-1} in PG and initially increased following biochar amendment from 1.93 to 2.56 mg kg^{-1} but then decreased to 1.15 mg kg^{-1} after 6 months, lower than the unamended PG. The phytoavailable Fe concentration was not affected by biochar addition. The phytoavailable Pb concentration was high in the unamended PG (3586 mg kg^{-1}) and decreased after 6 months of biochar amendment to 1263 mg kg^{-1} (Table 3).

In addition, the repartition of As and Pb in the different soil fractions was evaluated. Arsenic was mainly accumulated in the residual fraction, with more than 90% of As in the F4 fraction of PG (Fig. 2). The exchangeable fraction (F1) and reducible fraction (F2) were very low and increased following biochar amendment. The increase was more pronounced after 6 months (Fig. 2A). The oxidizable fraction (F3) was higher, but still low (around 5%). Arsenic content in F3 increased following biochar amendment, but this increase was reduced after 6 months (Fig. 2A). Finally, together with the increase of As content in F1, F2, and F3, the As content in F4 decreased following biochar addition (Fig. 2A).

Table 3 Soil pseudo-total and NH_4NO_3 extractable As, Fe, and Pb concentrations (mg kg^{-1}) of Pontgibaud technosol before biochar application (PG), immediately after biochar application (PB T0) and 6 months after biochar application (PB T6). Letters indicate significant differences between treatments ($p < 0.05$) ($n = 3 \pm SE$)

Treatments	Pseudo-total concentrations (mg kg^{-1})		NH_4NO_3-extractable concentrations (mg kg^{-1})		
	[As]	[Pb]	[As]	[Fe]	[Pb]
PG	752±66 ab	8098±859 a	1.93±0.15 b	11.9±5.7 a	3586±164 b
PB T0	752±52 b	9102±627 a	2.56±0.19 c	1.0±0.6 a	4023±433 b
PB T6	591±26 a	7419±465 a	1.15±0.06 a	4.0±0.9 a	1263±279 a

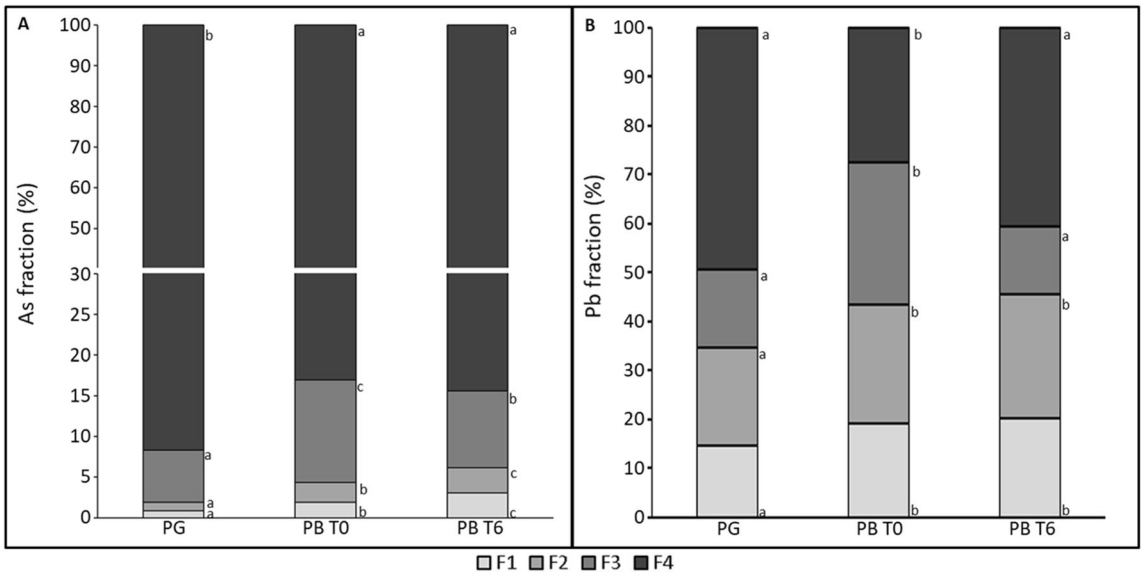

Fig. 2 As (A) and Pb (B) concentration distribution (%) according to the selected sequential extraction protocol (BCR) of the Pontgibaud technosol before biochar amendment (PG), immediately after the biochar application (PB T0) and 6 months after the biochar application (PB T6). Letters indicate significant differences between treatments ($p < 0.05$) ($n = 3 \pm SE$). F1, exchangeable fraction; F2, reducible fraction; F3, oxidizable fraction; F4, residual fraction

Half of the Pb in PG soil was located in the residual fraction (F4). The exchangeable fraction (F1) of Pb in PG was 15% of the total content and increased following biochar amendment. Pb in F2 (reducible fraction) was 20% and also increased after the addition of biochar. The percentage of Pb in F3 (oxidizable fraction) was 16% and increased with biochar application, but only at application time (T0). Only at application time (T0) did the residual Pb fraction decrease compared to PG or PBT6 (Fig. 2B).

Table 4 Elemental composition (% wt) of the unamended Pontgibaud technosol. S1 to S5 correspond to the different areas analyzed on Pontgibaud soil particles. The locations of the analysis sites are shown in Fig. 3A

Element	S1	S2	S3	S4	S5
C	nd	10.5	18.3	16.1	nd
O	50.4	25.1	44.4	31.6	49.1
Mg	0.7	0.3	0.5	nd	0.8
Al	12.5	3.5	4.7	3.3	9.7
Si	18.5	5.9	26.7	14.3	22.7
S	1	6.8	nd	3.7	1.1
K	5.9	nd	nd	nd	5
Fe	5.6	3.5	5.3	10.7	7.1
Zn	nd	nd	nd	nd	nd
As	nd	nd	nd	2.2	nd
Pb	5.3	44.5	nd	18.3	4.6
Total	100	100	100	100	100

nd, not detected

3.4 Microscopy analysis

3.4.1 The unamended Pontgibaud soil

The unamended PG soil was studied by SEM coupled with EDS in order to determine the speciation of the Pb and As present. The squares S1 to S12 visible in Fig. 3 A and B denote the specific locations of the EDS analysis; results are shown in Tables 4 and 5, respectively. The soil largely represents the volcanic rock it originates from, as well as the discarded mine tailings. Figure 3A shows mineral particles, whether or not surmounted by white areas, which were analyzed by EDS. The areas labeled S1, S3, S4, and S5 correspond to particles visible in black by microscopy (Fig. 3). These particles were composed of approximately 50% oxygen, Al between 3.3 and 12.5% and Si between 14.3 and 26.7% (Table 4). Thus, it can be assumed that these structures are Al-silicate particles. Regarding the white area S2 (Fig. 3A), it contains less oxygen, aluminum and silicon than the Al-silicate particles located in the black zone, but more sulfur and also a high level of Pb (44.5%) (Table 4) It can thus be assumed that this white zone constitutes Pb deposits, which are found as $PbCO_3$ (cerussite) and $PbSO_4$ (anglesite).

Figure 3B is a magnification of the surface of a soil particle. Similarly, it can be seen that most of the surface is visible in black in microscopy and some white spots can be observed. Seven zones (S6 to S12) were analyzed for their composition. Results are shown in Table 5. The square S6 corresponds to a region shown in black in the SEM picture, containing 46.16% oxygen, 14.09% aluminum, 18.73% silicon, 8.34% potassium and 3.42% iron, but no lead was detected. Thus, it can be supposed that this

Table 5 Elemental composition (% wt) of the unamended Pontgibaud technosol. S6 to S12 correspond to the different areas analyzed on one soil particle. The locations of the analysis sites are shown in Fig. 3B

Element	S6	S7	S8	S9	S10	S11	S12
C	8.6	7.39	8.8	5.35	8.27	6.75	5.82
O	46.16	34.1	37.64	34.09	41.18	35.39	34.26
Mg	0.67	nd	nd	nd	0.36	nd	0.38
Al	14.09	3	5.55	3.43	9.96	3.6	6.48
Si	18.73	3.88	7.82	4.95	12.09	4.8	8.59
S	nd	5.7	4.45	4.84	2.23	5.49	5.49
K	8.34	nd	nd	2.7	nd	nd	3.85
Fe	3.42	25.17	20.48	21.66	12.37	21.67	2.43
Zn	nd	1.53	nd	nd	0.49	nd	nd
As	nd	1.97	1.98	2.8	1.68	2.98	nd
Zr	0	0	0	3.11	0	2.66	0
Pb	nd	17.25	13.28	17.08	11.37	16.66	32.71
Total	100	100	100	100	100	100	100

nd, not detected

zone is constituted of K, Fe, and Al-silicate. Moreover, associating this elemental composition with the geological information of the region, it can be assumed that the sheet mineral is most likely biotite ($K(Mg,Fe)_3(AlSi3O10)(OH,F)_2$). This sheet mineral hosts a large number of smaller crystals, most of them smaller than 1 µm. The squares S7 to S12 show the location of the analyses of the small particles. The zones S7, S8, S9, S10 and S11 had similar elemental composition: high oxygen contents, between 34.09 and 37.64% and high iron contents between 12.37 and 25.17% whereas sulfur content was around 5%, arsenic between 1.68 and 2.98% and lead between 11.37 and 17.25% (Table 5). Such elemental composition could correspond to beudantite materials ($PbFe_3(AsO_4)SO_4(OH)_6$. Finally, the zone S12 contained similar oxygen content compared to the previous zones, but less iron (2.43%) and more lead (32.71%); it also contained 6.48% Al, 8.59% Si, and 3.85% K. Thus, this zone is supposedly a Pb-mineral with some background of the underlying biotite crystal.

3.4.2 Biochar

Figure 4 shows SEM images of the wood biochar used, before it was applied to the soil. Figure 4A is an SEM image of a biochar sample showing several biochar structures, whereas Fig. 4 B and C are magnifications of specific structures found in the biochar sample. These three images showed the porous structure of the biochar, consisting of very large channel structures with smaller channels around them. Figure 4B shows a magnification of the channel structure, in which are observed different tissues of the parental wood: xylem vessels, noted Xv, surrounded by xylem fibers, noted Xf, bordered by the ray parenchyma, noted Rp.

An elemental analysis was made on the parenchyma structure (square B1). The EDS analysis showed that it was made of more than 90% carbon and smaller amounts of oxygen (5.02%), potassium (1.62%) and calcium (2.05%) (Table 6). Moreover, on the biochar, surface specific structures, presenting as white spots on SEM, were noted. They are shown in Fig. 4C. An analysis revealed that, compared to the other biochar structure shown in Fig. 4B, these white spots contained less carbon (24.24%) but more oxygen (41.55%) and calcium (34.21%) and no potassium. From this, it can be suggested that these specific structures are $CaCO_3$ inclusions on the biochar surface.

Figure 5 shows a piece of the surface of a biochar grain recovered from the field after 6 months of aging in the Pontgibaud soil. On this SEM image, large white zones are visible on the porous black background. The zone B6, located on a particle shown in black under microscopy, is mainly

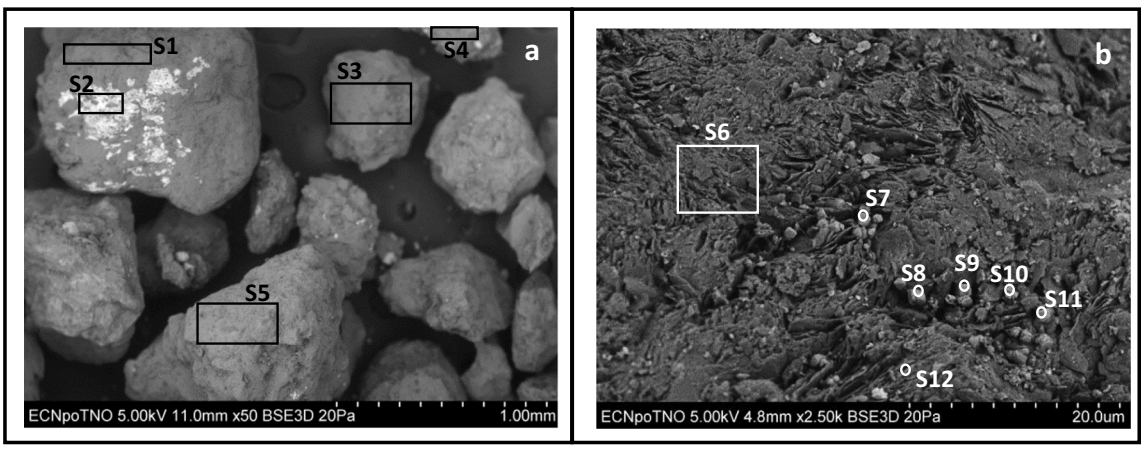

Fig. 3 Scanning electron microscopy images of the non-amended Pontgibaud technosol. (A) typical soil particles; (B) magnification of the surface of one soil particle; squares indicate the location of the analysis of element contents; the results are presented in Tables 4 and 5

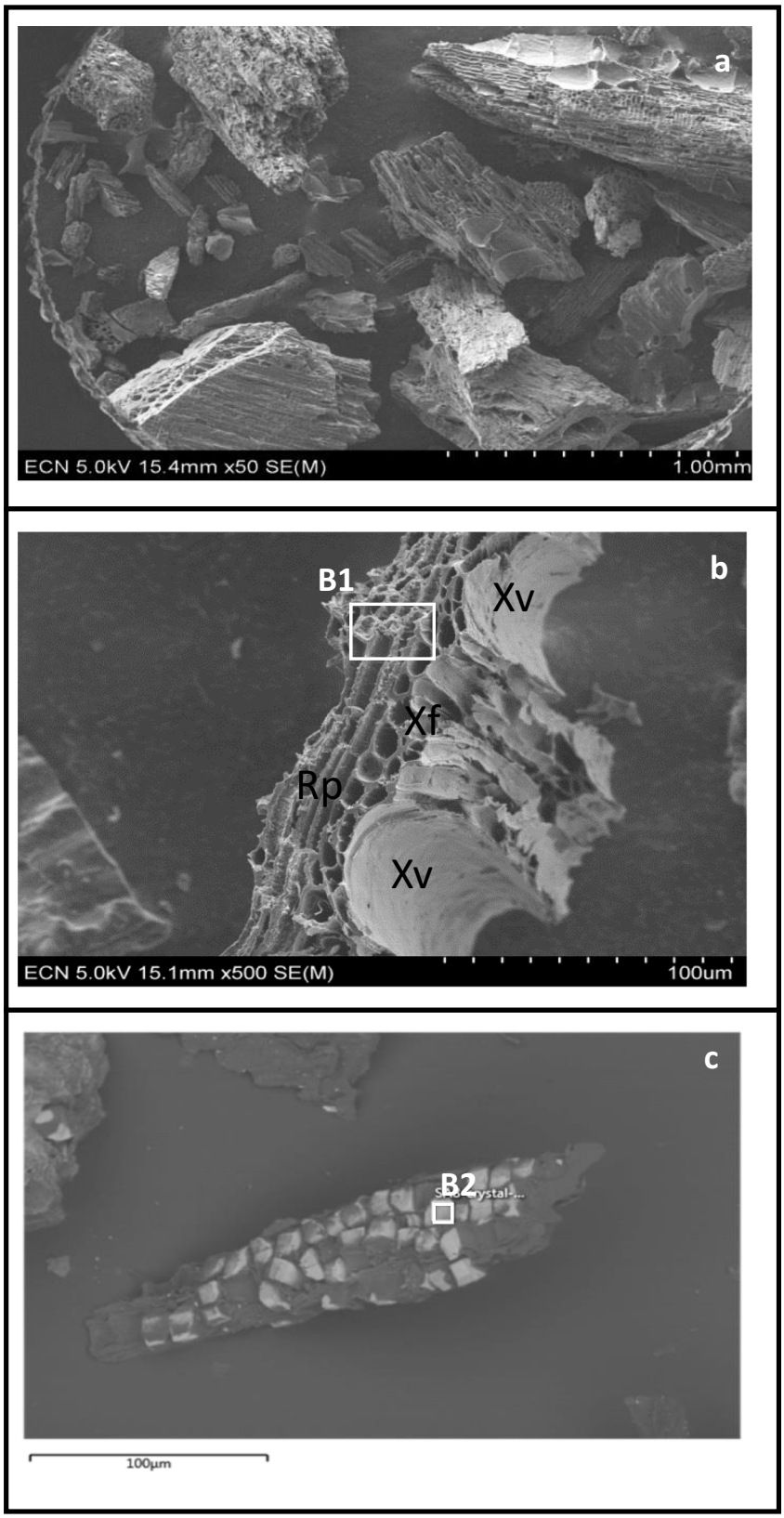

Fig. 4 Scanning electron microscopy images of the biochar before it was applied to Pontgibaud technosol. (A) corresponds to the biochar particles; (B) shows the magnification of the vessel structures, upon which typical tissue residues from raw biomass (Xv: Xylem vessel, Xf: Xylem fiber, Rp: Ray parenchyma) are visible; (C) shows a biochar particle containing calcium incrustations. Squares B1 and B2 indicate the location of the analysis of element contents

made of carbon (84.7%) with very small amounts of Al (1.1%), Si (1.9%), and Pb (1.3%) (Table 7), which is similar to the composition found in the analysis of the biochar before its incorporation to the soil. The zone B3 contained 25.1%

Table 6 Elemental composition (% wt) of the biochar before its application to the soil. B1 and B2 correspond to the different areas analyzed on the biochar surface. The locations of the analysis areas B1 and B2 are shown in Fig. 4 B and C

Element	B1	B2
C	91.31	24.24
O	5.02	41.55
Mg	nd	nd
Si	nd	nd
Si	nd	nd
K	1.62	nd
Ca	2.05	34.21
Total	100	100

nd, not detected

carbon, 31.9% oxygen and small amounts of Al (8.7%), Si (14.8%), K (3.2%), Fe (7.2%), Pb (6.9%), and very small amounts of sulfur (0.9%). It can be supposed that in this area, some of the soil material is fixed, i.e., Al, Fe and K silicates and Pb as $PbCO_3$ and $PbSO_4$. The zones B7 and B8 showed light gray particles composed of 20.2% to 38.9% oxygen, 2.3% to 10.3% Al, 4.3% to 31% Si, 0.9% to 3.8% K, 3.4% to 4.4% Fe, and 6.7% to 58.3% Pb. Such composition reflects the presence of Al-silicate with K and Fe, which are most likely the remnants of the eroded muscovite, biotite or other mica, and the fixation of Pb. Finally, the zones B4 and B5 correspond to large white areas composed mainly of Pb (70.4% to 78.2%), O (14.4% to 18.1%), and C (5.4% to 6.9%) (Table 7), demonstrating that Pb was fixed on the biochar as $PbCO_3$. However, in all the zones analyzed, arsenic was present only in very small amounts, showing that As likely did not fix to the biochar surface.

Table 7 Elemental composition (% wt) of the biochar 6 months after its incorporation into the soil. B3 to B8 correspond to the different areas analyzed on the biochar surface. The locations of the analysis sites are shown in Fig. 5

Element	B3	B4	B5	B6	B7	B8
C	25.1	6.9	5.4	84.7	2.3	3.7
N	nd	nd	nd	0	nd	nd
O	31.9	18.1	14.4	9	20.2	38.9
Na	0.1	nd	nd	nd	0	nd
Mg	0.6	0	0	0.2	0.2	0.6
Al	8.7	1.3	0.5	1.1	2.3	10.3
Si	14.8	2.1	0.7	1.9	4.3	31
Si	0.9	0	1	0.2	7.9	0.5
K	3.2	nd	nd	0.7	0.9	3.8
Ca	nd	nd	nd	0	nd	nd
Fe	7.2	1.2	0	1	3.4	4.4
Zn	nd	nd	nd	nd	nd	nd
As	0.6	0	0	0	0.3	0.2
Pb	6.9	70.4	78.2	1.3	58.3	6.7
Total	100	100	100	100	100	100

nd, not detected

4 Discussion

4.1 The effect of biochar on soil physico-chemical properties

Soil pH is a very important parameter, as it influences many processes in the soil and affects metal(loid) behavior [22]. Here, soil pH was acidic for unamended PG, which could lead to high solubility and availability of metal cations. Soil pH increased with biochar, thus potentially reducing metal mobility. Such pH increase has been observed in previous studies [14, 22, 23] and can be explained by the following: (i) the alkaline nature of the biochar added (pH 9), which is related to its ash content [23–25]; (ii) the dissolution of mineral elements such as oxides and carbonates [22]; and (iii) the presence of negative functional groups on the biochar surface that can bind H^+ ions [24].

Biochar was observed to induce an increase in organic matter in the soil. Organic carbon and organic matter are important in soil. They help in the retention of water and nutrients and provide substrates for microorganisms [26]. Moreover, organic matter has a high capacity to retain metal(loid)s [27]. Therefore, the increase in organic carbon and organic matter after biochar addition may have increased the soil capacity to retain metal(loid)s. As stated in the review of Janus et al. [11], the organic matter in biochar-amended soil is three times higher on average than unamended soil. Two mechanisms can explain the observed increase: (i) biochar contains high levels of carbon and organic fractions that are directly added to the soil [15, 28, 29]; and (ii) biochar addition can affect the microbial community, leading to changes in element cycles [15].

Regarding the nutrients available in the soil, phosphorus and potassium are necessary nutrients for plants. Their increase has been previously observed and explained by the reduction in runoff and leaching following biochar amendment, due to the porous nature of biochar [30]. In addition, the increase in soil pH following biochar amendment could have influenced the availability of P and K present in the soil, as soil pH influences nutrient behavior [16, 30]. Finally, nutrient increase can be explained by an increase in soil CEC and the addition of nutrients contained in biochar [24, 28, 31]. The reduction in available P after 6 months can be attributed to its leaching from the soil solution to groundwater.

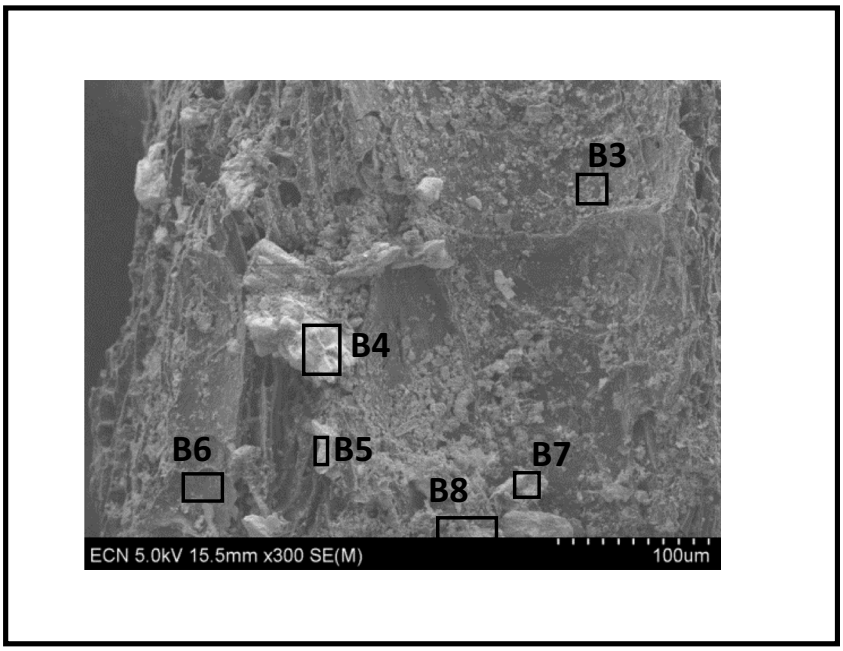

Fig. 5 Scanning electron microscopy image of a biochar particle 6 months after it was added to the soil. Squares B3 to B8 indicate the location of the analysis of element contents; results are shown in Table 7

4.2 The effect of biochar on the metal(loid) concentrations in the different fractions of the soil

The available concentration of arsenic increased following biochar amendment, which can be linked to the increase in soil pH that reduced the positive charges of soil colloids and thus the adsorption capacity of As [22]. However, after 6 months, As availability decreased, which was consistent with the study of Lebrun et al. [15] and could be attributed to its adsorption and complexation onto the biochar surface [15].

Similarly, Pb availability was highly reduced by biochar amendment. Different mechanisms can explain this observation. The increase in soil pH can lead to Pb immobilization [22]; Pb can adsorb and precipitate on biochar and soil surfaces [22] and Pb can complex with the functional groups of biochar [14, 22]. The increase in soil organic matter and soil organic carbon can increase soil microbial activity and the uptake of Pb within microorganisms [22]. Finally, as stated before, the increase in CEC and organic matter content can increase the capacity of soil to retain metal(loid)s [27, 32].

In the sequential extractions, the F1 fraction is considered the most easily soluble metal fraction, F2 corresponds to the metal(loid)s bound to poorly crystalline Fe/Mn oxyhydroxides and sulfates, F3 is the most oxidizable fraction, bound to organic matter and sulfides, and F4 is the most stable fraction, in which metal(loid)s are linked to the crystal structure of the soil [20].

Similarly to what was observed here, Pascaud et al. [20] observed low levels of As in F1 (0.6%), F2 (0.2%), and F3 (1% to 4%) and nearly 100% in F4, which is considered to be long-term stable with a low risk of contamination release into the environment. A similar pattern was also found for Pb, with 1% to 13% in F1, 3% to 25% in F2, 20% in F3, and 40% to 70% in F4 [20]. For both elements, when their contents in F1, F2 and F3 decreased, it was associated with an increase in F4. Rizwan et al. [25] observed that the immobilization of the metals was mainly transformed into the reducible fraction, and that the oxidizable and residual fractions of Pb increased following that. Moreover, as soil pH increases, Pb hydrolysis increases and thus more Pb is precipitated as $Pb_5(PO_4)_3Cl$ [25]. Such increase in soil pH also decreases the activities of free metals and thus reduces the acid-soluble fraction, related to a sorption of Pb onto the Fe-Mn oxides and chelation by the organic matter [25, 33], increasing F2 and F3 fractions. Moreover, the increase in the fraction related to organic matter can be attributed to the increase in organic matter content following biochar amendment [34].

4.3 Microscopy analysis

A good indication of the elemental localization and speciation, before and after the biochar amendment period, can be obtained by a scanning electron microscopy (SEM) study in combination with energy dispersive spectroscopy (EDS).

The SEM image of the unamended technosol shows the ore minerals are predominately combinations of Pb-sulfate (anglesite) and Pb-Carbonate (cerussite) deposited on Al-silicate mineral particles (Feldspars mostly). This demonstrates that sulfate and carbonate play an important role in Pb immobilization [35]. It is unclear how well-attached the small crystals in the sheet mineral are; as for most small

crystals, a large part of the 3-D surface can be seen. As the small crystals mostly show at the weathered platy locations, the most logical interpretation is that the crystals are incorporated in the biotite crystals and are freed as the sheet mineral is weathered. Of all the minerals mentioned in the geological description, the platy minerals such as muscovite/biotite will be the first to weather. Alternatively, the arsenic minerals can be adsorbed on the free platy surfaces, as other studies suggest that arsenic is easily adsorbed on the mica/platy crystals ([36], 2011). The higher concentrations of As are associated with higher Fe, Pb and S concentrations. This corresponds well with the finding of beudantite in the XRD analysis. Beudantite ($PbFe_3(AsO_4)SO_4(OH)_6$) is a mineral known to occur in oxidized zones of polymetallic ore minerals.

The use of biochar in the soil mixture was shown in the SEM study to have a major impact on the distribution of Pb and As. In combination with the weathered sheet minerals, the Pb and to a lesser degree As, was found attached on the biochar surfaces. This demonstrates that the Pb immobilization observed was due, at least partly, to an immobilization of the element on the biochar surface. Rees et al. [37] also demonstrated that Pb could be immobilized on the biochar surface, especially with the carbonate phase.

5 Conclusion

A field study was performed on a former mine technosol highly contaminated by As and Pb, to which a hardwood biochar was applied to evaluate the effect of biochar, after 6 months, on soil fertility and metal(loid) mobility. Soil properties such as pH, organic matter content and nutrient availability were improved by biochar immediately after its incorporation and these benefits were still observed after 6 months. Pb was immediately immobilized by biochar whereas As was first mobilized and then immobilized after 6 months. Finally, the microscopic analysis of the soil particles and the biochar before and 6 months after its incorporation to the soil showed that Pb was fixed on the biochar surface, mainly as $PbCO_3$, which could be one of the mechanism of Pb immobilization, in addition to precipitation due to pH increase.

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