

Original Research

Enhancement of Soil Arsenic Mobilization through Hydrochar from Arsenic-Contaminated *Boehmeria Nivea*: Roles of Alkali-Modification and Soil Microbial Modulation

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Abstract

The reuse of heavy-metal hyperaccumulator waste has emerged as a critical constraint in the implementation of phytoremediation to control heavy-metal pollution. Ramie (*Boehmeria nivea*) is a well-established arsenic (As) hyperaccumulator that has been widely used in the remediation of As-contaminated sites in recent years. To facilitate multipurpose development and the recycling of ramie straw contaminated with heavy metals, a study investigated the preparation of hydrochar (HC) at different temperatures and assessed the feasibility of reintroducing the HC into the soil. The results indicated higher yields, increased nutrient contents, and reduced As percentage levels at a hydrothermal carbonization temperature of 240°C. Treatment with a 300 mmol/L sodium hydroxide solution resulted in the removal of As from HC, ranging from 77.06% to 87.60%, effectively facilitating the reuse of HC. The application of alkali-modification HC in As-contaminated soil not only enhances soil properties but also augments the potential population of dominant microorganisms that promote soil arsenic mobilization, thereby improving arsenic mobilization capacity in mining soil. This study offers a viable strategy for enhancing the phytoremediation efficiency of As-contaminated soil and establishing a recycling system for As hyperaccumulator resources in the future.

Keywords: ramie, phytoremediation, hydrochar, alkali modification, soil microorganism

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Introduction

Arsenic (As) is one of the most toxic elements to humans and various other biota [1]. In nature, As in the soil primarily originates from the weathering of rocks, and subsequently enters the food chain through plant absorption [2]. In modern times, human activities such as mining and smelting have significantly intensified As pollution in the soil, posing a considerable threat to human health [3]. As contamination has been reported at numerous mining sites, reaching levels of several thousand mg/kg in the surrounding soil and several thousand $\mu\text{g/L}$ in freshwater [4, 5]. Prolonged exposure to elevated As levels can result in metabolic disorders, peripheral neuropathy, conjunctivitis, diabetes, hypertension, cardiovascular disease, organ damage, and cancer, affecting the skin, lungs, kidneys, and liver [6]. Therefore, it is imperative to reduce the risk of As pollution in soil.

Among the technologies established or under development for treating soils polluted with As, phytoremediation appears to be the most suitable for dealing with polluted mine sites, as it is a cost-effective method with minimal negative environmental impacts [7]. Hyperaccumulators, such as *Pteris vittata*, *Boehmeria nivea*, *Althernanthera ficoides*, and *Phyllanthus amarus*, have been widely employed in the remediation of As-contaminated sites in recent years [8, 9]. However, there is limited research on the disposal of As-enriched plants. Improper handling of post-harvest As hyperaccumulators can lead to secondary pollution. Currently, the post-harvest treatment of restoration plants worldwide mainly involves treating them as waste or hazardous materials through incineration or compression landfills, which can result in severe environmental pollution and safety incidents [10]. Therefore, it is crucial to develop a new, environment-friendly, and cost-effective technique for the disposal of As hyperaccumulators.

Carbonization, a globally advanced environmental protection technology, is widely used to treat crop waste, wood, plant tissues, and animal bones [7]. Typically, carbonization includes pyrolysis, combustion, gasification, and hydrothermal pretreatment [11, 12]. Hydrochar (HC), a carbon-rich solid-phase material, is produced by the hydrothermal carbonization of biomass under high-pressure and low-temperature conditions [13]. HC not only has a stable carbon structure and is cost-effective compared to conventional pyrolysis-produced biochar, but also displays a high heating value, diverse pore structure, and abundant surface oxygen-containing functional groups. These properties make it a promising material for diverse applications such as energy sources, adsorbents, soil amendments, and fertilizers [14, 15]. Recent studies have confirmed that biochar application enhances the potential mobility and bioavailability of heavy metals in soil [16, 17]. Biochar addition can boost As mobility by promoting As-reducing bacterial activity in the soil, consequently increasing As toxicity [18]. Moreover, biochar promotes the dissolution of iron by

acting as an electron shuttle between the iron-reducing bacteria and iron minerals. Biochar itself serves as an electron donor for iron reduction, and both processes may release As from iron hydroxides [19]. Biochar also acts as a carbon and nutrient source for microbial growth in the soil, stimulating microbially mediated As mobilization [20]. Hence, biochar has the potential to enhance plant restoration efficiency in As-contaminated soils in mining areas. However, biochar derived from agricultural waste for plant restoration often contains high levels of heavy metals because its preparation temperature is usually insufficient to reach the volatile threshold of these metals [7]. Previous studies have confirmed that the high available As content in the biochar produced directly from As-enriched *P. vittata* can result in secondary pollution [21]. Thus, it is necessary to modify the initial HC to eliminate heavy metals.

Surface modification of biochar is commonly performed using materials such as Fe-Mn oxides, zero-valent iron, humic acid, and chitosan. This enhances the surface characteristics of biochar and improves its adsorption and fixation of heavy metals [22-25]. For example, biochar modified with 3% (w/w) $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ reduced As concentration in soil, straw, and brown rice by 41.7%, 61.5%, and 80.1%, respectively [26]. Biochar derived from wheat straw modified with 1.5% (w/w) goethite led to a 32.2% reduction in As in rice roots and a 46.6% reduction in stems [27]. However, there has been limited research on the removal of heavy metals from biochar, particularly HC derived from As-rich biomass. Various chemicals, including acids, bases, chelates, and organic reagents, have been used to extract As from soil and biomass. Acids and bases, in particular, show superior As removal performance [28, 29]. For instance, oxalate effectively extracts arsenic bound to amorphous iron/aluminum hydrous oxides through the reductive and/or ligand-promoted dissolution of oxides. Furthermore, the reduction of As(V) to As(III) by oxalate enhances As extraction through desorption from the soil surface [30, 31]. Jang et al. demonstrated that sodium hydroxide (NaOH) exhibited higher and faster As extraction efficiencies than other acid extractants. This was attributed to the ligand displacement reaction of hydroxyl ions with As species and high pH conditions that prevented As readsorption [32]. However, the effectiveness of alkaline agents for removing As from As-rich HC remains unclear. Additionally, the impact of modified HC on promoting or inhibiting As mobilization in soil deserves further investigation.

Ramie (*Boehmeria nivea*) is an arsenic hyperaccumulator that has been extensively used in the remediation of arsenic-contaminated sites in recent years [33]. Ramie straw (RS), which is widely available and abundant in China, is used as agricultural biomass. Hence, this study suggests the use of RS to produce four types of HC at various hydrothermal pretreatment temperatures. Simultaneously, an alkaline modification was applied to remove As from HC and then to the soil in mining areas. This study aimed to 1) produce HC from RS using

hydrothermal carbonization technology, 2) investigate the heavy metal removal effect of alkaline-modified HC, and 3) evaluate the impact of alkaline-modified HC on As mobilization in mining area soil. This study aims to use modified HC to enhance As mobilization in mining area soil, improve the efficiency of plant restoration in As-contaminated soil, and establish a system for recycling and utilizing As hyperaccumulator resources.

Experimental

Materials and Reagents

All the reagents used in this study were purchased from Sinopharm Chemical Reagent Co., Ltd. (Shanghai, China). RS for this study were collected from As and antimony co-contaminated mining areas in Xikuangshan, Hunan Province, China (111°29'57.53" E, 27°47'3.18" N). The total As content in the RS was 6.40 mg/kg. The RS was air-dried, crushed, and sieved (0.45 mm) for the HC synthesis.

Hydrochar Synthesis

The HC was synthesized in a 100 mL lifttable high-pressure reactor at temperatures of 160°C, 200°C, 240°C, and 280°C. The hydrothermal carbonization process involved mixing 6 g of the RS powder with deionized water (solid:water = 1:10, w/w) in a sealed reactor container. The mixture was heated to the specified temperature and reacted for 4 hours under a pressure of 2.6–2.9 MPa [7]. After completing the reaction, solid and liquid products were separated when the internal temperature of the high-pressure reactor naturally dropped below 60°C. The liquid product was stored at 4°C, and the solid product was washed with distilled water to neutrality, dried at 105°C for 12 hours, ground, and stored for later use. The HC prepared at these temperatures are denoted as HC160, HC200, HC240, and HC280, respectively.

Removal of As from Hydrochar

Considering that heavy metal elements, especially As, may persist in HC during synthesis and affect As mobilization in the mining area soil, we employed sodium oxalate and NaOH at various concentrations to modify and extract heavy metal elements from HC240 [31, 32]. In summary, 2 g of HC was mixed with 20 mL of sodium oxalate (25, 50, and 75 mmol/L) or NaOH solution (100, 200, and 300 mmol/L) at a solid-liquid ratio of 1:10 (w/w), using deionized water as a blank control. The mixture was placed in a rotary oscillating device and rotated for 12 hours (70 r/min, 25°C). Subsequently, the samples were centrifuged at high speed for 10 minutes (8000 r/min), separating the HC from the extractant to obtain the modified HC samples. The entire extraction process was repeated three times, and each extraction was performed in triplicate. After each extraction step, the heavy metals remaining in the HC were determined

to assess the removal efficiency of heavy metals from the HC under different concentrations and extraction cycles.

Microverse Cultivation Experiments

A microcosm cultivation experiment was conducted to explore the impact of various dosages of alkaline-modified HC (highest As removal efficiency) on soil As mobilization. The heavy metal-contaminated soil in the experiment was also taken from Xikuangshan (111°29'56.904", 27°46'11.3196"N). Surface soil samples (0–20 cm) were air-dried, sieved (2 mm), and adjusted to 60% of the field water holding capacity before the experiments. Four treatments were applied: a control without HC application (CK), 0.5% HC (HC-0.5%), 1.0% HC (HC-1.0%), and 2.0% HC (HC-2.0%) into the soil, with each treatment conducted in triplicate. Specifically, 100 g of air-dried soil was placed in a 500 mL open glass bottle, and the soil moisture was adjusted to 60% of the field water-holding capacity. Subsequently, each uncovered glass bottle was filled with the designated amount of hydrothermal carbon and incubated at 25°C for 28 days. Soil sampling was conducted at 7-day intervals to evaluate the effects of varying HC concentrations on As mobilization and the physicochemical properties of the soil.

DNA Extraction, Amplification, and Sequencing

On the 28th day following the conclusion of the microcosm cultivation experiment, fresh soil (0.5 g) from each replicate was sieved, and total microbial DNA was extracted using the Bio Fast Soil Genomic DNA Extraction Kit (Bioer, Hangzhou, China), following the manufacturer's instructions. The quantity and purity of DNA were assessed using spectrophotometry (Nanodrop 2000, Thermo Scientific, USA) [34]. The bacterial 16S rRNA and fungal ITS rRNA genes were amplified using primers described by Ma et al. [35]. The purified PCR products were sequenced on an Illumina MiSeq PE300 platform (Illumina, San Diego, CA, USA). The raw tags obtained by sequencing were first spliced using Flash 1.2.7, and the paired-end reads were filtered using Trimmomatic 0.33. Uchime 8.1 was used to identify and remove ambiguous bases and chimeras to obtain valid data. Operational taxonomic units (OTUs) were clustered at 97% sequence similarity using UCLUST, and a representative sequence of each OTU was selected for taxonomic assignment. To perform the taxonomic assignment, we blasted the bacterial and fungal OTUs against the Silva 138.1 database and UNITE 7.2 fungi database, respectively [30].

Analysis of Physicochemical Properties of Hydrochar and Soil

The aqua regia heating method was employed for digestion to determine the heavy metals and nutrient elements in HC [36]. In brief, 0.1 g of HC or 5 mL hydrothermal liquid-phase product was weighed and placed in a conical flask, and then 6 mL of aqua regia

solution was added. The flask was covered with a bent-neck funnel and heated on an electric heating plate (PZ 28-2, Harry Gestigkeit, Germany), and aqua regia was gently boiled for 2 h. Heavy metals and nutrients in the digestion solution were determined using inductively coupled plasma mass spectrometry (ICP-MS, NexION 350, PerkinElmer, USA). The soil and HC pH were measured using a pH meter (PHS-3C, LEICI, China) with a soil/HC: water ratio of 1:2.5/20 [34, 37]. The in-situ redox state (Eh) was measured using a redox microelectrode (Pt805, Mettler Toledo, USA). The available As content in the soil was extracted with 0.5 mol/L NaHCO₃ at room temperature for 16 h and measured by ICP-MS [38]. Soil organic matter (SOM) was determined using the H₂SO₄-K₂Cr₂O₇ external heating method [39].

Statistical Analysis

Analyses of heavy metals and nutrients in the HC and soil physicochemical properties were performed using SPSS 26.0 (IBM, USA). One-way analysis of variance (ANOVA) with Duncan's multiple comparisons ($p < 0.05$) was applied to assess the data that met the homogeneity of variance criteria. Results for each treatment are presented as arithmetic means \pm standard error (SE) with $n = 3$, and data visualization was conducted using Origin 2024 software. Redundancy analysis (RDA) was employed to identify significant factors influencing the dominant bacterial/fungous phylum and the bioavailability of As, utilizing Canoco 5.0 (Microcomputer Power, USA).

Table 1. Effect of different hydrothermal temperatures on the yield and pH of hydrochar.

Hydrochar	Yield (%)	pH
HTC160	72.33 a	4.96 a
HTC200	61.70 b	4.97 a
HTC240	42.56 c	4.95 a
HTC280	32.67 d	5.03 a

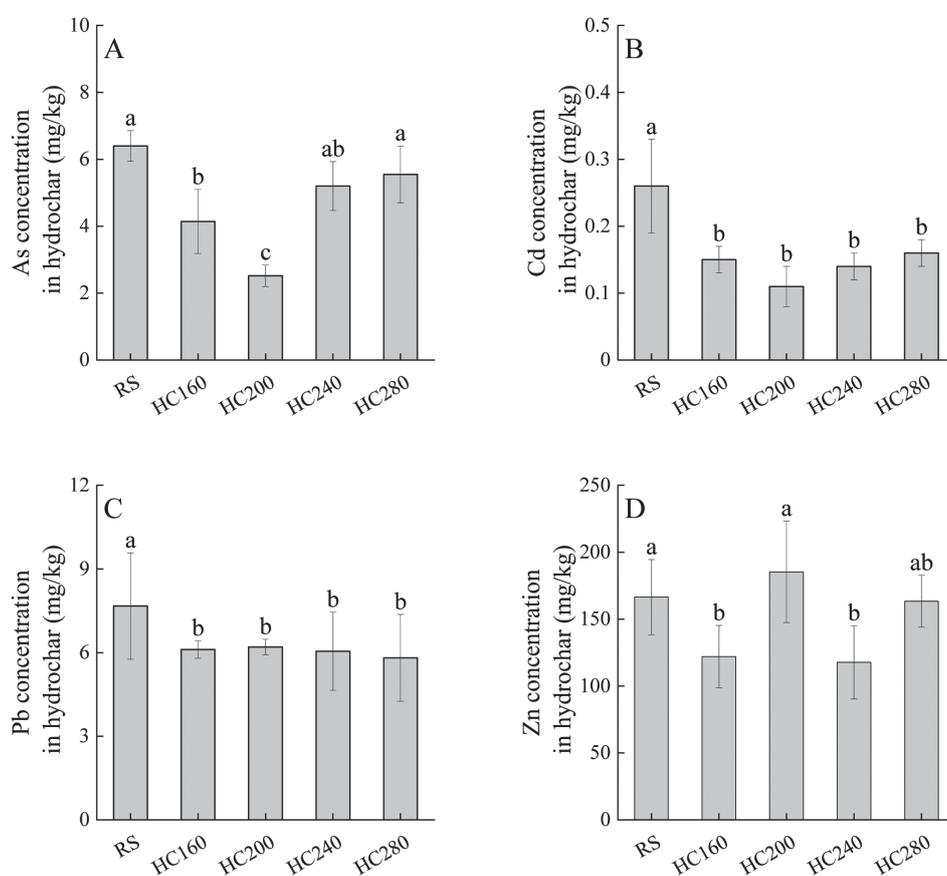


Fig. 1. Changes in heavy metal content of hydrochar carbonized at different temperatures. A) As; B) Cd; C) Pb; D) Zn. Different lowercase letters indicate significant differences between different treatments ($p < 0.05$).

Results

Influence of Temperature on Hydrochar Yield and pH

Table 1 presents the influence of different hydrothermal temperatures on HC production yield and initial pH. As the hydrothermal reaction temperature increased, the HC yield gradually decreased within the range of 32.67% to 72.33%, showing a significant difference. In contrast, the initial pH of HC followed the opposite trend to that of the yield variation. With an increase in the reaction temperature, the pH of the HC increased from 4.95 to 5.03. However, the HC produced at different temperatures did not exhibit significant pH differences.

Heavy Metal Distribution in the Prepared Hydrochar

The residual heavy metal content in the HC produced at various hydrothermal reaction temperatures is shown in Fig. 1. The As content in HC at different temperatures initially decreased, followed by an increase with increasing temperature. Specifically, the As content in HC160 and HC200 (4.14 and 2.52 mg/kg, respectively) was

significantly lower than in RS (6.40 mg/kg, $p < 0.05$). In contrast, HC240 and HC280 showed only modest decreases of 18.72% and 13.17%, respectively, compared to RS, with no significant difference. Furthermore, irrespective of the hydrothermal reaction temperature, the Cd and Pb contents in HC were significantly lower than those in RS ($p < 0.05$), indicating the effective reduction of Cd and Pb levels in the biomass through hydrothermal carbonization. Notably, compared to RS, only HC160 and HC240 significantly decreased the residual zinc (Zn) by 26.74% and 29.27% ($p < 0.05$), respectively, while HC200 processing resulted in Zn enrichment, which was 11.36% higher than that of RS.

Upon considering the distribution of heavy metals between the liquid and solid phases during the hydrothermal preparation process, it was observed that heavy metals primarily migrated from the solid phase to the liquid phase, thereby reducing the residual heavy metal content in the HC (Fig. 2). Except for Zn, the residual heavy metal content in the HC gradually decreased with increasing hydrothermal carbonization temperature. Interestingly, HC200 treatment indicated that 74% of the residual Zn was distributed in the solid phase, whereas HC240 and HC280 treatments significantly decreased this proportion.

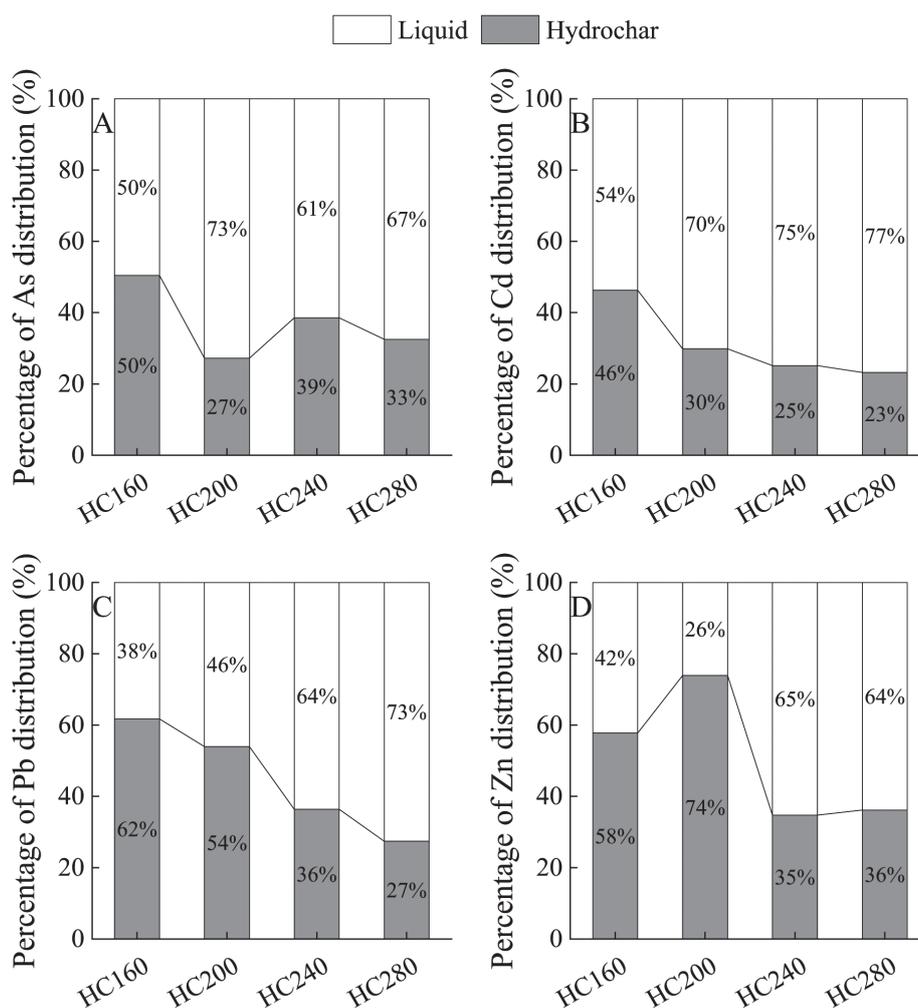


Fig. 2. Distribution of heavy metals in the solid and liquid hydrochar products A) As; B) Cd; C) Pb; D) Zn.

Considering the distribution of heavy metals in the solid and liquid phases, HC240 and HC280 exhibited the lowest residual heavy metal levels.

Residual Nutrient Elements in the Hydrochar

As the hydrothermal carbonization temperature increased, distinct variations were observed in the patterns of residual nutrient elements in HC (Fig. 3). The concentrations of calcium (Ca), potassium (K), and magnesium (Mg) initially increased and then decreased with increasing temperature. The HC240 treatment resulted in significantly higher concentrations of Ca and K compared to other treatments, at 6591.94 and 1942.93 mg/kg, respectively ($p < 0.05$). Residual phosphorus (P) concentrations in HC at different temperatures

followed the order HC280 (2606.98 mg/kg) > HC240 (1437.26 mg/kg) > HC200 (806.46 mg/kg) > HC160 (507.73 mg/kg). For iron (Fe), as the hydrothermal carbonization temperature increased, the retained Fe concentration in HC gradually decreased within the range of 282.30–524.25 mg/kg, with HC240 exhibiting the lowest Fe level. However, no significant difference was observed in the retained concentrations of sodium (Na) and manganese (Mn) in HC at different temperatures, ranging from 374.67–458.47 mg/kg and 8.11–14.49 mg/kg, respectively. Considering the retained levels of both heavy metals and nutrients in HC, the HC240 treatment resulted in lower levels of various heavy metals and higher levels of nutrient elements. Therefore, HC240 was selected as the research target for alkaline modification and practical applications.

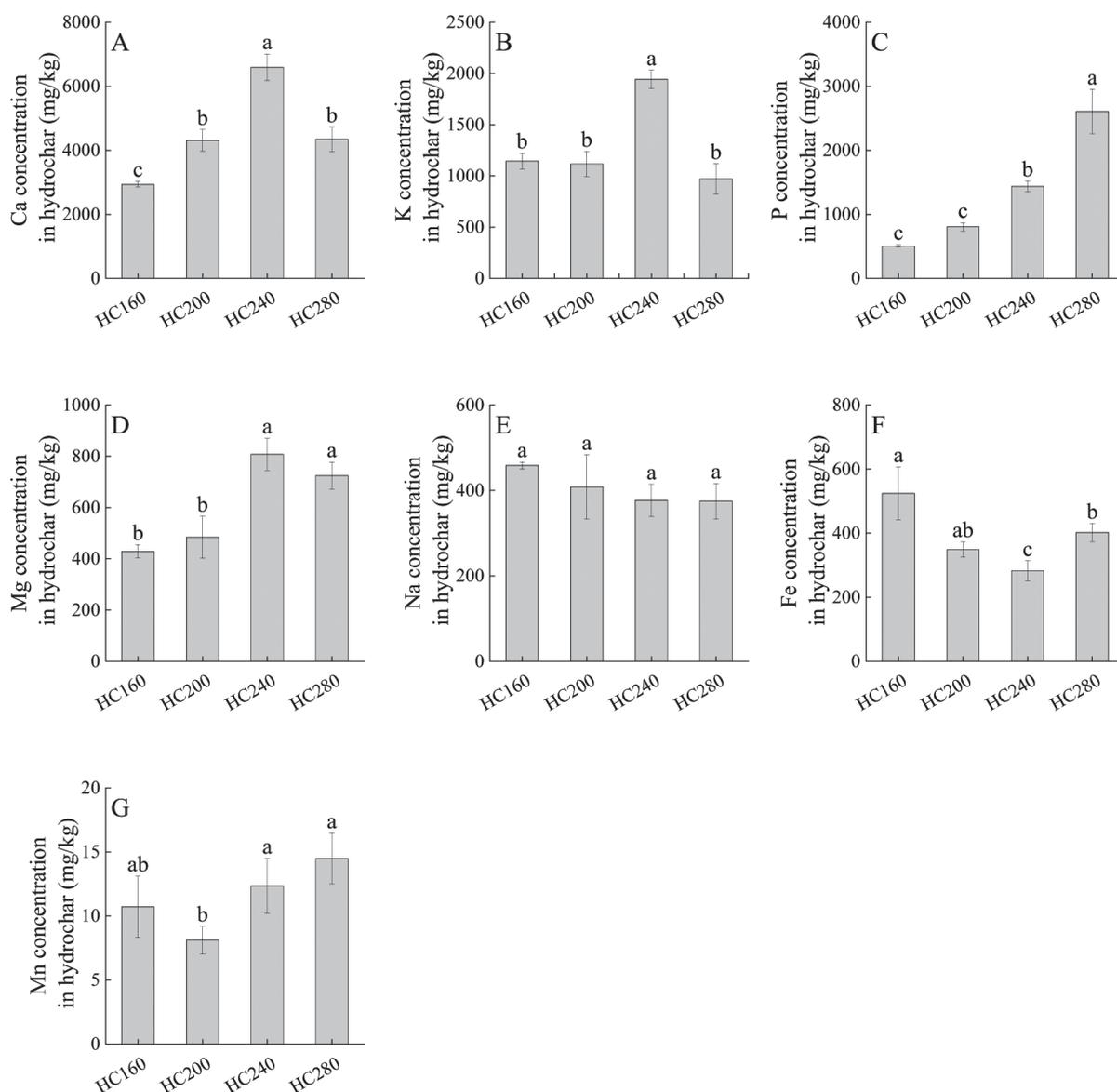


Fig. 3. Changes in the nutrient element content of hydrochar carbonized at different temperatures. A) Ca; B) K; C) P; D) Mg; E) Na; F) Fe; G) Mn. Different lowercase letters indicate significant differences between the treatments ($p < 0.05$).

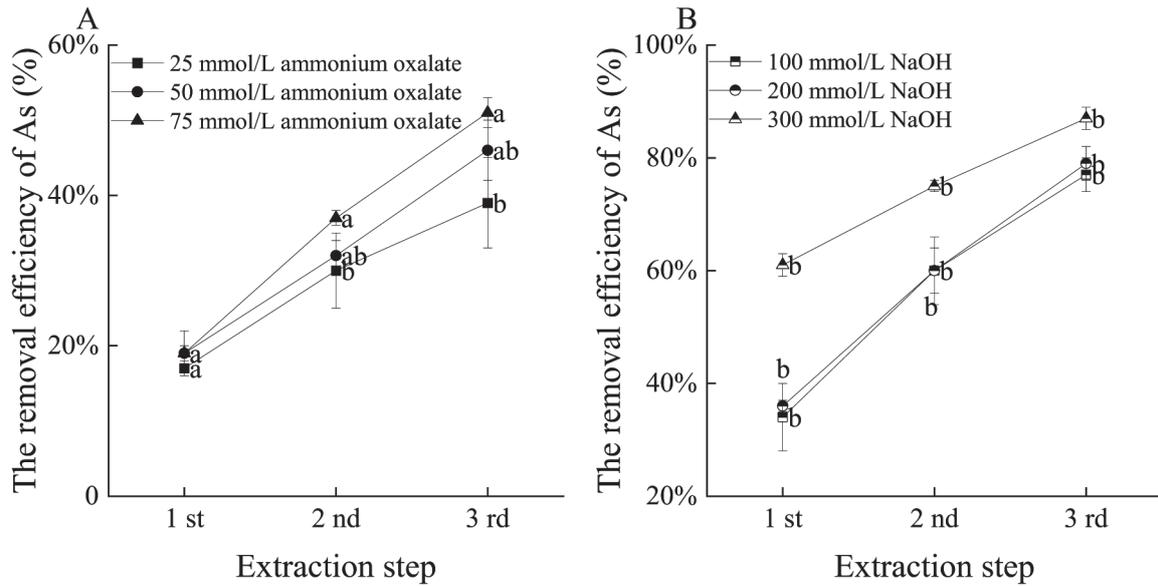


Fig. 4. Differences in the removal efficiency of As from hydrochar using different concentrations of ammonium oxalate and NaOH. A) As content in hydrochar extracted with ammonium oxalate; B) As content in hydrochar extracted with NaOH. Different lowercase letters indicate significant differences among the different extraction steps ($p < 0.05$).

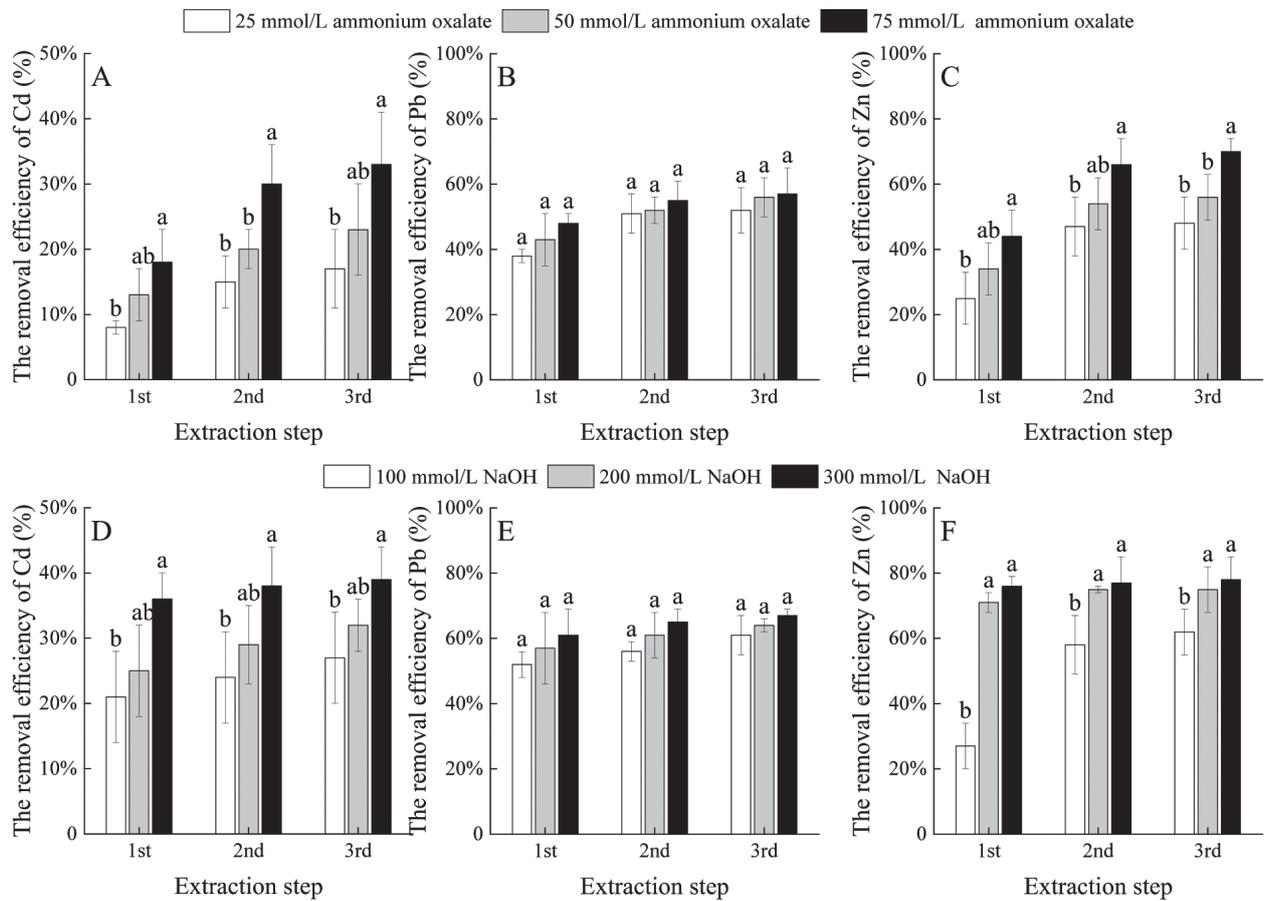


Fig. 5. Differences in removal efficiencies of Cd, Pb, and Zn from hydrochar using different concentrations of ammonium oxalate and NaOH. Cd (A), Pb (B), and Zn (C) content in hydrochar extracted with ammonium oxalate; Cd (D), Pb (E), and Zn (F) content in hydrochar extracted using NaOH. Different lowercase letters indicate significant differences among the different extraction steps ($p < 0.05$).

Effects of Alkali Modification on Heavy Metal Removal from Hydrochars

Fig. 4 illustrates the effects of three successive extractions with different concentrations of sodium oxalate and NaOH on the proportion of As. After the first extraction with sodium oxalate, the As concentration in the HC decreased significantly, and the removal efficiencies reached 17.40%, 19.43%, and 19.37%, respectively (Fig. 4A). As the number of extractions increased, the removal rate of As from the HC gradually increased with increasing concentrations of sodium oxalate. After three consecutive extractions with different concentrations of sodium oxalate, the As removal efficiencies reached

39.04% (25 mmol/L), 46.57% (50 mmol/L), and 51.15% (75 mmol/L). Similarly, different concentrations of NaOH were used as extraction agents to remove As from HC, with a significantly higher removal efficiency than sodium oxalate treatment, and the efficiency increased with the concentration and number of extractions (Fig. 4B). After three successive extractions, different concentrations of NaOH achieved removal efficiencies of 77.06% (100 mmol/L), 79.25% (200 mmol/L), and 87.60% (300 mmol/L) of As in HC. Notably, a single extraction using 300 mmol/L NaOH resulted in a high removal efficiency of 61.25%.

For the other heavy metals (Cd, Pb, and Zn), the removal efficiencies of both extraction agents were similar

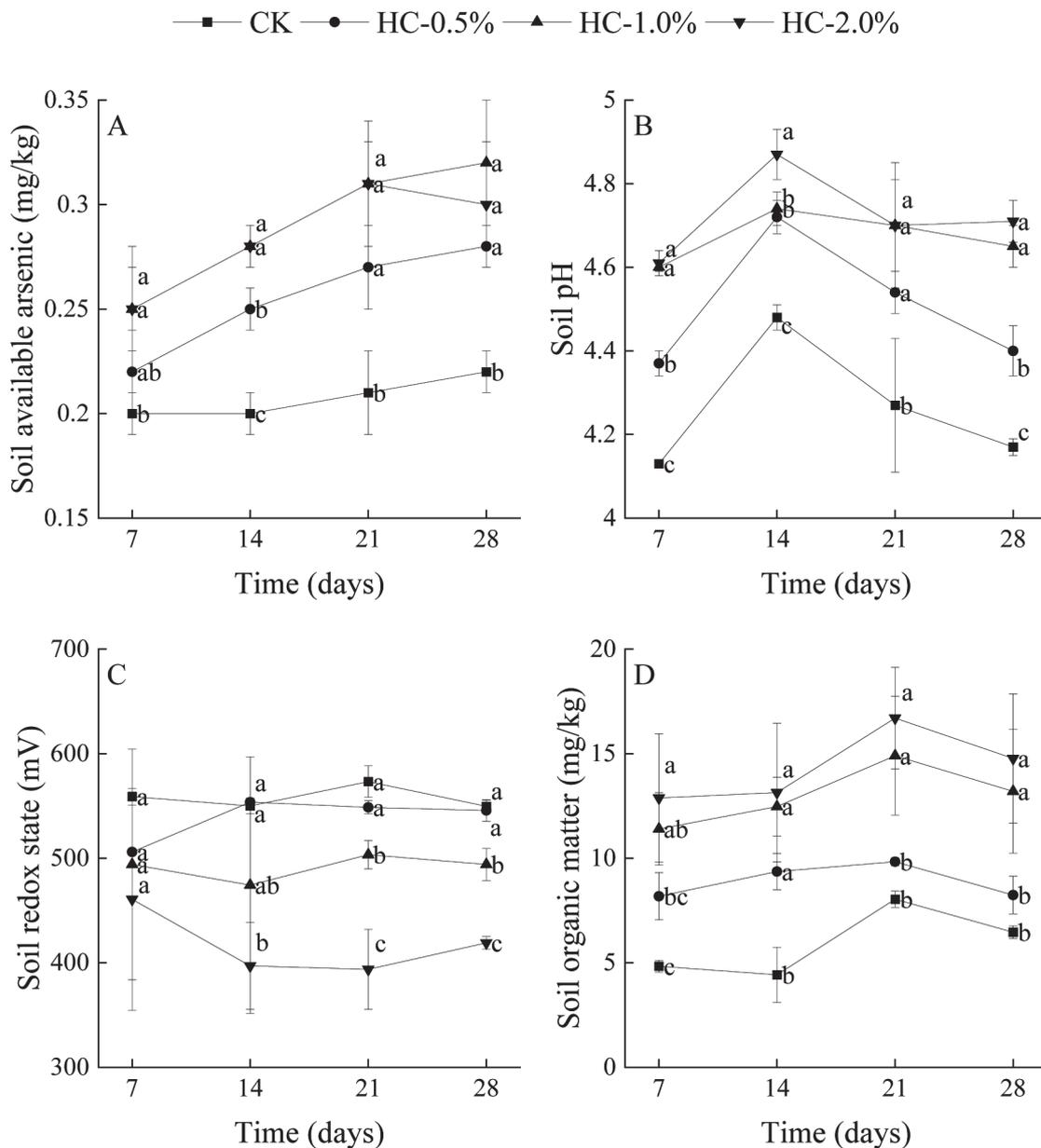


Fig. 6. Variations in soil properties under different hydrochar dosages applied during the entire cultivation period: A) Soil-available As; B) soil pH; C) soil redox state; D) soil organic matter. Different lowercase letters indicate significant differences between different hydrochar dosages ($p < 0.05$).

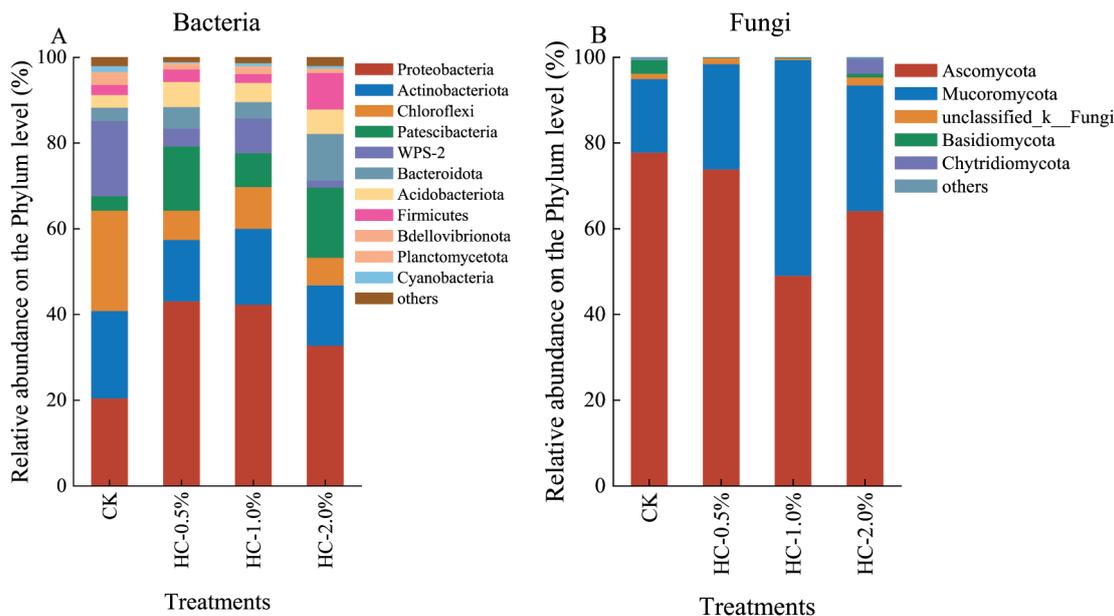


Fig. 7. Variation in relative microbial abundance at different hydrochar dosages. A) Bacterial abundance and B) fungal abundance at the phylum level.

to those of As, where NaOH had a stronger removal efficiency than sodium oxalate (Fig. 5). After three consecutive extractions with different concentrations of NaOH, the removal efficiencies of Cd in the HC ranged from 27.02% to 39.54%, Pb from 60.98% to 67.01%, and Zn from 61.44% to 78.20%. Therefore, considering the complexity, removal efficiency, and economic benefits, a single extraction with 300 mmol/L NaOH may be an effective strategy for removing heavy metals from HC.

Impact of Hydrochar Application on Soil Properties

The addition of HC at different dosages significantly promoted As mobilization, which increased with prolonged exposure time (Fig. 6A). At 28 d, all HC treatments significantly enhanced the bioavailability of soil As. The activation capability followed the sequence HC-1.0% (0.32 mg/kg) > HC-2.0% (0.30 mg/kg) > HC-0.5% (0.28 mg/kg) > CK (0.22 mg/kg), resulting in significant improvements of 47.69%, 37.50%, and 29.17%, respectively, compared to CK ($p < 0.05$). Regarding pH, a trend of initial increase and then decrease was observed throughout the cultivation period, reaching a peak at 14 d (Fig. 6B). At 28 d, the pH values of the HC-2.0% and HC-1.0% treatments were significantly higher than those of the other treatments, at 4.71 and 4.65, respectively ($p < 0.05$). Soil Eh values did not significantly change after HC was applied to the soil, with only the HC-2.0% treatment being significantly lower than that of the other treatments. Notably, the addition of HC significantly increased soil SOM content, reaching its peak at day 21. Specifically, the HC-1.0% and HC-2.0% treatments significantly increased SOM by 85.32% and 107.71%, respectively, compared to the CK (Fig. 6D). In

summary, the addition of HC significantly enhanced the soil pH and SOM, promoted the mobilization of As in the soil, and improved the efficiency of As-contaminated soil remediation for As hyperaccumulator plants.

Soil Bacterial and Fungal Community Responses to Hydrochar Application

Fig. 7A illustrates the relative abundance of bacterial taxa at the phylum level in the different treatments. In the CK treatment, Proteobacteria, Actinobacteria, Chloroflexi, and WPS-2 dominated, accounting for 20.50%, 20.42%, 23.40%, and 17.47% of the total bacterial population, respectively. In the HC-treated groups, Proteobacteria, Actinobacteria, and Patescibacteria were dominant. Compared to the CK, the relative abundance of Proteobacteria in the HC-0.5%, HC-1.0%, and HC-2.0% treatments increased by 22.64%, 21.85%, and 12.32%, respectively, and that of Patescibacteria increased by 11.57%, 4.52%, and 13.01%, respectively. The dominant bacterial taxa in the CK (Actinobacteria, Chloroflexi, and WPS-2) showed a decrease in relative abundance in the HC-treated groups, particularly in Chloroflexi and WPS-2. Similar to the changes in the bacterial communities, the HC-treated groups had a significant impact on the soil fungal community (Fig. 7B). In the CK treatment, Ascomycota (77.83%) and Mucoromycota (17.17%) dominated the fungal community at the phylum level. In the HC-treated groups, the fungal community composition in HC-0.5% was similar to that in the CK; however, HC-1.0% and HC-2.0% markedly decreased the dominant phylum Ascomycota (49.08% and 64.21%, respectively) and increased the relative abundance of Mucoromycota (50.33% and 29.30%, respectively). Additionally, HC-

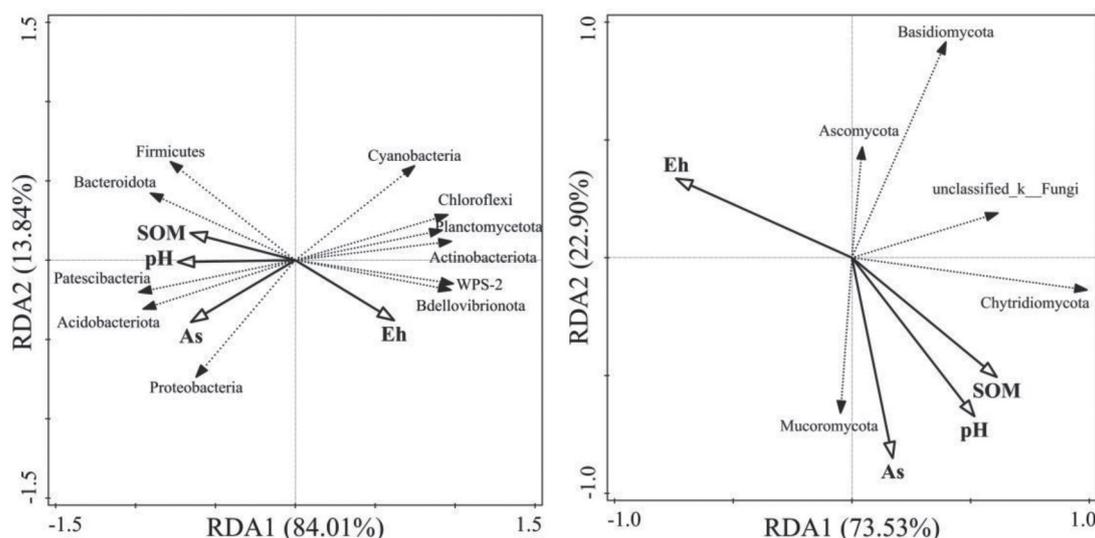


Fig. 8. Redundancy analysis (RDA) score charts for correlations between soil physicochemical variables and microbial taxa. A) Bacterial taxa; B) fungal taxa.

2.0% increased the relative abundance of the unique fungal phylum Chytridiomycota (3.48%). Overall, the addition of HC to the soil significantly improved the community structure of the soil microbes.

Redundancy Analysis

RDA was employed to explore the interactions among the soil physicochemical variables, available As, and microbial community composition (Fig. 8). The results revealed that soil properties and microbial community composition (bacteria and fungi) contributed 97.85% and 96.43% of the total variation, respectively. Soil available As exhibited a positive correlation with soil pH and SOM, but a negative correlation with Eh. Regarding the bacterial community, significant positive correlations with soil available As content were observed for Proteobacteria, Patescibacteria, Bacteroidetes, and Acidobacteria, whereas Actinobacteria, Chloroflexi, Bdellovibrionota, Planctomycetota, and Cyanobacteria were negatively correlated with the soil available As content (Fig. 8A). In terms of fungal community composition, only Mucoromycota showed a positive correlation with the soil available As content, whereas the remaining fungal taxa demonstrated negative correlations (Fig. 8B).

Discussion

Reuse of Phytoremediation Plants

The reutilization of plants for remediation has become a critical factor in implementing practical strategies to control heavy metal pollution in plants [40]. The development of these remediation plants not only helps offset economic costs, but also facilitates the redistribution of heavy metals. A straightforward strategy

is to use plant tissues without heavy metal enrichment; however, this is often unsuitable for tissues with transport functions, such as roots and stems [41]. *Boehmeria nivea*, which is recognized as an As hyperaccumulator, is widely used to restore As-contaminated soils [33]. However, inadequate and unscientific management of post-harvest residual straw from *Boehmeria nivea* frequently leads to secondary pollution incidents [10]. Therefore, it is crucial to conduct comprehensive property evaluations before considering the reutilization of residual straw.

Alkaline Modification to Remove Heavy Metal Pollution from Hydrochars

Currently, HC has gained significant attention as a soil amendment owing to its abundant raw material sources, cost-effective preparation, and contribution to carbon reduction [42]. In this study, higher yields and nutritional components, as well as lower proportions of As in HC were observed under hydrothermal carbonization at 240°C. Although most studies have confirmed the ability of HC to absorb heavy metals and enhance plant resistance, attention has also been directed towards the potential damage that HC may cause to soil ecosystems [43]. Previous studies support this perspective, indicating that the direct application of heavy-metal-contaminated straw biochar significantly increased the soil heavy-metal content by more than seven times compared to the control when heavy metals were not removed [44]. Hence, this study used NaOH to remove heavy metals from HC derived from the straw of As-hyperaccumulating plants. The results indicated that the removal efficiency of As from HC after treatment with 300 mmol/L NaOH ranged from 77.06% to 87.60%. The alkaline-modified HC achieved almost complete reutilization. This was attributed to the ligand displacement reaction of hydroxyl ions with As species and high pH conditions that prevented As

readsorption [32]. Under alkaline conditions, oxyanion species, such as As, are considered highly mobile and leachable, whereas other cations, such as Cd and Zn, are only moderately mobile [45]. Additionally, acidification is the primary reason for HC aging, and NaOH treatment is beneficial for the reactivation of HC [46]. Thus, the modified HC can be applied not only to soil but also further developed by exploring other application pathways for HC.

Effect of Soil Properties on Hydrochar Amendment

Changes in soil characteristics can significantly affect the bioavailability of As in soils. Our results revealed a notable increase in soil physicochemical variables (pH and SOM) in As-contaminated soil with increasing HC dosage. This observation aligns with a previous study that demonstrated that biochar supplementation elevated the rhizosphere pH and nutrient content [47]. This is primarily because alkaline modification increases the pH of HC itself. Furthermore, HC can bind H^+ ions to its surface through specific negatively charged functional groups, potentially releasing alkali salts into the soil [48]. Additionally, HC increased SOM by releasing fixed carbon content and enhancing humic substances, thus accelerating humification processes [49]. According to the RDA analysis (Fig. 8), a positive relationship was observed between soil pH, organic matter, and effective As content. Previous studies have confirmed a close correlation between soil pH, SOM, and plant availability of As [16]. This may be attributed to the presence of As in a negatively charged free state rather than a molecular form. The rise in pH following HC incorporation (Fig. 4) transforms the soil surface into a more negatively charged state (OH^-), potentially increasing the potential for As repulsion [50]. These findings are consistent with earlier reports demonstrating that soil amendments increasing pH enhance the bioavailability of As in the soil [51, 52]. Furthermore, SOM can facilitate As mobilization by mediating redox conditions and competing with As for adsorption sites on metal oxides, thus promoting As desorption [51, 53]. In addition, labile organic matter may stimulate Fe(III)-reducing bacteria, leading to the release of As into the soil solution [54]. Thus, an increase in soil pH and SOM content after HC application may be a potential driving factor altering the mobilization of As in As-polluted soils.

Responses of Soil Microbial Communities to Hydrochar Amendment

The adaptability of microorganisms to survive in a polluted biosphere can be leveraged for the bioremediation of heavy metals, such as As [6]. Bacteria play an essential role in the biogeochemical cycling of As by determining the speciation and bioavailability through oxidation/reduction and methylation/demethylation reactions [55]. In the present study, significant changes occurred in the microbial community structure, especially

among bacteria, after the application of HC (Fig. 7). HC application markedly increased the relative abundance of the dominant bacterial phyla, including Proteobacteria, Patescibacteria, and Acidobacteria, as well as the dominant fungal phylum Mucoromycota. Previous studies have indicated that Proteobacteria, typical As(V)-respiring bacteria, can promote the anaerobic reduction of As(V) in the soil through the *arrA* gene, facilitating the release of As(III) into pore water and, consequently, promoting the mobilization and accumulation of As [56]. Patescibacteria dominated under As-rich conditions, indicating that they may be related to As resistance and transformation [57]. Acidobacteria also exhibit dominance in As-enriched environments, with a positive correlation between bioavailable As and Acidobacteria [58]. Mucoromycota has been documented to function in metal tolerance and plant growth promotion [59]. Considering the results of the RDA regarding the association between the dominant bacterial and fungal phyla and As mobilization, the emphasis was mainly on As mobilization, which suggests that Proteobacteria, Patescibacteria, Acidobacteriota, and Mucoromycota were the dominant phyla that promoted soil As mobilization. Therefore, enhancing the activity and abundance of bacteria that potentially promote soil As mobilization may be a key driving force behind the HC promotion of soil As mobilization.

Ramie Plant Resource-Recycling System

The recycling mode is a crucial strategy for achieving sustainable development of plant remediation, with HC serving as a vital carrier for material cycling. In this study, HC was produced from heavy-metal-contaminated RS at different temperatures, and pure HC was obtained through alkali modification. Applying alkali-modified HC to As-contaminated areas increased soil pH and SOM, enhancing the microbial habitat and creating favorable conditions for potentially dominant phyla that promote soil As mobilization (Fig. 8). These results indicated that alkali-modified HC has the potential to enhance the efficiency of As hyperaccumulators in remediating As-polluted soils, particularly in mining areas. Moreover, the preparation and application of HC have achieved the goal of recycling agricultural waste. Therefore, a ramie-centered phytoremediation cycle system can be established. In this system, ramie plants are utilized to adsorb heavy metals, and after modification, ramie hydrochar can be repeatedly applied to the soil to improve soil conditions and enhance phytoremediation efficiency.

Conclusions

In this study, HC was produced from As-contaminated RS at varying hydrothermal carbonization temperatures. The HC underwent modification using sodium oxalate and NaOH. The research underscores the significance of heavy metal removal from HC for its reusability, showcasing that a 300 mmol/L NaOH solution effectively

removes As from HC. Applying modified HC not only improves soil properties but also enriches the soil with potentially dominant microbial phyla, which aid in soil As mobilization. Consequently, this enhancement contributes to an increased mobilization of As in mining soils. These findings propose a practical strategy to enhance the efficiency of phytoremediation in As-contaminated soil and lay the groundwork for establishing a recycling system for As hyperaccumulator resources in the future.

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Conflict of Interest

The authors declare no conflict of interest.

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