

Original Research

Remediation of Soil Contaminated by Heavy Metals Using Biochar: Strategies and Future Prospects

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Abstract

With the rapid development of industry and human society, massive heavy metals have been released into varied environments, the long-term accumulation of heavy metals is difficult to be removed and toxic to the ecological environment subtly. Global heavy metal pollution is serious, so it is necessary to study and develop economically feasible, green and effective remediation strategies to solve these pollution problems. In soil contaminated by heavy metals, especially in farmland, a vast number of heavy metals will directly or indirectly accumulate the food chain, and then lead to a severe threat to food production and security. Heavy metal pollution in soil threatens health and ecological sustainable development of all kinds of life, including human beings. Biochar, as a current and environmentally friendly material, has been widely used for remediation of water and soil polluted by heavy metals, carbon sequestration and other aspects, exhibiting the great application potential. The adsorption effect of biochar depends largely on its raw material source, preparation method and conditions, so the biochar obtained by traditional preparation method needs to be improved due to its limited adsorption capacity. Increasing scholars have focused on modification biochar in the past decade, and the combined application technology of biochar has also achieved huge progresses. Based on this, this review summarizes the current research progress of biochar in remediation of heavy metal pollution, the production, properties, repair mechanism, and modified methods. Simultaneously, the applications of biochar combined with microbial technology are introduced in detail, and we also propose the future research and development direction of biochar used for remediation of soil contaminated by heavy metals.

Keywords: biochar, soil heavy metals, adsorption efficiency, remediation mechanism, combined application

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Introduction

Soil, as a significant essential resource, is a basis for the survival and development of human beings, animals and plants. However, with the rapid development of industry, agriculture and urbanization, a massive number of heavy metal elements accumulate in soil and effectively accumulate in animals and plants. Heavy metals in soil will not only reduce soil fertility and damage the soil ecological environment, but also affect crop growth, reduce crop yield and quality. Moreover, it will damage the food chain and endanger human health [1], even cause irreparable destruction to the ecosystem. Heavy metal pollutants in the atmosphere, water, soil, sediment, plants and other systems, especially in cultivated farmland soil. From the pollution types, inorganic wastes play a leading role; from the pollutants exceeds bids, give priority to with Cd, Hg, Cu, Pb [2]. Land resource pollution represents a global environmental problem, so it is urgent to develop remediation and treatment technologies for soil heavy metal pollution [3].

Soil heavy metal pollution has attracted much attention due to its long-term accumulation, potential toxicity, high persistence and non-biodegradability [4]. According to the research, no matter what kind of source and type of soil heavy metals, their pollution characteristics make it difficult to rely on soil self-purification ability elimination method, so artificial control measures must be taken to effectively restore the contaminated soil. Traditional repair technology mainly includes physical remediation, replace without-soil, separation, heat treatment, etc., chemical remediation (soil leaching, chemical oxidation, passivation, etc.) and bioremediation (phytoremediation,

microbial remediation, animal remediation etc.) [5], to some extent, these techniques shown to achieve effective repair effect. However, these conventional technologies are also limited by the disadvantages of complex operation, high operation cost, poor feasibility, low removal rate and high secondary risk (Table 1). In order to overcome these shortcomings, combined with the characteristics of various soil remediation and soil pollution status, the application of combined remediation technology can be strengthened in the future. Therefore, the research on the remediation technology of heavy metal contaminated soil with low cost, high efficiency, environmental protection and sustainability has become the focus of the current environmental workers.

Biochar is a porous, carbon-rich substance with large functional group energy characteristics of porosity, specific surface area and abundance [24]. Because of its unique physical structure and chemical characteristics, biochar applied to soil can not only effectively reduce the bioavailability and migration of heavy metal ions in soil, but also reduce its harm to the ecological environment. It can also enhance soil fertility, increase carbon storage, and improve soil structure and microbial community [25-27]. Therefore, the application of biochar can achieve a win-win situation of environmental protection, clean and pollution-free, so showing a good application prospect in the treatment of heavy metal contaminated soil. However, the adsorption effect of traditional biochar is limited due to various factors. A large number of studies have found that biochar can achieve higher adsorption effect by modification or combined with other remediation materials [28-31], so the improvement of its application technology has become a research hotspot in recent

Table 1. Advantages and disadvantages of remediation technology application for heavy metal contaminated soil.

Remediation technologies	Mechanisms	Advantages	Disadvantages	References
Physical remediation	Soil replacement	Simple operation and fast effect	The amount of work is large, high cost, and damage soil structure	[6, 7]
	Landfilling	Thorough and stable	Occupy farmland, high cost	[8, 9]
	Thermal treatment	Works quickly	High energy consumption, high cost and secondary pollution	[10]
	Vitrification	Stable and efficient	High cost and damage to soil function	[11, [12]
Chemical remediation	Soil washing	Fast and efficient	High cost, secondary pollution	[13]
	Chemical reduction	Simple and efficient	Small application range	[14, 15]
	Solidification	Low cost and fast effect	Require long-term monitoring; Secondary pollution	[16, 17]
Bioremediation	Phytoremediation	Large processing area, low cost and can recover heavy metals	Long cycle time and the effect is slow	[18, 19]
	Microbial remediation	No damage to soil structure, low cost	Poor genetic stability of microorganisms	[20, 21]
	Animal remediation	No destroy the soil structure, improve the soil condition	Long cycle time and small application range	[22, 23]

years. Therefore, the application and improvement of biochar in remediation of soil heavy metal pollution were summarized, and the future development direction of biochar was prospected.

Heavy Metals in Soil and Remediation Methods

Sources and Pollution Hazards of Heavy Metals in Soil

With the rapid modernization of industry, agriculture and urbanization, environmental pollution caused by various wastes, typically heavy metal pollution, has become increasingly prominent and posing challenges to global ecological health. Heavy metals in soil mainly come from two parts: natural behavior and human activities. Under natural conditions, such as volcanic eruption and forest fire, many heavy metals enter the soil through various ways. Compared with natural pollution, heavy metal pollution caused by human activities, such as heavy metal mining, industrial manufacturing activities like leather production, landfill, coal-fired power generation and heating, is more serious [32]. Under various sources of heavy metal pollution, the quality of soil environment changes constantly, which has an impact on human life and ecological sustainable development. Soil, as a critical resource for the survival of animals, plants and humans, will cause soil pollution when the content of heavy metals exceeds the self-purification capacity of soil [33]. The quality of contaminated soil will decline, which will affect the development and yield of crops, and even cause crop death and soil degradation. At the same time, most crops live in the soil. If the soil is contaminated by heavy metals, heavy metals will enter the human body through the soil-plant system and food chain, endangering human health and threatening life safety [34].

Classification and Speciation of Heavy Metals

The pollutants in the soil are complex and diverse, among which typical heavy metals and polycyclic aromatic hydrocarbons pose the most significant threats to the safety of agricultural products, affecting human health and ecosystems [35]. In recent years, heavy metals with high attention can be divided into three categories: 1) Macro elements, such as Co and Mg; 2) Micronutrients, such as Fe, Cu, Mn, etc.; 3) Highly toxic elements, such as Pb, Hg, Zn, etc. [36]. Macronutrients and micronutrients, also known as essential nutrients, play a positive role in the growth, development and reproduction of plants, but have a negative effect when their content exceeds the plants growth need [37]. For highly toxic elements, harmful effects may occur even at low concentrations [38].

According to the survey on soil pollution in China in 2014, the point over-standard rates of eight major pollutants were 7.0%, 4.8%, 2.7%, 2.1%, 1.6%, 1.5%, 1.1%, and 0.9%, respectively [39]. Many studies have been carried out on the phenomenon of heavy metals exceeding the standard. Table 2 below lists China and its different regions and different average of soil heavy metal content in the national survey, found in a number of findings, there will be various in different areas of the phenomenon of heavy metal content is higher than that of soil background value. So, how to solve the problem of soil heavy metal pollution has attracted the attention of relevant scholars. At the same time, heavy metals can exist in various forms in soil. Tessier et al. divided heavy metals into five different forms such as exchangeable state and residual state [40], and different forms will also transform each other under different environmental conditions, so causing different toxic effects. Different from other pollutants, heavy metals possess stable chemical properties, are difficult to decompose and have poor biodegradability. They can accumulate in soil for a long time, with significant bio-concentration effect and a wide range of pollution [41].

Table 2. Comparison of soil heavy metal content in different regions of China and other countries.

Area	Sample size	Heavy metal content/(mg·kg ⁻¹)								References
		Cd	Ni	As	Cu	Hg	Pb	Cr	Zn	
Nationwide	368	0.24	28.17	10.71	28.34	0.13	32.07	62.18	83.29	[42]
Wushui River	49	1.28	68.32	72.44	54.62	0.27	72.29	-	158.42	[43]
Bohai Rim	185	0.42	25.3	9.18	27.72	0.13	25.07	57.35	71.70	[44]
Chongqing	1664	0.34	35.57	6.30	27.08	0.08	28.06	75.89	88.53	[45]
Yining City	51	-	35.2	0.128	31.3	0.037	61.6	33.2	61.33	[46]
UK	2604	0.6	35	-	43	-	50	113	147	[47]
Europe	23000	0.09	18.36	3.72	13.01	0.04	15.3	21.72	-	[48]

Notes: “-” represent not available, the same below

Table 3. Methods for analysis of heavy metals in soil.

Number	Test indicators	Analysis methods	Abbreviations
1	Pb	Emission spectroscopy	ES
2	Hg	Cold vapour-atomic fluorescence spectrometry	CV-AFS
3	As	Hydride generation-Atomic fluorescence spectrometry	HG-AFS
4	Cu, Zn, Cr	Inductively Coupled Plasma-Atomic Emission Spectrometry	ICP-AES
5	Cd, Pb, Ni, Zn	Inductively Coupled Plasma Mass Spectrometry	ICP-MS
6	As, Cr, Cu, Ni, Pb, Zn	X Ray Fluorescence	XRF

Therefore, the treatment of heavy metal pollution is urgent.

Methods for Analysis of Heavy Metals in Soil

In the process of detecting heavy metal pollution in soil, spectral detection technology is commonly used. Spectral method can detect and analyze heavy metal

content in soil samples with high sensitivity. With the development of detection technology, Inductively Coupled Plasma Mass spectrometry (ICP-MS) has become the main method to measure the content of heavy metal elements [49-50]. In order to obtain global authoritative and comparable geochemical observation data, Chinese scientists developed a set of high-quality analysis methods by analyzing elements, oxides, and other indicators [51]. Among them, Table 3

Table 4. Application characteristics of biochar under different biomass raw materials and different preparation conditions.

Biomass feedstock	Preparation conditions	Soil texture	Remediation effect	References
Rice straw	-	Ultisol	Decreased Cu and Pb by 20-100% and 19-77%, respectively.	[55]
Rice straw	350-550°C 1h	Red clay	Cd and Pb decreased in soil and plant after applications of the biochar.	[56]
Wheat straw	350-550°C -	Clay	Soil extractable Cd decreased by 55-71% and Pb by 65-80%.	[57]
Maize straw	450°C 4h	Moisture soil	It has good passivation effect on Cd and Pb in soil.	[58]
Rice hull	500°C -	Sandy-loam	Application of biochar reduced Cd by 97%, Cu (90%), Pb (100%) and Zn (100%).	[59]
Rice hull	500°C -	Reed soil	It reduced the content of exchangeable Cd and effectively prevented the HM Cd from entering the plant.	[60]
Bamboo and rice straw	500°C 30min	Sandy loam	Rice straw biochar reduced the concentration of Cu and Pb in the shoots by 46 and 71%, bamboo biochar reduced concentration of Cd in the shoot by 49%.	[61]
Rice husks and pig dung	400°C 3h	-	The contents of bioavailable forms of four kinds of HM decreased.	[62]
Fruit biochar	500°C -	-	Significantly reduced total and bioavailable HM concentrations.	[63]
Sugarcane bagasse	450°C 4h	Sandy loam silty clay	Cd decreased by 62-76%, Pb decreased by 17-49% and Cu by 15-38% in shoots.	[64]
Vegetable wastes	200°C, 500°C 1h	sandy loam	Achieved Pb immobilisation of 87%.	[65]
Agriculture residues	500°C 4-5h	Loamy sand	Achieved reduction of expendable Pb and Cd by 28.68% and 85.14%, respectively.	[66]
Sludge and cotton stalk	650°C 90min	-	Effectively adsorbing and fixing Cr in soil and reduces its mobility and bioavailability.	[67]
Potato	300°C, 400°C and 600°C 6h	Black soil and meadow soil	The passivation effects of Cd in the two soils were obvious.	[68]

is the analysis methods of heavy metal elements, which provides method guidance for laboratory sample analysis. It also lays a foundation for environmental assessment and continuous monitoring of future environmental changes.

The Application and Methods of Biochar Were Improved

Production, Characteristics and Application of Biochar

Biochar is a porous substance with high carbon content, which is produced by pyrolysis of waste biomass generated in agriculture and forestry at high temperature under aerobic or anaerobic conditions [25]. The raw materials for biochar preparation have a wide range of sources, such as sludge, sawdust, solid waste produced by industrial and agricultural activities, plant matter straw, fruit shell, and animal dung, etc. Biochar prepared from different biomass materials has a large difference in element content [52]. In addition, there are many methods to prepare biochar, including high-temperature pyrolysis, hydrothermal pyrolysis and flash pyrolysis. The yield and characteristics of biochar prepared by unique methods will also be affected [53]. As shown in Table 4, the application effects of some different biomass raw materials and biochar produced under different preparation conditions on the remediation of heavy metals in different types of soils are listed. In general, biochar can effectively reduce or fix the heavy metals in soil, and has a good application effect.

The surface properties of biochar can be analyzed from both physical and chemical perspectives [53-54], whose properties are mainly influenced by carbonization temperature, carbonization method and carbonization material. The porosity and specific surface area of the biochar obtained from the carbonization of biomass are higher than those of the original biomass and increase with the increase of pyrolysis temperature, but too high pyrolysis temperature will have the opposite effect. Compared with hydrothermal carbonization, the temperature of pyrolytic carbonization is higher, and the pore structure of pyrolytic carbon is richer and the specific surface area is larger. The surface morphology and pore size of biochar can be understood by electron microscope energy spectrum analysis. In addition, the higher the pyrolysis temperature, the less the surface functional group content of biochar, so the surface functional group under pyrolytic carbonization is lower than that under hydrothermal carbonization. Ash can provide mineral nutrients, and its content is affected by pyrolysis temperature and biomass materials. The higher the temperature, the higher the content and the content of sludge and feces is significantly higher than that of ordinary woody biological carbon. Through the analysis of X-ray diffraction technology, it is also found

that the crystallinity of crystal minerals on the surface of biochar at high temperature increased. Although some mineral crystals were lost, the amount and type of crystal crystals were improved.

In recent years, as an efficient and eco-friendly stable solid material, biochar has been widely used in soil improvement, carbon sequestration and heavy metal pollution remediation because of its unique physical structure and chemical properties [69]. which provides reference value for solving problems such as environmental protection, soil remediation and climate change to a certain extent.

Biochar Remediation of Heavy Metals in Soil and Its Improvement

Biochar as an important functional material has well-developed pore structure and rich surface functional groups, which makes it have strong adsorption capacity for heavy metals. It can effectively fix toxic heavy metals of soil in biochar, thus changing the form of heavy metals and improving the safety of soil use [70]. Studies have shown that adding rice straw biochar to soil contaminated by Cd and Pb can increase soil pH value and promote the transformation of Cd and Pb forms [71]. After using biochar in the remediation experiment of heavy metal contaminated soil, the solubility of Cu, Cd and Pb in the soil was reduced and the utilization of nutrient elements in plants was improved, hence the toxicity of heavy metals to plants reduced [72]. Wang et al. [67] found that Cr in soil could be effectively fixed by Sludge based biochar and the concentration of Cr would also change with the amount of sludge added. Some researchers [73] equally found that Sludge based biochar can effectively adsorb and fix heavy metal ions such as Pb, Mn, Cu, Zn and Cd in soil. Rizwan et al. [74] also found that biochar had a more significant passivation effect on soil heavy metals over time. Therefore, the application of biochar in soil can significantly affect soil environment and heavy metal toxicity, so it is a potential remediation technology for heavy metals in soil.

A large number of experiments show that biochar exerts remarkable effect on the adsorption of heavy metals, but the adsorption capacity of biochar obtained by the traditional preparation method is limited. Modified biochar by physical, chemical and biological methods can achieve stronger adsorption performance than the original biochar and improve the utilization rate of biochar [75]. Physical modification mainly changes the specific surface area, porosity, ash content and other physical structures of biochar through gas, in vitro, heat, ultrasound and electrochemistry [76]. Chemical modification is to include chemical agents to the soil to affect the surface functional groups of biochar, in which carboxyl and hydroxyl groups can significantly improve the adsorption efficiency of biochar for heavy metals [77]. Furthermore, the application of heteroatom oxides will also increase the specific surface area of biochar,

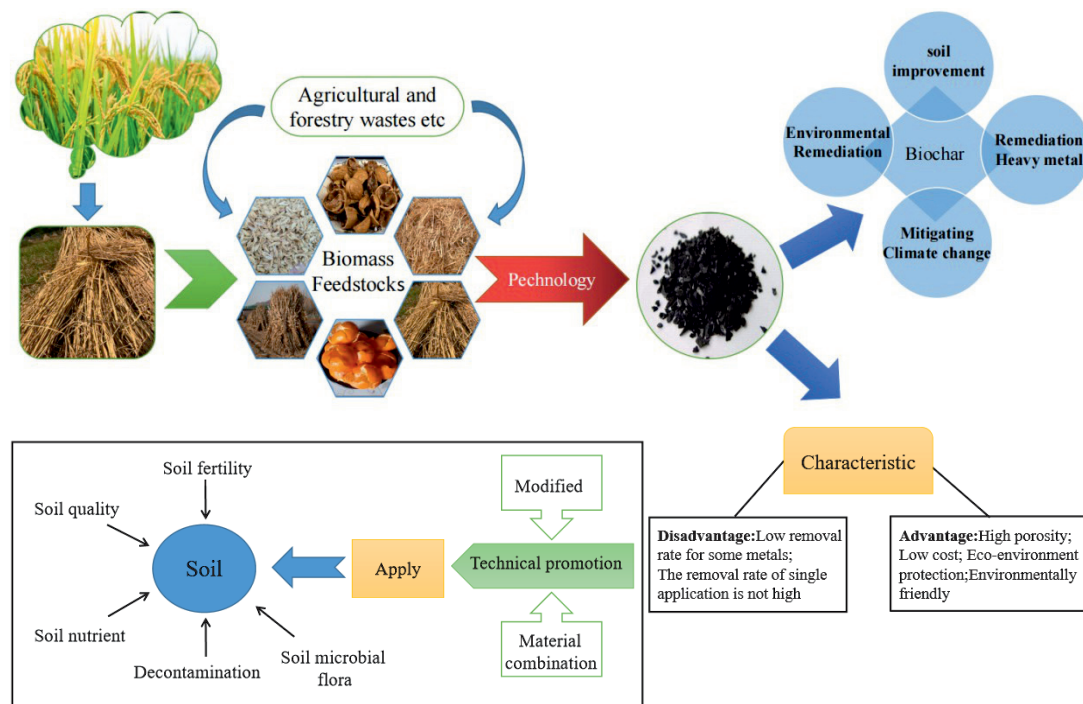


Fig. 1. Biochar characteristics, promotion methods and applications of biomass from agricultural and forestry wastes.

thereby improving its adsorption effect [78]. Biological modification is mainly through bioengineering [79] or inoculate microorganisms that can degrade pollutants on the surface of biochar [80] to promote the activity and richness of microbial communities in soils. In addition, the combined remediation technology is also an effective means to improve the adsorption performance of biochar, as shown in Fig. 1. This technology is also widely adopted in soil and has a good development prospect, so the research on the combined application of biochar and other materials should be strengthened.

Combined Application Technology of Biochar

Biochar can be combined with other materials in the remediation of soil heavy metals to obtain high binding sites and microbial activity to reduce the mobility and bioavailability of heavy metals. The existing joint remediation research has achieved many commendable achievements, such as the application of composite materials formed by the combination of biochar and degradants or microorganisms. The degradation products are rich in nutrients, which can provide microorganisms with substances needed for growth, improve the richness and activity of microbial population. After the addition of degradation products, there are positive effects on soil respiration and bacterial density, and the degradation rate of heavy metals is improved. Therefore, the combination of degradation products and biochar can

better degrade and remove heavy metals. Another way to fix and degrade heavy metals is add microorganisms to biochar to improve the remediation efficiency of biochar through bio-enhancement methods.

Repair Process and Mechanism

Bioremediation technology mainly uses biological enhancement to absorb, catalyze and degrade heavy metal pollutants in soil, and control and reduce the concentration and toxicity of heavy metals in soil. The organisms mentioned here include microorganisms and plants. Generally speaking, in terms of biological applications, bioremediation technologies can be divided into three categories [81]. Phytoremediation technology, Microbial remediation technology and Combined remediation technology. Phytoremediation technology mainly uses plants that can grow on heavy metal contaminated soil to absorb, fix and degrade heavy metals to purify the soil. Microbial remediation technology is mainly to add a large number of microorganisms to the soil to improve the diversity and activity of microbial communities. And it can repair the soil contaminated by heavy metals through adsorption, enrichment, degradation and dissolution. The use of combined remediation method is mainly because a single repair method is difficult to achieve the ideal repair effect. The use of plant and microorganism combined remediation methods can provide active play to the advantages of the two technologies and work together to achieve double effects.

Microorganisms are tiny organisms that can accumulate heavy metals in their bodies. They act on heavy metals in various mechanisms according to their own characteristics, thus alleviating or reducing the toxicity of heavy metals to avoid stronger toxicity caused by heavy metals. Various microorganisms such as bacteria, fungi and algae are implanted into biochar, and the heavy metals are absorbed and stored by the microbes after the accumulation process in the active

microbial community. Meanwhile, a variety of negatively charged functional groups form the cell wall of microorganisms. And the surface of negatively charged microorganisms can absorb positively charged metal ions in the soil [82] to achieve the effect of removing heavy metals in the soil. Table 5 lists the application mechanism and effect of some microorganisms in heavy metal remediation. Some scholars [83] also found that under the action of different charges, the interaction

Table 5. Comparison between microbial assisted biochar remediation and single remediation.

Metal	Technique	Mechanism	Remediation effect	References
Cd	Bacteria: Shewanella putrefaciens; Pseudomonas sp.	Cell wall adsorption, Intracellular accumulation	The maximum removal efficiency was 86.54%; Effective removal of Cd ²⁺ .	[93, 94]
	Fungi: Penicillium notatum; Absidia cylindrospora; Chaetomium atrobrunneum	Ion exchange, transportati-on across cell membrane	Highest biosorption rate for Cd at 10 ppm with 77.67%; Biosorbed more than 45% of Cd.	[95, 96]
	Algae: Asparagopsis armata, Codium vermilara et; Chlorella vulgaris	Alginate acid and its derivative	Fucus spiralis has the best adsorption effect; The highest cadmium biosorption efficiency was 76.448%.	[97, 98]
	Bacteria+Biochar: Pseudomonas sp.+Biochar; Bacillus subtilis+Corn straw and pig manure biochar	-	Decreased the bioavailability of Cd ²⁺ from 29.5 to 20.8 mg/kg; Concentrations in the edible part of lettuce decreased by 70-96%.	[79, 80, 99]
	Fungi+Biochar: White-rot fungi+biochar	-	The content of available Cd is effectively reduced (44.45%).	[100]
Pb	Bacteria: Enterobacter cloacae; Curtobacterium sp.	Enterobacter adsorption and intracellular accumulation	High removal rates (ca. 60%) of Pb; The maximum biosorption capacity to be 186.60 for Pb (II).	[101, 102]
	Fungi: Aspergillus niger; Phanerochaete chrysosporium	Sequestrations, metal detoxi-fication using organic acid production	It can remove copper (II) with a maximum specific uptake capacity of 15.6 mg/g; The maximum heavy metal ions adsorbed was 135.3 mg/g.	[103]
	Algae: Enteromorpha ; Gelidium amansii	Adsorption on cell wall surface, protein interaction	Adsorption capacity was 83.8 mg/g at pH 3.0; Effective in Pb ²⁺ removal.	[104, 105]
	Bacteria+Biochar: Bacillus subtilis+Corn straw and pig manure biochar	-	Concentrations in the edible part of lettuce decreased by 70-96%.	[99, 100]
	Fungi+Biochar: Arbuscular Mycorrhizal Fungi+Dry Olive Residue- Based biochar	-	AMF inoculation significantly decreased the mobile proportions of Pb.	[25]
Ni	Bacteria: <i>Curtobacterium</i> sp.; <i>Pseudomonas aeruginosa</i>	Ion exchange	The maximum biosorption capacity was 140.99 mg/g for Ni (II); Highest Ni uptake (with a maximum K _d of 1890 L/kg DW).	[103, 106]
	Fungi: <i>Umbelopsis isabellina</i> ; <i>Phanerochaete chrysosporium</i>	Adsorption	The highest inhibition of Ni (II) removal with 4-t-OP was 73.3 %; Maximum biosorption capacity of 46.50 mg/g for Ni ²⁺ .	[107, 108]
	Algae: <i>Cystoseria indica</i> ; <i>Euglena gracilis</i>	Intracellular accumulation	The maximum biosorption of Ni in binary- metal- component system was 18.17; Inhibit the accumulation of Ni.	[109, 110]
	Bacteria+Biochar: FD-17,KS-54, PsJN+ Biochar; Bacillus subtilis+Straw and sorghum biochar	-	When the biochar content was 2%, the content of Ni decreased by 40%; It has a good curing effect on Ni.	[92, 111]
	Fungi+Biochar: -	-	-	-

Table 5. Continued.

Cu	Bacteria: <i>Acinetobacter</i> sp. MA9	Adsorption	The removal of copper by bacteria was up to 68%.	[112]
	Fungi: <i>A. flavus</i> ; <i>Penicillium ochrochloron</i>	Adsorption, Intracellular accumulation by transport across cell membrane	The biosorption capacity for Cu (II) was 20.75-93.65 mg/g; Average biosorption capacity of Cu (II) was 7.53 mg/g and the maximum Cu (II) removal 75.0%.	[113, 114]
	Algae: <i>Chlorella</i> sp.; <i>Gelidiella acerosa</i>	Adsorption	The biosorption of <i>Chlorella</i> sp. for zinc ions was 28.5 mg/g; Maximum copper biosorption potential of 96.36%.	[115, 116]
	Bacteria+Biochar: NT-2+Biochar; <i>Bacillus subtilis</i> +Straw and sorghum biochar	-	Decreased the bioavailability of Cu ²⁺ from 127.3 to 73.4 mg/kg; It has a good curing effect on Cd.	[80, 92]
	Fungi+Biochar: <i>Ganoderma lucidum</i> fungus +Straw and sorghum biochar	-	It has a good curing effect on Cu.	[92]
As	Bacteria: Rhizobacteria; <i>Pseudomonas aeruginosa</i>	Nitrite-driven Fe (II) oxidation	<i>S. paucimobilis</i> showed the highest As biosorption capability (146.4±23.4 mg/g dry cell weight); Efficiently remove As with 98% efficiency.	[117, 118]
	Fungi: <i>Talaromyces</i> sp.; <i>Aspergillus</i> spp.	Ion exchange	The fungus was highly resistant to As, tolerating concentrations up to 1000 mg/L; <i>Aspergillus</i> spp APR-1 and APR-2 showed biosorption of 53.94 and 52.54%, respectively.	[119, 120]
	Algae: <i>Chlorellacoloniales</i> ; <i>Sarcodia suaie</i>	-	Can effectively remove As; Maximum absorption was obtained at 15°C.	[121, 122]
	Bacteria+Biochar: <i>Penicillium</i> +biochar	-	It can reduce the content of available As and improve the microbial environment in contaminated soil.	[123]
	Fungi+Biochar: AM+Wheat straw biochar	-	Two kinds of combination had the best effect on reducing the content of As in maize and the content of soil available As.	[124]
Zn	Bacteria: <i>Variovorax paradoxus</i> and <i>Arthrobacter viscosus</i> ; <i>Streptomyces</i> sp.	Intra-particle diffusion	The maximum removal efficiency of <i>A. viscosus</i> live and dead cells was 89.4 and 90.8%; The maximum biosorption capacity was 0.75 mmol/g.	[125, 126]
	Fungi: <i>Neopestalotis clavisporea</i> ; <i>Umbelopsis isabellina</i>	Adsorption	Maximum adsorption for Zn was 153.8±0.21 mg/g; The degradation of Zn was enhanced by 9%.	[127, 111]
	Algae: <i>Kappaphycus alvarezii</i> ; <i>Fucus vesiculosus</i>	External diffusion using ion exchange	Theoretical absorption capacities for metal oxide nanoparticles were 80% for ZnO; Relative to Zn, alginate is one of the main algae components responsible for metal binding.	[128, 129]
	Bacteria+Biochar: <i>Bacillus subtilis</i> and <i>ganoderma lucidum</i> fungus+biochar	-	It has a good curing effect on Zn.	[92]
	Fungi+Biochar: White-rot fungi+biochar	-	The content of available Zn is effectively reduced (43.22%).	[101]
Cr	Bacteria+Biochar: <i>Bacillus pasteurii</i>	Mixed adsorption and reduction	Soil leaching concentration and the amount of Cr (VI) in the soil decreased from 90 mg/L, 270 mg/kg to 1.08 mg/L, 5.14 mg/kg, respectively, and the remediation effect was the best.	[91]

between multiple combinations is more significant. Bioaccumulation is divided into multiple mechanisms, and in the repair of heavy metal contaminated soil, can accept somewhat bioaccumulation mechanism of microorganisms. Because of their tolerance to heavy metals, they are considered as ideal remediation species. However, such remediation mechanism fails to realize engineering, and microorganisms with multiple mechanisms still need to be combined to strengthen the study on the mechanism and characteristics of mixed culture. Bacterial remediation, fungal remediation and microalgae remediation

With the development of bioremediation technology, the research on metal remediation by bacteria, fungi and algae has attracted extensive attention in the field of environmental treatment. As the most common and effective method of bioremediation, bacterial remediation is mainly due to the large number and wide area of bacteria in nature, as well as a lot of bacteria in soil. These bacteria play their respective functions and jointly maintain a good soil ecological environment [84]. Gram-positive bacteria, as the largest bacterial phylum [85], are widely used in the removal of heavy metals. The study of Chang et al. found that the use of biochar containing *Pseudomonas* sp. DC-B1 and *Bacillus* sp. DC-B2 in pollution control can significantly reduce the content of Hg^{2+} in soil [86]. Generally, the bacteria used in remediation are taken from the soil heavily polluted by heavy metals. The extracted bacteria are purified and cultured after differentiation, and finally applied to the remediation of heavy metals.

The fungi used in fungal remediation technology are usually derived from plant roots. These fungi have strong biological activity and have a certain tolerance and enrichment ability to metals. In addition, they can form mycelium with plant roots to accumulate heavy metals and reduce the toxicity of heavy metals to plants. For example, Lu and other studies show that arbuscular mycorrhiza can reduce the content of heavy metals in soil and purify the soil environment [87]. Fungal remediation is an economic and effective biotechnology with a wide variety of fungi. The cultivation of dominant strains should be strengthened to realize the large-scale and market-oriented application of fungal remediation.

Microalgae remediation technology is currently mainly applied in wastewater treatment. Since the 1990s, some countries have started the application of microalgae in the treatment of heavy metal polluted water and has achieved some results, but its application in soil contaminated by heavy metals has been still immature [88]. It has been reported in some studies that microalgae, like bacteria, contain a large number of negatively charged functional groups on their surfaces that act as metal binding sites to attract positively charged metal ions [89]. Different from other microorganisms, microalgae need photosynthesis. Therefore, in remediation of heavy metal contaminated soil, microalgae are mainly used in rice field research.

Study on Microbial Assisted Biochar Restoration

In order to exert the remediation ability of biochar to a greater extent and solve the problem of single remediation effect, in recent years, some scholars found that the remediation effect of biochar on heavy metal contaminated soil was significantly improved after inoculating microorganisms into the soil. Table 5 compares the application of microbial assisted biochar remediation method and single remediation method. In general, compared with single application, the application effect of biochar composite materials in heavy metal remediation is more significant. In addition, adding microorganisms directly to the contaminated soil will cause the loss and phagocytosis of microorganisms and affect the remediation effect. Therefore, Wang chose to attach the test microorganism *Bacillus subtilis* to the biochar first, and then add it to the heavy metal contaminated soil [90]. At the same time, Xia et al. [91] studied the remediation effects of single *Bacillus Pasteurella* remediation, biochar remediation and the combination of *Bacillus Pasteurella* and biochar on chromium contaminated soil. The results show that compared with the single remediation of microorganism and biochar, microbial assisted biochar has the best remediation effect. In another study [92], two strains (*Bacillus subtilis* and *Ganoderma lucidum* fungi.) were selected to construct a microbial/biochar pot experiment to investigate the changes of soil physical and chemical properties, respiration intensity and heavy metal forms in the soil under the system.

The results showed the soil pH and organic matter were significantly reduced, and the microbial/biochar can inhibit the bioavailability of the heavy metals Cr, Ni, Zn and Cu in the soil and fungi have a greater effect on the degradation of organic matter than bacteria. In the soil polluted by Ni, Cd and Pb, Ejaz selected three different endophytic fungi FD-17, KS-54 and PsJN and applied biochar at the rate of 1% and 2%. When the application rate was 2%, the contents of heavy metals Ni, Cd and Pb showed a significant negative trend compared with the control under the combined treatment of FD-17 and KS-54 [111].

Although biological enhancement technology has shown great application potential in the remediation of heavy metal contaminated soil, the application of present technologies also display some limitations. For example, if exogenous microorganisms are put into use, it will produce nutrient competition with local microorganisms or trigger local microorganisms to prey on exogenous microorganisms. Therefore, the application of exogenous microorganisms to contaminated soil remediation may cause some problems, so it is a challenging problem to select appropriate microbial populations and strains. In addition, effective bioremediation technology should be adopted for specific pollution adjust measures to local conditions according to different pollution conditions and operating conditions.

Conclusions and Future Prospects

In conclusion, as a valuable soil conditioner, biochar has achieved remarkable achievements in the remediation of heavy metal pollution. Compared with traditional remediation methods, biochar, as an economical and environmental-friendly material, can restore heavy metals in soil, sequester carbon and improve soil quality. The removal rate and bioavailability of biochar for heavy metals in contaminated soil remediation mainly depend on the properties of biochar and heavy metals. However, the properties of biochar are affected by the raw materials, pyrolysis conditions and carbonization methods used in the preparation of biochar. In addition, biochar can be modified by specific engineering to improve its remediation effect. Overall, the application and efficacy of biochar were reviewed and introduces the mechanism and specific application of combined remediation. Although the research and results of biochar in the field of contaminated soil treatment have been widely reported, the above-mentioned joint auxiliary remediation method and remediation of mixed multi-metal pollution need to be further studied.

Moreover, biochar has become an ideal method due to its advantages of low cost, easy operation, high efficiency and environmental friendliness. However, there are still some limitations in the current research, which should be further deepened from the following aspects:

(1) At present, most of the experiments mainly focus on the remediation of single metal in the indoor scale. Field experiments and the remediation of multi-metal composite contaminated soil should be strengthened.

(2) There are deficiencies in the long-term impact of biochar application on soil environment, the long-term effects of biochar application amount and application frequency, and the understanding of heavy metal fixation efficiency and mechanism of aging biochar are still insufficient. These aspects still need our further understanding and research.

(3) There are significant differences in the characteristics of biochar prepared by different biomass raw materials and production processes. According to the characteristics of soil pollutants, method upgrades or production system optimization should be carried out to produce high-quality biochar with required characteristics.

(4) In the combined remediation application of biochar and microorganisms, the selection and supply of microorganisms and their flora that can effectively degrade heavy metals may be a challenge. In addition, the impact of exogenous microorganisms on the diversity and activity of local microorganisms needs to be considered.

(5) Whether the used biochar and biochar-based composite materials can be recycled needs to evaluate their properties and sustainability after regeneration in order to achieve sustainable utilization.

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Conflict of Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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