

Supplementary Material

Materials and methods

Metal content and speciation analysis

Total metal concentrations in the biochars were determined by an inductively coupled plasma atomic emission spectrometer (ICP-AES) after the digestion in HF-HClO₄-HNO₃.

Sequential extraction was carried out via the five-step sequential extraction procedure, including the exchangeable, bound to carbonates, bound to Fe/Mn oxides, bound to OM and sulfides, and residual fractions (Lin et al. 2003). Details are as follows.

–Exchangeable: One gram of biochar sample extracted with 8 mL of 0.5 mol·L⁻¹ MgCl₂ (pH 7) for 20 min, with continuous agitation at room temperature;

–Bound to carbonates or specifically adsorbed: the residue from the exchangeable fraction was extracted with 8 mL of 1 mol·L⁻¹ NaAc (pH 5) for 5 h, with continuous agitation at room temperature;

–Bound to Fe/Mn oxides: the residue from the carbonate or specifically adsorbed fraction was extracted with 20 mL of 0.04 mol·L⁻¹ NH₂OH/ HCl in 25% HAc for 6 h, with occasional agitation at 96 °C; After the extraction, the extract solution was diluted to 20 ml with deionized water (DIW) and subjected to continuous agitation for 10 min;

–Bound to OM and sulfide: the residue from Fe-Mn oxide fraction was extracted with 3 mL of 0.02 mol·L⁻¹ HNO₃ and 5 mL of 30% H₂O₂ (pH 2); the sample was heated progressively to 85 °C and maintained at this temperature for 2h with occasional agitation; then, 3 ml of 30% H₂O₂ was added, and mixture was heated to 85 °C for 3 h with intermittent agitation; after cooling, 5 mL of 3.2 mol·L⁻¹ CH₃COONH₄ were added, by diluting to a final volume of 20 mL with DIW, with the samples continuously agitated for 30 min;

–Residual: the residue from OM and sulfide fraction was digested with HNO₃-HCl-HClO₄.

After each extraction, separation was performed by centrifugation at 10000 rpm for 30 min. The metal concentrations in filtrate were determined by flame atomic absorption spectroscopy (MKIIM6, Thermo Electron, America).

Synthetic precipitation leaching procedure (SPLP) and Toxicity Characteristic Leaching Procedure (TCLP)

All of the samples (manure samples and biochar samples) were leached using the SPLP (synthetic precipitation leaching procedure, EPA Method 1312) and TCLP (Toxicity Characteristic Leaching Procedure, EPA Method 1311). The SPLP extraction solution (pH of 4.20±0.05) was designed to represent acidic rainwater. The extraction solution was prepared by adding a dilute sulfuric acid and nitric acid solution (60:40 mix) to a 2 L container and diluting to volume (2 L) with reagent water. Two grams of the sample was placed in a 150 mL conical flask, and 40 mL of the extraction solution was added. The slurry was mixed on horizontal vibration extractor at 110 ± 10 rpm for 18 ± 2 h and then filtered through a 0.45 µm filter. The filtrate was collected in 20 mL test tubes and preserved by adjusting to a pH of less than 2 using nitric acid. The metal concentrations in filtrate were measured using the AAS (MKIIM6, Thermo Electron, America). Every sample of the SPLP was carried out in triplicate.

The TCLP extraction solution was prepared by adding 11.4 mL of glacial acetic acid (CH₃COOH) to a 2 L volumetric flask, and the solution was brought to volume (2 L) with deionized water. The resulting solution pH was 2.88 ± 0.05. Two grams of the sample was placed in a 150 mL conical flask, and 40 mL of the extraction solution was added. The slurry was mixed on horizontal vibration extractor at 110±10 rpm for 18±2 h and then filtered through a 0.45 µm glass fiber filter. The filtrate was collected in 20 mL test tubes and preserved by adjusting to a pH of less than 2 using nitric acid. The metal concentrations in filtrate were measured using the AAS (MKIIM6, Thermo Electron, America). Every sample of the SPLP was carried out in triplicate.

Simple bioaccessibility extraction test (SBET)

The experiments were conducted at 37°C. A 0.4 mol·L⁻¹ glycine solution was prepared by mixing 30 g of 98% glycine in 1 L of reagent water. The pH of the solution was lowered to 1.5 using hydrochloric acid. This solution was designed to simulate the gastric fluid in a child's digestive system. Two grams of sample was placed in a 500 mL conical flask, and 200 mL of the extraction solution was added. The samples were extracted for 1 h on a horizontal vibration extractor at 110±10 rpm. During the extraction period, solution pH was monitored and maintained at approximately 1.5. After extraction, the sample was filtered using 0.45 µm filter. The metal concentrations in filtrate were measured using the AAS (MKIIM6, Thermo Electron, America). Every sample of the SBET was carried out in triplicate. Metal bioaccessibility was calculated by dividing the metal concentration (µg·g⁻¹) measured in the in vitro gastric solution by the total soil metal concentration (Equation 1).

$$\text{Metal bioaccessibility (\%)} = \text{In vitro metal} / \text{Total metal} \times 100 \quad (1)$$

Active pool for Cu and Zn

The active content of Cu and Zn were determined in duplicate after extraction with 0.43 mol·L⁻¹ HNO₃ solution. Five grams of the sample was placed in a 150 mL conical flask, and 50 mL of the extraction solution was added. The slurry was mixed on horizontal vibration extractor at 110±10 rpm for 2 h at room temperature and filtered through a 0.45 µm glass fiber filter. The metal concentrations in filtrate were measured using the AAS (MKIIM6, Thermo Electron, America).

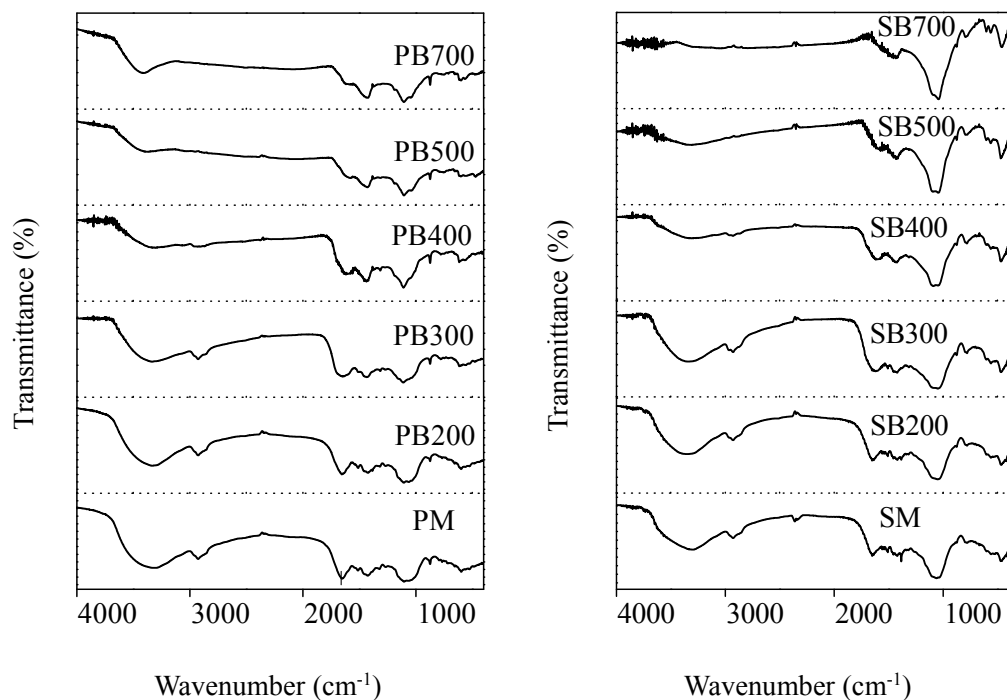


Fig. S1 The FTIR spectra of feedstocks and biochars. Poultry manure (PM), poultry manure derived-biochar (PB200, 300, 400, 500, 700), swine manure (SM) and swine manure derived-biochar (SB200, 300, 400, 500, 700)

Table S1 pH and concentrations of Cu and Zn in feedstocks and biochars

Sample ID ^a	Cu (mg/kg)	Zn (mg/kg)	pH
PM	184.31±6.43	321.85±4.82	6.97±0.02
PB200	188.04±4.06	331.85±4.28	6.45±0.01
PB300	248.97±11.41	448.65±21.68	6.85±0.01
PB400	312.84±5.23	565.72±10.83	8.28±0.04
PB500	370.97±7.26	664.39±9.45	8.90±0.08
PB700	398.17±8.22	633.19±14.00	9.54±0.12
SM	613.37±19.17	1044.25±26.86	6.99±0.09
SB200	628.31±35.29	1121.59±49.08	6.85±0.02
SB300	754.97±15.75	1235.59±25.48	7.60±0.31
SB400	881.51±29.51	1450.25±71.62	8.68±0.55
SB500	956.97±14.81	1619.59±28.10	9.29±0.18
SB700	1033.64±80.81	1598.92±117.85	9.98±0.44

Note: ^aPoultry manure (PM), Swine manure (SM), Poultry manure-derived biochar prepared under pyrolysis temperature of 200°C (P200), and so on

Table S2 Correlation coefficient of metal concentrations in the source material and biochars based on the data from XRF line-by-line scanning

	K	Ca	Mn	Fe	As	Zn
Swine manure (SM) (n=576)						
As	0.417	0.101	0.557	0.433		
Cu	0.694	0.236	0.498	0.392	0.509	0.893
Zn	0.698	0.317	0.619	0.536	0.576	
SB500 <63 μ m (n=576)						
As	0.452	0.164	0.586	0.449		
Cu	0.377	0.312	0.544	0.469	0.436	0.824
Zn	0.272	0.383	0.484	0.261	0.357	
SB500 63-800 μ m (n=576)						
As	0.399	-0.026	0.609	0.302		
Cu	0.624	0.258	0.473	0.621	0.233	0.811
Zn	0.643	0.281	0.624	0.736	0.214	
SB500 >800 μ m (n=576)						
As	0.590	0.239	0.414	0.437		
Cu	0.662	-0.025	0.213	0.736	0.566	0.629
Zn	0.715	0.684	0.852	0.567	0.583	