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Effects of bagasse biochar application on soil organic carbon fixation in manganese-contaminated sugarcane fields



Lening Hu¹, Yu Yang¹, Xue Hui Liu¹, SHuangli Li¹, Ke Li^{2*} and Hua Deng¹

Abstract

Background In recent years, there have been several studies on the remediation of heavy metal pollution in soil by the application of biochar. However, little attention has been paid to understanding the effects and underlying mechanisms of biochar on soil carbon sequestration in manganese-contaminated farmlands. Therefore, in this study, bagasse biochar was applied to the soil in different proportions (0%, 0.5%, 2%, and 5%) and the test was conducted indoors for 100 days at a constant temperature. Soil physical and chemical properties, organic carbon mineralization, organic carbon components, and enzyme activities were analyzed in this study.

Results In this study, when compared with the control, the application of 0.5%, 2%, and 5% bagasse biochar to the manganese-contaminated sugarcane field soil effectively reduced the cumulative CO_2 emissions, i.e., decreased by 123.18 mg·kg⁻¹, 208.28 mg·kg⁻¹, and 287.79 mg·kg⁻¹, respectively. Among the different treatment groups, the highest decrease in cumulative CO_2 emissions was observed in the 5% bagasse biochar-treated soil when compared with the control. The application of bagasse biochar increased the soil microbial biomass carbon content by 12.72 mg·kg⁻¹, 13.71 mg·kg⁻¹, and 15.10 mg·kg⁻¹, respectively when compared with the control. The soil nutrients and enzyme activities significantly increased with the increase in biochar application amount.

Conclusions The application of bagasse biochar to manganese-contaminated sugarcane soil field effectively inhibited the mineralization of soil organic carbon, improved the carbon sequestration potential of manganese-contaminated sugarcane field soil, and provided a theoretical basis for the carbon sequestration mechanism in manganese-contaminated farmland soil.

Keywords Biochar, Mineralization, Soil enzymes, Soil nutrient, Organic carbon fraction

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Introduction

Farmland carbon storage accounts for about 8-10% of terrestrial carbon storage, which forms an active part of the global carbon pool. This pool is easily affected by human activities, which alters the farmland carbon pool, which in turn can influence climate change [1]. Traditionally, China is an agrarian country, and agriculture is an important source of greenhouse gases. The CO₂, CH₄, and N₂O emissions from agricultural sources are 310 million tons, 9.685 million tons, and 665,000 tons, respectively [2]. According to the statistics, soil heavy metal pollution is a critical issue in China, with about 10 million hectares of farmland soil affected by heavy metal pollution [3]. Heavy metal pollution may affect CO_2 emissions. Bian et al. [4] observed that heavy metal pollution significantly increased soil CO₂ emission rate in heavy metalcontaminated farmland. Zhou et al. [5] studied heavy metal-contaminated soil and revealed that heavy metal pollution can reduce CO₂ emissions. The processes by which heavy metal pollution affects soil CO₂ emissions are still debatable. Therefore, it is of great significance to explore the mechanism underlying carbon sequestration in heavy metal-contaminated farmland soils.

Sugarcane is an important raw material for sugar production. According to the statistics, the total output of sugarcane in southern China is more than 70 million tons, with the annual production of bagasse of about 20 million tons [6]. At present, most of the bagasse in China is used for power generation and paper manufacturing. The utilization rate of bagasse is low, which not only leads to the wastage of resources but also leads to environmental pollution. The mass fraction of bagasse cellulose is about 43.80%, with a carbon content of about 44.17%, and bagasse cellulose is an excellent raw material for biochar preparation [7]. Biochar is a carbon-rich solid with a developed pore structure, high aromatization, and large specific surface area, which is obtained by the pyrolysis of animal and plant residues at high temperatures and oxygen-limiting conditions (350-600 °C) [8]. When applied to the soil, it can improve soil structure, increase soil nutrients and carbon sequestration, and reduce greenhouse gas emissions [9]. The application of biochar in heavy metal-contaminated paddy fields reduced soil CO₂ emissions by 16-24% [10]. Wu et al. [11] performed a 6-year field experiment and demonstrated that the application of biochar reduced CH₄ and N₂O emissions in soil by 11.2-17.5% and 19.5-26.3%, respectively. The conversion of bagasse into biochar can enhance its carbon sequestration capacity. In recent years, several studies were conducted on the remediation of heavy metal pollution in soil by biochar [12, 13], but only a few studies elucidate the mechanism underlying carbon sequestration in manganese-contaminated soil. Therefore, it is extremely important to study the greenhouse gas emissions from contaminated farmlands and improve the carbon sequestration capacity of contaminated soil to alleviate the global greenhouse effect.

Therefore, the soil from manganese-contaminated sugarcane fields in Guangxi, China was selected as the research object in this study. The different proportions of exogenous bagasse biochar were applied to carry out a 100-day incubation experiment at room temperature. The effects of bagasse biochar on the physical and chemical properties, organic carbon mineralization, organic carbon components, and enzyme activities of manganese-contaminated farmland soil were analyzed to provide a conceptual framework for the carbon fixation mechanism of biochar in manganese-contaminated farmlands.

Materials and methods

Experimental materials

The soil samples were collected from uncontaminated sugarcane fields $(23^{\circ}20'57'' \text{ N}, 108^{\circ}51'41'' \text{ E})$ in Nanning, Guangxi, and manganese-contaminated sugarcane fields $(23^{\circ}56'47'' \text{ N}, 109^{\circ}16'27'' \text{ E})$ in Laibin, Guangxi. The selected soil is a typical acidic red soil in southern China. After the removal of the stones and plant roots, the soil samples were placed at room temperature and dried naturally. The samples were crushed with a mortar and passed through 2 mm nylon sieves. The sieved samples were then placed in sealed bags for use, and their chemical properties were analyzed. The basic chemical properties of the test soils are shown in Table 1.

Biochar was prepared by pyrolysis of bagasse in a muffle furnace at 500 $^{\circ}$ C for 2 h under oxygen-limiting conditions. The bagasse required for the preparation of biochar was collected from Laibin, Guangxi. The basic properties of biochar are shown in Table 2.

Experimental design

The research experiment was started in 2021 at the Key Laboratory of Ecology of Rare and Endangered Species and Environmental Protection (Guangxi Normal University), Ministry of Education, China. The experimental design included sugarcane field soil+0% biochar, sugarcane field soil+0.5% biochar, sugarcane field soil+2% biochar, sugarcane field soil+5% biochar, manganese-contaminated sugarcane field soil+0.5% biochar, manganese-contaminated sugarcane field soil+0.5% biochar, manganese-contaminated sugarcane field soil+2% biochar, and manganese-contaminated sugarcane field soil+2% biochar, and manganese-contaminated sugarcane field soil+2% biochar, and manganese-contaminated sugarcane field soil+5% biochar, field soil+5% biochar, and manganese-contaminated sugarcane field soil+5% biochar, field soil+5% biochar, biochar, field soil+5% biochar, and manganese-contaminated sugarcane field soil+5% biochar, field soil+2% biochar, and manganese-contaminated sugarcane field soil+2% biochar, field soil+5% biochar, field soil+5% biochar, field soil+2% biochar, and manganese-contaminated sugarcane field soil+5% biochar, f

	рН	Productivity (%)	C%	H%	H/C
Bagasse biochar	8.56	28.13	78.19	2.87	0.51

respectively. A total of 24 treatments were carried out with each treatment repeated three times.

The air-dried soil passed through a 2 mm sieve was taken into a 2 L polyethylene bottle, and biochar was applied according to the experimental design mentioned above. After mixing biochar with soil, deionized water was added, and the field water holding capacity was maintained at 40-60% by the weighing method. The soil was continuously cultured in a constant temperature incubator at 25 °C. 150 g of soil samples were collected and analyzed on days 1, 3, 5, 10, 15, 20, 30, 40, 60, 80, and 100. Another batch of soil samples was set up and subjected to the same conditions for 1 week, with 50 g of soil and the corresponding proportion of biochar. A 10 mL beaker filled with a certain concentration of sodium hydroxide solution (0.1 mol· L^{-1}) was placed into a white polyethylene bottle, and the CO₂ emissions were analyzed on days 1, 3, 5, 10, 15, 20, 30, 40, 60, 80, and 100.

Measuring indices

Soil pH was measured by a pH meter (water-soil ratio of 2.5:1) [14]. The cation exchange capacity (CEC) was determined by the ammonium acetate method [15]. Available phosphorus (AP) was determined by the NaHCO₃ extraction-molybdenum antimony colorimetric method [16]. Available potassium (AK) content was determined by the ammonium acetate extraction method using a flame photometer [17]. Microbial biomass carbon (MBC) was determined by the chloroform fumigation extraction method [18]. Soil organic carbon (SOC) was determined by the potassium dichromate volumetric method with external heating [19]. Dissolved organic carbon (DOC) was extracted using deionized water and analyzed with a TOC analyzer [20]. Readily oxidized organic carbon (ROC) was determined using potassium permanganate oxidation [21]. Catalase activity was determined by potassium permanganate titration [22]. Urease activity was determined by the

 Table 1
 Chemical properties of soil

Properties	рН	AP (mg⋅kg ⁻¹)	AK (mg⋅kg ⁻¹)	SOC (g·kg ⁻¹)	CEC (cmol·kg ⁻¹)	Mn (mg⋅kg ⁻¹)
Uncontaminated sugarcane soil	6.59	18.52	5.39	1.01	18.9	/
Manganese-contami- nated sugarcane soil	5.35	3.20	2.13	0.86	8.40	2075.13

AP available phosphorus, AK available potassium, SOC soil organic carbon, CEC Cation exchange capacity

Calculations

The equation [24] for CO₂ emissions is given as follows:

$$CO_{2} (mLKg^{-1}) = \left\{ \left[(V_{0} - V) \times c \times 0.022 \times (22.4/44) \times 1000 \right] \times 2 \times 1000 \right\} / m$$

In Eq. (1), V₀ is the volume of standard hydrochloric acid consumed during blank titration, V is the volume of standard hydrochloric acid consumed during sample titration, c is the concentration of standard hydrochloric acid, 0.022 is the molar mass of carbon dioxide (1/2 CO₂), M (1/2 CO₂)=0.022 g·mmol⁻¹, 22.4/44 is milliliters per gram of CO₂ under standard conditions, and m is the weight of soil CO₂ during incubation.

CO₂ release rate (mg·kg⁻¹·d⁻¹) = amount of organic carbon mineralized/ Δt , where Δt is the culture interval (d).

Cumulative soil CO_2 emissions were calculated as the total CO_2 emissions from the 1 day of culture to the day of measurement.

The soil carbon mineralization under different culture conditions was fitted by a first-order kinetic equation [18].

$$C_t = C_0 \left(1 - e^{-kt} \right) \tag{2}$$

In Eq. (2), C_t is the cumulative mineralization amount at culture time t (d), C_0 is the potential soil carbon mineralization (mg·kg⁻¹); k is the rate constant of soil carbon mineralization, d⁻¹, and t is the culture time, d. Soil organic carbon stability coefficient [25] is given by

Eq. (3).

Soil organic carbon stability coefficient =
$$(SOC - ROC)/SOC$$

Statistical Analyses

Excel 2016 and SPSS 22.0 (IBM Corporation, United States) were used for data analysis and processing. The relevant index data were expressed as mean±standard deviation, and the correlation diagram was drawn using Origin 2023 (OriginLab Corporation, United States).

Results

Effects of biochar on soil pH and CEC

The application of bagasse biochar increased the pH of both manganese-contaminated sugarcane field soil and uncontaminated sugarcane field soil (Fig. 1). pH also increased with the increase in biochar application amount, but the change was not obvious with time. The pH of the uncontaminated sugarcane field with 0.5% biochar decreased with the culture time, while there was no significant change in the pH of the 2% and 5%



Fig. 1 Effects of biochar on soil pH. (a) Uncontaminated sugarcane field soil. (b) Manganese-contaminated sugarcane field soil

(1)

(3)

biochar-treated soil with time. The pH of manganesecontaminated sugarcane field soil with 0.5% biochar initially decreased and then increased with time, and the pH of the manganese-contaminated sugarcane field soil with 2% and 5% biochar decreased with time. The pH of the uncontaminated sugarcane field soil was higher than that of the manganese-contaminated sugarcane field soil. Biochar application had little effect on the pH of the manganese-contaminated sugarcane field soil.

As shown in Fig. 2, the CEC of uncontaminated sugarcane field soil during the entire incubation period was 3.56-61.01 cmol·kg⁻¹, and the CEC of manganese-contaminated sugarcane field soil was 3.62-49.13 cmol·kg⁻¹. Compared with that of the control, the CEC of uncontaminated sugarcane soil increased by 4.57-27.39%, 10.80-73.13%, and 17.57-74.80% when treated with 0.5%, 2%, and 5% biochar for 0-40 days, respectively. After 60 days of culture, the CEC increased gradually and then stabilized. The CEC of control (0% ZSB) increased to $61.01 \text{ cmol}\cdot\text{kg}^{-1}$, which was higher than that of other treatments. The CEC of biochar treatment in manganesecontaminated sugarcane field soil increased with the proportion of biochar as well as with time. When compared with that of the control (0% MSB), the CEC of 0.5% MSB, 2% MSB, and 5% MSB increased by 0.87-50.83%, 7.30-65.11%, and 26.59-175.64%, respectively. It increased slowly after 40 days of culture and gradually stabilized.

Effects of biochar on soil nutrients

From Fig. 3, it can be observed that during the entire culture process, the AP content in manganese-contaminated sugarcane field soil and uncontaminated sugarcane field soil increased with the increase in bagasse biochar amount. When compared with the control, the application of biochar can significantly increase the soil AP content in the soil. The AP content of the uncontaminated sugarcane field followed the order: 5% ZSB>2% ZSB>0.5% ZSB>0% ZSB. The AP content in 0.5% ZSB, 2% ZSB, and 5% ZSB increased by 143.85%, 417.06%, and 663.32%, respectively, when compared with that in the control (0% ZSB). The AP content in 2% ZSB and 5% ZSB was significantly higher than that in 0.5% ZSB. When biochar was applied to the manganese-contaminated sugarcane field soil, only a minimal change was observed in the AP content in each treatment during 0-40 days. After 40 days, the largest increase in the AP content was observed in 5% MSB followed by 2% MSB, and the smallest increase was observed in 0.5% MSB, which was significantly higher than that in the control (0% MSB). Thus, the increase in AP content followed the order: 5% MSB > 2% MSB > 0.5% MSB > 0% MSB.

As shown in Fig. 4, different bagasse biochar amounts applied to manganese-contaminated sugarcane field soil and uncontaminated sugarcane field soil during the entire culture process can increase the soil AK content to different degrees, which exhibited the following order: 5% ZSB > 2% ZSB > 0.5% ZSB > 0% ZSB and 5% MSB > 2%



Fig. 2 Effects of biochar on soil cation exchange capacity (CEC). (a) Uncontaminated sugarcane field soil. (b) Manganese-contaminated sugarcane field soil



Fig. 3 Effects of biochar on soil available phosphorus (AP). (a) Uncontaminated sugarcane field soil. (b) Manganese-contaminated sugarcane field soil



Fig. 4 Effects of biochar on soil available potassium (AK). (a) Uncontaminated sugarcane field soil. (b) Manganese-contaminated sugarcane field soil

MSB > 0.5% MSB > 0% MSB. During the incubation period, the AK content in 0.5% ZSB, 2% ZSB, and 5% ZSB increased by 5.95%, 48.85%, and 117.42% on average, respectively, when compared with that in the control

(0% ZSB). When compared with that in the control (0% MSB), the AK content in 0.5% MSB, 2% MSB, and 5% MSB increased by 54.82%, 210.11%, and 251.66% on average, respectively, and the AK content in the soil treated

with biochar was significantly higher than that in the control.

Effects of biochar on SOC mineralization

(a) 180

160

As shown in Fig. 5, when compared with the control, the application of bagasse biochar significantly reduced the CO₂ emission rates in both the sugarcane fields, and the emission rate decreased with the increase in biochar amount. During the entire incubation period, the CO₂ emission rates in the two biochar-applied sugarcane fields were lower than those in the control. During the early stage of culture, the CO₂ emission rates from biochar-applied uncontaminated sugarcane field soil followed the order: 0% ZSB>0.5% ZSB>2% ZSB>5% ZSB, and the CO₂ emission rates from biochar-applied manganese-contaminated sugarcane field soil followed the order: 0% MSB>0.5% MSB>2% MSB>5% MSB. In the uncontaminated sugarcane field soil, the CO₂ emission rate decreased rapidly within 0–10 days. Compared with the control (0% ZSB), the application of 0.5%, 2%, and 5% biochar decreased the CO₂ emission rates by 17.83 times, 18.27 times, and 25.08 times, respectively. In the manganese-contaminated sugarcane soil, the application of 0.5%, 2%, and 5% biochar decreased the CO₂ emission rates by 14.74 times, 17.42 times, and 19.17 times, respectively, when compared with the control (0% MSB). During the entire incubation period, the CO₂ emission from each treated soil can be divided into three stages: a rapid declining stage of the soil CO₂ emission rate during 0-10 days, a gradual decrease in the soil CO₂ emission rate during 10-40 days, and a gradual stabilization of the CO₂ emission rate after 40 days.

As shown in Fig. 6, the cumulative CO_2 emissions from the two sugarcane fields decreased with the increase in the bagasse biochar application amount. After 0-40 days of incubation, the cumulative CO₂ emissions from each treatment increased rapidly and then gradually stabilized. During the entire incubation process, the cumulative CO_2 emissions from the uncontaminated sugarcane soil treated with different proportions of biochar were significantly lower than those in the control (0% ZSB). At the end of the incubation period, the cumulative CO_2 emissions from ZSB followed the order: 0% ZSB>0.5% ZSB > 2% ZSB > 5% ZSB, and the cumulative CO_2 emissions from MSB followed the order 0% MSB>0.5% MSB>2% MSB>5% MSB. When compared with those from the control (0% ZSB), the cumulative CO₂ emissions from the uncontaminated sugarcane field soil treated with different proportions of biochar decreased by 35.29-57.29%. Similarly, when compared with those from the control (0% MSB), the cumulative CO_2 emission from the manganese-contaminated sugarcane field soil treated with different proportions of biochar decreased by 15.78-36.87%.

As shown in Table 3, the first-order kinetic equation accurately simulated the mineralization dynamics of SOC during the 100-day incubation period. In general, the potential mineralization amounts of organic carbon

0% MSB

0.5% MSB



(b) ¹⁴⁰

0% ZSB

Fig. 5 Effects of biochar on mineralization rate. (a) Uncontaminated sugarcane field soil. (b) Manganese-contaminated sugarcane field soil



Fig. 6 Effects of biochar on cumulative mineralization. (a) Uncontaminated sugarcane field soil. (b) Manganese-contaminated sugarcane field soil

Table 3 Kinetic parameters of soil carbon mineralization in biochar-treated soils

Different Treatments	Fitting Parameters				
	C₀/µg·g ⁻¹	k/d ⁻¹	R ²		
0%ZSB	1140.45 ± 48.48	0.069 ± 0.009	0.95		
0.5%ZSB	758.529 <u>+</u> 26.68	0.076±0.010	0.95		
2% ZSB	627.780 ± 20.03	0.099±0.012	0.96		
5% ZSB	481.268 ± 16.32	0.135 ± 0.019	0.94		
0%MSB	740.359 ± 24.49	0.089 ± 0.010	0.95		
0.5% MSB	634.238±15.97	0.101 ± 0.010	0.97		
2% MSB	557.396 <u>+</u> 12.47	0.093 ± 0.007	0.97		
5%MSB	486.594 <u>+</u> 9.368	0.091 ± 0.006	0.98		

ZSB indicates biochar applied to the uncontaminated sugarcane field soil, MSB indicates biochar applied to the manganese-contaminated sugarcane soil, C₀ represents the soil carbon mineralization potential, and k represents the soil carbon mineralization rate constant.

 (C_0) in soils treated with different proportions of biochar were significantly different. C_0 in the uncontaminated soils treated with biochar ranged from 481.268 to 1140.45 µg·g⁻¹, and in the manganese-contaminated soils treated with biochar ranged from 486.594 to 740.359 µg·g-1. It can be observed that the mineralization potential of soil decreased with the increase in biochar amount. However, the soil carbon mineralization rate constant (k) demonstrated an opposite trend. The k value of the ZSB group varied between 0.069 and 0.135 d^{-1} , while that of the MSB group varied between 0.089 and 0.101 d^{-1} .

Effects of biochar on soil active organic carbon components

The addition of different proportions of bagasse biochar to the uncontaminated sugarcane field soil and manganese-contaminated sugarcane field soil could increase the SOC content, with the increase in biochar amount leading to increasing SOC content (Fig. 7). At the end of the incubation period, different amounts of biochar were added to the uncontaminated sugarcane field soil. When compared with the control, the addition of 5% and 2% biochar significantly increased the SOC content by 100% and 58.87%, respectively. The addition of 0.5% biochar did not significantly increase the organic carbon content. The SOC content in each treatment followed the order: 5% ZSB > 2% ZSB > 0.5% ZSB > 0% ZSB. The SOC content in the manganese-contaminated sugarcane field soil with 5%, 2%, and 0.5% biochar increased by 132.70%, 114.56%, and 33.41%, respectively, when compared with that in the control. The organic carbon content followed the order: 5% MSB > 2% MSB > 0.5% MSB > 0% MSB.

The effects of bagasse biochar application on the stability coefficient of SOC in two sugarcane fields were analyzed. As shown in Table 4, the stability coefficient of SOC in two sugarcane fields generally increased with the increase in biochar application amount.



Fig. 7 Effect of biochar on soil organic carbon (SOC). (a) Uncontaminated sugarcane field soil. (b) Manganese-contaminated sugarcane field soil

Table 4 Stability coefficients of soil organic carbon (SOC) ineach treatment

Stability Factors	0%	0.5%	2%	5%
ZSB	0.47	0.51	0.48	0.54
MSB	/	0.07	0.50	0.53

Figure 8 shows the changes in MBC of manganesecontaminated sugarcane field soil and uncontaminated sugarcane field soil with different bagasse biochar application rates. During the entire culture period, the MBC in the soil increased with the increase in the biochar application amount and culture time. At the end of the incubation period, the MBC of the uncontaminated sugarcane soil with 5%, 2%, and 0.5% biochar application was 39.42 mg·kg⁻¹, 37.92 mg·kg⁻¹, and 28.49 mg·kg⁻¹, respectively. When compared with the control (0% ZSB), the MBC content of 5% ZSB, 2% ZSB, and 0.5% ZSB increased by 117.55%, 109.29%, and 57.23%, respectively. The MBC content of the manganese-contaminated sugarcane field soil with 5%, 2%, and 0.5% biochar was 15.10 mg·kg⁻¹, 13.71 mg·kg⁻¹, and 12.72 mg·kg⁻¹, respectively. When compared with the control (0% MSB), the MBC content of 5% MSB, 2% MSB, and 0.5% MSB increased by 180.15%, 154.36%, and 135.99%, respectively.

As shown in Fig. 9, the soil DOC content initially increased and then decreased with time when different

amounts of bagasse biochar were applied to the uncontaminated sugarcane field soil. During the entire culture period, the DOC varied from 32.24 to 14.99 mg·kg⁻¹, 37.03-15.64 mg·kg⁻¹, 43.50-16.11 mg·kg⁻¹, and 50.30-17.37 mg·kg⁻¹, respectively, when 0%, 0.5%, 2%, and 5% biochar were applied. The soil DOC content reached the maximum values of 32.24 mg·kg⁻¹, 37.03 mg·kg⁻¹, and 43.50 mg·kg⁻¹ on the 10th day of culture with the application of 0%, 0.5%, and 2% biochar, respectively. The soil DOC content reached the maximum values of 50.30 mg·kg⁻¹ on the 20th day with the application of 5% biochar. When different amounts of biochar were applied to the manganese-contaminated sugarcane field soil, the soil DOC content decreased with the increase in biochar amount. During the entire culture period, DOC varied from 43.82 to 13.96 mg·kg⁻¹, 34.44–13.59 mg·kg⁻¹, 30.95-13.00 mg·kg⁻¹, and 26.27-11.94 mg·kg⁻¹, respectively, with the application of 0%, 0.5%, 2%, and 5% biochar. The DOC content reached the maximum values of 34.44 mg·kg⁻¹ and 30.95 mg·kg⁻¹ on the 15th day when 0.5% and 2% biochar were applied, respectively. Similarly, the DOC content reached 43.82 mg kg^{-1} and 26.27 mg·kg⁻¹ on the 20th day of culture when 0% and 5% biochar were applied, respectively.

As shown in Fig. 10, the application of different amounts of bagasse biochar can increase the ROC content in uncontaminated sugarcane field soil and manganese-contaminated sugarcane field soil, with the increase in biochar application amount leading to



Fig. 8 Effects of biochar on soil microbial biomass carbon (MBC). (a) Uncontaminated sugarcane field soil. (b) Manganese-contaminated sugarcane field soil



Fig. 9 Effects of biochar on soil dissolved organic carbon (DOC). (a) Uncontaminated sugarcane field soil. (b) Manganese-contaminated sugarcane field soil



Fig. 10 Effects of biochar on soil readily oxidizable organic carbon (ROC). (a) Uncontaminated sugarcane field soil. (b) Manganese-contaminated sugarcane field soil



Fig. 11 Effects of biochar on soil catalase. (a) Uncontaminated sugarcane field soil. (b) Manganese-contaminated sugarcane field soil

increasing ROC content. When biochar was applied to the uncontaminated sugarcane field soil, the ROC content increased slowly from 0 to 40 days. After 40 days, the increase in 2% and 5% biochar-treated soil was more obvious, with ROC content increasing from $1.23 \text{ mg} \cdot \text{g}^{-1}$ to 2.20 mg·g⁻¹ and 1.37 mg·g⁻¹ to 2.25 mg·g⁻¹, i.e., an increase by 78.86% and 64.23%, respectively. The ROC content ranged from 1.11 to 2.25 mg·g⁻¹ during the entire culture period. The ROC content in the manganese-contaminated sugarcane field soil increased with the increase in biochar application amount and culture time. The ROC content increased by 6.63%, 17.13%, and 27.07%, respectively, with the application of 0.5%, 2%, and 5% biochar when compared with the control (0% MSB).

Effects of biochar on the soil enzyme activity

As shown in Fig. 11, the application of bagasse biochar in both the sugarcane fields could increase the catalase content. With the increase in biochar application amount, the catalase content in the uncontaminated sugarcane fields was much higher than that in the manganese-contaminated sugarcane fields. The catalase activity in the uncontaminated sugarcane fields increased with time. At the end of the culture period, the catalase activity of 0.5% ZSB, 2% ZSB, and 5% ZSB increased by 1.24 times, 1.33 times, and 1.47 times, respectively, when compared with that of the control (0% ZSB). The catalase content in the manganese-contaminated sugarcane field soil also increased with time. At the end of the culture period, the catalase activity of 0.5% MSB, 2% MSB, and 5% MSB increased by 2.19 times, 3.44 times, and 4.48 times, respectively, when compared with that of the control (0% MSB).

As shown in Fig. 12, the application of different amounts of bagasse biochar in the uncontaminated sugarcane field and manganese-contaminated sugarcane field soils could increase urease activity with time. The urease activity followed the order: 5% ZSB > 2% ZSB>0.5% ZSB>0% ZSB, 5% MSB>2% MSB>0.5% MSB > 0% MSB. In the uncontaminated sugarcane field soil, the urease activity reached the maximum value of 0.43 mg·g⁻¹·h⁻¹ on the 80th day with the application of 5% biochar. At the end of the culture period, the urease activities of 0.5% ZSB, 2% ZSB, and 5% ZSB were $0.28 \text{ mg}\cdot\text{g}^{-1}\cdot\text{h}^{-1}$, $0.34 \text{ mg}\cdot\text{g}^{-1}\cdot\text{h}^{-1}$, and $0.42 \text{ mg}\cdot\text{g}^{-1}\cdot\text{h}^{-1}$, respectively, which were 1.05 times, 1.29 times, and 1.59 times higher than those of the control (0% ZSB). Similarly, at the end of the culture period, the urease activities of 0.5% MSB, 2% MSB, and 5% MSB were 0.56 mg·g⁻¹·h⁻¹, 0.59 mg·g⁻¹·h⁻¹, and 0.60 mg·g⁻¹·h⁻¹, respectively, which were 1.03 times, 1.08 times, and 1.10 times higher than those of the control (0% MSB).

Correlation analysis

As shown in Fig. 13, when biochar was applied to the sugarcane field soil, CEC and ROC were significantly



Fig. 12 Effects of biochar on the soil urease. (a) Uncontaminated sugarcane field soil. (b) Manganese-contaminated sugarcane field soil



Fig. 13 Correlation analysis. (a) Uncontaminated sugarcane field soil. (b) Manganese-contaminated sugarcane field soil

positively correlated with AP; CEC was significantly positively correlated with ROC; and urease and catalase activities were significantly positively correlated with MBC. When biochar was applied to the manganesecontaminated sugarcane field soil, AK was significantly positively correlated with SOC; CEC and AP were significantly positively correlated with ROC; and urease and catalase activities were significantly positively correlated with MBC. Moreover, the cumulative CO₂ emission was negatively correlated with pH and positively correlated with CEC.

Discussion

Effects of biochar on soil pH and CEC

The application of biochar to the uncontaminated sugarcane field and manganese-contaminated sugarcane soil increased the field soils pH. Although the change in pH with time was not significant, the pH value was positively correlated with the biochar application amount. This indicated that the increase in pH value in this experiment may depend on the soil type and biochar application rate, and is not affected by the culture time, which is similar to research results from a previous study [26]. The application of biochar to the manganese-contaminated sugarcane field soil resulted in a slight increase in the pH value, probably due to the strong acidity of the soil (Table 1). Therefore, the addition of biochar has a little effect on the soil pH. This is consistent with the results of a study by Huang et al. [27], where a slight increase in soil pH was observed by the application of biochar to the heavy metal-contaminated soil. The increase in pH of the uncontaminated sugarcane field soil may be due to the application of bagasse biochar to the soil, which drives the oxygen-containing functional groups on the soil surface to combine with Al³⁺, reduces the exchangeable aluminum content, and increases the abundance of exchangeable alkali cations [28]. In this study, bagasse biochar also increased the pH of the manganese-contaminated soil. The application of 5% bagasse biochar increased the soil pH to 6.45, which improved the acidity of manganese-contaminated soil to a certain extent.

CEC is one of the important indicators for the measurement of soil quality, which can reduce soil nutrient leaching [29]. The application of bagasse biochar to manganese-contaminated soil increased soil CEC due to the negatively charged oxygen-containing functional groups on the biochar surface, thereby increasing CEC [30], as well as due to the presence of alkaline substances in biochar, higher ash content, and potassium, calcium, and magnesium ions [26]. When compared with the control, the CEC increased with the increase in biochar application amount during 0-40 days following the bagasse biochar application to the uncontaminated sugarcane field soil. This may be due to the influence of biochar surface oxidation and porous structure [31]. After 60 days, the application of biochar reduced CEC, which is similar to research results from a previous study [26]. In that study, it was hypothesized that the decrease in CEC was due to the blockage of biochar pores by humic acid and fulvic acid in the soil, which would further block its physical adsorption, thereby reducing CEC. The correlation results demonstrated a significant positive correlation between soil CEC and cumulative CO₂ emissions from the two soils, indicating that an increase in the cumulative CO₂ emissions from bagasse biochar-treated soils led to an increase in soil CEC.

Effects of biochar on soil nutrients

Bagasse biochar was applied to the manganese-contaminated sugarcane field soil. During 0-40 days, the AP content gradually increased. This may be due to the acidic nature of the manganese-contaminated sugarcane field soil and the high activity of iron and aluminum in the soil, which leads to the formation of large quantities of insoluble iron phosphate and aluminum phosphate, and even leads to a closed phosphorus cycle with lower effectiveness, with most of the soil phosphorus getting converted into fixed phosphorus [32]. After 40 days, the application of 5% biochar resulted in the largest increase in AP content when compared with the control. This may be because of the increase in soil pH by biochar application, and the increase in soil pH leads to a decrease in iron and aluminum activity, leading to an increase in soil AP content. The application of bagasse biochar significantly increased the AP content in manganese-contaminated sugarcane field soil, with the increase in biochar amount leading to enhanced AP content, which is in agreement with research results from a previous study [33]. In that study, corn straw biochar, rice husk biochar, and wheat straw biochar were applied to the chromiumcontaminated soil, and the results demonstrated a significant increase in the AP content, and this increase was influenced by the amount of biochar applied. Bagasse biochar exhibits a beneficial effect on enhancing AP levels in uncontaminated sugarcane field soil. This may be because biochar can adsorb soil phosphate and reduce AP leaching [34]. Huang et al. [35] demonstrated that the potassium and phosphorus content in sugarcane field soil was low. The ability of soil to supply phosphorus can be reflected by the AP content [36]. In this study, the application of raw bagasse biochar increased the soil AK content, with the increase in biochar amount leading to increasing AK content. This may be due to the porous adsorption and retention of soil potassium by biochar with a large specific surface area, leading to a reduction in the leaching of nutrients [28]. Similarly, Wang et al. [37] demonstrated that the application of biochar to tobacco fields increased soil AK content by 10.57%, which is consistent with the results of this study. In this study, the application of bagasse biochar to the manganese-contaminated sugarcane field soil alleviated the nutrient deficiency in the soil to a certain extent.

Effects of biochar on SOC mineralization

Biochar addition to soil can affect the basic physical and chemical properties of soil and soil structure, thereby influencing CO_2 emissions [28]. The application of biochar significantly reduced the CO_2 emission rates in both the sugarcane fields, which is similar to previous research results [38]. When compared with the control, the CO₂ emission decreased slightly with the application of bagasse biochar to the manganese-contaminated sugarcane field soil. The reasons for this are as follows: first, the quality of manganese-contaminated soil is poor, the nutrient content is low, and limited energy is provided by biochar for organic carbon mineralization in soil; second, the organic carbon mineralization is affected by microbial biomass. Manganese ions in the soil may be toxic and inhibitory to microorganisms, thus reducing the microbial decomposition of organic carbon and the mineralization rate of SOC [39]. Verma et al. [40] applied biochar to heavy metal-contaminated soils, and the results demonstrated that soil CO₂ emissions decreased with the increase in heavy metal concentration. In that study, it was revealed that heavy metal pollution may reduce soil microbial activity, thereby reducing soil respiration rate, which is consistent with the results of this study. Similarly, biochar can effectively reduce CO₂ emissions from heavy metal-contaminated soils, which has also been reported in other studies [41]. Manganese is an important metal element, which affects the soil carbon cycle by interfering with the soil anaerobic microbial redox under manganese-rich conditions [42]. A study by Tian et al. [43] has demonstrated that heavy metal pollution affects soil microbial communities, which in turn affects soil carbon emissions. The significant decrease in CO₂ emissions in the uncontaminated sugarcane field soil may be attributed to biochar, which contains Mg, Ca, and other elements. These elements combine with CO_2 in the soil to form $CaMg(CO_3)_2$ and other substances, thereby reducing CO₂ emissions [44]. Similarly, Mendez et al. [45] observed that the application of biochar to soil effectively reduced CO₂ emissions. In this study, soil pH was negatively correlated with cumulative mineralization, indicating that soil pH is also an important factor.

Effects of biochar on soil active organic carbon components

The application of biochar could increase the soil organic carbon (SOC) content in the two sugarcane fields, with the increase in biochar application amount leading to an increase in SOC, which is similar to previous research results [46]. SOC turnover is closely related to soil fertility and plays an important role in regulating ecosystem carbon cycle [47]. SOC can play a crucial role in providing carbon sources for soil microorganisms, plants, and animals [48]. When compared with the control, bagasse biochar had a limited impact on the increase in SOC content of manganese-contaminated sugarcane fields, because the soil DOC enters the biochar pores or is adsorbed on the biochar surface [49]. The increase in SOC content of the uncontaminated sugarcane field soil has a more positive effect, which may be due to the porosity of biochar

and the formation of aggregates, which enhances the protection of organic carbon and thus, inhibits the ability of microorganisms to decompose organic carbon [50]. Similarly, in a study by Liu et al. [51], the application of biochar to the soil increased SOC content by 7.88-30.05%, and the SOC content increased with the increase in biochar application, which was consistent with the results of this study. The SOC stability coefficient can be used to measure the stability of SOC, which can reflect the proportion of inert organic carbon in total soil carbon [52]. In this study, the stability coefficient of SOC increased with the amount of biochar application, indicating that biochar application could increase the stability of SOC to a certain extent. Moreover, the stability coefficients of SOC in manganese-contaminated sugarcane field soils were lower those that in uncontaminated sugarcane field soils, indicating that heavy metals may affect the stability of SOC to a certain extent. MBC is an important carbon source in the soil, and its variational trend indicates the carbon utilization efficiency of soil microorganisms [53]. Biochar had a minimal effect on the increase in MBC content of the manganese-contaminated sugarcane field soil, which may be partly due to the high manganese content of the sugarcane field soil, which destroys the microbial cell structure and function, thus inhibiting microbial activity. Another possible explanation for this is that microorganisms may slow down their growth and consume more energy to resist metal toxicity under the stress of manganese ions [54]. The MBC content in the uncontaminated sugarcane soil increased significantly, probably due to two major reasons. First, the porosity of biochar, makes the soil loose and improves soil aeration, thus increasing microbial activity [55]. Second, biochar has rich carbon content, which can provide additional carbon sources for microorganisms, thus providing good growth conditions for the microorganisms [55]. In a study by Jiang et al. [56], the application of pig manure biochar to tea garden soil increased MBC by 5.73-36.4%, which is consistent with the results of this study.

DOC and ROC are active components of SOC, which are directly involved in soil chemical cycling and provide nutrients for soil microbial growth [57]. This study demonstrated that the DOC content in two types of sugarcane field soils initially increased and then decreased with time, which is similar to previous research results [58]. It also demonstrated the same variational trend in the DOC content when the bagasse biochar was applied to the manganese-contaminated sugarcane field soil. In the early stage, biochar may adsorb manganese ions in the soil and reduce manganese toxicity in microorganisms [59]. Moreover, the application of exogenous organic carbon increased DOC content. In the later period, DOC may be used as a heavy metal ion carrier, which promotes the combination of manganese ions with DOC, thereby reducing the DOC content [60]. A similar phenomenon was also observed in a previous study [61]. In that study, the authors hypothesized that the surface functional groups of biochar and cations form complexes with DOC in the soil, thereby reducing the soil DOC content. The application of bagasse biochar to uncontaminated sugarcane field soil leads to a positive impact. This may be because the application of biochar can reduce the loss of soil nutrients, create a favorable environment for the growth of microorganisms, promote the microbial decomposition of soil organic matter, and increase DOC content [62]. The ROC content of the two sugarcane field soils increased with the application ratio of biochar. The application of 5% biochar exhibited the most significant impact, and 0.5% biochar yielded the least significant impact. The application of biochar to the soil can improve the soil structure and adsorb nutrients. Moreover, it can provide good growth conditions for microorganisms as well as promote soil respiration and organic carbon mineralization, thereby increasing the soil ROC content [63]. The correlation results demonstrated a significant positive correlation between ROC and AP, indicating that an increase in ROC content is also affected by AP.

Effects of biochar on soil enzyme activity

Soil enzyme activity is an important parameter to measure soil microbial activity. It is involved in the decomposition of soil organic matter and can affect soil fertility as well as the biological cycle [64]. The application of biochar to the manganese-contaminated sugarcane field soil and uncontaminated sugarcane field soil could increase the soil enzyme activity, with an increased amount of biochar application leading to increased enzyme activity, which is similar to previous research results [65]. When compared with the control, the soil catalase activity was lower during 0–40 days after the application of bagasse biochar to the manganese-contaminated sugarcane field soil, which may be due to the relatively stable catalase activity in the soil and low influence of the external environment. The application of biochar altered the soil moisture and inhibited the catalase activity [66]. After 40 days, the catalase activity increased significantly. Due to a large surface area and porous characteristics, biochar provides an environment conducive to the growth of microorganisms; enhances the flow of air, water, and nutrients in the soil; and improves microbial activity, thereby increasing enzymatic activity [67]. The inhibitory effect of catalase in the early stage following the application of biochar and then a promoting effect in the later stage was also reported in other studies [18]. The soil catalase activity was effectively improved in the uncontaminated sugarcane field soil, probably because

the application of biochar improved the soil structure by providing a habitat favorable for microbial growth and conducive to the oxidation activity of soil microorganisms, thereby increasing catalase activity [68]. The catalase activity increased with time, and this increasing trend was consistent with the variational trend of MBC, indicating a positive correlation of catalase activity MBC. Thus, the application of biochar enhances soil respiration, which in turn, improves the utilization of soil carbon.

Urease plays an important role in the transformation of soil nitrogen, which can catalyze the hydrolysis of urea in the soil to carbon dioxide and ammonium [69]. In this study, when compared with the control, the application of bagasse biochar had no significant effect on urease activity in the manganese-contaminated sugarcane field soil. The analysis revealed the following two reasons for this phenomenon: first, the combination of heavy metals and the active protein gene of the enzyme, can diminish the binding site of the enzyme, thereby inhibiting enzyme synthesis; second, heavy metals may deform proteins, thereby reducing enzyme activity and inhibiting enzyme synthesis [70]. The urease activity in the uncontaminated soil increase significantly when compared with that in the control. It is possible that the application of biochar increased urease activity by changing the composition of the soil microbial community and enhancing its biogeochemical cycle [71]. Urease activity was positively correlated with MBC, further indicating that microorganisms can affect urease activity.

Conclusions

This study applied different amounts of bagasse biochar to the manganese-contaminated sugarcane field soil to evaluate its effect on SOC sequestration. The results demonstrated significant differences in the mineralization characteristics of SOC between manganese-contaminated sugarcane field soil and uncontaminated sugarcane field soil. Among all the treatments, 5% bagasse biochar treatment significantly inhibited the mineralization of SOC. The application of bagasse biochar significantly increased the enzyme activity, organic carbon, MBC content, and ROC content in manganese-contaminated sugarcane field soil. Briefly, the application of 5% bagasse biochar demonstrated a good carbon sequestration effect on the manganese-contaminated sugarcane soil. The results of this study hold great significance in advancing our understanding of the mechanisms underlying carbon sequestration and emission reduction in manganese-contaminated farmland systems.

The applications of different amounts of bagasse biochar to both the manganese-contaminated sugarcane field soil and uncontaminated sugarcane field soil can improve the carbon sequestration capacity of the soil and this capacity increases with the increase in the biochar application amount. However, the carbon sequestration effects of bagasse biochar on the manganese-contaminated sugarcane field soil are slightly lower, probably due to the decrease in the soil carbon sequestration capacity of bagasse biochar mediated by manganese. However, the specific reason for this is not yet clear and needs further study.

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Author contributions

L.H: conceptualization and data curation. Y.Y: software, writing original draft preparation and data curation. X.H.L and S.L: data curation, supervision. H.D and K.L: supervised the experiment project and approved the final version. All authors have read and agreed to the published version of the manuscript.

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Availability of data and materials

All data are presented in the manuscript.

Declarations

Ethics approval and consent to participate

The manuscript is an original work that has not been published in other journals.

Consent for publication

All authors agreed to the publication.

Competing interests

The authors declare that they have no competing interests.

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