Greater microbial carbon use efficiency and carbon sequestration in soils: Amendment of biochar versus crop straws

Zhiwei Liu1,2 | Xiulan Wu1,2 | Wei Liu1,2 | Rongjun Bian1,2 | Tida Ge3
Wei Zhang4 | Jufeng Zheng1,2 | Marios Drosos1,2 | Xiaoyu Liu1,2 | Xuhui Zhang1,2
Kun Cheng1,2 | Lianqing Li1,2 | Genxing Pan1,2

Abstract
While high soil carbon stability had been well known for biochar-amended soils, how conversion of crop residues into biochar and subsequent biochar amendment (BA) would favor microbial carbon use and carbon sequestration had not been clearly understood. In this study, topsoil samples were collected from an upland soil and a paddy soil, both previously amended with straw and straw-derived biochar. These samples were incubated with 13C-labeled maize residue (LMR) for 140 days to compare carbon mineralization, metabolic quotient (qCO2), and microbial carbon use efficiency (CUE) under laboratory incubation. 13C-phospholipid fatty acid (13C-PLFA) was used to trace the use of substrate carbon by soil microorganisms. Comparing to straw amendment (SA), BA significantly decreased the native soil organic carbon (SOC) mineralization rates by 19.7%–20.1% and 9.2%–12.0% in the upland and paddy soils, respectively. Meanwhile, total carbon mineralization from the newly added LMR was significantly decreased by 12.9% and 11.1% in biochar-amended soils, compared with the straw-amended soils from the upland and paddy sites, respectively. Furthermore, compared to non-amended soils, the qCO2 value was unchanged in straw-amended soils, but was notably decreased by 15.2%–18.6% and 8.9%–12.5% in biochar-amended upland and paddy soils, respectively. Microbial CUE was significantly greater in biochar-amended soils than in straw-amended soils due to the increasing dominance of fungi in carbon utilization. Compared to SA, BA increased CUE by 23.0% in the upland soil and 21.2% in the paddy soil. This study suggests that BA could outperform SA in the long term to enhance the biological carbon sequestration potential of both upland and paddy soils. This could be due mainly to biochar input as a special substrate to promote microbial community evolution and increase the fungal utilization of carbon substrates, especially for the soil with lower SOC levels.

KEYWORDS
13C-phospholipid fatty acids, biochar, carbon sequestration, crop residue, microbial carbon use efficiency, soil amendment
1 | INTRODUCTION

Soil organic carbon (SOC) plays an important role in global C cycle and enhancing SOC sequestration is crucial for maintaining soil fertility and crop productivity, mitigating climate change, and improving agricultural sustainability (Lal, 2004, 2009; Lehmann & Kleber, 2015). To increase SOC storage, the main approach is to increase C inputs and/or decrease C losses (Ye et al., 2019). Several popular strategies recommended for C sequestration in agricultural soils include straw incorporation, organic fertilizer application, biochar amendment (BA), and conservation tillage. Incorporation of straw into soils is commonly used to improve soil fertility and crop yield by returning nutrient-rich organic matter to soils (Liu et al., 2014; Lu et al., 2009). However, straw incorporation may accelerate SOC decomposition and exert negligible or even negative impact on net amount of sequestered C, primarily because of increased microbial biomass, activities and C losses via mineralization and methanogenesis in aerobic and anaerobic soils (Wu et al., 2019; Zhu et al., 2018). As such, straw application may unintentionally increase CO$_2$ and CH$_4$ emissions to the atmosphere, thus contributing to global climate change and impeding sustainable agricultural development.

In contrast, incorporation of biochar into soils has been recognized as a promising strategy to increase C sequestration, mitigate greenhouse gas emissions, and promote soil health (Han et al., 2020; Lehmann, 2007; Smith, 2016; Xu et al., 2019). Biochar is pyrolyzed from organic materials under oxygen-limited conditions, and often contains a relatively larger proportion of condensed aromatic C that can persist in soils for hundreds to thousands of years (Lehmann et al., 2011). Thus, BA in soils as a C sequestration strategy has received much attention over the past decade (Smith, 2016; Sohi, 2012; Yousaf et al., 2017). Nonetheless, conflicting results were reported on whether biochar addition increased or decreased the mineralization of native SOC (i.e., the positive or negative priming effect; Jiang et al., 2019; Keith et al., 2011; Yousaf et al., 2017). These studies indicate that microbial utilization of C could be altered by BA in various ways, which, in turn, could variably affect the potential of C sequestration. Generally, biochar may have an intensively positive priming effect in the initial phase and a negative priming effect in the later period (>20 days; Maestrini et al., 2015). For example, Zimmerman et al. (2011) reported that following biochar addition to soils, the positive priming effect tended to decrease over time and could even switch to the negative priming. Although the long-term effect of BA on protecting C residues from decomposition was found in some studies (Hernandez-Soriano et al., 2016; Kerré et al., 2017), the persistent effect of biochar on microbial C use efficiency (CUE) remains elusive.

Microbial CUE is defined as the ratio of net soil microbial growth over substrate consumption (the sum of total microbial respiration, production and death; Fang, Singh, Luo, et al., 2018; Geyer et al., 2016). The microbial CUE is a critical parameter for modeling long-term SOC change, and a higher CUE implies an increased C sequestration potential in soils (Sinsabaugh et al., 2013; Zhran et al., 2020). In general, soil properties and management practices control microbial CUE, and thus affect microbial residues and C storage in soils (Chen et al., 2020; Cotrufo et al., 2015; Fang, Singh, Collins, et al., 2018). Microbial CUE is usually higher under lower environmental stress, as lower energy consumption is needed for coping with environmental stress and more organic C is available for microbial growth (Manzoni et al., 2012). In arable soils, the different SOC augmentation strategies (e.g., SA or BA) may influence soil microbial community structure and activities as fungi, archaea, and bacteria may respond differently to added C sources from straw or biochar. Bacteria and fungi are both capable of utilizing labile organic C initially, but fungi can more efficiently use complex C sources in aged substrates once labile C is depleted (Boer et al., 2005). In addition, it has been recognized that fungi have a greater resource use efficiency than bacteria (Keiblinger et al., 2010; Six et al., 2006). Therefore, it is important to characterize the changes in microbial CUE of soils receiving biochar or straw additions, as modulated by soil microbial community and its metabolic activities.

To address this question, we selected upland and paddy soils as representative global arable soils. In China, upland is mainly used for growing maize and wheat and is primarily distributed in the north of the Qinling Mountain and Huaihe River, whereas paddy soil is mainly distributed in the south of China. Considering the differences in climate, soil properties, and management practices, it is intriguing to characterize and compare the changes of SOC and microbial CUE in upland and paddy soils with SA or BA. Thus, this study aimed to investigate the shift of soil microbial communities and their C utilization in upland and paddy soils amended with straw or biochar using a well-controlled laboratory incubation experiment. Specifically, straw and biochar-amended soils were collected from two 3-year field study sites, respectively, and then received new addition of $^{13}$C-labeled maize residue (LMR) prior to incubation. CO$_2$ fluxes were determined during the 140-day incubation, and microbial biomass C (MBC), phospholipid fatty acids (PLFAs), and microbial CUE were analyzed at the end of incubation. We hypothesized that (1) C mineralization could be lowered to preserve the native SOC and newly added substrate in the biochar-amended soil and (2) soil microbial CUE could be increased due to improved soil habitat in the biochar-amended soil, corroborating with soil microbial community changes. This study specifically tested these two hypotheses and provided compelling insights into C dynamics in soils amended with straw or biochar.
2 | MATERIALS AND METHODS

2.1 | Field site description

In this study, the upland and paddy soils used in the laboratory incubation experiments were collected from two field experiments with crop straw and straw-derived BMs, respectively. The upland field site was located at Matouwang village (35°09′N, 113°51′E), Xinxiang Municipality, Henan Province, China. Situated in the central North China Plain, this region has a semi-humid temperate continental monsoon climate with an annual precipitation of 656.3 mm and a mean annual temperature of 13.9°C. Its calcareous cinnamon soil was derived from paleo-deposits of the Yellow River, and is classified as a loamy Aqufluvent (Soil Survey Staff, 1994). The soil texture is sandy loam with a clay (<0.01 mm) content of 13.1%. This region is traditionally an agricultural production area under the rotation of summer maize and winter wheat. The paddy field site was established in Kangbo village (31°30′N, 120°33′E), Changshu Municipality, Jiangsu Province, China. This area is located in the center of Tai Lake Plain, and is characterized by a subtropical monsoon climate with mean annual temperature of 16°C and precipitation of 1,100–1,200 mm. Its soil was formed from a clayey lacustrine deposit, and is classified as a Gleyic Stagnic Anthrosol (WRB, 2015) and a clay loam in texture with a clay content of 27.6%. This area has been farmed under the rotation of summer rice and winter wheat for hundreds of years and is agriculturally important in China. At both sites, the field experiments were initiated after wheat harvest in June 2014, and included three treatments, that is, no amendment as Control, straw amendment (SA), and BA. Incorporation of crop residues into soils after each crop harvest is a typical practice in these regions. For the SA treatments, wheat straw was incorporated into soils at 7.5 t/ha, followed by maize or rice straw at 9.0 t/ha each at the upland and paddy sites, respectively. Biochar was applied to soils at 20 t/ha in June 2014 (i.e., BA treatments). No more biochar was added in the subsequent crop seasons.

Before field application, crop straws were chopped into pieces of 2–3 mm. The biochar was provided by Sanli New Energy Company Henan, China, and was produced from pyrolysis of wheat straw under limited oxygen in a vertical kiln at 450°C with a residence time of 2 hr. The biochar was ground to pass through a 2 mm sieve and homogenized prior to field application. All amending materials were spread evenly onto soil surface and then incorporated into the plow layer with a rake, before sowing of next crop. Selected basic properties of the biochar and crop straws are presented in Table 1.

The field experiment was designed in a completely randomized block with three replicates in each treatment. Each field plot was 4 m × 5 m and separated by a 0.5 m wide buffer zone. Water management, tillage, and fertilizer and pesticide applications at the upland and paddy sites followed typical local crop management practices for each crop type and were kept consistent throughout the field experiments.

2.2 | Soil sampling and characterization

After the harvest of wheat in the upland and paddy sites on June 2017, topsoil samples (0–15 cm) were pooled from six soil cores randomly collected in each field plot using a stainless steel sampler (5 cm in diameter). The soil samples were stored in plastic sealing bags and shipped to the laboratory. Visible plant roots were removed from the soil samples, which were then naturally air-dried and mixed thoroughly to form composite samples. Each composite sample was passed through a 2 mm sieve and then divided into two subsamples. One subsample was used for soil physicochemical analyses and the other for the laboratory incubation experiment.

All analyses of soil properties were undertaken according to the method described by Lu (2000). Briefly, SOC was measured by the K2Cr2O7-H2SO4 oxidation method, total nitrogen (TN) by the micro-Kjeldahl method, and cation exchange capacity (CEC) by the sodium acetate-flame photometric method. Soil pH was measured in a 1:2.5 soil and water mixture. Soil bulk density was determined with a 100 cm³ cylinder in the field. The δ13C value of soil samples was measured by an isotope ratio mass spectrometer (IRMS, MAT253; Thermo Fisher Scientific). Selected physicochemical properties of the soil samples in all treatments are provided in Table 2.

2.3 | Laboratory mineralization incubation experiment

To assess the mineralization of newly added crop residues in the straw and biochar-amended soils, 13C-LMR

| Sample  | Organic C (g/kg) | Total N (g/kg) | Available P (g/kg) | Available K (g/kg) | Ash (%) |
|---------|------------------|----------------|--------------------|--------------------|--------|
| Maize straw | 439.5            | 6.77           | 2.08               | 12.96              | 5.35   |
| Wheat straw | 408.7            | 6.33           | 2.77               | 10.46              | 6.03   |
| Rice straw | 378.9            | 7.10           | 1.79               | 5.94               | 10.52  |
| Biochar   | 486.6            | 10.50          | 6.27               | 33.17              | 20.80  |

TABLE 1 Basic properties of maize, wheat and rice straws, and wheat biochar
LIU et al.

(OC of 48.0%, N of 1.23%, and a δ\textsuperscript{13}C of 557‰) was obtained through uniform \textsuperscript{13}CO\textsubscript{2} labeling in an automatically controlled gas-tight growth chamber according to the protocol described by Zhu et al. (2018) (Materials and Methods S1), and then used as the substrate in the laboratory incubation experiments. The dried LMR was ground to fine particle of 0.25 mm in size, and added as C substrate at 5 g/kg dry soil. The soils without addition of LMR were used for comparison. The incubation experiments had six treatments with three replicates for both the upland and paddy soils: (1) Control, (2) SA, (3) BA, (4) Control plus LMR (Control\textsubscript{M}), (5) SA plus LMR (SA\textsubscript{M}), and (6) BA plus LMR (BA\textsubscript{M}). Each soil sample of 60 g was placed in a 500 ml glass jar with or without an additional 0.3 g of LMR followed by adding water to 60% of soil water holding capacity. The jars were incubated for 140 days in dark at 25°C in an incubator and soil moisture was replenished with deionized water every day by weighing with a balance.

Carbon mineralization was analyzed by measuring CO\textsubscript{2} emission according to the method described by Liu et al. (2019) with minor modification. Briefly, the headspace air of each jar was completely removed by the standard air for 10 min at a rate of 300 ml/min before sampling. Then each jar was sealed and incubated for 4 hr at a constant temperature 25°C in the incubator. The headspace air samples of each jar at the start and 4 hr after jar closure were collected by a gas-tight-locking syringe and then injected into a 20 ml vacuum vial on Days 0.2, 1, 2, 3, 4, 5, 6, 7, 9, 11, 13, 15, 17, 20, 23, 26, 31, 36, 42, 49, 56, 65, 74, 83, 93, 104, 116, and 140 of incubation. The CO\textsubscript{2} concentration was analyzed by gas chromatography (GC; Agilent 7890A).

At the end of incubation, soil MBC was determined by the chloroform-fumigation method (Vance et al., 1987). Soil PLFAs were extracted according to the method described by Frostegard and Baath (1996). Briefly, 3.0 g of freeze-dried soil sample was extracted twice by 15.0 and 10.0 ml single-phase chloroform/methanol/citrate buffer mixture (v/v/v = 1/2/0.8; pH = 4.0). Phospholipids were separated from other lipids through a silicic acid column (Waters, Inc.) and then subjected to mild alkaline methanalysis to form fatty acid methyl esters (FAMEs). The FAME\textsubscript{s} were separated on a gas chromatograph equipped with a flame ionization detector (MIDI Inc.). Methyl nonadecanoate fatty acid (19:0) was added as an internal concentration standard to quantify the phospholipids before derivatization.

The δ\textsuperscript{13}C values of the emitted CO\textsubscript{2} and the freeze-dried MBC samples were measured using an IRMS. The values of δ\textsuperscript{13}C in the individual PLFA were analyzed by GC combustion isotope ratio mass spectrometry (GC–C–IRMS) using a TRACE GC Ultra gas chromatograph with combustion column attached to a GC Combustion III to a Delta V Advantage IRMS (Thermo Finnigan).

### 2.4 | Data analyses

A two-component isotopic mixing model (Balesdent & Balabane, 1992) was used to calculate the proportion (f%) of CO\textsubscript{2}-C or MBC derived from the LMR via Equation (1):

\[
f(\%) = \frac{\delta - \delta_{\text{soil}}}{\delta_{\text{LMR}} - \delta_{\text{soil}}},
\]

where δ is the δ\textsuperscript{13}C of CO\textsubscript{2} or MBC in the LMR amended soils (%), δ\textsubscript{LMR} is the δ\textsuperscript{13}C of the LMR (%), and δ\textsubscript{soil} is the δ\textsuperscript{13}C of the LMR-free treatments (%).

The δ\textsuperscript{13}C of CO\textsubscript{2} was calculated via Equation (2):

\[
\delta_{\text{CO}_2}^{13}C = \frac{\delta_2 \times C_2 - \delta_1 \times C_1}{C_2 - C_1},
\]

where δ\textsubscript{1} and δ\textsubscript{2} are the δ\textsuperscript{13}C values (%) of CO\textsubscript{2} sampled at the start and 4 hr after the jar closure, respectively. C\textsubscript{1} and C\textsubscript{2} are the concentration (μL/L) of CO\textsubscript{2} in the gases sampled at the start and 4 hr after the jar closure.

### TABLE 2

The soil physical and chemical properties at the end of the field trials

| Land-use type | Treatment | SOC (g/kg) | TN (g/kg) | CEC (cmol (+) kg\textsuperscript{-1}) | pH (H\textsubscript{2}O) | BD (g/cm\textsuperscript{3}) | δ\textsuperscript{13}C (‰) |
|--------------|-----------|------------|-----------|----------------|-------------------------|--------------------------|-------------------|
| Upland soil  | Control   | 9.37 ± 0.32 C | 0.90 ± 0.02 B | 14.70 ± 1.65 B | 8.16 ± 0.03 AB | 1.37 ± 0.02 A | −23.3 ± 0.3 A |
|              | SA        | 11.12 ± 0.41 B | 1.04 ± 0.06 A | 14.55 ± 1.80 B | 8.12 ± 0.06 B | 1.26 ± 0.03 B | −23.2 ± 0.1 A |
|              | BA        | 13.52 ± 0.34 A | 1.16 ± 0.06 A | 19.03 ± 2.28 A | 8.27 ± 0.05 A | 1.17 ± 0.04 C | −23.5 ± 0.3 A |
| Paddy soil   | Control   | 27.94 ± 0.39 c | 2.30 ± 0.09 a | 23.40 ± 3.99 b | 7.21 ± 0.04 a | 1.11 ± 0.02 a | −27.1 ± 0.2 a |
|              | SA        | 30.47 ± 0.70 b | 2.42 ± 0.15 a | 24.94 ± 1.62 ab | 7.20 ± 0.02 b | 1.04 ± 0.03 ab | −27.2 ± 0.1 a |
|              | BA        | 33.63 ± 0.62 a | 2.56 ± 0.13 a | 30.33 ± 2.12 a | 7.26 ± 0.05 a | 0.99 ± 0.02 b | −27.3 ± 0.2 a |

Note: Different capital and lowercase characters indicate difference (p < .05) between the treatments in the upland and paddy soils, respectively.

Abbreviations: BA, biochar-amended soil; BD, soil bulk density; CEC, cation exchange capacity; Control, non-amended soil; SA, straw-amended soil; SOC, soil organic carbon; TN, total nitrogen.

Different capital and lowercase characters indicate difference (p < .05) between the treatments in the upland and paddy soils, respectively.
The $\delta^{13}C$ of MBC was calculated using Equation (3) as described by De Troyer et al. (2011):

$$\delta^{13}C_{\text{MBC}} = \frac{(\delta C_{\text{fum}} \times C_{\text{fum}}) - (\delta C_{\text{nffum}} \times C_{\text{nffum}})}{C_{\text{fum}} - C_{\text{nffum}}},$$

where $\delta C_{\text{fum}}$ and $\delta C_{\text{nffum}}$ are the $\delta^{13}C$ values of the fumigated and non-fumigated soil extracts, and $C_{\text{fum}}$ and $C_{\text{nffum}}$ are the total MBC contents of the fumigated and non-fumigated soil extracts, respectively.

Then, the CO$_2$ emission or MBC amount derived from LMR was calculated by multiplying $f\%$ with the total CO$_2$ emission or total MBC amount.

We further calculated metabolic quotient ($q$CO$_2$; mg CO$_2$-C g$^{-1}$ MBC hr$^{-1}$) by dividing the hourly mean of total C mineralization rate with MBC.

The microbial use efficiency (CUE) of the LMR-derived C was calculated according to Equation (4) (Fang, Singh, Collins, et al., 2018):

$$\text{CUE} = \frac{13C_{\text{MBC}}}{13C_{\text{MBC}} + R_{\text{cum}}},$$

where $13C_{\text{MBC}}$ is the LMR-derived MBC (mg/kg soil) and $R_{\text{cum}}$ is the cumulative LMR-C mineralized (mg/kg soil) up to 140 days. The $\delta^{13}C$ of each PLFA molecule was corrected for the δ$^{13}$C of FAME and methanol (MeOH, $\delta^{13}C = -29.33\%$), respectively.

The amount of the LMR-derived C in each PLFA ($P_i$) was calculated according to Equation (6):

$$P_i = M_i \times \frac{\delta C_i - \delta C_c}{\delta C_{\text{LMR}} - \delta C_c},$$

where $M_i$ is the molecular C content of the individual PLFAs, $\delta C_i$ and $\delta C_c$ is the $\delta^{13}C$ of individual PLFAs in the samples with and without LMR addition, respectively, and $\delta C_{\text{LMR}}$ is the $\delta^{13}C$ of LMR. The PLFAs i14:0, a15:0, i15:0, i16:0, a17:0, and i17:0 were used as biomarkers for Gram-positive bacteria (G$^+$-bacteria); 16:1o5c, 16:1o7c, 16:1o9c, cy17:0, 17:1o8c, 18:1o7c, and cy19:0 for Gram-negative bacteria (G$^-$/bacteria); 18:2o6,9c and 18:2o6,9c for fungi 10Me16:0, 10Me17:0, and 10Me18:0 for actinomycete biomass; and 14:0, 15:0, 16:0, 17:0, and 18:0 straight-chain acids for unspecified (universal) soil microbes (Frostegård & Baath, 1996; Zelles, 1999; Zhu et al., 2017).

Data were expressed as mean of three replicates with standard deviation ($\pm SD$). Statistical analyses were carried out using SPSS 23.0 for Windows. The difference in means among various amendment types (Control, straw-, and biochar-amendment) was calculated with one-way ANOVA, followed by Duncan’s post-hoc test at $p < .05$. A two-way ANOVA was used to examine the effects of amendment type, LMR addition, and their interaction effect on the measured parameters across experimental treatments. PLFAs data were standardized to the molar percentages (mol%) of individual PLFAs and principal component analysis (PCA) was performed to examine the variation of microbial community structure using SPSS 23.0.

3 | RESULTS

3.1 | Mineralization of soil organic C and LMR-C

Over the 140-day incubation, the mineralization of SOC was significantly increased by 19.0% and 12.4% in the SA treatment, compared to the Control treatment in the upland soil (959 mg C/kg) and the paddy soil (1,750 mg C/kg), respectively (Figure 1a,b). However, the total SOC mineralization rates of the upland and paddy soils in the BA and Control treatments were similar ($p > .05$). With the LMR addition, the mineralization of SOC was significantly greater in the straw-amended soils, compared to those of non- and biochar-amended soils. In addition, there was no significant interactive effect on SOC mineralization between amendment type and LMR addition ($p > .05$; Table S1).

The LMR-C mineralization of all treatments occurred quickly in the first month and became gradually stable over time (Figure 2a,b). Total LMR-C mineralization amount (% of LMR-C added) ranged from 43.3% to 49.8% and from 45.3% to 51.0% after 140 days of incubation in the upland and paddy soils, respectively. The total mineralization amount of LMR-C in the soils was affected by the soil amendment type. There was no difference in the total LMR-C mineralization amount between SA$_M$ and Control$_M$ ($p > .05$), whereas the total LMR-C mineralization amount was decreased by 12.9% and 11.1% in the BA$_M$ treatment compared with that of the SA$_M$ treatment for the upland and paddy soils ($p < .05$), respectively. Compared to the Control$_M$ treatment, the BA$_M$ treatment decreased the total LMR-C mineralization amount by 10.8% in the upland soil ($p < .05$) and 6.4% in the paddy soil ($p < .1$), respectively.

3.2 | Microbial metabolic quotient and C use efficiency

The $q$CO$_2$ values were significantly affected by the soil amendment type and LMR addition ($p < .05$) without any interaction effect ($p > .05$; Figure 3a,b; Table S2). SA had
no effect on the $q_{CO_2}$ values, compared to the non-amended soil ($p > .05$). For the upland soil, the lowest $q_{CO_2}$ value was found in the BA treatment (1.66 mg CO$_2$ C g$^{-1}$ MBC hr$^{-1}$), which was 15.2% and 22.4% lower than that of the Control and SA treatments, respectively. Similarly, for the paddy soil, the lowest $q_{CO_2}$ value was 1.25 mg CO$_2$ C g$^{-1}$ MBC hr$^{-1}$ in the BA treatment and was 8.9% and 14.5% lower than that of the Control and SA treatments, respectively. With the LMR addition, the $q_{CO_2}$ values were increased by 93.7%–112.3% and 49.8%–56.0% in the upland and paddy soils, respectively.

Microbial CUE of the LMR-derived C was 0.039–0.049 and 0.046–0.052 across all treatments in the upland soils.
and paddy soils, respectively (Figure 4a,b). Compared to the SA_M treatment, the BA_M treatment significantly increased microbial CUE by 23.0% and 21.2% in the upland and paddy soils, respectively. Moreover, for the upland soil, microbial CUE was 24.6% greater in the BA_M treatment than that of the Control_M treatment ($p < .05$). For the upland and paddy soils, SA had no effect on the microbial CUE compared to that of the Control_M treatment ($p > .05$).

### 3.3 Microbial communities and assimilation of LMR-derived C into soil microbial biomass

Soil microbial community structure was clearly controlled by soil type and BA, as indicated by the pattern of PCA (Figure 5a). The biochar-amended soils mainly occupied the negative axis of PC2 due to the high molar percentages of $18:1\omega9c$ and $18:2\omega6,9c$ (the biomarkers for fungi). High
positive loadings on the PC2 axis, such as in the ControlM and SAM, correlated with greater molar percentages of i15:0 and i17:0 (G⁺-bacteria) and 18:1ω7c (G⁻-bacteria; Figure 5b).

The ¹³C-LMR was not equally distributed among different microbial groups, indicating different microorganisms had different abilities on their uptake and utilization of C-LMR (Figure 6a,b). G⁻-bacteria had the highest relative abundance of 36.8%–43.2%, whereas fungi had a low relative abundance of 10.1%–15.3% across all treatments at the end of incubation. Compared to those of the SAM treatment, the percentage of ¹³C-LMR distribution in G⁻-bacteria in the BAM treatment was significantly smaller by 8.8% and 8.2% in the upland and paddy soils, respectively. However, the BAM treatment significantly increased the ¹³C-LMR percentages in fungi by 13.3% and 28.3% from those of the SAM treatment in the upland and paddy soils, respectively.

4 | DISCUSSION

4.1 | Soil C sequestration potential: Straw incorporation versus biochar amendment

Straw incorporation and BA are important measures to increase soil C sequestration in agriculture ecosystems (Liu et al., 2014; Smith, 2016). In this study, BA significantly increased SOC content (Table 2) and reduced SOC mineralization in the upland and paddy soils, compared to straw incorporation (Figure 1). The measurements of the SOC mineralization rates further demonstrated the higher stability of SOC in the biochar-amended soils (Table S3), as straw incorporation did not change, but BA significantly decreased the mineralization rates of SOC. The lower C mineralization in soils after long-term BA has been reported in other studies. For example, Chen et al. (2018) found that 3 years after biochar addition to soils at 20 and 40 t/ha, soil basal respiration rates were significantly decreased by 12% and 20% during a 31-day incubation. Similarly, Liu et al. (2019) reported that a soil amended with biochar 6 years earlier had the total C mineralization decreased by 38.8% compared with that of the biochar-free Control during a 64-day incubation. The lower C mineralization in biochar-amended soils may be related to the protection of C substrates by sorption into biochar pore structures and the change of C pool composition. First, the C pool composition was changed due to SA and BA. Most of the straw was quickly decomposed once it was returned to soils as fresh organic matter, and the residues were transformed into labile and active soil organic C fractions (Liu et al., 2014). In contrast, only easily decomposable organic matter in biochar may have been preferentially decomposed in the field (Fang et al., 2017) and the remaining C in biochar would become increasingly recalcitrant (Yi et al., 2020). A meta-analysis found that the mean residence times of recalcitrant and labile C fractions in biochar were about 556 years and 108 days with pool sizes of 97% and 3%, respectively (Wang et al., 2016). Therefore, BA would increase the content of slow and resistant C pools in soils (Lehmann et al., 2006). In this study, we found that BA significantly decreased the size of the easily mineralizable C pool and the mineralization rate of the slow C pool, comparing with those of the Control and SA treatments (Table S3). Similarly, Ameloot et al. (2014) reported that the size of the easily mineralizable C pool and the mineralization rate of the slow C pool was significantly lower in the BA than in the Control treatment for a sandy clay loam soil. Furthermore, in the mineralization curves model, we also found that BA (cf. non-amendment) had no effect on the labile C pool decay rate of LMR, but increased the stable C fraction and decreased the stable C pool decay rate of LMR (Table S4). Our result was partly in agreement with Kerré et al. (2017) who found that the stable C fraction of added maize was larger and degradation rates were lower in the charcoal-enriched soil than in the adjacent non-charcoal soil. Second, biochar has a strong protective effect on C substrates. Because biochar has a myriad of macro-, meso-, and micro-pores and a relatively large surface area (Downie et al., 2009), soil organic matters (especially dissolved small aliphatic organic molecules) could be absorbed into the meso- and micro-pores of biochar (Smeebye et al., 2016), and thus become isolated from decomposing microbes. The high porosity and surface area of biochar also could stabilize other sources of organic compounds in soil through adsorption (Kleber et al., 2007). For example, BA could enhance the stabilization of plant-derived labile organic matter in soil (Fang et al., 2017; Weng et al., 2015). And Keith et al. (2011) found that wood biochar addition reduced the mineralization of labile organic matter (sugar cane residue), and the magnitude of this negative priming effect was enhanced with increasing rates of labile organic matter addition. This finding suggested that BA could improve the stability of LMR-C by strong sorption to the biochar due to its large porosity and surface area (Jiang et al., 2019).

4.2 | Microbial mechanisms of C sequestration in straw and biochar amendment

The qCO₂ parameter could be considered as an indicator for the microbial use of C substrates for their energy consumption (Anderson & Domsch, 1993). We found that qCO₂ was lower in the biochar-amended soils, and remained unchanged in the straw-amended soils, when comparing with that of the Control treatment (Figure 3). A lower qCO₂ value in the biochar-amended acidic paddy soil was also reported by Zheng et al. (2016), that is, the qCO₂ value was decreased from 0.84 in the Control treatment to 0.49–0.58 in the biochar treatments. Several studies had found that microorganisms colonized
within and near biochar particles, with the most likely area for microbial colonization referred as “charsphere,” which is the interface between soil matrix and biochar and provides a unique and preferred micro-environment for soil microbes (Luo et al., 2013). The coexistence of substrate, nutrients, and microorganisms at the “charsphere” may result in a high CUE (Lehmann et al., 2011). Soil microorganisms produced more cell mass per unit mass of consumed C (i.e., soil microbial CUE) in the biochar-amended soils than in the Control and straw-amended soils, and thus could help enhance the C sequestration in biochar-amended soil, as elucidated below.

It is now recognized that microbial C sequestration potential in soils depends on the microbial utilization efficiency of C substrates, namely the balance between microbial decomposition of organic C and stabilization of microbial assimilated C (Malik et al., 2019). Microbial CUE is related to the SOC quality and C/N ratio. In this study, BA significantly increased the soil C/N ratio (Table 2) and improved SOC quality as revealed by lower SOC mineralization rates (Table S3). As a result, a greater microbial CUE was observed in the biochar-amended soils than the straw-amended and Control soils (Figure 4; Silva-Sánchez et al., 2019). Additionally, soil microbial CUE may increase with soil oxygen content (Parsons & Smith, 1989). Thus, BA likely increases air-filled porosity in soils and subsequently soil oxygen concentration (Jiang et al., 2016) and microbial CUE, due to high porosity of biochar. Furthermore, nutrient deficiency could increase respiration or C excretion, resulting in a decline of the microbial CUE (Manzoni & Porporato, 2009). Biochar has been proposed as an effective means to improve soil fertility and nutrient retention (Lehmann et al., 2011). Soil nutritional stress is inversely proportional to the ratio of cyclopropyl fatty acids (cy17:0 + cy19:0)/monoenic precursors (16:1o7c + 18:1o7c; Bossio & Scow, 1998). The biochar-amended soils had the lowest ratio of cyclopropyl fatty acids/monoenic precursors (Figure S1), suggesting that the nutritional stress was alleviated in the BA (Fierer et al., 2003). It was suggested that high porosity in biochar could result in greater nitrogen retention in soils (Fiorentino et al., 2019; Shi et al., 2020), which would facilitate microbial assimilation of C substrates. Interestingly, we found that BA increased microbial CUE compared with the Control treatment, and this increase was more pronounced in the upland soils than in the paddy soils (Figure 4). Our results were supported by Jiang et al. (2016) who reported that 1% biochar addition increased microbial biomass and CUE, especially for the sandy clay loam soil in Colorado with the lowest SOC and TN. Thus, the lower SOC and TN in the upland soils than in the paddy soils (Table 2) may explain the more pronounced increase in microbial CUE in the upland soils.

Incorporation of straw and biochar into soils can stimulate microbial activities and increase microbial biomass (Jiang et al., 2016; Pan et al., 2016). In this study, SA and BA significantly increased total PLFA concentration (Figure S2a,b), and changed microbial community structure (Figure 5; Figure S2c,d), indicating the choice of substrates dictated specific growth and reproduction of soil microorganisms. The C/N ratio of the substrates regulated microbial community composition due to close relationship between the C/N ratio of substrates and decomposing microorganisms (Cui et al., 2020). In fact, fungi can utilize substrates of wider C/N ratio than that of substrates used by bacteria (Hogberg et al., 2007). Moreover, fungi could utilize complex C substrates in the later stages of organic matter decomposition due to their extensive range of extracellular enzymatic capabilities (Boer et al., 2005). Biochar residue had high C/N ratios and condensed aromatic substances, resulting in increases of fungi proportion in soils 3 years after BA (Figure S2c,d). However, despite high C/N ratios of straw, continuous incorporation of straw with high labile organic matter content into soils is more conducive to bacterial growth, especially for G+-bacteria. In addition, microbial community composition would be altered due to the changing soil physicochemical characteristics such as soil water holding capacity, porosity, pH, and CEC (Jiang et al., 2016; Spohn et al., 2016) in straw- and biochar-amended soils. The adaptive evolution of microbial community may further affect the microbial CUE due to long-term BA and continuous straw incorporation. In this study, the 13C-LMR was primarily assimilated into bacteria (G+-bacteria, G+ bacteria, and universal) biomass, accounting for about 70% of the total 13C-PLFAs (Figure 6). Thus, bacteria act as the main decomposer for added fresh residue, similar to the findings of Pan et al. (2016) who reported that the substrate of 13C-labeled rice straw assimilated into bacteria was 74.3% of total 13C-labeled PLFAs. However, the 13C-labeled fungi only accounted for a relatively small fraction (10.1%–15.3%) of the total 13C-labeled PLFAs. Nonetheless, a significantly higher percentages of 13C-labeled fungi was found in the biochar-amended soils than in non- and straw-amended soils (Figure 6). The different distribution of 13C-LMR into bacteria and fungi revealed the differences in the microbial CUE and the preferential utilization of C substrate for soil microbial communities in the non-, straw-, and biochar-amended soils. Fungal biomass is usually thought to contribute more to the formation of soil organic matter than bacteria biomass (Clemmensen et al., 2013) because fungal CUE is higher than bacterial CUE. Six et al. (2006) previously reported that soil microbial communities dominated by fungi can sequester more C than soils with higher bacterial abundance. Therefore, BA increased the fungal abundance and their ability to utilize organic matter, which, in turn, resulted in a higher microbial CUE than that of straw-amended soils. As a high CUE value indicates efficient microbial growth and relatively lower C release to the atmosphere (Manzoni et al., 2012), BA could be beneficial for sustaining microbial activities and improving soil C storage (Jiang et al., 2016).
This study investigated the effects of SA and BA on microbial CUE and C sequestration in both upland and paddy soils, and its findings have important implications in sustaining SOC levels and increasing C sequestration. BA significantly decreased the mineralization of SOC and newly added crop residue in both upland and paddy soils. Moreover, significantly lower $q_{\text{CO}_2}$ and higher microbial CUE values were found in the soils amended with biochar compared to those of straw-amended soils, suggesting greater C sequestration potential with BA. The $^{13}$C-PLFA analysis showed that enhancement of CUE in BA was attributed to the shift of microbial utilization of C substrates by increasing the proportion of fungi relative to bacteria when comparing with SA. Overall, this study suggested that conversion of straw to biochar and subsequent BA could increase C sequestration potential by enhancing microbial CUE in agricultural soils, especially for the upland soils with lower SOC levels. Even infrequent application of biochar into soils in cropping systems that otherwise continuously practice straw incorporation could be beneficial for increasing SOC levels and stabilizing labile C pool, thus improving agricultural sustainability.

ACKNOWLEDGEMENTS

This work was partially supported by the National Natural Science Foundation of China (grant numbers: 41877097, 41877096, 41371300, and U1612441), and Asia Hub Seed Funding (grant number: 2018-AH-02). We are grateful to many staff and graduate students involved in maintaining the field experiments who are not listed as co-authors.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

ORCID

Rongjun Bian https://orcid.org/0000-0002-5571-8242
Tida Ge https://orcid.org/0000-0003-0422-6122
Jufeng Zheng https://orcid.org/0000-0002-1304-2803
Kun Cheng https://orcid.org/0000-0002-6101-0558
Genxing Pan https://orcid.org/0000-0001-9755-0532

REFERENCES

Ameloot, N., Sletfelt, S., Case, S. D. C., Alberti, G., Mtnamara, N. P. Zavalloni, C., Vervisch, B., Vedove, G. D., & De Neve, S. (2014). C mineralization and microbial activity in four biochar field experiments several years after incorporation. Soil Biology and Biochemistry, 78, 195–203. https://doi.org/10.1016/j.soilbio.2014.08.004
Anderson, T. H., & Domsch, K. H. (1993). The metabolic quotient for CO$_2$ ($q_{\text{CO}_2}$) as a specific activity parameter to assess the effects of environmental conditions, such as pH, on the microbial biomass of forest soils. Soil Biology and Biochemistry, 25, 393–395. https://doi.org/10.1016/0038-0717(93)90140-7
Balesdent, J., & Balaban, M. (1992). Maize root-derived soil organic carbon estimated by natural $^{13}$C abundance. Soil Biology and Biochemistry, 24, 97–101. https://doi.org/10.1016/0038-0717(92)90264-X
Boer, W. D., Folman, L. B., Summerbell, R. C., & Boddy, L. (2005). Living in a fungal world: Impact of fungi on soil bacterial niche development. FEMS Microbiology Reviews, 29, 795–811. https://doi.org/10.1016/j.femsre.2004.11.005
Bossoio, D. A., & Scow, K. (1998). Impacts of carbon and flooding on soil microbial communities: Phospholipid fatty acid profiles and substrate utilization patterns. Microbial Ecology, 35, 265–278. https://doi.org/10.1007/s002489900082
Chen, J. H., Sun, X., Zheng, J. F., Zhang, X. H., Liu, X. Y., Bian, R. J., Li, L. Q., Cheng, K., Zheng, J. W., & Pan, G. X. (2018). Biochar amendment changes temperature sensitivity of soil respiration and composition of microbial communities 3 years after incorporation in an organic carbon-poor dry cropland soil. Biology and Fertility of Soils, 54, 175–188. https://doi.org/10.1007/s00374-017-1253-6
Chen, X., Xia, Y., Rui, Y., Ning, Z., Hu, Y., Tang, H., He, H., Li, H., Kuzyakov, Y., Ge, T., Wu, J., & Su, Y. (2020). Microbial carbon use efficiency, biomass turnover, and necromass accumulation in paddy soil depending on fertilization. Agriculture, Ecosystems & Environment, 292. https://doi.org/10.1016/j.agee.2020.106816
Clemmensen, K. E., Bahr, A., Ovaskainen, O., Dahlberg, A., Ekblad, A., Wallander, H., Stenlid, J., Finlay, R. D., Wardle, D. A., & Lindahl, B. D. (2013). Roots and associated fungi drive long-term carbon sequestration in boreal forest. Science, 339, 1615–1618. https://doi.org/10.1126/science.1231923
Corrufo, M. F., Soong, J. L., Horton, A. J., Campbell, E. E., Haddix, M. L., Wall, D. H., & Parton, W. J. (2015). Formation of soil organic matter via biochemical and physical pathways of litter mass loss. Nature Geoscience, 8, 776–779. https://doi.org/10.1038/ngeo2520
Cui, J., Zhu, Z., Xu, X., Liu, S., Jones, D. L., Kuzyakov, Y., Shibistova, O., Wu, J., & Ge, T. (2020). Carbon and nitrogen recycling from microbial necromass to cope with C:N stoichiometric imbalance by priming. Soil Biology and Biochemistry, 142. https://doi.org/10.1016/j.soilbio.2020.107720
De Troyer, I., Amery, F., Van Moorleghem, C., Smolders, E., & Merckx, R. (2011). Tracing the source and fate of dissolved organic matter in soil after incorporation of a $^{13}$C labelled residue: A batch incubation study. Soil Biology and Biochemistry, 43, 513–519. https://doi.org/10.1016/j.soilbio.2010.11.016
Downie, A., Crosby, A., & Munroe, P. (2009). Physical properties of biochar. In J. Lehmann & S. Joseph (Eds.), Biochar for environmental management: Science and technology (pp. 13–32). Earthscan.
Fang, Y., Singh, B. P., Collins, D., Li, B., Zhu, J., & Tavakkoli, E. (2018). Nutrient supply enhanced wheat residue-carbon mineralization, microbial growth, and microbial-carbon-use efficiency when residues were supplied at high rate in contrasting soils. Soil Biology and Biochemistry, 126, 168–178. https://doi.org/10.1016/j.soilbio.2018.09.003
Fang, Y., Singh, B. P., Luo, Y., Boersma, M., & Van Zwieten, L. (2018). Biochar carbon dynamics in physically separated fractions and microbial use efficiency in contrasting soils under temperate pastures. Soil Biology and Biochemistry, 116, 399–409. https://doi.org/10.1016/j.soilbio.2017.10.042
Fang, Y., Singh, B. P., Matta, P., Cowie, A. L., & Van Zwieten, L. (2017). Temperature sensitivity and priming of organic matter with different stabilities in a Vertisol with aged biochar. Soil Biology and Biochemistry, 115, 346–356. https://doi.org/10.1016/j.soilbio.2017.09.004
Fierer, N., Schimel, J. P., & Holden, P. A. (2003). Variations in microbial community composition through two soil depth profiles. Soil
Total Environment, 701, 134424. https://doi.org/10.1016/j.scitotenv.2019.134424

Silva-Sánchez, A., Soares, M., & Rousk, J. (2019). Testing the dependence of microbial growth and carbon use efficiency on nitrogen availability, pH, and organic matter quality. Soil Biology and Biochemistry, 134, 25–35. https://doi.org/10.1016/j.soilbio.2019.03.008

Sinsabaugh, R. L., Manzoni, S., Moorhead, D. L., & Richter, A. (2013). Carbon use efficiency of microbial communities: Stoichiometry, methodology and modelling. Ecology Letters, 16, 930–939. https://doi.org/10.1111/ele.12113

Six, J., Frey, S. D., Thiet, R. K., & Batten, K. M. (2006). Bacterial and fungal contributions to carbon sequestration in agroecosystems. Soil Science Society of America Journal, 70, 555–569. https://doi.org/10.2136/sssaj2004.0347

Smebye, A., Alling, V., Vogt, R. D., Gadmar, T. C., Mulder, J., Cornelissen, G., & Hale, S. E. (2016). Biochar amendment to soils changes dissolved organic matter content and composition. Chemosphere, 142, 100–105. https://doi.org/10.1016/j.chemosphere.2015.04.087

Smith, P. (2016). Soil carbon sequestration and biochar as negative emission technologies. Global Change Biology, 22, 1315–1324. https://doi.org/10.1111/gcb.13178

Sohi, S. P. (2012). Carbon storage with benefits. Science, 338, 1034–1035. https://doi.org/10.1126/science.1225987

Soil Survey Staff. (1994). Keys to soil taxonomy (6th ed., pp. 161–186). US Department of Agriculture. Soil Conservation Service.

Spohn, M., Pötsch, E. M., Eichorst, S. A., Woebken, D., Wanek, W., & Richter, A. (2016). Soil microbial carbon use efficiency and biomass turnover in a long-term fertilization experiment in a temperate grassland. Soil Biology and Biochemistry, 97, 168–175. https://doi.org/10.1016/j.soilbio.2016.03.008

Vance, E. D., Brookes, P. C., & Jenkinson, D. S. (1987). An extraction method for measuring soil microbial biomass-C. Soil Biology and Biochemistry, 19, 703–707. https://doi.org/10.1016/0038-0717(87)90052-6

Wang, J., Xiong, Z., & Kuyzakov, Y. (2016). Biochar stability in soil: Meta-analysis of decomposition and priming effects. GCB Bioenergy, 8, 512–523. https://doi.org/10.1111/gcbb.12266

Weng, Z., Van Zwieten, L., Singh, B. P., Kimber, S., Morris, S., Cowie, A., & Macdonald, L. M. (2015). Plant-biochar interactions drive the negative priming of soil organic carbon in an annual ryegrass field system. Soil Biology and Biochemistry, 90, 111–121. https://doi.org/10.1016/j.soilbio.2015.08.005

Wrh, I. W. G. (2015). World reference base for soil resources 2014, update 2015. International soil classification system for naming soils and creating legends for soil maps. World Soil Resources Rep. No. 106. FAO.

Wu, L., Zhang, W., Wei, W., He, Z., Kuyzakov, Y., Bol, R., & Hu, R. (2019). Soil organic matter priming and carbon balance after straw addition is regulated by long-term fertilization. Soil Biology and Biochemistry, 135, 383–391. https://doi.org/10.1016/j.soilbio.2019.06.003

Xu, X., Cheng, K., Wu, H., Sun, J., Yue, Q., & Pan, G. (2019). Greenhouse gas mitigation potential in crop production with biochar soil amendment—a carbon footprint assessment for cross-site field experiments from China. GCB Bioenergy, 11, 592–605. https://doi.org/10.1111/gcbb.12561

Ye, G., Lin, Y., Kuyzakov, Y., Liu, D., Luo, J., Lindsey, S., Wang, W., Fan, J., & Ding, W. (2019). Manure over crop residues increases soil organic matter but decreases microbial necromass relative contribution in upland Ultisols: Results of a 27-year field experiment. Soil Biology and Biochemistry, 134, 15–24. https://doi.org/10.1016/j.soilbio.2019.03.018

Yi, Q., Liang, B., Nan, Q., Wang, H., Zhang, W., & Wu, W. (2020). Temporal physicochemical changes and transformation of biochar in a rice paddy: Insights from a 9-year field experiment. Science of the Total Environment, 721, 137670. https://doi.org/10.1016/j.scitotenv.2020.137670

Yousaf, B., Liu, G., Wang, R., Abbas, Q., Imtiaz, M., & Liu, R. (2017). Investigating the biochar effects on C-mineralization and sequestration of carbon in soil compared with conventional amendments using the stable isotope (δ13C) approach. GCB Bioenergy, 9, 1085–1099. https://doi.org/10.1111/gcbb.12401

Zelles, L. (1999). Fatty acid patterns of phospholipids and lipopolysaccharides in the characterisation of microbial communities in soil: A review. Biology and Fertility of Soils, 29, 111–129. https://doi.org/10.1007/s003740050533

Zheng, J. F., Chen, J. H., Pan, G. X., Liu, X. Y., Zhang, X. H., Li, L. Q., Bian, R. J., Cheng, K., & Zheng, J. W. (2016). Biochar decreased microbial metabolic quotient and shifted community composition four years after a single incorporation in a slightly acid rice paddy from southwest China. Science of the Total Environment, 571, 206–217. https://doi.org/10.1016/j.scitotenv.2016.07.135

Zhran, M., Ge, T., Tong, Y., Deng, Y., Wei, X., Lynn, T. M., Zhu, Z., Wu, J., & Gunina, A. (2020). Assessment of depth-dependent microbial carbon use efficiency in long-term fertilized paddy soil using an 13O-H2O approach. Land Degradation & Development. https://doi.org/10.1002/ldr.3708

Zhu, Z., Ge, T., Hu, Y., Zhou, P., Wang, T., Shibistova, O., Guggenberger, G., Su, Y., & Wu, J. (2017). Fate of rice shoot and root residues, rhizodeposits, and microbial assimilated carbon in paddy soil – Part 2: Turnover and microbial utilization. Plant and Soil, 416, 243–257. https://doi.org/10.1007/s11104-017-3210-4

Zhu, Z., Ge, T., Luo, Y., Liu, S., Xu, X., Tong, C., Shibistova, O., Guggenberger, G., & Wu, J. (2018). Microbial stoichiometric flexibility regulates rice straw mineralization and its priming effect in paddy soil. Soil Biology and Biochemistry, 121, 67–76. https://doi.org/10.1016/j.soilbio.2018.03.003

Zimmerman, A. R., Gao, B., & Ahn, M.-Y. (2011). Positive and negative carbon mineralization priming effects among a variety of biochar-amended soils. Soil Biology and Biochemistry, 43, 1169–1179. https://doi.org/10.1016/j.soilbio.2011.02.005

SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section.

How to cite this article: Liu Z, Wu X, Liu W, et al. Greater microbial carbon use efficiency and carbon sequestration in soils: Amendment of biochar versus crop straws. GCB Bioenergy. 2020;12:1092–1103. https://doi.org/10.1111/gcbb.12763