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Effects of biochar amendment on soil carbon dioxide emission and carbon budget in the karst region of southwest China

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ABSTRACT

Knowledge about the influence of tobacco biochar (BC) on soil carbon dioxide (CO_2) emissions and carbon (C) budget in a karst region of southwest China is limited. The karst field experiment was conducted in randomly arranged plots and evaluated five treatments: a plot without BC addition (B_0), with 1.0 t BC ha^{-1} (B_1C_1), 10 t BC ha^{-1} ($B_{10}C_{10}$), 25 t BC ha^{-1} ($B_{25}C_{25}$) and 50 t BC ha^{-1} ($B_{50}C_{50}$). Soil CO_2 emissions were assessed using closed chambers at 10-day intervals during a tobacco-growing season. Soil temperature and moisture were simultaneously measured at 5-, 10-, and 20-cm depths. BC amendment increased tobacco productivity, soil organic matter, total nitrogen, available phosphorus, and available potassium contents but decreased total sulfur content. Compared with the control (B_0), daily average soil CO_2 fluxes significantly increased by 20.0%, 26.3%, 39.4% and 50.2% in the B_1, B_{10}, B_{25}, and B_{50} treatments, respectively. The cumulative soil CO_2 emissions for the entire tobacco-growing season were significantly higher in BC amendments than in the control. Furthermore, soil CO_2 fluxes were observed to be positively correlated with soil temperature but negatively correlated with soil moisture. Soil respiration sensitivity to temperature (Q_{10}) significantly increased at greater soil depths, whereas decreased in BC-amended soils. Moreover, the BC amendments significantly increased C gain from −1.17 t ha^{-1} in the B_0 treatment to 19.48 t ha^{-1} in the B_{50} treatment. Based on C budget, the increase in CO_2 emissions in the BC-treated soil was compensated by the higher tobacco biomass and soil C storages. These results suggest that the application of tobacco BC to karst farmland enhances the soil C sequestration.

1. Introduction

Carbon dioxide (CO_2) has been implicated in global climate warming based on higher concentrations emitted by the terrestrial ecosystem (Melillo et al., 2002). In 2010, total GHG emissions from agriculture were estimated to be 5.24 Gt CO_2 equivalents yr^{-1}, which corresponds to 11% of the total global anthropogenic emissions (Pearson et al., 2017). Hence, reducing GHG emissions by altering agricultural management schemes is warranted to mitigate climate change.

Biochar (BC) pertains to the carbon-rich solid that remains after pyrolysis of waste biomass (e.g., agricultural and forestry residues) using limited or absent oxygen conditions (Lehmann, 2007). The meta-analysis shows that BC has a very low turnover rate (0.0046% day^{-1}), which can persist in soils on the centennial time scale (Wang et al., 2016a). Therefore, BC amendment may be utilized as a strategy for mitigating climate change by increasing soil organic carbon (SOC) sequestration if BC-C itself is stable in soil for millennia or the interaction of BC with soil lasts for a long time (Lorenz and Lal, 2014). Meanwhile, BC addition can influence soil CO_2 emissions by altering the soil structure, moisture content, cation exchange capacity, enzyme activity, and soil biota (Lehmann et al., 2011). BC is obtained from various source materials that have different carbonization processes, and thus their properties and effects may greatly vary with local environmental conditions and employed management schemes (Saarnio et al., 2013). On average, meta-analysis showed that CO_2 emissions significantly increased from 22% to 28% in BC-amended soils, thereby reducing BC

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carbon sequestration potential (He et al., 2017; Zhang et al., 2019a). However, BC amendment affected soil CO_2 emissions with different magnitudes and even directions in a large number of individual studies (Liang et al., 2010; Ameloot et al., 2013; Lu et al., 2014). Consequently, effects of BC amendments on CO_2 emissions and climate change mitigation potential cannot be generalized as the effects are BC-, site-and plant-specific and thus need further investigation (Lorenz and Lal, 2014).

Recent studies on the effects of BC amendments on soil CO_2 emissions as well as C budget have been conducted in grasslands (Slavich et al., 2013), forests (Zhou et al., 2017; Ge et al., 2019) and croplands include such food crops as maize, rice, and wheat (Karhu et al., 2011; Fan et al., 2017; Zhang et al., 2017), whereas those using cash crops, such as tobacco are limited. Tobacco planting involves ridging, mulching, and transplanting, which are different from other crops. In addition, flue-cured tobacco is an important cash crop in southwest China, particularly in poor karst regions. Karst areas are known for their sensitivity to land degradation facilitated by human disturbances (e.g. agricultural activities) (Li et al., 2017). In the past, most of the arable soils in the karst areas have been degraded because of soil erosion and SOC loss (Yang et al., 2016). However, the distinct geological environment and climatic conditions of karst regions are essential production bases for generating high quality tobacco leaves, which are a very important component of the local economy. Tobacco stalks are the biggest byproducts of tobacco production, which is generally burned in the field after separating from the marketable leaves. This management scheme may thus result in severe environment pollution and pose health risks to humans (Chen et al., 2017). Theoretically, BC produced from tobacco stalk residues to soils could mitigate environmental pollution and hypothetically improve the tobacco productivity by the result of direct supply of nutrients and enhanced nutrient retention as well as changes in soil microbial community structure under BC amendments (Lehmann et al., 2011; Ding et al., 2016). Due to its proven longevity, BC addition to soil may mitigate climate change by increasing soil carbon sequestration, improving soil fertility, and enhancing agricultural productivity. However, current understanding of the impact of BC amendment on soil CO_2 flux and C budget specifically in a tobacco farming system in the karst region is limited.

Moreover, soil respiration is a major contributor to the rise of atmospheric CO_2, which mainly depends on soil temperature and moisture. BC amendment could change their patterns (He et al., 2016). However, little is known about the effect BC amendment on the temperature sensitivity of soil respiration (also refers to as Q_{10}, the rate of increase in soil respiration when temperature increases by 10 °C) (Fang et al., 2015). The Q_{10} value may change with changes in substrate availability, soil carbon quality and stabilization, and consequently alter the responses of soil CO_2 emissions (Conant et al., 2008; Frey et al., 2013). In general, BC can be considered a chemically and biologically aromatic C structure, and therefore may have a profound effect on soil carbon stabilization as well as microbial activities (Keith et al., 2011; Lehmann et al., 2011). We hypothesized that BC might also affect the sensitivity of soil respiration to temperature. Understanding this is critical for predicting the utility of BC as a strategy to mitigate and adapt to climate change. In doing so, we established a field experiment adding BC obtained from tobacco-stalk residues to karst soils to determine soil CO_2 emissions and C budget. The main objectives were to investigate the following: (1) effects of BC amendment on soil CO_2 emissions and the temperature sensitivity of soil CO_2 efflux (Q_{10}); (2) changes of tobacco growth in karst farmlands amended with BC at different levels; (3) alterations in C budget in the tobacco ecosystem after BC application in karst regions.

2. Materials and methods

2.1. Experimental site

The study was performed at the Pingba Tobacco Experimental Station (26°26′193″ N, 106°14′166″ E; elevation 1, 391 m above sea level), Guizhou Academy of Tobacco Science, Guiyang city, Guizhou province, China. The site experiences a subtropical humid monsoon climate that includes an average annual temperature of 14.3 °C, as well as an average annual rainfall of 1, 244 mm across four distinct seasons. The soil used for this study is classified as yellow soil in the Genetic Soil Classification of China (GSCC), equivalent to Ferric Alisols in the World Reference Base for Soil Resources (WRB). The soil of this station was clay loam in texture and had the following basic properties: pH (H_2O) of 6.58; soil organic matter of 36.77 g kg^{-1}; total N, P, and K contents of 2.10, 0.55, and 13.00 g kg^{-1}, respectively; and available N, P, and K contents of 145.56, 6.27,and 250.50 mg kg^{-1}, respectively.

2.2. BC amendment

The BC used in the field experiment was produced from the large-scale continuous production. Briefly, tobacco stalks were collected by farmers, then shipped to and air-dried in the warehouse. After that, BC was prepared from dry tobacco stalks at a temperature between 350 and 600 °C in a metallic kiln with oxygen-limiting conditions. For the field study, the BC was extensively mixed and then ground to pass through a 1-mm sieve prior to use as a soil amendment. The BC was characterized by pH (H_2O) of 8.34, 413.24 g kg^{-1}C, 12.67 g kg^{-1}N, and a C/N ratio of 32.63.

2.3. Field experiments

The experimental design consisted of a completely randomized block design with three replicates, with a buffer zone of 1.5 m between every two plots. BC was spread manually across the surface in April 2018, and extensively mixed into the plow layer (0–20 cm) at rates of 0, 1, 10, 25, and 50 t ha^{-1}. These treatments are hereafter referred to as B_0, B_1, B_{10}, B_{25}, and B_{50}, respectively. No BC amendment was performed in the following year. The special base fertilizer of flue-cured tobacco (N: P_2O_5: K_2O = 10:10:25) was applied at rates of 675 kg ha^{-1} across all plots before the high row ridge (30 cm). The uniformly sized tobacco seedlings of *Nicotiana tabacum L.* (K326, Northup King Seed Company) were directly transplanted by hand from the seedbeds and into the row ridges. For tobacco transplantation, 1.1-m row spacing and 0.55-m row distance were adapted. A total of 32 plants were planted in each plot, which had an area of 19.36 m^2. Tobacco production in the field experiment depends solely on natural precipitation. The consistent management practices (cultivation and pest control) were carried out following the local convention during the growing seasons. Soil temperature and moisture sensors were placed in each plot at soil depths of 5-, 10-, and 20-cm, which were recorded using data loggers (TR-6, Shunkeda, China). The field growth period of flue-cured tobacco was about 120 days.

2.4. Soil CO_2 emission monitoring

The CO_2 fluxes were assessed throughout the whole tobacco-growing season (i.e., May to September 2018) by static chamber-gas chromatography method. We inserted chamber bases (10-cm height and 30-cm diameter) with a circular-shaped groove at a depth of 7 cm in each plot. These bases were maintained on-site during the entire monitoring period. Once the moveable opaque polyvinyl chloride (PVC) chamber (50-cm height) was placed over the base, the groove was filled with water to act as air seal. The digital thermometers are arranged inside the chamber to record the air temperature during sampling. The chamber had an electric fan installed to ensure complete gas mixing during the collection. No plants were included in the champers. A 30-mL air sample was obtained from the chamber at different time points (0, 8, 16, and 24 min), followed by injection into a evacuated 12-mL glass bottle, which was vacuum-sealed using a butyl rubber stopper as well as a plastic cap. After tobacco transplanting, soil CO_2 emissions were conducted at 10-day intervals between 8:00 AM and 11:00 AM. At the same time, we also measured soil temperature and moisture using probes located at different soil depths (5, 10, and 20 cm). CO_2 concentrations were measured by gas chromatography (7890A, Agilent Technologies, Santa Clara, CA, USA) that was equipped with a Gilson autosampler (Sample Changer 223, Gilson Inc., Middleton, USA). We converted CO_2 into CH_4 using a nickel reforming furnace, followed by detection using a flame-ionization detector (FID) at a temperature of 250 °C.

To ensure continued measurement accuracy and reliability, the GC was calibrated using three certified standard gases, comprising 398 ppm, 603 ppm, and 809 ppm CO_2 (Chengdu Chenggang Messer Gas Products Co. Ltd, China). Calculation of CO_2 emission rates was performed using the increase in the concentration per unit surface area of the chamber at various time points. Gas sample sets were discarded unless these yielded a linear regression value of $r^2 > 0.90$. Soil CO_2 emission flux was calculated as follows (Beetz et al., 2013):

$F = k \times 273/(273 + T) \times (\Delta c/\Delta t) \times (V/A)$,where F is the CO_2 flux (mg CO_2-C $m^{-2}h^{-1}$), k is a unit conversion factor for calculating CO_2 flux rate (0.536 kg C m^{-3}), T is the temperature of chamber during sampling (°C), $\Delta c/\Delta t$ is the rate CO_2 concentration increase in the chamber (ppm h^{-1}), V is the volume of chamber (m^3), and A is the surface area of chamber (m^2).

Cumulative soil CO_2 emissions during growth were calculated using the following equation:

$M_{CO2} = \sum_{i=1}^{n} (F_i + F_{i+1})/2 \times (t_{i+1} - t_i) \times 24$, where M_{CO2} is the cumulative soil CO_2 emission, F_{i+1} and F_i are, respectively, the soil CO_2 emission rate at the $(i + 1)$ th and ith sampling dates, t_{i+1}-t_i is the time interval (days) between two adjacent measurements ($(i + 1)$ th and i th), and n is the total number of samplings conducted during the growing season.

An exponential-exponential function was used to describe the relationship between soil CO_2 emissions and soil temperature and moisture (Lai et al., 2012),

$F = ae^{bW}e^{cT}$where a, b, and c are fitted constants, W is the soil moisture (%), and T is the soil temperature (℃).

2.5. Soil collection and analyses

Soil samples were collected from five different locations within each plot after the tobacco plants leaves were harvest. These samples were then thoroughly mixed into a composite sample. Later, field-fresh soil samples were passed through a sieve with a 2-mm mesh size to exclude gravel and plant roots. The soil bulk density of each plot was assessed using the cylinder method. Soil organic matter (SOM) was determined by the potassium dichromate oxidation method. Soil total carbon (TC), total nitrogen (TN), and total sulfur (TS) concentrations were determined using an Elemental Analyzer (vario MACRO Analyzer, Elementar, Analysensysteme, GmbH, Germany) (Cheng et al., 2017). Soil total P (TP) and available P (AP) were measured by H_2SO_4-$HClO_4$ digestion and $NaHCO_3$ extraction, respectively. Total K (TK) and available K (AK) were determined using NaOH melting and CH_3COONH_4 extraction methods, respectively (Wang et al. 2016b). Soil available nitrogen (AN) was analyzed by the alkali solution diffusion method (Chen et al., 2015). The soil carbon stocks (SCS) was estimated as follows (dos Santos et al., 2016):

SCS (kg ha^{-1}) = BD × d × C × cf,where BD = bulk density (kg m^{-3}), d = soil depth (0.20 m), C = soil carbon concentration (kg kg^{-1}), and cf = the conversion factor (10,000 m^2 ha^{-1})

2.6. Plant collection and analyses

Tobacco biomass was measured using three plants by cutting excavating from each plot. The tobacco samples, including roots, stems, and leaves, were rinsed using tap water and then with deionized water, and later oven-dried at 105 °C for 10 min, and at 70 °C for 48 h to a constant weight. Subsequently, dried plant samples were weighed to obtain the biomass, which was grounded to determine the TC concentration using an elemental analyzer (vario MACRO Analyzer, Germany). The biomass C storage (BCS) of tobacco was calculated by multiplying the concentration and the biomass:

BCS (kg ha^{-1}) = $C_{root} \times B_{root} + C_{stem} \times B_{stem} + C_{leaf} \times B_{leaf}$,where C_{root}, C_{stem}, and C_{leaf} are the C concentrations (kg kg^{-1}) in the roots, stems, and leaves, respectively; B_{root}, B_{stem}, and B_{leaf} are the biomass (kg ha^{-1}) of the roots, stems, and leaves, respectively.

2.7. Estimation of C budget model

The C budget in the tobacco growing season was estimated using the following equation (Mukherjee et al., 2014):

C budget (t C ha^{-1}) = C_{input} - C_{output} = (amendment C (t C ha^{-1}) + BCS (t C ha^{-1})) - M_{CO2} (t C ha^{-1}),

where amendment C derived from BC application was applied to the estimation of C budget model.

2.8. Data analyses

All values were expressed as the means ± standard error. The normal distribution and homogeneous of variance tests were performed before subjecting to ANOVA. The differences among various BC treatments were examined with one-way ANOVA. The significance of observed differences was tested with the least significant difference (LSD) test at a level of 0.05. Regression analysis was employed to determine the significance of the observed fitted results on the soil CO_2 emissions and soil temperature and moisture at various depths (i.e., 5, 10, and 20 cm). All statistical analyses were performed using SPSS 16.0 (SPSS Inc., Chicago, IL, USA).

3. Results

3.1. Changes in soil chemical properties under different BC amendments

SOM content increased with the increasing BC application rates. Compared to the control (B_0), SOM content significantly increased by 3.5%, 84.5%, 157.7%, and 226.1% in the B_1, B_{10}, B_{25}, and B_{50} treatments, respectively ($p \leq 0.05$, Table 1). In addition to SOM, the contents of TC, TN, and AK also markedly increased with increasing amounts of BC amendment. However, BC addition resulted in a significant decrease in TS content in the control soil from 2.03 ± 0.81 g kg^{-1} relative to the B_{50} treatment to 0.35 ± 0.03 g kg^{-1} ($p \leq 0.05$). In addition, AN and AP contents initially increased and later decreased with increasing BC application, and the highest content was observed with the B_{10} treatment. Moreover, no significant differences in TK content were observed between BC-unamended and -amended soils, even at a BC application rate of 50 t ha^{-1}.

3.2. Effects of BC amendment to soil temperature, moisture, and CO_2 flux

Soil temperatures decreased along with increased soil depths among three growth periods of tobacco (Table 2). At the root extending period, the B_1, B_{10}, B_{25}, and B_{50} treatments decreased soil temperature at a depth of 5 cm by an average of 0.38 °C, 0.32 °C, 2.85 °C, and 2.71 °C, respectively, as compared to the B_0 treatment. Moreover, B_{25} and B_{50} treatments decreased soil temperature at a depth of 10 cm by an average of 0.97 °C and 1.66 °C, respectively, but B_1 and B_{10} treatments increased 0.17 °C and 0.67 °C, respectively. However, at a depth of 20 cm, there

Table 1
Soil chemical properties using various BC treatments.

	B_0	B_1	B_{10}	B_{25}	B_{50}
SOM (g kg^{-1})	36.77 ± 1.40^{d}	38.07 ± 1.22^{d}	67.85 ± 2.55^{c}	94.74 ± 5.53^{b}	119.91 ± 12.18^{a}
TC (g kg^{-1})	22.36 ± 0.28^{e}	24.05 ± 0.21^{d}	29.38 ± 0.05^{c}	31.11 ± 0.32^{b}	49.08 ± 0.38^{a}
TN (g kg^{-1})	2.10 ± 0.10^{c}	2.27 ± 0.08^{c}	2.59 ± 0.06^{b}	2.64 ± 0.06^{b}	2.97 ± 0.08^{a}
TP (g kg^{-1})	0.55 ± 0.10^{b}	0.65 ± 0.03^{b}	0.88 ± 0.02^{a}	0.87 ± 0.02^{a}	0.92 ± 0.05^{a}
TK (g kg^{-1})	13.00 ± 0.74^{a}	12.90 ± 1.10^{a}	13.97 ± 0.52^{a}	14.97 ± 0.44^{a}	14.07 ± 0.39^{a}
TS (g kg^{-1})	2.03 ± 0.81^{a}	0.86 ± 0.35^{ab}	0.58 ± 0.16^{b}	0.45 ± 0.01^{b}	0.35 ± 0.03^{b}
AN (mg kg^{-1})	145.56 ± 9.94^{c}	174.25 ± 1.91^{a}	179.12 ± 2.19^{a}	172.18 ± 3.19^{ab}	156.33 ± 3.05^{bc}
AP (mg kg^{-1})	5.27 ± 0.15^{c}	22.44 ± 1.50^{b}	29.15 ± 1.02^{a}	25.11 ± 2.28^{ab}	24.84 ± 0.87^{b}
AK (mg kg^{-1})	250.50 ± 0.90^{d}	342.80 ± 1.55^{d}	501.77 ± 3.89^{c}	1475.50 ± 33.10^{b}	1581.07 ± 58.84^{a}

B_0, B_1, B_{10}, B_{25}, and B_{50} represent soil amended with 0, 1.0, 10, 25 and 50 t BC ha^{-1}, respectively; SOM: soil organic matter content; TC: total carbon content; TN: total nitrogen content; TP: total phosphorus content; TK: total potassium content; TS: total sulfur content; AN, available nitrogen content; AP, available phosphorus content; AK, available potassium content. Different letters in columns indicate a significant difference ($p \leq 0.05$) (one-way ANOVA and LSD test)

Table 2
Effect of BC on soil temperature at different depths in different tobacco growth periods.

Growth periods	Treatment	Soil temperature (°C)		
		5 cm	10 cm	20 cm
REP	B_0	28.45 ± 0.87^{aA}	26.68 ± 0.54^{abAB}	26.27 ± 0.58^{aB}
	B_1	28.07 ± 0.81^{abcA}	26.85 ± 0.65^{abA}	26.31 ± 0.55^{aA}
	B_{10}	28.13 ± 0.91^{abA}	27.35 ± 0.74^{aA}	26.08 ± 0.48^{aA}
	B_{25}	25.60 ± 0.80^{cA}	25.71 ± 0.74^{abA}	25.89 ± 0.64^{aA}
	B_{50}	25.74 ± 1.07^{bcA}	25.02 ± 0.79^{bA}	24.76 ± 0.68^{aA}
VP	B_0	26.23 ± 0.24^{aA}	25.62 ± 0.18^{aB}	25.01 ± 0.19^{aC}
	B_1	26.67 ± 0.20^{aA}	25.69 ± 0.15^{aB}	24.99 ± 0.17^{aC}
	B_{10}	25.79 ± 0.44^{aA}	25.09 ± 0.32^{aA}	24.74 ± 0.41^{aA}
	B_{25}	26.14 ± 0.52^{aA}	25.21 ± 0.41^{aAB}	24.86 ± 0.41^{aB}
	B_{50}	26.61 ± 0.48^{aA}	25.89 ± 0.59^{aA}	25.59 ± 0.54^{aA}
MP	B_0	22.50 ± 0.70^{aA}	21.77 ± 0.64^{aA}	21.42 ± 0.58^{aA}
	B_1	22.76 ± 0.72^{aA}	21.90 ± 0.64^{aA}	21.52 ± 0.57^{aA}
	B_{10}	22.79 ± 0.73^{aA}	21.89 ± 0.63^{aA}	21.51 ± 0.58^{aA}
	B_{25}	20.52 ± 0.60^{bA}	20.24 ± 0.57^{abA}	20.36 ± 0.55^{aA}
	B_{50}	19.78 ± 0.55^{bA}	19.74 ± 0.55^{bA}	20.25 ± 0.53^{aA}

Different lowercase letters within a single column indicate significant differences at $p \leq 0.05$ between treatments. Different capital letters within a single line indicate significant differences at $p \leq 0.05$ between soil depths. REP: root extending period (30 days), VP: vigorous period (30 days), MP: mature period (60 days).

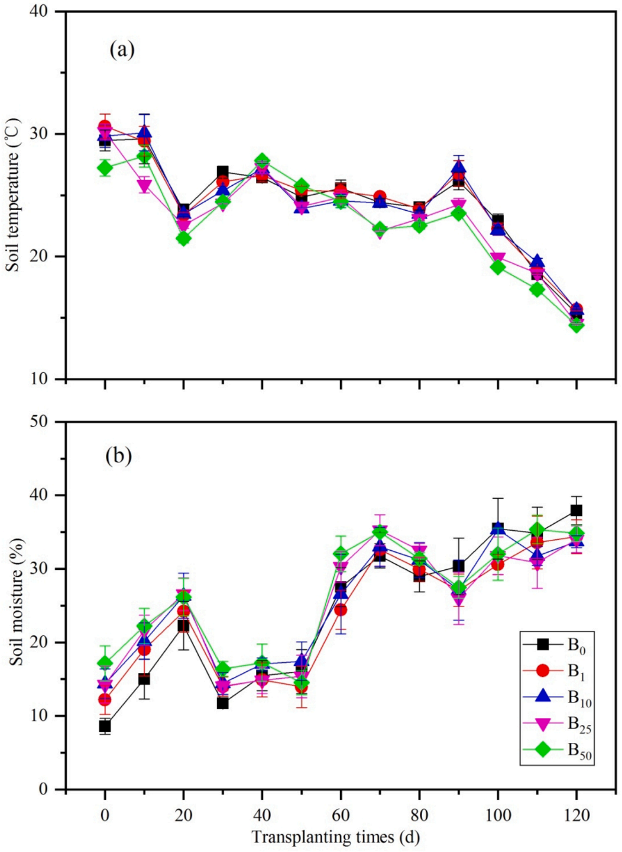

Fig. 1. Variations in soil temperature (a) and moisture (b) (0–20 cm) using various BC treatments during the tobacco-growing season.

were no significant differences between all treatments. At the vigorous period, the effect of BC amendment on temperature was much smaller at all soil depths compared to B_0, ranging from − 0.53 °C to 0.58 °C. At the mature period, the B_{50} treatment significantly decreased soil temperature at a depth of 5 cm and 10 cm by an average of 2.72 °C and 2.03 °C, respectively, as compared to the B_0 treatment.

Among the five BC treatments, the variation of soil temperature with transplanting time was a relatively consistent trend (Fig. 1a). The mean temperatures during the whole tobacco-growing season were 24.46 ± 1.09 °C for B_0, 24.56 ± 1.11 °C for B_1, 24.37 ± 1.09 °C for B_{10}, 23.20 ± 1.09 °C for B_{25}, and 22.97 ± 1.15 °C for B_{50}. Moreover, the soil moisture trend was similar, and significantly negatively correlated ($R = 0.65$–0.76) with soil temperature at different soil depths (Fig. 1b; Table 3). The mean soil moisture was $24.30 \pm 2.76\%$ for B_0, $23.92 \pm 2.29\%$ for B_1, $25.26 \pm 2.13\%$ for B_{10}, $25.19 \pm 2.26\%$ for B_{25}, and $26.29 \pm 2.18\%$ for B_{50}.

Soil CO_2 emissions with different BC treatments significantly varied with transplantation time, with the mean daily CO_2 emission rate ranging from 40.12 mg C $m^{-2}h^{-1}$ at 120 days to 137.63 mg C $m^{-2}h^{-1}$ at 30 days post seedling transplantation (Fig. 2a). Compared with the controls, significantly higher CO_2 emissions were measured in the BC-amended soils most of the time, and a few times, emissions from BC-amended soils were lower than the controls. Average soil CO_2 emission increased with increasing BC application levels. Compared to the control (B_0), the daily average rate of soil CO_2 fluxes significantly increased by 20.0%, 26.3%, 39.4%, and 50.2% in the B_1, B_{10}, B_{25}, and B_{50} treatments, respectively ($p \leq 0.05$, Fig. 2a). In addition, the cumulative CO_2 flux for the B_0 (2.36 ± 0.05 t C ha^{-1}) was significantly lower than those for the B_1 (2.86 ± 0.01 t C ha^{-1}), B_{10} (3.00 ± 0.07 t C ha^{-1}), B_{25} (3.30 ± 0.07 t C ha^{-1}), and B_{50} (3.52 ± 0.04 t C ha^{-1}) treatments. However, no significant differences in cumulative CO_2 fluxes were observed between the B_1 and B_{10} treatments (Fig. 2b).

3.3. *Correlation of soil CO_2 flux with soil moisture and temperature after BC amendment*

Soil CO_2 emissions were correlated with soil temperatures and moistures. In this study, a significant negative correlation was observed between rate of CO_2 emission and soil water levels (Table 3). A linear function could explain 26.2%-30.9% of the observed variations in CO_2

Table 3

Pearson correlation coefficients between CO_2 emissions and soil temperature and moisture.

	CO_2	T_{5cm}	T_{10cm}	T_{20cm}	SWC_{5cm}	SWC_{10cm}	SWC_{20cm}
CO_2	1.000	0.578**	0.631**	0.661**	−0.547**	−0.556**	−0.512**
T_{5cm}		1.000	0.980**	0.948**	−0.721**	−0.670**	−0.653**
T_{10cm}			1.000	0.984**	−0.745**	−0.710**	−0.700**
T_{20cm}				1.000	−0.757**	−0.735**	−0.737**
SWC_{5cm}					1.000	0.928**	0.910**
SWC_{10cm}						1.000	0.948**
SWC_{20cm}							1.000

Note: **Correlation is statistically significant at the 0.01 level (2-tailed). T_{5cm}, T_{10cm}, and T_{20cm} mean soil temperature at 5-, 10-, and 20-cm soil depths, respectively. SWC_{5cm}, SWC_{10cm}, and SWC_{20cm} indicate soil water content at soil depths of 5, 10, and 20 cm, respectively.

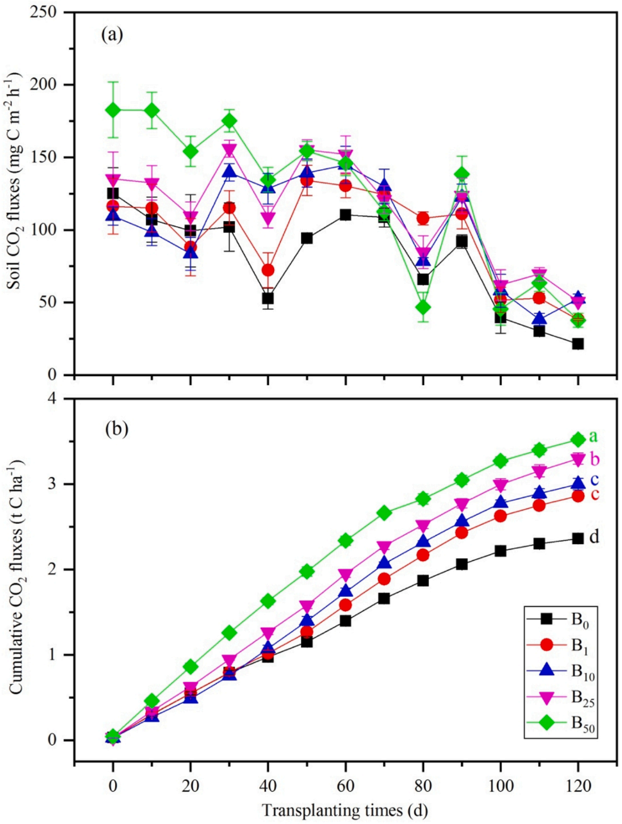

Fig. 2. Variations of soil CO_2 fluxes (a) and cumulative CO_2 fluxes (b) in various BC treatments during the tobacco-growing period. Various lower case letters indicate significant differences (LSD, $p \leq 0.05$), whereas the same lower case letters show non-significant differences among BC treatments.

fluxes, and a higher correlation coefficient was observed with the water contents at the 5- and 10-cm depths compared to the 20-cm depth. Soil CO_2 flux linearly and exponentially increased with soil temperature at soil depths of 5, 10, and 20 cm (Tables 3 and 4). At the 20-cm soil depth, the exponential model explained a large part of the variation (56.5%-71.5%) in CO_2 emissions for all BC treatments ($p \leq 0.05$; Table 4). Soil respiration sensitivity to temperature (Q_{10}) in B_1 was similar to that of B_{10} and B_{25}, but significantly lower compared to B_0 and B_{50}. Generally, the Q_{10} values were 38.6% higher at a soil depth of 20 cm than at a 5 cm depth for B_0, 29.0% for B_1, 40.1% for B_{10}, 11.0% for B_{25}, and 30.7% for B_{50}. When considering both soil temperature and soil moisture, exponential-exponential function could explain 49.0%-65.9%, 39.1%-64.8%, and 43.6%-62.6% of the variation of soil respiration in the five BC treatments at 5 cm, 10 cm and 20 cm depths, respectively. The R^2 values for B_0 and B_{50} treatments were higher than B_1, B_{10}, and B_{25} treatments, which indicated B_0 and B_{50} had better correlation with soil respiration than B_1, B_{10}, and B_{25} (Table 5).

3.4. *Effect of BC amendment on soil C budget*

Compared with the control (B_0), B_{50} significantly increased root, stem, and leaf biomass by 204.7%, 123.6%, and 64.0%, respectively. No significant differences in total tobacco biomass were detected between the B_0 and B_1 treatments. However, we observed significant differences in total biomass when we compared the B_{10}, B_{25}, and B_{50} treatments with the B_0 treatment (Fig. 3a). For all BC treatments, biomass C sequestration decreased in the order: leaf > root ≥ stem. The contribution of leaf to biomass C sequestration in the B_0 treatment was relatively higher than that in the other BC treatments (Fig. 3b).

Soil C sequestration increased by increasing BC application rates, which was 1.89-fold higher in the B_{50} treatment compared to the control (B_0) (Table 6). Compared with the B_0 treatment, the B_1, B_{10}, B_{25}, and B_{50} treatments increased biomass C sequestration by 44.6% (p = 0.064), 60.0% (p = 0.019), 91.4% (p = 0.002), and 96.4% (p = 0.001), respectively. Although BC amendment increases soil cumulative CO_2 emissions (21.1%–49.1%), the positive values of C budget were observed from the treatments amended with higher BC dosages, which benefits from the high relative proportion of C storage in BC to soil. The C budgets ranged from −1.17 t C ha^{-1} in the B_0 treatment to 19.48 t C ha^{-1} in the B_{50} treatment during the tobacco-growing season. C losses in the B_1 treatment were significantly lower than that observed in the B_0 treatment by 38.5%. Compared to the B_{10} treatment, C gains were 3.16 and 6.41 fold higher in the B_{25} and B_{50} treatments, respectively.

4. Discussion

4.1. *Effect of BC amendment on soil CO_2 emission in the karst region*

The increase in soil cumulative CO_2 emission ranged from 21.1% to 49.1% throughout the tobacco-growing season under different BC

Table 4

Parameters of the exponential model of soil CO_2 emission with soil temperature at 5- and 20-cm depths under different BC amendments.

	B_0 5 cm	B_0 20 cm	B_1 5 cm	B_1 20 cm	B_{10} 5 cm	B_{10} 20 cm	B_{25} 5 cm	B_{25} 20 cm	B_{50} 5 cm	B_{50} 20 cm
R^2	0.665	0.715	0.566	0.669	0.382	0.565	0.599	0.634	0.652	0.655
α	5.617	3.011	15.775	9.547	21.255	10.444	21.521	17.247	10.018	5.632
β	0.100	0.133	0.069	0.094	0.059	0.093	0.068	0.079	0.102	0.128
Q_{10}	2.729	3.781	1.988	2.565	1.811	2.537	1.978	2.195	2.762	3.611

Table 5
The exponential-exponential function of soil respiration with soil moisture and temperature under BC amendments at different soil depths.

Treatments	Soil depths		
	5 cm	10 cm	20 cm
B_0	$F = 22.67e^{-0.006W}e^{0.054T}$ $(R^2 = 0.53)$	$F = 3.64e^{0.006W}e^{0.119T}$ $(R^2 = 0.64)$	$F = 2.64e^{0.009W}e^{0.130T}$ $(R^2 = 0.59)$
B_1	$F = 26.00e^{0.002W}e^{0.050T}$ $(R^2 = 0.43)$	$F = 19.38e^{0.003W}e^{0.062T}$ $(R^2 = 0.50)$	$F = 5.15e^{0.015W}e^{0.104T}$ $(R^2 = 0.58)$
B_{10}	$F = 206.47e^{-0.031W}e^{-0.002T}$ $(R^2 = 0.53)$	$F = 98.00e^{-0.020W}e^{0.021T}$ $(R^2 = 0.39)$	$F = 15.29e^{0.003W}e^{0.075T}$ $(R^2 = 0.44)$
B_{25}	$F = 68.86e^{-0.011W}e^{0.030T}$ $(R^2 = 0.49)$	$F = 43.25e^{-0.005W}e^{0.046T}$ $(R^2 = 0.49)$	$F = 51.06e^{-0.008W}e^{0.044T}$ $(R^2 = 0.51)$
B_{50}	$F = 39.55e^{-0.013W}e^{0.059T}$ $(R^2 = 0.66)$	$F = 36.27e^{-0.013W}e^{0.065T}$ $(R^2 = 0.65)$	$F = 19.40e^{-0.009W}e^{0.090T}$ $(R^2 = 0.63)$

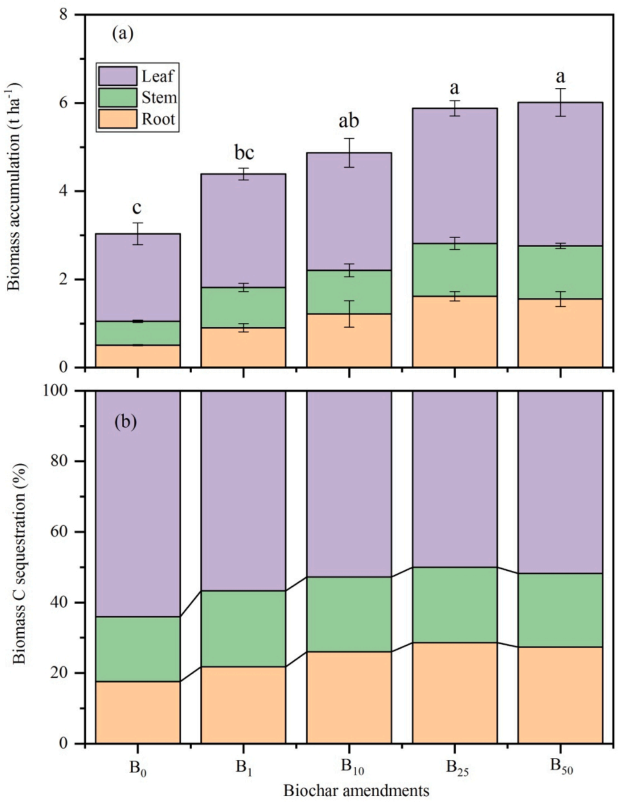

Fig. 3. Effect of BC application on leaf, stem, and root biomass (a) and carbon sequestration distribution (b). Various lower case letters indicate significant differences (LSD, $p \le 0.05$), whereas the same lower case letters show non-significant differences among BC treatments.

application rates compared with the control (Fig. 2), which is concordant to the results of He et al. (2017), who showed that BC amendment significantly increases soil CO_2 flux on average by 22.1% (*meta*-analysis). Similar results were reported by other studies (Novak et al., 2010; Sui et al., 2016), which showed higher CO_2 emissions in BC amendments. In contrast, some studies have demonstrated that BC amendments have no effect or reduce soil CO_2 emission (Karhu et al., 2011; Ameloot et al., 2013; Zhang et al., 2017). The discrepancy in this effect could be attributed to differences in BC characteristics, soil properties, and the experimental duration (Wang et al., 2016a).

In the present study, the reason for increasing CO_2 emissions by BC amendment is assumed to decompose and release the organic or inorganic C contained in BC (Zimmerman et al., 2011; Singla and Inubushi, 2014), especially in a short-term experiment (Lorenz and Lal, 2014). It has been demonstrated that the mean residence time (MRT) of labile BC carbon pool was estimated to be about 108 days and the average BC decomposition rate for studies shorter than 0.5 year was four times higher than that in those studies longer than 1 year (Wang et al., 2016a). Our study supports this conclusion as the experimental duration was about 120 days. Therefore, short-term application of BC can stimulate C mineralization and CO_2 production. Furthermore, the tobacco-growing soil in the present study was well-aerated for a long time due to the high ridging tillage (30 cm) for flue-cured tobacco cropping, which decreasing the formation of organic-mineral aggregates and interactions between BC and soils (Polifka et al., 2018). This missing physical protection could also increase the degradation of BC in a co-metabolic way, which results in higher CO_2 evolution (Polifka et al., 2018). Lastly, another possible explanation for the incresing of CO_2 emissions is that tobacco BC contains abundant nutrient substances (N, P, and K) (Table 1) and increases soil microbial activeities and SOM mineralization (Zavalloni et al., 2011).

4.2. Effect of environmental factors on soil CO_2 emission under BC amendments

Soil moisture and temperature are two major ecological factors that impact soil CO_2 emissions (Espeleta et al., 1999; Davidson et al., 2006). Here, a strongly negative correlation was observed between soil CO_2 emission and soil moisture irrespective of BC treatments (Table 2), which is concordant with the results of other studies (Lu et al., 2014; He et al., 2016). This finding may be attributable to the relatively high soil moisture during the majority of the sampling times (Fig. 1b). When soil water content is higher than a certain threshold (field capacity), high precipitation increases diffusional resistance and reduces soil CO_2 emissions (Espeleta et al., 1999). In addition, soil temperature is another common factor that influences seasonal variations in CO_2 efflux (Fang

Table 6
Estimation of C budget in different BC treatments during the tobacco-growing period (t C ha^{-1}).

	Soil C storage	Biomass C storage	Cumulative CO_2-C emission	BC-C amendment	C budget
B_0	46.59 ± 1.26^c	1.19 ± 0.08^c	2.36 ± 0.05^c	0.00 ± 0.00^e	-1.17 ± 0.13^e
B_1	49.54 ± 1.85^c	1.72 ± 0.10^{bc}	2.86 ± 0.01^c	0.41 ± 0.00^d	-0.72 ± 0.10^d
B_{10}	58.17 ± 1.59^b	1.91 ± 0.29^{ab}	3.00 ± 0.07^c	4.13 ± 0.04^c	3.04 ± 0.25^c
B_{25}	58.32 ± 1.76^b	2.28 ± 0.15^{ab}	3.30 ± 0.07^b	10.33 ± 0.09^b	9.31 ± 0.20^b
B_{50}	88.19 ± 1.84^a	2.34 ± 0.20^a	3.52 ± 0.04^a	20.66 ± 0.19^a	19.48 ± 0.23^a

Negative values of C budget indicate net CO_2 emission from soils.

and Moncrieff, 2001; Davidson et al., 2006). An exponential correlation between soil CO_2 flux and soil temperature has been reported under different BC applications (Lu et al., 2014; He et al., 2016; Shen et al., 2017). This relationship has also been observed in our study irrespective of BC amendment level (Table 4), and soil temperature accounts for 38.2%–71.5% of the observed variations in soil CO_2 emission.

Different management practices (cultivation, irrigation, or fertilization) can markedly alter the Q_{10} value (Sheng et al., 2010). In this study, Q_{10} values were between 1.81 and 2.76 at soil 5-cm depth during the tobacco-growing season, which was within the 1.3–3.3 range that has been reported for various biomes around the world (Raich and Schlesinger, 1992). Moreover, the Q_{10} values are influenced by the temperature measurement depths (Graf et al., 2008). The Q_{10} values were 11.0%-40.1% higher at a 20-cm soil depth than at a 5-cm soil depth for all BC treatments. Regardless of the depth, Q_{10} was significantly lower under B_1 than under B_0 and B_{50}, but was similar to B_{10} and B_{25} (Table 4), which is concordant with other studies (He et al., 2016). However, Zhou et al. (2017) found a positive relationship between Q_{10} and BC amendments in subtropical plantations. Because soil respiration is influenced by substrate availability, variations in temperature range and reference temperatures were used in the Q_{10} calculation (Gershenson et al., 2009; Nottingham et al., 2019). Here, the temperature sensitivity of soil respiration was reduced after BC amendment, particularly at low application rates. BC addition decreased soil temperature fluctuations and moderate soil temperature extremes (Fig. 1), which is attributable to the negative effect of BC application on soil thermal properties (capacity, conductivity, and diffusivity) by enhancing the total porosity, particularly in the meso- and macroporosity of BC-amended soils (Liu et al., 2018). Similar moderating soil high and low temperature with BC application was also previously reported (Zhang et al., 2013; Blanco-Canqui, 2017). The temperature regulation capability of BC can be strategic for reducing newly available substrate as well as native SOC degradation or root residues (Sun et al., 2014; He et al., 2016; Chen et al., 2019).

4.3. *Effect of BC on C budget in the agro-ecosystem*

Similar to previous studies (Mao et al., 2012; Ouyang et al., 2014), BC addition can directly increase agricultural soil C sequestration from 6.3% to 89.3% (Table 6), possibly due to high C contents and aromatic ring structures in the BC (Mao et al., 2012). Indirectly, BC change soil C inputs by influencing plant biomass during photosynthesis or bio-sequestration. Hence, increased biomass, C transport from plant to root symbionts, and root-derived C inputs after BC application may induce an increase in soil C storage (Ciais et al., 2010; Sohi et al., 2010). Compared with the control, tobacco biomass increased from 44.9% to 98.4% in BC treatments, which is in accordance with the mean increase ($\leq$10% to $\geq$200%) using a *meta*-analysis approach (Jeffery et al., 2011; Wang et al., 2012). Moreover, tobacco biomass increased with more BC application levels to the roots, stems, and leaves (Fig. 3). Similar data were found that BC derived from tobacco stalks increased the tobacco biomass, especially the yield of tobacco leaves (Zhang et al., 2019b). These positive results could be attributed to the high contents of TN, TP, and TK in the BC itself being easily dissolved in the soil solution, thus increasing the tobacco growth by enhancing soil fertility, particularly the SOM, AN, AP, and AK contents (Table 1).

C budget, which is dependent on the balance between C inputs (e.g., as crop residues and BC amendments) and outputs (e.g., as CO_2 from respiration), can be reflected in changes in soil C sequestration during the whole of cropping growing season. Except for the B_0 and B_1 treatments, the C inputs were higher than its outputs following BC amendment, indicating a strong characteristic of the C sink in the flue-cured tobacco ecosystem (Table 6). Although soil CO_2 emissions increased by increasing BC application rates during the growing season, the treatments with BC amended at higher levels showed larger C budget, as indicated by the higher net primary production (NPP) and soil C stock. BC contains a high amount of recalcitrant C, which could function in C sequestration in the soil for a relatively long time (Kuzyakov et al., 2009). Hence, agricultural fields with the integration of BC could store C more effectively compared with the integration of other amendments (e.g., plants, manure, green manure, and compost manure) (Kwapinski et al., 2010), indicating that BC is a useful approach for C sequestration of agricultural soil.

5. Conclusions

In the karst region, tobacco BC amendments induced a significant increase in tobacco yield, soil TC, TN, AP, AK, and SOM contents but reduced TS content. Moreover, soil CO_2 fluxes are significantly and negatively correlated with soil moisture but positively correlated with soil temperature. A significant reduction in the sensitivity of soil respiration to temperature was observed with BC amendment. Although BC-treated soils significantly increased the cumulative CO_2 emissions (21.1%–49.1%) in the short term, the BC amendments showed larger soil C budget because of enhanced both crop biomass and soil C storages during the tobacco–growing season. Consequently, returning BC to karstic agricultural soils enhances soil quality and may be potentially utilized for soil C sequestration. However, additional studies assessing whether these findings could be developed in the long–term BC treatment field experiments are warranted.

Declaration of Competing Interest

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

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