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HIGHLIGHTS

GRAPHICAL ABSTRACT

- The influence of straw/biochar on CO₂ emission and microbial community was tested.
- Straw significantly increased respiration and PLFAs than biochar and the control.
- ¹³C-PLFA profile in straw amendment was significantly different from biochar.
 Soil microorganisms utilized more straw
- Soil microorganisms utilized more straw-C than biochar-C
- The substrate composition/availability influenced the microbes and their function



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ABSTRACT

The application of straw and biochar to soil has received great attention because of their potential benefits such as fertility improvement and carbon (C) sequestration. The abiotic effects of these materials on C and nitrogen (N) cycling in the soil ecosystem have been previously investigated, however, the effects of straw or its derived biochar on the soil microbial community structure and function are not well understood. For this purpose, a short-term incubation experiment was conducted using ¹³C-labeled rice straw and its derived biochar (¹³Clabeled biochar) to deepen our understanding about soil microbial community dynamics and function in C sequestration and greenhouse gas emission in the acidic paddy soil amended with these materials. Regarding microbial function, biochar and straw applications increased CO₂ emission in the initial stage of incubation and reached the highest level (0.52 and 3.96 mg C kg⁻¹ soil h^{-1}) at 1 d and 3 d after incubation, respectively. Straw amendment significantly (p < 0.01) increased respiration rate, total phospholipid fatty acids (PLFAs) and ¹³C-PLFA as compared to biochar amendment and the control. The amount and percent of Gram positive bacteria, fungi and actinomycetes were also significantly (p < 0.05) higher in ¹³C-labeled straw amended soil than the ¹³Clabeled biochar amended soil. According to the ¹³C data, 23 different PLFAs were derived from straw amended paddy soil, while only 17 PLFAs were derived from biochar amendments. The profile of ¹³C-PLFAs derived from straw amendment was significantly (p < 0.01) different from biochar amendment. The PLFAs 18:1 ω 7c and cy17:0 (indicators of Gram negative bacteria) showed high relative abundances in the biochar amendment,

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while 10Me18:0, i17:0 and 18:2 ω 6,9c (indicators of actinomycetes, Gram positive bacteria and fungi, respectively) showed high relative abundance in the straw amendments. Our results suggest that the function, size and structure of the microbial community were strongly influenced by the substrate composition and availability. © 2016 Elsevier B.V. All rights reserved.

1. Introduction

Plant residuals have been applied to agriculture soils for centuries as a source of nutrients and to maintain soil organic matter contents, structure and moisture (Kogel-Knabner, 2002; Powlson et al., 2011). However, the incorporation of plant residuals can substantially increase the production of greenhouse gases from soil, which have an important influence on global climate change. The pyrolysis of residual biomass produces a charcoal-like substance, called biochar, that has the potential to be used as soil amendment because of its associated benefits (Biederman and Harpole, 2013; Khan et al., 2014). Recent studies have shown extensive application of biochar in land management practices with the intention to increase soil carbon (C) storage, mitigate greenhouse gas emissions, and minimize the availability and bioaccumulation of organic and inorganic contaminants (Case et al., 2012; Cayuela et al., 2014; Khan et al., 2013, 2015; Spokas et al., 2009; Stewart et al., 2013; Wagas et al., 2014). The causes and mechanisms of this reduction are still uncertain (Borchard et al., 2014; Spokas et al., 2012).

Microbial activity, the pivotal regulator of soil organic matter dynamics and nutrient cycling, has a great impact on organic matter degradation, though metabolic capacity varies with different microbial species. The composition of residues affects the decomposition rate, microbial respiration and microbial biomass (Xu et al., 2006). Microbial succession during the decomposition of rice straw is driven by the availability of limiting substrates and the ability of the microbial community to utilize the soil resources (Rui et al., 2009). After substrate addition, the rice straw derived C is rapidly incorporated into the soil microbial biomass (Murase et al., 2006). Ye et al. (2015) reported that bacterial PLFAs were increased by > 30% with the application of straw under anaerobic conditions but fungi did not show the same level of increase. Under anaerobic conditions, an increase in the size of the bacterial community depends on utilization of C derived from fungal decomposition of straw material (Meidute et al., 2008).

Recent studies have shown that biochar undergoes complicated biotic degradation processes depending on the type of biochar, soil characteristics, incubation time and climatic conditions (Farrell et al., 2013; Santos et al., 2012; Watzinger et al., 2014). In particular, the biochar pyrolytic activity and soil type significantly control the shifts in relative abundance and diversity of key soil microbial functional taxa, i.e. Gram-negative bacteria and actinomycetes that are able to degrade recalcitrant C compounds (Anderson et al., 2011; Prayogo et al., 2013; Khodada et al., 2011).

Rice is one of the most important agronomic plants in the world, and >135 million ha land is used for rice cultivation (Liesack et al., 2000). Increases in paddy soil organic C sequestration can favor of agricultural sustainable development and reduce global warming potential (Fornara et al., 2011; Lal, 2004). Soil microbial community is also a significant indicator for soil quality. Both straw and biochar could be used as soil amendment materials for C sequestration; however, there are huge differences in the physical and chemical characteristics of rice straw and its derived biochar and, undoubtedly, these will induce different C transformation routes, microbial communities and environmental functions. However, few studies have focused on the relative effects of rice straw and its biochar on the paddy soil microbial community, and the mechanisms involved in the microbial utilization of these two kinds of C sources are not clear. We tackle this critical gap and applied ¹³C-labeled rice straw and its derived biochar (¹³C-labeled biochar) into an acidic paddy soil and used the ¹³C-isotopic analysis of released CO₂ and phospholipid fatty acids (PLFAs) of the soil microorganisms to investigate the fates of rice straw and its biochar, and their effects on the soil microbial community structure and function.

2. Materials and methods

2.1. Soil ¹³C-labeled rice straw and ¹³C-labeled biochar

Paddy soil was collected from the plow layer (0-15 cm depth) from an arable field in Changde (N: 28°57′00″; E: 111°30′30.0″), located in the subtropical region of southern China, in September 2014. After the removal of visible roots and stones, the soil was sieved through a 2 mm mesh and thoroughly mixed. The soil texture was a clay loam with total C and nitrogen (N) contents of 16.6 g kg⁻¹ and 2.3 g kg⁻¹, respectively. Soil pH was 5.2 determined in a soil and water suspension (1:5 *w/v*, soil/water), NH₄⁺ and NO₃⁻ were extracted with 2 M KCI (1:5 *w/v*, soil/KCl) and were recorded as 7.5 mg kg⁻¹ and 4.5 mg kg⁻¹, respectively. The natural abundance δ^{13} C value of the soil was – 27.7‰.

To prepare ¹³C-labeled rice straw, rice plants at tillering stage were continuously labeled with ¹³CO₂ atmosphere. After one month, rice shoots were harvested and half of the straw was cut into 2 mm pieces before use as soil amendment. The basic characteristics of rice straw such as total C (365.2 g kg⁻¹), total N (13.6 g kg⁻¹), water extractable C (31.9 g kg⁻¹), water extractable N (2.7 g kg⁻¹) and the excess δ^{13} C (14,454.6‰) were determined using standard procedures (Nelson and Sommers, 1982).

The remaining half of the ¹³C-labeled rice straw was used to prepared biochar through the pyrolysis technique under no-oxygen conditions at 550 °C for 2 h. The furnace temperature was increased by a stepwise procedure at about 12.5 °C min⁻¹. After cooling, the biochar was sieved through <2 mm mesh to make it ready for soil addition. Biochar was analyzed for pH (11.0) with water (1:10 *w/v*, biochar/water), total C (429.4 g kg⁻¹), total N (13.0 g kg⁻¹), water extractable C (12.4 g kg⁻¹), water extractable N (0.83 g kg⁻¹) and the excess δ^{13} C (11,081.8‰).

Total C and N were analyzed using a CNS analyzer (Vario MAX C/N; Elementar, Germany), while soil water extractable C and N were determined using a total carbon analyzer (TOC-V CPH, SHIMADZU, Japan). The δ^{13} C values of soil, rice straw and biochar were determined using gas chromatography/combustion/isotope ratio mass spectrometry (GC-C-IRMS) (GA-C/Delta^{Plus} XP. Thermo Scientific, Germany).

2.2. Experimental design

The prepared soil was amended with ¹³C-labeled rice straw at an application rate of 3.652 g C kg⁻¹ and then thoroughly mixed. For biochar amendments, the ¹³C-labeled biochar was added in to paddy soil at the same application rate (3.652 g C kg⁻¹). The control treatment (soil without straw and biochar) was also prepared. After this, the equivalent of 10 g fresh, field-moist paddy soil (on a dry weight (d.w.) basis) was added in to brown serum bottles (100 mL) and a total 18 replicates were prepared for each treatment. All bottles were covered with needle-punctured aluminum foil to keep a stable aerobic condition. These experiments were carried out under laboratory conditions at 25 °C and at 95% relative humidity. During the incubation, deionized water was added by weighing the bottles to maintain the soil water content to 60% water holding capacity (WHC).

Three replicates (out of 18 for each treatment) were chosen for measuring the CO_2 emissions at five different time intervals: i.e. 0 (at 4 h

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