



Maize biochar addition rate influences soil enzyme activity and microbial community composition in a fluvo-aquic soil



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ABSTRACT

Biochar addition to soil has been proposed as a strategy to enhance soil quality and crop productivity, which may also affect microbial activity. However, the response of soil enzymes and microbial community composition to biochar addition and the main factors that drive their consequent behavior have rarely been studied. Therefore, to investigate the combined effect of different amounts of biochar (0, 0.5, 1.0, 2.5 and 5.0% by mass) and urea application on soil nutrients, enzymatic activities and microbial community in a fluvo-aquic soil, we conducted a 90-day laboratory study. Increased maize biochar addition led to significantly increased soil organic carbon (SOC), total N, and exchangeable K and reduced soil exchangeable Ca. Soil total N and exchangeable Ca were dominant factors affecting soil enzyme activities. Activities of soil extracellular enzymes involved in C and S cycling (except β -xylosidase) suggested lower amounts of biochar addition (0.5% by mass) could increase soil enzyme activities, while higher amounts of biochar addition reduce soil enzyme activities. However, the activities of L-leucine aminopeptidase and urease, both of which are involved in N cycling, increased with the increase of biochar addition rate. Total phospholipid fatty acid content and the relative abundance of bacteria were significantly reduced with increasing biochar addition rate. The relative abundance of fungi in the urea-amended soil was significantly higher than that in the other treated soils, and abundance of actinomycetes did not show a clear response to biochar addition. The changes in the microbial community composition were mainly related to SOC and total N contents, with a significant negative correlation. We concluded that the effect of biochar addition on soil enzymes and microbial community composition was highly variable. There is an urgent need to further estimate both the positive and negative long-term effects of biochar on the soil quality and crop productivity in this region.

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1. Introduction

Biochar is a by-product of biomass pyrolysis under oxygen-limited conditions and at relatively low temperatures (<700 °C). Biochar contains large amounts of carbon and macro or micro-nutrients depending on the feedstock and pyrolysis temperature (Enders et al., 2012; Ronsse et al., 2013; Wiedner et al., 2013). Recently, there has been a growing interest in applying biochar to amend acidic or nutrient-poor soil for soil ecological restoration while also sequestering carbon (Lehmann et al., 2003; Xu et al., 2013). Several studies have also reported biochar as a soil conditioner for enhancing soil fertility and crop productivity (Lehmann et al., 2006; Major et al., 2010). The enhancement of soil

fertility as a result of biochar addition has been attributed to increased cation exchange capacity (Liang et al., 2006), changes to soil pH, or direct nutrient contributions from the biochar (Enders et al., 2012; Quilliam et al., 2012). However, other studies have shown possible negative effects of biochar on soil quality and fertility parameters, such as short-term reductions in soil mineral N availability (Bruun et al., 2012; Tammeorg et al., 2014) and decreased performance of crops on calcareous soils (Van Zwieten et al., 2010). These results suggest that soil nutrient responses to biochar addition are dependent on soil type, biochar addition rate and other unknown factors (Liang et al., 2014). However, the exact mechanisms for these increases or decreases are still a matter of speculation (Sohi et al., 2010), but are certainly related to changes in soil physicochemical properties and biological functions (Biederman and Harpole, 2013).

Soil extracellular enzymes are the catalysts of organic matter decomposition and are involved in the biogeochemical cycling of

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nutrients (Burns et al., 2013). Understanding the effect of biochar on the activity of these key enzymes has been identified as a research priority. Recently, some studies have reported that biochar addition to soil usually increases the soil enzyme activities related to N and P cycling and reduces the soil enzyme activities involved in C cycling (Bailey et al., 2011). Conversely, other studies have found inconsistent results (Lammirato et al., 2011; Paz-Ferreiro et al., 2014), which suggest that biochar has variable effects on different soils, enzymes, and assay types. In addition, soil enzymes are catalysts that play an important role in modulating ecosystem responses to changes in abiotic (changes in soil nutrient status, or in the quality of soil organic matter) and biotic conditions (Stone et al., 2012; Trasar-Cepeda et al., 2007). However, to date, few studies have explored the response of soil extracellular enzymes to soil environment under combined urea and biochar addition with different levels of biochar application.

Biochar effects on the soil biological processes involved in C and N dynamics are not well understood (Lehmann et al., 2011) and the responses are highly variable (Jones et al., 2011). Biochar amendments to soils have been recently shown to affect the community structure and abundance of soil microorganisms (Meynet et al., 2012). Some studies have reported enhanced (Bamminger et al., 2014; Rutigliano et al., 2014) or inhibited microbial activity (Dempster et al., 2012) in response to biochar additions, whereas other studies have reported no effects on soil microbial biomass as a result of its recalcitrance (Kuzyakov et al., 2009; Zavalloni et al., 2011). In addition, biochar addition to soil can alter soil physicochemical properties, which influence the soil microbial biomass and activity as well as community composition (Lehmann et al., 2011). As a result of their sensitivity to environmental changes, soil microbial community abundance and structure have been widely used as indicators of soil quality changes (Chu et al., 2007; Marschner et al., 2003). Until now, only limited studies on the effects of biochar on individual microbial communities have been conducted, which suggest that community compositional responses to biochar addition vary according to biochar type, among other possible factors (Steinbeiss et al., 2009). However, few studies have explored the individual microbial compositional responses to soil environment changes under combined urea and biochar addition with different levels of biochar application.

Nitrogen fertilizer (N), especially urea, is one of the most important nutritious factors for crop productivity and grain quality. In the North China Plain (NCP), high rates of N fertilizer are often applied and have led to low fertilizer use efficiency and serious environmental problems (Ju et al., 2009). It has been reported that the combination of biochar and N fertilizer is effective for improving crop yield while reducing the N application rate (Steiner et al., 2007). In addition, the fluvo-aquic soil is a typical soil type in NCP, which accounts for 53% of the fluvo-aquic soils in China. However, there are few studies on microbial mechanisms between biochar and chemical N in a fluvo-aquic soil. Therefore, the specific objectives for this work were to: (1) investigate the short-term effects of combined urea and biochar

addition with different levels of biochar application (0, 0.5, 1.0, 2.5 and 5.0% by mass) on soil nutrients (pH, electrical conductivity, SOC, TN, NO_3^- -N, NH_4^+ -N, water soluble and exchangeable K, Na, Ca, Mg) and extracellular enzyme activities and microbial community in a fluvo-aquic soil; (2) illustrate the main factors that drive the changes in soil enzyme activity or microbial community composition after biochar addition. Soil enzyme activities involved in C, N, P, and S cycling and microbial community composition were determined by microbial fluorometric assay and phospholipid fatty acid (PLFA) analysis, respectively.

2. Materials and methods

2.1. Biochar and soil

Biochar was produced at 450 °C by slow pyrolysis (5 °C min⁻¹ heating and 1 h residence time in a Microwave Muffle Furnace (SX₂, Shanghai Rongfeng Scientific Instrument Inc, China)) of maize straw. Maize straw was taken from the main maize producing area in China; Zhengzhou, Henan Province, in the North China Plain. All biochar samples were mixed evenly, ground and sieved to <0.154 mm. Their physical and chemical properties are shown in Table 1.

Soil was collected in summer 2014, before sowing, from the top layer (0–20 cm) of a fluvo-aquic soil, in the Soil Fertility and Fertilizer Efficiency Monitoring Network Station, Zhengzhou, Henan Province, China (34°47'02" N, 113°39'25" E), with the soil parent material mainly originating from the alluvial deposits of the Yellow River. The soil texture is light loamy soil. The basic physical and chemical soil characteristics are shown in Table 1.

2.2. Incubation experiment

An incubation experiment was conducted over 90 days to investigate the effects of biochar on soil nutrients, enzyme activity and microbial community composition. The six treatments were control (CK), urea (U) and urea with maize biochar (MC) added separately at 0.5, 1.0, 2.5 and 5.0% by weight to soil (henceforth termed U+0.5%MC, U+1.0%MC, U+2.5%MC and U+5.0%MC, respectively). The experiment was arranged in a complete randomized block design with three replicates. Initially, 150 g of air-dried soil (<2 mm) was weighed into 500-ml plastic containers. A urea solution was added to each container (except CK) at the ratio of 200 mg N (kg soil)⁻¹. The moisture content of each sample was adjusted to 40–45% of the water-holding capacity, and readjusted by adding deionized water every 3 days. Each individual container was sealed with a polyethylene film containing 3 pin-sized holes to permit aeration. Temperature was kept constant at 25 °C during the entire experiment. The soil was sampled after 90 days and analyzed for SOC, total N, inorganic N, water soluble and exchangeable K, Na, Ca and Mg, extracellular enzyme activities and microbial community composition.

Table 1
The physical and chemical properties of experimental soil and biochar.

	Yield (%)	pH	Ash content (%)	EC (ms cm ⁻¹)	Surface area (m ² g ⁻¹)	SOC (%)	TN (%)	NH ₄ ⁺ -N (mg kg ⁻¹)	NO ₃ ⁻ -N (g kg ⁻¹)	Ws. K (g kg ⁻¹)	Ws. Na (g kg ⁻¹)	Ws. Ca (g kg ⁻¹)	Ws. Mg (g kg ⁻¹)	Ex. K (g kg ⁻¹)	Ex. Na (g kg ⁻¹)	Ex. Ca (g kg ⁻¹)	Ex. Mg (g kg ⁻¹)
Biochar	32.60	10.50	22.28	5.37	4.00	53.81	1.22	/	/	1.67	0.08	0.15	0.10	75.45	12.98	13.66	3.52
Soil		8.28	/	0.57	/	0.54	0.07	15.82	0.43	0.07	0.48	0.50	0.05	0.28	12.81	33.21	0.99

Abbreviations: Ex, exchangeable; Ws, water-soluble; TN, total nitrogen; SOC, soil organic carbon; EC, electrical conductivity. "/" not measured. Yield (%) = (weight of biochar) / (weight of feedstock) × 100.

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