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# Responses of soil microbial community structure changes and activities to biochar addition: A meta-analysis

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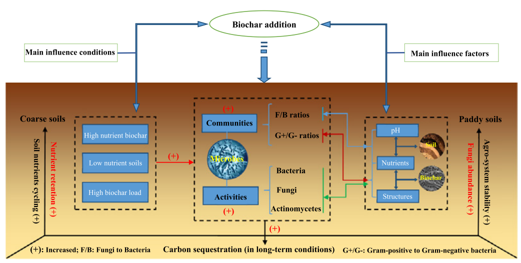
## HIGHLIGHTS

- Low temperature biochars addition in low pH soils greatly increased ratios of fungi to bacteria.
- Residue biochars application in dryland soils increased ratios of Gram-positive bacteria to Gram-negative bacteria the most.
- High load of biochar addition greatly enhanced microbial activities in low nutrients soils.
- Biochar nutrients and structural properties play the important role in soil microbial community structure changes and activities.

## GRAPHICAL ABSTRACT

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## ABSTRACT

The objective of this study was to investigate responses of soil microbial community structure changes and activities to biochar addition under different biochar characteristics, soil properties, and experiment conditions. A meta-analysis was conducted based on 265 datasets from 49 published studies. Results showed that biochar addition significantly increased the ratios of soil fungi to bacteria (F/B) and the ratios of Gram-positive bacteria to Gram-negative bacteria (G+/G−), and microbial biomass and activities. The enhancement of F/B ratios was most significant with addition of biochars produced at low temperatures to soils with lower pH and nutrients in a long-term condition, which improved ecosystem stability of agricultural soils. The F/B ratios were mainly affected by biochar nutrients, soil nutrients, and soil pH values. Biochar nutrients and structural properties (i.e., surface area and porosity) also played the important role in enhancing G+/G−, total microbial biomass, and activities of bacteria, fungi, and actinomycetes. The G+/G− ratios increased the most with addition of biochars produced with medium temperatures and residue accompanied with fertilizers in dry land (dried farmland) soils. High biochar load greatly improved the total phospholipid fatty acids, and activities of bacteria, fungi, and actinomycetes in fine/coarse, paddy soils, and soils with low nutrients, in turn increased the soil nutrient cycling. In addition, the structural properties of biochars were the most influencing factor to increase total microbial biomass and actinomycete activity. Overall, the enhancement of microbial activities and community structure shifts under biochar addition should promote soil nutrients cycling and carbon sequestration, and improve crop yields.



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## 1. Introduction

Biochar generally refers to a carbon-rich solid that is produced by the pyrolysis of biomass in oxygen-limited conditions (Lehmann et al., 2011). Biochar application in soils may be beneficial to carbon (C) sequestration, soil fertility, and ecosystem functioning (Cao et al., 2009; Liu et al., 2013). Soil microorganisms play a vital role in soil nutrient cycling, soil ecosystem stability, and soil C changes under biochar addition (Lehmann et al., 2011; Zhu et al., 2017). Microbial activities and community compositions are affected by biochar characteristics (e.g., feedstock materials, pyrolysis temperature, pH, and specific surface area (SSA)), soil physiochemical properties (e.g., pH, soil organic carbon (SOC), and soil total nitrogen (STN), and texture), and experiment conditions (e.g., biochar load, fertilization, and study duration) (Singh et al., 2010; Spokas et al., 2011; Biederman and Harpole, 2013; Rutigliano et al., 2014; Mitchell et al., 2015; Tian et al., 2016). Therefore, it is critical to determine responses of soil microbial community structure changes and activities to biochar addition with different biochar characteristics, soil properties, and experiment conditions.

Effects of biochar application on soil microbial community structure changes and activities remain controversial under different biochar characteristics, soil properties, and experiment conditions. Castaldi et al. (2011) showed that wood biochar addition had no or little effect on soil microbial activities and community compositions. However, addition of wood and cotton straw biochars or low pyrolysed biochar, particularly at high biochar addition rates, increased microbial activities and altered the community composition towards a more Gram-negative bacteria dominated (relative to fungi and Gram-positive bacteria) community (Nelissen et al., 2012; Gomez et al., 2014; Liao et al., 2016). Straw biochar addition increased activities and community structure changes of fungi, Gram-negative bacteria, and actinomycetes, while wheat husk biochar addition did not affect the microbes (Lu et al., 2014; Watzinger et al., 2014; Wang et al., 2015; Muhammad et al., 2016; Luo et al., 2017). In terms of soil properties, biochar addition in red soil reduced microbial activities of Gram-positive bacterial and fungal phospholipid fatty acids (PLFAs), whereas biochar amendment in black soil increased the ratios of fungi to bacteria, Gram-negative bacteria, and action-bacterial PLFAs (Wang et al., 2012). Biochar application in sandy clay loam and silt loam soils reduced the total PLFA, but increased the total PLFA in clay loam soil (Ameloot et al., 2014). In addition, biochar amendment in the fine textured soil reduced microbial activities compared to in coarse textured soil (Wang et al., 2017).

The experiment conditions, including different biochar application rates, experiment durations (residence time) of biochar addition in soil, and fertilization amendment with biochar, significantly influence soil microbial activities and community structure shifts. Mitchell et al. (2015) found PLFA concentrations specific to Gram-positive and Gram-negative bacteria as well as actinomycetes decreased during the first 16 weeks of biochar addition, while the PLFA concentrations and the ratio of bacteria to fungi increased during weeks 16–24 of biochar application. Soil Gram-positive bacterial and fungal activities decreased with 0.5% of biochar addition, but fungi activity increased with higher biochar application rates (Wang et al., 2012; Muhammad et al., 2014). Biochar addition with fertilizers significantly increased ratios of fungi to bacteria, but the opposite result was reported (Kelly et al., 2015; Luo et al., 2017). Therefore, it remains unclear how the soil microbial community structure changes and activities respond to different conditions of biochar amendment. So far a comprehensive synthesis to reveal the general responses of soil microbial activities and community structure changes to biochar addition under different conditions is still not available (Gul et al., 2015; Zhu et al., 2017).

Therefore, the purpose of this study was comprehensive and quantitative to synthesize responses of soil microbial community structure shifts and activities to biochar addition with different biochar characteristics, soil properties, and experiment conditions. Using the meta-analysis, we aimed to answer the following questions: How do soil microbial community structure changes and activities change with different biochar characteristics under different soil properties and experiment conditions? What are the main influencing factors on soil microbial community structure shifts and activities under biochar addition?

## 2. Materials and methods

### 2.1. Data sources

The peer-reviewed articles reporting effects of biochar addition on soil microorganisms with different biochars under different soil and experiment conditions were collected globally using Web of Science (http://apps.webofknowledge.com) and China National Knowledge Infrastructure (CNKI, http://www.cnki.net). Keywords and terms used for the literature online-searching were "biochar" and "soil microbial biomass, or soil microbial activity, or microbial community structure, or total PLFA, fungi, bacteria and actinomycetes" and "phospholipid fatty acids or PLFAs". Articles satisfying the following criteria were included for the meta-analysis: at least three replicates per treatment, biochar and control treatments in the same experimental site (i.e., the same experimental conditions), clearly reported biochar addition rates, and analyzed data of soil microbial activities (bacteria, fungi and actinomycetes), total microbial biomass and activities (total PLFA), and microbial community structure changes (fungi to bacteria ratios and Gram-positive bacteria to Gram-negative bacteria ratios) using the PLFA method (Kong et al., 2011). In addition, some datasets of soil bacterium, fungus and actinomycete activities, total microbial biomass, and microbial community structure changes were extracted from tables and figures of the publications, including values of the mean and standard deviation (SD) or standard error (SE). For two datasets with mean values but without SE or SD values, 1/10 of the mean values were assigned as the SD values (Luo et al., 2006; Luo et al., 2010; Liu et al., 2013).

Totally 265 biochar addition experiments from 49 papers met the criteria above and were utilized in this study (Text S1 and Tables S1–3). We only adopted the microbial community structure shifts and activities measured with the PLFAs, which was the most commonly used methods for microbial measurements and for similar meta-analysis (Z. Zhou et al., 2017). Total PLFA were used to estimate the total microbial biomass and activities. The activities of fungi (F), bacteria (B), and actinomycetes (Actino), Gram-positive bacteria (G+) and Gram-negative bacteria (G−) were measured with taxa-specific PLFAs. The datasets of community structure changes included the ratios of fungi to bacteria (F/B) and the ratios of Gram-positive bacteria to Gram-negative bacteria (G+/G−).

### 2.2. Data collection

The raw data were obtained numerically from the tables, texts, or extracted from the figures in the original papers with the Get-Data Graph Digitizer 2.26 software. To the multiple sampling dates, only the result of biochar effect on the latest sampling time and the uppermost soil layer was chosen (Geisseler et al., 2016). Data were collected based on paired measurements between the control and biochar treatments. The control was subject to the same experimental conditions without a biochar treatment (Nguyen et al., 2017). SE and pH ($CaCl_2$/KCl) were unified into SD and pH ($H_2O$), respectively (Liu et al., 2013; Jian et al., 2016; Nguyen et al., 2017). Biochar load was converted from t $ha^{-1}$ to % using soil bulk density and soil depth where biochar was applied. If bulk density was not reported, the Hydraulic Properties Calculator program was used to determine the bulk density based on soil texture (Biederman and Harpole, 2013; Nguyen et al., 2017).

The data were extracted from each study, including biochar characteristics (i.e., feedstock, pH, pyrolysis temperature, and SSA), soil properties (i.e., texture, pH, source, SOC, and STN), and experimental conditions, including biochar load (application rate), residence time of