Role of Biochar Amendment on Soil Carbon Mineralization and Microbial Biomass

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Abstract

To understand the influence of biochar properties (pyrolysis temperature and types) on soil physicochemical properties, we investigated the changes of soil organic carbon mineralization, nutrient contents and microbial biomass after 135 d incubation. Results showed that both corn straw (CB) and rice straw (RB) derived biochars increase the mineralization of organic carbon and nitrogen in the soil, and these biochars pyrolysised at 500 °C (CB500, RB500) significantly enhanced the mineralization of soil organic nitrogen. In comparison with control treatment, the application of biochar significantly increased the contents of soil organic carbon, available P and K in soil. Moreover, the activity of soil microbe was enhanced with biochar amendment. Among all treatments, RB500 significantly increased the content of soil microbial biomass carbon (379 ± 9 mg·kg⁻¹) in soil. Our results suggested that the application of biochars to soil improve soil quality, while the biochar type and pyrolysis temperature should be taken into consideration before its application in agro-ecosystem.

Keywords
Biochar, Soil, Soil Carbon Mineralization, Microbial Biomass

1. Introduction

With ~620 million tons of plant straw generated annually, China has an abundant biomass resource for various utilizations including its direct field return for increasing of soil organic carbon (SOC) accumulation (Zeng et al., 2007). One of the emerging strategies is to convert straw into biochar, a recalcitrant carbon-rich product obtained after pyrolysis under limited oxygen environment. (Lehmann et al., 2006) Biochar has advantages over fresh straw in enhancing the
capacity of long-term carbon (C) sequestration in soil. As one of the most important C pools on the global scale, the incorporation of biochar to soil serves as a long-term C sink (Lehmann, et al., 2006). The stabilization and transformation of C in biochar amended soil with respect to the inherent biochar C and the native SOC has been extensively investigated (Singh et al., 2014). However, there is a critical knowledge gap in how the influences of biochar pyrolysis temperatures and types on soil carbon mineralization.

Biochar derived from straw is not only an organic substance rich in carbon, but also contains a variety of nutrient elements such as nitrogen, oxygen, sulfur and inorganic carbonate components (Sanchez-Monedero et al., 2017). Studies have suggested that biochar amendment to soil can influence soil microbial biomass, activity and community structure (Farrell et al., 2013). Such microbial responses might occur because of the potential utilization of biochar as an energy source by microorganisms (Domene et al., 2014), as a habitat for microorganisms from grazers and as components to improve soil properties such as water holding capacity and nutrient availability (Wu et al., 2013). Although straw return to soil could also stimulate the growth and activity of microorganisms, increased emissions of greenhouse gas did always happen (Kuzyakov et al., 2014). In contrast, biochar amendment could decrease such emissions, and has a sustainable effect on soil microbial activity and diversity (Kolb, et al., 2009).

However, studies of biochar amendment on soil microbial properties were not consistent, likely due to variations in soil types (Gomez et al., 2014), biochar types and pyrolysis temperatures (Jones et al., 2012). Moreover, Jones et al. (Jones et al., 2012) also concluded that various effects of biochar on soil behaviour from laboratory studies were closely associated with its properties and incubation time. Therefore, we should pay attention to the role of biochar characteristics on the activities of soil microorganism.

2. Material and Methods

2.1. Soil Characterization

The bulk soil, located at 0 - 20 cm, was collected from Yingtan, Jiangxi province of China. This region is characterized as subtropical marine climate with mean annual precipitation of 1100 - 1785 mm, mean annual temperature of 13°C - 17°C. Collected soil was aired dried and sieved through 2 mm for future experiments. Soil pH was measured from a soil-water suspension (1:2.5 w/v) with a pH meter (Metler Toledo). Soil organic carbon (SOC) was determined by TOC analyzer (Multi N/C 3000). Soil total N was measured by Kjeldahl method, soil available N by the alkali hydrolysable method, soil available P by the Olsen method, and soil available K by the NH4OAC extraction. Soil total P and K were digested by HF-HClO4 and determined by the molybdenum-blue colorimetry and flame photometry, respectively. Measurement of soil CEC was accorded to the ammonium acetate compulsory displacement method (Yuan et al., 2011) (Table 1).
Table 1. The properties of tested soil.

| Property   | pH  | SOC g∙kg⁻¹ | Total N mg∙kg⁻¹ | Available N mg∙kg⁻¹ | Available P mg∙kg⁻¹ | Available K mg∙kg⁻¹ | CEC cmol∙kg⁻¹ |
|------------|-----|------------|-----------------|--------------------|-------------------|-------------------|--------------|
|            | 5.57| 20.87      | 2.17            | 261.33             | 54.63             | 130.00            | 8.37         |

2.2. Biochar Preparation

Two biochar materials were pyrolyzed from rice straw (RB), and corn straw (CB) under a limited-oxygen condition at different temperatures using a patented slow-pyrolysis process (China Patent No. ZL200920232191.9). Briefly, Straw materials were oven-dried for 12 h at 80° C before moved to a reactor, which was heated by 5˚C min⁻¹ to 300˚C, 400˚C, and 400˚C respectively. And then the reactor was maintained at designed temperature for 4 h till no further smoke exhaust. Biochar was then ground and sieved through a 0.25-mm mesh before further application. Detailed pyrolysis process was described in previous studies (Li et al., 2016). Then we obtained six biochar materials and denoted as RB300, RB400, RB500, and CB300, CB400, CB500. The characteristics of these obtained biochar samples were reported in our previous published papers (Ming, et al., 2016).

2.3. Soil Incubation and Microbial Analysis

100 g air-dried soil was weighed into a 500-mL plastic bottle, the obtained biochar samples were uniformly mixed with soil at a level of 1% (w/w) respectively. The biochar amended soil was adjusted to 60% of the saturated water-holding capacity every three days, and then incubated at 25˚C in a temperature-controlled incubator for 135 d. The bottles were covered with sterile membranes that permitted gaseous exchange and minimized moisture loss. Each treatment has three replications.

Soil samples were collected three times, on 45, 90 and 135 d, after which changes in soil microbial biomass carbon (MBC) contents were determined. Changes in soil properties such as the soil pH values, SOC, available N, available P and available K contents were measured at the end of incubation period (135 d).

2.4. Soil Carbon Mineralization in Soil

The mineralization of soil organic carbon was determined by indoor incubation and lye absorption method. Weigh 100 g of air-dried soil sample into 500 ml plastic bottle, add appropriate amount of deionized water to adjust to 60% of saturated water content, mix well, and pre-incubation for 1 week in a constant temperature incubator at 25˚C ± 1˚C. Seven treatments were designed. The biochar material was added at a level of 1% (w/w) to ensure that the material is thoroughly mixed with the soil, and each treatment was set to 3 replicates. The incubation bottles were sealed with a sterile membrane to ensure good aeration conditions in the bottle and the water was periodically added to maintain a con-
Y. M. Wang, M. Li

stant water content. The NaOH-containing absorption bottles were taken out on the 1st, 2nd, 4th, 7th, 14th, 21th, 28th, 42th, 56th and 72th days of the incubation period, and the excess BaCl₂ solution and the indicator were added respectively, and the hydrochloric acid was titrated to the end point with the calibration hydrochloric acid to calculate the amount of CO₂ released during the incubation period. The amount of total CO₂ evolution and net nitrogen mineralization were determined according to the alkali absorption method. Net nitrogen mineralization was calculated as \((\text{NO}_3^- + \text{NH}_4^+)_{72d} - (\text{NO}_3^- + \text{NH}_4^+)_{0d}\). Soil (5.0 g) was extracted with 50 mL 2 M KCl, shaken for 1 h, filtered and the amounts of \(\text{NH}_4^+\) and \(\text{NO}_3^-\) in solution were quantified using an automatic \(\text{NH}_4^+\) and \(\text{NO}_3^-\) analyzer (Bran + Lubbe AA3, Germany).

2.5. Data Analysis

Student-Newman-Keuls (S-N-K) of one-way ANOVA was applied to assess the significant differences (p < 0.05) among groups using SPSS 21.0. Data are presented as mean ± SD (n = 3). Charts and graphs were drawn by Sigmaplot 10.0.

3. Results and Discussion

3.1. Style and Spacing Change of Soil Organic Carbon Mineralization

During the incubation period, the mineralization rate of soil organic carbon gradually decrease, especially in the early stage of incubation, which is mainly related to the gradual decrease of available organic carbon in the soil (Figure 1). The results of different treatments showed that the addition of two biochar materials did not significantly affect the mineralization rate of organic carbon in the soil. The effects of the addition of biochar from different pyrolysis temperatures on the mineralization rate of soil organic carbon were not significant. However,

![Figure 1. Responses of C mineralization rate to different biochar addition during the incubation period.](image)

DOI: 10.4236/gep.2018.611013
the addition of RB300, induced a higher mineralization rate of soil organic carbon than other treatments, which may be related to the relatively high available organic carbon content in biochar material that pyrolysed at 300°C. Previous studies have shown that biochar materials prepared at lower temperatures usually richer in available organic carbon content than biochar materials prepared at higher temperatures.

The results of cumulative mineralization of organic carbon showed that the accumulation of organic carbon in RB300 treatment was the highest, which was 790 ± 15 mg C-CO$_2$ kg$^{-1}$ soil, which was significantly higher than that of the control treatment, while the other additions of biochar did not significantly change the cumulative mineralization of organic carbon. All the addition of biochars at different pyrolysis temperatures increased the mineralization of nitrogen in the soil to varying degrees, especially with the additions of RB500 and CB500, which significantly increased the nitrogen mineralization with 136 mg (NO$_3^-$ + NH$_4^+$) kg$^{-1}$ soil and 97 mg (NO$_3^-$ + NH$_4^+$) kg$^{-1}$ soil respectively. This indicates that the addition of biochar material can promote the conversion of soil organic nitrogen to inorganic nitrogen to some extent, which is consent with the previous studies (Table 2).

### 3.2. Change of Soil Chemistry Properties

After 135 days of incubation, the pH value of biochar treatments increased by an average of 0.16 units compared with the control, but the difference between the additions of biochars that pyrolysed at the same temperature was not significant (Table 3, $p < 0.05$). Compared to CK treatment, contents of organic carbon, available phosphorus and available potassium in biochar mediated soils increased by 26.1%, 20.6% and 282%, respectively. There was no significant difference in soil organic carbon content between the two straw biochar treatments; the addition of RB materials significantly increased the soil available potassium content, which was higher than the average treatment of corn straw biochar, while the incorporation of corn biochar induced 9.0% incensement on the average.

#### Table 2. The cumulative mineralization of organic carbon and nitrogen.

| Treatment | Cumulative mineralization C | Net mineralization | Cumulative mineralization N | Net mineralization |
|-----------|-----------------------------|--------------------|-----------------------------|--------------------|
|           | mg C-CO$_2$ kg$^{-1}$ soil   |                    | mg (NO$_3^-$ + NH$_4^+$) kg$^{-1}$ soil |        |
| CK        | 603 ± 18c                   | 283 ± 21a          |                             |                    |
| RB300     | 790 ± 15d                   | 188                | 313 ± 47ab                  | 30                 |
| RB400     | 567 ± 17ab                  | -36                | 318 ± 32ab                  | 34                 |
| RB500     | 549 ± 9a                    | -53                | 419 ± 19b                   | 136                |
| CB300     | 616 ± 12c                   | 14                 | 288 ± 38ab                  | 5                  |
| CB400     | 613 ± 9c                    | 11                 | 325 ± 53ab                  | 42                 |
| CB500     | 585 ± 14bc                  | -17                | 380 ± 60b                   | 97                 |
Table 3. Chemical properties of soils under different biochar amendments after 135 days of incubation.

| Treatment | pH       | SOC g kg$^{-1}$ | Available N mg kg$^{-1}$ | Available P mg kg$^{-1}$ | Available K mg kg$^{-1}$ |
|-----------|----------|-----------------|---------------------------|--------------------------|--------------------------|
| CK        | 4.33 ± 0.03a | 20.8 ± 0.7a     | 216.8 ± 7.3bc             | 82.1 ± 3.1a              | 123.1 ± 2.4a             |
| RB300     | 4.48 ± 0.04bc | 26.3 ± 0.6b     | 207.6 ± 9.2abc            | 94.4 ± 0.7b              | 528.1 ± 27.7d            |
| RB400     | 4.53 ± 0.04cd | 26.3 ± 0.3b     | 187.4 ± 7.3a             | 93.9 ± 2.7b              | 568.8 ± 21.7e            |
| RB500     | 4.58 ± 0.03d  | 25.0 ± 3.1b     | 190.8 ± 6.0a             | 93.9 ± 2.6b              | 623.8 ± 10.5f            |
| CB300     | 4.45 ± 0.02b  | 25.9 ± 0.7b     | 227.9 ± 19.9c            | 100.5 ± 1.4c             | 362.5 ± 10.2bc           |
| CB400     | 4.46 ± 0.02bc | 26.3 ± 0.8b     | 218.7 ± 3.7bc            | 102.7 ± 1.3c             | 390.6 ± 12.0c            |
| CB500     | 4.43 ± 0.04b  | 27.0 ± 1.2b     | 198.5 ± 6.0ab            | 104.3 ± 2.1c             | 350.0 ± 10.2b            |

...in soil available phosphorus than rice straw biochar ($p < 0.05$). The difference of soil organic carbon, available phosphorus and available potassium between the different pyrolysis temperatures of the same material did not significant ($p < 0.05$). The addition of low temperature pyrolysis straw biochar (300˚C) did not significant affect the soil alkali nitrogen content; while the application of RB400, RB500 and CB500 treatment significantly decrease the alkali nitrogen content by 10.4% and 8.1%, respectively ($p < 0.05$). Previous studies have also shown that the addition of biochar promotes the mineralization of soil organic nitrogen, thereby reducing the reduction of available organic nitrogen in the soil.

3.3. Change of Soil Microbial Biomass

The concentrations of microbial biomass carbon (MBC) in all treatments decreased during the incubation period, and the overall content of MBC was 45 d > 90 d > 135 d, indicating that the activity of microorganisms in the soil gradually reduced. Possible reasons due to the available nutrients in the soil. Results of different incubation time showed that the addition of biochar materials with different pyrolysis temperatures can promote the activity of microorganisms in soil to some extent, and the content of MBC in each treatment is higher than that of CK treatment. After 135 days of incubation, the soil microbial biomass carbon content was 185 ± 26 mg·kg$^{-1}$ in CK (Figure 2). The addition of straw biochar that pyrolysed at a low temperature (300˚C) did not significantly affect the MBC content, while the addition of straw biochars that prepared from 400˚C and 500˚C pyrolysis temperature significantly increased the MBC content by 50.1% and 84.3% ($p < 0.05$), respectively. With the addition of biochar pyrolysed at the same temperature, the effect of RB500 induced more incensement in MBC content than CB500, which is nearly 379 ± 9 mg·kg$^{-1}$ in the soil.

4. Conclusion

1) Biochar could increase the mineralization of organic carbon and nitrogen in the soil, especially the biochar prepared under low temperature conditions.
2) Biochar addition promoted the activity of microbial populations in the soil. Rice straw biochar pyrolysised at 500°C can significantly increase the content of soil microbial biomass carbon in soil.

**Acknowledgements**

This work was supported by the Foundation Research Project of Jiangsu Province (BK20160155; BK20160859).

**Conflicts of Interest**

The authors declare no conflicts of interest regarding the publication of this paper.

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