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MICROBIOLOGICAL CHARACTERISTICS OF BIOCHAR AMENDED ALLUVIAL MEADOW SOIL

GALINA PETKOVA; KOSTADINKA NEDYALKOVA; ANGELINA MIKOVA;
IRENA ATANASSOVA*
Nikola Poushkarov Institute of Soil Science, Agrotechnologies and Plant Protection, 1331 Sofia

Abstract

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Biochar (BC), produced by the pyrolysis of biomass under limited oxygen is highly stable and resistant to microbial decay and can serve as a long-term sink of carbon. During the last years biochar from different origin is studied as a soil amendment in many countries. The ecological importance of biochar addition is connected with decreasing of greenhouse gasses emissions and associated with them climate changes, and improvement of soil structure and fertility. The effect of biochar amendment on microbial amount and activity of Alluvial meadow soil was studied in field experiments with wheat and maize cultivated in crop rotation. The number of the main groups of soil microorganisms, CO_2-production and microbial biomass were determined. A stimulating effect of biochar amendment on the soil microflora was established in the study. The CO_2-production and the bacterial amounts were positively affected to the greatest extent. The results obtained confirm the importance of biochar addition as a promising method for soil fertility conservation.

Key words: biochar, soil microorganism, CO_2-production, microbial biomass, Alluvial- meadow soil

Introduction

The irrational use of soil resources in the last decades has lead to reduction of soil organic matter in many countries. This negatively affects not only soil fertility but also the associated global environmental problems, climate changes, biodiversity loss, desertification of large areas. In order to counteract the processes of soil organic matter decrease and maintain a positive balance of humic substances, it is required to utilize variable sources of organic matter. Recently, biochar has been of great interest in this regard. It is obtained by pyrolysis (incomplete combustion) of biomass and is a subject of research in many countries (Great Britain, New Zealand, Japan, USA, etc.) as a means of improving soil structure and fertility (Ogawa, 1994; Steiner et al., 2004; Lehmann, 2007). The presence of polycyclic and aromatic structures in BC makes it very resistant to decomposition in the soil. During its slow mineralization, compounds involved in the humus substances synthesis are formed. At the same time, BC has a high capacity to adsorb chemical substances, preventing the nutrient elements leaching by surface water. In addition, the decomposition of agricultural wastes, due to their rapid mineralization is connected with emission of large amounts of CO_2 in the atmosphere. Carbon in BC is in a stable form, therefore during its decomposion smaller amounts of CO_2 are released leading to reduced emissions of CO_2 in the atmosphere.

Most of the studies on the use of BC in agriculture are from agronomic standpoint. Less research has been conducted on the hydro-physical and water-retention properties

**Corresponding author*: i.d.atanassova@abv.bg

of biochar (Kinney et al. 2012; Gray et al. 2014). Often the hydrophobic character in biochar treated soil was attributed to fungal colonisation (Abel et al. 2013). In recent years, data were obtained proving its positive effect on particular groups of soil microorganisms, nodule-forming bacteria, (Mia et al., 2015), mycorrhizal fungi (Ishii and Kadoya, 1994; Warnock et al., 2007) and also on general indicators of soil biological activity (Kolb et al., 2008; Steiner et al., 2008). Other authors establish no impact of BC addition on soil microflora (Ameloot, 2014; Sun et al., 2014). Sun et al. (2014) reported that soil respiration, nitrification and arylsulphatase activity of two soils were not affected by biochar amendment. Obviously, the effect of BC on soil microorganisms varies depending on its composition, conditions of production, its physical properties, soil type, rates of application and duration of BC degradation in soil. Until now, there is no evidence of a negative impact of BC on plant roots.

The aim of the present study is to determine the influence of BC addition on the microbiota of Alluvial meadow soil depending on the duration of the period following BC application in soil.

Materials and Methods

The study was conducted in April 2017 in the experimental field of Nikola Poushkarov Institute of Soil Science, Agrotechnologies and Plant Protection in the village of Tsalapitza. The samples from a field experiment with wheat "Sadovo 1" variety on Alluvial meadow soil (Fluvisol) were analyzed. The following fertilization was applied: N10 (in the form of urea urea), P12 (as triple superphosphate) - and K10 (applied as potassium sulphate). The P and K fertilizers were introduced prior to the main soil tillage and N fertilizer before the presowing treatment. There was also spring nourishment with ammonium nitrate in March. General soil characteristics were the following: total organic C $\%_{2012,\ 2016,\ 2017}$ (0.8%-1%), total $N2_{012,\ 2016,\ 2017}$(0.09-0.1%) $pH_{soil\ 2012}$ (5.7-6.0); $pH_{soil\ 2016}$ (6.0-6.3); $pH_{soil\ 2017}$ (6.0-6.1). Grids Δ 2 m, ~ 40 m^2 were constructed at the four plots and samples for microbiological analysis were taken at six points at 0-10 cm depth. Sample water-repellency (soil hydrophobicity) was measured by the water drop penetration time (WDPT) method (Doer et al. 2002). The soil samples were weighed and subsequently equilibrated at the ambient air humidity before measuring water drop penetration time in the laboratory at recorded humidity and temperature. The WDPT measured at the laboratory for all soils treated with biochar including the control variant, and the biochar used in the field trial showed that sample were hydrophilic (non-water repellent, WDPT < 5s).

The following treatments were included in the study: 1. Soil without biochar amendment; 2.Soil amended in 2012 with BC (200 kg/da), produced by pyrolysis of maize residues at 500^0C; 3. Soil amended in 2016 with BC (230 kg/da), produced by pyrolysis of rice straw at the same conditions; 4. Soil amended with biochar, produced by pyrolysis of oak residues at the same conditions (applied at rate of 300 kg/da in 2017). The samples of treatments 2 and 3 were taken from parcels where the wheat was cultivated with maize in crop rotation. All samples were collected from a soil depth 0-10 cm in the phase tillaring and analyzed for the following microbiological parameters: 1. Colony-forming units (CFU) number of the main groups of soil microorganisms by dilution plate method (Gushterov et al., 1977). The following nutrient media were utilized: meat-peptone agar for ammonifying bacteria; stach-ammonium agar for actinomycetes and Chapek agar for microscopic fungi; 2. CO_2-production (total biological activity) (Alef, Nannipieri, 1998); 3. Microbial biomass quantity was assessed by the method of Anderson-Domsch (Anderson-Domsch,1978).

All analyses were done using three replicates per treatment. The least significant differences (LSD) were determined at $P \leq 0.05$ using ANOVA (the test of Duncan).

Results and Discussion

The data presented in Table 1 show that the number of ammonifying bacteria was significantly higher than that of the control soil (up to two times) in all biochar amended treatments regardless of the year of its application to the soil. Unlike the mentioned group of microorganisms, the numbers of actinomycetes and microscopic fungi were clearly dependent on the duration of the period following biochar addition. For example, in treatment 4 (BC applied in the 2017 year), the amount of actinomycetes was close to that of the control. Unlike actinomycetes, the number of microscopic fungi was higher, however the difference with the control is not statistically proven. In treatment 3 (BC applied in 2016) the amounts of these groups of microorganisms were significantly higher than treatment 1. Higher values for the number of main groups of soil microorganisms in the treatment with five-year long period following BC addition to the soil prove its slow mineralization. This is probably connected with a gradual release of substances used as nutrient sources by the soil microorganisms. The results on the amounts of major groups of microorganisms are in accordance with the data of Prayogo et al. (2008) and Petkova et al. (2015), which established that biochar addition stimulated the development of soil bacterial populations.

The values obtained for CO_2-production and quantity of microbial biomass (Figures 2 and 3) are in agreement with the data related to the number of soil microorganisms and prove the positive effect of the biochar addition on the soil microflora. The CO_2 production values for treatments with BC amendment applied in 2017 and 2016 were significantly higher than that of the control. In treatment 2 the CO_2-production was close the control, probably due to the delayed biochar mineralization. Relatively easily assimilated by the soil microorganisms chemical compounds in BC are likely to be depleted and mineralization processes occur with low intensity. The amount of microbial biomass is higher in all biochar amended treatments compared to the control, and the differences are statistically proven for treatments with BC applied in the present year and a year ago. The results obtained confirmed the data of Kobl et al. (2008), which reported for increase of microbial biomass and total biological activity after BC addition to soil.

The presented results show a long-term positive impact of the BC amendment on the soil microorganisms. The mechanisms of impact of BC on the size and composition of soil biota have not been elucidated yet. Keech et al. (2005) suggest that BC adsorbs substances with allelopathic activity, phenolic compounds and others, and reduce the concentration or deactivating micro-toxic compounds in the soil. According to Ameloot (2013), local BC particles in the soil create areas with increased nutrient concentration and amounts of microorganisms. Other authors (Zackrisson, 1996; Warnock et al., 2007) consider that the high porosity of BC may provide favoraruble habitats for microorganisms, altering predation rates by soil microfauna.

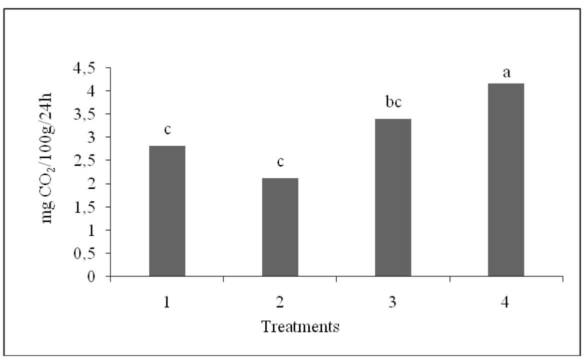

Fig. 1. CO_2-production of Alluvial Meadow Soil Amended with Biochar

1.Control; 2. + BC (applied in 2012); 3.+ BC (applied in 2016); 4.+ BC (2017);

Table 1. Number of the microorganisms (CFU/g) of Alluvial meadow soil amended with BC

Treatments	Ammonifying bacteria	Actinomycetes	Microscopic fungi
	1.10^5	1.10^5	1.10^3
1. Control	3.33^c	0.5^b	0.073^b
2.+ BC applied in 2012	7.53a	0.93^{ab}	0.14^a
3.+ BC applied in 2016	6.81^{ab}	1.4^a	0.15^a
4.+ BC applied in 2017	5.63^b	0.58^b	0.13^{ab}
LSD $P \leq 0.05$	1.56	0.54	0.061

*Values in the same column, which are followed by different letters are different at $P \leq 0.05$

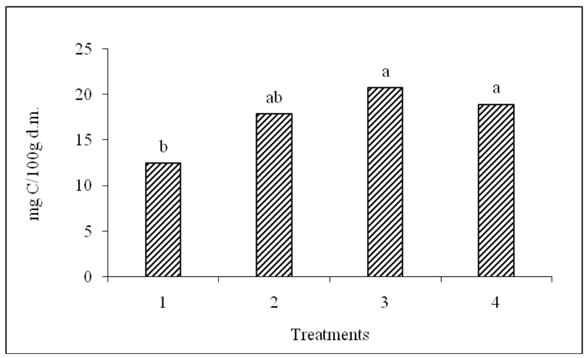

Fig. 2. Microbial Biomass of Biochar Amended Alluvial Meadow Soil

1.Control; 2. + BC (applied in 2012); 3.+ BC (applied in 2016); 4.+ BC (2017);

*Values in the same column, which are followed by different letters are different at $P \leq 0.05$

Conclusion

Key outcomes of the study are the following: (i) A long-term beneficial effect of biochar amendment in Aluvial-meadow soil on microbial biomass and the growth of soil microorganisms has been established. The bacterial populations and CO_2-production were stimulated to the greatest extent; (ii) the results obtained confirm the importance of biochar addition as a promising method for soil fertility conservation.

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