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## Impact of biochar application to a Mediterranean wheat crop on soil microbial activity and greenhouse gas fluxes

S. Castaldi <sup>a,\*</sup>, M. Riondino <sup>a</sup>, S. Baronti <sup>b</sup>, F.R. Esposito <sup>a</sup>, R. Marzaioli <sup>a</sup>, F.A. Rutigliano <sup>a</sup>, F.P. Vaccari <sup>b</sup>, F. Miglietta <sup>b,c</sup>

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#### ABSTRACT

Biochar has been recently proposed as a management strategy to improve crop productivity and global warming mitigation. However, the effect of such approach on soil greenhouse gas fluxes is highly uncertain and few data from field experiments are available. In a field trial, cultivated with wheat, biochar was added to the soil (3 or 6 kg m $^{-2}$ ) in two growing seasons (2008/2009 and 2009/2010) so to monitor the effect of treatments on microbial parameters 3 months and 14 months after char addition. N<sub>2</sub>O, CH<sub>4</sub> and CO<sub>2</sub> fluxes were measured in the field during the first year after char addition. Biochar incorporation into the soil increased soil pH (from 5.2 to 6.7) and the rates of net N mineralization, soil microbial respiration and denitrification activity in the first 3 months, but after 14 months treated and control plots did not differ significantly. No changes in total microbial biomass and net nitrification rate were observed. In char treated plots, soil N<sub>2</sub>O fluxes were from 26% to 79% lower than N<sub>2</sub>O fluxes in control plots, excluding four sampling dates after the last fertilization with urea, when N<sub>2</sub>O emissions were higher in char treated plots. However, due to the high spatial variability, the observed differences were rarely significant. No significant differences of CH<sub>4</sub> fluxes and field soil respiration were observed among different treatments, with just few exceptions. Overall the char treatments showed a minimal impact on microbial parameters and GHG fluxes over the first 14 months after biochar incorporation.

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#### 1. Introduction

Biochar from crop residues is one of the most recent environmental management approaches proposed for both improving crop productivity and global warming mitigation (Lehmann et al., 2006). In fact, addition of biochar to soils is proposed as a mean for long term C sequestration (Lehmann et al., 2006). The rational beyond this idea is that the C-CO<sub>2</sub> fixed by plants, during the photosynthetic process, will undergo a much slower decomposition (turnover rate from hundreds to thousand years) if transformed in the much more stable form "biochar" (Lehmann, 2007), compared with untreated residues, thus creating a fast in/slow out carbon dynamic into the soil. Moreover, biochar has been often found to further improve the terrestrial C sink by enhancing crop productivity (Lehmann and Rondon, 2006; Lehmann et al., 2006; Rondon et al., 2007; Steiner et al., 2007; Zhang et al., 2010; Vaccari et al., 2011), thanks to its effects on physical, chemical and biological soil characteristics (Lehmann et al., 2003; Lehmann and Joseph, 2009;

Atkinson et al., 2010; Graber et al., 2010; Uchimiya et al., 2010) or, as recently hypothesized by Graber et al. (2010) by inducing hormesis. However, agricultural ecosystems also represent a critical environment for the emission/consumption balance of other greenhouse gases, apart from CO<sub>2</sub>. Cultivated soils are the main source of  $N_2O$  in terrestrial ecosystems (IPCC, 2007), moreover agricultural management practices strongly decrease the soil CH<sub>4</sub> sink and often enhance CH<sub>4</sub> production (Smith et al., 2000; IPCC, 2007). The flux balance of these two greenhouse gases is strictly related to soil properties and substrate inputs, which can be strongly altered by soil management practices. Hence, a relevant question might be if the proposed practice of biochar incorporation to soil has an influence on soil-atmosphere exchanges of N<sub>2</sub>O and CH<sub>4</sub> and, if so, in which direction. From a theoretical point of view, biochar might enhance N<sub>2</sub>O emissions by creating more favorable conditions for soil microflora involved in N cycle. Charcoal is known to have properties which allow for NH<sub>4</sub> (Berglund et al., 2004; Lee et al., 2005; Lehmann et al., 2006) and gas absorption (Bagreev et al., 2001; Hitoshi et al., 2002), which improve soil water retention and aeration (Yanai et al., 2007), and increase soil cation exchange capacity and soil pH (Tryon, 1948; Liang et al., 2006; Cheng et al., 2008). This latter effect, more evident in acidic soils, could increase phosphorus

<sup>&</sup>lt;sup>a</sup> Dipartimento di Scienze Ambientali, Seconda Università degli Studi di Napoli, via Vivaldi 43, 81100 Caserta, Italy

<sup>&</sup>lt;sup>b</sup> Institute of Biometeorology (IBIMET), National Research Council (CNR), Via Caproni 8, 50145 Firenze, Italy

<sup>&</sup>lt;sup>c</sup> Foundation Edmund Mach, Via E. Mach 1, 38010 San Michele all'Adige (TN), Italy

<sup>\*</sup> Corresponding author. Tel.: +39 0823274646; fax: +39 0823274605. E-mail address: simona.castaldi@unina2.it (S. Castaldi).

availability and reduce the availability of Al, thus reducing its potential toxic effect on plants and microorganisms (Hammes and Schmidt, 2009). Less acidic soil conditions are expected to favor microbial activity and in particular to increase autotrophic nitrifying bacteria activity (De Luca et al., 2009).

Available studies on the effect of biochar incorporation into the soil on greenhouse gas fluxes are not always consistent, being N2O found to decrease in char treated trials in some but not all the reported experiments (Oviedo and Sanz, 2005; Yanai et al., 2007; Spokas and Reicosky, 2009; Van Zwieten et al., 2009; Singh et al., 2010; Zhang et al., 2010; Scheer et al., 2011; Taghizadeh-Toosi et al., 2011). Equally uncertain is the effect of biochar on CH<sub>4</sub> fluxes (Spokas and Reicosky, 2009; Zhang et al., 2010; Scheer et al., 2011). If evidence will be provided that biochar stimulates crop productivity and limits or reduces N<sub>2</sub>O emissions, being neutral for soil CH<sub>4</sub> sink and production, the climate mitigation potential of this agronomical practice might be even more successful than expected. Vice versa, an enhancement of N<sub>2</sub>O production and an increase of the ratio CH<sub>4</sub> produced/CH<sub>4</sub> oxidized might in part counter balance the positive effect of biochar on long term C-CO<sub>2</sub> sequestration. At present, few data are available to support these hypotheses at field scale and studies are needed where the effects of biochar treatments on plant growth yield, soil microbial activity and greenhouse gas fluxes are simultaneously evaluated.

This paper analyses the impact of biochar incorporation into the soil at different rates (3 and 6 kg m<sup>-2</sup>) on several soil microbial activities involved in C and N cycle and on field fluxes of greenhouse gases (N<sub>2</sub>O, CH<sub>4</sub> and CO<sub>2</sub>). Results presented in this paper refer to field trials made with durum wheat over two consecutive seasons in which positive effects of biochar on crop yields were observed (Vaccari et al., 2011).

#### 2. Materials and methods

#### 2.1. Site and experimental design

The experiment was carried out with durum wheat (cv. *Neolatino*) during the growing seasons 2008/2009 and 2009/2010, in Pistoia (Tuscany, Italy, Lat. 43°56′N, Long. 10°54′E, 65 m a.s.l.). Total rainfall from September to July was 1159 mm in 2008/2009 and 1222.8 mm in 2009/2010 while mean air temperature was 13.9 °C (2008/2009) and 15.1 °C (2009/2010) (data from an automatic weather station at site). The soil is a silty-loam (USDA, soil classification) with a bulk density of 1.2 (Mg m $^{-3}$ ), pH 5.4, organic C content of 21 g kg $^{-1}$  (dry soil), total N of 1.2 g kg $^{-1}$  (soil dry weight) and a CEC of 18 meq 100 g $^{-1}$  (soil dry weight).

For the present experiment three treatments were compared: wheat crop (control), wheat crop treated with 3 kg biochar m<sup>-2</sup>, wheat crop treated with 6 kg biochar m<sup>-2</sup>. We also aimed to compare the effect of these three treatments, on several soil parameters, 3 months and 14 months after biochar addition to the soil. To reach this goal, we incorporated biochar into the soil in two growing seasons, 2008/2009 and 2009/2010, in different plots and soil was sampled using a synchronic approach in 2010. At the date of soil sampling, 3 months had passed since biochar incorporation into 2009/2010 plots and 14 months had passed since biochar incorporation into 2008/2009 plots. Each treatment was replicated using 4 subplots of 25 m<sup>2</sup> each. The subplots were distributed over an area of 5000 m<sup>2</sup> using a randomized block design.

Soil was tilled in October (2008 and 2009) and biochar was manually applied before sowing (on 16th January 2009 for experimental plots 2008/2009 and on 14th December 2009 for experimental plots 2009/2010), and partially buried with a rotary hoeing tillage over 15 cm depth. Wheat was sown in rows (450 seeds per  $\rm m^2$ ) and partially buried with a rotary hoeing till-

age. Nitrogen–phosphate and phosphorous fertilizer was distributed at sowing ( $2.2 \,\mathrm{g \, m^{-2}}$  of N and  $5 \,\mathrm{g \, m^{-2}}$  of  $\mathrm{P_2O_5}$ ), a second fertilization was done in mid March ( $10 \,\mathrm{g \, N \, m^{-2}}$  N as ammonium nitrate) and a third fertilization was done at the end of April ( $10 \,\mathrm{g \, N \, m^{-2}}$  as urea).

The applied biochar was a commercial horticultural charcoal provided by Lakeland Coppice Products (England) derived from coppiced woodlands (beech, hazel, oak and birch), obtained by pyrolysis at 500 °C. The used biochar has the following characteristics: total C (g kg $^{-1}$ ) 840, total N 12 g kg $^{-1}$ , available N 0.03 g kg $^{-1}$ , pH (1:2.5 H $_2$ O) 7.2, max water absorption 4.5 g g $^{-1}$  of d. m., bulk density 1.8 Mg m $^{-3}$ . Other chemical characteristics are reported by Vaccari et al. (2011).

#### 2.2. Soil and gas field sampling

A synchronic soil sampling was performed in March 2010 in all subplots to quantify microbial biomass and the rates of several microbial processes involved in the C and N cycle (soil respiration, net N mineralization, net nitrification, potential net nitrification, denitrification enzyme activity). Soil pH and available mineral N were also measured on sampled soils.

A composite sample (four soil cores sampled at 0–10 cm depth, 5 cm diameter) was obtained for each subplot (4 subplots per each treatment). The four composite samples were truly mixed, sieved at 2 mm (mesh sieve) and stored in plastic bags at 4 °C for subsequent biological and chemical analyses.

Field soil respiration (SR) was measured in the growing season 2008/2009 using an infrared gas analyzer coupled with a closed dynamic chamber (EGM-2 with SRC-1, PPSystems, Hitchin, UK). The chamber was inserted on PVC collars (10 cm height, 10 cm inner diameter) for measurements. Three collars (12 per each treatment), inserted 7 cm into the soil, were placed in each plot in January 2009 and were left in place throughout the course of the experiment until June 2009. Measurements were always made between 11:00 am and 02:00 pm.

N<sub>2</sub>O and CH<sub>4</sub> fluxes were measured only in the plots treated with char in the season 2009/2010 using closed static chambers (4 replicates per treatment, one chamber in each subplot). Chambers (Hutchinson and Mosier, 1981; Smith et al., 1995) were made of high-density polyvinyl chloride (15 cm high × 15 cm in diameter) and, during gas sampling, were placed on a base (7 cm high  $\times$  15 cm in diameter) inserted 5 cm in the ground. Three gas samples were taken from each chamber using a gas sampling port (time zero and 30 and 60 min) and stored in 20 mL air-tight evacuated glass vials crimped with butyl rubber lids and aluminum crowns. Concentrations of N<sub>2</sub>O and CH<sub>4</sub> were determined, within a couple of days, using a gas chromatograph (Series 800 Fisons, Milan, Italy). A modified system from Loftfield et al. (1997) was set up to analyze N<sub>2</sub>O and CH<sub>4</sub> on the same gas sample. Gas was loaded on a 2 mL loop connected to a 10-ports valve (Valco Europe, Switzerland). A pre-column of 1 m (external diameter 1/8", internal diameter 0.08"), filled with Porapak 80-100 Q and maintained at 60 °C, was connected to the 10-port valve in order to operate front-flush and back-flush. From the pre-column, the gas passed into the main column (T Porapak 80-100 Q, external diameter 1/8", internal diameter 0.08", 2 m length), also held at 60 °C. Then it was directed, via a 4-ports valve (Valco Europe, Switzerland) firstly to a flame ionization detector (FID) and, after 86 s, to an electron capture detector (ECD), held at 280 °C. Pure nitrogen was used as carrier gas at a flow rate of 40 cm<sup>3</sup> min<sup>-1</sup>. Calibrated standards (Air Liquide Italia) were used and were injected on duplicate every 20 samples to allow for instrumental drifting. Flux rates were determined via linear regression of the three sampling points for each chamber and by applying a temperature and pressure correction.

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