# Charcoal (biochar) as a carbon sequestration approach and its effect on soil's functions

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#### **Abstract**

We compared the microbial biomass, basal respiration, dehydrogenase activity, mineral-N (nitrogen) and hot-water extractable carbon (HWC) of the soil with and without charcoal as a biological and a chemical property. Using quantitative polymerase chain reaction (qPCR) we tested for the presence of microbial groups and genes associated with methane metabolism as indicators of any change in microbial diversity resulting from the addition of charcoal. As a soil physical property we analysed if the addition of charcoal increased soil water repellency.

Our preliminary analysis showed that the addition of charcoal resulted in no significant change to microbial biomass, basal respiration or HWC. The qPCR analysis showed that there was also no significant change to biodiversity. The soil in the orchard prior to the addition of charcoal was not hydrophobic; after the addition of charcoal this did not change.

#### **Key Words**

Basal respiration, microbial biomass, hot water carbon, qPCR, carbon sequestration.

## Introduction

Biochar is a term reserved for the plant biomass derived materials contained within the black carbon. This definition includes chars and charcoal, and excludes fossil fuel products or geogenic carbon (Lehmann *et al.* 2006). Work on terra preta de índio (TP) soil, the fertile Amazonian Dark Earths, has served as a major inspiration for the use of biochar as a promising soil additive promoting crop growth and carbon storage (Glaser *et al.* 2002; Glaser and Woods 2004; Lehmann *et al.* 2006; Glaser 2007). The sequestering of carbon through the addition of charcoal to agricultural and horticultural soils is a strategy that has recently gained interest as a way to mitigate climate change. To assess the practical feasibility of this strategy the impact of the addition of charcoal on soil biophysical properties needs to be understood. As yet, only limited field trials have been conducted to investigate this. The objective of this study is to measure the effects of the addition of charcoal to soil in an integrated research apple orchard in Havelock North (Figure 1a), on a number of biophysical soil properties. The apple trees in the integrated orchard system were 12 yr old. The apple variety was Pacific Rose, and the rootstock variety was 'MM.106'. The tree spacing was 3.4 m within the rows and 4.5 m between the rows. A 0.5-m wide strip under the trees was kept bare by regular herbicide applications. The apple trees were drip-irrigated during the vegetative period. The irrigation, nutrient, and pest management followed the guidelines of integrated fruit production (Wiltshire 2003).

## Methods

Charcoal was added at a rate of 2 kg/m<sup>2</sup> (20 t/ha), and mixed with the top 0.1 m of the soil (Figure 1b). This served as the soil-plus-charcoal treatment. The charcoal was added to three sampling sites within a single tree row. Three separate sampling sites in the same tree row, but without the addition of charcoal served as the control. The 6 sites had the same soil type and climate, and had received the same orchard management.

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Figure 1a. Left. Havelock North integrated apple orchard. Figure 1b. Right. Mixing charcoal into the soil

We compared the microbial biomass according to the method of Höper (2006). Microbial respiration was determined by using basal respiration (Őhlinger *et al.* 1996), Dehydrogenase activity (Chandler and Brooks 1991) and microbial biodiversity of the soil with and without charcoal as biological soil properties.

Detecting microorganisms by polymerase chain reaction (PCR) amplification of Deoxyribonucleic acid (DNA) extracted from environmental samples has an advantage over culture techniques which requires recovery and growth of active organisms (Johnston and Aust 1994; Sivakumaran et al. 1997). DNA was purified from three samples of each soil type using PowerSoil DNA kit (MoBio Laboratories, Carlsbad, Ca, USA) following the maker's instructions. Microbial biodiversity was analysed using quantitative polymerase chain reaction (qPCR). Sixteen microlitre qPCR reactions were performed in triplicate on two separate plates on a LightCycler 480 (Roche Applied Science, Indianapolis, IN, USA) using 25 ng of DNA, 0.5 µM primers with LightCycler® 480 SYBR Green I Master (Roche Applied Science, Indianapolis, IN, USA). Cycling conditions included an initial hot start at 95 °C for 5 min, followed by 40 cycles of 95 °C for 10 s, 53 °C for 10 s and 72 °C for 30 s. Each qRT-PCR was ended by the addition of a dissociation curve analysis of the amplified product. This involved denaturation at 95 °C for 5 s, cooling to 65 °C for 1 minute and then gradual heating at 0.21 °C/s to a final temperature of 97 °C. Raw crossing points were converted to quantities representing relative expression levels using a modified comparative method (Pfaffl 2001) and with correction for different amplification efficiencies (Ramakers et al. 2003). Primers used in qPCR of 16S rRNA in Eubacteria, and sulfate-reducing bacterial groups Desulfovibrionaceae, Desulfobacteraceae and Desulfobulbus were 9/27f, 519f, 519r, 907r, DSVIB679r, DSBA355f and DSBb279f from Stubner (2004); primers for urease gene wereURE1F and URE2R from Koper et al. (2004); primers for type I and II methanogtroph 16S rRNA were taken from Chen et al. (2007) and for methane oxidases from Holmes et al. (1995). We tested for the presence of microbial groups and genes associated with methane metabolism.

Hot water extractable carbon (HWC) (Ghani *et al.* 2003) and mineral-N (nitrogen) content (Keeney and Nelson 1982) were compared as chemical soil properties.

As a soil physical property we analysed if the addition of charcoal caused the soil to become hydrophobic. (Roy and McGill 2002).

Data was analysed using 95% confidence intervals.

## Results Biological properties

Addition of charcoal had no significant impact on basal respiration or microbial biomass (Figure 2a & b).

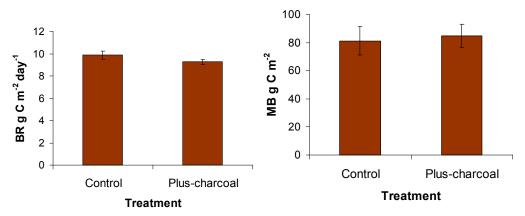


Figure 2a. Left. Basal respiration measured in soil with and without the addition of charcoal. (Error bars are mean  $\pm$  s.e., n = 9). Figure 2b. Right. Microbial biomass measured in soil with and with out the addition of charcoal. (Error bars are mean  $\pm$  s.e., n = 9).

Following experimentation we found that dehydrogenase activity cannot be reliably quantified in soil if charcoal amendments have been added, since the charcoal interferes with the absorbance readings.

There was no significant change in population size of methanogens, methanotrophs, ammonia-oxidising bacteria, eubacteria, fungi, archaea,  $\alpha$ -proteobacteria and  $\beta$ -proteobacteria resulting from the addition of charcoal to the soil.

## Chemical properties

The hot water extractable carbon (HWC) content was significantly lower in the soil-plus-charcoal than in the control (Figure 3a).

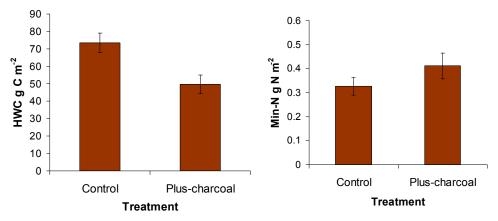


Figure 3a. Left. Hot water extractable carbon (HWC) content of soil with and with out the addition of charcoal (Error bars are mean  $\pm$  s.e., n = 9). Figure 3b Right. Mineral-N (nitrogen) content of soil with and without the addition of charcoal. (Error bars are mean  $\pm$  s.e., n = 6).

The addition of charcoal led to no significant change in mineral-N content (Figure 3b).

### Physical properties

The soil in the orchard prior to the addition of charcoal was not hydrophobic; after the addition of charcoal this did not change. When testing for hydrophobicity it was noted that pure charcoal was slightly water repellent.

## Conclusion

Most of the measured properties of the orchard soil were not significantly affected by the addition of charcoal during the five months of this trial. However, the difference in HWC content could be an early indicator of changes that may become more significant over a longer period of time. Comparing the N-

mineralisation rates of the two treatments would further indicate if changes are occurring. To properly assess what effect the addition of charcoal has on a soil's biophysical functions further trials and analysis are required.

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