



Changes in the fungal-to-bacterial respiratory ratio and microbial biomass in agriculturally managed soils under free-air CO₂ enrichment (FACE) – A six-year survey of a field study

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ABSTRACT

In soil ecology, microbial parameters have been identified as sensitive indicators of changes in the soil environment. The Braunschweig FACE project provided the opportunity to study the effects of elevated CO₂ (550 μmol mol⁻¹) as compared to ambient CO₂ (370 μmol mol⁻¹) on total microbial biomass (C_{mic}), C_{mic}-to-C_{org} ratio and the fungal-to-bacterial respiratory ratio together with total C_{org}, N_t, C:N ratio and pH over a six-year period. Field management followed a typical crop rotation system of this region with either a crop-related full nitrogen supply (N100) or 50% reduced N supply (N50). The soil microbial parameters responded to the elevated CO₂ treatment in varying intensities and time spans. The fungal-to-bacterial respiratory ratio was the most sensitive parameter in responding to an elevated CO₂ treatment with highly significant differences to ambient CO₂-treated control plots in the third year of CO₂ fumigation. After six years bacterial respiratory activity had increased in ascending order to 34% in FACE-treated plots (N50 and N100) as compared to control plots. Soil microbial biomass (C_{mic}) responded more slowly to the FACE treatment with highly significant increases of >12% after the fourth year of CO₂ fumigation. The C_{mic}-to-C_{org} ratio responded very late in the last two years of the CO₂ treatment with a significant increase of >7.0% only in the N100 variant. Total C_{org} and N_t were slightly but significantly increased under FACE around 10.0% with ascending tendency over time starting with the second year of CO₂ treatment. No significant FACE effects could be recorded for the C:N ratio or pH.

These results suggest that under FACE treatment changes in the soil microbial community will occur. In our study the fungal-to-bacterial respiratory ratio was superior to total C_{mic} as microbial bioindicators in reflecting changes in the soil organic matter composition.

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1. Introduction

The threat of global climate change, particularly the rising CO₂ concentrations in the atmosphere and the possible impacts of these changes on the biosphere initiated a boom in ecosystem research during the last three decades. The majority of former studies had primarily focused on impacts of CO₂ elevation on plant systems (the primary producer) in natural or managed soil ecosystems. Since the turnover of soil organic matter (SOM) and nutrient cycling is linked to the soil microbial decomposer community, changes in organic

litter from the primary producer, both as a change in quality or amount, would directly affect the microbial decomposers and research here has gained importance over time.

The bulk of literature with relevance to the microbial decomposer community report that elevated CO₂ stimulates photosynthesis and plant growth above and below-ground in most types of plant systems (e.g. Norby, 1994; Canadell et al., 1996; Rogers et al., 1997; Sadowsky and Schortemeyer, 1997; Allen et al., 2000; Körner, 2000) with concomitant increases in rhizodeposition (e.g. Paterson et al., 1997; Sadowsky and Schortemeyer, 1997; Cheng, 1999; Pendall et al., 2004). This input of labile carbon (C) into the soil (Torbert et al., 1997; Hoosbeek et al., 2004; Jastrow et al., 2005; Martens et al., 2009) increased microbial biomass as long as other soil nutrients were not limited (Hu et al., 1999; Johnson et al., 2004; Lagomarsino et al., 2007). On the other hand, the C:N ratio of the plant litter reaching the soil can increase (Cotrufo et al., 1998;

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Körner, 2000; Norby et al., 2001a; Johnson et al., 2004; Pendall et al., 2004; Weigel et al., 2005) leading to nitrogen (N) limitations for microbes with consequences of altered decomposition rates of SOM (Ball, 1997; Cotrufo et al., 1998; Hoosbeek et al., 2004; Peralta and Wander, 2008). Here, variable results with respect to litter quality and decomposition rates have been observed, ranging from no effects on litter quality and decomposition rates (Hoorens et al., 2003; De Graaff et al., 2006) to accelerated decomposition rates (Sowerby et al., 2000, 2005; Heath et al., 2005; Bazot et al., 2006; Taneva and Gonzales-Meler, 2008) or decreased decomposition rates (Gorissen, 1996; Frederiksen et al., 2001; Marhan et al., 2008; Van Groenigen et al., 2005). These inconsistent results must reflect different responses of the microbial decomposer community to a changing nutrient and energy supply underground. In short-term or long-term experiments no change in the fungal-to-bacterial ratio (PLFA profiles) or bacterial diversity was reported (Bruce et al., 2000; Ebersberger et al., 2004) but rather an increase in bacterial metabolic activity (Grayston et al., 1998; Hodge et al., 1998). There was a lack of long-term experiments such as the propagated free-air CO₂ enrichment (FACE) approach (Allen, 1992; Hendrey, 1992; Norby et al., 2001b) for the *in situ* study of soil systems to better understand elevated CO₂ impacts on the microbial community structure and its metabolic activity.

The FACE experiment in Braunschweig, northern Germany, set up in 1999, attempted to follow the effects of an elevated atmospheric CO₂ concentration in fields with crop rotations (Weigel and Dämmgen, 2000; Weigel et al., 2006) under local traditional crop and soil management. At that time, there was a gap in FACE field studies for the maritime temperate climatic zone. The bulk of the acquired knowledge attained so far was derived from case studies of grassland and forests of semiarid, arid or continental temperate climatic zones (Nösberger et al., 2006). While the Braunschweig experiment concentrated primarily on CO₂ effects on plant growth and plant-ecophysiological parameters (Weigel et al., 2005, 2008), it provided the opportunity to follow soil microbial parameters (respiratory fungal-to-bacterial ratio, microbial biomass carbon (C_{mic}) and the microbial biomass carbon to total organic carbon (C_{org}) (the C_{mic}-to-C_{org} ratio)) which we had tested in the past as soil indicators to assess soil quality (e.g. Blagodatskaya and Anderson, 1998; Insam, 2001; Anderson, 2003). Particularly, the fungal-to-bacterial respiratory ratio was very quick in responding to environmental impacts (Blagodatskaya and Anderson, 1998, 1999). With the exception of the fungal-to-bacterial respiratory ratio, they also have been applied in CO₂ enrichment studies in field and forest trials (Islam et al., 2000; Moscatelli et al., 2005). Considering the knowledge drawn from the citations above, we tested the following hypotheses:

- The fungal-to-bacterial respiratory ratio should change in favor of bacteria should rhizodeposition increase under FACE, because these organisms respond more quickly to easily available C sources since they have a somewhat shorter turnover time than fungi (Rousk and Bååth, 2007).
- Total microbial biomass must increase should the total plant residue increase under FACE without concurrent nutrient limitation, while under nutrient limitation it should decrease. The parameter microbial biomass carbon (C_{mic}) to total organic carbon (C_{org}) (the C_{mic}-to-C_{org} ratio) should as well reflect the latter but should increase if soil carbon becomes more easily available to the microbial community (Anderson and Domsch, 1989).

The underlying open question is: how quickly will the microbial pool respond to a change in organic matter quality, thereby acting as an indicator of the fate of SOM.

The present paper reports on the response of these soil microbial parameters to a six-year free-air CO₂ enrichment period in an agriculturally managed crop rotation system together with the monitoring of total soil C_{org}, N_t, pH and C:N ratio.

2. Materials and methods

2.1. Experimental site and soil properties

In autumn 1999, a 22 ha field experiment was established, equipped with the FACE technology developed by the Brookhaven National Laboratory (New York, USA) (see Hendrey, 1992) on agricultural land with a long history of crop rotation and liquid manuring. The experimental site is located at the former Agricultural Research Centre (FAL) (now Johann Heinrich von Thünen Institute (vTI)) in Braunschweig, Lower Saxony, Germany (geographical coordinates: 52°18' North, 10°26' East). The location has an average annual temperature of 8.8 °C, a mean July temperature of 17 °C and the annual total precipitation amounts to 618 mm. The soil is a sandy loam (dystric Cambisol) which developed from a diluvial underground during the ice age epoch ("Bodenkundliche Detailkartierung der FAL" by Sauerbeck, G., 2005, not published) with a soil texture of 69% sand, 24% silt and 7% clay, and has a mean pH of 6.5. Total soil organic carbon (C_{org}) of the site is in the range of 0.9–1.3%. The soil is of low to intermediate fertility.

The field experiment was managed according to local practices with respect to crop cultivation and fertilization including liquid manure application. However, from July 2000, fertilization with liquid manure was canceled and changed to a mineral liquid fertilizer application of AHL (an ammonium nitrate–urea solution) because of interference with proper soil sampling and its impact on microbial and soil parameters.

The present experiment was applied to a crop rotation system which is typical for this location, with three crops starting with winter barley (*Hordeum vulgare*) in September 1999–June 2000, followed by a rye grass mixture (*Lolium*) as cover crop from July 2000 to October 2000, then sugar beet (*Beta vulgaris*) from April 2001 to September 2001, and finally winter wheat (*Triticum aestivum*) from November 2001 to July 2002. The rotation cycle was repeated once and the CO₂ experiment terminated in July 2005 (six years in total). More detailed information can be found by Weigel et al. (2005) and Manderscheid et al. (2009). Table 1 depicts information on crops, sowing and harvesting dates, mean temperature and water supply during the growing season, from the start to the end of the experiment.

2.2. Experimental design

The FACE system consisted of six circular plots (rings), each with a \varnothing of 20 m (for a detailed description of the system see Hendrey, 1992; Weigel and Dämmgen, 2000). Treatments were as follows: 2 rings were equipped with blowers and purged with CO₂ (FACE) of a concentration of 550 $\mu\text{mol mol}^{-1}$, 2 rings were operated with blowers and ambient air (370 $\mu\text{mol mol}^{-1}$ CO₂, control) and 2 rings were without blowers (control). Exposure to enriched CO₂ occurred only at daylight hours and during the growing season of the crops (Weigel et al., 2005). At high wind velocity of >6.0–6.5 m s⁻¹ and air temperatures below <5 °C CO₂ fumigation was stopped.

A nitrogen (N) fertilization variant was included into the experiment in order to test interactive effects of elevated CO₂ and supply of N to the crops. Nitrogen application was restricted to 50% (N50) of adequate N (N100) in half of each of the six rings, resulting in a CO₂ × N split plot design with 2 replicates for each treatment (Weigel et al., 2005, 2006) by dividing each ring in 4 quadrants (with an area of 78.5 m² each), designated A + B and C + D.

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