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Soil microbial indices as bioindicators of environmental changes in a poplar plantation

M.C. Moscatelli [a,*], A. Lagomarsino [b], S. Marinari [a], P. De Angelis [b], S. Grego [a]

[a] *Dip. di Agrobiologia e Agrochimica, Università della Tuscia, Viterbo, Italy*
[b] *Dip. di Scienze dell'Ambiente Forestale e delle sue Risorse, Università della Tuscia, Viterbo, Italy*

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Abstract

An understanding of microbial biomass and microbial activity as part of belowground processes as affected by elevated CO_2 is crucial in order to predict the long-term response of ecosystems to climatic changes. The ratio of biomass C to soil organic C (Cmic:Corg), the metabolic quotient (the specific soil respiration of the microbial biomass, qCO_2), the C mineralization quotient (the fraction of total organic C mineralized throughout the incubation, qM), the microbial biomass change rate quotient (qC) and soil inorganic nitrogen content were determined on soil samples taken during 3 years (Fall 2000–Fall 2003) in a poplar plantation exposed to increased atmospheric CO_2 by means of FACE (Free Air CO_2 Enrichment) technique and nitrogen fertilization. A competition for nitrogen between plants and microrganisms, stronger in FACE plots, induced a stress condition within microbial community. FACE treatment provided C for microbial growth (Cmic:Corg), but reducing nitrogen availability, led to a higher microbial loss over time (qC). Nitrogen fertilization decreased microbial mortality lowering energetic maintenance requirements (qCO_2) and induced a short-term shift in favour of microrganisms more rapid in the use of the resources. The C mineralization quotient (qM) was not affected by either FACE nor fertilization treatment meaning that the fraction of total organic carbon mineralized during the incubation period did not vary significantly.

Keywords: Soil; Elevated CO_2; N fertilization; Microbial biomass; Soil respiration; Indices; Poplar

1. Introduction

Elevated atmospheric CO_2 may affect the microbe–soil–plant root system indirectly by modifying soil water content and by increasing root growth and rhizodepositions rates (Hungate et al., 1997; Janssens et al., 1998). Therefore changes in microbial population, community structure and activity of soil- and rhizosphere-associated microrganisms are likely to occur under elevated CO_2 (Sadowsky and Schortemeyer, 1997).

Microrganisms in fact are the driving force of nutrient supply in soils and are the primary recipients of increased photoassimilates from plants growing in elevated atmospheric CO_2. Moreover long-term effects of elevated CO_2 on ecosystem carbon (C) sequestration

* Corresponding author. Tel.: +39 0761 357329; fax: +39 0761 357242.
E-mail address: mcm@unitus.it (M.C. Moscatelli).

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are highly dependent on the factors affecting C sequestration in mineral soils and the interactions of C with other nutrients (Cardon, 1996). Depending on soil C/N ratio, the interactions of C and nitrogen (N) are particularly important being N the nutrient most commonly limiting plant and microbial growth and soluble C the main energy source for microrganisms.

Terrestrial ecosystems response to CO_2 fertilization is therefore linked to the knowledge of belowground processes and particularly those performed by the microbial pool (Zak et al., 2000). Microbiological parameters related to soil weight are often correlated or combined as an index in order to evaluate the significance of microbial populations and microbial activity in the cycling of elements in soils of different ecosystems in situ (Nannipieri, 1994). Brookes (1995) recommends to combine microbial parameters in order to have an ''internal control'' such as biomass C as the percentage of soil organic matter. The same author also reports that combining microbial activity and population measurements (biomass specific respiration or metabolic quotient) appears to provide more sensitive indications of soil pollution than either activity or population measurements alone (see also Dilly and Munch, 1998). Ecophysiological indices (metabolic quotients) are generated by basing physiological performances (respiration, growth/death, carbon uptake) on the total microbial biomass per unit time. Any environmental impact which will affect members of a microbial community should be detectable at the community level by a change of a particular total microbial community activity which can be quantified (qCO_2, etc.) (Anderson, 2003).

The ratio of biomass C to soil organic C (Cmic:Corg) reflects the contribution of microbial biomass to soil organic carbon (Anderson and Domsch, 1989). It also indicates the substrate availability to the soil microflora or, in reverse, the fraction of recalcitrant organic matter in the soil; in fact this ratio declines as the concentration of available organic matter decreases (Brookes, 1995). The qCO_2 (the community respiration per biomass unit or the metabolic quotient) has been widely used in literature and is originally based on Odum's theory of ecosystem succession. Although its reliability as a bioindicator of disturbance or ecosystem development has been recently criticised by some authors, it is recognized to have valuable application as a relative measure of how efficiently the soil microbial biomass is utilizing C resources and the degree of substrate limitation for soil microbes (Wardle and Ghani, 1995; Dilly and Munch, 1998). The qM (mineralization quotient) expresses the fraction of total organic carbon mineralized throughout the incubation time (Dommergues, 1960; Pinzari et al., 1999). The qC (microbial biomass change rate quotient) expresses the daily enrichment or loss of soil microbial C and is calculated based on qD as reported by Anderson and Domsch (1990). In the present study Cmic:Corg, qCO_2, qM, qC and inorganic nitrogen content were determined on soil samples taken during 3 years (Fall 2000–Fall 2003) in a poplar plantation exposed to increased atmospheric CO_2 by means of FACE (Free Air CO_2 Enrichment) technique and fertilized during the last 2 years. Aim of this paper was to assess the validity of the microbial indices as bioindicators of microbial processes induced by the two treatments: FACE and N fertilization.

2. Materials and methods

2.1. Site description

POPFACE experimental plantation and FACE facility are located in central Italy, Tuscania (VT) (42°22′N, 11°48′E, alt 150 m). The soil is loam/silt-loam, total C range is 0.65–1.18%, total N range is 0.11–0.14%. For further information on soil physical and chemical properties, see Hoosbeck et al. (2004). The mean values of precipitation and temperature (calculated over a period of 14 years, from meteorological data collected at POPFACE site) are of 14.1 °C and 818 mm, respectively. Clones of *Populus alba*, *Populus nigra* and *Populus* × *euramericana* were grown, since 1999, in six 314 m^2 plots treated either with atmospheric (control) or enriched (550 μmol mol^{-1} CO_2) CO_2 concentration with FACE technology (Free Air CO_2 Enrichment). Each plot is divided into six triangular sectors, with two sectors per poplar genotype: three species × two nitrogen levels. Nitrogen fertilization started in July 2002, it was executed once per week during the growing season and lasted for 16 weeks. Fertilizer was supplied weekly in constant dose to a final total amount of 212 kg N ha^{-1}. In the 2003 growing season the fertilizer was supplied weekly in amounts proportional to the growth rate for 20 weeks and provided a total amount of 290 kg N ha^{-1}.

2.2. Soil sampling

After removal of litter layer two soil cores per genotype (10 cm wide, 20 cm long) were taken inside each of the three sectors in each plot, for a total of 36 soil cores in not fertilized sub-plots from October 2000 until October 2001 and 72 soil cores from June 2002 to October 2003 in fertilized and not fertilized sub-plots. In June 2002 soil samples were collected also in fertilized sub-plots although the addition of nitrogen started the following month, however data related to these samples are not considered in the calculation of the fertilization effect. Soil samples were immediately sieved (<2 mm) and the moisture content adjusted to 60% of their water holding capacity (WHC). The soil samples were then left to equilibrate at room temperature in the dark for 1 day prior to biochemical analyses.

2.3. Chemical and microbiological analyses

Inorganic nitrogen was assessed as the sum of ammonium and nitrate: ammonium was extracted in 1 M KCl and was determined following Anderson and Ingram (1993) while nitrate was determined colorimetrically after extraction in 0.5 M K_2SO_4 (Cataldo et al., 1975). Microbial biomass carbon (MBC) was estimated following the Fumigation Extraction (FE) method: two portions of moist soil (20 g oven-dry soil) were weighed, the first one (non-fumigated) was immediately extracted with 80 ml of 0.5 M K_2SO_4 for 30 min by oscillating shaking at 200 rpm and filtered (Whatman no. 42); the second one was fumigated for 24 h at 25 °C with ethanol-free $CHCl_3$ and then extracted as described above. Organic C in the extracts was determined after oxidation with 0.4 N $K_2Cr_2O_7$ at 100 °C for 30 min (Vance et al., 1987). Microbial biomass was calculated as follows: biomass C = E_C:k_{EC}, where E_C is the difference between organic C extracted from fumigated soils and organic C extracted from non-fumigated soils and $k_{EC} = 0.38$. POPFACE soil characteristics allow the use of this factor since caution is required in soils recently amended with organic matter (Harden et al., 1993a), in waterlogged soils (Inubushi et al., 1991) and in organic layers of forest soils (Scholle et al., 1992). For measuring microbial respiration 20 g (oven-dry basis) of moist sample were placed in 1 l stoppered glass jars. The CO_2 evolved was trapped, after 24, 72, 168, 240 h of incubation, in 2 ml 1 M NaOH and determined by titration of the excess NaOH with 0.1 M HCl (Badalucco et al., 1992). The CO_2 evolved during the 10th day of incubation was used as the basal respiration value because, after that period, the soil reached a relatively constant hourly CO_2 production rate. Total organic carbon (TOC) was estimated following the method reported by Springer and Klee (1954). Microbial indices were calculated as follows:

Cmic:Corg = μg of biomass C μg total organic carbon^{-1} (Anderson and Domsch, 1989);
qCO_2 = (μg C-$CO_{2\ basal}$ h^{-1} × μg biomass C^{-1}) × 10^3 (Dilly and Munch, 1998);
qM = μg C-$CO_{2\ cumulative}$ μg total organic carbon^{-1} (Pinzari et al., 1999);
$qC = ((\mu g\ Cmic_{t_1} - \mu g\ Cmic_{t_2})/\mu g\ Cmic_{t_1}/(t_2 - t_1))$.

qC is calculated as reported by Anderson and Domsch (1990) for qD. Positive and negative values indicate a daily loss and an enrichment, respectively, of microbial biomass carbon in the ecosystem.

These indices can be considered as potential indicators of soil biological properties and processes since they have been obtained analyzing soils through laboratory standard procedures (sieving, controlled temperature and moisture) that do not necessarily reflect in situ conditions.

2.4. Statistical analysis

Analysis of variance (ANOVA) was performed to evaluate the main effects of FACE, fertilization, time and their interaction on parameters analyzed. Data were tested for normality with the Shapiro-Wilk statistic and normalized with a square root transformation. qC was linearly transformed. A randomized block design was applied using the general linear model procedure with CO_2, N, time and blocks as factors. The two replicates for each plot were averaged and the plot (three control plots and three FACE plots) was the unit of replication. The significance of FACE, time and the interaction between the two factors was determined in not fertilized plots (years 2000–2003, $n = 36$). The significance of fertilization and its interaction with FACE and time was determined in

Table 1
Inorganic N, Cmic:Corg (microbial quotient), qCO_2 (metabolic quotient), qM (C mineralization quotient) and MR24 h (microbial respiration after 24 h) measured in control, control + N, FACE, FACE + N plots from Fall 2000 to Fall 2003

	October 2000	June 2001	October 2001	June 02	October 2002	June 2003	October 03
Inorganic N (μg N-NH_4 + N-NO_3) g^{-1}							
Control	41.8 (4.1)	37.0 (1.2)	29.0 (3.2)	9.5 (0.7)	5.6 (0.4)	7.1 (0.6)	6.6 (0.7)
Control + N				13.2 (0.8)	26.9 (3.4)	11.4 (1.2)	12.8 (1.3)
FACE	42.1 (3.0)	35.6 (1.9)	9.9 (1.2)	6.9 (0.4)	3.9 (0.3)	3.8 (0.3)	5.4 (0.6)
FACE + N				9.0 (0.5)	13.4 (1.6)	14.7 (1.5)	14.8 (1.2)
Cmic:Corg (μg C biomass μg total organic C^{-1})							
Control	6.8 (0.5)	3.1 (0.4)	1.5 (0.1)	1.05 (0.2)	1.27 (0.1)	2.19 (0.2)	1.03 (0.1)
Control + N				1.01 (0.1)	1.38 (0.1)	1.79 (0.2)	1.58 (0.1)
FACE	9.1 (0.8)	5.1 (0.8)	2.2 (0.3)	1.65 (0.2)	1.43 (0.2)	2.16 (0.3)	1.32 (0.2)
FACE + N				1.76 (0.1)	1.32 (0.1)	2.34 (0.2)	1.83 (0.1)
qCO_2 (μg C-CO_2 h^{-1} μg C biomass^{-1}) × 10^3							
Control	0.78 (0.2)	3.36 (0.4)	2.46 (0.6)	5.44 (0.7)	2.17 (0.3)	3.11 (0.3)	3.86 (0.5)
Control + N				6.08 (1.1)	2.95 (0.6)	2.04 (0.4)	1.86 (0.2)
FACE	0.26 (0.1)	3.06 (0.6)	2.60 (0.5)	2.34 (0.5)	2.36 (0.3)	3.88 (0.4)	2.90 (0.4)
FACE + N				2.44 (0.3)	2.57 (0.5)	1.27 (0.2)	1.43 (0.2)
qM (μg C-$CO_{2\ cumulative}$ μg total organic C^{-1})							
Control	1.54 (0.12)	0.87 (0.05)	0.99 (0.08)	1.05 (0.09)	0.91 (0.06)	1.41 (0.11)	0.85 (0.07)
Control + N				0.95 (0.05)	0.73 (0.07)	1.42 (0.09)	0.98 (0.07)
FACE	1.32 (0.12)	1.10 (0.1)	1.29 (0.13)	1.11 (0.11)	0.90 (0.10)	1.69 (0.12)	0.85 (0.06)
FACE + N				0.98 (0.07)	0.86 (0.15)	1.56 (0.12)	0.95 (0.06)
MR24 h (μg C-CO_2 g^{-1} 24 h^{-1})							
Control	19.9 (3.0)	21.4 (2.1)	23.3 (1.6)	18.6 (2.3)	13.2 (1.7)	29.6 (2.3)	14.2 (1.1)
Control + N				16.5 (1.8)	22.8 (1.7)	74.1 (6.1)	27.7 (2.2)
FACE	16.7 (3.9)	28.7 (1.6)	25.4 (1.0)	17.0 (2.5)	14.5 (1.9)	26.2 (1.9)	14.7 (2.3)
FACE + N				19.5 (2.2)	24.4 (1.4)	59.9 (2.6)	28.1 (2.2)

Standard error is reported in parentheses.

fertilized and not fertilized plots from 2002 to 2003 ($n = 72$). Because there were no significant variations due to the different poplar species, data from different poplar genotypes were pooled together. When interactions were not significant they were excluded from analysis. In the results section the effect of FACE and/or fertilization treatments has been reported as percentage variation with respect to the control. It has been calculated on the average values of all sampling dates for FACE effect and from October 2002 for the fertilization effect: June 2002 is, in fact, not included since fertilization was started the following month. All statistical analysis were performed with the Systat 11.0 statistical software package (SPSS Inc.).

3. Results

A strong reduction of soil inorganic nitrogen was observed, in not fertilized plots (FACE and control), as from the year 2000; the depletion of inorganic nitrogen was about 85% after 3 years (Table 1). Moreover, FACE treatment reduced inorganic nitrogen availability, during the whole period of study, with respect to control plots (−20%, $p < 0.001$) (Fig. 1A). The fertilization produced a significant increase of soil inorganic nitrogen (+123% in FACE and +160% in control plots, $p < 0.001$) although it did not re-establish the original values of October 2000 (Fig. 1A and Table 1).

The trend of microbial quotient (Cmic:Corg ratio), during the 3 years of observation, parallels the trend of inorganic nitrogen, as also shown by the linear regression on these two parameters in Fig. 2. Cmic:Corg significantly decreases after the first year and assesses its value to less than 2% until the end of 2003 (Table 1). However, although the contribution of microbial biomass to total organic carbon is very low in this soil, FACE treatment induced a significant increase of Cmic:Corg ratio in not fertilized plots (+35%, $p < 0.001$) (Fig. 1B).

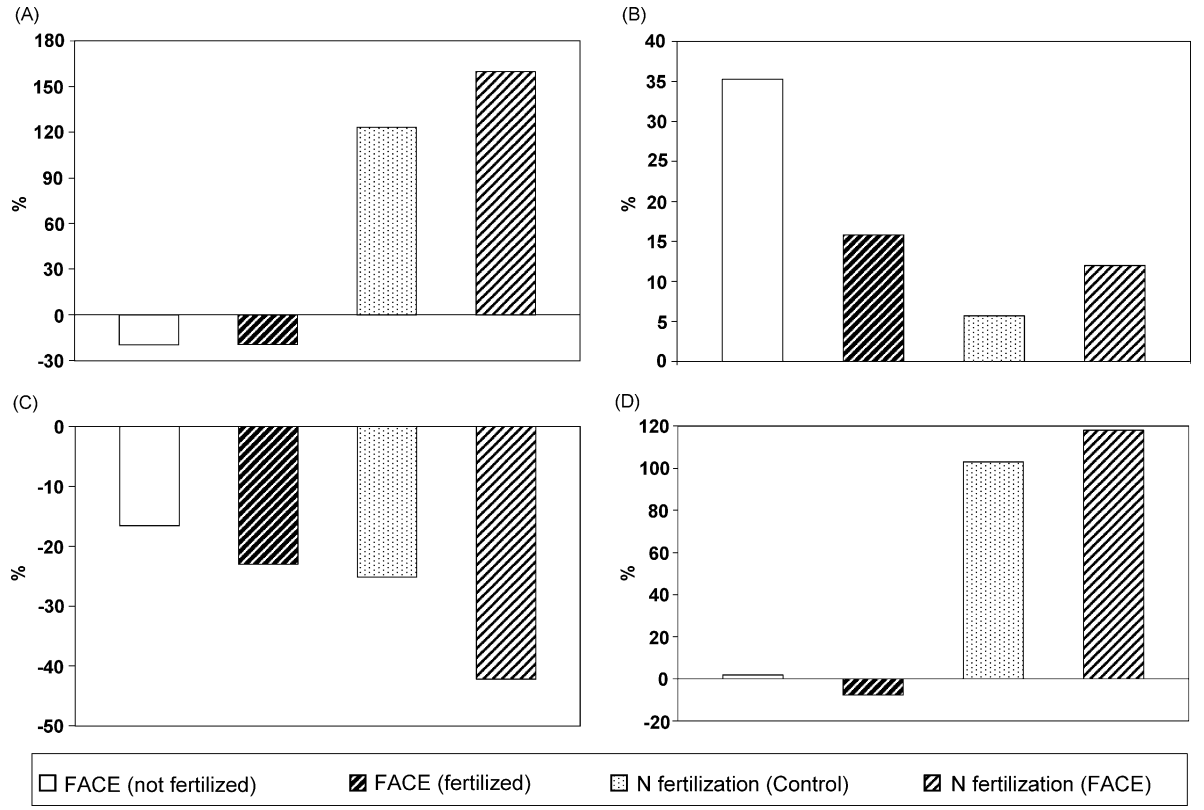

Fig. 1. Mean percentage effects of treatments (FACE and N fertilization) calculated from 2000 to 2003 as relative variation with respect to the control. (A) Inorganic nitrogen ($N\text{-}NH_4 + N\text{-}NO_3$), (B) microbial quotient (Cmic:Corg), (C) metabolic quotient (qCO_2) and (D) microbial respiration (24 h).

Tables 2 and 3 report the qC measured in not fertilized (Fall 2000–Fall 2003) and fertilized plots (Spring 2002–Fall 2003), respectively. The mean qC for FACE plots during the whole period of study was 2.30 versus 0.60 μg biomass C loss μg biomass C^{-1} $day^{-1} \times 10^{-3}$ in control plots. Microbial loss thus increased under elevated CO_2 where the depletion in inorganic nitrogen seems to be the driving variable of microbial physiological status, in fact the addition of nitrogen lowers the qC (−1.08 and −1.77 in FACE + N and control + N, respectively) (Table 3).

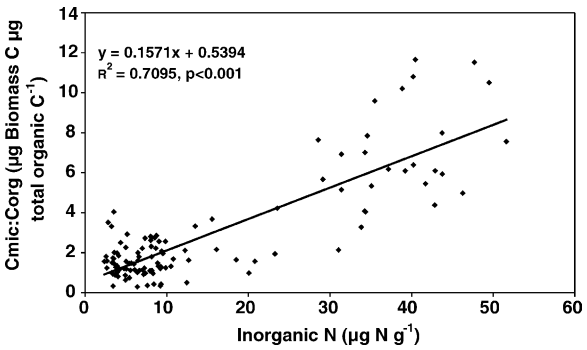

Fig. 2. Linear regression between inorganic nitrogen and Cmic:Corg measured from 2000 to 2003 in all plots ($n = 121$).

The metabolic quotient is negatively and significantly affected by FACE and N fertilization treatments (Tables 1 and 4). Face lowers qCO_2 by 17% in not fertilized and by 23% in fertilized plots while the addition of nitrogen causes a further decrease of this index by 25 and 42% ($p < 0.001$) in control and FACE plots, respectively (Fig. 1C). In fact qCO_2 reaches, at the end of 2003, values of 1.86 and 1.43 for control + N and FACE + N versus 3.86 and 2.90 for control and FACE (Table 1).

An inverse correlation is generally observed between qCO_2 and Cmic:Corg ratio indicating a strict interdependency between microbial growth and

Table 2
Microbial biomass change rate quotient (qC) (($\mu g\ Cmic_{t_1} - \mu g\ Cmic_{t_2}$)/$\mu g\ Cmic_{t_1}/(t_2 - t_1)$) $\times 10^{-3}$ measured in FACE and control plots from Fall 2000 to Fall 2003

Period	Days	FACE	Control
October 2000–March 2001	129	5.50 (±0.29)	5.45 (±0.31)
March 2001–June 2001	82	0.25 (±0.1)	−4.89 (±1.99)
June 2001–August 2001	98	2.13 (±1.13)	1.22 (±0.94)
August 2001–October 2001	53	7.85 (±1.46)	3.90 (±1.06)
October 2001–June 2002	240	0.98 (±0.29)	0.78 (±0.35)
June 2002–October 2002	145	0.47 (±0.64)	−2.75 (±1.60)
October 2002–June 2003	220	−1.36 (±0.55)	−2.20 (±0.75)
June 2003–October 2003	156	2.54 (±0.53)	3.27 (±0.49)
Average		2.29	0.60

Standard error is reported in parentheses.

Table 3
Microbial biomass change rate quotient (qC) (($\mu g\ Cmic_{t_1} - \mu g\ Cmic_{t_2}$)/$\mu g\ Cmic_{t_1}/(t_2 - t_1)$) $\times 10^{-3}$ measured in FACE + N and control + N plots from Spring 2002 to Fall 2003

Period	Days	FACE	Control	FACE + N	Control + N
June 2002–October 2002	145	0.47 (±0.64)	−2.75 (±1.60)	1.69 (±0.50)	−2.72 (±1.22)
October 2002–June 2003	220	−1.36 (±0.55)	−2.20 (±0.75)	−5.70 (±1.24)	−3.82 (±1.18)
June 2003–October 2003	156	2.54 (±0.53)	3.27 (±0.49)	0.78 (±0.73)	1.21 (±0.67)
Average		0.55	−0.56	−1.08	−1.77

Standard error is reported in parentheses.

maintenance. In this study the correlation coefficient between the two indices is $r = -0.371$ ($n = 177$; $p < 0.001$) and indicates that to a low qCO_2 corresponds a high Cmic:Corg ratio.

In the attempt to get further insight into microbial respiration activity, CO_2 output after 24 h of incubation and the cumulative value of CO_2 evolved after 10 days were considered. This was to emphasize the known difference between the flush of CO_2 following rewetting of soil and the basal respiration activity (Wang et al., 2003). CO_2 production after 24 h (MR24 h) is not modified by FACE treatment while the fertilization caused a significant increase in both FACE and control plots: the mean fertilization effect was in fact +118 and +103% ($p < 0.001$), respectively (Table 1 and Fig. 1D).

The C mineralization quotient (qM) provides information on the fraction of total organic carbon

Table 4
Analysis of variance of Cmic:Corg, qC, qCO_2, qM, microbial respiration (24 h) and inorganic nitrogen measured in FACE, control, FACE + N and control + N from Fall 2000 to Fall 2003

	Cmic:Corg	qC	qCO_2	qM	MR24 h	Inorganic N
Time	***	***	***	***	***	***
FACE	**	**	*	ns	ns	***
Fertilization	ns	*	***	ns	***	***
Time × FACE	***	ns	*	ns	ns	*
Time × fert.	ns	*	**	ns	***	*
FACE × fert.	ns	ns	ns	ns	ns	ns
Time × FACE × fert.	ns	ns	ns	ns	ns	**

ns: not significant.
* $p < 0.05$.
** $p < 0.01$.
*** $p < 0.001$.

mineralized throughout the incubation time (10 days in this study) (Dommergues, 1960; Pinzari et al., 1999). qM ranged from 0.849 to 1.686 in FACE plots and from 0.734 to 1.541 $\mu g\ C\text{-}CO_{2\ cumulative}\ TOC^{-1}$ in control plots (Table 1). It was not affected by either FACE nor fertilization treatments.

4. Discussion

In many studies microbiological parameters were correlated or combined as an index (Nannipieri, 1994). Nevertheless ratios between microbiological parameters have often been used for evaluating the microbial ecophysiology implying an interlinkage between cell-physiological functioning under the influence of environmental factors (Anderson, 2003).

In this study the responses of Cmic:Corg ratio, qCO_2 (metabolic quotient) and qC (microbial change rate quotient) to FACE and nitrogen fertilization treatments, observed during 3 years, seemed to be strongly affected by the nutritional *status* of the soil. In fact a strong reduction of soil inorganic nitrogen was detected and it was probably due to enhanced plant uptake linked to the increase of biomass under elevated CO_2 as shown by Calfapietra et al. (2003). The microbial pool is strongly dependent on nitrogen and probably suffered from a competition with plants for this element (Allen and Schlesinger, 2004). This nutritional "stress" could explain the decrease of Cmic:Corg ratio, in not fertilized plots, to values lower than 2.0 which is considered a critical threshold for soils with neutral pH (Anderson, 2003). Moreover, it is reasonable to assume that a nutritional unbalance between C and N may have altered the physiological state of microbes with changes in microbial size over time. The decrease of qC after fertilization suggests an improvement of microbial nutritional conditions as nitrogen in easily available forms was provided.

Anderson (2003) refers to the same critical value, mentioned for Cmic:Corg, also with reference to qCO_2, affirming that values higher than 2.0 of metabolic quotient indicate an energetically less efficient microbial community. Changes in nutrient availability can modify microbial maintenance energy requirements. The low Cmic:Corg and the high qCO_2 reflect a less efficient use of organic substrates by microbial biomass (Anderson, 2003; Pinzari et al., 1999). Nutrients acquisition activity is an energetically expensive process particularly when microbes are forced to degrade stable SOM to get new available substrates. qCO_2 decreases under FACE treatment but this reduction is more pronounced when both treatments (FACE and N fertilization) are applied. In fact, in FACE + N plots, C and N are easily available in soil, therefore a more efficient use of energy in nutrient acquisition activity is permitted.

In elevated CO_2 environments it is assumed that, because of faster root turnover or increased production of root exudates, more C is available for microbes (Cardon, 1996; Cheng, 1999; Schortmeyer et al., 2000). In another study that we performed at POPFACE experimental station, elevated CO_2 induced a significant increase of soil labile carbon fractions (+19% of water soluble carbon and + 21% of K_2SO_4-extractable carbon) indicating a flux of soluble C forms that could lead to the microbial immobilization process observed (Moscatelli et al., in press). We can therefore hypothesize that, in our experimental conditions, the extra C made available for microbes has been used to build up more microbial biomass as the significant increase of microbial quotient under FACE treatment suggests.

The response of microbial respiration to nitrogen fertilization was significant in the first 24 h of incubation, particularly in June 2003 when the highest increase of this parameter was recorded. At this purpose it should be considered that June 2003 was just 1 month after the beginning of the fertilization and this could be the reason for the consistent flush of CO_2 measured. It is well known that a sudden increase of CO_2 output from soil is generally observed after the addition of easily available organic substrates or of inorganic nitrogen fertilizers to the soil. This phenomenon, the so-called priming effect, is due to an increase of microbial activity resulting in an acceleration of soil organic matter mineralization as substrate and energy source (Kuzyakov et al., 2000). The addition of inorganic nitrogen could have provoked, likewise, a short-term selection inside the microbial community in favour of microrganisms more efficient in the use of the nutrient resources. To support this hypothesis we have evidence that microbial biomass C/N ratio decreased significantly in June 2003 after fertilization by 61% in control + N and 48% in FACE + N indicating a shift towards bacterial communities (data not shown).

The qM, or the potential C mineralization activity (measured under controlled conditions of temperature and humidity) as defined by Dommergues (1960), did not show significant changes meaning that neither FACE treatment nor N fertilization did affect the capacity of the soil to store carbon.

In conclusion, as far as the aim of this paper is concerned, microbial indices proved to be sensitive to changes occurred to soil processes under FACE and N fertilization. We hypothesize that a competition for nitrogen between plants and microrganisms occurred, strongly in FACE plots, and that it probably induced a stress condition within microbial community. FACE treatment provided C for microbial growth, but reduced nitrogen availability and increased microbial loss. Nitrogen fertilization, conversely, promoted soil microbial biomass enrichment, lowering energetic maintenance requirements. Although we need further investigation on microbial C mineralization kinetics, particularly during a longer incubation experiment, a not consistent change on carbon sequestration soil capacity has been observed.

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