

More new carbon in the mineral soil of a poplar plantation under Free Air Carbon Enrichment (POPFACE): Cause of increased priming effect?

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[1] In order to establish suitability of forest ecosystems for long-term storage of C, it is necessary to characterize the effects of predicted increased atmospheric CO₂ levels on the pools and fluxes of C within these systems. Since most C held in terrestrial ecosystems is in the soil, we assessed the influence of Free Air Carbon Enrichment (FACE) treatment on the total soil C content (C_{total}) and incorporation of litter derived C (C_{new}) into soil organic matter (SOM) in a fast growing poplar plantation. C_{new} was estimated by the C3/C4 stable isotope method. C_{total} contents increased under control and FACE respectively by 12 and 3%, i.e., 484 and 107 gC/m², while 704 and 926 gC/m² of new carbon was sequestered under control and FACE during the experiment. We conclude that FACE suppressed the increase of C_{total} and simultaneously increased C_{new}. We hypothesize that these opposite effects may be caused by a priming effect of the newly incorporated litter, where priming effect is defined as the stimulation of SOM decomposition caused by the addition of labile substrates. *INDEX TERMS*: 0315 Atmospheric

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1. Introduction

[2] Will rising atmospheric CO₂ concentrations increase the productivity of plants and ecosystems, and will increased productivity translate into increased carbon storage [Norby *et al.*, 2002]? Elevated CO₂ does initially increase net primary production (NPP) of certain plant communities [e.g., Gielen and Ceulemans, 2001]. However, the critical uncertainty in view of C sequestration is whether this increase in NPP will result in an increase of plant biomass, increased build up of litter layer, and/or an increase of

organic carbon content of the mineral soil. On a global scale, the vegetation and the combined litter and soil pools were estimated to contain 610 and 1580 Gt C, respectively [Schimel, 1995]. With estimated global NPP and respiration rates of 61.4 and 60 Gt C yr⁻¹, respectively, the turnover time of the vegetation pool is about a decade. Turnover times of forest litter layers range from less than a year to several decades, while organic matter fractions in mineral soils may have turnover times of less than a year to thousands of years depending on climatic, edaphic, and biotic factors [Schimel *et al.*, 1994].

[3] At a timescale of several decades, increased C storage in woody biomass of forests is relevant to policy decisions [Norby *et al.*, 2002; Schulze *et al.*, 2000]. However, the possible increase of long-term C storage in mineral soils is relevant at larger timescales as a mechanism to mitigate increasing atmospheric CO₂ concentrations [Houghton *et al.*, 1998; Schimel, 1995].

[4] Several Free Air CO₂ Enrichment (FACE) experiments were established in forest ecosystems by different research groups to test hypotheses involving increased C storage in forest biomass, litter, and soils under increased atmospheric CO₂ concentrations [Hendrey *et al.*, 1999]. In general, FACE treatment effects on soil C content are hard

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to detect because the soil C pool is usually very large compared to the annual C input, and the soil C turnover time is long compared to the duration of most experiments [Hungate *et al.*, 1996]. Furthermore, the usually high spatial variability in soil C content tends to blur effects of C sequestration. One way to improve the sensitivity of detecting FACE effects on soil C input is based on the use of $\delta^{13}\text{C}$ analyses. For instance, at the Duke and Swiss grass-FACE experiments, the CO_2 added to the elevated CO_2 plots is derived from fossil fuel and is thus depleted in ^{13}C ($\delta^{13}\text{C}$ respectively -43% and -48% versus the Pee Dee Belemnite standard) compared to ambient CO_2 ($\delta^{13}\text{C} -8\%$) [Schlesinger and Lichter, 2001; Van Kessel *et al.*, 2000b]. Mixing of ^{13}C -depleted and ambient CO_2 during application and photosynthetic fractionation determines the isotopic signal of the plant remains. Incorporation of this signal into the soil provides insight into soil C input dynamics. Unfortunately, this method can only be used in the elevated CO_2 plots.

[5] To quantify C sequestration due to an increase of atmospheric CO_2 concentration, the difference in the amount of newly sequestered C under ambient and elevated CO_2 concentrations must be determined. The C3/C4 stable isotope method as outlined by Balesdent *et al.* [1988] has been applied for this purpose in several FACE studies [Leavitt *et al.*, 2001; Van Kessel *et al.*, 2000a]. The abundance of ^{13}C relative to ^{12}C is less in plant tissue than in atmospheric CO_2 due to isotope discrimination, where plants using the C4 photosynthetic pathway discriminate less than C3 plants do. The mean ^{13}C composition of soil C reflects the ^{13}C signal of the long-term vegetative inputs. A shift in the ^{13}C content of plant input followed by changes in soil ^{13}C content can be used to estimate new soil C input. Alternatively, soil under an existing vegetation can be replaced by soil with a different $\delta^{13}\text{C}$ signature [Ineson *et al.*, 1996; Van Kessel *et al.*, 2000a]. This method provides for the ambient versus elevated CO_2 comparison and was used in the POPFACE experiment. In this paper we report on the effects of FACE treatment and poplar species on the total soil carbon pool and on the annually newly sequestered soil carbon as estimated by the use of stable C isotopes.

2. Methods

[6] Early in 1999 the POPFACE experiment was established on former agricultural fields near Viterbo ($42^\circ37'04''\text{N}$, $11^\circ80'87''\text{E}$, alt 150 m), Italy. Nine hectare were planted with *Populus euramericana* hardwood cuttings at a spacing of $2 \times 1 \text{ m}^2$. Three FACE (plots 1, 4, and 5) and three controls (plots 2, 3, and 6) of 22 m in diameter were randomly assigned under the condition of minimum CO_2 enrichment pollution. The $\sim 350 \text{ m}^2$ circular plots were divided into six sectors, where two sectors each were planted with the following species at a spacing of $1 \times 1 \text{ m}^2$: *P. alba*, *P. nigra*, and *P. euramericana*. Carbon enrichment was achieved by injection of pure CO_2 through laser-drilled holes in tubing mounted on telescopic poles [Miglietta *et al.*, 2001]. The average CO_2 concentration in the FACE plots was 544 ± 48 , 532 ± 83 , and $554 \pm 95 \mu\text{mol mol}^{-1}$ during the first, second, and third growing seasons (early

April until early November). The plantation was drip irrigated at a rate of 6 to 10 mm per day during the growing seasons.

[7] After the fields were ploughed during the fall of 1998 and before the establishment of the plantation, a soil survey took place and soil samples were collected. Soil profile descriptions were made according to the USDA system at the end of 1998 and 2002 [Munsell, 1994; Soil Survey Division Staff, 1993; Soil Survey Staff, 1994]. Soil pH was measured in a KCl extract ($n = 4$ per plot) [Van Lagen, 1996]. The POPFACE plantation and adjacent fields had been under forest until about 1950. Since then a variety of agricultural C3 crops have been grown on these former forest soils until the inception of the POPFACE plantation. The annual precipitation is on average 700 mm with dry summers (Xeric moisture regime). The soils were formed in consolidated volcanic ash of about 600,000 years old. During the last glacial period the soils were rejuvenated, or covered, by blown-in loess. The POPFACE plots are located in an undulating landscape (slope gradient 1–6%). The presented profile descriptions and data are based on the survey of 1998 with the exception of the O horizons (Table 1). The descriptions of the mineral horizons did not change between the end of 1998 and 2002. However, the O horizons were absent in 1998 and were formed under the plantation during that period.

[8] After the initial survey and sampling, soil samples were collected from each sector within the three control and three FACE plots in November of 1999, 2000, and 2001. Bulk density samples were taken with metal rings, with a diameter and height of 10 cm, at 0–10, 10–20, and 20–30 cm below the surface of the mineral soil. The samples were dried at 105°C for 3 days. Bulk densities were calculated based on dry weight and ring volume. Next, the soil samples were crushed by hand, and live roots were removed. Carbon and nitrogen were determined by flash combustion in an elemental analyzer (EA 1108) [Van Lagen, 1996]. The soil C, N, and bulk density data were used to calculate the C and N contents per 10 cm depth increment and 1 m^2 surface, i.e., gram C and N per 0.1 m^3 . To be able to detect FACE and species effects, the spatial variability of the initial soil C and N contents was taken into account (Table 2). Insufficient degrees of freedom prohibited the use of plot effect as a factor in the statistical analyses. Instead, the differences between the initial C and N contents and the C and N contents measured at the end of 2000 and 2001 were calculated.

[9] The CO_2 gas used for fumigation at the POPFACE site had a $\delta^{13}\text{C}$ of -6% versus the PDB standard, which is close to the ambient value of about -8% . Therefore, after mixing of fumigated and ambient CO_2 in the FACE plots, the same $\delta^{13}\text{C}$ of CO_2 was assumed in control and in FACE plots.

[10] The C4 soil used to fill the ingrowth cores was taken from Udine (north eastern Italy) and had been under continuous corn for at least 45 years. This soil had a loam texture and C and N percentages of respectively 0.90 ± 0.01 and 0.10 ± 0.001 . The root ingrowth cores had a diameter of 4 cm and length of 40 cm and were covered with a 2-mm mesh that allowed roots to grow in [Lukac *et al.*, 2003]. Care was taken to compact the soil to the original bulk density of the

Table 1. Soil Profile Descriptions^a

Plot	Soil	Horizon	Depth, cm	Description
1 and 2	Pachic Xerumbrept	Oi (L)	2–1	poplar leaf and small branches litter; very loose and low density
		Oe (F)	1–0	fragmented and partly decomposed litter; earthworms and arthropods present
		A	0–60	loam (48% sand, 36% silt, 16% clay), 10YR3/2 moist, strong fine and medium granular peds, friable consistence, pH 5.0 ± 0.1, 1.10 ± 0.08%C, 0.14 ± 0.01%N
		Bw	6 – ±200	loam, 10YR4/3 moist, moderate medium subangular blocky peds, firm consistence
3 and 4	Pachic Xerumbrept	Oi (L)	2–1	poplar leaf and small branches litter; very loose and low density
		Oe (F)	1–0	fragmented and partly decomposed litter; earthworms and arthropods present
		A	0–60	silt loam (22% sand, 53% silt, 25% clay), 5YR3/3 moist, strong fine and medium granular peds, friable consistence, pH 4.8 ± 0.0, 1.15 ± 0.01%C, 0.14 ± 0.01%N
		Bw	60–	silt loam, 5YR4/3 moist, moderate medium subangular blocky peds, firm consistence, rounded Fe concretions and Fe deposits along former root channels, black Fe-Mn concretions, below 100 cm yellow-white weathered pommish (BC horizon below 100 cm)
5	Dystric Xerochrept	Oi (L)	2–1	poplar leaf and small branches litter; very loose and low density
		Oe (F)	1–0	fragmented and partly decomposed litter; earthworms and arthropods present
		A	0–45	loam (48% sand, 39% silt, 13% clay), 10YR3/3 moist, strong fine and medium granular peds, friable consistence, pH 4.9 ± 0.0, 0.65 ± 0.07%C, 0.11 ± 0.01%N
		Bw	45–70	loam, 10YR4/3 moist, moderate medium subangular blocky peds, firm consistence
		BC	70–	loam with pockets of yellow-white weathered pommish
6	Pachic Xerumbrept	Oi (L)	3–1	poplar leaf and small branches litter; very loose and low density
		Oe (F)	1–0	fragmented and partly decomposed litter; earthworms and arthropods present
		A	0–70	loam (41% sand, 44% silt, 15% clay), 10YR3/2 moist, strong fine and medium granular peds, friable consistence, pH 5.5 ± 0.0, 1.18 ± 0.04%C, 0.13 ± 0.01%N
		BC	70–80	loam, sedimentary layers
		2BC	80–	loam, firm consistence, rounded Fe concretions and Fe deposits along former root channels, black Fe-Mn concretions, weathered volcanic ash with pockets of yellow-white weathered pommish

^aThe classification and descriptions of A and B horizons did not change between the end of 1998 and 2002. The O horizons were formed during that period.

undisturbed soil [Steingrobe *et al.*, 2000]. The cores were placed in the soil, between 0 and 40 cm depth, in March and collected in November of 2000 and 2001. Two subsamples of each core at 10 and 30 cm depth were taken, mixed and kept at 4°C until analysis. The samples were crushed by hand, and live roots were removed. Carbon was determined by flash combustion in an elemental analyzer and ¹³C abundance was determined after conversion of total C to CO₂, purified by CuO and Ag, in a VG/SIRA 9 Mass Spectrometer. Results were expressed as δ¹³C (‰) versus the PDB standard. The fraction of C in the soil derived from leaf and root litter during the incubation period (f), was calculated with a simple mixing model as described by Balesdent *et al.* [1988] and others [Leavitt *et al.*, 2001; Van Kessel *et al.*, 2000a],

$$f = (\delta^{13}\text{C}_{\text{incubated soil}} - \delta^{13}\text{C}_{\text{initial C4soil}}) / (\delta^{13}\text{C}_{\text{poplar}} - \delta^{13}\text{C}_{\text{initial C4soil}}).$$

The amount of newly sequestered soil C during the incubation period (C_{new}) was obtained by multiplying the C content of the incubated sample by f. The SPSS (v 10.1) general linear model was used to calculate univariate analysis of variance and to evaluate treatment effects.

3. Results

[11] At the end of years 2000 and 2001 total soil C contents (C_{total}) under ambient CO₂ treatment had increased for all species at all depths (Table 3). However, under FACE, C_{total} had decreased for *P. nigra* and *P. euramericana* at 10–20 cm depth in 2000 and at all depths in 2001. In addition, in 2000 at 0–10 and 20–30 cm depths, C_{total} increases were the greatest under ambient CO₂ for two out of three species. At the end of 2001, average C_{total} contents showed smaller increases or declined under FACE treatment as

Table 2. Initial C and N Contents ($\text{g C} \times 0.1 \text{ m}^{-1} \text{ depth} \times \text{m}^{-2}$) and C/N Ratio of the Mineral Soil Per Plot

Plot	Depth, cm	Initial C, g/0.1m ³ (n = 6)	Standard Error	Initial N, g/0.1m ³ (n = 6)	Standard Error	Initial C/N (n = 6)	Standard Error
2 (control CO ₂)	0–10	1205	34	128	5	9.4	0.2
	10–20	1279	31	140	5	9.2	0.2
	20–30	1286	37	144	5	9.0	0.2
3 (control CO ₂)	0–10	1271	18	138	3	9.2	0.2
	10–20	1237	33	136	3	9.1	0.2
	20–30	1240	24	132	2	9.4	0.1
6 (control CO ₂)	0–10	1475	58	152	5	9.7	0.1
	10–20	1395	55	140	4	9.9	0.1
	20–30	1393	35	143	3	9.8	0.1
1 (FACE CO ₂)	0–10	1353	58	144	6	9.4	0.3
	10–20	1337	49	137	7	9.8	0.4
	20–30	1314	56	133	7	9.9	0.3
4 (FACE CO ₂)	0–10	1230	47	136	5	9.1	0.1
	10–20	1173	17	128	3	9.2	0.2
	20–30	1152	12	130	2	8.9	0.1
5 (FACE CO ₂)	0–10	859	54	98	5	8.7	0.3
	10–20	815	43	92	4	8.9	0.1
	20–30	754	59	85	7	8.9	0.5

compared to control. This trend was significant at the 0–10 cm depth only. On average, C_{total} contents increased under control and FACE, respectively, by 12 and 3%, i.e., 484 and 107 gC/m^2 , during the experiment. Changes in total N content were always negative under FACE at the end of 2001. Significantly more N was lost from the soil at 0–10 cm depth under FACE.

[12] The local soil of the plantation had an average $\delta^{13}\text{C}$ of $-24.86 \pm 0.14\%$. Poplar leaves and roots had $\delta^{13}\text{C}$ values of $-29.45 \pm 0.6\%$ and $-29.56 \pm 0.6\%$, respectively. The C4 soil used for incubation had an average initial $\delta^{13}\text{C}$

value of $-18.31 \pm 0.07\%$. These values together with the $\delta^{13}\text{C}$ values of the samples collected after incubation were used to calculate the amount of soil C that was sequestered during the active growing seasons of 2000 and 2001 (Table 4).

[13] Between March and November of 2000, more C_{new} was sequestered under FACE in the mineral topsoil planted with *P. alba* and *P. nigra*. However, under *P. euramericana* the opposite effect was observed. During 2001, significantly ($P = 0.000$) more C_{new} was sequestered under FACE in the mineral topsoil for all three species. On average, 704 and

Table 3. FACE and Species Effects on Total Soil C and N Content and C/N Ratio of the Mineral Soil Expressed as the Difference Between the Initial and End of Year Values

Depth, cm	Species <i>P.</i>	CO ₂ Treatment	Initial		2000				2001					
			C, g/0.1m ³ (n = 6)	Standard Error	N, g/0.1m ³ (n = 6)	Standard Error	ΔC , g/0.1m ³ (n = 6)	Standard Error	ΔN , g/0.1m ³ (n = 6)	Standard Error	ΔC , g/0.1m ³ (n = 6)	Standard Error	ΔN , g/0.1m ³ (n = 6)	Standard Error
0–10	<i>alba</i>	control	1320	71	137	7	45	44	3	6	252 ^a	37	18 ^b	4
		FACE	1128	100	130	11	67	56	-5	2	207 ^a	79	-2 ^b	9
	<i>nigra</i>	control	1379	68	143	6	78	73	4	6	202 ^a	59	11 ^b	7
		FACE	1149	134	124	11	20	49	-5	6	-3 ^a	106	-8 ^b	8
	<i>euram</i>	control	1252	39	138	4	177	36	8	6	197 ^a	41	9 ^b	5
		FACE	1165	83	125	10	48	70	7	6	-47 ^a	84	-8 ^b	17
10–20	<i>alba</i>	control	1339	53	140	5	4	29	-5	7	4	30	-2	4
		FACE	1096	111	123	13	4	44	-7	4	45	60	-5	8
	<i>nigra</i>	control	1341	49	143	3	37	70	-4	6	8	59	-8	5
		FACE	1117	116	117	9	-53	32	-5	5	-60	68	-12	8
	<i>euram</i>	control	1231	32	133	4	92	45	4	7	59	63	2	9
		FACE	1112	85	116	7	-6	41	2	5	-38	39	-8	4
20–30	<i>alba</i>	control	1311	49	140	5	68	22	2	7	41	32	-2	4
		FACE	1088	120	121	13	28	37	-3	6	7	78	-8	10
	<i>nigra</i>	control	1343	44	141	4	61	66	-4	7	52	59	-1	2
		FACE	1044	136	113	11	35	19	4	3	-8	74	-6	7
	<i>euram</i>	control	1264	25	138	4	59	69	2	10	14	57	-8	8
		FACE	1089	82	113	9	81	44	9	5	-7	58	-1	5

^aSignificant FACE treatment effect ($P = 0.009$).

^bSignificant FACE treatment effect ($P = 0.020$).

Table 4. Newly Sequestered Soil C (C_{new}) in the Mineral Soil as Estimated With the $^{13}\text{C}/^{12}\text{C}$ Isotopic Method

Species	CO ₂ Treatment	C, % (n = 9)	Standard Error	$\delta^{13}\text{C}$, ‰ (n = 9)	Standard Error	f (n = 9)	Standard Error	C_{new} , g/m ² (n = 9)	Standard Error
<i>Incubation Period March–November 2000</i>									
<i>P. alba</i>	control	0.89	0.08	−19.52	0.15	0.13	0.01	365	34
	FACE	0.96	0.07	−19.80	0.27	0.15	0.02	462	35
<i>P. nigra</i>	control	1.08	0.03	−19.46	0.09	0.12	0.01	445	42
	FACE	1.03	0.05	−19.61	0.15	0.13	0.01	469	54
<i>P. euram</i>	control	1.06	0.01	−19.67	0.15	0.14	0.01	518	54
	FACE	1.08	0.02	−19.54	0.19	0.13	0.02	488	87
Species	CO ₂ Treatment	C, % (n = 15)	Standard Error	$\delta^{13}\text{C}$, ‰ (n = 15)	Standard Error	f (n = 15)	Standard Error	C_{new} , g/m ² (n = 15)	Standard Error
<i>Incubation Period March–November 2001</i>									
<i>P. alba</i>	control	0.90	0.01	−19.02	0.10	0.08	0.01	227 ^a	34
	FACE	0.81	0.05	−19.79	0.21	0.15	0.02	408 ^a	62
<i>P. nigra</i>	control	0.89	0.01	−19.06	0.19	0.08	0.02	244 ^a	67
	FACE	0.95	0.04	−19.75	0.14	0.15	0.01	480 ^a	42
<i>P. euram</i>	control	0.88	0.01	−19.28	0.22	0.10	0.02	313 ^a	71
	FACE	0.96	0.02	−19.66	0.16	0.14	0.01	470 ^a	60

^aSignificant FACE treatment effect ($P = 0.000$).

926 g/m² C_{new} was sequestered, respectively, under control and FACE conditions during the experiment.

4. Discussion

[14] The soils of the POPFACE plantation are characterized by thick A horizons. The many fine granular peds of which these A horizons consist are the result of the activity of earthworms, arthropods, and other soil biota. The combined activity of these soil organisms causes bioturbation and formation of soil structure [Van Breemen and Burman, 2002]. Bioturbation causes mixing of above and below-ground litter with mineral soil parts. During subsequent stages of humification, soil organic matter may become increasingly protected against decomposition by physical protection in aggregates [Pulleman, 2002] and/or chemical protection by bonding with metallic surfaces [Wattel-Koekkoek et al., 2003, 2001] resulting in long-term storage of C in the soil.

[15] Several research efforts were directed at explaining the effect of elevated CO₂ on potential C sequestration of tree dominated ecosystems. In the Duke FACE experiment, Schlesinger and Lichter [2001] found a significant accumulation of C in the litter layer after 3 years of growth at increased CO₂ concentrations, but found no accumulation of carbon in the mineral soil layers. Because the potential carbon sink in the litter layer is limited by a turnover time of about 3 years, they suggested that significant long-term net carbon sequestration in forest soils is unlikely.

[16] At another FACE facility in a sweetgum (*Liquidambar styraciflua*) plantation at Oak Ridge, Tennessee, NPP increased on average by 21% during 3 years of elevated CO₂ treatment [Norby et al., 2002]. During this period a shift in allocation of the extra photosynthate in CO₂ enriched trees was observed. At first, most extra C was allocated to woody biomass (80%), but during subsequent years this portion declined (54 and 25%). This trend was matched by increased fine-root production, which offers possibilities of increased C sequestration in soil organic matter pools with relatively longer turnover times. As long

as more C enters the system due to increased atmospheric CO₂ concentrations, it will be necessary to look to the soil for evidence of C sequestration [Norby et al., 2002].

[17] In the POPFACE experiment an increase of above-ground biomass was observed for *P. alba* (27%), *P. nigra* (15%), and *P. euramericana* (26%) under FACE treatment [Calfapietra et al., 2003]. This was reflected below ground, where total root biomass increased by 47%, 76%, and 71% for *P. alba*, *P. nigra*, and *P. euramericana*, respectively [Lukac et al., 2003]. In view of these observations, we expected greater increases of C_{total} and C_{new} under FACE than under control. The overall FACE effect on soil C pools is presented in Figure 1. The old soil C pool (C_{old}) was defined as C_{total} minus C_{new} , while the respired C was calculated as the difference between C_{old} at the beginning and the end of the experiment. C_{total} ($C_{\text{old}} + C_{\text{new}}$) increased by 12 and 3%, i.e., 484 and 107 g/m², under control and FACE, respectively. During the same time span, 704 and 926 g/m² C_{new} was sequestered under control and FACE. The C_{old} pool lost relatively more C under FACE resulting in

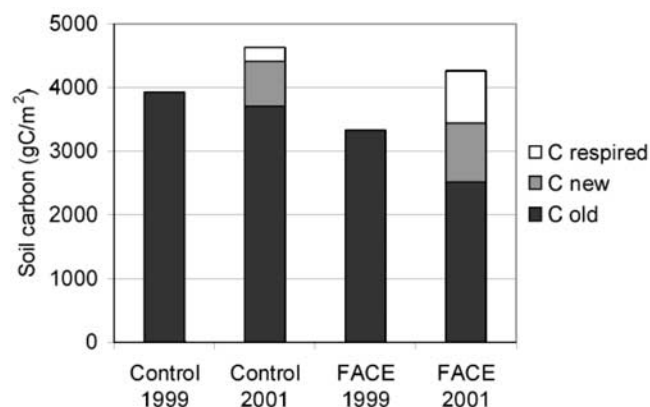


Figure 1. FACE effect on C pools of the mineral soil during the experiment.

a loss of C by respiration of, respectively, 220 and 819 g/m² under control and FACE.

[18] The soil at the Duke FACE experiment was classified as a low-fertility Ultic Alfisol with a topsoil of up to 1 m deep and a pH of 5.8. After 3 years, *Schlesinger and Lichter* [2001] found a significant increase in litter layer mass under FACE, but observed no FACE effect on the C and N contents of the mineral soil compartments. Why did 3 years of FACE treatment have no significant effect on soil C content at the Duke plantation, while it had significant opposite effects on C_{total} and C_{new} at the POPFACE plantation? Both soils have well-developed deep A horizons that are slightly acidic, but have been under different land use for the past several decades. Both soils were formed under forest, but the Duke plantation was established 20 years ago, whereas the POPFACE plantation was planted 3 years ago on a soil that had been under agricultural management for more than 45 years. As a result, the soil C/N ratios in the Duke plantation range between 16 and 19, while in the POPFACE plantation this range is between 9 and 10.

[19] At both plantations, FACE-induced measured increases of above and below ground production. In addition, increased low-molecular-weight C compounds exudation by roots is reported when plants are grown under elevated atmospheric CO₂ concentrations [*Fox and Comerford*, 1990; *Delucia et al.*, 1997; *Jones et al.*, 1998]. Under CO₂ enrichment around natural CO₂ springs in central Italy, leaves of deciduous *Quercus pubescens* contained twice as much non-structural carbohydrates (NSC) inside as compared to outside of the vent area [*Körner*, 2003; *Körner and Miglietta*, 1994]. After 1 year of FACE treatment at the alpine treeline experiment, NSC contents of *Larix decidua* and *Pinus uncinata* increased, respectively, with 17 and 38% [*Hättenschwiler et al.*, 2002]. At the small-pot scale it has been demonstrated that compared to ambient CO₂ concentrations, elevated CO₂ increases the amount of C allocated to the rhizosphere by enhancing root deposition, or total rhizosphere respiration [*Cheng*, 1999; *Cheng and Johnson*, 1998; *Gorissen*, 1996; *Hungate et al.*, 1997; *Ineson et al.*, 1996]. To draw a tentative conclusion from all these observations, in addition to increasing the amount of above and below ground litter, FACE probably also increases the fraction of easily metabolizable C.

[20] In a low-fertility soil with a relatively high C/N ratio as in the Duke plantation, the FACE-induced increases in the amount and degradability of litter input may have a limited effect on the soil microbial population due to a limitation of N or other nutrient. Moreover, a meta-analysis of data from naturally senesced leaves in field experiments showed that the N concentration in leaf litter was 7.1% lower under FACE conditions [*Norby et al.*, 2001]. Therefore the turnover rate of the soil organic C in a low-fertility soil may not be affected by FACE. In contrast, in a fertile soil the extra and more easily degradable C may increase the growth of the microbial population [*Dalenberg and Jager*, 1989; *Jenkinson and Rayner*, 1977; *Van Veen et al.*, 1984]. This will increase the respiration rate of the C_{new} fraction, and may simultaneously increase respiration rates of older C fractions and shorten their turnover times.

[21] We hypothesize that the opposite effects of FACE on soil C_{total} and C_{new} at the POPFACE plantation were caused by a priming effect of the newly incorporated litter. Priming effect was defined as the stimulation of SOM decomposition caused by the addition of labile substrates [*Cheng*, 1999; *Dalenberg and Jager*, 1989]. This FACE-induced priming effect may have caused increased respiration rates. This trend is opposite from recent predictions of a large potential sink of C in soils under future increased atmospheric CO₂ concentrations [*Cox et al.*, 2000; *Harrison et al.*, 1993].

[22] The new questions which need to be answered are: (1) For how long will this hypothesized FACE induced priming effect continue to accelerate the respiration of older SOM fractions? (2) At which point in time will the net soil C sequestration become larger under FACE than under ambient conditions? We suspect that the answers to these questions are closely related to the nutrient status of the soil or, more specifically, to the rate at which nitrogen is lost from the soil by nitrification, leaching of nitrate and harvest.

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