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Elevated CO₂ influences nutrient availability in young beech-spruce communities on two soil types

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Abstract The objectives of this study were to investigate how different soil types and elevated N deposition (0.7 vs 7 g N m⁻²a⁻¹) influence the effects of elevated CO₂ (370 vs 570 µmol CO₂ mol⁻¹) on soil nutrients and net accumulation of N, P, K, S, Ca, Mg, Fe, Mn, and Zn in spruce (*Picea abies*) and beech (*Fagus sylvatica*). Model ecosystems were established in large open-top chambers on two different forest soils: a nutrient-poor acidic loam and a nutrient-rich calcareous sand. The response of net nutrient accumulation to elevated atmospheric CO₂ depended upon soil type (interaction soil × CO₂, $P < 0.05$ for N, P, K, S, Ca, Mg, Zn) and differed between spruce and beech. On the acidic loam, CO₂ enrichment suppressed net accumulation of all nutrients in beech ($P < 0.05$ for P, S, Zn), but stimulated it for spruce ($P < 0.05$ for Fe, Zn). On the nutrient-rich calcareous sand, increased atmospheric CO₂ enhanced nutrient accumulation in both species significantly. Increasing the N deposition did not influence the CO₂ effects on net nutrient accumulation with either soil. Under elevated atmospheric CO₂, the accumulation of N declined relative to other nutrients, as indicated by decreasing ratios of N to other nutrients in tree biomass (all ratios: $P < 0.001$, except the N to S ratio). In both the soil and soil solution, elevated CO₂ did not influence concentrations of base cations and available P. Under CO₂ enrichment, concentrations of exchangeable NH₄⁺ decreased by 22% in the acidic loam and increased by 50% in the calcareous sand (soil × CO₂, $P < 0.001$). NO₃⁻ concentrations decreased by 10–70% at elevated CO₂ in both soils ($P < 0.01$).

Keywords Allocation · Competition · Fertility · Forest ecosystem · N deposition

Introduction

Since forest ecosystems contain 62–78% of the total terrestrial carbon (Dixon et al. 1994; Perruchoud and Fischlin 1995), the response of forests to the rising atmospheric CO₂ concentrations is crucial for the global carbon cycle. Elevated atmospheric CO₂ concentrations could potentially increase C storage in ecosystems, which would slow future rises in atmospheric CO₂ (Woodwell and Mackenzie 1995). However, it is questionable whether forests can sequester more carbon in a future CO₂-rich world since many forests are limited by the availability of other resources such as nutrients. Therefore, the magnitude of the response of forest ecosystems to elevated atmospheric CO₂ has been linked to soil fertility (Kirschbaum et al. 1998; Niklaus et al. 1998; Maurer et al. 1999). In the long term, elevated CO₂ might decrease nutrient availability through increased storage of nutrients in both plant biomass and unreactive soil organic matter (Comins and McMurtrie 1993; Schimel 1995; Kirschbaum et al. 1998). Under CO₂ enrichment, extra plant-fixed carbon is allocated below-ground, which in turn may stimulate microbial activity and immobilization of nutrients by soil microbes (Diaz et al. 1993; Zak et al. 1993; Cotrufo and Gorissov 1997). Plants grown under elevated CO₂, however, can counterbalance reduced nutrient availability in the soil by better nutrient-use efficiency and by increased nutrient acquisition through stimulation of mycorrhiza and root growth (Berntson and Bazzaz 1996).

Since many forest ecosystems are limited by nitrogen (Vitousek and Howarth 1991), elevated N deposition might increase C storage in forests. The magnitude of this effect, however, is not known exactly (Townsend et al. 1996; Nadelhoffer et al. 1999). Increased N deposition might also influence the response of forests to elevated atmospheric CO₂ by removing N limitations, but the reported interactive effects of N and CO₂ on trees are inconclusive, ranging from negligible to significant (Hättenschwiler and Körner 1998; Mikan et al. 2000; Walker et al. 2000). The inherent nitrogen status of the

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soil may be one reason for this variety of responses to increased N deposition.

Most studies on feedback mechanisms of elevated CO_2 concentrations with nutrients have focussed mainly on nitrogen (Diaz et al. 1993; Cotrufo and Gorissón 1997; Hungate et al. 1999). Usually, CO_2 enrichment reduces plant N concentrations (reviewed by Cotrufo et al. 1998) and often increases the immobilization of N in soils (Niklaus et al. 1998; Hungate et al. 1999), indicating that elevated atmospheric CO_2 stimulates the competition of plants and microbes for N. The effects of CO_2 enrichment on other nutrients are less consistent. In plant tissues, they range from decreased to increased concentrations under elevated CO_2 (Roberntz and Linder 1999; Gifford et al. 2000; Walker et al. 2000). In the soil, reports on the impact of CO_2 enrichment on exchangeable nutrients other than N are scarce, but they suggest that these CO_2 effects are relatively small (Johnson et al. 2000). Cycling of nutrients under elevated CO_2 may differ between N and other nutrients, because the largest fractions of base cations occur in inorganic forms in the soil, whereas N is mainly organically bound (Marschner 1995). In the long term, this might change nutrient cycling and induce imbalances in the nutrient supply of plants.

In diverse plant communities, the responsiveness of various plants to elevated CO_2 can be different, which may lead to changes in plant community composition and biodiversity (Leadley et al. 1999; Körner 2000; Joel et al. 2001). Differences in the species-level growth response to elevated CO_2 may be driven, by and large, by differences in the ability to acquire nutrients (Berntson et al. 1998). In addition, elevated atmospheric CO_2 can also change the competitiveness of plants indirectly by increasing or decreasing the availability of nutrients in soils. Therefore, soil fertility is probably one important co-factor determining shifts in competitiveness of different species under CO_2 enrichment (Joel et al. 2001).

In this present study, model ecosystems with spruce and beech were exposed to elevated atmospheric CO_2 concentrations and to increased N deposition in large open-top chambers for 4 years. The juvenile spruce and beech forests were established on two different soils: a nutrient-poor acidic loam and a nutrient-rich calcareous sand. We quantified the net nutrient accumulation in spruce and beech trees. In the final year, we also measured concentrations of nutrients in the soil solution and exchangeable nutrients in soils. The following hypotheses were tested: (1) The soil type will determine the response of nutrient accumulation in spruce and beech to elevated CO_2 . (2) Increased N deposition will enlarge the responsiveness of trees to elevated CO_2 on the nutrient-poor soil only, where it removes N limitations. (3) Nitrogen in plants and soils, which is mainly organically bound in soils, will respond differently to elevated CO_2 than other nutrients occurring in inorganic forms in soils.

Materials and methods

Experimental set-up

Model forest ecosystems were established in 16 large open-top chambers (height 3 m, diameter 3 m, depth 1.5 m, soil layer 1 m) in 1994 (Landolt et al. 1997). The following four combinations of treatments were applied: ambient CO_2 (370 $\mu\text{mol mol}^{-1}$) + low N deposition (0.5–0.7 g $\text{NH}_4\text{NO}_3\text{-N m}^{-2}\text{a}^{-1}$); elevated CO_2 (570 $\mu\text{mol mol}^{-1}$) + low N; ambient CO_2 + high N (5–7 g $\text{NH}_4\text{NO}_3\text{-N m}^{-2}\text{a}^{-1}$); elevated CO_2 + high N. The open-top chambers were arranged in a Latin square design with four replications for each $\text{CO}_2 \times \text{N}$ treatment. All treatments were studied in two soils, each in one of the two separate gravitational lysimeters (surface area 3 m^2) of each open-top chamber. One of the soils was an acidic sandy loamy Haplic Alisol (FAO 1990), referred to as 'acidic loam'. The other soil was a calcareous loamy sandy Fluvisol (FAO 1990), quoted as 'calcareous sand'. There were no organic layers on the soils in the lysimeters. The acidic loam consisted of 55, 29, and 16% of sand, silt, and clay, while the calcareous sand had a sand, silt, and clay content of 84, 10, and 6%, respectively. Both topsoils (0–40 cm depth) had an initial organic C content of about 13 g C per kg soil. Other properties of the topsoils are shown in the tables. The subsoil of the acidic loam was poor in soil organic matter (2.3 g C per kg soil), while that of the calcareous sand was the same as the topsoil. The soils were transferred in layers from natural beech-spruce forest sites in Switzerland into the lysimeters in spring 1994. Each model ecosystem consisted of eight beech (*Fagus sylvatica* L.) and eight spruce (*Picea abies* Karst) trees as well as of five typical understory species (*Carex sylvatica*, *Geum urbanum*, *Ranunculus ficaria*, *Viola sylvatica* and *Hedera helix*). At planting, beeches, which had been grown from seeds collected from four selected provenances (two individuals per provenance), were 2–3 years old. Spruce trees were 4 years old at planting; two spruce trees were seedlings from two Swiss provenances, whereas six trees were clonal cuttings from six genotypes (for details see Egli et al. 1998). The planting positions of trees were fixed but randomized for populations/genotypes, i.e. the spatial arrangement of all model ecosystems was identical. Further details about the experimental set-up can be found in Landolt et al. (1997) and Egli et al. (1998).

The CO_2 enrichment started at the end of January 1995. The incoming air was blown through textile tubes at a rate of 3000 $\text{m}^3 \text{h}^{-1}$. The chambers were irrigated with electro-osmotically purified tap water with ions added in concentrations comparable to those usually found in rain water. Nitrogen was added as NH_4NO_3 to the irrigation water.

Net nutrient accumulation in trees

After four treatment years, trees were harvested in September 1998. Fine roots, coarse roots, stems, twigs, and foliage were collected separately, dried at 60°C and weighed. Needles were separated according to their different age classes. Subsamples of each tree fraction were ground with a milling machine. Plant tissues were digested with a MLS UltraClave (Milestone, Sorisole, Italy) by adding 2 ml of 40% HNO_3 and 80 μl of concentrated HF to 200 mg of sample. After 15–20 min at room temperature, the samples were heated for 55 min at 240°C and 134 hPa. The samples were diluted with H_2O and analysed for nutrients by inductively coupled plasma atomic emission spectrometry (ICP-AES Optima 3000, Perkin Elmer, Norwalk, C.T.). Carbon and N in plant samples were analysed with a C/N analyser (NA 1500, Carlo Erba, Milan, Italy). The net accumulation of nutrients in trees was calculated by multiplying the mass of the tree fractions with their nutrient concentrations. From the sum of all nutrient masses, the initial masses of nutrients in the planted trees (<6% of the final mass) randomly selected at planting were subtracted.

Nutrients in soil solution and in soils

Soil solution was sampled during the 4th and final growing season in 1998. Two suction cups (SoilMoisture Equipment Corp) per

lysimeter were installed at depths of 5, 25, and 50 cm in March 1998. Prior to their installation, all suction cups were flushed with 1 N HCl, then with distilled water and finally with soil solution. The suction cups were connected to a 100 ml glass bottle and evacuated with 30 kPa twice a week.

Soil samples were taken at the end of the experiment in September 1998. Samples from 0–10 cm depth were collected with two cores (diameter 10 cm) from each lysimeter. Samples from 50–60 and from 90–100 cm depth were taken with a soil corer (diameter 5 cm).

To determine soil mineral N concentrations, the soil was bulked depthwise, stored at 4°C and extracted within 24 h after sampling. For analysis, roots were removed, the soil was homogenized and a subsample was extracted for 1 h with 1 M KCl at a 1:4 soil/solution ratio. For the other analysis, the soil was dried at 50°C, passed through a 2-mm sieve and stored at room temperature. Soil pH was measured potentiometrically in 0.01 M CaCl₂ with a solid/extractant ratio of 1:2. Exchangeable cations were extracted with 1 M NH₄Cl on an end-over-end shaker using a solid/extractant ratio of 1:10. Available phosphorus was estimated by extracting the soil with NaHCO₃ at pH 8.5 and determining the concentration of P photometrically with the molybdenum blue method (Olsen et al. 1954).

In the soil extracts, soil solution, and drainage water, concentrations of cations were measured by ICP-AES (Optima 3000, Perkin Elmer, Norwalk, Conn.). Ammonium was determined colorimetrically by automated flow injection analysis (PE Fias-300, Perkin-Elmer). Anions (Cl⁻, NO₃⁻, SO₄²⁻) in the soil solution were measured by ion chromatography (DX-120, Dionex, Sunnyvale, Calif.). NO₃⁻ in the soil was determined photometrically at 210 nm according to Norman and Stucki (1981) with an UV-160 spectrophotometer (Shimadzu, Tokyo, Japan).

Statistical analyses

The significance of treatment effects was tested by analyses of variance (ANOVA). Following the experimental design, the main effects of CO₂ and N and their interaction were tested against the 'chamber mean squares', while the main effects of soil type and its interactions with CO₂, N and CO₂ × N were tested against the 'lysimeter mean squares'. The responses of the tree community and the those of the trees species alone were all analysed with separate ANOVAs. In separate analyses, the significance of the effects of CO₂ and N were tested for the acidic and the calcareous soil. All statistical analyses were performed with SAS (SAS System 6.12, Cary, N.C.).

Results

Net nutrient accumulation in spruce and beech

The impact of elevated CO₂ on tree growth and on net accumulation of nutrients was different for beech and spruce and depended upon the soil type (interaction of species × soil type, $P < 0.01$; Fig. 1). The net nutrient acquisition by spruce was stimulated by elevated CO₂ on both soil types (up to +70%). On acidic loam, this effect was smaller than the CO₂-induced growth enhancement, showing that CO₂ enrichment led to a dilution in nutrient concentrations of spruce and to a widening of the C:nutrient ratios. In contrast to spruce, growth of beech and the amount of all nutrients in beech decreased under elevated CO₂ on acidic loam (−2% for Fe, up to −17% for Zn), but increased on the calcareous sand (+15% for Mn, up to +66% for P), except for the amount of N and S. The interaction between CO₂ and soil type was signifi-

Fig. 1 Change of the net accumulation of C, N, P, K, S, Ca, Mg, Fe, Mn, and Zn in spruce and beech during 4 years due to CO₂ enrichment. The CO₂-induced change of element accumulation relative to control was estimated by dividing element accumulation under elevated CO₂ by that under ambient CO₂. Data were pooled across both N treatments since CO₂ and N showed no significant interaction. Means and standard errors of eight model ecosystems

cant for the accumulation of all nutrients in spruce and of P, K, Mg and Zn in beech. On calcareous sand, the increase in total net nutrient accumulation in the total community biomass under elevated CO₂ was largest for P (+60%) and smallest for N (+12%; Fig. 1, Table 1), indicating that the ratio of N to P decreased under CO₂ enrichment (−30%, $P < 0.001$). The concentrations of P and Zn in total tree biomass were higher under elevated than at ambient CO₂ on the calcareous sand (P +28%; Zn +23%; $P < 0.001$).

Nutrient allocation

Figure 2 shows the impact of elevated CO₂ on the allocation of nutrients into below-ground biomass. For spruce, atmospheric CO₂ did not influence net accumulation of nutrients in roots relative to those in the total biomass on either of the soils. In contrast, beech growing on the acidic loam allocated more nutrients into

Table 1 Effects of elevated atmospheric CO₂ and N deposition on total net nutrient accumulation (g m⁻²) in the total community biomass (spruce + beech) during 4 years. Means of 4 model ecosystems (*n.s.* not significant)

	N	CO ₂	ex. N	P	K	S	Ca	Mg	Fe	Mn	Zn
Acidic loam	Low	Ambient	15.1	2.43	8.8	1.23	13.6	2.11	0.333	1.63	0.242
	Low	Elevated	15.0	2.71	10.5	1.40	14.3	2.06	0.392	1.73	0.277
	High	Ambient	23.9	3.07	13.3	1.72	17.6	2.99	0.540	2.02	0.327
	High	Elevated	22.9	2.63	11.7	1.55	18.4	2.90	0.572	2.12	0.332
Significance	CO ₂		<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>
	N		***	**	***	**	***	***	*	***	***
	CO ₂ × N		<i>n.s.</i>	**	<i>n.s.</i>	*	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>
Calcareous sand	Low	Ambient	33.4	4.77	14.4	2.52	37.5	3.86	0.295	0.044	0.240
	Low	Elevated	34.7	7.20	19.1	2.79	48.2	4.87	0.509	0.060	0.379
	High	Ambient	35.2	3.70	13.1	2.51	34.8	3.31	0.274	0.041	0.219
	High	Elevated	42.7	6.50	18.9	3.07	44.8	4.43	0.405	0.052	0.336
Significance	CO ₂		**	***	***	***	***	***	**	**	***
	N		***	**	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	*	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>
	CO ₂ × N		<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>
Soil effects	Soil Type		***	***	***	***	***	***	<i>n.s.</i>	***	<i>n.s.</i>
	Soil × CO ₂		***	***	***	**	***	**	<i>n.s.</i>	<i>n.s.</i>	***
	Soil × N		***	***	***	<i>n.s.</i>	**	***	*	***	***

P*<0.05, *P*<0.01, ****P*<0.001 **Fig. 2** Fractions of net below-ground accumulation of C, N, P, K, and S in spruce and beech as compared with the total net element accumulation (above + below) at an N deposition of 0.5–0.7 g N m⁻² a⁻¹. Means and standard errors of four model ecosystems

roots under elevated than at ambient CO₂ (*P*<0.05), but this effect was more pronounced under low (+22% for K to +31% for S) than under high N deposition (+2% for K to +7% for N) (interaction CO₂ × N: *P*<0.05). On the calcareous sand, elevated CO₂ had no effect on nutrient allocation in beech, except for a greater net accumulation of P in roots under CO₂ enrichment (+29%, *P*<0.001). For both species, the allocation of all nutrients into below-ground biomass was similar to that of the biomass at both CO₂ levels (Fig. 2), indicating that trees did not allocate specific nutrients preferentially into their rooting systems.

Accumulation of N in comparison to other nutrients

We compared net accumulation of N in trees receiving a low N input of 0.7 g N m⁻² a⁻¹ with that of other nutrients by dividing the amount of N in trees by the amounts of other nutrients (Fig. 3). Elevated CO₂ decreased all ratios of N to other nutrients for both tree species (*P*<0.001 except the N to S ratio), indicating that, under CO₂ enrichment, accumulation of N declined in comparison to other nutrients. The response depended upon soil type (interaction soil × CO₂ for all ratios: *P*<0.01, except the N/S ratio). The decrease was more pronounced on calcareous sand than on acidic loam (Fig. 3). Increasing the N deposition from 0.7 to 7 g N m⁻² a⁻¹ had an effect opposite to that of atmospheric CO₂: as expected, it increased the N:nutrient ratios (all ratios: *P*<0.05). The interaction between N and CO₂ was not significant, indicating that elevated CO₂ decreased the ratios of N to other nutrients similarly under high and low N deposition.

Table 2 Effects of elevated atmospheric CO₂ and N deposition on soil pH, KCl-extractable N, available P, and NH₄Cl-extractable cations of the soil at 0–10 cm depth after 4 experimental years. Means of 4 model ecosystems (*n.s.* not significant)

	N	CO ₂	pH	ex. N	P (Olsen)	CEC	Ca	Mg
				mg (kg soil) ⁻¹		mmol _c (kg soil) ⁻¹		
Acidic loam	Low	Ambient	4.2	2.30	2.1	41.6	14.3	2.07
	Low	Elevated	4.2	1.72	2.3	41.5	12.1	1.74
	High	Ambient	4.1	4.96	2.3	42.0	11.8	1.80
	High	Elevated	4.1	4.20	2.3	43.6	12.2	1.70
Calcareous sand	Low	Ambient	7.1	3.66	6.5	127	124	1.77
	Low	Elevated	7.2	4.23	6.7	126	123	2.04
	High	Ambient	7.2	6.45	5.6	129	126	2.26
	High	Elevated	7.1	6.36	5.7	130	126	2.10
Significance	CO ₂		<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>
	N		<i>n.s.</i>	***	**	*	<i>n.s.</i>	<i>n.s.</i>
	CO ₂ × N		<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>
Soil effects	Soil Type		***	***	***	***	***	<i>n.s.</i>
	Soil × CO ₂		<i>n.s.</i>	*	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>
	Soil × N		<i>n.s.</i>	<i>n.s.</i>	*	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>

P*<0.05, *P*<0.01, ****P*<0.001

Fig. 3 Change in the ratios of N to other nutrients in total community biomass at an N deposition of 0.5–0.7 g N m⁻²a⁻¹ due to CO₂ enrichment. The CO₂-induced change of the N:nutrient ratios relative to control was estimated by dividing the ratios under elevated CO₂ by those under ambient CO₂. Means and standard errors of four model ecosystems

Soil nutrients

The acidic loam had a lower pH and lower concentrations of exchangeable base cations, of exchangeable N, and of available P than the calcareous sand (Table 2). The effects of CO₂ enrichment on most exchangeable nutrients and on nutrients in the soil solution were not significant (Tables 2 and 3). Elevated CO₂ increased the concentrations of exchangeable K (+13%), but atmospheric CO₂ concentrations did not affect K concentrations in the soil solution. In contrast to the other nutrients, the effects of elevated CO₂ on KCl-extractable N and N in the soil solution were large but different for NH₄⁺ and NO₃⁻ and depended upon the soil type (inter-

Fig. 4 Effects of CO₂ enrichment and of N deposition on KCl-extractable ammonium and nitrate of soil from 0–10 cm depth. Low N deposition was 0.5–0.7 g N m⁻²a⁻¹ and high N deposition was 5–7 g N m⁻²a⁻¹. Means and standard errors of four model ecosystems

action soil × CO₂ for extractable total N, NH₄⁺, NO₃⁻; *P*<0.05). In the acidic loam, concentrations of extractable NH₄⁺ and NO₃⁻ were lower under elevated than at ambient CO₂ concentrations (–22% and –11%, Fig. 4). In the calcareous sand, concentrations of extractable NH₄⁺

Table 3 Effects of elevated atmospheric CO₂ and N deposition on nutrients of the soil solution (μmol l⁻¹) at 5 cm depth and on concentrations of NH₄⁺ and NO₃⁻ at 50 cm depth after 4 experimentalyears. Soil solution was collected with suction cups. Means of 4 model ecosystems (– below detection limit, *n.s.* not significant)

	N	CO ₂	5 cm depth					50 cm depth	
			Ca	Al	K	NH ₄	NO ₃	NH ₄	NO ₃
Acidic loam	Low	Ambient	72.6	12.1	–	2.44	7.6	1.08	0.7
	Low	Elevated	40.5	11.3	–	2.01	6.1	0.15	–
	High	Ambient	45.4	10.4	–	2.29	55.0	1.47	1.6
	High	Elevated	70.0	7.1	–	1.88	16.9	0.32	1.3
Calcareous sand	Low	Ambient	1118	0.67	1.7	0.25	26.1	2.09	3240
	Low	Elevated	1224	0.86	1.8	0.52	9.0	0.98	50
	High	Ambient	1449	0.68	3.0	1.11	412.4	1.99	3360
	High	Elevated	1244	0.66	5.0	0.50	227.1	3.41	1880
Significance	CO ₂		<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	*	<i>n.s.</i>	*
	N		<i>n.s.</i>	*	<i>n.s.</i>	<i>n.s.</i>	**	<i>n.s.</i>	<i>n.s.</i>
	CO ₂ × N		<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	*	<i>n.s.</i>	<i>n.s.</i>
Soil effects	Soil Type		***	***	***	***	***	*	***
	Soil × CO ₂		<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	*
	Soil × N		<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	***	<i>n.s.</i>	<i>n.s.</i>

P*<0.05, *P*<0.01, ****P*<0.001

increased (+50%), but those of NO₃⁻ decreased under CO₂ enrichment (–33%). In the soil solution, concentrations of NH₄⁺ were not affected by CO₂ concentrations in both soils, whereas NO₃⁻ declined under elevated CO₂, particularly in the calcareous sand at 50 cm depth (–70%; Table 3). The concentrations of total N in the soil were not affected by elevated CO₂, but were significantly increased by high N deposition (*P*<0.05).

Discussion

Soil-dependent CO₂ effects on nutrient accumulation in beech and spruce

The response of tree growth and net nutrient accumulation to elevated CO₂ differed between beech and spruce (Fig. 1), which indicates that CO₂ enrichment alters the competition of tree species for nutrients, with spruce winning over beech. The CO₂-induced changes of net nutrient accumulation mainly reflected changes in tree growth (Fig. 1). Soil fertility was an important co-factor for the species-dependent effects of elevated CO₂. On the acidic loam, CO₂ enrichment suppressed net accumulation of all nutrients in beech, but stimulated it for spruce, while on the nutrient-rich calcareous sand increased atmospheric CO₂ enhanced nutrient accumulation in both species significantly. The reason for the dependency of the CO₂ effects from soil types might be that the availability of nutrients becomes limiting for one of the species under elevated CO₂ (here beech), while for the other (here spruce), the nutrient supply is still adequate. The increasing nutrient limitation under CO₂ enrichment for beech might be either imposed by the larger CO₂ sensitivity of the competitor (spruce) or it might be caused indirectly by a greater nutrient immobilization at

elevated CO₂ (Joel et al. 2001). However, under adequate resource availability (here on the calcareous sand), the response of tree species to CO₂ enrichment depends solely on the responsiveness of each tree species itself. Water supply, as another potentially limiting resource, did not play an important role in this experiment, because the model ecosystems were sufficiently irrigated and in the acidic loam CO₂ enrichment increased the soil water content (Sonnleitner et al. 2001). The soil-dependent response of the two tree species to elevated CO₂ could be explained by different rooting systems of beech and spruce relative to distributions of available N in soils. Spruce developed shallow roots, while beech roots were more evenly distributed throughout all soil depths, which is consistent with rooting patterns in mature mixed spruce-beech forests (Schmid and Kazda 2001). In the acidic loam, 87% of the spruce but only 50% of the beech fine roots are located in the uppermost 20 cm of the soil (Phillip Egli, personal communication). In the acidic loam, the plant-available N was mainly concentrated in the topsoil, as indicated by concentrations of N in the soil solution (Table 3). Thus, spruce with its shallow rooting system probably had a greater capacity to acquire nutrients from the acidic loam and to profit more from the CO₂ enrichment than beech. Beech, in turn, allocated more biomass and nutrients into roots at elevated CO₂ and under low N deposition (Fig. 2), most likely increasing its competitiveness for nutrients in the acidic loam. On calcareous sand, which had a nutrient-rich subsoil, net nutrient accumulation in both species was substantially increased by elevated atmospheric CO₂; however, the gain was larger for spruce than for beech (Fig. 1).

The dependency of the CO₂ effects on tree growth and on net nutrient accumulation from soil fertility supports the concept that the spatial and temporal availability

ty of resources determines the response of plants to CO₂ enrichment (Field et al. 1992; Körner 2000). Because the juvenile forests did not fully exploit the soil resources in our model ecosystems at harvest, especially not in the calcareous sand, we expect that differences in the CO₂-response of spruce and beech would have increased further with time.

Interaction of atmospheric CO₂ and N deposition

Since many forests are limited by N, increased N deposition could potentially remove these N limitations and enlarge the response of trees to CO₂ enrichment (Townsend et al. 1996). We studied this potential interaction between N deposition and atmospheric CO₂ for the net nutrient accumulation in trees on two soils with a differing N availability. In the acidic loam, N was most likely limiting, as indicated by concentrations of N in current-year foliage (spruce 1.1%; beech 1.8%) being below critical values (Bergmann 1992) and by a significant increase in tree growth under elevated N deposition (Egli et al. 1998). In contrast to the acidic loam, trees on the calcareous sand seemed adequately supplied with N (current year foliage: spruce 1.7%; beech 2.1%) and tree growth did not respond to increased N inputs. This is consistent with KCl-extractable N in soils (Table 2) and with N in the soil solution (Table 3): concentrations of N were significantly lower in the acidic loam than in the calcareous sand. Moreover, significant leaching losses of NO₃⁻ occurred only in the calcareous sand, indicating N saturation (Hagedorn et al. 2000).

Increasing the N deposition to 5–7 g N m⁻²a⁻¹ did not affect the response of net nutrient accumulation in trees to CO₂ enrichment (the interactions between CO₂ and N deposition were not significant on either soil except for P and S on the acidic loam; Table 1). Therefore, N inputs did not have the same level of influence on the CO₂-effect as did the soil type. The most likely reason was that on the N-saturated calcareous sand, trees were already adequately supplied with N at low N deposition and, thus, additional N inputs were luxury. On the acidic loam, the lack of an interaction between CO₂ and N deposition for tree growth and net nutrient accumulation agrees with a similar experiment on a nutrient-poor podzolic soil, in which young spruce trees did not respond to the combination of elevated CO₂ and N deposition (Hättenschwiler and Körner 1998). One reason could be that trees were also limited by resources other than CO₂ and N. For instance, P concentrations of tree foliage were rather low on the acidic loam (beech and spruce 1.7 g kg⁻¹). Increasing N deposition even decreased concentrations of P in spruce (–20%, $P < 0.01$) and beech (–15%, $P < 0.01$). This suggests that elevated N deposition induced a P limitation, which is in accordance with increased N/P ratios and lower P concentrations under elevated N depositions in mature forests (Aber et al. 1989; Boxman et al. 1998). Another reason for the lacking interaction between CO₂ × N could be that the addi-

tion of 5–7 g N m⁻²a⁻¹ did not remove N limitation completely in the acidic loam, as suggested by negligible effects of elevated N deposition on concentrations of dissolved N in the soil solution at 25 cm and deeper in the soil (see Table 3 for 50 cm depth). This explanation is supported by Nadelhoffer et al. (1999), who showed that the largest part of increased N deposition is immobilized in the uppermost soil, which in turn decreases the uptake of N by trees and the effects of N deposition on tree growth.

Elevated CO₂ decreases acquisition of N in comparison to other nutrients

Net N accumulation in trees responded differently to elevated CO₂ than did accumulation of other nutrients, as shown by declining ratios of N to other nutrients under CO₂ enrichment (Fig. 3). Within the realm of CO₂ research with forest trees, investigations of nutritional responses other than those of N have been infrequent, but where nutrients other than N were measured, the responses to elevated CO₂ were either not significant or most often smaller than those of N (Murray et al. 1996; Hättenschwiler and Körner 1998; Gifford et al. 2000; Walker et al. 2000). A common explanation for reduced concentrations of plant nutrients under elevated CO₂ is dilution by larger concentrations of non-structural carbohydrates (Roberntz and Linder 1999; Körner 2000). However, this dilution would be the same for all nutrients and, thus, the decrease of the N:nutrient ratios indicates that the reduction in tree N goes beyond a simple dilution signal. This might have had two causes: either a greater ability of trees to stretch their C:N ratios in comparison to other nutrients or a greater decrease in plant-available soil N than in other soil nutrients in response to elevated CO₂. Our results support both explanations. Decrease in tissue N is often related to a lower N requirement for photosynthesis under CO₂ enrichment (Cotrufo et al. 1998), which corresponds to lower contents of Rubisco in tree foliage in our experiment (unpublished data). However, concentrations of N were not only lower under elevated CO₂ in photosynthetic active tissues, but equal reductions were found for concentrations of N in foliage, in woody tissues, and in roots. This suggests that the larger reduction of N contents in trees under elevated CO₂ relative to other nutrients was caused partly by a decrease in available N, which was greater than that of other nutrients in soils.

Soil nutrients

In the soil as well as in the soil solution, N was indeed the only nutrient that responded to CO₂ enrichment, but the effects were different for NH₄⁺ which decreased in the acidic loam and increased in the calcareous sand, and for NO₃⁻, which decreased under elevated CO₂ in both soils (Tables 2 and 3, Fig. 4). The close linkage of C and

N cycling might explain why N and not the other soil nutrients responded to CO₂ enrichment (Gifford et al. 2000). Available N for plants is almost wholly derived from mineralization of organic matter, whereas the largest fractions of other nutrients occur in inorganic forms (Marschner 1995). Phosphorus, with a large portion also bound organically, is more accessible for microorganisms than is N, because phosphorus is usually ester-bonded, while N is directly carbon-bonded and more difficult to decompose (Vitousek and Howarth 1991).

Base cations derive primarily from rock weathering and their availability is mainly controlled by the soil mineral phase. In short-term experiments, it is unlikely that changes in C cycling due to CO₂ enrichment influence weathering and the cation exchange complex. Another potential reason for a decreased availability of base cations in soils under elevated CO₂ is an increased accumulation of cations in plant biomass (Comins and McMurtrie 1993). In this experiment, however, net accumulation of base cations in trees was not affected by atmospheric CO₂ concentrations on the acidic loam. On the calcareous sand, where elevated CO₂ caused increased accumulation of base cations in trees (Table 1), this increase was not reflected in the soil, because it was quantitatively negligible in comparison to the high contents of exchangeable base cations in the soil (Table 2). An indirect effect of elevated CO₂ on base cations in soils could be increased or decreased leaching rates of NO₃⁻ (Körner and Arnone 1992; Torbert et al. 1996), which are accompanied by leaching of base cations (Aber et al. 1989). Since concentrations of NO₃⁻ in the soil solution and export of NO₃⁻ from the model ecosystems decreased under elevated CO₂ (Hagedorn et al. 2000), CO₂ enrichment would rather increase than decrease the contents of base cations in soils. However, we found no indication of an impact of elevated CO₂ on base cations in soils, which is consistent with the negligible effects on soils planted with ponderosa pine (Johnson et al. 2000).

The decreases in NO₃⁻ concentration in soils and in the soil solution under elevated CO₂ are in agreement with other experiments in agroecosystems and grasslands (Torbert et al. 1996; Niklaus et al. 1998), but are in contrast to findings in an artificial tropical forest ecosystem (Körner and Arnone 1992). This decrease might result from increased uptake of N by trees or stimulated immobilization of N by soil microbes under elevated CO₂. The amount of N in tree biomass was not increased by CO₂ enrichment on the acidic loam and only by 13% on the calcareous sand (Fig. 1), which agrees with observations in grasslands (Arnone 1997; Niklaus et al. 1998) as well as in spruce model ecosystems (Hättenschwiler and Körner 1998). Therefore, the lower concentrations of NO₃⁻ are most likely due to increased immobilization of N by soil microbes or to suppressed net nitrification through an increased substrate availability and root exudation at elevated CO₂ (Cotrufo and Gorissón 1997; Hungate et al. 1999; Mikan et al. 2000; Niklaus et al. 2001). The opposite response of NH₄⁺ and NO₃⁻ to ele-

vated CO₂ in the calcareous sand may be due to altered root uptake capacity of the different N forms, as observed by BassiriRad et al. (1997). The authors found that CO₂ enrichment increased absorption rates of NO₃⁻ in pine, but decreased those of NH₄⁺. They argued that this resulted from a greater ability of plants growing under elevated CO₂ to invest fixed C into the energy-demanding NO₃⁻ reduction. The findings of decreased soil NO₃⁻ concentrations under elevated CO₂ shows that CO₂ enrichment might reduce NO₃⁻ leaching from forest ecosystems, which in turn might increase the capacity of ecosystems to retain atmospheric N deposition.

Conclusions

Our results show that soils play a prominent role in the response of trees to CO₂ enrichment. The effects of elevated atmospheric CO₂ on net nutrient accumulation depended upon the soil type and they differed between spruce and beech. The distribution of roots relative to soil nutrients is most likely the reason for the species-dependent response of net nutrient accumulation on the two soil types. On the acidic loam, where most available N was concentrated in the topsoil, atmospheric CO₂ enrichment stimulated net nutrient acquisition in the shallow-rooting spruce, but suppressed it in the deep-rooting beech. On the calcareous sand, where N concentrations were equally high in top- and subsoils, increased atmospheric CO₂ stimulated net nutrient accumulation in both species.

The response of N in trees and soils to elevated CO₂ was different from the response of other nutrients. Under CO₂ enrichment, the accumulation of N in tree biomass declined compared with other nutrients. Consistently, elevated CO₂ decreased concentrations of NO₃⁻ in soils, but had no effects on other soil nutrients.

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