Responses of N fluxes and pools to elevated atmospheric CO₂ in model forest ecosystems with acidic and calcareous soils

Frank Hagedorn^{1,*}, Jürg B. Bucher¹, David Tarjan¹, Peter Rusert² and Inga Bucher-Wallin³

¹Swiss Federal Institute for Forest, Snow and Landscape Research (WSL), Zürcherstr. 111 CH-8903 Birmensdorf, Switzerland; ²Institute of Plant Biology / Microbiology, University of Zürich, Zollikerstr. 107, CH-8008 Zürich, Switzerland and ³Institute of Terrestrial Ecology, ETH Zürich, Grabenstr. 11, CH-8952 Schlieren, Switzerland

Received 25 November 1999. Accepted in revised form 25 May 2000

Key words: elevated CO₂, forest ecosystem, N deposition, nitrate leaching, soil solution, soil type

Abstract

The objectives of this study were to estimate how soil type, elevated N deposition (0.7 vs. 7 g N m⁻²y⁻¹) and tree species influence the potential effects of elevated CO₂ (370 vs. 570 μmol CO₂ mol⁻¹) on N pools and fluxes in forest soils. Model spruce-beech forest ecosystems were established on a nutrient-rich calcareous sand and on a nutrient-poor acidic loam in large open-top chambers. In the fourth year of treatment, we measured N concentrations in the soil solution at different depths, estimated N accumulation by ion exchange resin (IER) bags, and quantified N export in drainage water, denitrification, and net N uptake by trees. Under elevated CO₂, concentrations of N in the soil solution were significantly reduced. In the nutrient-rich calcareous sand, CO₂ enrichment decreased N concentrations in the soil solution at all depths (–45 to –100%). In the nutrient-poor acidic loam, the negative CO₂ effect was restricted to the uppermost 5 cm of the soil. Increasing the N deposition stimulated the negative impact of CO₂ enrichment on soil solution N in the acidic loam at 5 cm depth from –20% at low N inputs to –70% at high N inputs. In the nutrient-rich calcareous sand, N additions did not influence the CO₂ effect on soil solution N. Accumulation of N by IER bags, which were installed under individual trees, was decreased at high CO₂ levels under spruce in both soil types. Under beech, this decrease occurred only in the calcareous sand. N accumulation by IER bags was negatively correlated with current-years foliage biomass, suggesting that the reduction of soil N availability indices was related to a CO₂-induced growth enhancement. However, the net N uptake by trees was not significantly increased by elevated CO₂. Thus, we suppose that the reduced N concentrations in the soil solution at elevated CO₂ concentrations were rather caused by an increased N immobilisation in the soil. Denitrification was not influenced by atmospheric CO₂ concentrations. CO₂ enrichment decreased nitrate leaching in drainage by 65%, which suggests that rising atmospheric CO₂ potentially increases the N retention capacity of forest ecosystems.

Introduction

In many short-term experiments, atmospheric CO₂ enrichment increased plant productivity, suggesting that elevated CO₂ is partially sequestered in terrestrial ecosystems (Gorisson, 1996; Wullschlegel et al., 1995). However, it is questionable if such short-term observations can be extrapolated to natural ecosystems, which are often nutrient limited (Canadell et al.,

1996; Johnson et al., 1995; Schimel, 1995). For instance, in nutrient-poor grassland ecosystems, the response of plant productivity to increased CO₂ is rather low (Arnone, 1997; Diaz et al., 1993; Schäppi and Körner, 1996). Furthermore, several experiments and models have raised serious concerns of whether negative ecosystem-level feedbacks might constrain plant productivity (Canadell et al., 1996; Comins and McMurtrie, 1993; Schimel, 1995). Negative feedbacks are related to decreasing nutrient availability brought about by an increased storage of nutrients in plant

* FAX No: 004117392215; E-mail: Hagedorn@wsl.ch

material due to increases in net primary productivity, increased C/N ratios leading to reduced mineralisation rates, and an increased nutrient immobilisation by soil microbes (Berntson and Bazzaz, 1996; Diaz et al., 1993; Hungate et al., 1996; Torbert et al., 1998). However, reported impacts of elevated CO₂ on nutrient availability in the soil are not consistent. Results from Körner and Arnone (1992) and from Zak et al. (1993) suggest that N availability increases under high CO₂ as a result of a stimulated net N mineralisation.

The nutrient status of the soil appears to be one of the key factors controlling the CO₂ response of plant productivity and its consequences for N availability (Egli et al., 1998; Gorisson, 1996; Paterson et al., 1997). In nutrient-rich ecosystems, where greater plant growth can be sustained under elevated CO₂, extra plant-fixed carbon is allocated below-ground through rhizodeposition, which in turn stimulates microbial activity (Diaz et al., 1993; Van Ginkel and Gorisson, 1998). In ecosystems where primary productivity becomes nutrient limited, the effects of CO₂ enrichment on microbial populations and N availability is small (Arnone, 1997; Kandeler et al., 1998). However, there are too few experiments which properly estimated the effects of soil fertility on the response of soil N availability to elevated CO₂. Furthermore, most studies were conducted with one soil type only. In short-term and pot experiments, soil disturbance causes excessive N mineralisation which can seriously override soil N deficiency (Johnson et al., 1995).

In this study, we measured the response of N fluxes and pools to CO₂ enrichment and N deposition in spruce-beech model ecosystems that were established in large open-top chambers with two soil types: An acidic loam with a low N availability and a calcareous sand with a high N availability. The objectives were to estimate the effects of soil type, elevated N deposition and of tree species on the response of soil N dynamics to elevated atmospheric CO₂.

Materials and methods

Experimental design

Effects of atmospheric CO₂ concentrations on soil N dynamics were studied in 4-year old spruce (*Picea abies* Karst) – beech (*Fagus sylvatica* L.) model ecosystem in 16 open-top chambers (height 3 m, diameter 3 m, depth 1.5 m; Figure 1, details see Egli et

al., 1998). Each chamber was divided into two soil compartments of 3 m² surface area each. The compartments served as non-weighable lysimeters, and had a 0.5 m thick layer of quartz sand and a drainage outlet at the bottom (details see Sonnleitner et al., 2000). The treatments were combined as follows: ambient CO₂ (370 µmol CO₂ mol⁻¹) + low N deposition (0.7 g NH₄NO₃-N m⁻²yr⁻¹); elevated CO₂ (570 µmol CO₂ mol⁻¹) + low N; ambient CO₂ + high N (7 g NH₄NO₃-N m⁻²yr⁻¹); elevated CO₂ + high N. The treatments were arranged in a Latin square design with four replications for each CO₂ × N treatment. Atmospheric CO₂ enrichment started in January 1995. The incoming filtered air was blown through textile tubes at a rate of 3000 m³ per hour (Landolt et al., 1998). The chambers were irrigated with electro-osmotically purified tap water with ions added in concentrations usually found in rain water. Nitrogen was added as NH₄NO₃ to the simulated rain. The simulated rainwater was applied through nozzles just above the plant canopy. To avoid external water and nutrient inputs by rain, a transparent roof closed over the open-top chambers when it rained. There was no water stress for the trees during the fourth year of treatment as indicated by water potentials between -1 and -30 kPa. All treatments were applied to two soils sampled from natural beech-spruce forest sites in Switzerland. One of the soils was an acidic sandy loamy derived from a Haplic Alisol (FAO, 1990), referred to as 'acidic loam'. The other soil was a calcareous loamy sand deriving from a Calcaric Fluvisol (FAO, 1990), quoted henceforth as 'calcareous sand'. Each soil was filled into one of the two separate gravitational lysimeters of each open-top chamber in spring 1994. The acidic loam was transferred in two layers, a 0.4 m topsoil layer and a 0.6 m subsoil layer, into the lysimeters. The calcareous sand, which had no distinct soil horizons, was transferred in one layer. Soil properties are given in Table 1. To mitigate the effects of soil disturbance such as nutrient losses, oats and barley were cropped in the summer 1994. In October 1994, one model ecosystem was established in each of the 32 lysimeters (16 per soil type). Each model ecosystem was composed of eight beech and eight spruce trees with five typical non-woody forest understory species (*Carex sylvatica*, *Geum urbanum*, *Ranunculus ficaria*, *Viola sylvatica* and *Hedera helix*). At planting, beech trees were 2–3 years and spruce trees 4 years old. Trees had been grown from seeds collected from selected provenances (beech) and from clonal cuttings (spruce) (details see Egli et al., 1998).

Table 1. Selected properties of the soil prior to the experiment

	pH (CaCl ₂)	CO ₃ -C ^a g (kg soil) ⁻¹	C _{org.}	N _{tot}	CEC _{eff} ^b mmol _c (kg soil) ⁻¹	base saturation %	P (Olsen) ^c mg (kg soil) ⁻¹	sand ^d	silt ^d	clay ^d
								%	%	%
Acidic loam (0–40 cm)	4.11	< d.l.	12.9	0.83	42	40	2.3	55	29	16
Acidic loam (40–100 cm)	3.80	< d.l.	2.3	0.30	60	50	< d.l.	55	27	18
Calcareous sand	7.16	19.0	13.1	0.76	127	100	6.0	84	18	6

^a: determined according to Nelson (1982).

^b: extracted with 1 N NH₄Cl.

^c: measured at the end of the experiment.

^d: from Sonnleitner et al. (2000).

d.l.: detection limit.

Soil and drainage water

Soil solution was sampled in 1998 during the fourth and last growing season of the experiment. Two suction cups (SoilMoisture Equipment Corp., Ca) per lysimeter were installed at each of the three depths of 5, 25 and 50 cm in March 1998. Prior to their installation, all suction cups were flushed first with 1 N HCl, then with distilled water and finally with soil solution. The suction cups were connected to a 100 cm³ glass bottle and were evacuated with 30 kPa twice a week. Soil solution samples were collected from the end of April to mid August and pooled every 2 weeks for each depths in each lysimeter. Drainage water was collected during the entire 4 years of treatment. Each week, the drainage water was pumped out of the sampling container of each lysimeters and was pooled over 3 weeks. Soil water content was measured weekly by time domain reflectometry (TDR). TDR probes of 25 cm length were installed at 25, 50 and 75 cm depth. Three TDR probes per depth were used in one lysimeter of each CO₂ × N treatment, while one probe was taken for the other lysimeters. TDR signals were calibrated and analyzed according to Roth et al. (1989).

Soil samples from 0 to 10 cm depth were collected with 6 cores (diameter 3 cm) from each lysimeter. Samples from 10 to 15, 35 to 50, 50 to 60 and from 90 to 100 cm depth were taken with a soil corer (diameter 5 cm, two samples per lysimeter).

Ion exchange resin (IER) bags

IER bags effectively absorb plant available anions and cations from the soil and are a relatively nondestructive method to estimate N availability *in situ* (e.g. Binkley and Matson, 1983). In order to study the influence of tree species on N availability, in each lysimeter,

two IER bags were installed at a distance of 10 cm from spruce stems, two bags 10 cm from beech stems and two bags in the middle between spruce and beech trees. Resin bags were prepared by filling cylindrical nylon mesh (10 cm long, 1 cm in diameter, 0.6 mm mesh) bags with a mixed-bed IER (DOWEX 50WX4, H⁺-form; DOWEX 1X8, Cl⁻-form saturated with OH⁻). The IER bags were closed with stainless steel staples and labeled with a cord. In March 1998, the bags were inserted into a precut slit at a 60° angle to a depth of 3 cm. All IER bags were recovered mid August 1998. To remove soil particles, the resin bags were washed with distilled water. Absorbed N was extracted with 150 ml 1 N KCl for 12 h on the day after recovery.

N₂O-fluxes

Static chambers were used to measure N₂O-fluxes. Each chamber, one per lysimeter ($n=32$), consisted of a PVC tube (30 cm length, 30 cm diameter), which was pushed 10 cm into the soil during the winter before the last growing season. Fluxes of N₂O were measured every 14 days. For each measurement, the chambers were sealed with a saran foil. The headspace in the chambers was sampled with a 250 ml syringe through a septum 0, 45 and 90 min after closure. The gas samples were injected into a Hungate tube. Concentrations of N₂O were measured with a gas chromatograph (Fractovap, model 8500) equipped with a ⁶³Ni electron capture detector. Finally, the N₂O flux was calculated from the slope of the N₂O concentration increase in the chambers.

Net N uptake in tree biomass

After 4 treatment years, the standing biomass was harvested in September 1998. Tree compartments (coarse

Figure 1. Experimental set-up of a spruce-beech model ecosystem. (a) Profile view of an open-top chamber, containing two lysimeters with an acidic loam and a calcareous sand. (b) Plan view of the spatial arrangement of trees and soils in the open-top chambers.

roots, stems, twigs and foliage) were collected separately from individual trees, dried at 60 °C and ground. Fine roots biomass of each lysimeter was sampled with 3 soil cores of 3.5 cm in diameter down to a soil depths of 40 cm.

Laboratory analyses

Determination of NO_3^- in the resin extracts was conducted photometrically at 210 nm according to Norman and Stucki (1981) using an UV-160 spectrophotometer (Shimadzu Corp., Kyoto, Japan). Samples of soil solution and drainage water were collected and stored in acid-washed polyethylene bottles, and passed through 0.45 μm cellulose-acetate filters (Schleicher&Schuell, ME25) within the next 48 h. Subsamples were acidified to 2.5% HNO_3 by volume for cation analyses. Water samples were analysed for major cations (Ca^{2+} , Mg^{2+} , Na^+ , K^+) by induced-coupled plasma atomic emission spectrometry (ICPAES OPTIMA 3000, Perkin Elmer, Norwalk, CT) and anions (Cl^- , NO_3^- , SO_4^{2-} , H_2PO_4^-) by ion chromatography (DX-120, Dionex, Sunnyvale, Ca). Ammonium in both resin extracts and water samples was measured colorimetrically by automated flow injection analysis (PE FIAS-300, Perkin-Elmer). For extractable C measurements, 25 g fresh soil were

extracted with 100 ml of KCl for 1 h and then filtered (Schleicher&Schuell 589/3). Extractable C and dissolved organic carbon (DOC) in the soil solution were determined with a Shimadzu TOC-500 analyzer. Carbon and N concentrations of plant tissues and soils were measured with a C/N analyzer (NA 1500, Carlo Erba Instr., Milan, Italy).

Calculations and statistical analyses

To estimate the amount of N in the soil solution of the whole lysimeter at harvest, N concentrations were multiplied with measured water contents. Both N concentrations and water contents were linearly interpolated between the measured depths. From 50 to 150 cm depth, the water contents of 75 cm depth and the mean of the N concentrations of 50 cm depth and of the drainage water were taken for the estimation. Losses of N in drainage were summed up over the entire experimental period.

Treatment effects were identified by analyses of variance (ANOVA). Following the experimental design, the main effects of CO_2 and N and their interaction were tested against the 'chamber mean squares', while the main effects of soil type and its interactions with CO_2 , N, and $\text{CO}_2 \times \text{N}$ were tested against the 'lysimeter mean squares'. Effects of CO_2

Table 2. Effects of elevated CO₂ on selected properties of the soil at 0–10 cm depth and of the soil solution at 5 cm depth. Values are the mean of two N treatments (*n*=8)

	CO ₂	pH (CaCl ₂)	Ca ²⁺	Al ³⁺	K ⁺	NO ₃ ⁻	NH ₄ ⁺	DOC	Extractable C mg (kg soil) ⁻¹
			mg l ⁻¹						
Acidic loam	ambient	4.11	2.4	0.30	< d.l.	1.95	0.013	12.9	17.6
	elevated	4.22	2.1	0.26	< d.l.	0.71	0.008	13.7	15.7
Calcareous sand	ambient	7.16	51.4	0.01	0.09	13.6	0.038	9.1	12.0
	elevated	7.16	49.5	0.02	0.14	7.32	0.039	8.4	12.6
Significanc	soil type	0.0001	0.0001	0.0001	0.03	0.0001	0.0002	0.0001	0.0001
	CO ₂	n.s.	n.s.	n.s.	n.s.	0.02	n.s.	n.s.	n.s.

d.l.: detection limit.

and N were additionally tested in separate analyses for the acidic and the calcareous soil. Soil solution samples were first analysed by an overall ANOVA across all depths and then separately for each depth. The impact of the location of IER bags on absorbed N was tested against the 'tree per lysimeter mean squares' (= residual MS). Furthermore, the effects of CO₂, N, soil type, and their interaction were tested separately for each species. All statistical analyses were performed with SAS (SAS System 6.12, Cary, NC).

Results

Soil solution

As expected, soil pH and base cations in the soil and in the soil solution were significantly higher in the calcareous sand than in the acidic loam (Tables 1 and 2). Concentrations of NH₄⁺ and NO₃⁻ in the soil solution were increased in the calcareous sand compared to the acidic loam (*p*<0.001). This supports our initial assumption that the calcareous sand was representative for a nutrient-rich soil with a high N availability and the acidic loam for a nutrient-poor soil with a low N availability.

Inorganic N occurred almost completely as NO₃⁻. Ammonium concentrations were often below the detection limit of 0.01 mg NH₄⁺-N l⁻¹, which indicates that NH₄⁺ was rapidly removed from the soil solution; probably by sorption to negatively charged soil surfaces or by NH₄⁺ immobilisation. The two soil types showed a contrasting depth distribution of NO₃⁻ (Figure 2; interaction depth × soil, *p*<0.001). In the calcareous sand, NO₃⁻ concentrations increased down

to 50 cm depth, which demonstrates a net NO₃⁻ production (nitrification - [plant uptake + denitrification + immobilisation]). Contrary to this, concentrations of NO₃⁻ decreased with depth in the acidic loam, indicating a net immobilisation of NO₃⁻.

Elevated N deposition significantly increased NO₃⁻ concentrations in the soil water (*p*<0.05; pooled across depths). However, when the effect of N deposition was tested separately for each depth, it was significant only at 5 cm depth and not deeper in the soil. Concentrations of NH₄⁺ did not respond to N additions.

The enrichment with CO₂ for 4 years had no effect on concentrations of nutrients in the soil solution, except to decrease NO₃⁻ (*p*=0.002, pooled across all depths, Figure 2). However, the response of NO₃⁻ concentrations depended on the soil as indicated by a significant interaction between CO₂ and soil type (*p*=0.002). In the calcareous sand, CO₂ enrichment caused a strong reduction in NO₃⁻ concentrations at all depths (*p*<0.001). Under elevated CO₂, NO₃⁻ concentrations at 5, 25 and at 50 cm depth were 50%, 95% and 75% lower than at ambient CO₂, respectively. In the acidic loam, elevated CO₂ reduced NO₃⁻ concentrations at 5 cm depth (*p*<0.05), but not deeper in the soil. The interaction between CO₂ and N deposition was significant in the acidic loam (*p*=0.04) and not significant in the calcareous sand. This indicates that increased N inputs stimulated the negative response of NO₃⁻ to elevated CO₂ in the N-poor acidic loam, but not in the N-rich calcareous sand.

Drainage water

The export of inorganic N in drainage water occurred only as NO₃⁻. In the acidic loam, export of NO₃⁻ by drainage was close to the detection limit (Table 3), in-

Figure 2. Depth distribution of NO_3^- concentrations in the soil solution and in the drainage water. Means and standard errors of four replications per treatment averaged over the growing season 1998. Low N represents a N deposition of $0.7 \text{ g N m}^{-2} \text{ yr}^{-1}$, high N a N deposition of $7 \text{ g N m}^{-2} \text{ yr}^{-1}$. Note the different scales for NO_3^- -N concentrations in the acidic loam and in the calcareous sand.

dicating that N inputs were retained in the ecosystem. In contrast, NO_3^- export was high in the calcareous sand. At low N deposition and ambient CO_2 , drainage losses of N even exceeded the inputs during the last growing season.

As expected, treatment effects were similar to those observed for soil solution N (Table 3). In the calcareous sand, increased atmospheric CO_2 decreased the NO_3^- losses by approximately 90% at low N and by 70% at high N deposition. The substantial decrease of NO_3^- export under elevated CO_2 was a product of the significantly lower NO_3^- concentrations in the drainage water (–80% at low N and –60% at high N) and a reduced drainage (–25%) as a result of enhanced evapotranspiration through an higher tree biomass (Sonnleitner et al., 2000).

Denitrification

Fluxes of N_2O were low compared to the amount of N in the soil solution and that lost in the drainage (Table 3). In accordance with NO_3^- concentrations

in the soil solution, denitrification was significantly higher in the calcareous sand than in the acidic loam. Elevated N deposition increased N_2O fluxes significantly. CO_2 enrichment had no overall effect, but denitrification tended to be higher under high CO_2 in the acidic loam ($p=0.10$).

Nitrogen accumulation by resin bags

In contrast to N concentrations of the soil solution and of the drainage water, N accumulation by IER bags was higher in the acidic loam than in the calcareous sand ($p<0.001$; Figure 3). This was probably due to the finer texture of the acidic loam, because smaller pores increase the soil volume from which ions can be absorbed by the IER bags. In addition, a finer substrate creates a better contact of IER bags with soil material. Another reason could have been the higher soil water potentials and higher water contents in the acidic loam than in the calcareous sand (data not shown).

At high N deposition, IER bags captured more N than at low N inputs ($p<0.001$, pooled across all treat-

Table 3. N losses by drainage water during the growing season 1998 (until 1.September). Means and standard deviation of four lysimeters per treatment. The N addition was 0.7 and 7 g N m⁻²yr⁻¹ for the low and high N deposition, respectively

	N	CO ₂	Drainage g N m ⁻²	N ₂ O-flu g N m ⁻²
Acidic	low	ambient	0.04	0.008
Loam	low	elevated	0.02	0.009
	high	ambient	0.03	0.009
	high	elevated	0.02	0.010
Signif cance	CO ₂		n.s.	n.s. (0.10)
	N		n.s.	0.028
	CO ₂ × N		n.s.	n.s.
Calcareous	low	ambient	1.20	0.014
Sand	low	elevated	0.1	0.015
	high	ambient	3.17	0.026
	high	elevated	0.95	0.021
Signif cance	CO ₂		0.001	n.s.
	N		0.003	0.001
	CO ₂ × N		n.s.	n.s.

Table 4. Nitrogen concentrations of current-year foliage. Mean and standard deviations of four provenances, each with four replications

	N	CO ₂	Beech	Spruce
			% N	
Acidic	low	ambient	1.90	1.24
Loam	low	elevated	1.78	0.96
	high	ambient	2.11	1.45
	high	elevated	1.97	1.24
Signif cance	CO ₂		0.012	0.002
	N		0.0007	0.002
	CO ₂ × N		n.s.	n.s.
Calcareous	low	ambient	2.27	1.91
Sand	low	elevated	1.90	1.53
	high	ambient	2.26	1.96
	high	elevated	2.19	1.89
Signif cance	CO ₂		0.0004	0.03
	N		0.001	0.04
	CO ₂ × N		0.005	n.s. (0.11)

Figure 3. Accumulation of N by ion-exchange resin bags installed in a distance of 10 cm from spruce and beech stems. Means and standard errors of eight replication (two per lysimeter). Low N represents a N deposition of 0.7 g N m⁻² yr⁻¹, high N a N deposition of 7 g N m⁻² yr⁻¹.

Figure 4. Relation between N accumulation by ion exchange resin bags and current-year foliage biomass. Data points represent the mean of four replications.

ments and tree species). As observed for the soil solution, the impact of CO₂ was different for NH₄⁺ and NO₃⁻. NO₃⁻ accumulation declined in response to CO₂ enrichment ($p=0.006$, calcareous sand), whereas resin-absorbed NH₄⁺ showed no significant CO₂ effect. There was a significant interaction between atmospheric CO₂ concentrations and soil type ($p<0.01$; Figure 3). CO₂ enrichment reduced N accumulation by 45% in the calcareous sand ($p=0.007$), but it did not significantly influence N captured by IER bags in the acidic loam.

Accumulation of N by IER bags was approximately 20% lower under trees than in the middle between trees. However, this effect was not significant. The effect of CO₂ enrichment was different for IER bags under spruce and under beech (Figure 3). Under

spruce, NO₃⁻ captured by IER bags was lower at elevated CO₂ concentrations in both soil types ($p<0.05$). Under beech, accumulated N showed no overall response to CO₂, but a significant interaction between soil type and CO₂ ($p<0.05$). In the calcareous sand, CO₂ enrichment caused a reduction in N accumulation ($p<0.05$), while in the acidic loam, N accumulation did not differ between ambient and elevated CO₂ under beech ($p=0.20$).

Tree N

Nitrogen concentrations in leaves of beech and spruce were significantly lower in the acidic loam than in the calcareous sand ($p<0.001$; Table 4), which is consistent with the measured indices of soil N availability. Increasing the N input caused an increase in foliar

Figure 5. Storage of N in tree biomass (above- and below-ground) and N content in the soil solution and drainage water at harvest mid August. Soil water N content was estimated by multiplying N concentrations with water contents of the soil at harvest. N export in drainage was summed up over the entire 4 treatment years. Means and standard errors of 4 replications per treatment.

N concentrations in both soils. CO₂ enrichment decreased N concentrations of spruce and beech leaves significantly (Table 4). These decreases were not caused by dilutions with non-structural carbohydrates. At harvest, non-structural carbohydrates accounted for 4–7% of the dry weight only and CO₂-effects were negligible (W. Landolt, pers. comm.).

Total net uptake of N by trees was about 80% higher in the calcareous sand than in the acidic loam ($p < 0.001$; Figure 4). Elevated N deposition stimulated total tree N uptake in the acidic loam by 80%, but had no effect on the calcareous sand without CO₂ enrichment. Despite a significant increase in tree biomass at high CO₂ levels (+20%; D. Spinnler, pers. comm.), total net uptake of N by trees showed no overall significant CO₂ effect (Figure 4) as a result of the lower N concentrations of plant tissues at high CO₂. However, CO₂ enrichment stimulated total tree N uptake on the calcareous sand that received N fertilization ($p < 0.05$; Figure 4).

Discussion

Soil type determines the effects of elevated CO₂ on soil N availability

Atmospheric CO₂ enrichment generally reduced indices of N availability in the soil as indicated by lower N concentrations of the soil solution, by a reduced N accumulation of IER bags and by a decreased N export through drainage (Table 5).

This is in agreement with studies in agroecosystems and in Mediterranean grasslands, where elevated CO₂ caused greater N immobilisation and reduced NO₃⁻ leaching (Hungate et al., 1997, Torbert et al., 1996, 1998). However, our results contrast with studies in mesocosms and in the laboratory; in which increased NO₃⁻ leaching (Körner and Arnone, 1992) and N availability at high CO₂ levels were found as a result of a stimulated N mineralisation (Zak et al., 1993). In N-limited and nutrient-poor grasslands, Arnone (1997) detected no changes of N availability and Kandeler et al. (1998) found only minimal impacts on soil microbial processes under increased CO₂ concentrations. The reason for the contrasting effects of atmospheric CO₂ on N availability may be the soil type used and/or the degree of disturbance of the soil

Table 5. Summary table of CO₂ effects on indices of N availability

	Acidic loam		Calcareous sand	
	low N	high N	low N	high N
NO ₃ ⁻ in soil solution (5 cm depth)	-20%	-69% *	-66% **	-46% *
NH ₄ ⁺ in soil solution (5 cm depth)	+100%	-55%	-18%	-18%
N in drainage	-45%	-28%	-92% *	-70% **
NH ₄ ⁺ accumulation by IER bags	-2%	-8%	-13%	-47% *
NO ₃ ⁻ accumulation by IER bags	+3%	+7%	-33% *	-45% *
total N of the soil (0-10 cm)	+6%	+4%	-1%	+3%
N uptake by trees	+3%	-2%	+3%	+19% **

p*<0.05; *p*<0.01

(Arnone, 1997, Johnson et al., 1995). Studies which have reported relatively large effects of elevated CO₂ (positive or negative) were conducted in highly disturbed artificial or nutrient-rich systems. Low impacts of increased CO₂ on N availability and microbial processes were usually found in nutrient-poor soils. The results of our experiment, using a nutrient-rich and a nutrient-poor soil with different N inputs, underlines the importance soil type plays in the response of N availability to elevated CO₂. The decreases of N concentrations in the soil solution, N export in drainage and N accumulated by IER bags through increased CO₂ concentrations were large in the nutrient-rich and N saturated calcareous sand, but low in the nutrient-poor and N-limited acidic loam (Table 5). Increasing the N deposition reduced some of the differences between the two soil types. In the acidic N-poor loam, the high N inputs increased the effects of elevated CO₂ on N concentrations in the soil solution, whereas in the N-rich calcareous sand, the effect was independent of the N addition rate.

Effects of tree species

Our data show that the effect of CO₂ enrichment on indices of soil N availability did not solely depend on the soil type. It was also influenced by tree species. Under spruce, N accumulation by IER bags was significantly reduced at high CO₂ in both soil types (Figure 3). Under beech, the increased CO₂ concentrations caused a significant decrease of accumulated N in the calcareous sand and a statistically not significant increase in the acidic loam. Tree biomass was opposite to this: Spruce profited from the CO₂ enrichment in both soil types, while beech responded negatively to elevated CO₂ in the acidic loam and positively in the calcareous sand (D. Spinnler, pers. comm.; data

from 1996/97 see Egli et al., 1998). The significance of tree species for the CO₂ effects on soil N availability is consistent with experiments in agro-ecosystems, where the response of NO₃⁻ leaching and N immobilisation to elevated CO₂ differed between crop species (Torbert et al., 1996, 1998). In our study, N accumulation by IER bags was negatively correlated with current-year foliage biomass (Figure 5), which suggests that the negative impact of elevated CO₂ on soil N availability indices was caused by a stimulated tree growth under CO₂ enrichment. Consistently, mean N concentrations of the soil solution were negatively correlated with total tree biomass in the calcareous sand (*r* = -0.73, *p* < 0.001). In the acidic and N-limited loam with low CO₂-effects, no significant correlation existed between tree biomass and N concentrations in the soil solution.

Potential reasons for lower soil N availability under elevated CO₂

Direct effects of increased above-ground CO₂ concentrations on N transformation processes in the soil are unlikely, due to the high pCO₂ in the soil atmosphere (Paterson et al., 1997; Santruckova and Simek, 1997). There are several potential indirect reasons for the negative impact of CO₂ on soil N availability: (1) reduced N-mineralisation rates because of greater C/N ratios of decomposing litter and root residues (Torbert et al., 1998); (2) increased storage of N in plant biomass (Comins and McMurtrie, 1993); (3) increased rhizodeposition, which stimulates N immobilisation by soil microbes through the provision of an easily available C-source (Cheng and Johnson, 1998; Van Ginkel and Gorissen, 1998).

Reduced mineralisation rate?

A reduced N release by slower litter decomposition was probably not quantitatively important. The soil solution data showed that the absolute and relative decrease of NO_3^- through elevated CO_2 was higher in the subsoil than in the topsoil of the calcareous sand (Figure 2). If a lower N release by a slower decomposing litter would have been the major reason for the reduced NO_3^- concentrations, then the CO_2 effect would have been largest in the uppermost soil layers closest to the litter and within the major rooting zone, but not in the subsoil. This conclusion is supported by the negligible impact of CO_2 enrichment on mass loss of foliar litter and on NO_3^- leaching in a laboratory experiment with beech litter collected from the lysimeters (Betschar, 1999).

Increased N uptake by trees?

The growth enhancement in response to elevated CO_2 (+20%; D. Spinnler, pers. comm.; data from 1996/97 see Egli et al., 1998) did not lead to a significantly greater N uptake (Figure 4), since N concentrations of plant tissues were lower under elevated CO_2 . In the calcareous sand, where the CO_2 effect was largest, net N uptake by trees increased by maximal 6.4 g N m^{-2} . The CO_2 -induced reduction in dissolved N of the soil compartment (soil solution + drainage water) at harvest 1998 was approximately 12 g N m^{-2} , when N concentrations in the collected soil solution were multiplied with measured water contents. This rough mass balance is consistent with the findings of an accompanying experiment, which showed that uptake of applied ^{15}N by trees was not significantly affected by atmospheric CO_2 (S. Maurer, pers. comm.). Thus, we suppose that a stimulated net N uptake by trees alone could not account for the strongly decreased N concentrations in the soil and drainage water.

Increased immobilisation of N in the soil?

As a consequence of a stimulated below-ground activity under elevated CO_2 , C availability in the soil is expected to be increased, which in turn may promote N immobilisation (Diaz et al., 1993, Hungate et al., 1997). In our forest model ecosystem, we have indications that below-ground activity was stimulated by higher atmospheric CO_2 . CO_2 enrichment increased the root biomass (acidic loam: +12%; calcareous sand: +34%) and the respiration rate (D. Spinnler, pers. comm.). However, a stimulated N immobilisation under elevated CO_2 is hardly detectable in the total soil

N pool, because the changes are too small compared to the large storage of N in the soil (Table 5).

CO_2 enrichment reduces nitrate leaching

In industrialized regions of the Northern Hemisphere, N deposition increases simultaneously with levels of atmospheric CO_2 . It was hypothesized that an increase in atmospheric CO_2 may decrease the susceptibility of forests to nitrogen saturation (Berntson and Bazzaz, 1996). In agroecosystems, Torbert et al. (1996) showed that CO_2 enrichment reduced NO_3^- leaching. In our experiment, N leaching losses from the spruce-beech model ecosystems in the calcareous sand were high, indicating N saturation according to the definition of Aber et al. (1989). Leaching of N from these model ecosystems and concentrations of N in the soil solution were substantially decreased by elevated atmospheric CO_2 . In the acidic loam, NO_3^- leaching in drainage was negligible during the last treatment year (Table 3). In the soil solution of the acidic loam in 5 cm depth, however, elevated N deposition stimulated the CO_2 -induced decrease of NO_3^- concentrations (Table 5). These findings support the hypothesis of Berntson and Bazzaz, (1996) that increased atmospheric CO_2 concentrations counteract N saturation by increasing the capacity of ecosystems to retain N deposition. However, our model ecosystem consisted of a juvenile spruce-beech forest. The reduction of N concentrations in the soil solution and in the drainage were linked either to an increased tree growth or to a higher C availability in the rhizosphere. Thus, it is questionable if a comparable decrease in N leaching will occur in mature forests or in forest ecosystems that are limited by other nutrients.

Conclusions

In the spruce-beech model ecosystem, concentrations of N in the soil solution and the export of N in drainage were decreased at elevated atmospheric CO_2 . Our results show that the effect of elevated atmospheric CO_2 on N dynamics in the soil depend on the characteristics of the ecosystem such as soil type and tree species (Table 5). Reduction of N concentrations in the soil solution through increased CO_2 concentrations were large in the nutrient-rich calcareous sand and low in the nutrient-poor acidic loam. Under spruce, N accumulation by IER bags decreased at high CO_2 in both soil types, whereas under beech, this decrease

occurred only in the calcareous sand. Increasing the N deposition stimulated the negative impact of CO₂ enrichment on soil solution N in the acidic loam, but not in the calcareous sand. Our results suggest that CO₂ enrichment decreases soil N availability in forest ecosystems, which may restrict CO₂-induced growth enhancements.

The finding that elevated atmospheric CO₂ reduced NO₃⁻ leaching indicate that elevated atmospheric CO₂ increased the capacity of ecosystems to retain N deposition. Thus, rising atmospheric CO₂ has the potential to counteract N saturation and to lower the risk of groundwater contamination with NO₃⁻.

Acknowledgements

This experiment was designed by Ch. Körner, J.B. Bucher and W. Landolt as part of the Swiss contribution to COST 614 under the coordination of CH. Brunold. The CO₂ enrichment was maintained by U. Bleuler and W. Landolt. We are grateful to M. Bundt, P. Egli and to two anonymous reviewers for helpful discussions and carefully reading the manuscript. Furthermore, we thank the central laboratories of the WSL for performing the anion and cation analyses. Funding was provided by the Board of the Swiss Federal Institute of Technology.

References

- Aber J D, Nadelhoffer K J, Steudler P and Melillo J M 1989 Nitrogen saturation in northern forest ecosystems. *BioScience* 39, 378-386.
- Arnone III J A 1997 Indices of plant N availability in an alpine grassland under elevated atmospheric CO₂. *Plant Soil* 190, 61-66.
- Berntson G M and Bazzaz F A 1996 Belowground positive and negative feedbacks on CO₂ growth enhancement. *Plant Soil* 187, 119-131.
- Betschar D 1999 Effekt von Asseln auf Abbau von Laubstreu verschiedener Qualität. Diploma Thesis. University Basel. 48 p. (unpublished).
- Binkley D and Matson P 1983 Ion exchange resin bag method for assessing forest soil nitrogen availability. *Soil Sci. Soc. Am J.* 47, 1050-1052.
- Boerner R E J and Rebbeck J 1995 Decomposition and nitrogen release from leaves of three hardwood species grown under elevated O₃ and/or CO₂. *Plant Soil* 170, 149-157.
- Canadell J G, Pitelka L F and Ingram J S I 1996 The effects of elevated [CO₂] on plant - soil carbon below-ground: A summary and synthesis. *Plant Soil* 187, 391-400.
- Cheng W and Johnson D W 1998 Elevated CO₂, rhizosphere processes, and soil organic matter decomposition. *Plant Soil* 202, 167-174.
- Comins H N and McMurtrie R E 1993 Long-term response of nutrient limited forests to CO₂ enrichment: Equilibrium behaviour of plant-soil models. *Ecol. Appl.* 3, 666-681.
- Cotrufo F M, Ineson P and Scott A 1998 Elevated CO₂ reduces the nitrogen concentration of plant tissues. *Global Change Biol.* 4, 43-54.
- Diaz S, Grime J P, Harris J and McPherson E 1993 Evidence of a feedback mechanism limiting plant response to elevated carbon dioxide. *Nature* 364, 616-617.
- Egli P, Maurer S, Günthardt-Goerg M and Körner C 1998 Effects of elevated CO₂ and soil quality on leaf gas exchange and above-ground growth in beech-spruce model ecosystems. *New Phytol.* 140, 185-196.
- FAO 1990 FAO-UNESCO Soil map of the world. Food and Agricultural Organization. Rome. Italy. 119 p.
- Gorissen A 1996 Elevated CO₂ evokes quantitative and qualitative changes in carbon dynamics in a plant/soil system: mechanisms and implications. *Plant Soil* 187, 289-298.
- Hungate B A, Lund C P, Pearson H L and Chapin III F S 1997 Elevated CO₂ and nutrient addition alter soil N cycling and N trace gas fluxes with early season wet-up in a California annual grassland. *Biogeochemistry* 37, 89-109.
- Johnson D W, Walker R F and Ball J T 1995 Lessons from lysimeters: soil N release from disturbance compromises controlled environment study. *Ecol. Appl.* 5, 395-400.
- Kandeler E, Tschirko D, Bardgett R D, Hobbs P J, Kampichler C and Jones T H 1998 The response of soil microorganisms and roots to elevated CO₂ and temperature in a terrestrial model ecosystem. *Plant Soil* 202, 251-262.
- Körner C and Arnone III J A 1992 Responses to elevated carbon dioxide in artificial tropical ecosystems. *Science* 257, 1672-1674.
- Landolt W, Bucher J B, Schulín R, Körner C and Brunold C 1998 Effects of elevated CO₂ concentration and N deposition on spruce-beech model ecosystem. In *Impacts of Global Change on Tree Physiology and Forest Ecosystems*. Eds Mohren G M J, Kramer K, and Sabaté S. pp 317-324. Kluwer Academic Publisher, Dordrecht.
- Nelson R E 1982 Carbonate and gypsum. In *Methods of Soil Analysis, Part 2: Chemical and microbial properties*. Ed. AL Page. pp 181-197. Agronomy 9. ASA-SSSA Publishers. Madison, WI, USA.
- Norman R J and Stucki J W 1981 The determination of nitrate and nitrite in soil extracts by ultraviolet spectrophotometry. *Soil Sci. Soc. Am. J.* 45, 347-353.
- Paterson E, Hall J M, Rattray E A S, Griffiths B S, Ritz K and Killham K 1997 Effect of elevated CO₂ on rhizosphere carbon fl w and soil microbial processes. *Global Change Biol.* 3, 363-377.
- Roth K, Schulín R, Flühler H and Attinger W 1989 Calibration of time domain reflectometry for water content measurements using a composite dielectric approach. *Wat. Resour. Res.* 26, 2267-2273.
- Santruckova H and Simek M 1997. Effect of soil CO₂ concentrations on microbial biomass. *Biol. Fert. Soils* 25, 269-273.
- Schimel D S 1995 Terrestrial ecosystems and the carbon cycle. *Global Change Biol.* 1, 77-91.
- Schäppi B and Körner Ch 1996 Growth responses of an alpine grassland to elevated CO₂. *Oecologia* 105, 43-52.
- Sonnleitner M A, Günthardt-Goerg M S, Bucher-Wallin I K, Attinger W, Reis S and Schulín R 2000 Influence of soil type on the effects of elevated atmospheric CO₂ and N deposition on the water balance and growth of a young spruce and beech forest. *Wat. Air, and Soil Pollut. (In press)*.
- Torbert H A, Prior S A, Rogers H H, Schlesinger W H, Mullins G L and Runion G B 1996 Elevated atmospheric carbon dioxide

- in agroecosystems affects groundwater quality. *J. Environ. Qual.* 25, 720-726.
- Torbert H A, Prior S A, Rogers H H and Runion g B 1998 Crop residues decomposition as affected by growth under elevated atmospheric CO₂. *Soil Sci.* 163, 412-419.
- Van Ginkel J H and Gorissen A 1998 In situ decomposition of grass roots as affected by elevated atmospheric carbon dioxide. *Soil Sci. Soc. Am. J.* 62, 951-958.
- Wullschleger S D, Post W M and King AW 1995 On the potential for a CO₂ fertilization effect in forest trees. An assessment of 58 controlled-exposure studies and estimates of the biotic growth factor. In *Biotic Feedbacks in the Global Climatic System: Will the Warm Feed the Warming?* Eds Woodwell G M, Mackenzie. pp 85-107. Oxford University Press, Oxford.
- Zak D R, Pregitzer K S, Curtis P S, Teeri J A, Fogel R and Randle D L 1993 Elevated atmospheric CO₂ and feedback between carbon and nitrogen cycles. *Plant Soil* 151, 105-117.

Section editor: R Aerts