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We assessed the effects of doubling atmospheric CO_2 concentration, [CO_2], on C and N allocation within pedunculate oak plants (*Quercus robur* L.) grown in containers under optimal water supply. A short-term dual $^{13}CO_2$ and $^{15}NO_3^-$ labelling experiment was carried out when the plants had formed their third growing flush. The 22-week exposure to 700 µl l^{-1} [CO_2] stimulated plant growth and biomass accumulation (+53% as compared with the 350 µl l^{-1} [CO_2] treatment) but decreased the root/shoot biomass ratio (–23%) and specific leaf area (–18%). Moreover, there was an increase in net CO_2 assimilation rate (+37% on a leaf dry weight basis; +71% on a leaf area basis), and a decrease in both above- and below-ground CO_2 respiration rates (–32 and –26%, respectively, on a dry mass basis) under elevated [CO_2]. ^{13}C acquisition, expressed on a plant mass basis or on a plant leaf area basis, was also markedly stimulated under elevated [CO_2] both after the 12-h $^{13}CO_2$ pulse phase and after the 60-h chase phase. Plant N content was increased under elevated CO_2 (+36%), but not enough to compensate for the increase in plant C content (+53%). Thus, the plant C/N ratio was increased (+13%) and plant N concentration was decreased (–11%). There was no effect of elevated [CO_2] on fine root-specific ^{15}N uptake (amount of recently assimilated ^{15}N per unit fine root dry mass), suggesting that modifications of plant N pools were merely linked to root size and not to root function. N concentration was decreased in the leaves of the first and second growing flushes and in the coarse roots, whereas it was unaffected by [CO_2] in the stem and in the actively growing organs (fine roots and leaves of the third growth flush). Furthermore, leaf N content per unit area was unaffected by [CO_2]. These results are consistent with the short-term optimization of N distribution within the plants with respect to growth and photosynthesis. Such an optimization might be achieved at the expense of the N pools in storage compartments (coarse roots, leaves of the first and second growth flushes). After the 60-h ^{13}C chase phase, leaves of the first and second growth flushes were almost completely depleted in recent ^{13}C under ambient [CO_2], whereas these leaves retained important amounts of recently assimilated ^{13}C (carbohydrate reserves?) under elevated [CO_2].

Key words – Allocation, ^{13}C, elevated CO_2, gas exchange, growth, ^{15}N, oak, *Quercus robur*, stable isotope.

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Introduction

In C_3 plants, which are limited in photosynthesis by the ambient CO_2 concentration ([CO_2]), doubling atmospheric [CO_2] stimulates net CO_2 assimilation rates and whole plant carbon supply (Jarvis 1989, Stitt 1991, Ceulemans and Mousseau 1994). The metabolism and within-plant allocation of C and N may be modified by environmental parameters, but are closely related in all phases of plant development (Dickson 1989). Most of the knowledge substantiating functional models on C/N balance (Davidson 1969, Reynolds and Thornley 1982, Ingestad and Ågren 1991, Thornley 1995) has been obtained under stable present-day [CO_2]; only N availabil-

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ity in the soil was considered a variable factor. In the context of rising atmospheric [CO_2], the extent to which the plant C/N balance and the within-plant allocation of C and N will be modified by the greater plant C supply, remains largely undetermined (Kirschbaum et al. 1994, Luo et al. 1994).

Nitrogen absorption by trees and their associated ectomycorrhizal symbionts is affected not only by pH, temperature and mineral composition of the soil solution but also by carbohydrate supply to the roots (Bernardo et al. 1984). Nitrogen assimilation in trees, which occurs almost entirely in root tips (Martin and Botton 1993), is carbon- and energy-consuming (Salsac et al. 1987). Energy costs for NO_3^- reduction to NH_4^+ by nitrate reductase and nitrite reductase and transport or storage of assimilated nitrogen compounds (Dickson 1989) are met by the supply of reduced cofactors, ATP and carbon compounds. However, in fine roots, growth and metabolic processes compete for carbon. It could therefore be expected that the increased availability of photo-assimilates in fine roots brought about by elevated [CO_2] (Vivin et al. 1995) would stimulate N acquisition and assimilation, leading to increased N availability for plant growth (Luxmoore et al. 1986, Thomas et al. 1991, Billes et al. 1993, Rouhier et al. 1994).

The use of heavy stable isotopes is a powerful approach to assess C and N interactions (Deléens et al. 1994, Maillard et al. 1994), especially in the below-ground compartment. To our knowledge, such an approach has not been used so far in elevated CO_2 studies. In the present study, we investigated the effects of doubling atmospheric [CO_2] on the plant C and N allocation within oak seedlings (*Quercus robur* L.) grown in containers under optimal watering conditions. Dual $^{13}CO_2$ and $^{15}NO_3^-$ labelling was used to follow C and N acquisition and distribution within the plant. Results were analyzed with respect to functional models on C/N balance at the plant level.

Abbreviations – CER, carbon exchange rate; GE, growth efficiency; LAR, leaf area ratio; LMR, leaf mass ratio; RMR, root mass ratio; R/S, root/shoot ratio; SLA, specific leaf area; SMR, stem mass ratio.

Materials and methods

Plant culture

Acorns of pedunculate oak (*Quercus robur* L., provenance Manoncourt) were collected in Autumn 1993 in a parent stand close to Nancy (Lorraine, north-eastern France), soaked in fungicide (Rhodiasan, Rhône Poulenc, Paris, France) and overwintered at –1 °C in a plastic bag.

In March 1994, the acorns were peeled, soaked in water and sown in 5-l cylindrical plastic containers (20 cm deep) filled with a sphagnum peat-sand mixture (1:1, v/v). The substrate was fertilized with delayed release Nutricote 100 (NPK 13:13:13, + trace elements, NH_4:NO_3 1:1; 5 kg m^{-3}). Thirty containers were randomly placed in two transparent 50 µm thick polypropylene mini-greenhouses (30 m^3) located at INRA, Champenoux. Irradiance was about 60% of the outside conditions. Immediately after sowing, containers were exposed to either ambient (350 µl l^{-1}) or elevated (700 µl l^{-1}) [CO_2] and were watered twice a week to compensate for evapotranspiration. The CO_2 control and monitoring system as well as the growth conditions have been previously described by Guehl et al. (1994) and Vivin et al. (1995). Maximum irradiance at the plant level was about 1 200 µmol m^{-2} s^{-1}. Average daily temperatures were 26°C (maximum) and 11°C (minimum), and relative air humidity ranged between 40 and 70%.

Plant CO_2 exchange and labelling experiment

At the end of August (week 22 after sowing), 8 plants were randomly selected within each CO_2 treatment. These plants were placed in a semi-closed climatized chamber (23°C, 70% RH) and submitted to a short-term dual ^{13}C and ^{15}N labelling. The semi-closed labelling system, described in detail by Vivin et al. (1995), was designed (1) to measure carbon dioxide exchange in both above- and below-ground compartments of the plant-soil system and (2) to supply a constant ^{13}C-enriched CO_2 pulse to the shoots.

Carbon dioxide concentrations in these compartments were separately measured by two infrared gas analysers and monitored by a homemade regulation system (Vivin et al. 1995). Carbon dioxide was either added from an industrial CO_2 cyclinder to compensate for the shoot photosynthetic activity, or eliminated in a soda lime trap to compensate for the shoot dark respiration (night) and the below-ground CO_2 efflux. Three high-pressure sodium lamps (Philips SONT) provided a photosynthetic photon flux density of 350 µmol m^{-2} s^{-1} at the plant level, which was close to the mean irradiance level received by the plants in the mini-greenhouses. Gas exchange rates were calculated from the time course of [CO_2] as previously described (Vivin et al. 1995). The proportion of the below-ground respiration due to microbial respiration could not be assessed in this study.

The $^{13}CO_2$ enrichment of the aboveground compartment was obtained by the injection of $^{13}CO_2$ from a cylinder containing a $^{13}CO_2$ and N_2 mixture (1:99, v/v; Eurisotop, Saint-Aubin, France) in the main CO_2 supply flow (cylinder with industrial CO_2 at 1.08 atom% ^{13}C). The mixing ratio of the flows from the two cylinders was set so as to provide a labelling air with 1.5 atom% ^{13}C, that is ca 0.4% over the ambient atmospheric level. This labelling level was a good compromise between the precision in the determination of new and pre-existing C pools in the plants and the risk of contamination of the mass spectrometer (Deléens et al. 1994). In the aboveground plant compartment, $^{13}CO_2$ was supplied for 8 h from the beginning of the light period and its injection was stopped 4 h before the light was switched off, in order to eliminate possible interference with plant isotope

discrimination and to ensure a complete assimilation of $^{13}CO_2$ (Deléens et al. 1994).

Each plant received 100 ml of a 1 m*M* $K^{15}NO_3$ solution (Eurisotop, Saint-Aubin, France) at the beginning of the $^{13}CO_2$ labelling period, so that isotopic enrichment was about 2 atom% ^{15}N in the soil. Contrarily to the ^{13}C labelling, no distinct chase phase was applied for the ^{15}N labelling since the ^{15}N load could not be interrupted simultaneously to the cessation of the ^{13}C load.

Sampling and analysis

For each [CO_2], 3 plants were harvested at the end of the labelling phase (12 h); the 5 remaining plants within each [CO_2] treatment were harvested after a 60-h chase period (3 nights and 2 days). In addition, 3 unlabelled plants were harvested to obtain natural baseline ^{13}C abundances. Plants were separated into leaves, stems, coarse roots (comprising mainly the tap root) and fine roots (< 2 mm diameter). The leaf material from the 3 aerial growth flushes produced (flush 1 denotes the oldest flush) was separated. Leaf area was measured using a planimeter (ΔT Devices, UK). Plant organs were then oven dried at 60°C for 48 h, and ground to a fine homogeneous powder in a vibrating mill. Biomass partitioning among the plant components was assessed by determining the leaf mass ratio (LMR), the stem mass ratio (SMR), the root mass ratio (RMR) and the root/shoot ratio (R/S). Plant specific leaf area (SLA) and leaf area ratio (LAR) were calculated as the ratio leaf area/leaf dry weight and the ratio leaf area/plant dry weight, respectively. Growth efficiency (GE) was defined as stem dry weight divided by leaf area at harvest (Norby and O'Neill 1989).

Powdered plant tissues were combusted (3% O_2 + He, 1 050°C) and their C and N concentrations and isotope ratios of ^{13}C and ^{15}N (i.e. the molar ratios $^{13}C/^{12}C$ and $^{15}N/^{14}N$) were measured using an elemental analyser coupled with an isotope ratio mass spectrometer (Delta S, Finnigan-Mat, Bremen, Germany).

Calculations and statistical analysis

Absolute molar proportions (A) of the heavy isotope per 100 atoms, also called abundances, were calculated from the isotope ratios (R) as (Deléens et al. 1994):

$$A\% = \frac{R}{R+1} \times 100$$

In order to estimate the recently incorporated pools of heavy isotope relative to the pre-existing pools, A% results were converted into abundance excess according to the formula:

$$A\%\ \text{excess} = (A\%_l - A\%_c)$$

where subscripts l and c refer to samples from labelled and control plants, respectively. Moreover, recently assimilated ^{13}C and ^{15}N pools were calculated for the different plant components according to their dry masses and C and N concentrations:

$$\text{New } ^{13}C \text{ (or } ^{15}N\text{) content} = \frac{A\%\ \text{excess}}{100} \times \text{dry mass} \times \text{C (or N) concentration}$$

The partitioning of new ^{13}C and ^{15}N among plant components was determined as:

$$\text{Partitioning (\%)} = \frac{\text{New } ^{13}C \text{ (or } ^{15}N\text{) in the component}}{\text{New } ^{13}C \text{ (or } ^{15}N\text{) in the plant}} \times 100$$

Data were analyzed as a completely randomized design including two CO_2 treatments (350 or 700 µl l^{-1} [CO_2]) and 3 different labelling conditions (unlabelled controls, labelled plants at the end of the 12-h $^{13}CO_2$ pulse phase, labelled plants at the end of the 72-h labelling experiment) with 3 to 5 replicates. Statistical tests were carried out by using one-way or two-way analysis of variance.

Results

Growth, biomass partitioning, N concentration and C/N ratio

Doubling atmospheric [CO_2] modified the aerial growth dynamics of the plants. Growth periods, especially the third flush, were initiated earlier in plants grown under elevated [CO_2] (Fig. 1). Moreover, CO_2 enrichment significantly increased stem height, root collar diameter, the number of leaves per plant, leaf area of the third growing flush, but decreased SLA (Fig. 1, Tab. 1). The area of a single leaf was unaffected by CO_2 enrichment. Plant dry weight was 1.54-fold higher under elevated [CO_2] than under ambient [CO_2]. LMR and SMR were increased under elevated [CO_2], whereas RMR and R/S were decreased. Leaf area ratio was unaffected by [CO_2] and growth efficiency was increased by 32% under elevated [CO_2].

Elevated [CO_2] significantly decreased N concentrations and increased the C/N ratio in the oldest leaves (first and second flushes), coarse roots and whole-plant, but not in the stem, fine roots and expanding leaves (third flush) (Tab. 2). Leaves from the different flushes did not display differences in N content per unit area between the two [CO_2] (Tab. 2).

Carbon dioxide exchange

Diurnal CER of the above- and below-ground compartments was measured during the labelling period (week 22 after sowing). In the aboveground compartment, the daily mean value of net CER expressed per unit aerial dry mass was 1.37-fold higher under elevated [CO_2] than under ambient [CO_2] (Fig. 2). This stimulating effect of elevated [CO_2] on net CO_2 assimilation was even higher on a leaf area basis (+71%, data not shown). In contrast,

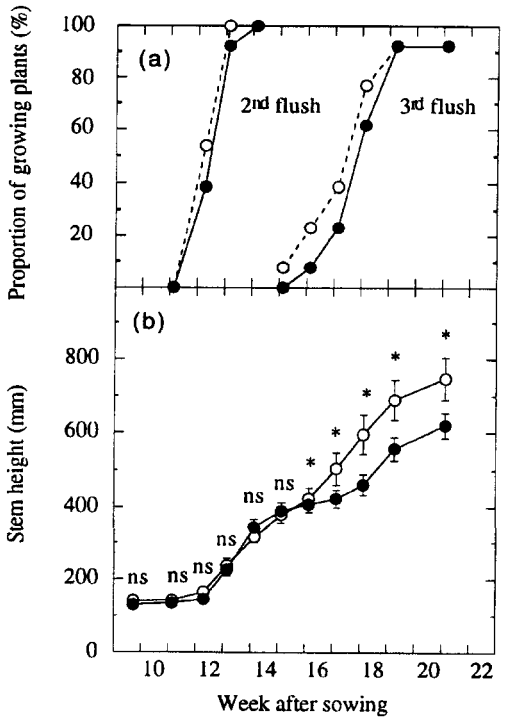

Fig. 1. (a) Time course of aerial elongation dynamics and (b) mean stem height (± SE) in *Q. robur* plants grown under two CO_2 concentrations. In (a) the proportion of plants elongating in a given flush is shown (n=13 to 16). Asterisks denote significant differences ($P<0.05$) among [CO_2] treatments for a given date; ns, non-significant.

nocturnal CER of the aerial compartment, as well as CER of the below-ground compartment, expressed on a mass basis, were about 30% lower under elevated [CO_2] than under ambient [CO_2] (Fig. 2).

On a whole-plant basis, diurnal CO_2 assimilation rate was 2.2 times higher under elevated than under ambient [CO_2] (data not shown). Nocturnal respiration of the whole aerial component was 1.2 times higher under elevated [CO_2] conditions than under ambient [CO_2]. The respiration rate of the below-ground component (root and microbial respiration) expressed on a plant basis, or on a soil area (or volume) basis, was also 1.3 times higher under elevated [CO_2] than under ambient [CO_2] (data not shown).

^{13}C uptake and within-plant allocation

The acquisition of ^{13}C, expressed either on a plant basis or on a whole-plant leaf area basis, was markedly stimulated under elevated [CO_2] (Tab. 3). The pools of recently photoassimilated ^{13}C, expressed per unit dry mass, considered either immediatly after the labelling, or after the 60-h chase phase, were significantly higher under elevated than under ambient [CO_2] in all plant components (Fig. 3). During the 60-h chase period, the recently photoassimilated ^{13}C was translocated from the leaves, in which the proportion of new C decreased, to

Tab. 1. Growth parameters in *Q. robur* plants grown under 350 or 700 µl l^{-1} CO_2 concentration for 22 weeks. The significance of CO_2 treatments is indicated for the different parameters ; NS, non-significant ; *, $P<0.05$ (n=13 to 16).

Parameter	CO_2 (µl l^{-1})		P
	350	700	
Root collar diameter (mm)	9.3	10.7	*
Stem height (mm)	619	775	*
Number of leaves per plant	34.0	39.9	*
Leaf area (dm^2)			
1st flush	2.1	2.0	NS
2nd flush	4.8	5.3	NS
3rd flush	2.6	5.9	*
Plant	9.5	13.2	*
Plant dry mass (g)	26.8	40.8	*
LMR (g g^{-1})	0.22	0.24	*
SMR (g g^{-1})	0.24	0.28	*
RMR (g g^{-1})	0.54	0.48	*
R/S (g g^{-1})	1.25	0.96	*
Fine root mass:plant mass (g g^{-1})	0.18	0.18	NS
SLA (dm^2 g^{-1})			
1st flush	1.50	1.25	*
2nd flush	1.55	1.26	*
3rd flush	1.86	1.40	*
Plant	1.61	1.32	*
LAR (dm^2 g^{-1})	0.36	0.32	NS
Growth efficiency (g dm^{-2})	66.9	88.3	*

Tab. 2. Nitrogen concentration, C/N ratio and whole plant C and N content in *Q. robur* plants grown under 350 or 700 µl l^{-1} CO_2 concentration for 22 weeks. The significance of CO_2 treatments is indicated for the different variables; NS, non significant; *, $P<0.05$ (n=13 to 16).

Parameter	CO_2 (µl l^{-1})		P
	350	700	
N concentration (mg g^{-1})			
Leaves 1st flush	37.7	29.5	*
Leaves 2nd flush	33.1	28.1	*
Leaves 3rd flush	26.5	24.5	NS
Stem	9.8	9.5	NS
Coarse roots	16.3	14.3	*
Fine roots (< 2 mm)	17.5	16.8	NS
Plant	18.6	16.5	*
Leaf N content (g m^{-2})			
1st flush	2.51	2.36	NS
2nd flush	2.14	2.23	NS
3rd flush	1.42	1.75	NS
Plant	2.02	2.07	NS
C/N ratio (g g^{-1})			
Leaves 1st flush	11.6	14.7	*
Leaves 2nd flush	14.1	15.9	*
Leaves 3rd flush	17.9	19.5	NS
Stem	45.1	46.9	NS
Coarse roots	26.5	30.5	*
Fine roots (< 2 mm)	23.3	25.0	NS
Plant	23.4	26.5	*
Plant C content (g $plant^{-1}$)	11.6	17.9	*
Plant N content (g $plant^{-1}$)	0.50	0.68	*

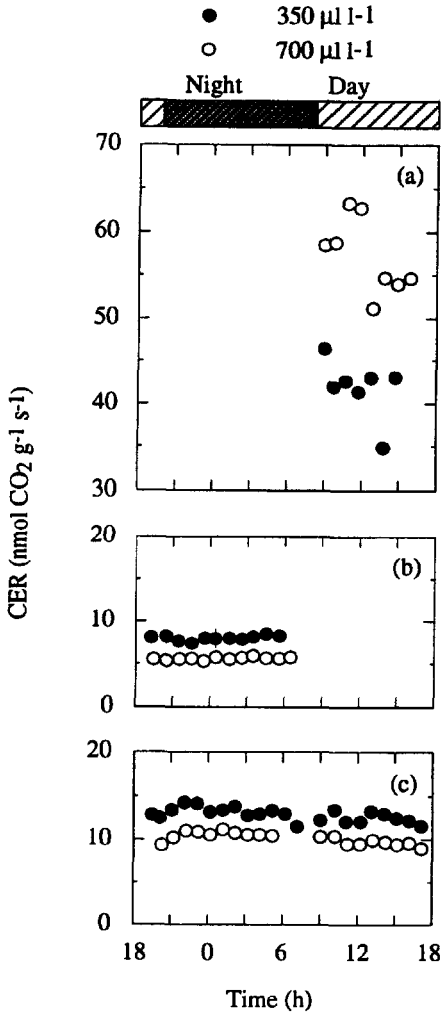

Fig. 2. Daily course of (a) net CO_2 assimilation, (b) aerial respiration and (c) below-ground respiration rates, expressed on a dry weight basis, in *Q. robur* plants grown under two CO_2 concentrations for 22 weeks. Data represent a single measurement on a set of 8 plants.

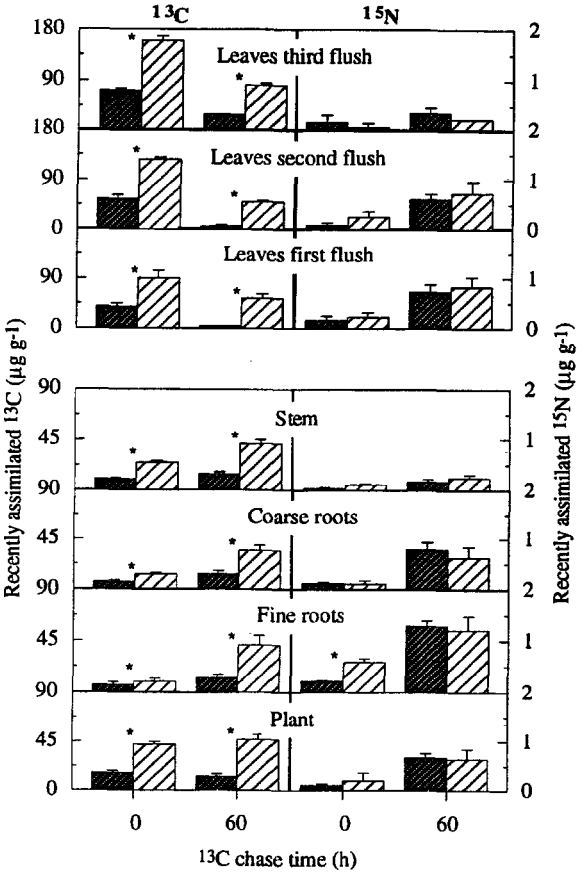

Fig. 3. Recently assimilated ^{13}C and ^{15}N proportions expressed on a dry mass basis, at the whole-plant level and in the different components of *Q. robur* plants grown under two CO_2 concentrations for 22 weeks. A short-term (total duration 72 h) dual labelling experiment was carried out. The different parameters were assessed at the end of the 12-h $^{13}CO_2$ pulse phase (0 h) and at the end of the ^{13}C chase phase (60 h). The ^{15}N load was not interrupted over the 72-h period. (▨), 350 µl l^{-1} CO_2; (▨), 700 µl l^{-1} CO_2. Asterisks denote significant differences ($P<0.05$) among $[CO_2]$ treatments for a given date (n=3 to 5).

the stem and roots, in which this proportion increased. Under ambient $[CO_2]$, the proportion of new ^{13}C was practically annulled after the 60-h chase phase in the leaves of the first and second flushes whereas the amount of new ^{13}C remained high under elevated $[CO_2]$. The increase in new ^{13}C during the chase phase was particularly pronounced in the fine roots under elevated $[CO_2]$. Elevated $[CO_2]$ led to a decrease in the short-term

Tab. 3. Plant ^{13}C and ^{15}N acquisition parameters in *Q. robur* plants grown under 350 or 700 µl l^{-1} CO_2 concentration for 22 weeks. A short-term (total duration 72 h) dual labelling experiment was carried out. The different parameters were assessed at the end of the 12-h $^{13}CO_2$ pulse phase (0 h) and at the end of the ^{13}C chase phase (60 h). The ^{15}N load was not interrupted over the 72-h period. The significance of CO_2 treatments and of time effects is indicated for the different variables; NS, non significant; *, $P<0.05$ (n=3 to 5).

Parameter	0 h		60 h		ANOVA (P<0.05)	
	350	700	350	700	CO_2	Time
Newly assimilated ^{13}C per plant (mg)	0.43	1.55	0.34	1.90	*	NS
Newly assimilated ^{13}C per unit leaf area (mg m^{-2})	5.4	13.3	3.5	15.5	*	NS
Newly assimilated ^{15}N per plant (µg)	4.01	4.77	17.7	27.1	*	*
Newly assimilated ^{15}N per unit fine root mass (µg g^{-1})	0.94	0.94	3.81	3.77	NS	*

^{13}C partitioning to the roots while the ^{13}C partitioning to leaves and the stem was increased (Fig. 4). Using biomass and C concentration measures to assess the long-term C partitioning to the different plant components, we also found lower values for the roots under elevated than under ambient $[CO_2]$ (Fig. 4). Elevated $[CO_2]$ did not significantly affect plant C concentrations (average value 43.3%).

^{15}N uptake and within-plant allocation

There was no $[CO_2]$ effect on specific ^{15}N uptake calculated as the pool of ^{15}N recently assimilated by the plant divided by the mass of fine roots (Tab. 3). The pools of receunaffected by $[CO_2]$ in all plant components but the fine roots (Fig. 3). In this latter component a transitory (12 h after the beginning of ^{15}N labelling) enhancement in the proportion of new ^{15}N was observed under elevated $[CO_2]$. Elevated $[CO_2]$ slightly increased the N partitioning to the aerial plant component both at the short-term (^{15}N data) and time-integrated (final biomass) scales (Fig. 4).

Discussion

At the end of the third aerial growing flush, the *Q. robur* plants still had markedly stimulated net CO_2 uptake rates under elevated $[CO_2]$ as shown by both the aerial CER (Fig. 2) and short-term ^{13}C acquisition data (Tab. 3).

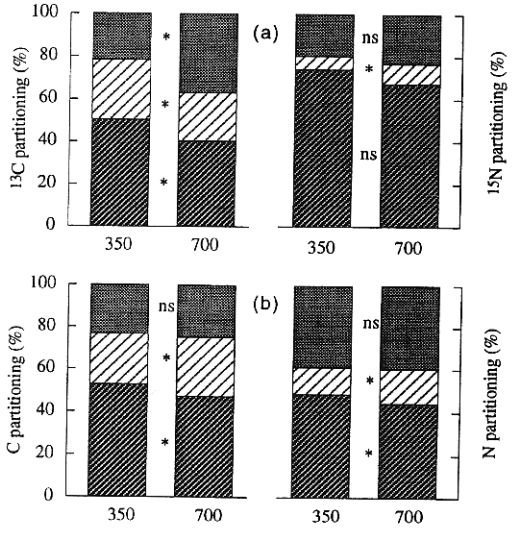

Fig. 4. (a) Short-term ^{13}C and ^{15}N partitioning values in the different components of *Q. robur* plants grown under two CO_2 concentrations for 22 weeks. A short-term (total duration 72 h) dual labelling experiment was carried out. Plants were harvested after a 12-h $^{13}CO_2$ pulse phase followed by a 60-h ^{13}C chase period (see Materials and methods). The ^{15}N load was not interrupted over the 72-h period. (b) Long-term biomass C and N partitioning. The significance of CO_2 treatments is indicated for the different variables; (ns) non-significant; (*) $P<0.05$ (n=3 to 12). (■) Leaf; (▨) stem; (▨) root.

These results are consistent with previous findings in *Quercus* species (Norby and O'Neill 1989, Sharkey et al. 1991, Gunderson et al. 1993, Picon et al. 1996) pointing to an absence of photosynthetic down-regulation during the aerial growth phase. Photosynthetic acclimation responses under elevated $[CO_2]$ have mainly been attributed to modifications in C source-sink relationships (Stitt 1991, Vivin et al. 1995) and have been related to the genetic capacity of plants to increase the size or the number of C sink organs in response to increasing $[CO_2]$ (Kaushal et al. 1989, Ceulemans and Mousseau 1994). The morphogenic plasticity vs $[CO_2]$ observed in the *Q. robur* plants (Fig. 1, Tab. 1) could be a crucial feature for the sustained stimulation of CO_2 assimilation rates. However long-term down-regulation of photosynthetic capacity induced by doubling $[CO_2]$ has also been reported in *Quercus* species (Bunce 1992) such as in *Q. robur* at the end of the growing season (Vivin et al. 1995, 1996).

Elevated $[CO_2]$ decreased both nocturnal shoot and below-ground respiration rates expressed on a dry mass basis, suggesting that apart from effects on photosynthesis, decreased respiration at high CO_2 may contribute to increased biomass accumulation of CO_2-enriched plants (Gifford et al. 1985). Microbial and plant components of below-ground respiration could not be disentangled here. Several studies have pointed to a stimulation of microbial respiration under elevated $[CO_2]$ due to increased C inputs in the soil (Körner and Arnone 1992, Norby et al. 1992). There is no general trend for the plant respiratory response to $[CO_2]$ (Amthor 1991, Poorter et al. 1992, Ziska and Bunce 1993, Bunce 1994, Wullschleger et al. 1994). Decreased shoot dark respiration has been related to lower maintenance respiration (Norby et al. 1986), especially when nitrogen concentration was decreased (Ryan 1991). The decreases in respiration rates we found could only be partly related to plant N status, since the decrease in shoot dark respiration rates, expressed on a N content basis, remained lower under 700 than under 350 $\mu l\ l^{-1}$ $[CO_2]$ (data not shown). Griffin et al. (1995) reached similar conclusions in North American pines. A direct inhibition of respiration by elevated $[CO_2]$ has been reported both for herbaceous (Azcon-Bieto et al. 1994) and woody (El Kohen et al. 1991) species. This inhibition has been attributed to a CO_2-induced regulation of the concentration and activity of some enzyme in the mitochondrial electron transport chain (Azcon-Bieto et al. 1994).

Original insights into within-plant C-N interactions in response to doubling $[CO_2]$ were provided by the dual ^{13}C- and ^{15}N- labelling approach. The proportion of recently assimilated ^{13}C was increased in all plant components under elevated $[CO_2]$ and particularly in fine roots. However this increased availability in C did not bring about a stimulation in ^{15}N acquisition as shown by the constancy among the two $[CO_2]$ of fine root specific ^{15}N uptake (Tab. 3) and of the proportion of new ^{15}N in the different plant components (Fig. 3). In the present study,

fertilization was determined theoretically so as to provide optimal N supply over the experimental period. However, we do not have measurements substantiating the absence of N supply limitations, particularly under elevated [CO_2]. Therefore it is not possible here to determine whether the absence of stimulation of ^{15}N acquisition is due to limiting N availability in the soil or to the absence of modulation of N assimilation by the C availability for metabolism. Further dual labelling experiments under controlled N supply conditions could provide decisive information to elucidate this question. Whatever the underlying mechanism, the uncoupling between ^{13}C and ^{15}N acquisition by the plant was in accord with the decrease in C/N ratio and N concentration at the plant and time integrated level (Tab. 2) as observed in other tree species (El Kohen et al. 1992, Billes et al. 1993, Conroy and Hocking 1993, Overdieck 1993).

Both short-term ^{13}C labelling and biomass results (Fig. 4) point to a decreased C partitioning to roots under elevated [CO_2], resulting in a decrease in the R/S ratio, although plant N concentration was significantly lower under elevated CO_2 concentration.

A similar result was observed in *Q. petraea* (Guehl et al. 1994) and *Q. alba* (Norby and O'Neill 1989), although in many studies with *Quercus* species (Sharkey et al. 1991, Bunce 1992, Bazzaz and Miao 1993, Gunderson et al. 1993, Lindroth et al. 1993), dry weight partitioning among the different plant components was not affected by the CO_2 concentration, under non-limiting nutrient availability.

The effects of [CO_2] on R/S are not in accord with the functional balance hypothesis of Davidson (1969) which predicts that increased shoot activity (photosynthesis) should always be balanced by increased relative C allocation to roots when the plant N concentration is decreased. However according to the model proposed by Luo et al. (1994), modifications in the R/S balance are determined by a number of adaptation mechanisms not necessarily involving a higher C partitioning to roots. No decrease in N concentration was observed in stems and in the metabolically active organs (i.e. growing fine roots or expanding leaves of the third growing flush) as already reported in *Castanea sativa* (Rouhier et al. 1994). Furthermore, in accord with the marked stimulation of CO_2 assimilation, there was no decrease in N content per unit leaf area under elevated [CO_2]. Thus, our results could reflect an optimization of the N distribution within the plant with respect to growth and photosynthetic capacity. This could be realized at the expense of the N pools in storage plant tissues as suggested by the lower N concentrations found in coarse roots and in leaves of the first and second growth flushes under elevated [CO_2] (Tab. 2). Furthermore, these leaves retained considerably higher amounts of recently assimilated ^{13}C under elevated than under ambient [CO_2]. Although no biochemical analyzes were performed, one may speculate that non-stuctural carbohydrates, especially starch, accumulated in these fully expanded leaves (Stitt 1991, Thomas and Strain 1991). Further investigations will be needed to assess plant responses over a longer period, particularly to determine changes in N vs C storage pools and their remobilization within the plant (Millard and Proe 1993). An altered C-N balance in the reserve pools accumulated in a given year might have consequences on plant growth in the following year.

Reduced N concentration in plant tissue induced by elevated [CO_2] have been suggested to result in lowered energetic construction costs (Griffin et al. 1993) or changes in chemical composition of the tissues (Lindroth et al. 1993). Our data do not support these conclusions, since N concentrations in growing organs were not affected.

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