24. X. D. Song, *Phys. Earth Planet. Inter.* **124**, 269 (2001).
25. Materials and methods are available as supporting material on *Science* Online.
26. F. Waldhauser, W. L. Ellsworth, *Bull. Seism. Soc. Am.* **90**, 1353 (2000).
27. X. D. Song, X. X. Xu, *Geophys. Res. Lett.* **29**, 10.1029/2001GL013822 (2002).
28. J. E. Vidale, P. S. Earle, *Nature* **404**, 273 (2000).
29. V. F. Cormier, X. Li, *J. Geophys. Res.* **107**, 10.1029/2002JB001796 (2002).

30. We thank the Data Management Center of the Incorporated Research Institutions for Seismology and the Alaska Seismic Network for making the data openly accessible. Digital data from the U.S. Atomic Energy Detection System stations were provided by the Air Force Tactical Applications Center. X.S. thanks J. Bass for discussion. This work was supported by the NSF and by the Natural Science Foundation of China. This is Lamont-Doherty Earth Observatory contribution number 6807.

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Materials and Methods
Figs. S1 to S11
Tables S1 to S5
References

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Carbon Flux and Growth in Mature Deciduous Forest Trees Exposed to Elevated CO_2

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Whether rising atmospheric carbon dioxide (CO_2) concentrations will cause forests to grow faster and store more carbon is an open question. Using free air CO_2 release in combination with a canopy crane, we found an immediate and sustained enhancement of carbon flux through 35-meter-tall temperate forest trees when exposed to elevated CO_2. However, there was no overall stimulation in stem growth and leaf litter production after 4 years. Photosynthetic capacity was not reduced, leaf chemistry changes were minor, and tree species differed in their responses. Although growing vigorously, these trees did not accrete more biomass carbon in stems in response to elevated CO_2, thus challenging projections of growth responses derived from tests with smaller trees.

How forest trees, the largest biomass carbon (C) pool on Earth, will respond to the continued rise in atmospheric CO_2 is unknown (*1*). Is there a potential for more growth, and perhaps more C storage, as a result of CO_2 fertilization (*2*)? Are trees in natural forests already carbon saturated, given that CO_2 concentrations have already reached twice the glacial minimum concentration (*3*)?

Experimental ecology has made important advances in recent years answering such questions, but unlike grass and shrub vegetation, adult forest trees do not fit any conventional test system with elevated CO_2. Free air CO_2 enrichment (FACE) is currently applied to fast-growing plantations (*4*–*7*), but to date, tall trees in a natural forest have not been studied because of overwhelming technical difficulties. We solved this problem with a technique called web-FACE (*8*) that releases pure CO_2 through a fine web of tubes woven into tree canopies with the help of a construction crane (Fig. 1) (*9*). Here, we present responses of 32- to 35-m-tall trees in a near-natural deciduous forest in Switzerland (*9*) to a 530 parts per million (ppm) CO_2 atmosphere over 4 years. Given the size and species diversity of the study trees, this project inevitably is tree- and not ecosystem-oriented, in contrast to other FACE projects, which use smaller trees. Because plant responses to CO_2 are species-specific (*10*, *11*), insight from single-species approaches remains limited, no matter how large the test scale. The statistical power of our approach is limited at the species level because of reduced intraspecific replication, but tree-ring analysis helps to account for much of the variation in tree vigor. The ultimate effect of rising CO_2 will remain concealed within our limited time scales, yet knowing the dynamics of tree responses over a number of years helps to estimate the nonlinear, longer-term trends. The project is thus a compromise between realism and precision, given that there is no way to maximize both (*12*).

Web-FACE uses CO_2 gas with a constant carbon isotope composition ($\delta^{13}C$) of $-29.7 \pm 0.3‰$ (mean ± SE of four annual means). Mixture with ambient-air CO_2 ($\delta^{13}C = -8‰$) results in a ^{13}C tracer signal in photoassimilates that was monitored at ~50 canopy positions with "iso-meters" (small containers planted with a grass using the C_4 photosynthetic pathway), which yielded a 4-year mean of 5.8 ± 0.5‰ ^{13}C tissue depletion. Once assimilated by leaves, the signal penetrates the various biomass compartments and allows one to track the fate of carbon. During the first season, leaves accreted 40% (*Quer-*

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Fig. 1. Canopy CO_2 release by a web of porous tubes (left), with "iso-meters" (front) and gas sampling lines (back) for control, all operated from a 45-m-high crane (right).

cus), 65% (*Fagus*), 67% (*Acer*), 79% (*Carpinus*), and 100% (*Tilia*) of new carbon as compared with the iso-meter mean (*9*). By the end of the first season, new assimilates contributed 71% and 73% to newly formed leaf and wood tissue across all trees (Fig. 2), suggesting very close coupling between leaf and tree-ring construction (*13*). Tree rings of *Quercus* and *Tilia* consisted almost completely of new carbon after the first year, whereas in *Fagus*, the tree-ring signals were weaker and lagged. By the end of the fourth year, 99% of all leaf mass and 91% of all wood mass consisted of new C. After 4 years the new <1-mm fine root fraction (*9*) reached 40% (2.3‰ ^{13}C depletion in 2004), suggesting a ~10-year fine root turnover, similar to the data for other deciduous forests (*14*, *15*).

Soil CO_2 sampled in 170 gas-wells at 3- to 11-cm soil depth across the crane area was 1‰ more depleted after 7 weeks in the treatment zone and 1.8‰ after 4 months, and remained at 2.2 ± 0.2‰ during the remaining test period, indicating a near steady-state efflux of new C (38% of total CO_2 emitted). Soil CO_2 concentrations were 25% higher in the elevated CO_2 area as compared with the control area (P = 0.03) in June 2001 and reached +44% in 2002. Mycorrhizal fungi carried a 64% new carbon signal already by the end of the first year (*16*). CO_2 enrichment thus rapidly produced isotopic fingerprints in the whole leaf-soil continuum. The strong soil-air signal indicates a rapid flux of new carbon through this system, and increased soil CO_2 concentrations provide clear evidence for enhanced metabolic activity in soils under CO_2-enriched trees (*16*), as was found in smaller test systems (*17*–*19*) and in four forest FACE studies (*4*, *20*).

We found no downward adjustment of photosynthetic capacity in response to growth in elevated CO_2 (*21*), similar to findings for other forest FACE experiments (*22*–*24*) but contrasting with those for some smaller test systems (*25*). Hence, it is not surprising that leaf nitrogen concentrations in the canopy were little affected (Table 1). Only *Carpinus* showed a significant mean reduction in leaf N by 15%, and across all trees the mean depletion was not significant when the dilution by nonstructural carbohydrates (NSC) was accounted for. *Quercus* and the subdominant taxa showed a significant +22% to +25% (P = 0.005) increase of NSC in leaves in elevated CO_2, similar to that for sweetgum in the Oak Ridge FACE (*23*). Across all trees, NSC increased by 12% (Table 1). NSC concentrations in branch wood were not significantly affected. The slight reductions in N and increases in NSC across trees were largely driven by the presence of two taxa, whereas the 5 to 8% reduction in specific leaf area (SLA) was not species-specific. *Fagus*, the only species with a periodic growth response (see below), did not show a CO_2 response in any of these traits.

Foliage per unit land area (leaf area index, LAI) and leaf duration did not exhibit consistent changes. Elevated CO_2 increased mean leaf duration for 2002 to 2004 in *Carpinus* and *Fagus* by 5 to 6 days and reduced it by 5 days in *Quercus* (marginally significant for the species × CO_2 interaction for *Quercus* and *Fagus*; P = 0.068). Annual litter production did not respond to CO_2 (Fig. 3). It indicates a constant LAI of ~5, in line with data for younger monocultures (*7*, *17*–*19*, *26*).

Total C and N in fresh fallen litter did not change significantly (*9*), but NSC concentrations (varying from 1.8 to 5.7%) rose, suggesting an overflow of photoassimilates channeled to the decomposer pathway (Table 2). Lignin concentrations ranging from 6.5 to 15% across species in ambient CO_2 were significantly reduced in elevated CO_2, largely driven by *Fagus* and the subdominant taxa. *Quercus* litter was hardly affected. These results indicate a shift in carbon fractions from recalcitrant to more labile compounds. Consequently, litter mass loss was enhanced after 220 days of in situ decay (largely driven by *Carpinus* and the subdominant taxa). In summary, litter became richer in starch and sugar, poorer in lignin, and decomposed faster, but the effect was species-specific. Ecosystem consequences will thus reflect species presence and abundance.

Using pretreatment radial growth rates of trees (tree-ring analysis) (*9*), we standardized basal stem area increments during the treatment period derived from girth tapes. *Fagus* showed a sharp growth stimulation in the first year (+ 92%, P = 0.026), whereas no effects were observed in the other taxa at any time (Fig. 4). The CO_2 response in *Fagus* became insignificant in 2002, but recovered in 2003 (P = 0.028) during a centennial heat wave during which elevated CO_2 improved plant water relations (*27*). The cumulative 4-year response of *Fagus* was only marginally significant at P = 0.07. We performed a second analysis for trees irrespective of species that yielded no significant basal area effect, because the *Fagus* signal was diluted by other species signals and, hence, the cumulative CO_2 signal for all trees in 2004 was zero (P = 0.79), suggesting C saturation (*3*, *28*). Tree-ring chronologies illustrate that the four treatment seasons included a pair of best and worst years over 9 years of growth data, partly explaining the greater variation. Tagged and revisited leading shoots in the top canopy showed no consistent length growth response across species and years (mean annual increment ~15 cm). Fruiting was un-

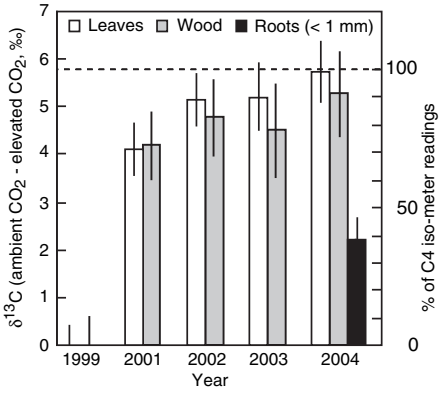

Fig. 2. New carbon in leaves, tree rings, and fine roots as assessed by ^{13}C tracer signals (means ± SE). The maximum steady-state carbon isotope difference of 5.8‰ between ambient and elevated CO_2 is represented by the iso-meter reference value (C_4 grass growing in the canopy, straight dashed line).

Table 1. Leaf nitrogen (N), nonstructural carbohydrates (NSC), and specific leaf area (SLA) across taxa for 13 trees growing in elevated CO_2 versus 16 control trees [repeated measures analysis of variance (ANOVA), with pretreatment values for each individual tree as covariable]; data are for 2001 to 2004, and means for early and late summer are pooled. Dry matter was used as a common reference, either including or excluding NSC.

Leaf trait	Difference (%)	P-value	Main driver
Leaf NSC	+12	0.007	Quercus, TAP*
N, total	−10	0.027	Carpinus, TAP
N, NSC-free	−7	0.136	Carpinus
SLA, total	−8	0.002	Nonspecific
SLA, NSC-free	−5	0.025	Nonspecific

*TAP is a mix of rare taxa of the genera *Tilia*, *Acer*, and *Prunus*.

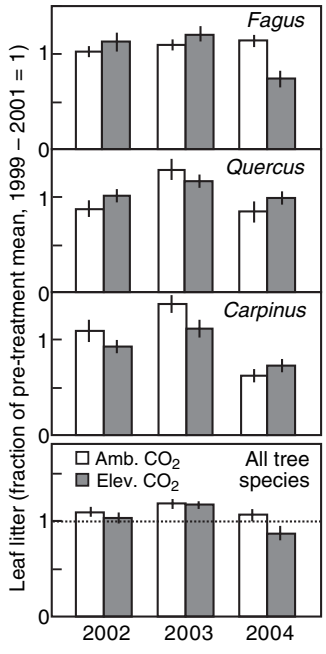

Fig. 3. Leaf litter production standardized (divided) by the mean for two pretreatment years and the year 2001 (*9*) for each litter trap (218 ± 9 g m^{-2} for 56 traps) to account for a priori spatial variability (means ± SE).

affected except for a gain in a single masting year in *Carpinus* (Fig. 5).

The data suggest no lasting growth stimulations by CO_2 enrichment in these mature trees after 4 years. It should be emphasized that the dominant study trees have reached only half of their average life span, have reached about two-thirds of their maximum size, and are growing vigorously by forestry standards (*9*). Other forest FACE experiments with younger trees have either shown a continuous growth stimulation (*Pinus taeda*) (*4*) or a transitory response, very similar to the one shown here for *Fagus* (*Liquidambar styraciflua*) (*5*). A lack of a response in stem growth or leaf litter production does not preclude a faster rate of below-ground (root) production, as was shown for the Oak Ridge FACE (*15*), but these are transitory C pools, which could translate into more recalcitrant forms of carbon in soil humus, mineral nutrients permitting.

Initially we asked whether forest trees will reduce their C uptake or trap more carbon in a CO_2-rich world. Our data suggest that they instead "pump" more carbon through their body. The few secondary effects found varied with species. Had we studied only single species, each would have led us to draw different conclusions: one species reducing its protein concentration, another consistently increasing its leaf carbohydrates, litter being affected in one group of species and not in others, and one species showing a transitory growth stimulation and others not. The Swiss forest FACE study thus points at the crucial role of tree species identity (*29*) and so far does not support expectations of greater carbon binding in tree biomass in such deciduous trees. The lack of a growth response or the transient response in one species is unlikely to be associated with N shortage (*30*), given the overabundance of N in this region (*9*). Broader stoichiometric constraints, soil microbial feedback (*18*, *31*), or counteracting ambient ozone (*6*, *32*) may hold answers. In summary, we find no evidence that current CO_2 concentrations are limiting tree growth in this tallest forest studied so far.

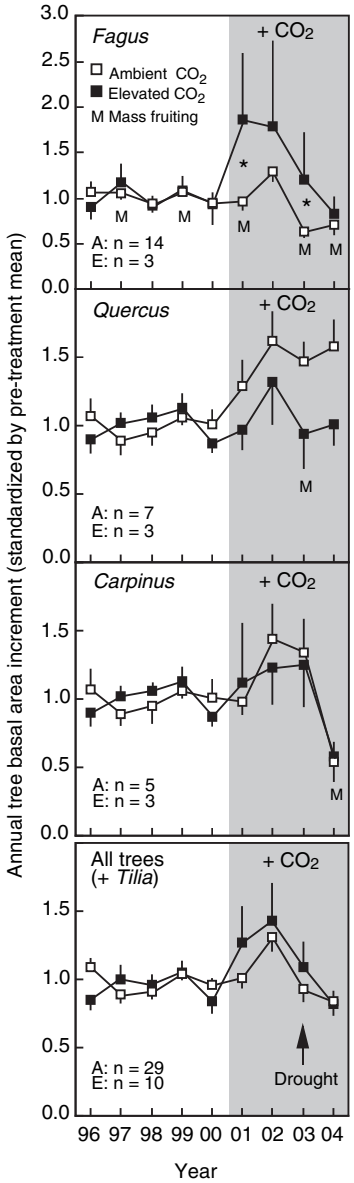

Fig. 4. Stem basal area (BA) growth of trees, standardized by the mean pretreatment BA increment for each tree. The two asterisks in *Fagus* indicate the only significant stimulation; 2003 was a drought summer; no significant $CO_2 \times$ year interaction in any species). The bottom diagram is for all trees that showed measurable growth, i.e., 10 in elevated CO_2 versus 29 controls (means ± SE).

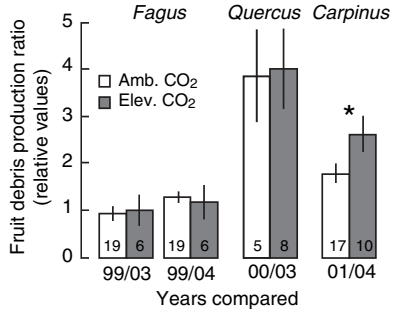

Fig. 5. Fruit debris collected with litter traps for three different tree species in years of mass fruiting. Pretreatment-to-treatment ratios for years with mass fruiting were used to account for spatial variability (numbers within bars for *n* traps). For *Fagus* we show the two latest mass fruiting episodes. The height of the bars denotes the degree of mass fruiting in the respective years ($P = 0.047$ for *Carpinus* in 2004; mean ± SE).

Table 2. Change in litter composition (% dry matter) as well as 220-day decomposition of fresh leaf litter collected in the canopy (two-factorial ANOVA with CO_2 and species as fixed factors).

Litter trait	Difference (%)	*P*-value	Main driver
Total C	+2	0.03	Nonspecific
Total N	−1	0.73	–
NSC*	+21	0.002	*Fagus*, TAP†
Lignin‡	−11	0.007	*Fagus*, TAP
Litter mass loss	+14	0.001	Carpinus, TAP

*Nonstructural carbohydrates (sugar, starch). †TAP is a mix of rare taxa of the genera *Tilia*, *Acer*, and *Prunus*. ‡Correcting for NSC-free litter mass changed the relative difference in lignin between CO_2 treatments from −10.7 to −10.0% ($P = 0.01$) but had no influence on the relative difference in nitrogen concentration.

References and Notes

1. R. J. Norby, K. Kobayashi, B. K. Kimball, *New Phytol.* **150**, 215 (2001).
2. J. G. Canadell, D. Pataki, *Trends Ecol. Evol.* **17**, 156 (2002).
3. C. Körner, *J. Ecol.* **91**, 4 (2003).
4. J. G. Hamilton *et al.*, *Oecologia* **131**, 250 (2002).
5. R. J. Norby, J. Ledford, C. D. Reilly, N. E. Miller, E. G. O'Neill, *Proc. Natl. Acad. Sci. U.S.A.* **101**, 9689 (2004).
6. J. G. Isebrands *et al.*, *Environ. Pollut.* **115**, 359 (2001).
7. B. Gielen *et al.*, *Glob. Change Biol.* **9**, 1022 (2003).
8. S. Pepin, C. Körner, *Oecologia* **133**, 1 (2002).
9. Materials and methods are available as supporting material on *Science* Online.
10. C. Körner, *Ecol. Appl.* **10**, 1590 (2000).
11. L. Tangley, *Science* **292**, 36 (2001).
12. C. Körner, in *Ecology: Achievement and Challenge*, M. C. Press, N. J. Huntly, S. Levin, Eds. (Blackwell Science, Oxford, 2001), pp. 227–247.
13. V. S. Chevillat, R. T. W. Siegwolf, S. Pepin, C. Körner, *Basic. Appl. Ecol,* in press.
14. J. B. Gaudinski *et al.*, *Oecologia* **129**, 420 (2001).
15. R. Matamala, M. A. Gonzalez-Meler, J. D. Jastrow, R. J. Norby, W. H. Schlesinger, *Science* **302**, 1385 (2003).
16. K. Steinmann, R. T. W. Siegwolf, M. Saurer, C. Körner, *Oecologia* **141**, 489 (2004).
17. C. Körner, J. A. Arnone III, *Science* **257**, 1672 (1992).
18. S. Hättenschwiler, C. Körner, *Oecologia* **113**, 104 (1998).
19. P. Egli *et al.*, *Oikos* **92**, 279 (2001).
20. J. S. King *et al.*, *Global Change Biol.* **10**, 1027 (2004).
21. G. Zotz, S. Pepin, C. Körner, *Plant Biol.* **7**, 369 (2005).
22. J. D. Herrick, R. B. Thomas, *Plant Cell Environ.* **24**, 53 (2001).
23. J. D. Sholtis, C. A. Gunderson, R. J. Norby, D. T. Tissue, *New Phytol.* **162**, 343 (2004).
24. L. Blaschke *et al.*, *Plant Biol.* **3**, 288 (2001).
25. P. Jarvis, Ed., *European Forests and Global Change* (Cambridge Univ. Press, Cambridge, 1998).
26. R. J. Norby, J. D. Sholtis, C. A. Gunderson, S. S. Jawdy, *Oecologia* **136**, 574 (2003).
27. S. Leuzinger, G. Zotz, R. Asshoff, C. Körner, *Tree Physiol.* **25**, 641 (2005).
28. G. Hoch, A. Richter, C. Körner, *Plant Cell Environ.* **26**, 1067 (2003).
29. I. T. Handa, C. Körner, S. Hättenschwiler, *Ecology* **86**, 1288 (2005).
30. R. Oren *et al.*, *Nature* **411**, 469 (2001).
31. D. R. Zak, K. S. Pregitzer, J. S. King, W. E. Holmes, *New Phytol.* **147**, 201 (2000).
32. W. M. Loya, K. S. Pregitzer, N. J. Karberg, J. S. King, C. P. Giardina, *Nature* **425**, 705 (2003).

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