

LETTER

Forest productivity under elevated CO₂ and O₃: positive feedbacks to soil N cycling sustain decade-long net primary productivity enhancement by CO₂

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Abstract

The accumulation of anthropogenic CO₂ in the Earth's atmosphere, and hence the rate of climate warming, is sensitive to stimulation of plant growth by higher concentrations of atmospheric CO₂. Here, we synthesise data from a field experiment in which three developing northern forest communities have been exposed to factorial combinations of elevated CO₂ and O₃. Enhanced net primary productivity (NPP) (*c.* 26% increase) under elevated CO₂ was sustained by greater root exploration of soil for growth-limiting N, as well as more rapid rates of litter decomposition and microbial N release during decay. Despite initial declines in forest productivity under elevated O₃, compensatory growth of O₃-tolerant individuals resulted in equivalent NPP under ambient and elevated O₃. After a decade, NPP has remained enhanced under elevated CO₂ and has recovered under elevated O₃ by mechanisms that remain un-calibrated or not considered in coupled climate–biogeochemical models simulating interactions between the global C cycle and climate warming.

Keywords

Elevated CO₂, elevated O₃, forest productivity, global C cycle, N-cycle feedbacks.

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INTRODUCTION

Coupled climate–biogeochemical models portray greater rates of terrestrial productivity on a CO₂-enriched Earth, which increase C storage on land, ameliorate the rate of anthropogenic CO₂ increase in the atmosphere and slow the pace of climate warming (Cramer *et al.* 2001; Friedlingstein *et al.* 2006). Although these simulations reveal the response of terrestrial ecosystems to elevated CO₂ (eCO₂) has important consequences for climate warming, there is considerable uncertainty regarding the extent to which greater rates of plant productivity will be sustained on a CO₂-enriched Earth. For example, biogeochemical feedbacks between C and nitrogen (N) cycles in terrestrial ecosystems, as well as other facets of climate change like elevated ground-level O₃ (eO₃), could constrain the degree to which eCO₂ enhances terrestrial net primary productivity (NPP), and hence, the degree to which the Earth's climate is projected to warm (Stitch *et al.* 2007; Thornton *et al.* 2007). This is especially true for northern temperate forests, which compose a globally important sink for atmospheric CO₂ and whose long-term enhancement of NPP by eCO₂ remains a critical gap in our knowledge of ecosystem response to global change (Norby *et al.* 2010; Drake *et al.* 2011). Presently, it is uncertain whether eCO₂ and eO₃ will interact to influence forest NPP, and how the response of forest NPP to these trace gases will be constrained, or not, by feedbacks to the terrestrial N cycle.

Future concentrations of ground-level ozone (O₃) could diminish the eCO₂ enhancement of NPP in forests as well as other terrestrial

ecosystems (Booker *et al.* 2009). Many portions of the Earth are already experiencing ground-level O₃ concentrations exceeding 40 nmol mol⁻¹, and concentrations are expected to reach 70 nmol mol⁻¹ throughout the Northern Hemisphere by 2100 (Stitch *et al.* 2007). Current and projected O₃ concentrations can damage the photosynthetic capacity of plants (Reich 1987), decrease plant productivity (Mauzerall & Wang 2001) and, in some cases, counter-balance the growth-enhancing effects of eCO₂ on forest productivity (King *et al.* 2005). Biogeochemical models incorporating the negative effects of eO₃ on terrestrial NPP suggest current concentrations could globally reduce NPP on land by *c.* 30–45%, even when the growth-enhancing effects of eCO₂ are considered (Felzer *et al.* 2005; Stitch *et al.* 2007). Reductions of this magnitude are projected to dramatically reduce C storage on land, allowing anthropogenic CO₂ to further accumulate in the atmosphere and accelerate climate warming (Stitch *et al.* 2007). Although plants differ in their sensitivity to ground-level eO₃ (Matyssek *et al.* 2010; Feng *et al.* 2011), few studies have addressed how these differences could, over time, influence the degree to which eO₃ diminishes terrestrial NPP, as well as the extent to which eCO₂ might counter this effect (Kubiske *et al.* 2007). Coupled climate–biogeochemical models are particularly sensitive to these dynamics, but they remain relatively uncalibrated regarding the response of different plants and plant communities to eO₃ (Stitch *et al.* 2007).

The degree to which eCO₂ stimulates productivity in northern temperate forests, as well as other terrestrial ecosystems, is also highly

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contingent on a series of biogeochemical feedbacks between C and N cycles, which control the growth-limiting supply of soil N to plants (Zak *et al.* 1993; Reich *et al.* 2006a; Norby *et al.* 2010; Drake *et al.* 2011). Some have argued that the greater production of plant litter under eCO₂, especially woody detritus, could act as a governor on the CO₂ enhancement of forest NPP by negatively impacting the terrestrial N cycle (i.e. progressive N limitation; Luo *et al.* 2004; Johnson 2006). Under such scenarios, the microbial decay of plant detritus slows, the accumulation of N from plant detritus into long-lived soil organic matter increases and the supply of growth-limiting soil N to plants declines. This series of events could dampen or eliminate the extent to which eCO₂ enhances NPP, having global implications for C storage on land as well as the future concentration of CO₂ in the atmosphere. Coupled climate–biogeochemical models that incorporate N cycle feedbacks on terrestrial NPP have demonstrated the sensitivity of climate warming to these C–N cycle interactions (Thornton *et al.* 2007); however, the magnitude and duration of this feedback in terrestrial ecosystems remains undefined as does how this feedback will be influenced by reductions in plant growth under eO₃. Recent syntheses provide evidence for increases, decreases and no change in the supply of N to plants growing under eCO₂ (de Graff *et al.* 2006; Reich *et al.* 2006b), implying that the eCO₂ enhancement of NPP could display ecosystem-specific responses; the combined effects of eCO₂ and eO₃ feedbacks to the terrestrial N cycle remain undocumented in the context of long-term, replicated field experiments. Resolving the uncertainty surrounding feedbacks between C and N cycles under eCO₂, and how they might be modified by eO₃, is necessary to refine estimates of future C sinks in terrestrial ecosystems as well as to predict the trajectory of climate warming.

Here, we report the results of a decade-long field experiment in which three developing northern forest communities have been exposed to factorial combinations of ambient and elevated CO₂ and O₃ concentrations. These communities were grown under free-air exposure technology (FACE) from 1997 to 2008, and we used tracer amounts of ¹⁵N applied to soil to follow the flow of N within them. Our objectives were to determine: (1) how future CO₂ and O₃ concentrations will interact to influence the productivity of contrasting forest communities, and (2) how N-cycle feedbacks will modify growth responses to these accumulating trace gases. We demonstrate that the eCO₂ enhancement of NPP has been sustained throughout the experiment via an acceleration of soil N cycling, and equivalent rates of NPP have occurred under ambient and eO₃ at the end of the experiment, despite an initial reduction in NPP by eO₃ (King *et al.* 2005). These observations provide new insight into the mechanisms sustaining NPP in forests experiencing atmospheric CO₂ and O₃ concentrations predicted to occur in the near future, and call for re-evaluating current conceptions regarding the timing and magnitude of feedback mechanisms on NPP as currently portrayed in coupled climate–biogeochemical models.

METHODS

Experimental design

The Rhinelander FACE experiment (49°40.5′ N, 89°37.5′ E; 490 m elevation) is composed of factorial CO₂ (ambient and 560 μmol mol⁻¹) and O₃ (ambient and 50–60 nmol mol⁻¹) treatments arranged in a split-plot, randomised complete block ($n = 3$) design. The

treatments are delivered in twelve 30-m FACE rings, each of which is divided into three plant communities. In 1997, one-half of each FACE ring was planted with trembling aspen (*Populus tremuloides*) genotypes of differing CO₂ and O₃ sensitivity (genotypes 8, 42, 216, 259, and 271); one-quarter of each ring was planted with a single aspen genotype (genotype 216) and paper birch (*Betula papyrifera*); the remaining ring quarter was planted with the same single aspen genotype and sugar maple (*Acer saccharum*). Aspen ramets and seedlings of birch and sugar maple were *c.* 3–4 months old at the time of planting. All plant communities were established at a density of one stem m⁻² (Dickson *et al.* 2000; Karnosky *et al.* 2003, 2005). As previously reported (Kubiske *et al.* 2007; Zak *et al.* 2007b), three aspen genotypes have responded positively to eCO₂ (genotypes 42, 216 and 271) and two genotypes have not (genotypes 8 and 259). Whereas, eO₃ caused growth declines in genotypes 271 and 216, compensatory growth increases in genotype 8 and no response in genotypes 42 and 259. Although aspen and birch both responded positively to eCO₂ and negatively to eO₃, birch increased growth to a greater extent under eCO₂ and decreased growth to a lesser extent under eO₃ than aspen (genotype 216; Kubiske *et al.* 2007; Zak *et al.* 2007a,b); we have not previously documented the response of aspen and maple growing in the mixed community.

In June 2003, each 30-m-diameter FACE ring was labelled with tracer quantities of ¹⁵N to follow the flow of N in the plant–soil system. Backpack sprayers were used to evenly disperse (0.034 L m⁻²) a dilute solution of ¹⁵NH₄Cl (99.98% ¹⁵N) over the forest floor. We applied ¹⁵NH₄⁺ to follow the microbial release of NH₄⁺ during litter decay into plants, the soil microbial community and soil organic matter. The isotope was applied at the rate of 15 mg ¹⁵N m⁻², which represented 3% of the inorganic N pool in mineral soil (0–10 cm depth). Immediately following application to the forest floor, 1.6 L m⁻² of water was applied to move the ¹⁵N into mineral soil.

Net primary productivity

In 2005, 2006, 2007 and 2008, the diameter of each tree in all FACE rings was measured, and the biomass of aboveground plant components (leaves, branches and stem) was estimated using allometric biomass equations developed from the destructive harvest in 2009. We developed separate equations to estimate the biomass of each genotype and species in our experiment. Production of aboveground litter was estimated using 0.15 m² baskets; the contents of 6–10 baskets in each plant community were collected bimonthly from August to November. Aboveground litter in each basket was composited over time, dried and weighed. Additionally, the aboveground biomass of herbaceous understory plants was collected from four 0.5 m² plots randomly located in each community; their fine roots could not be collected separately from those of overstory trees. The coarse roots of overstory trees were recovered from aspen ring halves in 1-m deep soil pits (2 m × 5 m); in the mixed community ring quarters, pits were 1-m deep and 2-m × 3-m in size. Coarse root biomass from 2005 to 2008 was estimated using the ratio of aboveground biomass to coarse root biomass from the 2009 harvest.

The biomass (g m⁻²) of fine roots (< 1 mm) was estimated by collecting ten 4.8-cm diameter cores (25 cm deep) in each ring section from 2006 to 2008 (*sensu* Pregitzer *et al.* 2008); samples were collected in July of each year. Fine-root productivity (mm mm⁻¹ year⁻¹) and mortality (mm mm⁻¹ year⁻¹) were estimated using minirhizotrons

installed in each ring section (Pregitzer *et al.* 2008); both production and mortality were quantified in 2003 and 2004, and we used these rates to estimate fine-root production in subsequent years. Because fine-root productivity did not differ by year, treatment or community (Pregitzer *et al.* 2008), we estimated fine-root production ($\text{g m}^{-2} \text{ year}^{-1}$) as the product of fine-root biomass (2006 to 2008; g m^{-2}) and mean fine-root productivity ($2.15 \text{ mm mm}^{-1} \text{ year}^{-1}$). NPP was estimated as the annual increment in branch, stem, coarse root and herbaceous understory biomass, plus the annual production of aboveground litter and fine roots. We report NPP as grams of dry matter annually produced per meter square ($\text{g m}^{-2} \text{ year}^{-1}$). We used a repeated-measures ANOVA for a split-plot, randomised complete block design to test the hypothesis that NPP has been sustained under eCO_2 , and to determine whether the compensatory growth of eO_3 -tolerant individuals caused NPP to recover over time (King *et al.* 2005).

Ecosystem N pools and ^{15}N tracing

We followed the movement of tracer ^{15}N into soil organic matter over time (7 days to 5 years) by randomly collecting ten 2.5-cm diameter cores in the three plant communities growing in each FACE ring (Zak *et al.* 2007a); the natural abundance of ^{15}N was initially determined in all ecosystem pools before isotope addition. Cores were composited within each plant community in each ring, dried and analysed for N and ^{15}N using a Finnigan Delta Plus IRMS interfaced with a Europa Scientific CN analyser (Zak *et al.* 2007a). In 2008, we sampled the biomass of each ecosystem pool as previously described (Zak *et al.* 2007a) and analysed each sample for N and ^{15}N as described above (Zak *et al.* 2007a). Ecosystem N and ^{15}N pools were calculated as the product of the biomass in each ecosystem pool (g m^{-2}) and the N or ^{15}N concentration ($\text{g of N or } ^{15}\text{N g}^{-1}$); ^{15}N concentrations were corrected for natural abundance of the isotope, which was determined prior to ^{15}N addition (Zak *et al.* 2007a,b). A repeated-measures ANOVA for a split-plot randomised block design was used to determine whether eCO_2 , eO_3 , or their interaction had influenced the amount of N and ^{15}N in soil organic matter beneath plant communities (King *et al.* 2005; Zak *et al.* 2007a). We used a split-plot randomised block design ANOVA to determine whether our experimental treatments influenced the amount of N and ^{15}N residing in total plant biomass, leaf litter production and forest floor (Oi & Oe).

RESULTS AND DISCUSSION

Although the eCO_2 enhancement of forest NPP was eliminated by N limitation in a long-term (11 years) experiment with sweetgum trees (*Liquidambar styraciflua*; Norby *et al.* 2010), we found no evidence of this effect after 12 years of eCO_2 exposure in the forest communities composing our experiment. Relative to NPP under ambient CO_2 (aCO_2), NPP was significantly enhanced under eCO_2 by 40% in 2006 ($P = 0.009$), 14% in 2007 ($P = 0.013$) and 25% in 2008 ($P = 0.009$), which corresponded to the 10th–12th years of the experiment (main effect means; Fig. 1); this represents a substantial and sustained increase in plant productivity. Despite eO_3 -induced reductions in plant growth that occurred early in the experiment (i.e. after 3 years of exposure; King *et al.* 2005), eO_3 had no effect on NPP during the 10th–12th years of exposure (main effect means; Fig. 1; $P = 0.128$ – 0.887). This response appears to result from the compensatory growth of eO_3 -tolerant genotypes and species as the growth of eO_3 -sensitive

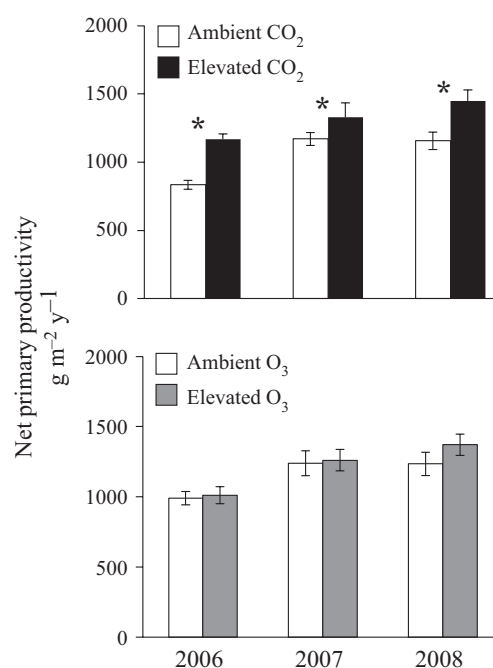


Figure 1 Net primary productivity (NPP) during the 10th to 12th years of eCO_2 (upper panel) and eO_3 (lower panel) exposure. Values are main effect means ($\pm$ SEM) averaged across plant community type; interactions between and among community type, CO_2 and O_3 were not significant, indicating that the three plant communities responded in the same manner to these trace gases. NPP was estimated as the grams of dry matter annually produced per meter square ($\text{g m}^{-2} \text{ year}^{-1}$).

individuals declined over time (Kubiske *et al.* 2007; Zak *et al.* 2007b), thereby causing NPP to attain equivalent levels under ambient O_3 (aO_3) and eO_3 . For example, eO_3 increased the growth and N acquisition of one aspen genotype (8) over the others in the aspen community (Kubiske *et al.* 2007; Zak *et al.* 2007b). In the aspen-birch community, which contained a single aspen genotype, eO_3 caused a decline in aspen growth and N acquisition, while increasing that of paper birch. The same aspen genotype was also planted with sugar maple, but unlike its negative response to eO_3 when growing with paper birch, eO_3 had no effect on aspen or maple growth (Kubiske *et al.* 2007). Collectively, these prior observations support our conclusion that compensatory growth under eO_3 resulted in equivalent NPP under aO_3 and eO_3 during the final years of our experiment (Fig. 1). Given the degree to which eO_3 has been projected to decrease global NPP (Felzer *et al.* 2005), the compensatory growth of eO_3 tolerant plants in our experiment should be considered in future simulations and, depending on the generality of this response, could dramatically diminish the negative effect of eO_3 on NPP and C storage on land as well as projected increases in anthropogenic CO_2 and climate warming (Stitch *et al.* 2007). A current gap in our knowledge of ecosystem response to eO_3 is the abundance and geographic distribution of eO_3 tolerant plant genotypes and species, whose growth could be favoured as O_3 accumulates in the Earth's lower atmosphere.

In no case did we find significant interactions among plant community, CO_2 or O_3 , indicating that NPP in the three plant communities responded similarly to both eCO_2 and eO_3 . Moreover, the lack of interaction between CO_2 and O_3 treatments indicated that NPP responded similarly to eCO_2 regardless of ground-level

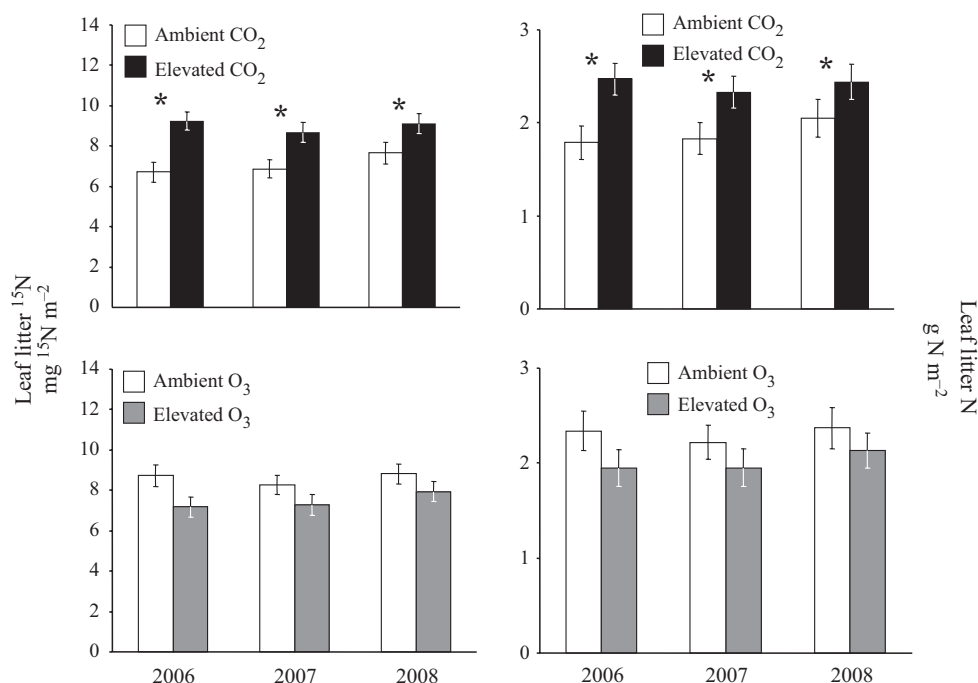


Figure 2 The amount of tracer ¹⁵N (left panels) and N (right panels) returned to soil in leaf litter fall during the 10th to 12th years of CO₂ (upper panels) and O₃ exposure. Values are main effect treatment means ($\pm$ SEM) averaged across plant community type; CO₂, O₃ or plant community type displayed no significant two- or three-way interactions.

O₃; the converse is also true. As such, we have no evidence to suggest that responses to either eCO₂ or eO₃ were additive or opposing among the three plant communities. If forests of similar composition growing throughout north-eastern North America respond in the same manner as those in our experiment (Cole *et al.* 2009), then enhanced forest NPP under eCO₂ may be sustained for a longer duration than previously thought (Norby *et al.* 2010). Furthermore, the negative effect of eO₃ may be diminished by compensatory growth of eO₃-tolerant plants as they begin to dominate forest communities (Kubiske *et al.* 2007; Zak *et al.* 2007b), suggesting that aspects of biodiversity like genetic diversity and species composition are important components of ecosystem response to this agent of global change.

In our experiment, several lines of evidence strongly indicate that greater NPP under eCO₂ was sustained by more rapid rates of soil N cycling. Foremost, the amount of N and ¹⁵N returned to soil via leaf litter fall was significantly greater under eCO₂, and this was true in 2006, 2007 and 2008 (Fig. 2). On average, 22–35% more N and 18–34% more tracer ¹⁵N, that was initially applied to soil, were returned to the soil surface via leaf litter fall under eCO₂. If the greater production of leaf litter under eCO₂ (172 vs. 230 g m⁻²; $P_{2006 \text{ to } 2008} = 0.001\text{--}0.002$) caused microbial decay to slow, then forest floor mass, N content and ¹⁵N content should be significantly greater under eCO₂; this was not the case. In 2008, we found no effect of eCO₂ on forest floor mass (ambient vs. elevated; 458 vs. 553 g m⁻², $P = 0.294$), nor did eCO₂ influence the amount of N residing in forest floor (5.2 vs. 5.6 g N m⁻², $P = 0.610$). Further, the amount of tracer ¹⁵N recovered in forest floor was also not influenced by eCO₂ (611 vs. 483 μ g ¹⁵N m⁻², $P = 0.271$). Greater amounts of N and ¹⁵N returned to soil via leaf litter fall under eCO₂, together with equivalent amounts residing in forest floor under aCO₂ and eCO₂, indicate that microbial decay and net N mineralisation have hastened under eCO₂, thereby

accelerating the rate at which N has cycled through forest floor. This observation clearly does not support the idea that changes in plant litter production and biochemical composition under eCO₂ will slow microbial litter decay and decrease N availability to plants (Luo *et al.* 2004; Johnson 2006). Additionally, eO₃ had no effect on forest floor mass ($P = 0.679$), N content ($P = 0.535$) or ¹⁵N content ($P = 0.673$), indicating equivalent rates of forest floor turnover (forest floor mass/litter production) under aO₃ and eO₃.

Greater amounts of N cycling through forest floor under eCO₂ have several potential fates, and distinguishing between them has important consequences for whether higher levels of NPP can be sustained over time by eCO₂. If enhanced litter decay led to the greater formation of soil organic matter, then such a response would sequester N in soil organic matter and decrease N availability to plants, which, in turn, would dampen or eliminate the growth-enhancing effects of eCO₂. In contrast, if a large proportion of N (and ¹⁵N) in leaf litter fall was released as inorganic N during microbial decay, then such a response would increase N availability to plants, thereby sustaining greater rates of NPP (Fig. 1). We have followed the movement of ¹⁵N into soil organic matter over the course of our experiment, and neither eCO₂ nor eO₃ have influenced the amount of ¹⁵N (Fig. 3; $P_{\text{CO}_2} = 0.632$ and $P_{\text{O}_3} = 0.635$) residing in soil organic matter; soil N content (g N m⁻²) was also not influenced by these treatment main effects ($P_{\text{CO}_2} = 0.798$ and $P_{\text{O}_3} = 0.274$). However, the amount of N contained in living plant biomass (28.8 vs. 37.5 g N m⁻², $P = 0.0172$), as well as the amount of tracer ¹⁵N recovered in it (2.44 vs. 2.89 mg ¹⁵N m⁻², $P = 0.017$), increased significantly under eCO₂; ozone had no effect on the amount of N ($P = 0.417$) or ¹⁵N ($P = 0.911$) contained in plant biomass. These observations indicate that plant acquisition of soil N was greater under eCO₂, and dispels the notion that N availability in our experiment was diminished by the incorporation of leaf litter N into long-lived soil organic matter.

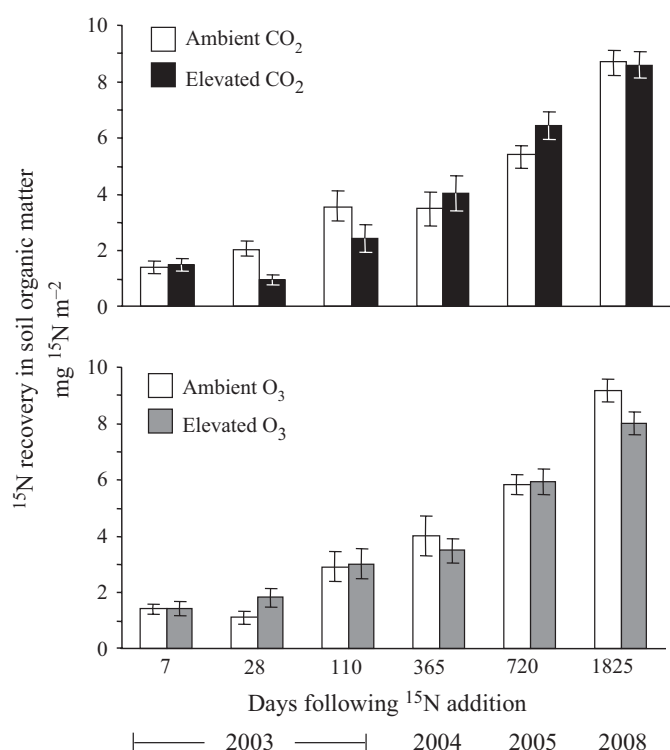


Figure 3 The amount of tracer ^{15}N recovered in soil organic matter under CO_2 (upper panel) and O_3 (lower panel) treatments. We followed the flow of ^{15}N from 7 days to 5 years after application. Values are main effect treatment means $\pm$ 1 SEM.

The greater exploitation of soil resources by fine roots appears to be one mechanism whereby plants have acquired additional N and ^{15}N under eCO_2 . Averaged over 2006 to 2008, fine-root biomass (154 vs. 172 g m^{-2} ; $P = 0.002$) and production (329 vs. $377 \text{ g m}^{-2} \text{ year}^{-1}$; $P = 0.002$) were significantly greater under eCO_2 . Although fine-root biomass ($P < 0.001$) and production ($P < 0.001$) were also significantly greater under eO_3 , this same response did not significantly increase plant N acquisition ($\text{aO}_3 = 28.8 \text{ g N m}^{-2}$; $\text{eO}_3 = 37.5 \text{ g N m}^{-2}$; $P = 0.417$). Moreover, between the 10th and 12th years of our experiment, plants continued to expand their fine-root system regardless of treatment, an indication that they had not yet fully exploited soil for growth-limiting resources. For example, averaged across CO_2 and O_3 treatments, fine-root biomass was 155 g m^{-2} in 2005 and gradually increased over subsequent years to 168 g m^{-2} by 2008. Similarly, fine-root production increased from $344 \text{ g m}^{-2} \text{ year}^{-1}$ in 2006 to $362 \text{ g m}^{-2} \text{ year}^{-1}$ in 2008; however, increases in fine-root biomass and production from 2005 to 2008 were not statistically significant ($P_{\text{biomass}} = 0.108$; $P_{\text{production}} = 0.105$). The similar responses of fine-root biomass and production under eCO_2 and eO_3 , in combination with contrasting quantities of soil N acquired by plants growing under these conditions, suggest that additional mechanisms, beyond the greater exploitation of soil by fine roots, increased the amounts of soil N plants acquired under eCO_2 .

Accumulating evidence suggests that eCO_2 can supplement the supply of soil N to plants by increasing the production of root exudates, which, in turn, facilitate the decay of soil organic matter and the subsequent release of inorganic N for plant uptake (Zak *et al.* 1993; Langley *et al.* 2009; Rütting *et al.* 2010; Drake *et al.* 2011; Phillips *et al.* 2011). This mechanism has sustained enhanced NPP under eCO_2

in a loblolly pine (*Pinus taeda*) forest (Drake *et al.* 2011), as well as a scrub-oak forest (Langley *et al.* 2009), and several lines of evidence indirectly indicate that it may have contributed to the greater plant acquisition of soil N under eCO_2 that we report here. In our experiment, soil organic matter has accumulated at a significantly slower pace under eCO_2 , despite the fact that both above- and belowground litter production have significantly increased under eCO_2 (Talhelm *et al.* 2009). This observation indicates that the decay of soil organic matter is occurring at a more rapid rate under eCO_2 , a response that has occurred in parallel with the increased rate of forest floor N cycling we report here. These findings support the idea that greater belowground plant growth under eCO_2 has accelerated organic matter decay and increased the supply of N to plants, thereby sustaining the enhancement of NPP under eCO_2 .

Under eCO_2 , higher rates of NPP sustained throughout our experiment could have increased the ecosystem-level retention of N against leaching losses, thereby providing an additional mechanism further enhancing the accumulation of N in plant biomass (Luo *et al.* 2006). For example, our experiment was initiated from bare ground into which we planted seedlings or ramets that were < 1 year old. The export of N from forest ecosystems known to be the greatest when rates of net ecosystem production (NEP) are low (Vitousek & Reiners 1975), which undoubtedly occurred during the early phase of our experiment. As both NPP and NEP increase over time, N losses decline and the accrual of N in living plant biomass increases (Yang *et al.* 2011), especially under eCO_2 (Luo *et al.* 2006). Because the eCO_2 enhancement of NPP occurred throughout our experiment, it is conceivable that N accrued at a greater rate in living plant biomass relative to N accrual under aCO_2 . This mechanism is consistent with the greater amounts of N and ^{15}N residing in plant biomass at the end of our experiment, and may be an additional mechanism by which NPP was sustained under eCO_2 (Luo *et al.* 2006).

Despite the fact that eCO_2 has enhanced NPP for over a decade in our experiment, it is unlikely that this response could be maintained over the timeframe of forest development (i.e. > 100 years). Sustained eCO_2 enhancement of NPP could occur during forest development if plant N-use efficiency increased (i.e. biomass produced per unit of N assimilated), thereby alleviating N limitation. This has not occurred in our experiment, nor has it occurred in other forest FACE experiments on unmanaged soil (Finzi *et al.* 2007). In our study, the degree to which greater root exploitation for soil resources or more rapid rates of soil N cycling have individually increased plant N acquisition is uncertain. However, as the forests in our experiment further develop, fine roots will eventually fully exploit soil resources, regardless of current differences in fine-root biomass or production under aCO_2 and eCO_2 . In developing sweetgum forests, the loss of eCO_2 enhancement of NPP coincided with fine roots attaining equivalent levels under aCO_2 and eCO_2 (Norby *et al.* 2010). Such a response suggests that differential root exploitation for soil N will plausibly diminish as the forests in our experiment mature. Moreover, throughout the course of forest development, NPP declines as a natural cause of several factors, including nutrient and hydraulic constraints on GPP (Ryan *et al.* 1997; Drake *et al.* 2010). Our results suggest that plants growing under aCO_2 and eCO_2 will eventually face these same constraints on GPP as they reach maturity (Drake *et al.* 2010), albeit plants growing under eCO_2 will reach that point more rapidly. Although the extent to which accelerated rates of organic matter decay can supplement N-limited tree growth under eCO_2 is uncertain, the fact that plant N-use efficiency has not increased in our

experiment suggests that the present enhancement of NPP under eCO₂ could diminish as forests in our experiment mature. Collectively, our results indicate that eCO₂ has accelerated the pace of forest development; however, it remains uncertain if eCO₂ will increase C stored in the biomass of future forests. Amounts of C stored in forests will depend on rates of NPP through stages of forest development (Pregitzer & Euskirchen 2004; Norby *et al.* 2010; Drake *et al.* 2011), human utilisation of forests globally and the long-term impacts of forest NPP on storage of C in soil under eCO₂ (Talhelm *et al.* 2009).

The accumulation of anthropogenic CO₂ in the Earth's atmosphere, and hence the pace of climate warming, is sensitive to the uptake of anthropogenic CO₂ by plants and the global storage of C on land (Thornton *et al.* 2007). These coupled climate–biogeochemical dynamics are modulated by feedbacks between C and N cycles in terrestrial ecosystems as well as other aspects of climate change, such as eO₃, that can impact and potentially interact with eCO₂ enhancement of plant growth. Therefore, it is imperative to accurately portray the magnitude and duration over which these feedbacks develop and are sustained in terrestrial ecosystems, because they have global climatic and biogeochemical implications (Stitch *et al.* 2007; Thornton *et al.* 2007). Here, we have demonstrated that positive feedback between C and N cycles under eCO₂ has sustained greater forest productivity over a decade in young developing forest ecosystems, regardless of differences in forest composition. The duration over which accelerated rates of organic matter decay supplement the supply of N to plants growing under eCO₂ remains uncertain and can only be determined by longer-term experimentation. Additionally, compensatory growth by eO₃-tolerant genotypes, as eO₃-sensitive individuals decline, has the potential to diminish the negative effect of eO₃ on forest productivity and must be considered to accurately predict how this driver of global change will impact terrestrial NPP, the subsequent storage of C on land as well as the course of climate warming. Collectively, our results indicate that eCO₂ may accelerate the rate at which biomass accumulates in forest ecosystems, and eO₃ will impact forest composition, but may not dramatically reduce NPP on land as current models suggest (Felzer *et al.* 2005; Stitch *et al.* 2007).

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AUTHOR CONTRIBUTIONS

K.S.P., M.E.K. and D.R.Z. designed the research; D.R.Z., K.S.P., M.E.K. and A.J.B. performed the research; D.R.Z., K.S.P., M.E.K.

and A.J.B. composed and analysed the data; D.R.Z. wrote the paper. All authors discussed the results and commented on the manuscript.

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