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Photosynthesis, respiration, and net primary production of sunflower stands in ambient and elevated atmospheric CO_2 concentrations: an invariant NPP:GPP ratio?

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Abstract

The effect of elevated CO_2 on photosynthesis, respiration, and growth efficiency of sunflower plants at the whole-stand level was investigated using a whole-system gas exchange facility (the EcoCELLs at the Desert Research Institute) and a ^{13}C natural tracer method. Total daily photosynthesis (GPP), net primary production (NPP), and respiration under the elevated CO_2 treatment were consistently higher than under the ambient CO_2 treatment. The overall level of enhancement due to elevated CO_2 was consistent with published results for a typical C3 plant species. The patterns of daily GPP and NPP through time approximated logistic curves under both CO_2 treatments. Regression analysis indicated that both the rate of increase (the parameter 'r') and the maximum value (the parameter 'k') of daily GPP and NPP under the elevated CO_2 treatment were significantly higher than under the ambient CO_2 treatment. The percentage increase in daily GPP due to elevated CO_2 varied systematically through time according to the logistic equations used for the two treatments. The GPP increase due to elevated CO_2 ranged from approximately 10% initially to 73% at the peak, while declining to about 33%, as predicted by the ratio of the two maximum values. Different values of percentage increase in GPP and NPP were obtained at different sampling times. This result demonstrated that one-time measurements of percentage increases due to elevated CO_2 could be misleading, thereby making interpretation difficult. Although rhizosphere respiration was substantially enhanced by elevated CO_2, no effect of elevated CO_2 on R:P (respiration:photosynthesis) was found, suggesting an invariant NPP:GPP ratio during the entire experiment. Further validation of the notion of an invariant NPP:GPP ratio may significantly simplify the process of quantifying terrestrial carbon sequestration by directly relating total photosynthesis to net primary production.

Keywords: carbon-13, CO_2, mesocosm, photosynthesis, respiration, sunflower

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Introduction

The trend of continuous increase in atmospheric CO_2 has been well-documented in global environmental change research (Keeling *et al.* 1989). An increase in atmospheric CO_2 concentration often results in a significant increase in primary productivity as measured by leaf-level gas exchange (Curtis 1996). To understand ecosystem responses to global change, however, it is critical to integrate key processes regulating material and energy flows across spatial and temporal scales (Mooney 1991).

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Given the restrictions of current experimental methods, our knowledge in this area is limited mostly to spatial scales ranging from the whole-plant to the biochemical level. Knowledge at these scales has been obtained primarily from noncontinuously measured pot studies of individual plants and is generally inadequate to use for predicting system-level responses (Mooney *et al.* 1991).

Several field studies at both the microcosm and mesocosm scale have been reported (Strain & Thomas 1992), including studies of the arctic tundra (Billings *et al.* 1984; Oechel *et al.* 1994), tallgrass prairie (Owensby *et al.* 1993), Californian grasslands (Field *et al.* 1995), alpine grassland (Schäppi & Körner 1996), and a wetland (Drake 1992). These field studies have increased our understanding in the following research areas: (i) system carbon fluxes (i.e. input and output); (ii) carbon and nitrogen interactions; and (iii) carbon, nitrogen, and water relationships. In most of these studies, plant photosynthesis and respiration of aboveground components were not measured continuously, thereby limiting the usefulness of the data for modelling and integration (Körner 1995). Root respiration and rhizosphere activities could not be determined in these field studies because of the mixing of soil-derived carbon with plant-derived carbon. A time-course analysis of photosynthesis, respiration, and net primary production of whole-plant and soil systems at the mesocosm scale has been lacking, due primarily to methodological difficulties.

Plant growth and net primary production depend on the balance between carbon gain through photosynthesis and carbon loss through respiration. Processes occurring at smaller and shorter term scales may not be directly applicable to larger and longer-term scales due to component interactions and time-delay responses (Luo & Reynolds 1999). A review of recent literature (Curtis 1996) confirmed that plant responses vary depending on the spatial and temporal scales studied.

Plant respiratory CO_2 efflux is a major physiological process that influences the carbon balance of plant communities and ecosystems (Ryan 1991). Plant respiration may be inhibited directly by increased concentrations of CO_2 or altered indirectly by elevated CO_2 through a possible acclimation response, due either to changes in tissue nitrogen or to construction costs. Short-term, small-scale studies of the effects of elevated CO_2 on plant respiration have reported potentially contradictory results. In response to elevated CO_2, plant respiration has been found to decrease (Wullschleger & Norby 1992; Bunce 1995), increase (Thomas *et al.* 1993; Thomas & Griffin 1994), or show no effect (Wullschleger *et al.* 1995; Bouma *et al.* 1997), depending on types of plant tissues, species, or experimental conditions. Studies at the whole-plant level with longer measurement periods (Gifford 1994, 1995; Tjoelker *et al.* 1999) have shown that plant respiratory loss of C as a proportion of total photosynthesis C gain (i.e. R:P ratio) is not affected by an elevated CO_2 concentration, indicating an invariant R:P (or interchangeably the ratio of net primary production to gross primary production — NPP:GPP) ratio under changing atmospheric CO_2 concentrations. It is an open question, however, whether this hypothesized invariant R:P ratio holds true at the mesocosm scale and above. A substrate-utilization/single-leaf model has been developed to provide a mechanistic explanation for this phenomenon (Dewar *et al.* 1998, 1999). Based on analysis with this model, an invariant NPP:GPP ratio is predicted to be true at the whole-plant and larger spatial scales. (Gifford 1994, 1995; Tjoelker *et al.* 1999)

In our study, we used the EcoCELLs, a plant growth facility (Griffin *et al.* 1996a) at the Great Basin Environmental Research Laboratory of the Desert Research Institute. The unique characteristics of this facility enabled us to continuously measure the fluxes of carbon in whole-plant and soil systems. Using the ^{13}C natural tracer technique (Cheng 1996), plant-derived carbon was monitored separately from soil-derived carbon. By using the ^{13}C natural tracer technique in the EcoCELL facility, we were able to overcome some methodological difficulties and continuously measure whole-system photosynthesis, respiration, and net primary production. The main objectives of this study were: (i) to investigate the effect of elevated atmospheric CO_2 on photosynthesis and respiration both aboveground and belowground at the whole-plant stand level; (ii) to determine whether net primary production increases in proportion to the increase in total photosynthesis as a response to elevated CO_2 during the initial vegetative growth stage; and (iii) to examine which parameter(s) of the plant growth curve are affected significantly by elevated CO_2 at the whole-plant stand level.

Materials and methods

Experimental system

A detailed description of the EcoCELLs can be found in Griffin *et al.* (1996a). Briefly, the EcoCELLs are a series of environmentally controlled, naturally lit, open-flow, mass-balance systems at the mesocosm scale. The EcoCELLs have the same theory of operation as leaf-level gas-exchange systems but work at a much larger scale, measuring whole-system fluxes continuously. The dimensions of each EcoCELL are $7.3 \times 5.5 \times 4.5$ m (L × W × H), providing a total volume of 183.5 m^3. There is a circulating volume of 162.5 m^3 and a soil volume of 20.1 m^3 in each cell. The soil medium in each cell is contained in three $2.85 \times 1.3 \times 1.8$ m (L × W × H)

acrylic-walled boxes. These boxes comprise the lysimeter/rhizotron system and adjoin each other closely, leading to the formation of a continuous plant canopy. Each lysimeter/rhizotron is mounted on a set of four load cells capable of discriminating a change in weight of 250 g out of a total weight of 2.0×10^6 g. The air temperature, relative humidity, and CO_2 concentration in each EcoCEll are controlled automatically.

Two EcoCELLs were used in this experiment, one under ambient atmospheric CO_2 and the other under elevated CO_2 (ambient plus 350 μL L^{-1}). The CO_2 levels in the ambient treatment were dependent on the outside air. Thus, the lower-end set-point was constrained by the global average CO_2 concentration and local anthropogenic source emissions. The 350 μL L^{-1}increase in the CO_2 concentration in the elevated treatment was controlled by a three-stage system. First, a needle valve was used to inject a constant amount of CO_2, approximately 80–90% of the required addition. The CO_2 then passed through a mass flow controller for coarse control (0–100 L min^{-1} with 15 L min^{-1} steps), followed by a fine control mass flow controller (0–15 L min^{-1}). Using this three-stage approach, we were able to obtain CO_2 concentrations well within 2% of the desired set point.

A soil profile was constructed in each soil container in the EcoCELLs. The soil profile consisted of the following three layers: (i) 0–0.4 m, 1:1 mix of top soil from Kansas Tallgrass prairie and washed river sand; (ii) 0.4–0.8 m, washed river sand; and (3) 0.8–1.8 m, washed river bed pebbles. The top soil was obtained from the Konza Prairie Long-Term Ecological Research site. The soil carbon was predominantly C4 plant-derived carbon (^{13}C enriched), which had a $\delta^{13}C$ value of -14.2 ± 0.14‰. The soil was first sieved through a 12.7-mm screen. Plant material and large stones were the removed from the soil by hand-picking before use. The 1:1 mix of the soil with sand was made using a cement mixer. The amount of soil:sand mix applied to each soil container was measured using electronic scales.

Sunflowers (*Helianthus annus*) were planted in each soil container and grown for 53 days in the two EcoCELLs under nearly identical air temperature, humidity, soil moisture, and irradiance conditions. At the end of the experiment, no floral initiation was visible. There were 108 plants in each EcoCELL. Water was supplied based on whole-system weight data and through frequent hand watering. The soil water content of both EcoCELLs was kept between 60% and 100% of field-water holding capacity. Daytime air temperature was set at 28 °C and night-time at 13 °C. Daytime relative humidity was set at 30% and night-time at 60%. The environmental control system was able to maintain the air temperature in the EcoCELLs within ± 0.5 °C and the relative humidity within ± 5%. Most of the days during the experimental period were cloudless. The appearance of the plants was normal and healthy.

We chose to use sunflowers for this research because they: (i) represent a common C3 plant; (ii) show a typical response to elevated CO_2; and (3) allow for measurement of belowground CO_2 with relative ease.

Whole-system carbon fluxes

Whole-system carbon fluxes include: (i) gross primary production (GPP) or total carbon fixed by photosynthesis; (ii) net primary production (NPP), which equals GPP minus total plant respiration; (iii) total plant respiration, which consists of shoot respiration and rhizosphere respiration defined as the sum of root respiration and microbial respiration supported by root-derived substrates; (iv) soil respiration; and (v) net ecosystem production (NEP), which equals total carbon input from photosynthesis minus total carbon output from plant–soil respiration. The five fluxes have the following relationships:

$$\text{NEP (net ecosystem production)} = \text{GPP} - R_{\text{shoot}} - R_{\text{rhizo}} - R_{\text{soil}}$$

or

$$\text{NEP} + R_{\text{soil}} = \text{GPP} - R_{\text{shoot}} - R_{\text{rhizo}}$$

Because $\text{NPP} = \text{GPP} - R_{\text{shoot}} - R_{\text{rhizo}}$ by definition, then

$$\text{NPP} = \text{NEP} + R_{\text{soil}} \quad (1)$$

$$\text{GPP} = \text{NPP} + R_{\text{shoot}} + R_{\text{rhizo}} \quad (2)$$

where NEP = net ecosystem production or whole-cell net carbon gain, measured using a whole-cell gas exchange system; GPP = gross primary production or total carbon fixed by plant photosynthesis; NPP = net primary production; R_{shoot} = shoot dark respiration, i.e. total dark respiration measured using whole-cell gas exchange minus belowground respiration (CO_2 loss from roots and soil); R_{rhizo} = rhizosphere respiration, or root respiration plus microbial respiration utilizing root-derived substrates, equal to total belowground respiration minus soil respiration, measured by an open-flow gas exchange system combined with ^{13}C analysis; and R_{soil} = soil respiration (C loss from original soil organic matter), measured by an open-flow gas exchange system combined with ^{13}C analysis.

NEP: net ecosystem production

Net ecosystem production was measured continuously using the EcoCELL gas exchange system (Griffin *et al.*

1996a). Briefly, three infrared gas analysers (IRGAs) were dedicated to the CO_2 monitoring system. Two IRGAs were run continuously in differential mode to record the net flux of CO_2 across each EcoCELL. The third IRGA ran in absolute mode and sequentially sampled a standard gas, as well as the gas entering and exiting each EcoCELL. The reference gas was in a closed loop continuously scrubbed of CO_2 and H_2O by soda-lime and magnesium perchlorate. All three IRGAs were sampled at 5-s intervals and recorded as 60-s averages. Each IRGA was zeroed and spanned with NIST traceable standards (99% accuracy standards) on an every-day schedule to account for any drift in the calibration coefficients.

The mass flow rate of air entering each EcoCELL was measured directly using a multipoint hot-wire anemometer. Each mass-flow meter was calibrated individually *in situ* using a trace gas addition technique (Field *et al.* 1991). System calibration of the airflow was carried out prior to planting and after the completion of the experiment to account for any drift in the volumetric flow measurements.

Before the start of the experiment, all equipment in the whole gas exchange system was calibrated either by the manufacturer or by DRI laboratory personnel. During the experiment, the accuracy of the whole-system gas exchange system was checked five times by injecting a known amount of CO_2 gas through a calibrated mass-flow meter with an accuracy of 98%.

Data points affected by the presence of human activities inside the EcoCELLs, as well as door openings, were corrected using daily regression curves of hourly data points between photosynthetic active radiation (PAR) and NEP during daytime and the average of unaffected data points before and after the affected point(s).

Soil and rhizosphere respiration (R_{soil}, R_{rhizo})

The loss of original soil carbon due to soil respiration was determined by subtracting rhizosphere-respired carbon (i.e. C3 plant-derived carbon) from the total respired carbon from the belowground system. The release of CO_2 from the belowground system during a 24-h period was measured weekly using a continuous open-flow gas exchange system (Cheng *et al.* 2000). Six sampling units (inverted boxes) were employed in each EcoCELL. The open-flow gas exchange system for measuring soil surface CO_2 fluxes was calibrated before planting using the whole EcoCELL gas exchange system. The $\delta^{13}C$ value of belowground CO_2 was determined weekly using a closed-circulation CO_2 trapping system (Cheng *et al.* 2000) and subsequent analysis of ^{13}C abundance in the trapped CO_2 during each 24-h period (Harris *et al.* 1997). Plant-derived, rhizosphere-respired carbon in total belowground CO_2 was partitioned from soil-derived carbon using the ^{13}C natural tracer method (Cheng 1996). Daily values of soil and rhizosphere respiration were obtained by extrapolating data of weekly measurements using curve fitting.

The ^{13}C natural tracer method is based on the difference in ^{13}C:^{12}C ratio (often reported as the $\delta^{13}C$ value) between C3 and C4 plants (Smith & Epstein 1971), and on the subsequent difference between soil organic matter derived from the two types of plants. Soil organic matter derived from C4 plant-dominated vegetation (C4-derived soil), such as tall-grass prairie and tropical grasslands, has $\delta^{13}C$ values ranging from -12 to $-20‰$, whereas $\delta^{13}C$ values of soil organic matter derived from cold and temperate forest (C3-derived soil) range from -24 to $-29‰$. By using a C4-derived soil in a C3 plant-dominated system such as sunflowers, or vice versa, the carbon entering the soil via roots will have a different $\delta^{13}C$ value than the $\delta^{13}C$ value of the soil. The following equation can be used to partition soil-derived C4 carbon from plant-derived C3 carbon:

$$C_3 = C_t\,(\delta_t - \delta_4)/(\delta_3 - \delta_4), \qquad (3)$$

where $C_t = C_3 + C_4$ and is the total belowground CO_2-carbon; C_3 = the amount of carbon derived from C3 plants; C_4 = the amount of carbon derived from C4 soil; δ_t = the $\delta^{13}C$ value of the C_t carbon; δ_3 = the $\delta^{13}C$ value of the C3 plant carbon; and δ_4 = the $\delta^{13}C$ value of the C4 soil carbon.

Shoot respiration

Shoot respiration during each night was calculated by subtracting belowground respiration from total EcoCELL respiration during the dark period. Daytime shoot respiration was calculated based on the night-time rate with adjustment for the air temperature difference using a Q_{10} value equal to 2 because shoot respiration for most agricultural crops has Q_{10} values centred around 2 (Amthor 1984). Different Q_{10} values (1.5 and 2.5) was also used in sensitivity analysis.

Results and discussion

The visual appearance of plants under both CO_2 treatments was healthy and normal during the entire experiment. Neither herbivorous insects nor diseases were noticed. Nutrient supply to plant growth seemed adequate because nutrient-deficient symptoms did not appear. The cloudless condition during this experiment simplified our analysis through time since photosynthetic active radiation was relatively constant. Replications were not included in the experimental

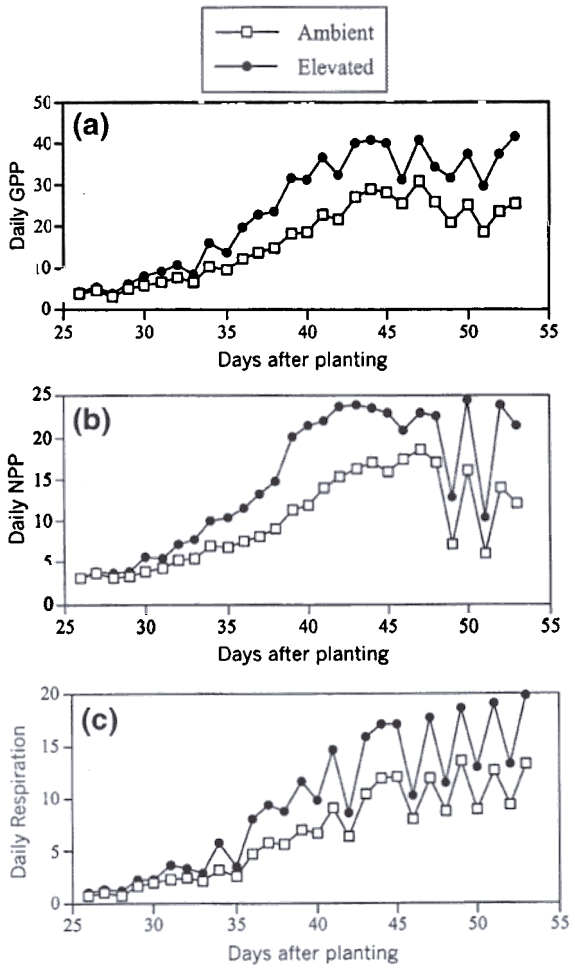

Fig. 1 (a)Daily gross primary production (GPP), (b) daily net primary production (NPP), and (c) daily total plant respiration (respiration) in $g C m^{-2} d^{-1}$.

design because there were only two EcoCELLs available at the time of this experiment. Statistical inference was limited because CO_2 treatments were not replicated. The only intended and detectable difference between the two EcoCELL treatments was the concentration of atmospheric CO_2. Other experimental conditions including air temperature, humidity, lighting, and soils were almost identical (Cheng *et al.* 2000).

Changes in GPP and NPP through time

As plant growth progressed, whole-stand daily gross primary production (GPP) initially increased exponentially and later approached asymptote under both CO_2 treatments (Fig. 1a), except for some cloudy days toward the end of this experiment. Daily GPP under the elevated CO_2 treatment was consistently higher than under the ambient treatment. Whole-stand daily net primary production (NPP) showed a similar pattern through time (Fig. 1b). Whole-stand daily plant respiration, including rhizosphere respiration, increased as GPP increased but with much more day-to-day variation (Fig. 1c). Daily total plant respiration under the elevated CO_2 treatment was consistently higher than under the ambient treatment. This overall increase in total photosynthesis, net primary production, and total plant respiration in response to elevated CO_2 is consistent with many published reports (as reviewed in Curtis 1996).

Changes in daily GPP and daily NPP through time roughly took the form of logistic curves. Maximum daily GPP and NPP occurred approximately 43 days after planting. To compare the effect of elevated CO_2 on logistic parameters, logistic equations were fitted to the daily GPP and NPP data after transforming the equation to a linear form. A general logistic equation was used, which took the following form:

$$GPP_t \text{ (or } NPP_t) = K/(1 + A\,e^{-rt}),$$

where GPP_t is daily gross primary production at time t; K is the maximum GPP; A is a conversion constant; r is the instantaneous rate of change; and t is time, in days after planting (or DAP). Some data points near the end of the experiment were excluded due to cloudiness (Fig. 2a,b). The regression equations for both CO_2 treatments were highly significant ($P > 0.001$), indicating that sunflowers grown under elevated CO_2 had higher rates of increase for daily GPP and daily NPP than under ambient CO_2. Maximum daily GPP and NPP were also higher under elevated CO_2 than under ambient CO_2. The rate of increase in stand photosynthesis was determined by both the speed of canopy development and the photosynthetic rate per unit of leaf area, whereas maximum stand-level photosynthesis was controlled mainly by the leaf-level photosynthesis integrated across the whole stand after the canopy fully developed. Earlier studies in the EcoCELLs indicated that leaf position in the canopy is an important factor in determining total canopy photosynthesis (Sims *et al.* 1999), and that canopy quantum yield varies with canopy development and light levels (Luo *et al.* 2000).

The ratio of daily GPP under elevated CO_2 to daily GPP under ambient CO_2 (GPP_e:GPP_a) over time showed a bell-shaped curve, initially increasing to a maximum of approximately 1.7 and declining later (Fig. 3a). This trend could also be derived from the regression curves of the logistic equations, indicating that the change of GPP_e:GPP_a over time resulted from both the higher rate of increase and higher maximum daily GPP under elevated CO_2. The ratio of daily NPP under elevated CO_2 to daily NPP under ambient CO_2 (NPP_e:NPP_a) over

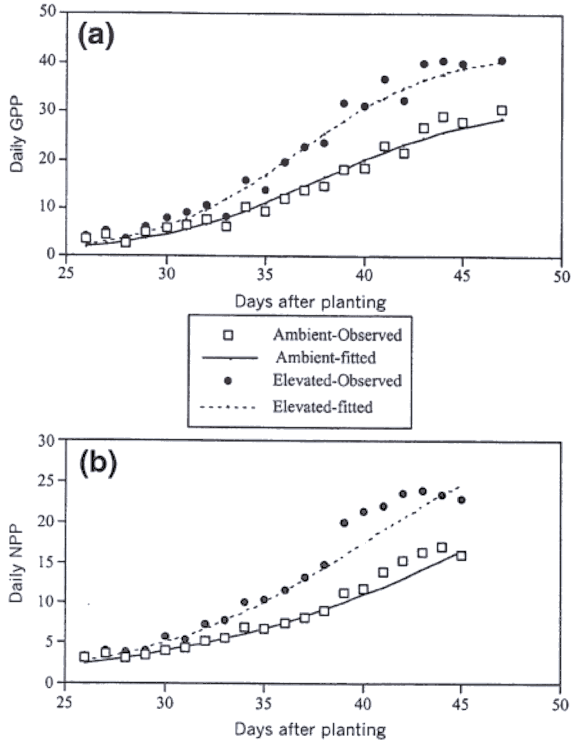

Fig. 2 Logistic curves fitted to (a) daily gross primary production (GPP) and (b) daily net primary production (NPP) in $g C m^{-2} d^{-1}$ under ambient CO_2 (□) and elevated CO_2 (●).

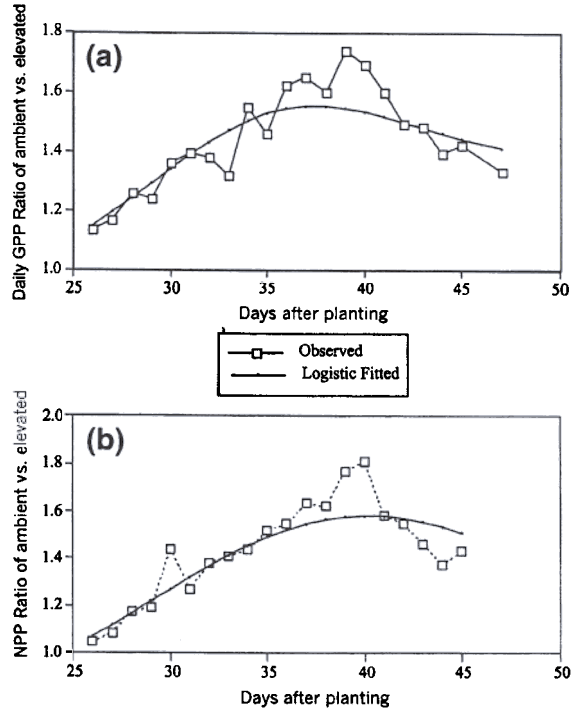

Fig. 3 (a) Ratios of daily GPP under elevated CO_2 to ambient CO_2 and (b) ratios of daily NPP under elevated CO_2 to ambient CO_2. The smooth curves are from logistic equations shown in Fig. 2.

time showed a similar pattern (Fig. 3b). These results indicate that the percentage increase in daily GPP due to elevated CO_2 varied systematically through time according to logistic equations of the two treatments (from approximately 10% initially to as high as 73% at the peak and declining to about 33%, as predicted by the ratio of the two maximum values at the end). Different percentage increases in GPP or NPP could be obtained simply by changing the sampling times, demonstrating that one-time measurements of percentage increases due to elevated CO_2 are often difficult to interpret and can be misleading.

If the logistic curve is a common growth pattern for most annual plants, the parameters of the curve may offer useful information for scaling over time. A significant increase of the parameter r in the logistic curve of daily GPP, as a result of elevated CO_2, can be interpreted as an accelerated expansion of plant photosynthesis capacity, which may lead to an early reach of the maximum photosynthesis rate under elevated CO_2. Alternatively, a significant increase of the parameter K in the logistic curve of daily GPP may indicate the overall enhancement of closed-canopy photosynthesis due to elevated CO_2. As has been discussed by Coleman & Bazzaz (1992), plant responses to elevated CO_2 may depend on ontogenies. Different responses to elevated CO_2 may occur at different developmental stages. Our results support the assertion that continuous or multiple-point measurements through time are essential for understanding plant response to an elevated CO_2 environment, especially for longer-lived plant communities such as forests.

Daily relative increase of NPP (DRINPP) and growth efficiencies

Daily relative increase of NPP (DRINPP), defined as $DRINPP = [(NPP_{tj} - NPP_{ti})/NPP_s]$, where NPP_s is the sum of NPP from t_0 to t_j], declined through time for both CO_2 treatments (Fig. 4), similar to commonly reported data in the literature (Pooter 1993; Gifford *et al.* 1996). This was due primarily to the faster increase in the total NPP denominator. DRINPP under elevated CO_2 was higher than under ambient CO_2 until 40 days after planting and lower than under ambient CO_2 after that date. The fortieth day also corresponds with the first turning point on the curves of GPP and NPP ratios in Fig. 3 and with canopy closure as the leaf area index

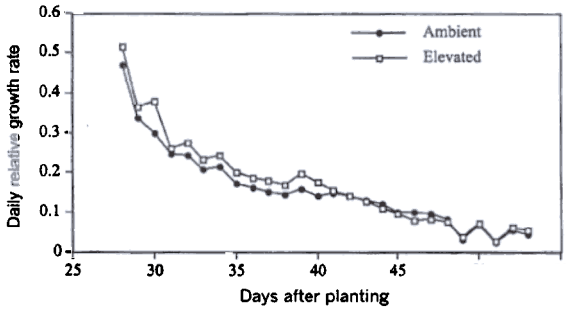

Fig. 4 Daily relative growth rates ($g C g C^{-1} d^{-1}$ under ambient CO_2 and elevated CO_2.

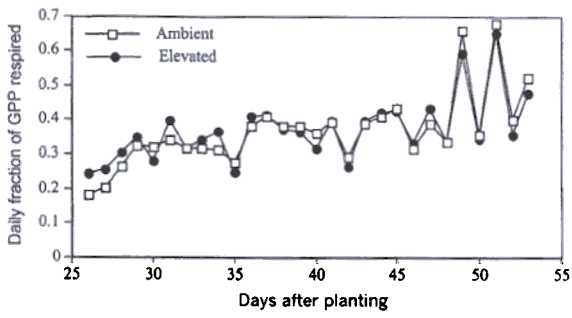

Fig. 5 Daily fraction of GPP respired by the plants under ambient CO_2 and elevated CO_2.

reached 2 (see Sims *et al.* 1999). This suggests that the magnitude of the elevated CO_2 effect on total photosynthesis could be higher under open canopy than under closed canopy. In another study, we found that photosynthetic acclimation to elevated CO_2 occurred only in older leaves of lower canopy positions, and not in younger leaves at the top of the canopy (Sims *et al.* 1999). Although the photosynthetic capacity of older, lower leaves under elevated CO_2 was down-regulated due to acclimation, the maximum GPP under elevated CO_2 was still 33% higher than under ambient CO_2 after canopy closure, probably because leaves at the top of the canopy contributed more photosynthesis than the leaves below them.

Whole-stand daily respiration expressed as a percentage of daily GPP for both CO_2 treatments increased during the early part of the experiment, stabilized around 40% during the middle period, and fluctuated during the end period primarily due to cloudiness (Fig. 5). The percentage GPP utilized in respiration was slightly higher under elevated CO_2 than ambient CO_2 during the early period and became similar under both CO_2 treatments later. Total plant respiration as a proportion of GPP, or plant growth efficiency, was not affected significantly by elevated CO_2. Root/rhizosphere respiration as a proportion of total plant respiration, however, was consistently higher under elevated CO_2 than under ambient CO_2 (Fig. 6). Root:shoot ratio was also higher under elevated CO_2 than under ambient CO_2 (data reported in Cheng *et al.* 2000). These results suggest that plants grown under elevated CO_2 allocated more photosynthate to belowground components than did plants under ambient CO_2, even though total plant respiration as a proportion of GPP was not affected by elevated CO_2. Similar results have been reported from several small-scale studies using potted plants in greenhouses or growth chambers (Kuikman *et al.* 1990; Billes *et al.* 1993; Cheng & Johnson 1998). These results indicate that analysis of plant growth and carbon allocation need

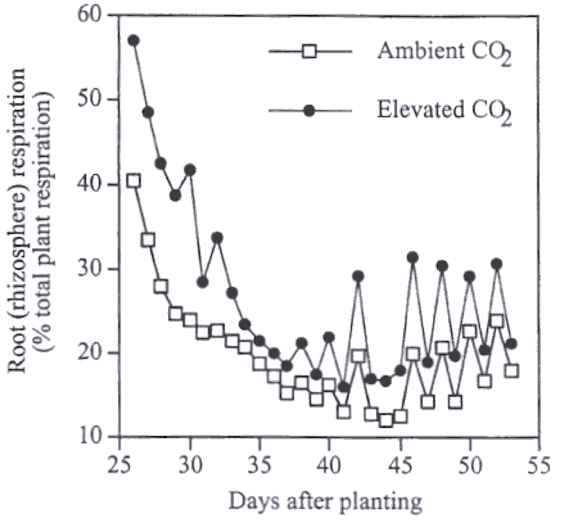

Fig. 6 Root/rhizosphere respiration as percentage of total daily plant respiration under ambient CO_2 (□) and elevated CO_2 (●).

to consider both aboveground and belowground components, and also include the temporal dimension. The absence of measurable plant growth response to elevated CO_2 despite a significant increase in photosynthesis, as reported in several published studies (Norby *et al.* 1992; Diemer 1994; Körner *et al.* 1996), may have been caused by the exclusion of either belowground components or time-course analysis.

Invariant NPP:GPP ratio

Daily NPP was linearly correlated with daily GPP under both CO_2 treatments (Fig. 7). The relationships between daily NPP and daily GPP were similar for both CO_2 treatments as indicated by slope and intercept values. This result suggests that growth efficiencies at the stand

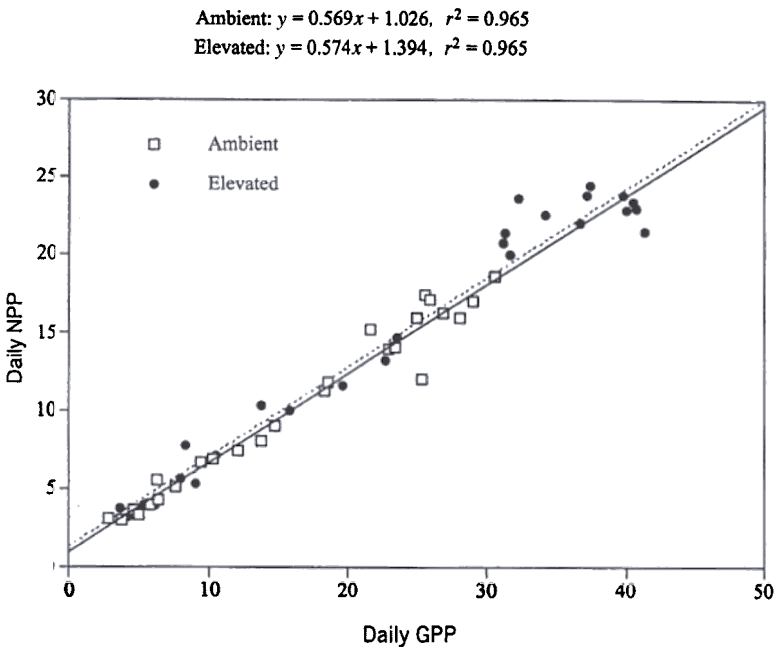

Fig. 7 Linear correlation between daily NPP and daily GPP under ambient CO_2 and elevated CO_2.

level were virtually the same for both CO_2 treatments and throughout the period of the experiment. Similar results were also reported in a study using whole-plant carbon balancing of wheat plants (Gifford 1995). Based on results from this experiment using wheat plants, Gifford (1995) proposed that plant growth efficiency or R:P ratio might be independent of several environmental variables such as CO_2 concentration and air temperature. Similar results were also reported by Tjoelker *et al.* (1999).

As described previously in the materials and methods section, GPP was determined based on three components, i.e. NPP, shoot respiration, and rhizosphere respiration. Night-time shoot respiration was measured directly, whereas daytime shoot respiration was calculated based on the corresponding night-time rate, with adjustment for the temperature difference between day and night using a fixed Q_{10} value. The choice of different Q_{10} values might potentially influence the result of an invariant NPP:GPP ratio. To investigate the effect of different Q_{10} values on NPP:GPP ratio, a sensitivity analysis was conducted by calculating NPP:GPP ratio using Q_{10} values of 1.5, 2.0, and 2.5, respectively. This sensitivity analysis indicated that using different Q_{10} values did not change the results described above, i.e. NPP:GPP ratio was invariant for both CO_2 treatments and throughout the period of the experiment (Fig. 8). However, using different Q_{10} values in the calculation resulted in different values of NPP:GPP: approximately 0.65 when Q_{10} was 1.5; 0.58 when Q_{10} was 2.0; and 0.52 when Q_{10} was 2.5, for both CO_2 treatments. This was expected because using larger Q_{10} values enlarged shoot respiration, and therefore increased the respiration portion of GPP, and decreased the NPP portion.

Our mesocosm-scale results are among the first to actually support the proposition that plant growth efficiency or whole-stand NPP:GPP ratio is independent of several environmental variables such as CO_2 concentration. Given the complexity in plant respiratory responses to elevated CO_2 shown in published reports (e.g. Griffin *et al.* 1996b; Amthor 1997), the relative constancy of plant-growth efficiency (R:P ratio) or of NPP:GPP is of importance in prediction, model construction, and integration across multiple scales. If NPP:GPP is invariant, the process of quantifying terrestrial carbon uptake can be simplified by directly relating total plant photosynthesis to net primary production. If the CO_2 fertilization factor on C3 photosynthesis is relatively constant and independent of other variables (e.g. temperature, soil nutrition, and species) as indicated by Luo *et al.* (1996), and NPP:GPP is relatively independent of other variables as supported by our results, then the increase of NPP due to CO_2 fertilization should be relatively independent of other environmental variables. Although some theoretical support for this hypothesis has been provided by a modelling study (Dewar *et al.* 1999) using substrate utilization as the underlying mechanism, further testing is definitely needed to answer critical questions. Is the NPP:GPP ratio truly invariant across ecosystem types or invariant for each particular ecosystem? Is the NPP:GPP ratio a

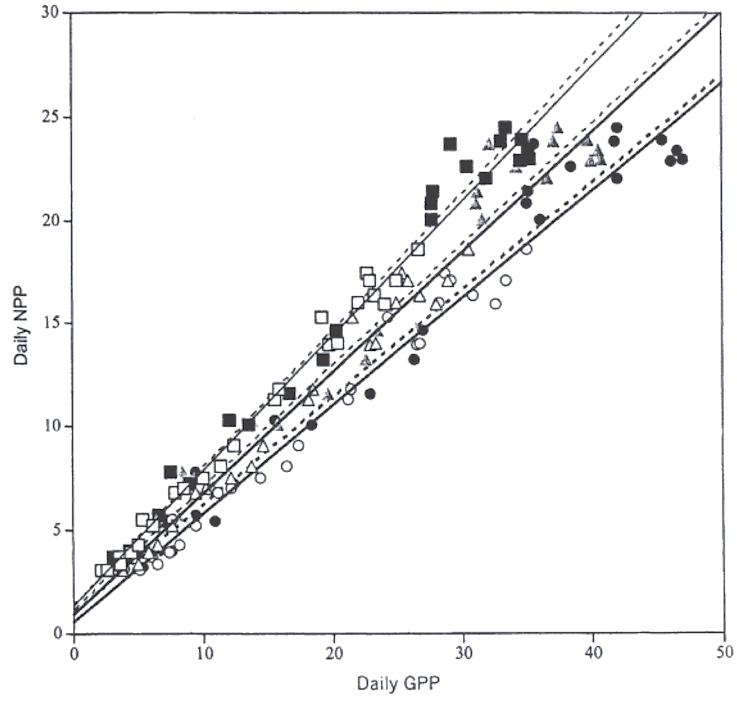

Fig. 8 Linear correlation between daily NPP and daily GPP under ambient CO_2 and elevated CO_2 when three different Q_{10} values are used to calculate daytime shoot respiration. Open symbols are data points of the ambient CO_2 treatment. Closed symbols are data points of the elevated CO_2 treatment. Square symbols are data points when $Q_{10}=1.5$, triangle symbols are data points when $Q_{10}=2.0$, and circle symbols are data points when $Q_{10}=2.5$. Dotted lines are linear fits of data points of the ambient CO_2 treatment, and solid lines are linear fits of data points of the elevated CO_2 treatment. The middle two lines are linear fits of data points when $Q_{10}=2.0$, which are the same as shown in Fig. 7. The upper two lines are linear fits of data points when $Q_{10}=1.5$, and the lower two lines are linear fits of data points when $Q_{10}=2.5$.

constant for all forest ecosystems as suggested by Waring *et al.* (1998)? What are the mechanisms stabilizing NPP:GPP in addition to substrate utilization (as suggested by Dewar *et al.* 1999)? Is NPP:GPP species-dependent?

Caution should be exercised before the notion of invariant NPP:GPP can be generally applied. Our results presented here are obtained from an experiment without replications. Whether or not NPP:GPP is invariant in undisturbed, mature ecosystems remains to be tested. Mechanisms responsible for this potentially invariant ratio need to be explored further. To stimulate further research and discussion on this topic, the concept of 'ecosystem incorporation' proposed in *Hierarchical Concept of Ecosystems* (O'Neill *et al.* 1986) may be considered as a general mechanism through which NPP:GPP stabilizes (i.e. at and above the whole-plant level, NPP:GPP tends to stabilize because the biotic system is complex enough that environmental perturbations and variations get incorporated and are no longer uncontrollable) (O'Neill *et al.* 1986, p. 169).

Conclusions

Whole-system daily photosynthesis (GPP), net primary production (NPP), and respiration under an elevated CO_2 treatment were higher than under an ambient CO_2 treatment. Changes in daily GPP and NPP through time approximated logistic curves under both CO_2 treatments. Both the rate of increase (r) and the maximum value (k) of daily GPP and NPP under the elevated CO_2 treatment were significantly higher than under the ambient CO_2 treatment. The percentage increase in daily GPP due to elevated CO_2 varied systematically through time according to logistic equations of the two treatments, from approximately 10% initially to as high as 73% at the peak and declining to about 33% as predicted by the ratio of the two maximum values. Different values of percentage increase in GPP or NPP could be obtained simply by changing the sampling time, indicating that one-time measurements of percentage increases due to elevated CO_2 are often difficult to interpret and could be misleading. Although rhizosphere respiration was enhanced substantially by elevated CO_2, no effect of elevated CO_2 on whole stand R:P was found, suggesting an invariant NPP:GPP ratio during the entire experiment.

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