Plant and Soil 207: 77–86, 1999.

Predominance of ecophysiological controls on soil CO_2 flux in a Minnesota grassland

Joseph M. Craine[1,*], David A. Wedin[2] and F. Stuart Chapin, III[3]
[1]*Department of Integrative Biology, University of California at Berkeley, Berkeley, CA 94720, USA;* [2]*School of Natural Resource Sciences, University of Nebraska, 104 Plant Industry Bldg., Lincoln, NE 68583, USA and* [3]*Institute of Arctic Biology, University of Alaska, Fairbanks, AK 99775, USA*

Key words: Cedar Creek, Minnesota, light, root respiration, *Scizachyrium scorparium*, soil CO_2 flux, temperature

Abstract

Ecosystem studies often study soil CO_2 flux as a function of environmental factors, such as temperature, that affect respiration rates by changing the rate of utilization of carbon substrates. These studies tend not to include factors, such as photosynthesis, that affect the supply of carbon substrates to roots and root-associated processes. We examined the role of decreased carbohydrate source on soil CO_2 flux and root respiration in an annually-burned grassland through manipulations of light intensity and removal of above ground biomass. We also quantified the contribution of root respiration to soil CO_2 flux by measuring the respiration rates of excised roots. Two days of shading caused a 40% reduction in soil CO_2 flux, while clipping was associated with a 19% reduction in soil CO_2 flux. Both reductions were independent of soil and air temperature at the time of measurement. The relative decrease in soil CO_2 flux observed in the clipping experiment was similar in magnitude to an observed decrease in root respiration per gram of root, linking decreased root activity and soil CO_2 flux. From these experiments, we conclude that variation in factors that affect carbon availability to roots can be important determinants of soil CO_2 flux and should be included explicitly in studies that measure or model soil CO_2 flux.

Abbreviations: NPP – net primary production; SOM – soil organic matter; SRR – specific root respiration

Introduction

Accurate predictions of carbon exchange between terrestrial ecosystems and the atmosphere require an understanding of factors influencing patterns of soil CO2 flux. Temperature has been the abiotic factor most associated with soil CO_2 flux in reviews of global patterns of soil CO_2 flux (Lloyd and Taylor, 1994; Raich and Potter, 1995), models of ecosystem carbon exchange (Keith et al, 1997; Zhang et al., 1994), and global change models (Ryan, 1991). This focus on temperature comes from the systems tradition of ecosystem ecology in which soils are modeled as a compartment whose behavior can be predicted with simple thermodynamic principles such as a Q_{10} relationship between respiration and temperature. At a physiological level, an emphasis on temperature implies that respiration of carbon substrates in roots and by microbes is more dependent on factors that affect the rate at which available carbon substrates are respired ('pulls' *sensu* Amthor, 1994) rather than factors that affect the rate at which these substrates are supplied below ground ('pushes' *sensu* Amthor, 1994). However, some studies show weak relationships between temperature and soil CO_2 flux or root respiration (Ham et al., 1995; Lambers et al., 1996) without identifying other 'pulling' determinants.

Even when temperature and soil CO_2 flux are well-correlated, this association may not be causal. Heating of the soil is primarily caused by solar radiation and consequently, soil temperature and light availability to canopies are often correlated at diurnal (e.g., Thomas et al., 1996) and annual time scales (Lloyd and Taylor, 1994). The relative importance of temperature and

* FAX No: 5106436264. E-mail: jcraine@socrates.berekeley.edu

light in correlative studies may be difficult to separate. The responses of cellular processes to changes in temperature are thought to be immediate and are consistent with the results of lab and field observations that show strong correlations between short-term changes in temperature and the two components of soil CO_2 flux: root-associated activity (respiration of recently fixed carbon by roots, mycorrhizae, and the rhizosphere) and the decomposition of litter and/or soil organic matter (Kirschbaum, 1995; Peterjohn et al., 1994; Zak and Pregitzer, 1995). Yet the relationships between temperature and soil CO_2 flux are often derived at monthly time scales, long enough for roots to acclimate to changes in temperature (Amthor, 1994; Bryla et al., 1997; Ryan, 1991). Although soil temperature and soil CO_2 flux may be generally well-correlated, this relationship may not adequately predict soil CO_2 flux under situations where the relationship between light and soil temperature is altered (e.g., climate warming).

Ecophysiological studies consistently demonstrate tight linkage between photosynthesis and carbohydrate supply to roots as and between carbohydrate supply and root respiration (Amthor, 1994; Lambers et al., 1996; Szaniawski and Kielkiewicz, 1982). Therefore, studies of factors affecting root carbohydrate status, such as light or defoliation, often find that factors influencing carbohydrate supply explain patterns of root respiration better than does temperature (Bassirirad et al., 1993; Fukuoka et al., 1996; Percival et al., 1996). Autotrophic respiration is thought to comprise 40–60% of total soil CO_2 flux (Raich and Schlesinger, 1992), an assertion supported by physiological studies (Bouma et al., 1996; Van der Werf et al., 1996) and empirical studies (Ewel et al., 1987; Haynes and Gower, 1995; Lamade et al., 1996; Landsberg and Waring, 1997). It is reasonable to predict that controls over plant carbon gain and allocation could provide key links between light availability, its interception and soil CO_2 flux. This ecophysiological perspective has generally not been applied to ecosystem studies of soil CO_2 flux.

To address the influence of factors that affect carbohydrate source on soil CO_2 flux at the daily time scale, we examined the effect of decreased carbohydrate source on soil CO_2 flux and root respiration in an annually burned, nitrogen-limited grassland through manipulations of light intensity and removal of aboveground biomass. We also quantified the contribution of root respiration to soil CO_2 flux and examined correlations between soil CO_2 flux and soil temperature, air temperature and root biomass. We hypothesized that with decreased light supply or removal of biomass, soil CO_2 flux would decline. If declines in soil CO_2 flux in our experiments are driven by decreased carbohydrate supply rates, they should correspond to declines in the respiration rates of roots.

Materials and methods

Study area

The study was performed in late July and early August (1997) at the Cedar Creek Natural History Area in east-central Minnesota, USA (mean annual temperature = 5.5 °C, mean annual precipitation = 726 mm). Cedar Creek lies on a glacial outwash sandplain and, prior to settlement, had upland vegetation dominated by oak savanna, oak woodlands and tallgrass prairie. Plots for this study were located in a former agricultural field that was plowed, repeatedly disked, and planted to native prairie grasses three years prior to the study (1994). Soils in the field were sandy (94% sand, 6% silt plus clay), with low soil carbon (0.45% C in 0–20 cm horizon). The predominant grass in the plots was *Schizachyrium scoparium* (Michx.) Nash (little bluestem), a native perennial bunchgrass with the C_4 photosynthetic pathway. Cover was noncontinuous with *S. scoparium* clumps approximately 50 cm apart and surrounded by mostly bare ground. Previous studies at Cedar Creek indicate that *S. scoparium* can approach steady-state belowground biomass within three years in experimental monocultures or prairie restoration plots (Tilman and Wedin, 1991). Thus, root biomass values and aboveground vegetative cover in our plots at the time of the study were comparable to areas with *S. scoparium*-dominated vegetation in late successional old fields and native prairies at Cedar Creek. Because the field had been burned in the spring, aboveground dead biomass in the plots was minimal. The period of the experiments corresponded to the period of peak aboveground live biomass in C_4-dominated vegetation at Cedar Creek.

Clipping experiment

In the clipping experiment, we established 28 1 × 2 meter plots, separated by 1 m buffer strips on July 22, 1997. Each plot was separated into two 1 × 1 m subplots that were randomly assigned to be clipped or left unclipped as control subplots. At the beginning of the experiment, we clipped the vegetation in four subplots

to a height of 2 cm and their measured parameters in two locations in the clipped subplots and one in the control subplots in each of the four plots (see below). These four plots were then remeasured followed by the addition of four new plots, whose clipped subplots had just been clipped. This serial addition of plots continued for the entire experiment, such that 16 plots were clipped the first day, 8 the second day, and 4 the third day. This experimental design reduces the correlation between the time since a subplot was clipped and the time into the experiment. Therefore, trends in abiotic factors (e.g. weather) that occur over the course of the experiment can be better distinguished from trends associated with increasing time since a subplot was clipped.

Prior to the clipping of a subplot, a *S. scoparium* clump was identified in the central area of each subplot and marked. After clipping, plastic (PVC) collars were inserted into the ground on top of the marked clumps 'clip') and 5 cm away from the edge of the clump ('clip-adjacent'). These two treatments would allow examination of the response of soil CO_2 flux to clipping directly on top of plants as well as adjacent to them. In the unclipped control subplots, one collar was placed 5 cm away from of the clump ('control-adjacent') to serve as a control for the clip-adjacents. Measurement of soil CO_2 flux requires that there is no aboveground vegetation at the spot of measurement. As it is necessary to remove any aboveground vegetation from the point of measurement, there could be no control for measurements made on top of clumps that were clipped. Collars were 10 cm across and 5 cm high and inserted approximately 2.5 cm into the ground. Measurements of soil CO_2 flux were performed with the LI-COR 6200 gas exchange system (LI-COR, Lincoln. NE, USA) fitted with the LI-COR 6400-09 soil respiration chamber. At the beginning of each measurement, we measured the ambient CO_2 concentration at the surface of the soil and then scrubbed CO_2 from the chamber using soda lime such that the concentration of CO_2 was approximately 20–30 ppm below ambient concentrations. CO_2 flux was measured during 6 five-second observations over 90 s during which CO_2 concentration increased from sub-ambient to supra-ambient concentrations. In addition, we measured the temperature of the soil at 10 cm ($Tsoil_{10}$) and chamber air temperature (Tair). All measurements took place between the hours of 8:00 and 20:00. Measurements were temporarily halted on the first and third days due to rain. Rain was unlikely to strongly affect CO_2 diffusion rates in these porous, sandy soils and should not differentially affect treatments. Estimated soil CO_2 flux for each measureme was calculated at ambient CO_2 concentration from the linear regression of soil CO_2 flux and CO_2 concentrations over time for each measurement using S-Plus 4.0 (MathSoft).

On the morning following the last measurements, we collected cores that were 5 cm in diameter and 25 cm deep from the center of each collar. This depth contains approximately 90% of the total root biomass for vegetation at this site dominated by *S. scoparium* (Wedin, unpublished data). Roots were washed free of soil and chilled in an insulated container. Within 12 hours, the respiration of the root sample was determined at 25 °C at approximately 370 ppm with an ADC LCA3 infrared gas exchange system (Analytical Development Company, Hoddesdon, UK) in a PLC-C conifer chamber with the system in an open configuration. This system has been used previously to measure rates of root respiration in the past (Reich et al., 1998). Flow rates were set to 350 mL min^{-1}, and magnesium perchlorate removed water vapor from the analyzer airstream. The entire sample of roots from the core, or a representative subsample, was placed in the chamber. We recorded the respiration rate after the differential between CO_2 concentrations entering and leaving the chamber stabilized (typically four minutes). Roots were then dried at 60 °C for 72 h and weighed. From these data, we calculated the dry mass of roots per unit ground area, respiration per gram dry root (specific root respiration, SRR), and the respiration of roots on a ground-area basis.

Shading experiment

For the shade experiment, we chose 24 *S. scoparium*-dominated plots in a nearby 15 × 150 m area of the same field. Each 4 m × 4 m plot contained three 1.5 × 1.5 m subplots, separated by a 0.5 m buffer, that were randomly assigned to three treatments: 'shade', 'sun' and 'shade-sun'. Shading was applied to shade and shade-sun plots for two days (August 9–10, 1997) before measurements began for this experiment on August 11. The shade treatment consisted of double-layer 80% black shade cloth that provided a 95% reduction in light intensity. The shade cloth was supported with a 50 cm tall frame, and the edges of the shade cloth extended to the ground on three sides with the northern face open. The sun subplot was unmanipulated and served as the control. The shade-sun treatment was identical to the shade treatment, but immediately

before a plot was first measured, we removed the shade cloth and the subplot remained unshaded for the duration of the experiment. This treatment allows examination of the response of soil CO_2 flux following an increase in light supply after shading.

Just as in the clipping experiment, at the beginning of the measurement period four plots were measured, then immediately remeasured, followed by four additional plots. These eight plots were then measured again followed by four new plots. Serial addition of plots continued in this manner such that sixteen plots were added to the measurement cycle the first day and eight new plots added the second day. Before a plot was measured, collars were placed 5 cm away from the edge of a *S. scoparium* clump in the center of each subplot. In the four shade-sun subplots, shade cloths were removed at this time. The measurement protocol for Tair, Tsoil10, soil CO_2 flux, and root biomass were the same as in the clipping experiment. No measurements of root respiration were made on these samples.

The first two days of the experiment, during which the shade and shade-sun treatments were applied, were cloudless (1600 μmol m^{-2} s^{-1} photosynthetically active radiation (PAR)). The third and fourth days of the experiment during which all measurements were taken were cloudy, with values for PAR approximately 400 μmol m^{-2} s^{-1} at peak intensity, except during the last measurement cycle when there was low cloud cover and higher light intensities observed (PAR not measured). Measurements were halted for 1–2 h midmorning each day due to rain. While soil moisture was not measured in either experiment, rain was frequent during this period, and there was no obvious relationship between rain events and soil CO_2 flux, suggesting that soil moisture had minimal effect on respiration or CO_2 diffusion during the experimeni and should have had little relative effect vith different treatments.

Analysis

For each experiment, we used ANOVA (JMP-IN 3.1.5, SAS Institute) to compare parameters between different treatments. The ratio of root respiration to soil CO_2 flux was calculated for each collar in the clipping experiment and then the treatment mean calculated from the average of the ratios. Also, for each experiment, we regressed the plot means of soil CO_2 flux taken over the entire measurement period against the relevant explanatory variables (see below). These regressions were also performed on the last set of measurements taken during each experiment. Analysis of the data for all measurements allows comparisons that are representative of the environmental conditions over the entire experiment, while analysis of the last data set is more directly comparable to root respiration values derived from excised roots. We also performed repeated measures analysis over the entire data set with within- and between-plot differences determined using the ANOVA procedure of S-Plus 4.0, including an error term based on plot. For both experiments differences were minor between the results of the within-plot variation of the repeated measures analysis and the plot means for the regression analysis. The results of the regression of soil CO_2 flux and plot means are presented and discussed to facilitate interpretation.

For the clipping experiment, the soil CO_2 flux was regressed with all measured independent parameters: soil temperature at 10 cm ($TSoil_{10}$), temperature of the air in the respiration chamber (TAir), biomass of roots per unit area (RootMass), specific root respiration (SRR), time since the beginning of the experiment (ExperimentalTime), time since the plot was clipped (TimeSinceClip), Treatment, and the interactions between the continuous variables and Treatment. For the shade experiment, soil CO_2 flux was regressed with $Tsoil_{10}$, Tair, RootMass, ExperimentalTime, the time since the shade cloth was removed (TimeSincePull), Treatment, and the interactions between the continuous variables and Treatment. The distribution of data for each parameter was centered at zero by adding a constant to each value, except for temporal data for which a constant was added to each data point such that the first measurement occurred at time zero. With this centering technique, the intercept of the model represents the average flux rate for all treatments at the beginning of the measurement period at mean values for all other measured parameters.

Only significant parameters ($p < 0.05$) were allowed to remain in the final regression model after stepwise deletion of non-significant parameters, except for Tair in the regression of data from the last cycle of the clipping experiment. In this model, soil CO_2 flux appeared to decrease with increasing Tair, which was significantly correlated with TimeSinceClipping ($r = -0.39$, $p < 0.001$), an apparent consequence of increasing temperature during this period and plots having been measured in order of TimeSinceClip. The relationship between Tair and soil CO_2 flux only affected those plots that were clipped and not unclipped plots, indicating that TimeSinceClip was the driving variable and the relationship with Tair coincidental.

Removing Tair from the model produced a model for which TimeSinceClip became significant and the resultant r^2_{adj} decreased by only 0.02.

Standard errors reported are based on pooled variance. For the results of the regression, if treatment was significant in the model we also present the value of the treatment effect for each treatment. Combination of this constant and the value of the intercept provides the value for the soil CO_2 flux rates at the beginning of the measurement period at mean parameter values for each treatment. If an interaction term between treatment and a parameter was significant, then the specific value of this interaction for each treatment is also presented. Combination of this value with the value of the relationship between soil CO_2 flux and the parameter for which the interaction with treatment is significant provides the relationship between soil CO_2 flux and the parameter for each treatment. This process can be considered to adjust the slope of the relationship between the pararmeter and soil CO_2 flux for the specific treatment.

Results

Clipping experiment

Analysis of the data from the last set of measurements of soil CO_2 flux (taken on average approximately 24 h after clipping) revealed that soil CO_2 flux was 19% less adjacent to clipped plants than adjacent to unclipped plants, even though these treatments did not significantly differ in soil temperature, air temperature, or root biomass (Table 1). Respiration rate per unit dry mass of excised root (SRR) was approximately 14% less adjacent to clipped plants than adjacent to unclipped plants and root respiration comprised a similar proportion (38–40%) of total soil CO_2 flux in the two treatments (Table 1). This implies that microbial respiration would have decreased also with clipping. These results indicate that clipping not only affected root respiration but also total belowground respiration through its effect on carbohydrate supply rather than through its effect on environment or on root biomass.

Beneath clipped plants all of the soil CO_2 flux is accounted for by root respiration (Table 1). This large value may be unrealistically high as the roots beneath a clump of *S. scoparium* are composed of mainly crowns and other coarse roots and these roots may respond to the measurement process by increasing their respiration rates. However, it is not unreasonable that these roots are relatively non-mycorrhizal and decompose slowly and the large value calculated indicative of a large fraction of the soil CO_2 flux beneath plants mainly being generated by respiring roots and not microbes.

Five factors were significant in modeling experimental mean soil CO_2 flux in the clipping experiment: TAir, $TSoil_{10}$, Treatment, TimeSinceClip, and the interaction between Treatment and TimeSinceClip (Table 2a) There was no significant difference between the values of the Treatment effect for clip-adjacent (-0.51 ± 0.30) and control-adjacent (-0.70 ± 0.30) plants. The difference between these two values represents the difference between the soil CO_2 flux at the beginning of the experiment for all plots and the individual treatment. The lack of a significant difference between the two values reveals that there was no difference in the soil CO_2 flux rate at the time of clipping for these two treatments. Thus, any differences between these two treatments were not due to initial differences.

Combining the value for the TimeSinceClip parameter and the value of the Control-adjacent*TimeSinceClip provides the relationship between soil CO_2 flux and TimeSinceClip for the control-adjacent treatment. This shows that there was no marked change in soil CO_2 flux over the three days of the experiment for control-adjacent plots (0.03 μmol m^{-2} s^{-1} d^{-1}). The analogous process for clip and clip-adjacent treatments reveals that the soil CO_2 flux directly beneath and adjacent to the aboveground parts of the bunchgrass decreased at similar absolute rates after clipping (-1.47 and -1.41 μmol m^{-2} s^{-1} d^{-1}, respectively).

Since the value of the relationship between soil CO_2 flux and Tair was negative, soil CO_2 flux decreased with increasing Tair. Conversely, soil CO_2 flux increased with increasing $Tsoil_{10}$ Results for regressions performed on the values from the last set of measurements taken during the experiment were similar to the model of the experimental mean soil CO_2 flux, except that no temperature component was significant in the model (Table 2b).

No root parameters were significant components of the model of soil CO_2 flux in the clipping experiment. This does not mean that there is no relationship between root mass or root respiration rates and soil CO_2 flux, since there were significant correlations between mean soil CO_2 flux and both root biomass and SRR (data not shown). However, the root effects are mostly contained in the treatment effect. Removal of Treatment from the model produces a model that in-

Table 1. Effect of clipping on soil CO_2 flux, temperature of the air (Tair), temperature of the soil at 10 cm ($Tsoil_{10}$), root mass per unit ground area (Rootmass), the specific rate of respiration of excised roots (SRR), the rate of root respiration per unit ground area (RootResp), and the ratio of root respiration to soil CO_2 flux (% soil CO_2 flux from roots). Treatments include measurements made on top of clipped plants (clip), adjacent to the clipped plants (clip-adjacent) and adjacent to unclipped plants in the control plots (control-adjacent). Treatment means and standard errors are provided for (1) the entire data set where data was first averaged for all measurements taken in a plot's treatment (entire set), and (2) the last set of measurements of the experiment (last set). Significant differences ($p < 0.05$) between treatments are denoted by different superscript letters

Parameter	Units	Data set	Clip	Clip-adjacent	Control-adjacent
Soil CO_2 flux	μmol m^{-2} s^{-1}	last set	6.58 ± 0.24^a	4.85 ± 0.14^b	6.01 ± 0.15^c
Soil CO_2 flux	μmol m^{-2} s^{-1} m2	entire set	7.6 ± 0.2^a	5.7 ± 0.2^b	6.5 ± 0.2^c
Tair	°C	last set	26.0 ± 0.3^a	26.3 ± 0.3^a	26.1 ± 0.3^a
Tair	°C	entire set	24.8 ± 0.2^a	25.0 ± 0.2^a	24.9 ± 0.2^a
$Tsoil_{10}$	°C	last set	22.7 ± 0.1^a	22.7 ± 0.1^a	22.5 ± 0.2^a
$Tsoil_{10}$	°C	entire set	22.1 ± 0.1^a	22.1 ± 0.1^a	21.9 ± 0.1^a
Rootmass	g m^{-2}		1307 ± 116^a	201 ± 29^b	201 ± 15^b
SRR	nmol g^{-1} s^{-1}		5.67 ± 0.54^a	10.2 ± 0.6^b	11.9 ± 0.4^c
RootResp	μmol m^{-2} s^{-1}		6.58 ± 0.43^a	1.82 ± 0.20^b	2.40 ± 0.21^b
% soil CO_2 flux from roots		last set	100 ± 8^a	38 ± 4^b	40 ± 4^b

cludes these two parameters as significant (results not shown).

Shading experiment

Averaging measurements of soil CO_2 flux for a subplot over the entire shading experiment, soil CO_2 flux was 35–39% lower for plots that were shaded than plots that were unshaded, regardless of whether the shade cloth was removed during the experiment or not (Table 3). There was on average less than 1°C difference in soil temperature at 10 cm between treatments during the time of measurement (Table 3) despite an average of 10 °C daily variation (data not shown). There were no significant differences in root biomass between treatments (Table 3). These results show that, as in the clipping experiment, shading influenced soil CO_2 flux primarily through its effect on carbohydrate supply as we measured little effect on environment.

Four factors were significant components of the model of soil CO_2 flux averaged over the entire experiment: Treatment, $Tsoil_{10}$, Rootmass, and the interaction between $Tsoil_{10}$ and Treatment (Table 4b). The Treatment effect shows that at the beginning of the measurements unshaded subplots had a higher soil CO_2 flux than did the subplots that were shaded during the previous two days of the experiment (shade-sun, shade treatments), with no significant difference between subplots where shade cloths were removed in the experiment and subplots where the shade cloths remained for the entire experiment (Table 4b). The lack of response from removal of the shade cloth is probably associated with the overcast conditions of this portion of the experiment, such that any increase in soil CO_2 flux would have been too small to detect.

There were relationships between temperature and soil CO_2 flux associated with temporal changes during the experiment, but these were variable in sign and magnitude. Results for regressions performed on the values from the last set of measurements taken during the shading experiment were similar to the model of the experimental mean soil CO_2 flux, except the sign of the relationship between $Tsoil_{10}$ and soil CO_2 flux was positive (Table 4a), whereas in the model of the last set of measurement the sign of the relationship was negative (Table 4b). We observed that for the last set of measurements, air and soil temperature declined while cloud cover declined causing an increase in light intensity. This may have caused the apparent negative relationship between soil CO_2 flux and soil temperature.

Discussion

Our results show that factors affecting plant canopy carbon status and carbohydrate supply to roots had large effects on soil respiration in this grassland

Table 2. Results of regression of soil CO_2 flux and significant independent variables for the last set of measurements made in the clipping experiment (Table 2a) and the treatment means over the entire experiment (Table 2b). The coefficients presented represent the relationship between soil CO_2 flux that given parameter, the intercept represents the average soil CO_2 flux rate for all treatments at the beginning of the measurement period at mean values for all other measured parameters. If Treatment and/or the interaction between Treatment and another parameter was significant ($p < 0.05$) in the model, values for the specific treatment are presented for each treatment (see text for details). Standard error values are derived from the pooled variance. The r^2_{adj} for the last set of measurements and entire data set were 0.48 and 0.43, respectively

	Coefficient	*F* ratio	*p* value
(a)			
Intercept	7.35 ± 0.11		<0.05
Tair	−0.24 ± 0.11	4.9	<0.05
$Tsoil_{10}$	1.55 ± 0.43	12.6	<0.001
Treatment		8.5	<0.001
clip-adjacent	−0.51 ± 0.30		<0.1
clip	1.21 ± 0.29		<0.001
control-adjacent	−0.70 ± 0.30		<0.05
TimeSinceClip	−0.93 ± 0.29	10.3	<0.01
TimeSinceClip * Treatment		3.9	<0.05
TimeSinceClip * clip-adjacent	−0.54 ± 0.36		<0.13
TimeSinceClip * clip	−0.48 ± 0.36		<0.19
TimeSinceClip * control	0.96 ± 0.36		<0.01
(b)			
Intercept	6.39 ± 0.24		<0.001
Treatment		6.4	<0.01
clip-adjacent	−0.61 ± 0.35		<0.1
clip	1.21 ± 0.34		<0.001
control-adjacent	−0.60 ± 0.34		<0.1
TimeSinceClip	−0.40 ± 0.15	7.1	<0.01
TimeSinceClip * Treatment		3.2	<0.05
TimeSinceClip * clip-adjacent	−0.22 ± 0.21		<0.3
TimeSinceClip * clip	−0.31 ± 0.21		<0.13
TimeSinceClip * control	0.53 ± 0.21		<0.01

ecosystem, independent of the physical environment (temperature). Ecophysiological studies consistently show that factors affecting photosynthesis and whole-plant carbon gain strongly influence carbon allocation belowground and root respiration at the daily time scale. This study clearly links whole plant carbon gain and soil CO_2 flux, which has been historically modeled ignoring factors that are associated with carbohydrate sources to roots. Although we did not measure root respiration in the shade experiment, the results are consistent with Fukuoka et al. (1996), where shaded cabbage seedlings had lower resuts of root respiration than unshaded seedlings. This would imply that in our shading experiment, the decreases in soil CO_2 flux in shaded plots partly were due to decreases in root respiration. The ecophysiological controls over whole plant carbon gain may be more important than is generally recognized in ecosystem studies of soil CO_2 flux.

Root respiration comprised 38–40% of soil CO_2 flux for areas adjacent to clipped and unclipped plants, although total soil CO_2 fluxes were approximately 20% less in the adjacent to clipped plants. The estimate of 40% of soil CO_2 flux for root respiration agrees with reported values. Physiological studies estimate approximately 40–60% of the carbon allocated to root is respired, depending on the species and nu-

Table 3. Effect of shading on soil CO_2 flux, temperature of the air (Tair), temperature of the soil at 10 cm ($Tsoil_{10}$), and root mass per unit ground area ($Rmass{\cdot}m^{-2}$). Treatments include measurements made from subplots that were unshaded for the entire experiment (Sun), shaded for the entire experiment (Shade), and shaded for the a portion of the experiment and left unshaded for the duration of the experiment (Shade-sun). Treatment means and standard errors are provided for 1) the entire data set where data was first averaged for all measurements taken in a plots treatment (entire set), and 2) the last set of measurements of the experiment (last set). Significant differences ($p < 0.05$) between treatments are denoted by different superscript letters

Parameter	Units	Data set	Sun	Shade-sun	Shade
Soil CO_2 flux	$\mu mol\ m^{-2}\ s^{-1}$	last set	8.89 ± 0.57^a	6.57 ± 0.43^b	5.41 ± 0.36^b
Soil CO_2 flux	$\mu mol\ m^{-2}\ s^{-1}$	entire set	9.28 ± 0.68^a	6.02 ± 0.44^b	5.66 ± 0.36^b
Tair	°C	last set	29.2 ± 0.31^a	28.6 ± 0.31^a	26.8 ± 0.4^b
Tair	°C	entire set	27.3 ± 0.3^a	26.2 ± 0.26^b	25.6 ± 0.47^b
$Tsoil_{10}$	°C	last set	31.0 ± 0.3^a	30.8 ± 0.37^a	30.1 ± 0.28^b
$Tsoil_{10}$	°C	entire set	28.1 ± 0.47^a	27.6 ± 0.48^a	27.2 ± 0.46^a
Rootmass	$g\ m^{-2}$		735 ± 77^a	793 ± 109^a	605 ± 72^a

Table 4. Results of regression of soil CO_2 flux and significant independent variables for the last set of measurements made in the experiment (Table 4a) and the treatment means of all measurements made over the entire experiment in a plot (Table 4b) of the shading experiment. Values of coefficients for intercepts, treatments and interaction terms are as in Table 2. The r^2_{adj} for the last set of measurements and entire data set were 0.61 and 0.55, respectively

	Coefficient	*F* ratio	*p* value
	(a)		
Intercept	6.36 ± 0.24		<0.001
Treatment		10.4	<0.001
shade	-0.68 ± 0.33		<0.05
shade-sun	-0.88 ± 0.33		<0.01
sun	1.56 ± 0.33		<0.001
$Tsoil_{10}$	0.46 ± 0.1	21.0	<0.001
Rootmass	2.2 ± 0.5	16.5	<0.001
$Tsoil_{10}$ * Treatment		4.8	<0.05
$Tsoil_{10}$ * shade	-0.26 ± 0.14		<0.1
$Tsoil_{10}$ * shade-sun	-0.16 ± 0.14		<0.2
$Tsoil_{10}$ * sun	0.42 ± 0.14		<0.01
	(b)		
Intercept	6.96 ± 0.21		<0.001
Treatment		25.8	<0.001
shade	-1.64 ± 0.32		<0.001
shade-sun	-0.46 ± 0.31		<0.13
sun	2.10 ± 0.32		<0.001
Rootmass	2.1 ± 0.5	16.4	<0.001
$Tsoil_{10}$	-0.54 ± 0.14	14.7	<0.001

trient availability (Van der Werf, 1996). This ratio would be the ratio of root respiration to soil CO_2 flux at steady-state with no transfer of carbon to mycorrhizae or exudation. Reported values for the ratio of

root respiration (or root-associated respiration) to soil respiration range from 24% to 75% (Horwarth et al., 1994; Lamade et al., 1996; Raich and Potter, 1995).

We could not separate the contribution of mycorrhizal respiration, respiration of exudates, and decomposition of soil organic matter (SOM) to soil CO_2 flux, but our data suggest that these components of hetrotrophic respiration also respond sensitively to plant transport of carbohydrates below ground. Decomposition is often considered to be independent of plant activity, although there is evidence to the contrary (Zagal, 1994). Although recalcitrant SOM fractions with turnover rates on the scales of decades to millennia comprise the bulk (>90%) of SOM, these fractions contribute little to heterotrophic soil respiration. Schimel et al. (1994) estimated that 80% of heterotrophic soil respiration came from cletrital and microbial pools, which comprise less than 10% of SOM. Changes in the relative abundance and/or physiological status of a dominant grass can lead to relatively rapid changes in the small fraction of soil organic matter that dominates microbial activity (Holland et al., 1992; Seastedt et al., 1991; Wedin and Pastor, 1993).

Relationships between soil CO_2 flux and temperature, when they existed, were variable in sign and magnitude depending on the time period, time scale, and experiment. Although models based on temperature rather than light can often accurately explain patterns of soil CO_2 flux (e.g. Keith et al., 1997), there has been little investigation on the influence of the temporal resolution of these models and their robustness to changes in the correlation between parameters, such as light and temperature. This is important, for example, in analyses of climate warming, which would entail an increase in temperature without a change in light supply. In such a scenario, the pre-warming relationship between temperature and soil CO_2 flux may not apply.

The work in this study has strong implications for the measurement and modeling of soil CO_2 flux. Methodologically, accurate measurement of soil CO_2 flux in grasslands requires removal of a portion of the canopy since soil CO_2 flux in the clipping experiment was greater directly beneath the aboveground parts of the bunchgrass. Yet, removing a portion of the canopy has consequences for soil CO_2 flux. Therefore, measurements of soil CO_2 flux in which the canopy is removed need to account for the time since removal of the aboveground vegetation.

For modeling soil CO_2 flux, this work indicates that measurement of factors that affect the rate at which carbohydrates are metabolized by roots need to be joined with measurement of factors that affect the supply of carbohydrates to roots. More complete mechanistic analyses of soil CO_2 flux need to include factors that affect total carbohydrate transfers to roots, such as light availability and leaf area.

Acknowledgements

We would like to thank Paul Fine, Rachel Gallery, Jenny Goth, and Rachel Melzer for their assistance in the field, Mark Tjoelker for assistance with determination of root respiration, Paul Grogan and two anonymous referees for reviewing this manuscript, and the Cedar Creek LTER for establishment and maintenance of the study site. JMC acknowledges support by the National Science Foundation for a graduate fellowship.

References

Amthor J S 1994 Plant respiratory responses to the environment and their effects on the carbon balance. *In* Plant-environment interactions. Ed. R E Wilkinson. pp 501–54. Marcel Dekker, Inc, New York.

Bassirirad H, Caldwell M M and Mott K A 1993 Effect of root cooling on photosynthesis of *Artermisia tridentata* seedlings under different light levels. Bot. Acta 106, 22–227.

Bouma T J, Broekihuysen A G M and Veen B W 1996 Analysis of root respiration *Solanum tuberosum* as related to growth, ion uptake and maintenance of biomass. Plant Physiol. Biochem. 34, 795–806.

Bryla D R, Bouma T J and Eissenstat D M 1997 Root respiration in citrus acclimates to temperature and slows during drought. Plant Cell Environ. 20, 1411–1420.

Ewel K, Cropper W and Gholz H 1987 Soil CO_2 evolution in Florida slash pine plantations. II. Importance of root respiration. Can. J. For. Res. 17, 330–333.

Fukuoka N, Yoshioka H, Shimizu E and Fujiwara T 1996 Effect of shading cabbage seedlings on their physiological processes and rooting ability after transplanting to the nursery. J. Japan. Soc. Hortic. Sci 65, 545–551.

Ham J M, Owensby C E, Coyne P I and Bremer D J 1995 Fluxes of CO_2 and water vapor from a prairie ecosystem exposed to ambient and elevated atmospheric CO_2. Agric. For. Meteorol. 77, 73–93.

Haynes B E and Gower S T 1995 Belowground carbon allocation in unfertilized and fertilized red pine plantations in northern Wisconsin. Tree Physiol. 15, 317–325.

Holland E A, Parton W J, Detling J K and Coppock D L 1992 Physiological responses of plant populations to herbivory and their consequences for ecosystem nutrient flow. Am. Nat. 140, 685–706.

Horwath W R, Pregitzer K S and Paul E A 1994 ^{14}C allocation in tree-soil systems. Tree Physiol. 14, 1163–1176.

Keith H, Raison R J and Jacobsen K L 1997 Allocation of carbon in a mature eucalypt forest and some effects of soil phosphorus availability. Plant Soil 196, 81–99.

Kirschbaum M U F 1995 The temperature dependence of soil organic matter decomposition, and the effect of global warming on soil organic C storage. Soil Biol. Biochem. 27, 753–760.

Lamade E, Djegui N and Leterme P 1996 Estimation of carbon allocation to the roots from soil respiration measurements of oil palm. Plant Soil 181, 329–339.

Lambers H, Scheurwater I and Atkin O K 1996 Respiratory patterns in roots in relation to their functioning. *In* Plant roots: the hidden half Ed. Y Waisel, A Eshel and U Kafkafi. pp 323–362. Marcel Dekker, Inc., New York.

Landsberg J J and Waring R H 1997 A generalized model of forest productivity using simplified concepts of radiation-use efficiency, carbon balance and partitioning. For. Ecol. Manag. 95, 209–228.

Lloyd J and Taylor J A 1994 On the temperature dependence of soil respiration. Funct. Ecol. 8, 315–323.

MacDonald N W, Zak D R and Pregitzer K S 1995 Temperature effects on kinetics of microbial respiration and net nitrogen and sulfur mineralization. Soil Sci. Soc. Amer. J. 59, 233–240.

Percival D C, Proctor J T A and Tsujita M J 1996 Whole-plant net CO_2 exchange of raspberry as influenced by air and root-zone temperature, CO_2 concentration, irradiation, and humidity. J. Am. Soc. Hortic. Sci. 121, 838–845.

Peterjohn W T, Melillo J M, Steudler P A, Newkirk K M, Bowles F P and Aber J D 1994 Responses of trace gas fluxes and N availability to experimentally elevated soil temperatures. Ecol. Appl. 4, 617–625.

Raich J W and Potter C S 1995 Global patterns of carbon dioxide emissions from soils. Global Biogeochem. Cycles 9 23–36.

Raich J W and Schlesinger W H 1992 The global carbon dioxide flux in soil respiration and its relationship to vegetation and climate. Tellus Ser. B Chem. Phys. Meteorol. 44, 81–99.

Reich P B, Walters M B, Tjoelker M G, Vanderklein D, Buschena C 1998 Photosynthesis and respiration rates depend on leaf and root morphology and nitrogen concentration in nine boreal tree species differing in relative growth rate. Funct. Ecol. 12, 395–405.

Ryan M G 1991 Effects of climate change on plant respiration. Ecol. Appl. 1, 157–167.

Seastedt T R, Parton W W and Ojima D S 1992 Mass loss and nitrogen dynamics of decaying litter of grassland: The apparent low nitrogen immobilization potential of root detritus. Can. J. Bot. 70, 384–391.

Schimel D S, Braswell B H, Holland E A, McKeown R, Ojima D S, Painter T H, Parton W J and Townsend A R 1994 Climatic, edaphic, and biotic controls over storage and turnover of carbon in soils. Global Biogeochem. Cycles 8, 279–293.

Szaniawski R K and Kielkiewicz M 1982 Maintenance and growth respiration in shoots and roots of sunflower plants grown at different root temperatures. Physiol. Plant. 54, 500–504.

Thomas S M, Whitehead D, Adams J A, Reid J B, Shetlock R R and Leckie A C 1996 Seasonal root distribution and soil surface carbon fluxes for one-year-old *Pinus radiata* trees growing at ambient an elevated carbon dioxide concentration. Tree Physiol. 16, 1015–1021.

Tilman D and Wedin D 1991 Plant traits and resource reduction for five grasses growing on a nitrogen gradient. Ecology 72, 685–700.

Townsend A R, Vitousek P M Desmarais D J and Tharpe A 1997 Soil carbon pool structure and temperature sensitivity inferred using CO_2 and $^{13}CO_2$ incubation fluxes from five Hawaiian soils. Biogeochemistry 38, 1–17.

Van Der Werf A 1996 Growth, carbon allocation, and respiration as affected by nitrogen supply: aspects of the carbon balance. *In* Dynamics of Roots and Nitrogen in Cropping Systems of the Semi-Arid Tropics Eds. O Ito, C Johnasen, J J Adu-Gyamfi, K Katayama, J V D K Kumar Rao, T J Rego. pp 145–158. Japan International Research Center for Agricultural Sciences.

Wedin D A and Pastor J 1993 Nitrogen mineralization dynamics in grass monocultures. Oecologia 96, 186–192.

Zagal E 1994 Influence of light intensity on the distribution of carbon and consequent effects on mineralization of soil nitrogen in a barley (*Hordeum vulgare* L.)-soil system. Plant Soil 160, 21–31.

Zhang Y, Reed D D, Cattelino P J, Gale M R, Jones E A, Liechty H O and Mroz G D 1994 A process-based growth: model for young red pine. For. Ecol. Manag. 69, 21–40.

Section editor: H Lambers