Elevated Atmospheric Co2 Effects and Soil Water Feedbacks on Soil Respiration Components in a Colorado Grassland

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Elevated atmospheric CO$_2$ effects and soil water feedbacks on soil respiration components in a Colorado grassland

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[1] The shortgrass steppe is a semi-arid grassland, where elevated CO$_2$ reduces stomatal conductance and promotes soil moisture storage. Enhanced biomass growth from elevated CO$_2$ has been attributed in part to soil moisture effects. However, the influence of this soil moisture feedback on C cycling has received little attention. We used open-top chambers to increase atmospheric CO$_2$ concentrations to twice-ambient for four growing seasons. Soil respiration rates and stable C isotopes of soil CO$_2$ were measured during the third and fourth seasons. Elevated CO$_2$ increased soil respiration rates by $\sim$25% in a moist growing season and by $\sim$85% in a dry season. Stable C isotope partitioning of soil respiration into its components of decomposition and rhizosphere respiration was facilitated on all treatments by a ^{13}C disequilibrium between currently growing plants and soil organic matter. Decomposition rates were more than doubled by elevated CO$_2$, whereas rhizosphere respiration rates were not changed. In general, decomposition rates were most significantly correlated with soil temperature, and rhizosphere respiration rates were best predicted by soil moisture content. Model simulations suggested that soil moisture feedbacks, rather than differences in substrate availability, were primarily responsible for higher total respiration rates under elevated CO$_2$. By contrast, modeling demonstrated that substrate availability was at least as important as soil moisture in driving CO$_2$ treatment differences in soil organic matter decomposition rates.   *INDEX TERMS:* 1610 Global Change: Atmosphere (0315, 0325); 1615 Global Change: Biogeochemical processes (4805); 1851 Hydrology: Plant ecology; 1866 Hydrology: Soil moisture; *KEYWORDS:* decomposition, rhizosphere respiration, stable isotopes, ^{13}C/^{12}C, soil C cycling, shortgrass steppe

1. Introduction

[2] Atmospheric concentrations of carbon dioxide (CO$_2$) are expected to double within the next century [*Houghton et al.*, 2001], and will doubtlessly bring about changes in the functioning and structure of ecosystems. The primary direct effect of elevated CO$_2$ on terrestrial ecosystems is growth enhancement, resulting from increased C assimilation rates of most plants, which may or may not be transient. Numerous indirect effects have been documented in a variety of experiments at the ecosystem level, including stimulation of the rate of C cycling through soils [*Hungate et al.*, 1997b; *Lin et al.*, 1999; *Pendall et al.*, 2001b]; alteration of the soil N cycle [*Hungate et al.*, 1997a; *Thornley and Cannell*, 2000; *Hu et al.*, 2001]; changes in soil microbial communities, including mycorrhizal symbionts [*Rice et al.*, 1994; *Williams et al.*, 2000]; improved soil/plant water relations and increased soil water use efficiency [*Owensby et al.*, 1999; *Morgan et al.*, 2001; *Volk et al.*, 2000]; and shifts in aboveground species composition [*Owensby et al.*, 1999; *Morgan et al.*, 2003]. Feedbacks among these various indirect effects have the tendency to confound predictions about the influence of elevated CO$_2$ on C cycling.

[3] Despite a recent emphasis on research on the belowground responses to elevated CO$_2$, many questions regarding the fate of C assimilated by plants remain unresolved. Many studies report increased rates of soil respiration and a general enhancement of belowground C cycling [*Zak et al.*, 2000]. However, quantifying the individual components of the soil respiration flux, rhizosphere respiration (including root respiration and microbial turnover of recent rhizode-

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posits) and organic matter decomposition, is critical for predicting soil C sequestration potential [*Canadell et al.*, 1996; *Edwards and Norby*, 1999]. These two components may respond to climatic changes and feed back on C cycling in contrasting ways. For example, increased rates of rhizosphere respiration might be predicted under elevated CO_2 as a result of increased belowground C allocation, root growth and root turnover, but would have no net effect on C storage in soil [*Cheng and Johnson*, 1998; *Edwards and Norby*, 1999]. If organic matter decomposition is stimulated in response to a substrate induced priming effect (i.e., stimulation of microbial decomposition rates by enhanced availability of labile substrates), net losses of soil C are possible if inputs do not keep pace [*Jenkinson*, 1966; *Zak et al.*, 1993; *Rice et al.*, 1994; *Hungate et al.*, 1997b]. Organic matter mineralization would have a positive feedback on plant growth if nutrients were mobilized, allowing continued C inputs to soil [*Hu et al.*, 1999]. On the other hand, limited soil N availability may suppress decomposition rates and favor soil C storage [*Hu et al.*, 2001].

[4] Most field methods currently available for partitioning soil respiration components disturb the roots or rhizosphere, whether it be measuring specific respiration rates from excavated root systems, killing aboveground vegetation, or creating root-exclusion zones [*Edwards and Norby*, 1999; *Hogberg et al.*, 2001; *Hanson et al.*, 2000]. An exception is the use of stable isotopes in experiments where growing plants can attain an isotopic composition that is distinct from that of soil organic matter [*Balesdent et al.*, 1988; *Rochette and Flanagan*, 1997; *Lin et al.*, 1999, 2001]. The stable isotope method allows partitioning of the soil CO_2 flux into "old," or pre-label, and "new," or current growing season, components [e.g., *Pendall et al.*, 2001b]. In this paper, the old C flux is referred to as decomposition of pre-existing soil organic matter (SOM), and the new C flux is referred to as rhizosphere respiration. Strict separation of heterotrophic and autotrophic respiratory components is not possible with isotopes, because the rhizosphere microbial community rapidly acquires the isotopic signature of current photosynthate, for example, from turnover of labile root exudates (however, see *Cheng et al.* [1994]). Nonetheless, this method of partitioning provides a relatively easy way to follow new C inputs derived from elevated CO_2 treatments through the plant-soil system, because the CO_2 used for fumigation is usually derived from ^{13}C depleted fossil fuel sources [e.g., *Leavitt et al.*, 2001]. It has also been successfully used in agroecosystems where C_4 crops are planted into C_3 soils [*Balesdent et al.*, 1988; *Rochette and Flanagan*, 1997].

[5] Soil moisture is often conserved under elevated CO_2 because reduced stomatal conductance enhances the water-use efficiency of plants [e.g., *Morgan et al.*, 2001]. Soil moisture content is known to influence soil respiration rates, although the nature of the relationship between water content and CO_2 flux is poorly quantified [*Davidson et al.*, 2000]. Increased soil moisture content under elevated CO_2 was invoked as the mechanism stimulating gross N mineralization in a California annual grassland [*Hungate et al.*, 1997a]. Microbial activity in a tallgrass prairie was found to be enhanced under elevated CO_2 when soils were

dry [*Williams et al.*, 2000]. One goal of the current research is to evaluate the role of soil water feedbacks to C cycling under elevated CO_2 on the semiarid shortgrass steppe.

[6] Model predictions of the response of soil respiration to elevated CO_2 have rarely been tested. Here we present comparisons of measured soil respiration and decomposition fluxes with modeled fluxes using the DAYCENT parameterization (S. J. Del Grosso et al., Modeling CO_2 emissions from decomposition of soil organic carbon, manuscript in preparation, 2003) (hereinafter referred to as Del Grosso et al., manuscript in preparation, 2003). This empirically based model assumes that the maximum rate of decomposition is controlled by labile C substrates, with temporal variations driven by soil temperature and limited by insufficient soil moisture. Comparing model results with observations allows us to evaluate mechanisms responsible for changes in soil respiration and decomposition rates.

[7] We hypothesized that elevated CO_2 may stimulate soil respiration and decomposition rates in a semiarid grassland soil in Colorado. Aboveground and belowground biomass production have been stimulated by 15–35% by doubled CO_2 in this open-top chamber experiment [*Morgan et al.*, 2001, 2003]. A 5‰ ^{13}C disequilibrium between currently growing plants and existing SOM has resulted at the site because of a reduction in livestock grazing ∼20 years ago. We were therefore able to partition the soil respiration flux into new and old components in all four treatments, elevated CO_2 chambers (EC), ambient CO_2 chambers (AC), non-chambered control plots (NC), and fallow plots without vegetation (Fallow).

2. Methods

2.1. Field Methods

[8] The experiment was conducted in the shortgrass steppe region of northeastern Colorado, at the USDA-ARS Central Plains Experimental Range (CPER; latitude 40°40′N, longitude 104°45′W), about 55 km northeast of Fort Collins. The most abundant species at the study site were the C_4 grass, *Bouteloua gracilis* (H.B.K) Lag. (blue grama), and the C_3 grasses *Stipa comata* Trin and Rupr. (needle-and-thread grass) and *Elymus smithii* (Rydb.) Gould (western wheatgrass). Root biomass (including crowns) is responsible for ∼70% of net primary production (NPP) in this ecosystem: belowground biomass NPP averages 223 g m^{-2} yr^{-1}, root crowns contribute 57 g m^{-2} yr^{-1}, while aboveground biomass NPP averages 109 g m^{-2} yr^{-1} [*Milchunas and Lauenroth*, 2001]. The long-term mean annual precipitation averages 320 mm, and mean annual temperature averages 15.6°C in summer and 0.6°C in winter [*Lauenroth and Milchunas*, 1991]. The soil at the site was classified as an Ustollic Camborthid, in the Remmit fine sandy loam series. Soil water content at field capacity was determined to be ∼18%, and at the wilting point, ∼4% by volume [*Morgan et al.*, 2001].

[9] Beginning in 1997, open-top chambers (4.5m diameter) were used to evaluate the effects of CO_2 on the shortgrass steppe ecosystem, with three replicate chambers at ambient (360 ± 20 ppmv) and elevated (720 ± 20 ppmv) CO_2. Three unchambered plots of the same area allowed

evaluation of any chamber effects. Chambers were placed on the plots before growth started in late March or early April, and removed at the end of the growing season in late October. Blowers with ambient or elevated CO_2 ran continuously. Fans placed at chamber outlets maintained pressure equilibrium, which was monitored on several occasions; no significant pressure differentials were detected. The experimental and chamber design was described in detail by *Morgan et al.* [2001]. In addition to the three main treatments, a "Fallow" plot was established in spring, 1999, to evaluate the contribution of recent C substrates to heterotrophic decomposition. This plot, ~100 m from the OTC plots, was trenched to 1 m depth, lined with weed barrier, and sprayed with a broad-spectrum herbicide to kill all vegetation.

[10] Beginning in 1999, soil gas samples were collected from stainless steel tubes (1/8" OD) inserted horizontally 15 cm into a pit face at 6 depths (3, 5, 10, 15, 25 and 50 cm). The tube ends that extended into the soil were perforated over ~5 cm. The tubes were bent at the pit face and reached ~10 cm above the surface. They remained closed with plugs between sampling times. Tubes were installed in the autumn before sample collection began, to allow equilibration after soil disturbance. For sampling, we removed the plugs and inserted a water trap made of a 10-cm long piece of glass filled with magnesium perchlorate that was capped on one end with a rubber septum. A glass syringe was used to remove 1.5 times the volume of air in each tube (+ water trap). After waiting at least 30 min for the diffusion gradient to stabilize, soil gas samples were collected into gas-tight glass syringes (greased with Apiezon M). Volumes of 10 cc were collected at all depths, except for the 3-cm depth, where 6 cc were collected, to ensure that no atmospheric air was pulled into the syringe. Atmosphere samples were also collected on each sampling date, from each chamber or plot into 0.5-L flasks at ~1.5 m above the ground, after flushing ~10 flask volumes.

[11] Soil temperature was recorded continuously in each plot at 5-cm depth. Volumetric soil moisture content in the top 15 cm was measured by time domain reflectometry (TDR) on a weekly basis (except for a gap during 2000 due to instrument failure). These values were used in calculation of soil diffusivity and for modeling soil CO_2 fluxes (see below).

2.2. Analysis of CO_2 and $\delta^{13}C$

[12] Soil gas samples were analyzed within 24 hours of collection for CO_2 concentration using an infrared gas analyzer with a precision of ± 3 μmol mol^{-1} over a concentration range of 360 to 8000 μmol mol^{-1} (Model LI-6251, LICOR, Inc., Lincoln, NE). A power function was developed using four standards to calculate unknown concentrations. Soil gas samples were analyzed for stable isotopes of CO_2 using gas chromatography-isotope ratio mass spectrometry (GC-IRMS; Isoprime model, Micromass, U.K.) [*Miller et al.*, 1999]. Tests indicated that standards stored in these greased syringes kept for up to one week without significant leakage or isotopic exchange. To ensure a linear response of the mass spectrometer, sample sizes varying from ~7 to ~250 μL of soil gas were injected into a carrier gas stream, which was further split before being introduced

into the mass spectrometer. This allowed the peak height of the sample to be within ~10% of the peak height of the standard; precision of $\delta^{13}C$ in soil CO_2 was better than 0.1‰. Flasks of atmospheric air were analyzed for CO_2 mixing ratio by infrared absorption (precision 0.1 μmol mol^{-1} [*Conway et al.*, 1994]) and for $\delta^{13}C$ by dual-inlet mass spectrometry (precision 0.01‰ [*Trolier et al.*, 1996]).

2.3. Data Analysis

[13] We used soil CO_2 gradients and diffusivities to calculate belowground respiration rates. This approach ensured that aboveground biomass was not disturbed, and that aboveground respiration of growing vegetation was not included, as it would be in a chamber method. Diffusivity was calculated from soil temperature and moisture using an approach that accounts for inter- and intra-aggregate differences in diffusion rates [*Potter et al.*, 1996]. CO_2 gradients were determined by linear regression using values from the atmospheric flasks and soil depths of 3, 5, 10 and 15 cm. Linear fits to the data were significant 86% of the time (n = 144 profiles, with 4 or 5 data points); poor linear fits usually resulted from sample loss (<4 data points) or soils that were wet at the surface.

[14] A "Keeling" plot approach was used to estimate the $\delta^{13}C$ value of soil respiration ($\delta^{13}C_{SR}$) [*Pendall et al.*, 2001b; *Keeling*, 1958, 1961], in order to eliminate the influence of variable amounts of atmospheric CO_2 that diffuse down into the soil profile. When $\delta^{13}C$ is plotted against the inverse of the CO_2 concentration, the y-intercept reflects the flux weighted average $\delta^{13}C$ value of the biological source of CO_2, resulting from root/rhizosphere respiration and microbial decomposition. The intercepts were calculated using geometric means to account for variability in both independent and dependant variables [*Sokal and Rohlf*, 1995]. We subtracted 4.4‰ from the y-intercept values to account for kinetic fractionation during diffusion because the soil CO_2 was sampled from within the soil rather than from the soil surface [*Cerling*, 1984; *Amundson et al.*, 1998].

[15] The soil respiration flux was partitioned into "new" and "old" components using a simple two-component mixing model [*Balesdent et al.*, 1988; *Pendall et al.*, 2001b],

$$\delta^{13}C_{SR} = F_{old}(\delta^{13}C_{old}) + (1 - F_{old})(\delta^{13}C_{new}), \qquad (1)$$

where F_{old} is the proportion of CO_2 generated by decomposition of intermediate-pool C. Aboveground samples of the dominant grass species were collected from each chamber or plot periodically in 1999 and 2000 for analysis of $\delta^{13}C$ by elemental analyzer-IRMS. These $\delta^{13}C$ values, weighted by aboveground biomass amounts, were used as the "new" C end-member signatures ($\delta^{13}C_{new}$). The three dominant species maintain similar shoot:root ratios at ambient and elevated CO_2, and we assumed that root activities were proportional to biomass [*Morgan et al.*, 1994]. Root biomass was not used as the "new" end-member because roots have long residence times (4–6 years [*Milchunas and Lauenroth*, 2001]). Soil samples collected at the end of the 1999 growing season were subjected to long-term laboratory incubations after removing roots [*Paul*

et al., 2001; *Pendall et al.*, 2001a]. Average $\delta^{13}C$ values (n = 4 for each of 2 depths) of CO_2 evolved after 100 days of incubation (representing decomposition of intermediate pool C) were used as the "old" C end-member signatures ($\delta^{13}C_{old}$). End-members on Fallow plots were assumed to be the same as on NC plots. Partitioning of the soil respiration flux on EC treatments was facilitated by the depleted CO_2 used for elevating the concentration. On AC, NC and Fallow treatments, partitioning was made possible by a reduction in grazing at our field site 20 years prior to the experiment. When grazing pressure is reduced in shortgrass steppe, C_3 grass abundance tends to increase [*Milchunas et al.*, 1988]. A land-use change disequilibrium in ^{13}C of 5‰ has resulted because there is a lag between the time vegetation changes and when SOM reflects that change.

2.4. Modeling of Soil Respiration and Decomposition

[16] The DAYCENT abiotic CO_2 flux submodel calculates the relative effects of soil temperature and moisture stress on soil respiration and decomposition fluxes [*Parton et al.*, 1998; Del Grosso et al., manuscript in preparation, 2003]. We used soil temperature measured at 5 cm depth, and volumetric soil moisture data corrected to Relative Water Content (RWC = (%H_2O-Wilting Point)/(Field Capacity-Wilting Point)) as the main model drivers for each treatment (except the Fallow plot, which was not monitored for soil moisture or temperature). The model is not scaled to an absolute flux rate, which is determined by labile C availability for decomposition and root activity for root respiration. Therefore, we optimized a site-specific multiplier for each treatment by comparing the ratio of observed and simulated CO_2 flux rates. A single multiplier of 0.68 was sufficient for scaling the model to the observations for all three treatments for total soil respiration.

[17] Our isotopically derived decomposition observations include only decomposition of "old" SOM, including substrates older than ~2 years, but the DAYCENT model simulates total decomposition, including labile substrates. For comparison between measured and modeled values, we normalized our observed values to include heterotrophic respiration of labile ("new") C as well as more refractory compounds: we simply divided the F_{old} values from AC, EC and NC by the Fallow plot F_{old} values. This assumed that during the first summer following herbicide application, the Fallow plot respiration rate represented total decomposition, and that the proportion of "new" C contributing to respiration rates on Fallow plots was similar to the other plots. As with total soil respiration, we calculated a multiplier for model simulations of each treatment to account for differences in labile C; NC simulations were multiplied by 1.19, AC by 0.88, and EC by 1.52. These factors were based on ratios of average (normalized) observations to average simulations, and allowed us to evaluate relative changes in decomposition rates driven by differences in soil temperature and RWC.

2.5. Statistics

[18] Differences in soil respiration rates and F_{old} between CO_2 treatments were evaluated using repeated measures analysis of variance (ANOVAR) for 1999 and 2000 separately, and using ANOVA on individual dates [*Sokal and Rohlf*, 1995]. Post-hoc probability values were calculated by Fisher's PLSD, and significance was evaluated at P < 0.05, unless otherwise noted. Uncertainties in estimating F_{old} were determined by accounting for variability in all of the components, including $\delta^{13}C_{SR}$, $\delta^{13}C_{old}$, and $\delta^{13}C_{new}$, following a first-order Taylor series approach [*Phillips and Gregg*, 2001]. Uncertainties in the end-members were estimated as follows: $\delta^{13}C_{new}$ error was the square root of the sum of the squared standard errors of % biomass and $\delta^{13}C$ of the dominant grass species; $\delta^{13}C_{old}$ error was the standard error associated with laboratory incubations; and $\delta^{13}C_{SR}$ error was the standard error of the least squares intercepts of Keeling plots.

3. Results

3.1. Site Conditions and Soil Respiration Rates

[19] Precipitation amounts in the two years under investigation contrasted strongly; for 1999, the total was 557 mm (nearly twice the annual average of 320 mm); for 2000, it was 311 mm, much of which fell after mid-August. Volumetric soil water content in the top 15 cm (θ) at the OTC site reflected a very wet spring in 1999, and a very dry spring and summer in 2000 (Figure 1a). During the growing season of 1999, θ was significantly lower in AC (8.4 ± 0.6%) than NC (12.9 ± 1.8%) or EC (11.6 ± 1.3%) treatments (P < 0.001 for both comparisons). During the growing season of 2000, θ was significantly lower in AC (5.1 ± 0.9%) than EC (7.8 ± 1.5%) but not NC (7.0 ± 1.6%) (P < 0.05). The difference in θ (lower in the AC treatment) carried through the unchambered winter period of 1999–2000. During the first 2 years of the shortgrass steppe (SGS) OTC experiment, 1997–1998, similar treatment effects on θ were observed [*Morgan et al.*, 2001]. Soil temperatures at 5 cm depth tended to be slightly higher in the chambered treatments (AC and EC) than in the NC treatment, except during the unchambered winter period (Figure 1b). During the growing season of 1999, AC and EC treatments both averaged 23.8 ± 0.5°C, and NC averaged 23.1 ± 0.4°C (P > 0.05). During the growing season of 2000, AC averaged 23.1 ± 0.3°C, EC averaged 23.3 ± 0.1°C, and NC averaged 22.0 ± 0.4°C, with AC and EC significantly higher than NC (P < 0.001).

[20] Bimodal peaks of soil respiration in summer of both years, centered around day of year (DOY) 150 and again at DOY 225–250, reflect the influence of soil moisture conditions (Figures 2a and 2b). On DOY 146 in 1999 and 227 in 2000, rates from all treatments were very low, probably because measurements were made immediately following a large rainstorm, when diffusivities of CO_2 were very low. Soil respiration rates were significantly higher in NC than AC or EC plots in 1999 (P < 0.05), and rates in EC were almost always higher than in AC plots (Figure 2a; P > 0.05). In 2000, EC plots had significantly higher respiration rates than AC plots during the middle of the growing season (Figure 2b; P < 0.10 for all dates marked with an asterisk). These dates correspond to a period when soil moisture was substantially higher in EC than AC treatments (Figure 1a). In 1999, the Fallow plot generally had soil respiration rates lower than NC plots, but similar to or higher than AC or EC
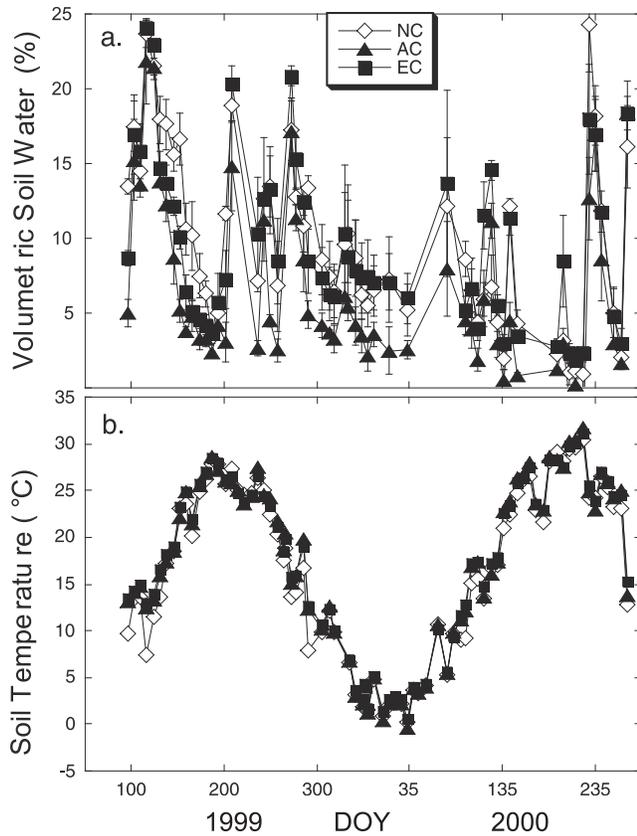
Figure 1. Soil water and temperature during the study period in Elevated CO_2 chambers (EC), Ambient CO_2 chambers (AC), and Non-Chambered control plots (NC). (a) Volumetric water content in top 15 cm as measured by TDR. (b) Soil temperature measured at 5 cm depth. DOY is day of year.

Figure 2. Soil respiration rates measured by the flux-gradient method for (a) 1999 and (b) 2000. Asterisks denote dates when the High CO_2 chambers had significantly higher fluxes than Ambient chambers ($P < 0.1$). Treatments are abbreviated as in Figure 1 and include the Fallow plot where living vegetation was removed. Error bars represent standard error of the measurement.

plots (Figure 2a). In 2000, the Fallow plot had soil respiration rates similar to NC plots (Figure 2b).

[21] Total C fluxes from the growing seasons of 1999 and 2000, the winter/spring of 1999–2000, and the year from May, 1999 to April, 2000 were estimated by integrating the area under the curves in Figure 2 (Table 1). The largest stimulation of soil respiration occurred during the growing season of 2000, when EC flux rates were ~85% higher than AC rates. Over the annual period (1999 DOY 146 to 2000 DOY 145), EC flux rates were about 30% greater than AC rates.

[22] In general, soil respiration rates showed seasonal patterns, with lowest values in winter, and highest values in summer, demonstrating the primary influence of temperature and plant phenology (Figures 2a and 2b). Significant exponential relationships between soil respiration and soil temperature were found for EC and NC ($P < 0.01$), but the relationship was marginal for the AC treatment ($P = 0.055$). Soil moisture alone (at least in the top 15 cm) was not an adequate predictor of temporal variations in soil respiration rate.

[23] Model simulations of soil respiration rates tended to agree with the general pattern of field measurements,
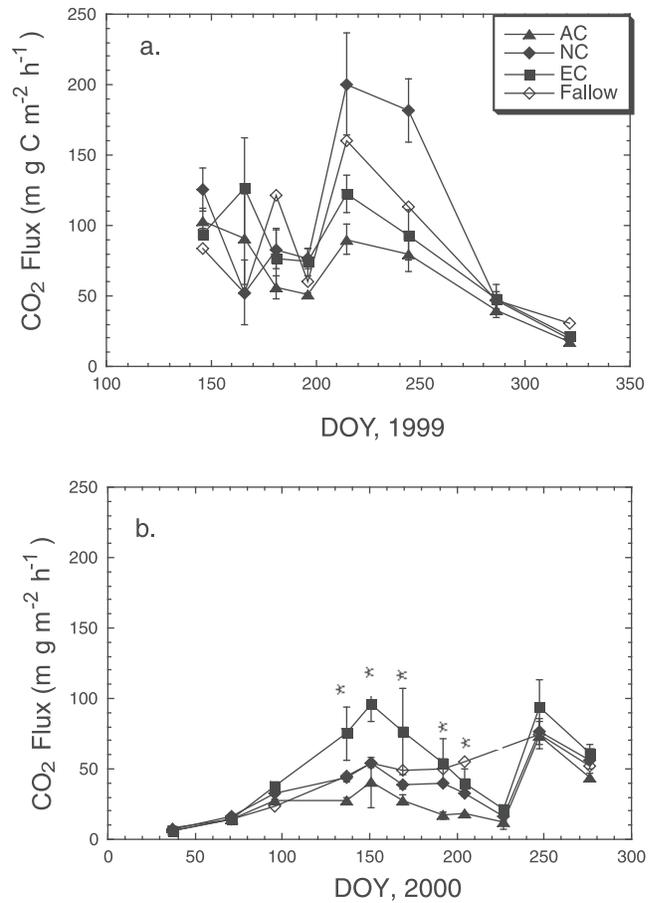
capturing seasonal variations due to temperature as well as soil moisture (Figure 3). In particular, the model was able to simulate differences between EC and AC respiration rates, especially during the dry summer of 2000. The model

Table 1. Growing Season and Annual Soil Respiration Flux Rates, kgC m^{-2} yr^{-1}, and the Ratio of EC:AC Rates[a]

	Growing Season		Winter/Spring	Annual
	1999	2000	1999–2000	1999–2000
	Treatment			
AC	0.233[b]	0.107[b]	0.113[b]	0.346[b]
EC	0.295[b]	0.197[c]	0.163[b]	0.458[b]
NC	0.392[c]	0.141[b]	0.138[b]	0.530[b]
	Ratio			
EC:AC	1.27	1.84	1.44	1.32

[a]Growing season fluxes calculated for DOY 146–276; Winter/Spring flux calculated for DOY 277 (1999) to DOY 145 (2000). Annual total = growing season 1999 plus Winter/Spring 99–00.

[b,c]Values within a column followed by the same superscript are not significantly different from each other (ANOVAR, $P > 0.05$).

also correctly simulated high respiration rates from NC plots in 1999.

3.2. The δ^{13}C of Atmosphere, Plants, and Soil Respiration

[24] δ^{13}C values of CO$_2$ in ambient air collected over NC and Fallow plots (~1.5 m above the ground) averaged −8.06 ± 0.20‰ during the growing season in 1999, and −8.35 ± 0.10‰ during the October−March dormant period. Air in the AC treatment was well within the standard deviations of the NC and Fallow plot air, averaging −8.07 ± 0.28 in the growing season of 1999, and −8.38 ± 0.12‰ during the dormant (non-chambered) period. Air in the EC treatment reflected the ^{13}C-depleted CO$_2$ that was used to double the ambient CO$_2$ concentration, averaging −24.67 ± 1.43‰ during summer of 1999. EC air samples collected during the dormant (non-chambered) period were statistically the same as air from the other treatments (−8.30 ± 0.13‰). In April of 2000, a CO$_2$ tank was delivered from a ^{13}C-enriched CO$_2$ well, and δ^{13}C values of EC air averaged −7.29 ± 0.32‰ until the tank was replaced in mid-June. The changing δ^{13}C values introduced a major element of variability to the δ^{13}C values of the plants growing in EC chambers, and we therefore restrict our discussion of stable isotope results to data from 1999.

[25] In 1999, plants were sampled at the peak of growth (DOY 201) and in the fall after growth had ceased (DOY 307). Aboveground plant biomass data and δ^{13}C values of new C inputs are shown in Table 2. Aboveground rather than belowground biomass and isotopic compositions were used because the belowground biomass reflects C inputs derived from the last 4−6 years [Milchunas and Lauenroth, 2001]. This long residence time made it impossible to use root δ^{13}C values as "new" end-members; roots do not reflect newly assimilated C. The δ^{13}C value of EC air imparted a strongly ^{13}C-depleted tracer to the plants, while δ^{13}C values of AC and NC plants were not statistically different (Table 2; t-test, P > 0.05 for C$_3$ and C$_4$ groups). We

Table 2. Aboveground Biomass and δ^{13}C Values in July 1999 (Averages of three Replicates With Standard Errors Shown in Parentheses) and Estimated δ^{13}C Values of "New" C Inputs From Rhizosphere Respiration (Standard Error Includes Variability in δ^{13}C Values and% Biomass)

Treatment	C$_3$ Biomass, %	δ^{13}C C$_3$, ‰	δ^{13}C C$_4$, ‰	δ^{13}C "New" C, ‰
AC	72 (3)	−24.1 (0.85)	−15.4 (0.21)	−21.6 (0.88)
EC	81 (4)	−42.5 (1.85)	−33.2 (0.81)	−40.7 (2.02)
NC	67 (7)	−26.0 (0.26)	−15.4 (0.27)	−22.5 (0.39)

did not observe significant changes in proportional biomass or plant δ^{13}C values over the course of the growing season. In NC plots in 2000, δ^{13}C values of S. comata leaves collected at 7 times from June 1 to October 4 varied by only ±0.17‰ (data not shown). This temporal variability falls well within the range of spatial variability observed among replicates, and is included in our error estimates (Table 2).

[26] At all sampling times, soil CO$_2$ concentration increased, and δ^{13}C values of soil CO$_2$ decreased, with increasing depth, as predicted by diffusion theory (e.g., Figures 4a and 4b) [Cerling, 1984]. For each sampling date, we plotted the δ^{13}C values of soil CO$_2$ against the inverse of CO$_2$ mixing ratios, and calculated the y-intercept (using geometric means), which represents the biological source, δ^{13}C$_{SR}$ (e.g., Figure 4c) [Keeling, 1958, 1961]. For a given treatment, low values correspond to greater plant activity; higher values to relatively more decomposition. In general, in 1999, δ^{13}C$_{SR}$ was highest in the Fallow treatment, slightly lower in AC and NC treatments, and lowest in EC treatments (Figure 5). In all treatments, δ^{13}C$_{SR}$ was lowest around DOY 150−170 and again around DOY 215-245.

3.3. Decomposition, Rhizosphere Respiration and Their Environmental Controls

[27] Decomposition of SOM and root/rhizosphere respiration both contributed to the soil respiration flux. These components were quantified using the two-part mixing model (equation (1)). The δ^{13}C values of "new" C, derived from root/rhizosphere respiration, are shown in Table 2. The δ^{13}C values of "old" C averaged −17.8 ± 0.5‰ on NC soils, −17.0 ± 0.5‰ on AC soils, and −21.9 ± 1.5‰ on EC soils, reflecting decomposition of mineralizable SOM at an approximate steady state during laboratory incubations. We assumed that these values represent "old" C available for microbial decomposition in the field. We note that these "old" C values are always more ^{13}C depleted than bulk SOM, which reflects the historically higher proportion of C$_4$ grasses at the site.

[28] The fraction of soil respiration from decomposition of "old" C (F$_{old}$) was higher on EC than AC treatments throughout the growing season (Figure 6; P = 0.001), and greater on NC than AC treatments (P = 0.077). F$_{old}$ on Fallow plots was higher than on all other treatments (Figure 6; Table 3; P < 0.05), and averaged <100% because δ^{13}C$_{SR}$ values were also influenced by "new" C (decomposition of recently growing plants, root turnover, etc.). We normalized F$_{old}$ values of the other treatments for each date
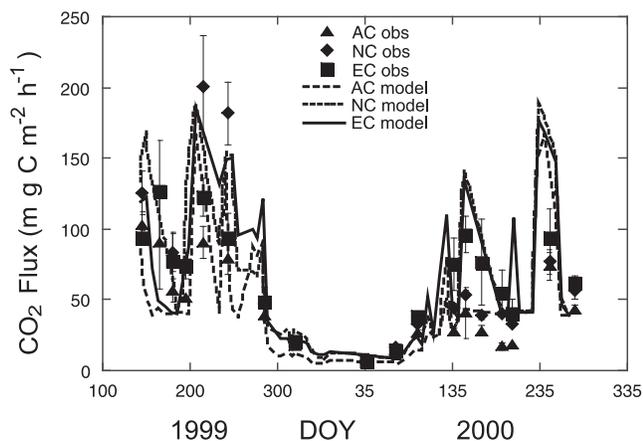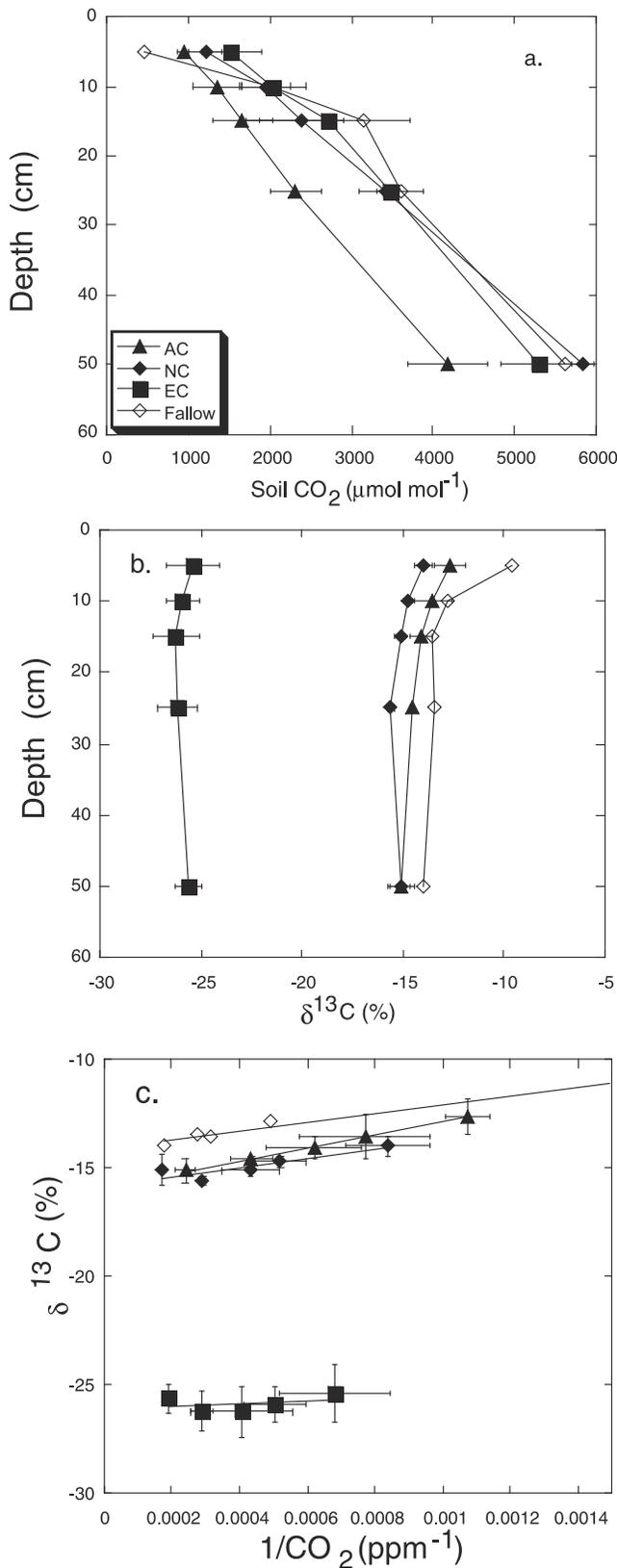

Figure 3. Soil respiration rates modeled by DAYCENT [Parton et al., 1998; Del Grosso et al., manuscript in preparation, 2003] ("Model," lines) compared with measured rates ("Obs," symbols). Treatments are abbreviated as in Figure 1.

in 1999 to the Fallow values to estimate the total proportion of decomposition (including turnover of recent substrates): normalized values averaged 50% for NC, 30% for AC, and 75% for EC treatments. These field based measurements suggest that total decomposition was more than doubled by elevated CO_2 (Table 3).

[29] We multiplied normalized F_{old} values by the soil respiration rates to compare our measurements with model simulations of heterotrophic respiration (including decomposition of recent substrates; Figure 7a). Derivation of different scaling factors ("multipliers") for each treatment suggests that substrate availability (in addition to soil temperature and moisture content) was partly responsible for differences in mean decomposition rates between treatments. Decomposition measurements early in the growing season were higher than model predictions, possibly demonstrating a greater importance of substrate induced decomposition (priming) when soils were moist and root exudation may have been highest. Later in the growing season, modeled values were close to observations. Modeled values for 2000 assumed the same scaling factors used for 1999, and suggest that EC treatments continued to have higher decomposition rates than AC treatments, in part because EC soils were wetter (Figure 7b).

[30] Growing season C losses via decomposition of old C, and rhizosphere respiration of new C, were estimated by integrating the area under the curves defined by the observations for 1999 (Table 4). Total decomposition loss (old flux) from EC plots was more than double that from AC plots (ANOVAR, $P < 0.001$), and similar to that from NC plots ($P > 0.05$). The amount of organic carbon in the different soils did not explain the differences in the decomposition losses, because EC and AC soils have similar SOC contents (Table 4). Total rhizosphere respiration loss (new flux) was similar from all treatments (except Fallow; Table 4). Elevated CO_2 appears to suppress rhizosphere respiration on a per unit root basis, either measured as belowground net primary production from root ingrowth cylinders, or as total root biomass C (D. Milchunas, unpublished data, 2001; Table 4). This suggests that roots grown under elevated CO_2 respire less, and/or that turnover and decomposition of "new" rhizodeposits are slower, possibly because of higher C:N ratios or differences in structural components such as lignin.

[31] We investigated the environmental controls over soil respiration components by correlating decomposition and rhizosphere respiration (old and new fluxes, respectively, as determined by isotopes) with relative water content (RWC) and soil temperature during the 1999 growing season. An exponential fit with soil temperature was the best predictor

Figure 4. (opposite) "In situ" soil gas samples collected 30 June, 1999. Samples from other dates followed the same patterns. (a) Soil CO_2 concentration with depth (note nearly constant gradients with depth); (b) $\delta^{13}C$ of soil CO_2; and (c) "Keeling" plot showing linear relationship between $\delta^{13}C$ and the inverse of CO_2 concentration. Error bars show standard deviations of the measurements ($n = 3$). Treatments are abbreviated as in Figure 1.
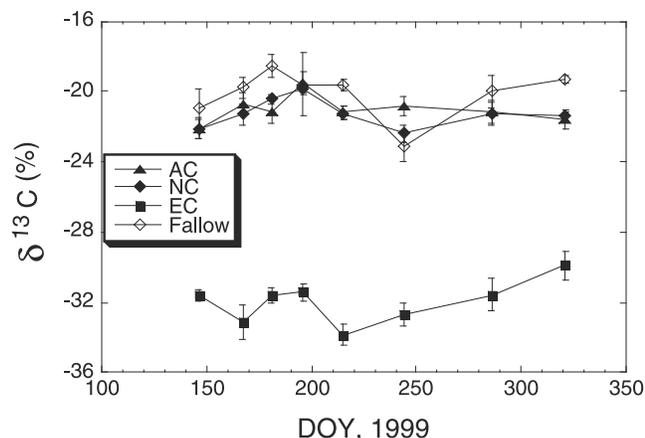
Figure 5. The $\delta^{13}C$ of soil respiration. Intercepts of "Keeling" plots (geometric mean intercepts) minus 4.4‰ are plotted for 1999. Error bars represent the standard error of the intercept. Treatments are abbreviated as in Figure 1.

of decomposition for all treatments, and with rhizosphere respiration on EC treatments (P < 0.05 for all regressions), whereas a linear fit with RWC was the best predictor of rhizosphere respiration on AC and NC treatments (P < 0.05; Table 5; Figure 8). Q_{10} values for decomposition were 4.1 on AC, 1.8 on EC, and 3.0 on NC treatments, and Q_{10} for rhizosphere respiration on EC treatments was 2.5. Relationships between decomposition and RWC, and between rhizosphere respiration and temperature on AC and NC plots, were not significant.

4. Discussion

4.1. Elevated CO_2 Effects on Total Soil Respiration

[32] Elevated CO_2 stimulated soil respiration in this experiment, but significant responses were observed only when soil moisture content was significantly greater than under ambient conditions. Respiration rates in the moist
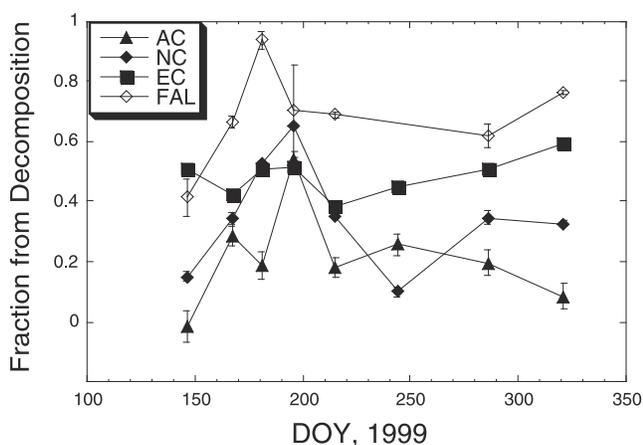

Figure 6. Fraction of soil respiration derived from "old" C (F_{old}) for 1999. Error bars represent combined errors from all steps of the partitioning [*Phillips and Gregg*, 2001]. Treatments are abbreviated as in Figure 1.

Table 3. Fraction of Soil Respiration Derived From Decomposition of "Old" SOM (F_{old}), and Those Values Divided by the Fallow Values (Normalized F_{old})[a]

Treatment	F_{old}, %	Normalized F_{old}, %
AC	21 (16)[b]	31 (24)[b]
EC	48 (7)[c]	75 (21)[c]
NC	35 (18)[bc]	50 (22)[b]
Fallow	68 (16)[d]	100

[a]Averages for the 1999 growing season. Standard deviations in parentheses (n = 8).
[b,c,d]Values within a column followed by the same superscript are not significantly different from each other (ANOVAR, P > 0.05).

growing season of 1999 were 27% higher in EC than AC treatments, when θ was ∼10% and 8%, respectively (P = 0.2), but in the dry growing season of 2000, they were 84% higher, when θ was ∼3% and 0.5%, respectively (P = 0.004). These values broadly bracket the average soil respiration stimulation by elevated CO_2 of 51% in 11 grassland species [*Zak et al.*, 2000]. In this semi-arid grassland
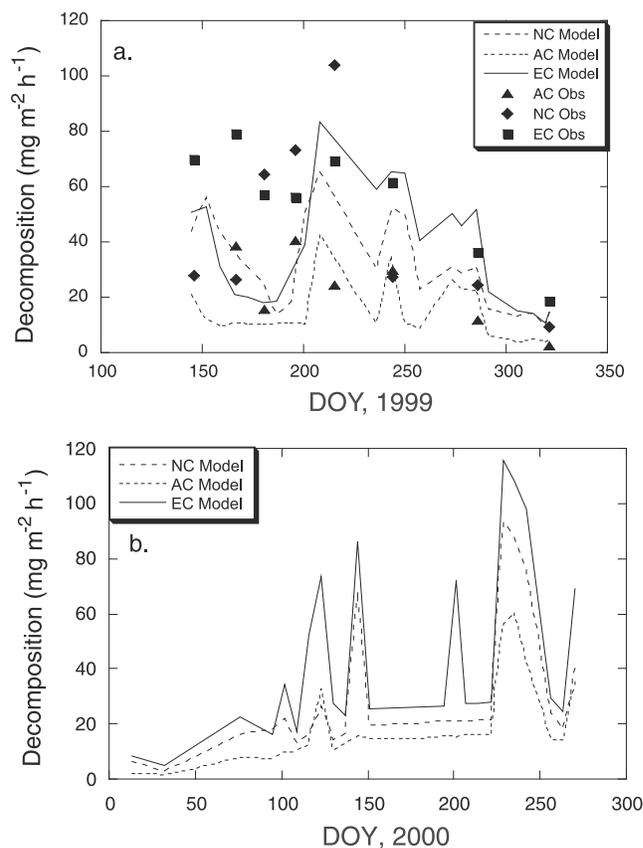
Figure 7. Decomposition rates in two growing seasons. (a) Measured ("Obs," symbols) and simulated ("Model," lines) estimates of the decomposition flux for 1999, and (b) simulated estimates for 2000. Measured values were partitioned using ^{13}C and then normalized to Fallow plot values to include all decomposition components. DAYCENT abiotic decomposition submodel was used for the simulations (Del Grosso et al., manuscript in preparation, 2003).

Table 4. Growing Season Losses by Decomposition of Old C and Rhizosphere Respiration of New C, for DOY 146-321 in 1999[a]

Treatment	Old Flux, kg C m^{-2} season^{-1}	Kg Old/kg SOC, kg C m^{-2} season^{-1} kg SOC^{-1}	New Flux, kg m^{-2} season^{-1}	Kg New/kg BNPP, kg C m^{-2} season^{-1} kg BNPP^{-1}	Kg New/kg Root C, kg C m^{-2} season^{-1} kg Root C^{-1}
AC	0.060[b]	0.014	0.208[b]	5.128	0.150
EC	0.154[c]	0.035	0.183[b]	3.303	0.126
NC	0.122[c]	0.025	0.314[b]	9.563	0.216
Fallow	0.246[d]	NA	0.110[c]	NA	NA
EC:AC	2.550	2.557	0.880	0.644	0.843

[a]Decomposition was normalized to total soil organic C (SOC) content, and rhizosphere respiration was normalized to belowground NPP (BNPP, measured as new root growth, kg C per season) and to total root C content.
[b,c,d]Values within a column followed by the same superscript are not significantly different from each other (ANOVAR, $P > 0.05$).

ecosystem, soil moisture content appears to be an important feedback on soil C cycling. This is not surprising in light of known plant physiological responses to elevated CO_2. Soil moisture conservation associated with reduced stomatal conductance under elevated CO_2 has been documented in several studies, including the present one [*Nie et al.*, 1992; *Volk et al.*, 2000; *Morgan et al.*, 1994, 2001]. At the SGS OTC site, $\delta^{18}O$ values of soil water showed that evaporation rates were lower from EC soils, probably because increased biomass and litter cover reduced bare ground [*Ferretti et al.*, 2003]. Aboveground and belowground biomass and photosynthetic rates in grassland species were found to have stronger responses to elevated CO_2 at low to moderate irrigation rates [*Volk et al.*, 2000]. Furthermore, enhanced N mineralization rates were attributed to greater soil moisture content under elevated CO_2 in a California grassland [*Hungate et al.*, 1997a]. Thus, a general stimulation of belowground C and N cycling might be expected in semi-arid regions as a result of soil moisture feedbacks to elevated CO_2.

[33] The influence of soil moisture was also seen in the bimodal peaks of soil respiration in both years; rates were high following the moist spring months of April and May, then declined through June and July, and increased again in August in response to monsoon rains. Although soil moisture by itself was not an adequate predictor of soil respiration rates, it improved soil respiration predictions when combined with soil temperature. It must be kept in mind, however, that temperature and moisture are simply drivers of plant and microbe physiological activity, which, together with substrate availability, are the ultimate determinants of soil respiration rates.

[34] Stimulation of soil respiration under elevated CO_2 has generally been attributed to increased C allocation belowground, including greater root biomass, increased fine root turnover, or possibly higher rates of root/rhizosphere respiration per unit of root material [*Fitter et al.*, 1997; *Edwards and Norby*, 1999; *Pregitzer et al.*, 2000]. In some

cases, higher decomposition rates may contribute to the stimulation of soil respiration, driven primarily by increased root exudation and rhizodeposition (the "priming effect") [e.g., *Diaz et al.*, 1993; *Billes et al.*, 1993; *Loiseau and Soussana*, 1999]. At the shortgrass steppe, soil moisture is likely to affect both decomposition and rhizosphere respiration to some degree. Methodological difficulties in separating root from microbial respiration in the field have for the most part limited the ability to ascribe mechanisms driving differences in soil respiration rates, with few exceptions [*Hungate et al.*, 1997b; *Cardon et al.*, 2001]. We discuss soil respiration components in greater detail below.

[35] Model estimates of soil respiration, based on a combination of soil temperature and moisture content, followed observed seasonal patterns and differences between treatments well. Whereas we found significant exponential relationships between soil respiration and temperature, the model used an arctangent function that allowed a variable Q_{10}, especially at lower temperatures. Nonetheless, the model fit winter data quite well. Soil temperature did not strongly differ among treatments, and a single multiplier was adequate for simulating soil respiration for all three treatments, suggesting that the main difference between treatments was driven by soil moisture, rather than by substrate availability or root activity. Exceptionally high rates of respiration from NC plots in early 1999 that were not quite matched by the model may have been driven by high soil moisture contents below 15 cm. In some instances, our measurements may have missed peaks of respiration following wetting events.

4.2. Elevated CO_2 Effects on Soil Respiration Components

[36] On the shortgrass steppe, decomposition rates were more than doubled by elevated CO_2, at a time when total soil respiration rates were not significantly altered. This occurred during a moist growing season, when soil moisture

Table 5. Relationships Between Decomposition and Rhizosphere Respiration Fluxes (mg C m^{-2} h^{-1}) and Environmental Parameters for the 1999 Growing Season[a]

Treatment	Decomposition Flux	R	P	Rhizosphere Respiration Flux	R	P
AC	C = 0.48 e^(0.14T)	0.57	0.012	C = 33.9 + 45.9 RWC	0.65	0.112
EC	C = 8.99 e^(0.06T)	0.58	0.026	C = 5.16 e^(0.09T)	0.48	0.029
NC	C = 2.13 e^(0.11T)	0.86	0.011	C = 6.96 + 127 RWC	0.77	0.024

[a]T = Soil T in °C at 5 cm; RWC = relative volumetric water content in top 15 cm; R = correlation coefficient; P = probability level.
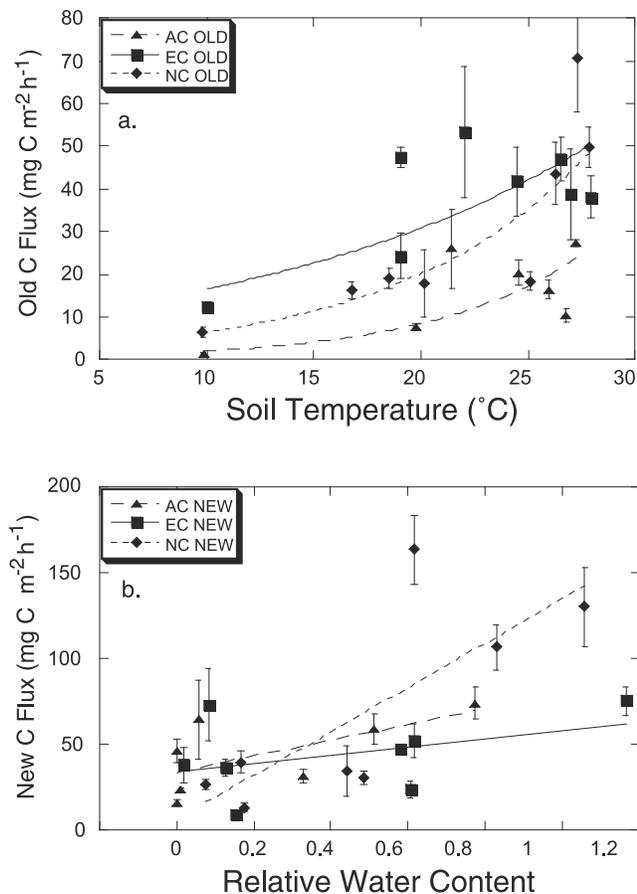
Figure 8. (a) Relationships between measured estimates of decomposition and soil temperature, and (b) relationships between measured estimates of rhizosphere respiration and relative soil water content (RWC). Functions and their associated statistics are shown in Table 5. Treatments are abbreviated as in Figure 1. Error bars represent accumulated errors [*Phillips and Gregg*, 2001].

differences between EC and AC treatments were small. Enhancement of decomposition rates was apparently driven more by differences in substrate availability than by differences in moisture or temperature between CO_2 treatments. Concurrent incubation experiments showed that active pool C was roughly doubled by elevated CO_2 [*Pendall et al.*, 2001a], and higher microbial biomass was found in soils harvested in October of 1999 (A. Parsons, unpublished data, 2001). Simulations also support the assertion that the observed differences in decomposition rates were driven by substrate availability. In contrast to total soil respiration where a single multiplier was sufficient and the model suggested that treatment differences were driven by soil water content, optimized multipliers for decomposition were different among treatments (NC = 1.2, AC = 0.9, EC = 1.5). The multipliers were derived after including the effects of temperature and water, so model results also imply that substrate availability was higher in the elevated CO_2 treatment.

[37] Increased rates of decomposition under elevated CO_2 have been considered likely because of the potential for an enhanced priming effect: added labile C belowground stimulates growth and activity of the microbial community, and decomposition of native SOM [*Jenkinson*, 1966; *Billes et al.*, 1993; *Zak et al.*, 1993]. Laboratory studies have generally shown enhancement of microbial respiration by elevated CO_2, although only a few have demonstrated significant effects [*Zak et al.*, 1993; *Rice et al.*, 1994; *Niklaus and Korner*, 1996; *Zak et al.*, 2000]. However, field studies to date show conflicting results, possibly because of methodological differences. Elevated CO_2 stimulated decomposition from a C_4 soil planted with C_3 wheat with added N fertilization, and suppressed decomposition where N was limiting [*Cheng and Johnson*, 1998]. ^{13}C-pulse labeling showed stimulation of heterotrophic respiration by elevated CO_2 only with N fertilization, and had no effect on decomposition without added N [*Hungate et al.*, 1997b]. Turnover rates of mineral organic matter, roots, and litter were stimulated by elevated CO_2 only with added N, as shown by ^{13}C values of SOM pools [*Loiseau and Soussana*, 1999]. Heterotrophic respiration was suppressed under elevated CO_2 when N was limiting [*Hu et al.*, 2001]. By contrast, elevated CO_2 suppressed decomposition in a high nutrient soil planted with annual grasses and forbs from the California grassland [*Cardon et al.*, 2001].

[38] Apparently, the N status of the ecosystem plays a key role in regulating SOM mineralization. *Parnas* [1976] postulated that greatest SOM mineralization would occur if added substrate had a C:N ratio of ~25. However, the soil N status likely interacts with the C:N ratio of substrate. It has been hypothesized that as mineral N becomes depleted, microbes begin to favor older SOM, with a lower C:N ratio, over recent substrates [*Lekkerkerk et al.*, 1990]. Interestingly, the balance of evidence suggests the opposite, i.e., that under N limitation, plants outcompete microbes for nutrients, thereby limiting decomposition [e.g., *Hu et al.*, 2001]. At the shortgrass steppe OTC experiment in 1999, the C:N ratios of new roots were ~34 on EC and ~22 on AC plots (P < 0.0001), but N concentrations in surface soils were slightly higher on EC than AC plots (P = 0.09; A. R. Mosier et al., unpublished data, 2001). Decomposition of older SOM, with C:N of ~9, was favored over new substrates with high C:N, and N was apparently mineralized, allowing continued enhanced plant growth. More accurate predictions of the effects of elevated CO_2 on decomposition may need to evaluate more factors than simply soil N status or residue C:N ratios. Additional controls over decomposition rates may include lignin content, the age and origin of pre-existing SOM, changes in soil microbial communities, and length of exposure to elevated CO_2.

[39] In our field experiment, soil temperature was the best predictor of decomposition rates for all treatments; soil water content was not well correlated with decomposition rates, nor did it improve correlations significantly when combined with temperature. This contrasts with results from the tallgrass prairie, where microbial activity (measured on disturbed samples) was correlated with soil moisture content [*Williams et al.*, 2000]. That experiment was based on 7

years of data, covering a greater range of soil moisture conditions than in the one year of our experiment. Clayey soils on the tallgrass prairie may also interact with soil moisture effects. In a laboratory incubation experiment, soil water potential was shown to influence decomposition rates on loamy, but not on sandy, soils [*Scott et al.*, 1996]. Q_{10} values for decomposition, evaluated at growing season temperatures, demonstrated a CO_2 interaction with temperature response: EC treatment had the lowest Q_{10}, and AC the highest. This suggests that a feedback mechanism may hinder temperature-driven increases in decomposition in a greenhouse world.

[40] In the shortgrass steppe OTC experiment, we were able to demonstrate that decomposition rates were stimulated under elevated CO_2 relative to ambient conditions, because in addition to the ^{13}C-depleted CO_2 added to EC chambers, a ^{13}C-disequilibrium (of $\sim5‰$) between growing plants and intermediate pool C provided a natural tracer for AC and NC treatments. Reduction of livestock grazing over the last ~20 years has favored C_3 species (especially *S. comata*) over the dominant C_4, *B. gracilis*, at our field site. Although the isotopic partitioning method we used is robust, uncertainties related to the pool of organic matter being decomposed and the mixture of C_3 and C_4 grasses contributing to rhizosphere respiration should not be neglected. We acknowledge that SOM is a complex of compounds with varying isotopic compositions, and that microbes may utilize different compounds over time and/or space as availability of C and N shifts. A steady state in the $\delta^{13}C$ value of microbially respired CO_2 after 100 days of laboratory incubation [*Pendall et al.*, 2001a] defined the pool of ''intermediate'' SOM used in decomposition. Probably the largest contributor to unquantified uncertainties in this study relates to variable activity of the dominant C_3 and C_4 species over the growing season. However, the high spatial heterogeneity of this ecosystem, which was included in our error estimates, likely overwhelms phenological differences in functional group activity. Another source of bias may stem from our use of leaves rather than roots for the $\delta^{13}C_{new}$ end-member. In C_3 plants, roots have been found to be about 1‰ more enriched than leaves [*Cheng and Johnson*, 1998], but in C_4 plants, roots were either similar to leaves or slightly depleted [*Trouve et al.*, 1994; *Schweitzer et al.*, 1999]. In this experiment, it was not feasible to separate roots by species, so we cannot evaluate the magnitude of potential uncertainty, but this should be considered for future studies of SOM isotopic dynamics.

[41] Our partitioning approach demonstrated that elevated CO_2 did not enhance rhizosphere respiration rates on the shortgrass steppe. This is somewhat surprising, considering the significantly greater belowground NPP and root biomass found on EC plots (Milchunas et al., unpublished data). Therefore, specific respiration rates (per unit of root material) were apparently suppressed by elevated CO_2. Dark respiration by aboveground tissues often declines under elevated CO_2, possibly because biomass with higher C:N ratios requires less energy input [*Bunce*, 1994]. This mechanism should be relevant to root respiration as well. Suppression of specific root respiration by elevated CO_2 has

been found in white oak and in tussock sedge [*Norby*, 1996; *BassiriRad et al.*, 1996]. Higher specific respiration of maple roots under elevated CO_2 was found in one study, but those authors suggest that enhanced root growth probably overwhelms small differences in specific respiration rates in most studies [*Edwards and Norby*, 1999]. Our measurements of root biomass and BNPP were made only at the end of the growing season, and thus did not reflect seasonal dynamics of root turnover. More direct measurements are needed to confirm our findings of slight suppression of specific root respiration rates.

[42] At the shortgrass steppe, rhizosphere respiration rates increased with increasing soil water content. Soil moisture is clearly a key limiting factor to plant growth on the shortgrass steppe; our results demonstrate its importance in soil C cycling. Many ecosystem models use only soil temperature and root biomass to simulate root respiration rates, but our data suggest that in semi-arid grasslands, soil moisture effects should not be neglected. Rhizosphere respiration on EC plots was more strongly related to soil temperature than to soil water, suggesting that turnover of recent exudates by microbes may contribute a larger proportion of the new C flux on EC than AC and NC treatments.

4.3. Modeling of Decomposition and Implications for Soil C Storage

[43] We applied the latest heterotrophic respiration submodel used in DAYCENT, which uses a variable Q_{10} temperature function derived from dormant season data, and a moisture function derived from year-round data [*Parton et al.*, 1998; *DelGrosso et al.*, in preparation]. The model captured the temporal variability in decomposition rates well, although it underpredicted rates from EC and NC plots early in the growing season. Soil moisture content below 15-cm depth was not used for modeling, but was probably an important factor in microbial respiration as well as production of labile substrates, particularly early in the growing season of 1999. This may also explain the poor correlations we observed between decomposition and soil moisture.

[44] Comparison of observed with simulated decomposition rates was facilitated by normalizing the flux of ''old'' C from CO_2 treatments to Fallow plot values, to include turnover of recent substrates. On the Fallow plots, isotopic partitioning indicated that $\sim70\%$ of decomposition was derived from ''old'' C, and $\sim30\%$ came from decomposition of ''new'' C, including turnover of fine roots and decomposition of exudates and other rhizodeposits. Our estimate of 50% total heterotrophic respiration on NC plots for the growing season is in agreement with other estimates from the shortgrass steppe (Del Grosso et al., manuscript in preparation, 2003).

[45] The scaling factors used to account for differences in substrate availability for the three treatments (based on the ratio of averaged observations to averaged simulations) suggest a substrate-mediated stimulation of decomposition rates of $\sim70\%$ by elevated CO_2. This compares with an observed stimulation of $\sim250\%$, which encompasses both soil moisture and substrate effects (soil temperatures were

the same on EC and AC treatments). The observations and model results taken together imply that the higher decomposition rates observed under doubled CO_2 are driven by both increased substrates and higher soil water content. Earlier CENTURY modeling efforts predicted $\sim$30% stimulation of "abiotic" decomposition rates by doubled CO_2 in temperate steppe regions; this stimulation was driven by soil moisture effects related to reduced stomatal conductance [*Parton et al.*, 1995]. Our observed stimulation of EC relative to AC decomposition rates may be an overestimate because it includes possible chamber effects. Higher soil temperatures over the first 3 years of the experiment likely enhanced decomposition rates in AC relative to NC plots; mineralizable SOM on AC plots may have been depleted, leading to artificially suppressed decomposition rates. Nonetheless, our observations support the model result that elevated CO_2 should stimulate decomposition rates on the shortgrass steppe. We predict that plant growth enhancement by elevated CO_2 will probably be sustained by enhanced N mineralization rates, particularly in dry years when soil moisture conservation is significant. Soil C storage is therefore likely to increase on the shortgrass steppe, despite increased decomposition rates.

5. Conclusions

[46] We investigated belowground C cycling responses to elevated CO_2 on the shortgrass steppe in northeastern Colorado. Elevated CO_2 stimulated soil respiration rates by 25% during a moist summer and by 85% during a dry summer, although the stimulation was only significant for the driest period. Stable C isotopes allowed partitioning of soil respiration into decomposition and rhizosphere respiration components on all treatments. Decomposition rates were significantly enhanced by elevated CO_2 during a moist summer, when differences in soil moisture between treatments were small. Greater labile C combined with high C:N ratios of belowground biomass under elevated CO_2 apparently stimulated microbial decomposition of native SOM. This is evidence for enhanced substrate induced priming by elevated CO_2 in these grassland soils. Rhizosphere respiration rates were unaffected by elevated CO_2, despite greater belowground biomass and NPP. Model simulations of total soil respiration generally captured important differences between treatments that were mediated by soil moisture feedbacks to elevated CO_2. Simulations of decomposition rates required scaling factors to account for a probable enhancement of substrate availability under elevated CO_2.

References

Amundson, R., L. Stern, T. Baisden, and Y. Wang, The isotopic composition of soil and soil-respired CO_2, *Geoderma*, *82*, 83–114, 1998.

Balesdent, J., G. Wagner, and A. Mariotti, Soil organic-matter turnover in long-term field experiments as revealed by C-13 natural abundance, *Soil Sci. Soc. Am. J.*, *52*, 118–124, 1988.

BassiriRad, H., D. T. Tissue, J. F. Reynolds, and F. S. Chapin III, Response of *Eriophorum vaginatum* to CO_2 enrichment at different soil temperatures: Effects on growth, root respiration and PO_4^{3-} uptake kinetics, *New Phytol.*, *133*, 423–430, 1996.

Billes, G., H. Rouher, and P. Bottner, Modifications of the carbon and nitrogen allocations in the plant (*Triticum aestivum* L.) soil system in response to increased atmospheric CO_2 concentration, *Plant Soil*, *157*, 215–225, 1993.

Bunce, J. A., Responses of respiration to increasing atmospheric carbon dioxide, *Physiol. Plant.*, *90*, 427–430, 1994.

Canadell, J. G., L. F. Pitelka, and J. S. I. Ingram, The effects of elevated [CO_2] on plant-soil carbon below-ground: A summary and synthesis, *Plant Soil*, *187*, 391–400, 1996.

Cardon, Z. G., B. A. Hungate, C. A. Cambardella, F. S. Chapin III, C. B. Field, E. A. Holland, and H. A. Mooney, Contrasting effects of elevated CO_2 on old and new soil carbon pools, *Soil Biol. Biochem.*, *33*, 365–373, 2001.

Cerling, T. E., The stable isotopic composition of modern soil carbonate and its relationship to climate, *Earth Planet. Sci. Lett.*, *71*, 229–240, 1984.

Cheng, W., and D. W. Johnson, Elevated CO_2, rhizosphere processes, and soil organic matter decomposition, *Plant Soil*, *202*, 167–174, 1998.

Cheng, W., D. C. Coleman, C. R. Carroll, and C. A. Hoffman, Investigating short-term carbon flows in the rhizospheres of different plant species using isotopic trapping, *Agron. J.*, *86*, 782–791, 1994.

Conway, T. J., P. P. Tans, L. S. Waterman, K. W. Thoning, D. A. Kitzis, K. A. Masarie, and N. Zhang, Evidence of interannual variability of the carbon cycle from the NOAA/CMDL global air sampling network, *J. Geophys. Res.*, *99*, 22,831–22,855, 1994.

Davidson, E. A., L. V. Verchot, J. H. Cattanio, I. L. Ackerman, and J. E. M. Carvalho, Effects of soil water content on soil respiration in forests and cattle pastures of eastern Amazonia, *Biogeochemistry*, *48*, 53–69, 2000.

Diaz, S., J. P. Grime, J. Harris, and E. McPherson, Evidence of a feedback mechanism limiting plant response to elevated carbon dioxide, *Nature*, *364*, 616–617, 1993.

Edwards, N. T., and R. J. Norby, Below-ground respiratory responses of sugar maple and red maple saplings to atmospheric CO_2 enrichment and elevated air temperature, *Plant Soil*, *206*, 85–97, 1999.

Ferretti, D. F., E. Pendall, J. A. Morgan, J. A. Nelson, D. LeCain, and A. R. Mosier, Partitioning evapotranspiration fluxes from a Colorado grassland using stable, isotopes: Seasonal variations and ecosystem implications of elevated atmospheric CO2, *Plant Soil*, in press, 2003.

Fitter, A. H., J. D. Graves, J. Wolfenden, G. K. Self, T. K. Brown, D. Bogie, and T. A. Mansfield, Root production and turnover and carbon budgets of two contrasting grasslands under ambient and elevated atmospheric carbon dioxide concentrations, *New Phytol.*, *137*, 247–255, 1997.

Hanson, P. J., N. T. Edwards, C. T. Garten, and J. A. Andrews, Separating root and soil microbial contributions to soil respiration: A review of methods and observations, *Biogeochemistry*, *48*, 115–146, 2000.

Hogberg, P., A. Nordgren, N. Buchmann, A. Taylor, A. Ekblad, M. Hogberg, G. Nyberg, M. Ottosson-Lofvenius, and D. Read, Large-scale forest girdling shows that current photosynthesis drives soil respiration, *Nature*, *411*, 789–792, 2001.

Houghton, J. T., Y. Ding, D. J. Griggs, M. Noguer, P. J. van der Linden, X. Dai, K. Maskell, and C. A. Johnson (Eds.), The carbon cycle and atmospheric carbon dioxide, in *Climate Change 2001: The Scientific Basis*, pp. 183–237, Cambridge Univ. Press, New York, 2001.

Hu, S., M. Firestone, and F. S. Chapin III, Soil microbial feedbacks to atmospheric CO_2 enrichment, *Trends Ecol. Evol.*, *14*, 433–437, 1999.

Hu, S., F. S. Chapin III, M. Firestone, C. Field, and N. Chiariello, Nitrogen limitation of microbial decomposition in a grassland under elevated CO_2, *Nature*, *409*, 188–191, 2001.

Hungate, B., F. S. Chapin III, H. Shong, E. Holland, and C. Field, Stimulation of grassland nitrogen cycling under carbon dioxide enrichment, *Oecologia*, *109*, 149–153, 1997a.

Hungate, B. A., E. A. Holland, R. B. Jackson, F. S. Chapin III, H. A. Mooney, and C. B. Field, The fate of carbon in grasslands under carbon dioxide enrichment, *Nature*, *388*, 576–579, 1997b.

Jenkinson, D. S., The priming effect, in *The Use of Isotopes in Soil Organic Matter Studies*, pp. 199–208, Pergamon Press, New York, 1966.

Keeling, C. D., The concentration and isotopic abundance of atmospheric carbon dioxide in rural areas, *Geochim. Cosmochim. Acta*, *13*, 322–334, 1958.

Keeling, C. D., The concentration and isotopic abundance of carbon dioxide in rural and marine air, *Geochim. Cosmochim. Acta*, *24*, 277–298, 1961.

Lauenroth, W. K., and D. G. Milchunas, The shortgrass steppe, in *Natural Grasslands*, edited by R. T. Coupland, pp. 183–226, Elsevier Sci., New York, 1991.

Leavitt, S., E. Pendall, E. Paul, T. Brooks, B. Kimball, P. Pinter, H. Johnson, and G. Wall, Stable-carbon isotopes and soil organic carbon in the 1996 and 1997 FACE wheat experiments, *New Phytol.*, *150*, 305–314, 2001.

Lekkerkerk, L., A. Van de Geijn, and J. Van Veen, Effects of elevated atmospheric CO_2-levels on the C economy of a soil planted with wheat, in *Soils and the Greenhouse Effect*, edited by A. Bouwman, pp. 423–429, John Wiley, New York, 1990.

Lin, G., J. Ehleringer, P. Rygiewicz, M. Johnson, and D. Tingey, Elevated CO_2 and temperature impacts on different components of soil CO_2 efflux in Douglas-fir mesocosms, *Global Change Biol.*, *9*, 157–168, 1999.

Lin, G. H., P. T. Rygiewicz, J. R. Ehleringer, M. G. Johnson, and D. T. Tingey, Time-dependent responses of soil CO_2 efflux components to elevated atmospheric $[CO_2]$ and temperature in experimental forest mesocosms, *Plant Soil*, *229*, 259–270, 2001.

Loiseau, P., and J. F. Soussana, Elevated $[CO_2]$, temperature increase and N supply effects on the turnover of below-ground carbon in a temperate grassland ecosystem, *Plant Soil*, *210*, 233–247, 1999.

Milchunas, D. G., and W. K. Lauenroth, Belowground primary production by carbon isotope decay and long-term root biomass dynamics, *Ecosystems*, *4*, 139–150, 2001.

Milchunas, D., O. E. Sala, and W. K. Lauenroth, A generalized model of the effects of grazing by large herbivores on grassland community structure, *Am. Nat.*, *132*, 87–106, 1988.

Miller, J., D. Yakir, J. W. C. White, and P. Tans, Measurement of $^{18}O/^{16}O$ in the soil-atmosphere CO_2 flux, *Global Biogeochem. Cycles*, *13*, 761–774, 1999.

Morgan, J. A., H. W. Hunt, C. A. Monz, and D. R. LeCain, Consequences of growth at two carbon dioxide concentrations and two temperatures for leaf gas exchange in *Pascopyrum smithii* (C3) and *Bouteloua gracilis* (C4), *Plant Cell Environ.*, *17*, 1023–1033, 1994.

Morgan, J., D. LeCain, A. Mosier, and D. Milchunas, Elevated CO_2 enhances water relations and productivity and affects gas exchange in C_3 and C_4 grasses of the Colorado shortgrass steppe, *Global Change Biol.*, *7*, 451–466, 2001.

Morgan, J. A., A. R. Mosier, D. G. Milchunas, D. R. LeCain, J. A. Nelson, and W. J. Parton, CO_2 enhances productivity of the shortgrass steppe, alters species composition and reduces forage digestibility, *Ecol. Appl*, in press, 2003.

Nie, D., H. He, G. Mo, M. B. Kirkham, and E. T. Kanemasu, Canopy photosynthesis and evapotranspiration of rangeland plants under doubled carbon dioxide in closed-top chambers, *Agric. For. Meteorol.*, *61*, 205–217, 1992.

Niklaus, P. A., and C. Korner, Responses of soil microbiota of a late successional alpine grassland to long term CO_2 enrichment, *Plant Soil*, *184*, 219–229, 1996.

Norby, R. J., Oak in a high-CO_2 world, *Ann. Sci. For.*, *53*, 413–429, 1996.

Owensby, C., J. Ham, A. Knapp, D. Bremer, and L. Auen, Biomass production and species composition change in a tallgrass prairie ecosystem after long-term exposure to elevated CO_2, *Global Change Biol.*, *5*, 497–506, 1999.

Parnas, H., A theoretical explanation of the priming effect based on microbial growth with two limiting substrates, *Soil Biol. Biochem.*, *8*, 139–144, 1976.

Parton, W. J., J. M. O. Scurlock, D. S. Ojima, D. S. Schimel, D. O. Hall, and S. G. Members, Impact of climate change on grassland production and soil carbon worldwide, *Global Change Biol.*, *1*, 13–22, 1995.

Parton, W., M. Hartman, D. Ojima, and D. Schimel, DAYCENT and its land surface submodel: description and testing, *Global Planet. Change*, *19*, 35–48, 1998.

Paul, E. A., S. J. Morris, and S. Bohm, The determination of soil C pool sizes and turnover rates: Biophysical fractionation and tracers, in *Assessment Methods for Soil Carbon*, edited by R. Lal et al., pp. 193–206, Lewis, Boca Raton, Fla., 2001.

Pendall, E., J. King, A. Mosier, J. Morgan, and J. White, Changes in turnover rates and pool sizes of soil organic matter under elevated CO_2 on the shortgrass steppe, *Eos Trans. AGU*, *82*(47), Fall Meet. Suppl., B11A-08, 2001a.

Pendall, E., et al., Elevated CO_2 stimulates soil respiration in a FACE wheat field, *Basic Appl. Ecol.*, *2*, 193–201, 2001b.

Phillips, D. L., and J. W. Gregg, Uncertainty in source partitioning using stable isotopes, *Oecologia*, *127*, 171–179, 2001.

Potter, C. S., E. A. Davidson, and L. V. Verchot, Estimation of global biogeochemical controls and seasonality in soil methane consumption, *Chemosphere*, *32*, 2219–2245, 1996.

Pregitzer, K. S., D. R. Zak, J. Maziasz, J. DeForest, P. S. Curtis, and J. Lussenhop, Fine root growth, mortality, and morphology in a factorial elevated atmospheric CO_2 × soil N availability experiment, *Ecol. Appl.*, *10*, 18–33, 2000.

Rice, C., F. Garcia, C. Hampton, and C. Owensby, Soil microbial response in tallgrass prairie to elevated CO_2, *Plant Soil*, *165*, 67–74, 1994.

Rochette, P., and L. Flanagan, Quantifying rhizosphere respiration in a corn crop under field conditions, *Soil Sci. Soc. Am. J.*, *61*, 466–474, 1997.

Schweitzer, E., J. Fear, and G. Cadisch, Isotopic (C-13) fractionation during plant residue decomposition and its implications for soil organic matter studies, *Rapid Commun. Mass Spectrom.*, *13*, 1284–1290, 1999.

Scott, N. A., C. V. Cole, E. T. Elliott, and S. A. Huffman, Soil textural control on decomposition and soil organic matter dynamics, *Soil Sci. Soc. Am. J.*, *60*, 1102–1109, 1996.

Sokal, R. R. and F. J. Rohlf, *Biometry*, 3rd ed., 887 pp., Freeman, W.H., New York, 1995.

Thornley, J. H. M., and M. G. R. Cannell, Dynamics of mineral N availability in grassland ecosystems under increased $[CO_2]$: Hypotheses evaluated using the Hurley Pasture Model, *Plant Soil*, *224*, 153–170, 2000.

Trolier, M., J. W. C. White, P. P. Tans, K. A. Masarie, and P. A. Gemery, Monitoring the isotopic composition of atmospheric CO_2: Measurements from the NOAA global air sampling network, *J. Geophys. Res.*, *101*, 25,897–25,916, 1996.

Trouve, C., A. Mariotti, D. Schwartz, and B. Guillet, Soil organic carbon dynamics under Eucalyptus and Pinus planted on savannas in the Congo, *Soil Biol. Biochem.*, *26*, 287–295, 1994.

Volk, M., P. Niklaus, and C. Korner, Soil moisture effects determine CO_2 responses of grassland species, *Oecologia*, *125*, 380–388, 2000.

Williams, M., C. Rice, and C. Owensby, Carbon dynamics and microbial activity in tallgrass prairie exposed to elevated CO_2 for 8 years, *Plant Soil*, *227*, 127–137, 2000.

Zak, D. R., K. S. Pregitzer, P. S. Curtis, J. A. Teeri, R. Fogel, and D. L. Randlett, Elevated atmospheric CO_2 and feedbacks between carbon and nitrogen cycles in forested ecosystems, *Plant Soil*, *151*, 105–117, 1993.

Zak, D. R., K. Pregitzer, J. King, and W. Holmes, Elevated atmospheric CO_2, fine roots, and the response of soil microorganisms: a review and hypothesis, *New Phytol.*, *147*, 201–222, 2000.

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