

# Feedback from plant species change amplifies CO<sub>2</sub> enhancement of grassland productivity

H. WAYNE POLLEY\*, VIRGINIA L. JIN\*† and PHILIP A. FAY\*

\*Grassland, Soil & Water Research Laboratory, US Department of Agriculture, Agricultural Research Service, Temple, Texas 76502, USA, †Agroecosystem Management Research Unit, US Department of Agriculture, Agricultural Research Service, Lincoln, Nebraska 68583, USA

## Abstract

Dynamic global vegetation models simulate feedbacks of vegetation change on ecosystem processes, but direct, experimental evidence for feedbacks that result from atmospheric CO<sub>2</sub> enrichment is rare. We hypothesized that feedbacks from species change would amplify the initial CO<sub>2</sub> stimulation of aboveground net primary productivity (ANPP) of tallgrass prairie communities. Communities of perennial forb and C<sub>4</sub> grass species were grown for 5 years along a field CO<sub>2</sub> gradient (250–500 μL L<sup>-1</sup>) in central Texas USA on each of three soil types, including upland and lowland clay soils and a sandy soil. CO<sub>2</sub> enrichment increased community ANPP by 0–117% among years and soils and increased the contribution of the tallgrass species *Sorghastrum nutans* (Indian grass) to community ANPP on each of the three soil types. CO<sub>2</sub>-induced changes in ANPP and *Sorghastrum* abundance were linked. The slope of ANPP-CO<sub>2</sub> regressions increased between initial and final years on the two clay soils because of a positive feedback from the increase in *Sorghastrum* fraction. This feedback accounted for 30–60% of the CO<sub>2</sub>-mediated increase in ANPP on the upland and lowland clay soils during the final 3 years and 1 year of the experiment, respectively. By contrast, species change had little influence on the ANPP-CO<sub>2</sub> response on the sandy soil, possibly because *Sorghastrum* increased largely at the expense of a functionally similar C<sub>4</sub> grass species. By favoring a mesic C<sub>4</sub> tall grass, CO<sub>2</sub> enrichment approximately doubled the initial enhancement of community ANPP on two clay soils. The CO<sub>2</sub>-stimulation of grassland productivity may be significantly underestimated if feedbacks from plant community change are not considered.

**Keywords:** aboveground net primary productivity, C<sub>4</sub> grasses, plant species composition, soil texture, soil water content, subambient CO<sub>2</sub>, tallgrass prairie

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## Introduction

Experimental enrichment of atmospheric CO<sub>2</sub> usually increases the productivity of grassland and other herbaceous vegetation (Morgan *et al.*, 2004b). Rarely is the initial stimulation of growth maintained, however. Growth responses to CO<sub>2</sub> may diminish with time for a variety of reasons, ranging from down-regulation of photosynthetic capacity (Crous *et al.*, 2010) to a progressive decrease in N recycling that leads to greater N limitation of growth (Luo *et al.*, 2004; Reich *et al.*, 2006). Less frequently, community growth responses to CO<sub>2</sub> increase with time (Stöcklin *et al.*, 1998). The CO<sub>2</sub> sensitivity of productivity changes because plant responses to CO<sub>2</sub> are regulated by a set of biotic and abiotic variables that are sensitive to feedbacks from CO<sub>2</sub> effects on plant growth (Polley *et al.*, 2011b). These variables, which include soil resource supply

and the functional composition of plant communities, interact dynamically with ecosystem processes to determine the sensitivity of community productivity to CO<sub>2</sub>. Feedbacks from CO<sub>2</sub>- and climate-caused changes usually dampen CO<sub>2</sub> effects on community productivity (Leuzinger *et al.*, 2011; Polley *et al.*, 2011b). Positive feedbacks on CO<sub>2</sub> or climate responses are fewer, but may occur most frequently when mediated through changes in plant species composition or relative abundances (defined herein as 'species change'). For example, a shift to greater C<sub>4</sub> dominance enhanced the positive effects of warming on aboveground net primary productivity (ANPP) of tallgrass prairie by increasing growth per unit of plant N (Niu *et al.*, 2010).

There is ample evidence that CO<sub>2</sub> enrichment alters species contributions to grassland productivity (Leadley *et al.*, 1999; Edwards *et al.*, 2001; Niklaus *et al.*, 2001; Polley *et al.*, 2003; Morgan *et al.*, 2004a), but little documentation that species change enhances the community ANPP-CO<sub>2</sub> response. Why? We suggest five general

Correspondence: H. Wayne Polley, tel. + 254 770 6629, fax + 254 770 6561, e-mail: wayne.polley@ars.usda.gov

reasons. First, feedbacks from species change simply may not have been considered when analyzing experimental data. Second, experiments may have been terminated before species feedbacks were fully expressed. Species change and any resulting feedbacks on the community ANPP-CO<sub>2</sub> response may not be evident in short-term, even multi-year, experiments in which CO<sub>2</sub> effects on resource levels develop slowly. Changes in N cycling that lead ultimately to a change in the ANPP-CO<sub>2</sub> response may require years to manifest (Gill *et al.*, 2002, 2006; Hungate *et al.*, 2004; Jin & Evans, 2007), as would species feedbacks on community ANPP, for example. Third, responsive species may not have been present in plant communities either because the plant species pool was limited or species change was inhibited by management activities or other factors. Fourth, species feedbacks will be difficult to detect when the ANPP-CO<sub>2</sub> relationship differs widely among years because of variation in other factors that regulate plant growth or CO<sub>2</sub> response. Year-to-year variability in environmental factors, such as the amount of photosynthetically active radiation and precipitation (Owensby *et al.*, 1999; Morgan *et al.*, 2004b; Kubiske *et al.*, 2006), could mask species effects on the ANPP-CO<sub>2</sub> relationship. Fifth, species change at elevated CO<sub>2</sub> simply may not have become large enough to significantly affect the ANPP-CO<sub>2</sub> relationship. How large must the shift in species be? The answer depends partly on the magnitude of functional differences among co-occurring species and on whether species shifts result from opposing changes in the productivity of dominant species or simply an increase in productivity of one or more species. The greater the differences in response to CO<sub>2</sub> or CO<sub>2</sub>-caused changes in water and N availability among species, the greater, likely, will be the change in ANPP-CO<sub>2</sub> response per unit of change in species relative abundances.

A positive feedback of species change on the ANPP-CO<sub>2</sub> response implies that the increase in community productivity per unit of increase in CO<sub>2</sub> will be greater following species change than before it. We have shown that increasing CO<sub>2</sub> from subambient to elevated concentrations increased the contribution of the tallgrass species *Sorghastrum nutans* (L.) Nash (Indian grass) to ANPP of tallgrass prairie assemblages grown on three soil types of strongly contrasting texture (Polley *et al.*, 2012). The shift to greater *Sorghastrum* abundance during the first 4 years of CO<sub>2</sub> treatment resulted from a CO<sub>2</sub>-caused increase in soil water content and was most strongly expressed on the two soils on which the per-unit effect of CO<sub>2</sub> on soil water was greatest. Herein, we test the prediction that the increase in *Sorghastrum* fraction amplified the amount by which CO<sub>2</sub> stimu-

lated ANPP during 5 years of treatment. We hypothesized that feedback effects of *Sorghastrum* on ANPP, like the magnitude of species change, would be greatest for the soils for which the water-savings effect of CO<sub>2</sub> enrichment was greatest. We assumed that *Sorghastrum* is more productive than species that declined in relative abundance under the wetter conditions at high CO<sub>2</sub>.

## Materials and Methods

### CO<sub>2</sub> chambers

We used unique, elongated field chambers to expose assemblages of tallgrass prairie species to a continuous gradient in CO<sub>2</sub> spanning pre-industrial to elevated concentrations (Polley *et al.*, 2008; Fay *et al.*, 2009). The Lysimeter CO<sub>2</sub> gradient (LYCOG) facility is located in central Texas, USA (31°05' N, 97°20' W) at the Grassland, Soil & Water Research Laboratory. It consists of two transparent, tunnel-shaped chambers each 1.2 m wide and tall and 60 m long, aligned parallel along a north-south axis. Each chamber is divided into 10 consecutive compartments each 5 m long. Aerial growth of chambered vegetation is enclosed in a transparent polyethylene film which transmits >90% of incident irradiance with minimal effects on spectral quality. Chambers depend upon photosynthesis during daylight and upon respiration at night to create CO<sub>2</sub> gradients. Air introduced into chambers during daylight is progressively depleted of CO<sub>2</sub> by photosynthesizing plants as it is moved by blowers toward the outlet of the system. The direction of air flow is reversed at night, and respiration by the enclosed ecosystem progressively increases the CO<sub>2</sub> concentration of chamber air. Desired CO<sub>2</sub> concentration gradients are maintained by automatically varying the rate of air flow through chambers in response to changes to photosynthesis (daylight) or respiration rates (night). During daylight, pure CO<sub>2</sub> is injected into air introduced to the south end of one chamber (elevated chamber) to elevate CO<sub>2</sub> to 500  $\mu\text{L L}^{-1}$ . The CO<sub>2</sub> level is depleted to 370  $\mu\text{L L}^{-1}$  at the north end of the chamber. Ambient air is introduced into the south end of the second chamber (subambient chamber) to initiate an ambient to subambient CO<sub>2</sub> gradient (370 – 250  $\mu\text{L L}^{-1}$ ). Night-time CO<sub>2</sub> levels are regulated at about 130  $\mu\text{L L}^{-1}$  above daytime values along each chamber. Air temperature and vapor pressure deficit are regulated near ambient values by cooling and dehumidifying air at 5-m intervals along chambers. CO<sub>2</sub> treatments have been maintained each growing season from early April through October since CO<sub>2</sub> control was initiated in 2006.

### Soil monoliths

We installed four steel boxes containing intact soil monoliths (each 1 m wide and long  $\times$  1.5 m deep) beneath each of the 20 5-m long compartments. Three soil types of contrasting physical and hydrological properties are represented. All are located in the Blackland Prairie region of central Texas. Soils

include an upland clay Mollisol from the Austin series (Udorthentic Haplustolls), a lowland clay Vertisol from the Houston Black series (Udic Haplusterts), and a sandy loam Alfisol from the Bastil (or Bastrop) series (Udic Paleustalfs). Two monoliths of each of two soil types were randomly placed beneath each 5-m long compartment. Twelve compartments contain two monoliths each of the Mollisol and Vertisol (40–45% and 49–55% clay content, respectively; Fay *et al.*, 2009). Remaining compartments contain two monoliths of the Alfisol (60–73% sand content, 0–50 cm depth) and two monoliths of either the Mollisol or Vertisol.

Perennial species characteristic of tallgrass prairie in central Texas were transplanted into 60 monoliths in June 2003, 3 years before CO<sub>2</sub> control was initiated. Eight plants of each of four C<sub>4</sub> grass species [*Bouteloua curtipendula* (Michx.) Torr. (side-oats grama), *Schizachyrium scoparium* (Michx.) Nash (little bluestem), *Sorghastrum nutans*, *Tridens albescens* (Vasey) Wootton & Standl. (white tridens)] and three forb species [*Salvia azurea* Michx. Ex Lam. (pitcher sage), *Solidago canadensis* L. (Canada goldenrod), and the legume *Desmanthus illinoensis* (Michx.) MacMill. ex B.L. Rob. & Fernald (Illinois bundleflower)] were transplanted into each monolith (total density = 56 plants m<sup>-2</sup>; hereafter referenced by genus). Monoliths received ambient precipitation for most of the period prior to CO<sub>2</sub> treatment (Polley *et al.*, 2012). We excluded precipitation after initiating CO<sub>2</sub> control in 2006 and irrigated each monolith to simulate the average of growing season precipitation in central Texas (560 mm). Water input was scheduled to mimic the typical seasonal pattern of precipitation during 2007 and 2009, with a maximum during spring and minimum during late summer (August–September). This pattern was reversed in 2008 by adding larger amounts of water in summer than spring. The seasonal total of irrigation was applied in equal monthly increments during 2006. Volumetric soil water content was measured weekly (bi-weekly in 2006) to 0.9 m depth in the center of each monolith with a neutron probe.

ANPP was estimated by clipping vegetation in each monolith to 5 cm height at the end of each growing season, near the time of peak biomass, beginning in 2005 the year prior to CO<sub>2</sub> treatment. Peak biomass provides a reasonable estimate of ANPP in this system because aboveground tissues represent current-year growth only (standing tissues are removed at harvest each year) and all species, with the exception of two minor components of the community, *Desmanthus* and *Tridens* (1.9–8.4% and 0.2–3.3% of peak biomass, respectively), continue growth through autumn. At harvest, plants from each monolith were sorted by species. Harvested tissues were weighed after oven drying for 72 h at 60 °C. Community ANPP was calculated by summing species-specific values of ANPP for each monolith and year. No fertilizer was added. To minimize effects of harvesting on element cycling, harvested material was returned to the monolith from which it was removed prior to the next growing season. Plant material was shredded with a wood chipper to simulate effects of late-season mowing before it was returned.

We used between-year change in the proportional contribution of the dominant species, the grass *Sorghastrum*, to community

ANPP ( $\Delta$  fraction) as an index of vegetation change. Change was calculated for each monolith by subtracting the dominant's contribution to total community production in 1 year from its contribution to production in a preceding year.

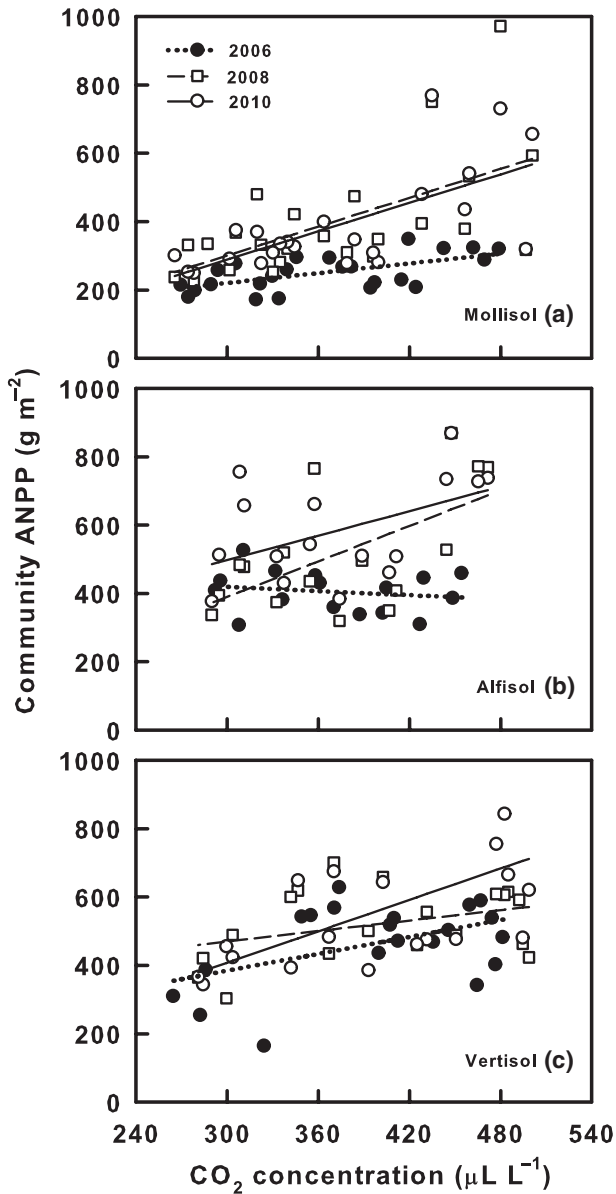
The mean CO<sub>2</sub> concentration to which each monolith was exposed was calculated using the linear relationship between CO<sub>2</sub> during daylight and distance along chambers (Fay *et al.*, 2009). CO<sub>2</sub> was used as the independent variable in regression analyses with between-year changes in community ANPP, species-specific ANPP, and species fractions.

The CO<sub>2</sub> response of community ANPP is regulated by multiple, often inter-correlated, factors (Polley *et al.*, 2011b). We used a statistical modeling approach, structural equation modeling (SEM), to evaluate causal relationships among interacting variables that together affected ANPP (Shipley, 2000; Grace, 2006). SEM with observed variables (path modeling) was used to partition the net effect of CO<sub>2</sub> on ANPP into a direct effect and indirect effects mediated through changes in soil water content and the *Sorghastrum* fraction of community ANPP for each soil type (Supporting Information, Fig. S1). The SEM model included direct paths from each variable to ANPP and indirect paths from CO<sub>2</sub> to ANPP through soil water content and the *Sorghastrum* fraction. Bivariate relationships between modeled variables were linear to slightly curvilinear. The ln-transformation of variables rarely improved the linearity of these relationships and did not alter the general relationships among variables that were derived using SEM. We used AMOS 4.0 software (SmallWaters Corp., Chicago, IL, USA) to fit the SEM model to data from each year and soil type. Each path coefficient was divided by its standard error to assess significance. Coefficients with  $P < 0.10$  were considered significant. To obtain a significant fit to data for the different soil types, we used a slightly different SEM model for the sandy soil vs. clay soils (Fig. S1). Non-significant paths in the models used for each soil type were retained in final models. Significance in most statistical tests of treatment effects is demonstrated by rejecting a null hypothesis. The reverse is true when testing the adequacy of the full SEM (Shipley, 2000). The hypothesized relationship among variables in the SEM is considered to be consistent with data when the probability level of the statistical test (chi-squared statistic) is greater than the chosen significance level (typically,  $P = 0.05$ ).

## Results

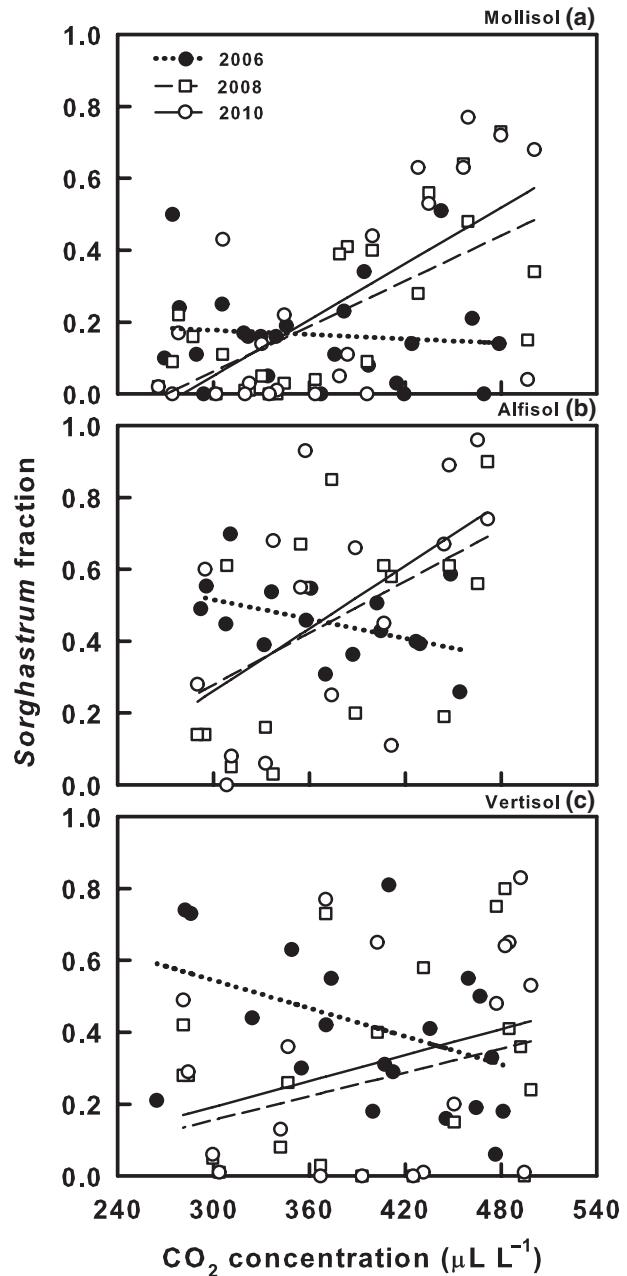
### *Temporal trends in ANPP and Sorghastrum relative abundance*

CO<sub>2</sub> enrichment increased both community ANPP (Fig. 1) and the contribution of *Sorghastrum* to community productivity on all three soil types (Fig. 2). Linear increases in community ANPP ranged from 0 (not significant) to 173 g m<sup>-2</sup> per 100  $\mu$ L L<sup>-1</sup> rise in CO<sub>2</sub> among soils and years, and were larger following 3–5 years of CO<sub>2</sub> exposure than during the initial 2 years of treatment for all soil types (Table 1). At these rates of ANPP increase, community ANPP rose by 45–117%



**Fig. 1** Relationships between the ANPP of prairie communities (community ANPP) and CO<sub>2</sub> concentration for each of three soil types for the first, third, and fifth years of treatment (2006, 2008, 2010). Lines are linear regression fits. See Table 1 for regression statistics.

(Mollisol), 0–98% (Alfisol), and 0–81% (Vertisol) from 280 to 480  $\mu\text{L L}^{-1}$ . The *Sorghastrum* fraction varied widely among monoliths along the CO<sub>2</sub> gradient in a given year (Fig. 2), but slopes of fraction-CO<sub>2</sub> regression relationships were greater following 2–3 years of exposure than during the initial year of treatment for Mollisol and Alfisol soils (Table 1). The *Sorghastrum* fraction was a significant positive function of CO<sub>2</sub> for the Vertisol in 2009 only, but was negatively correlated to CO<sub>2</sub> following the first year of treatment (2006). Con-



**Fig. 2** Relationships between the fractional contribution of *Sorghastrum* to ANPP of prairie communities and CO<sub>2</sub> concentration for each of three soil types for the first, third, and fifth years of treatment (2006, 2008, 2010). Lines are linear regression fits. See Table 1 for regression statistics.

sequently, CO<sub>2</sub> enrichment increased the contribution of *Sorghastrum* to community ANPP between the pre-treatment year and fifth year of CO<sub>2</sub> exposure on all soils (Fig. S2).

CO<sub>2</sub> enrichment increased the *Sorghastrum* fraction of community productivity by increasing *Sorghastrum* ANPP and, on the Alfisol soil, reducing the amount by

**Table 1** Linear regressions relating community ANPP and the *Sorghastrum* fraction of ANPP to CO<sub>2</sub> concentration for each of three soil types and each year of CO<sub>2</sub> treatment. Regression slopes represent the change in ANPP or *Sorghastrum* fraction per 100  $\mu\text{L L}^{-1}$  increase in CO<sub>2</sub> concentration

Soil/Year	ANPP ( $\text{g m}^{-2}$ )				<i>Sorghastrum</i> fraction					
	Intercept	Slope	$r^2$	<i>P</i> -value	Intercept	Slope	$r^2$	<i>P</i> -value	<i>n</i>	
<b>Mollisol</b>										
2006	77.95	47.52	0.34	0.002	0.237	-0.021	0	0.65	24	
2007	3.70	72.68	0.52	<0.0001	-0.219	0.089	0.28	0.006	24	
2008	-122.31	140.93	0.33	0.002	-0.568	0.212	0.46	0.0002	24	
2009	-172.87	158.51	0.51	<0.0001	-0.803	0.268	0.56	<0.0001	23	
2010	-126.78	138.49	0.43	0.0004	-0.730	0.261	0.42	0.0005	23	
<b>Alfisol</b>										
2006	478.64	-19.98	0	0.51	0.785	-0.088	0.12	0.1	16	
2007	-80.94	126.76	0.53	0.0008	-0.656	0.282	0.32	0.01	16	
2008	-130.47	173.33	0.31	0.01	-0.442	0.241	0.22	0.04	16	
2009	-26.95	170.38	0.47	0.003	-0.383	0.239	0.22	0.04	16	
2010	139.04	119.41	0.19	0.05	-0.609	0.294	0.26	0.03	16	
<b>Vertisol</b>										
2006	137.32	82.31	0.18	0.04	0.934	-0.135	0.14	0.06	20	
2007	116.82	66.10	0.16	0.04	0.036	0.048	0	0.44	20	
2008	316.00	51.15	0.08	0.12	-0.175	0.114	0.05	0.17	19	
2009	370.71	42.70	0.04	0.2	-0.413	0.178	0.22	0.03	19	
2010	-50.91	152.96	0.35	0.005	-0.168	0.122	0.05	0.19	19	

which the combined ANPP of remaining species increased between the pretreatment (2005) year and fifth year of exposure (Fig. 3). CO<sub>2</sub> enrichment reduced the 5-year change in the combined ANPP of all species other than *Sorghastrum* on the Alfisol largely by reducing the increase in ANPP of two subdominant grasses, *Bouteloua* and *Schizachyrium* (Table S2). CO<sub>2</sub> enrichment also reduced the amount by which *Bouteloua* ANPP increased during 5 years on the Mollisol, but the combined ANPP of all species other than the dominant *Sorghastrum* was not affected by CO<sub>2</sub> on this upland clay soil (Fig. 3). Together, *Sorghastrum* and *Bouteloua* accounted for an average of 0.60 and 0.73 of community productivity on the clay Mollisol and Vertisol soils, respectively, following 5 years of CO<sub>2</sub> treatment. By contrast, *Sorghastrum* and *Schizachyrium* together accounted for a mean 0.89 of ANPP on the sandy Alfisol after 5 years.

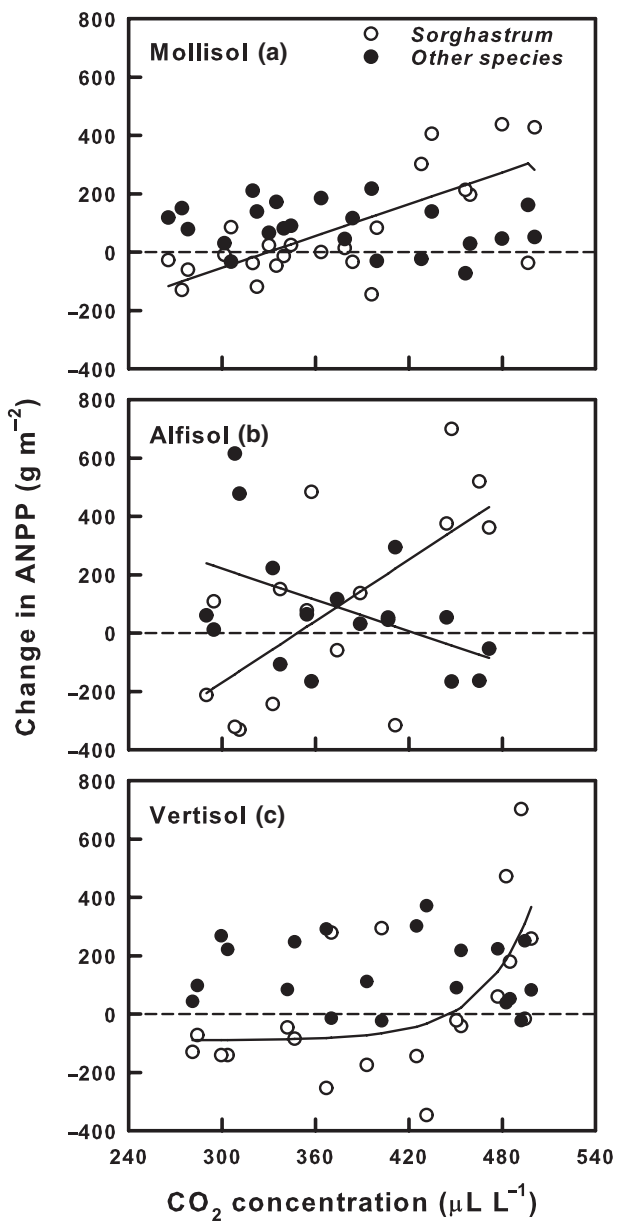
The shared tendency for ANPP and the *Sorghastrum* fraction to increase more with time at elevated than subambient CO<sub>2</sub> resulted in a positive correlation between changes in ANPP and the *Sorghastrum* fraction for each soil type (Fig. 4). These correlations between cumulative changes in community properties imply a direct causal linkage between the *Sorghastrum* fraction and ANPP sensitivity to CO<sub>2</sub>. However, both ANPP and the *Sorghastrum* fraction varied with CO<sub>2</sub> (Table 1), evidence that ANPP was regulated directly and

indirectly by CO<sub>2</sub>, the indirect effect mediated partly through among-year change in the *Sorghastrum* fraction (i.e.,  $\Delta \text{fraction} = \text{Sorghastrum fraction}_{(t)} - \text{Sorghastrum fraction}_{(t-1...n)}$ ). In addition, community ANPP during any given year likely was sensitive to the current *Sorghastrum* fraction, as determined partly by the initial composition of vegetation, rather than to CO<sub>2</sub>-mediated change in the fraction only. We used structural equation modeling to determine the contribution of CO<sub>2</sub>-caused shifts in both the *Sorghastrum* fraction and soil water content to the response of community ANPP to CO<sub>2</sub>.

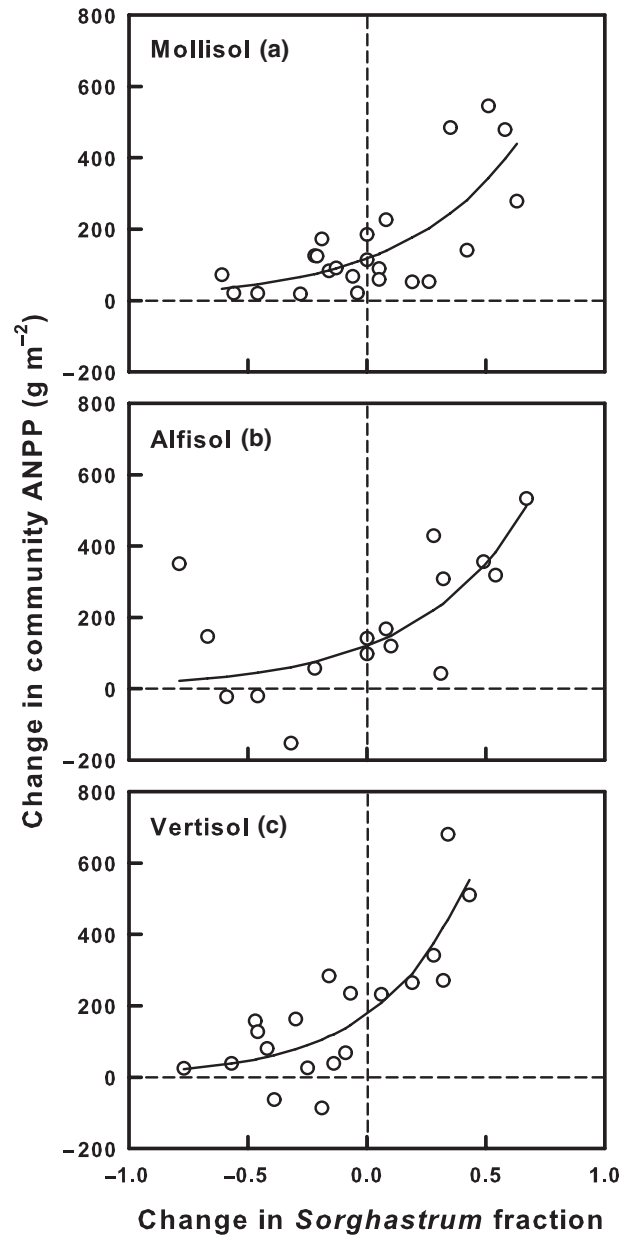
#### Structural equation models

SEM models adequately fit data from each year of CO<sub>2</sub> treatment and soil type ( $\chi^2_3 = 0.2\text{--}6.3$ ,  $P = 0.97\text{--}0.10$ ) and explained 36–54%, 3–56%, and 9–38% of the variance in ANPP for the Mollisol, Alfisol, and Vertisol soils, respectively.

**Mollisol.** CO<sub>2</sub> enrichment increased community ANPP on the Mollisol soil during each year of treatment, with linear increases of 48–159  $\text{g m}^{-2}$  per 100  $\mu\text{L L}^{-1}$  increase in CO<sub>2</sub> (Fig. 5). The CO<sub>2</sub> effect on community ANPP was 2-fold greater during the final 3 years than first 2 years of treatment because CO<sub>2</sub> increased ANPP by increasing the *Sorghastrum* fraction. The pathway

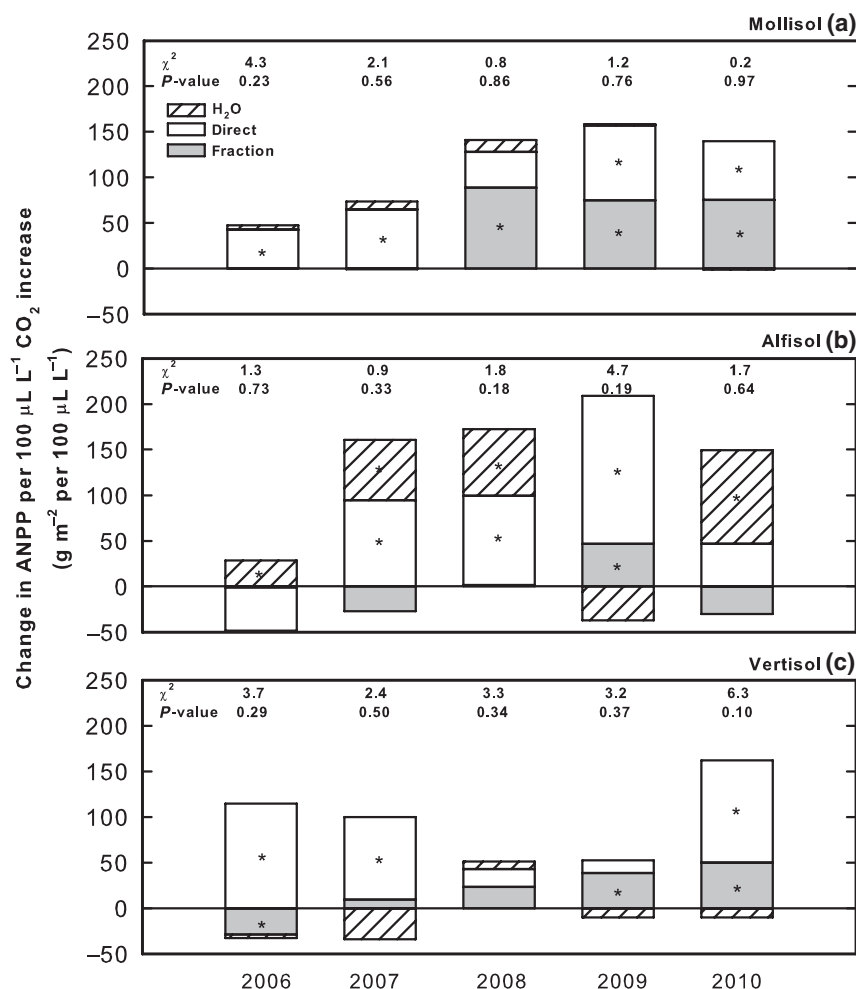


**Fig. 3** Relationships between the 5-year change in ANPP of the dominant grass species *Sorghastrum* and of all remaining (other) species in prairie communities combined and CO<sub>2</sub> concentration for each of three soil types. Change in ANPP was calculated by subtracting the ANPP for the year prior to CO<sub>2</sub> treatment (2005) from ANPP following year 5 of treatment (2010). Lines are significant linear or exponential regression fits to changes in species ANPP for Mollisol (*Sorghastrum*,  $r^2 = 0.45$ ,  $P = 0.0003$ ,  $n = 23$ ), Alfisol (*Sorghastrum*,  $r^2 = 0.39$ ,  $P = 0.006$ ; other species,  $r^2 = 0.16$ ,  $P = 0.07$ ,  $n = 16$ ), and Vertisol soils, respectively (*Sorghastrum*,  $r^2 = 0.29$ ,  $P = 0.02$ ,  $n = 20$ ;  $r^2 = 0.17$ ,  $P = 0.09$  following exclusion of a single point for which the change in ANPP > 600 g m<sup>-2</sup>). The regression model with greatest  $r^2$  value was selected. The change in combined ANPP of species other than *Sorghastrum* was not related to CO<sub>2</sub> for the Mollisol ( $P = 0.29$ ) or Vertisol soils ( $P = 0.72$ ), consequently no lines are shown.



**Fig. 4** Relationships between the 5-year change in ANPP of prairie communities and change in the fractional contribution of *Sorghastrum* to community ANPP for each of three soil types. Change in ANPP or *Sorghastrum* fraction was calculated by subtracting values measured the year prior to CO<sub>2</sub> treatment (2005) from values measured after 5 years of treatment (2010). Lines are exponential regression fits to changes in community ANPP for Mollisol ( $r^2 = 0.53$ ,  $P < 0.0001$ ,  $n = 23$ ), Alfisol ( $r^2 = 0.45$ ,  $P = 0.003$ ,  $n = 16$ ), and Vertisol soils, respectively ( $r^2 = 0.66$ ,  $P < 0.0001$ ,  $n = 19$ ).

linking CO<sub>2</sub> to community ANPP through the *Sorghastrum* fraction (Fig. 6) accounted for 47–63% of the CO<sub>2</sub>-caused increase in ANPP during 2008–2010 (Fig. 5). Among years, the total effect of CO<sub>2</sub> on community



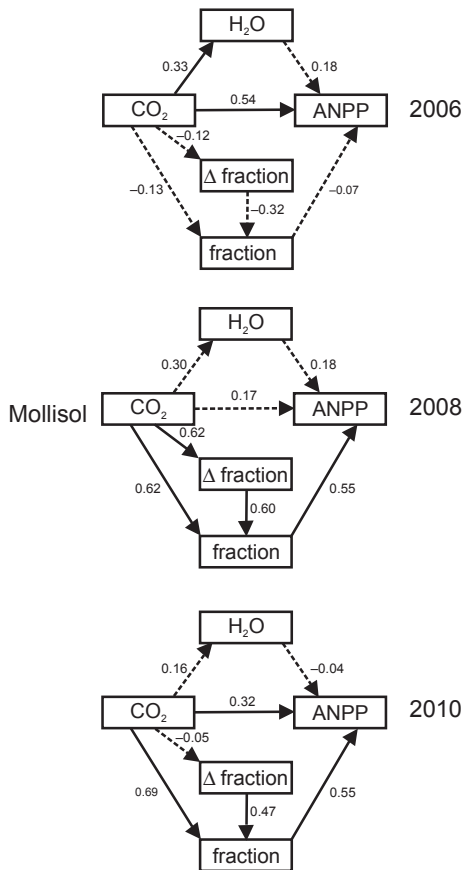
**Fig. 5** Direct and indirect components of the CO<sub>2</sub> effect on ANPP of prairie communities that were grown on each of three soil types for 5 years. CO<sub>2</sub> effects were partitioned using SEM (Table S1, Fig. S1). Indirect effects of CO<sub>2</sub> on ANPP were mediated through changes in soil water content (H<sub>2</sub>O) and the fractional contribution of the dominant C<sub>4</sub> grass species *Sorghastrum* to community productivity (fraction). Values for each effect were calculated using unstandardized path coefficients and are expressed as the change in ANPP (g m<sup>-2</sup>) per 100 μL L<sup>-1</sup> increase in CO<sub>2</sub> concentration. Significant effects are denoted with an asterisk. An indirect effect was considered significant if coefficients for one or more pathways linking CO<sub>2</sub> to ANPP were significant. The overall effect of CO<sub>2</sub> on ANPP as mediated by species change, for example, represents the sum of the effects derived from paths linking CO<sub>2</sub> to ANPP through the *Sorghastrum* fraction. Chi-square (χ<sup>2</sup>) and P-values for the full SEM model are listed. The full model is considered to be consistent with data when the P-value of the chi-squared statistic is >0.05. The df = 3 for all soils except the Alfisol in 2007–2008 for which a simplified version of the SEM was required (df = 1).

ANPP was positively correlated with the magnitude of the indirect effect of CO<sub>2</sub> through the increase in *Sorghastrum* fraction ( $r^2 = 0.87$ ;  $P = 0.01$ ). Conversely, paths linking CO<sub>2</sub> to ANPP through changes in soil water content were not significant.

**Alfisol.** CO<sub>2</sub> enrichment increased community ANPP on the Alfisol in all but the first year of treatment (Fig. 5). The total CO<sub>2</sub> effect included large direct effects in 3 years, depended relatively little on the *Sorghastrum* fraction, and was mediated in 3 of 4 years (2007, 2008, 2010) through an increase in soil water

content. The *Sorghastrum* fraction was negatively correlated with CO<sub>2</sub> following 2006, the first year of treatment (Fig. 7), apparently because of variability in vegetation that developed during the establishment period. In years 4 and 5 (2009–2010), the coefficient for the pathway linking CO<sub>2</sub> and the *Sorghastrum* fraction was positive, indicating that CO<sub>2</sub> enrichment increased *Sorghastrum*'s contribution to community productivity. Paths linking CO<sub>2</sub> to community ANPP through the *Sorghastrum* fraction were significant only for 2009, however. ANPP of prairie communities increased by 46.8 g m<sup>-2</sup> per 100 μL L<sup>-1</sup> increase in CO<sub>2</sub> as a result

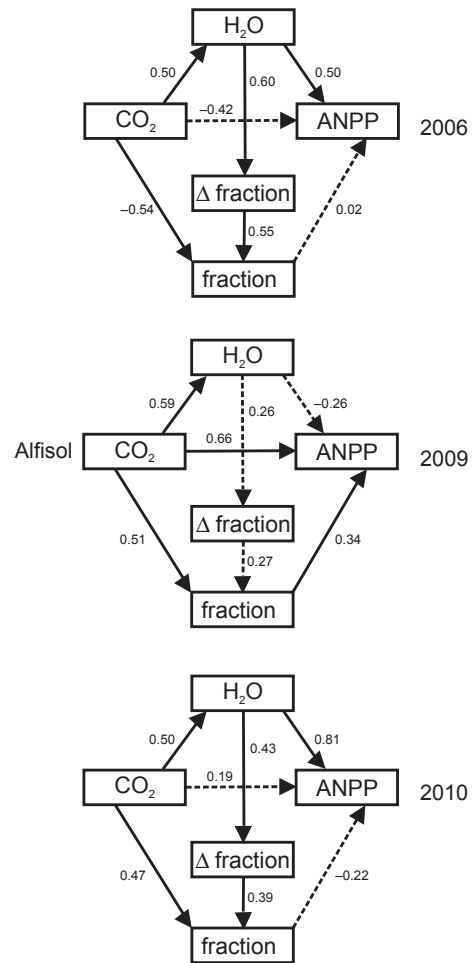




**Fig. 6** Structural equation models describing direct and indirect effects of CO<sub>2</sub> on ANPP of prairie communities grown on a Mollisol for the first, third, and fifth years of treatment (2006, 2008, 2010; models for remaining years were omitted for brevity). Maximum likelihood techniques were used to calculate path coefficients that minimize the difference between the calculated covariance matrix and the covariance matrix present in the data. Standardized coefficients are calculated by first standardizing each variable by subtracting its mean and dividing by its standard deviation. Non-significant pathways are indicated by dashed lines. Standardized coefficients are listed beside each path. Modeled are indirect effects of CO<sub>2</sub> through soil water content, change in the *Sorghastrum* fraction of community ANPP during a given year ( $\Delta$  fraction), and the *Sorghastrum* fraction of ANPP (fraction).

(Fig. 5). This indirect effect of CO<sub>2</sub> on community ANPP was only 28% of the magnitude of the direct CO<sub>2</sub> effect.

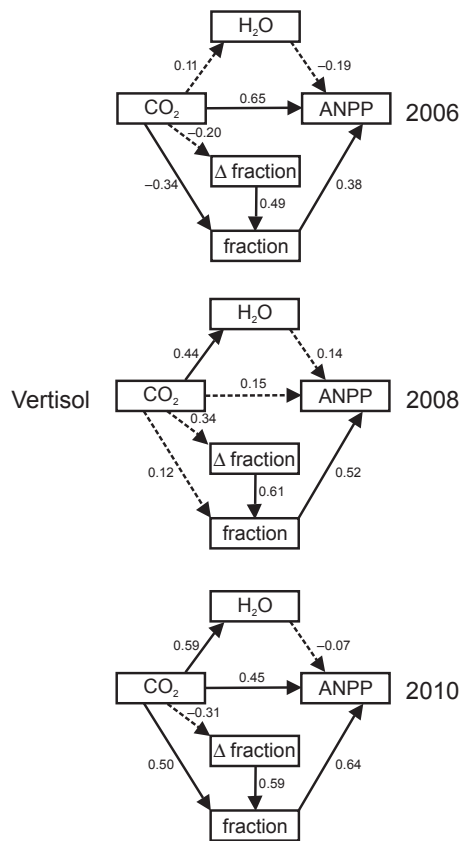
*Vertisol*. The bivariate community ANPP-CO<sub>2</sub> relationship was positive each year for the Vertisol but was significant in 2006, 2007, and 2010 only (Table 1). The positive effect of CO<sub>2</sub> on ANPP of prairie communities was entirely direct during the first 2 years, but resulted from or included a significant indirect effect mediated through an increase in the *Sorghastrum* fraction in 2010



**Fig. 7** Structural equation models describing direct and indirect effects of CO<sub>2</sub> on ANPP of prairie communities grown on an Alfisol for the first, fourth, and fifth years of treatment (2006, 2009, 2010; models for remaining years were omitted for brevity). Non-significant pathways are indicated by dashed lines. Standardized coefficients are listed beside each path. Modeled are indirect effects of CO<sub>2</sub> through soil water content, change in the *Sorghastrum* fraction of community ANPP during a given year ( $\Delta$  fraction), and the *Sorghastrum* fraction of ANPP (fraction).

(as well as in 2009; Fig. 5). Community ANPP was positively correlated with the *Sorghastrum* fraction each year (Fig. 8), in contrast to trends observed for Mollisol and Alfisol soils, with linear increases in ANPP of 199–413 g m<sup>-2</sup> per unit increase in the *Sorghastrum* fraction (not shown). However, the coefficient for the pathway linking CO<sub>2</sub> and the *Sorghastrum* fraction was negative after year one of treatment (2006), meaning that *Sorghastrum* contributed less to community ANPP at elevated than subambient CO<sub>2</sub>. This pattern was reversed in later years. The temporal trend for *Sorghastrum* to increase at elevated CO<sub>2</sub>, combined with the strong





**Fig. 8** Structural equation models describing direct and indirect effects of CO<sub>2</sub> on ANPP of prairie communities grown on a Vertisol for the first, third, and fifth years of treatment (2006, 2008, 2010; models for remaining years were omitted for brevity). Non-significant pathways are indicated by dashed lines. Standardized coefficients are listed beside each path. Modeled are indirect effects of CO<sub>2</sub> through soil water content, change in the *Sorghastrum* fraction of community ANPP during a given year ( $\Delta$  fraction), and the *Sorghastrum* fraction of ANPP (fraction).

positive link between community ANPP and the *Sorghastrum* fraction in all years, caused the net CO<sub>2</sub> effect on ANPP through the *Sorghastrum* fraction to increase from 1 year to the next (Fig. 5). Paths linking CO<sub>2</sub> to community ANPP through changes in soil water content were not significant for the Vertisol, as for the other clay soil.

## Discussion

Increasing CO<sub>2</sub> from subambient to elevated concentrations increased ANPP of perennial prairie communities on each of the three soil types we studied (see also, Fay *et al.*, in press). Linear increases ranged from 0–173 g m<sup>-2</sup> per 100  $\mu$ L L<sup>-1</sup> increase in CO<sub>2</sub> among soils and years, and were larger following 3–5 years of CO<sub>2</sub> exposure than during the initial 2 years of treatment

for all soil types. CO<sub>2</sub> also increased the relative contribution of the tallgrass *Sorghastrum* to community productivity on all three soils by increasing *Sorghastrum* ANPP and, on the alluvial sandy soil (Alfisol), reducing the amount by which the combined ANPP of remaining species increased during the 5-year experiment. Importantly, these two general trends, the increase in community ANPP and increase in the *Sorghastrum* fraction of productivity at elevated CO<sub>2</sub>, were positively correlated as predicted on the three soils. The response of community ANPP to CO<sub>2</sub> increased with time on the two clay soils because of a positive feedback from the CO<sub>2</sub>-caused increase in the *Sorghastrum* fraction. This feedback accounted for 47–63% of the CO<sub>2</sub>-mediated increase in community ANPP on the upland clay soil (Mollisol) during the final 3 years and 33% of the ANPP increase on the lowland clay soil (Vertisol) during the final year. CO<sub>2</sub> also favored *Sorghastrum* on the Alfisol, but species change had little effect on the ANPP-CO<sub>2</sub> response of communities. Rather, CO<sub>2</sub> enrichment increased community ANPP on this sandy soil both directly and through an increase in soil water content that likely resulted from reduced transpiration.

We hypothesized that feedback effects of species change on community ANPP would be greatest for the soils for which the water-savings effect of CO<sub>2</sub> was greatest under the assumption that water savings would favor species that are more productive under wetter conditions. Our hypothesis was only partially supported. Per unit of rise in CO<sub>2</sub>, the increase in soil water content was greater by more than a factor of 1.8 and 2.4 on the Alfisol than Mollisol and Vertisol soils, respectively (Polley *et al.*, 2012). Yet, the positive feedback of increased *Sorghastrum* abundance on community ANPP was least on the sandy Alfisol.

Species shifts that involve opposing changes in the productivity of dominant species likely will enhance the sensitivity of community ANPP to CO<sub>2</sub> only when the increase in productivity of the favored species exceeds any decrease in productivity of disadvantaged species. Species change had little effect on the ANPP-CO<sub>2</sub> response of the Alfisol because CO<sub>2</sub> enrichment increased *Sorghastrum* at the expense of other species, including subdominant C<sub>4</sub> grasses. Element or other limitations on community ANPP may lead to offsetting changes in productivity among species.

CO<sub>2</sub> enrichment may change species relative abundances by increasing the production of one or more species without affecting biomass of other dominant and co-dominant species. This ‘favored-species’ route to species change is the most frequently reported ‘mechanism’ by which CO<sub>2</sub> affects grassland communities (Leadley *et al.*, 1999; Morgan *et al.*, 2004a). CO<sub>2</sub> increased the contribution of *Stipa comata* to

aboveground production of shortgrass steppe but did not affect growth of other dominant grasses (Morgan *et al.*, 2004a), for example. CO<sub>2</sub> increased *Sorghastrum* productivity on the two clay soils (Mollisol and Vertisol), but did not significantly affect the combined ANPP of species other than *Sorghastrum*. The result was a direct correlation between the increase in relative abundance of the tallgrass and increase in community ANPP by year 5 of the experiment.

Our results highlight the obvious, but important role that the presence of productive and CO<sub>2</sub>-responsive species play in determining the sensitivity of community ANPP to CO<sub>2</sub>, as well as the importance of multi-year CO<sub>2</sub> experiments. Community ANPP was a significant and positive function of the *Sorghastrum* fraction on the Vertisol even early in the experiment when the *Sorghastrum* fraction was negatively correlated to CO<sub>2</sub> (2006). Community response to CO<sub>2</sub> apparently was depressed because less-responsive species usurped growth-limiting resources from *Sorghastrum*. Wang (2007) similarly invoked resource usurpation by weakly responsive species to explain the smaller CO<sub>2</sub> effect on growth of communities than species monocultures. By preferentially increasing *Sorghastrum* productivity, CO<sub>2</sub> contributed to a positive feedback on the ANPP-CO<sub>2</sub> response of prairie communities on the Vertisol. Results for this clay soil also demonstrate that initial responses to CO<sub>2</sub> enrichment may be misleading. Simple bivariate relationships between community ANPP and CO<sub>2</sub> were weaker and generally exhibited smaller slopes for the Vertisol than other soils (see also Fay *et al.*, in press). Interpreted from a physiological perspective, prairie communities on this lowland clay were the least responsive to CO<sub>2</sub>. Only with time did it become evident that the ANPP-CO<sub>2</sub> response was regulated largely by a biotic feedback.

Clearly, factors in addition to CO<sub>2</sub> can influence the ANPP-CO<sub>2</sub> relationship by modifying vegetation and its response to CO<sub>2</sub>. Disturbances, such as grazing and fire, or N fertilization may alter CO<sub>2</sub> effects on species by facilitating seed dispersal and seedling recruitment (grazing; Newton *et al.*, 2006) or changing competitive interactions among plants (grazing, fire, N fertilization; e.g., Henry *et al.*, 2006). For example, N fertilization initially enhanced the CO<sub>2</sub> stimulation of productivity in herbaceous brackish wetland, but promoted a shift from C<sub>3</sub> to C<sub>4</sub> species that suppressed CO<sub>2</sub> effects on community productivity by the third and fourth years of treatment (Langley & Megonigal, 2010). Because of the vagaries of seed dispersal and disturbance effects, species relative abundances may vary more in space at a given time than over time in response to CO<sub>2</sub>, even in a given community type on a given soil. Species effects on the ANPP

thus may also be reflected in spatial variation in the ANPP-CO<sub>2</sub> response.

Increasing CO<sub>2</sub> from pre-Industrial to elevated levels contributed to a positive feedback on the ANPP-CO<sub>2</sub> response of prairie communities by favoring a widely distributed tallgrass species. The feedback accounted for 30–60% of the CO<sub>2</sub>-mediated increase in community ANPP on two of three soil types during the final 1–3 years of treatment. Feedback effects were smallest for the soil type on which the increase in *Sorghastrum* was largely compensated for by a decline in the productivity and relative abundances of two sub-dominant C<sub>4</sub> grasses. We anticipate that species change will enhance the community ANPP-CO<sub>2</sub> response provided that shifting species exhibit different functional responses to CO<sub>2</sub> and the shift in species abundances is sufficiently large relative to the magnitude of functional differences in species. The smaller are species differences in response to CO<sub>2</sub> or accompanying changes in water and N availability, the greater, likely, must be the change in species abundances to influence the ANPP-CO<sub>2</sub> relationship. Simulations from dynamic global vegetation models indicate that CO<sub>2</sub>-caused changes in vegetation will be greatest in grasslands and savannas (Cramer *et al.*, 2001). Our results highlight the importance of accounting for species change and composition when predicting CO<sub>2</sub> effects on community productivity and other aspects of the functioning of these ecosystems (e.g., Polley *et al.*, 2011a).

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### Supporting Information

Additional Supporting Information may be found in the online version of this article:

**Table S1.** Description of variables included in structural equation models.

**Table S2.** Linear regression relationships between the 5-year change in ANPP of sub-dominant perennial grasses and CO<sub>2</sub> concentration for each of three soil types.

**Figure S1.** The theoretical structural equation model used to explore direct and indirect effects of CO<sub>2</sub> enrichment on ANPP of prairie communities.

**Figure S2.** Relationships between the 5-year change in the fractional contribution of *Sorghastrum* to ANPP of prairie communities and CO<sub>2</sub> treatment for each of three soil types.

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