

# Interactive responses of old-field plant growth and composition to warming and precipitation

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## Abstract

As Earth's atmosphere accumulates carbon dioxide (CO<sub>2</sub>) and other greenhouse gases, Earth's climate is expected to warm and precipitation patterns will likely change. The manner in which terrestrial ecosystems respond to climatic changes will in turn affect the rate of climate change. Here we describe responses of an old-field herbaceous community to a factorial combination of four levels of warming (up to 4 °C) and three precipitation regimes (drought, ambient and rain addition) over 2 years. Warming suppressed total production, shoot production, and species richness, but only in the drought treatment. Root production did not respond to warming, but drought stimulated the growth of deeper (> 10 cm) roots by 121% in 1 year. Warming and precipitation treatments both affected functional group composition, with C<sub>4</sub> grasses and other annual and biennial species entering the C<sub>3</sub> perennial-dominated community in ambient rainfall and rain addition treatments as well as in warmed treatments. Our results suggest that, in this mesic system, expected changes in temperature or large changes in precipitation alone can alter functional composition, but they have little effect on total herbaceous plant growth. However, drought limits the capacity of the entire system to withstand warming. The relative insensitivity of our study system to climate suggests that the herbaceous component of old-field communities will not dramatically increase production in response to warming or precipitation change, and so it is unlikely to provide either substantial increases in forage production or a meaningful negative feedback to climate change later this century.

**Keywords:** biodiversity, Boston-Area Climate Experiment, C<sub>4</sub> grasses, climate change, drought, old-field community, precipitation, primary production, root production, warming experiment

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## Introduction

Part of the uncertainty in climate change projections and their consequences stems from our still limited knowledge of climate change impacts on the terrestrial carbon cycle and its feedbacks to global climate change (Meir *et al.*, 2006; Arneth *et al.*, 2010). Variables of particular interest to climate change modellers include carbon uptake by, and loss from, terrestrial ecosystems, which will impact how fast climate will change (Arneth *et al.*, 2010; Bond-Lamberty & Thomson, 2010), and changes in plant functional types, which may impact regional climate by altering albedo, physical structure of the land surface and the rate of evapotranspiration (Meir *et al.*, 2006). Currently, the uncertainty due to carbon cycle parameters is roughly 40% that of more widely debated physical climate properties (i.e., equilibrium climate sensitivity and global heat capacity) in

Earth system and global climate models (Huntingford *et al.*, 2009). Research clarifying carbon cycle responses to climate change is vital to improving projections of how fast climate will change. To address these questions, greenhouse and mesocosm experiments are useful to determine rate changes in key biological processes in response to single and multiple effects of climate change (e.g., Fang *et al.*, 2005; Knorr *et al.*, 2005; Zavalloni *et al.*, 2008). Ultimately, though, field experiments are needed to determine whole-ecosystem responses under conditions when primary production is linked with concurrent changes in decomposition and nutrient cycling rates, and potentially also with varying species pools (e.g., Jentsch *et al.*, 2011).

Evidence that environmental changes such as warming, changes in precipitation regimes, and rising CO<sub>2</sub> concentrations are affecting ecosystem processes is accruing rapidly (e.g., Rustad, 2008). Experiments in a range of vegetation types have shown that warming strongly affects many ecosystem and community processes. In three meta-analyses of ecosystem

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responses to experimental warming across > 30 field sites, warming of ~1–6 °C boosted plant productivity, although responses ranged from strongly positive to negative across the sites (Rustad *et al.*, 2001; Lin *et al.*, 2010; Wu *et al.*, 2011). Wu *et al.* (2011) and Rustad *et al.* (2001) also found that warming increased soil respiration, and the latter study documented increases in nitrogen mineralization. In a multi-site warming experiment focused on arctic tundra, Arft *et al.* (1999) found that herbaceous plant species responded more vigorously to warming than woody species. A variety of experiments have also examined ecosystem effects of changes in precipitation amount (e.g., Dukes *et al.*, 2005; Johnson *et al.*, 2008; Sowerby *et al.*, 2008) frequency (e.g., Swemmer *et al.*, 2007; Fay *et al.*, 2008; Miranda *et al.*, 2009), and seasonality (e.g., Bates *et al.*, 2006; Chou *et al.*, 2008; Chimner *et al.*, 2010). These studies provide abundant evidence that changes in precipitation regime strongly influence community and ecosystem properties and can be expected to influence responses to warming (Weltzin *et al.*, 2003; Knapp *et al.*, 2008; Wu *et al.*, 2011).

Increasingly, researchers have recognized the advantages of studies examining ecosystem responses to multiple, interacting elements of global change (Norby & Luo, 2004; Heimann & Reichstein, 2008; Luo *et al.*, 2008; Rustad, 2008). The nature and strength of the interactive effects of warming and precipitation are of particular relevance, as climate warming has the potential to decrease soil moisture (e.g., Bell *et al.*, 2010), which could limit positive effects of warming on ecosystem processes in systems that experience water stress. In a modelling study in which four ecosystem models were parameterized for seven ecosystems in different climate zones, Luo *et al.* (2008) found that warming in combination with doubled precipitation consistently increased NPP, whereas warming in combination with halved precipitation interactively reduced NPP. So far, meta-analyses indicate that the interactive effects of warming and precipitation on NPP observed in field studies to date are small (Wu *et al.*, 2011) or not significant (Lin *et al.*, 2010). However, few field studies included in these meta-analyses manipulated both precipitation and temperature. More nuanced studies investigating the nature and magnitude of such interactions, and how they are affected by short- and long-term shifts in composition and diversity, can improve our understanding of the net effects of climate change on productivity. To date, about fifteen studies have factorially manipulated temperature and precipitation in field experiments (e.g., Shaw *et al.*, 2002; Dermody *et al.*, 2007; Mikkelsen *et al.*, 2008; Niu *et al.*, 2008; Sherry *et al.*, 2008; Charles & Dukes, 2009; Volder *et al.*, 2010), although several of these have only recently begun.

Many ecosystem processes exhibit non-linear responses to temperature or soil moisture gradients. At small scales, many biological processes exhibit exponential (e.g., decomposition, soil respiration) or quadratic (e.g., photosynthesis) responses to temperature and soil moisture changes. Even over regional and continental scales, aboveground net primary production (ANPP) may show logistic responses to mean annual temperature (Schuur, 2003; Luo *et al.*, 2004) and either linear (Sala *et al.*, 1988; Zhou *et al.*, 2002) or unimodal (Schuur, 2003) responses to precipitation. It is not clear that community and ecosystem responses to intermediate or more extreme climate changes can be reliably inferred from the many experiments with two levels of an environmental variable. For example, in a multi-level precipitation experiment with planted assemblages of tallgrass prairie species, Fay *et al.* (2008) observed diminishing returns of increased primary productivity and soil respiration (20–56%) to rain additions (100–250%, 400–1000 mm) as overall water limitations were relieved. This example and others (e.g., McHale *et al.*, 1998) highlight the importance of examining ecosystem responses over multiple levels of climate change factors.

The Boston-Area Climate Experiment (BACE) was designed to test multi-factor, multi-level effects of precipitation and temperature on ecosystem processes in a mesic old field. Old fields, whether transient or maintained through human activities such as grazing and mowing, are a common grassland type in the temperate and tropical regions of the world (Cramer *et al.*, 2008). Grasslands, including steppes and savannah, cover more than 25% of Earth's vegetated area (Ramankutty & Foley, 1999), providing ecosystem services such as forage production and carbon sequestration. Grasslands have high inter-annual variation in productivity (Fang *et al.*, 2001; Knapp & Smith, 2001), suggesting that they are particularly sensitive to climate change. The BACE tests the nature of this sensitivity via two primary hypotheses: ecosystem process responses to changes in precipitation and temperature (1) are not linear, and (2) interact.

In this article, we examined responses of herbaceous biomass production and species composition in the first two full years of treatment application. We hypothesized that drought would suppress plant growth and reduce species richness. Given that warming stimulates evapotranspiration and reduces soil water availability (e.g., Harte & Shaw, 1995; Rustad *et al.*, 2001; Niu *et al.*, 2008), we hypothesized that warming effects would depend on precipitation treatments. Specifically, (1) drought treatments would have increasingly negative responses to warming; (2) treatments receiving ambient precipitation would have a unimodal response, such

that intermediate levels of warming would stimulate plant growth, but plants in the warmest treatments would incur moisture stress and thus exhibit reduced plant growth again; and (3) plant growth would increase along the entire warming gradient in treatments receiving supplemental precipitation. We further hypothesized that wet treatments would not differ from ambient treatments at any but the warmest temperatures, because the old-field community at the BACE is a fairly mesic community in which additional rain would likely run off. We hypothesized that warming would increase plant growth by lengthening the growing season in years with relatively cool, wet springs, but that warming would suppress growth in years with relatively hot, dry summers, as even small temperature increases would push the system farther from its soil moisture and photosynthetic optima. Tests of these hypotheses can provide new insight on the character of grassland responses to climate change, and the potential for grassland carbon uptake and sequestration under a variety of future climate scenarios.

## Materials and methods

### Site description

The BACE was constructed in an old-field community at the Suburban Experiment Station of the University of Massachusetts in Waltham, MA, USA (42° 23' 3" N, 71° 12' 52" W). The site's mean annual temperature is 9.3 °C (mean  $T_{\text{Dec,Jan,Feb}} = -2.6$  °C, range = -5.1–0.2 °C; mean  $T_{\text{Jun,Jul,Aug}} = 20.7$  °C, range = 18.7–21.1 °C), the mean growing season length is 186 days, and the mean annual precipitation is 1194 mm, with roughly equal amounts of precipitation falling in each month (NOAA National Climatic Data Center Cooperative Station ID 190535, years 1960–2008). The experimental area has loam topsoil (0–30 cm) over gravelly sandy loam subsoil. At the onset of the experiment (in 2008), the plant community was a mixture of native and introduced  $C_3$  and  $C_4$  forbs and grasses, which was largely dominated by  $C_3$  species (mean contribution to aboveground biomass production in plots = 99%, range = 85–100%) and perennials (mean = 80%, range = 29–100%), with grasses more abundant than forbs (mean = 73% grass, range = 15–100%). Prior to the onset of the experiment, the old-field community was maintained through occasional mowing. Dominant old-field species at BACE include the perennial grasses *Dactylis glomerata*, *Elymus repens*, *Phleum pratense* and *Poa* spp., the annual  $C_4$  grasses *Setaria glauca* and *S. viridis*, the intermittently flowering perennial forbs *Plantago lanceolata* and *Taraxacum officinale*, and the summer-flowering perennial forbs *Achillea millefolium*, *Potentilla argentea*, *Silene latifolia* and *Tanacetum vulgare*. While the old fields at BACE originally contained a fairly uniform mix of up to 42 species, we seeded 13 forb species commonly found at our field site (Supplementary Data Table S1) into all field plots in early spring 2008 to further increase uniformity.

### Experimental design

The experiment was designed as a randomized, complete block, split-plot design with factorial combinations of three precipitation treatments and four temperature treatments in each of three blocks (Fig. 1). The three precipitation treatments consisted of 'wet', ambient, and 'drought' conditions. Rainout shelters over the drought treatments deflected 50% of incoming precipitation throughout the year. During the growing season (early May to mid-November), this water was routed to sprinklers that immediately reapplied it onto the wet plots, increasing precipitation by 50%. The sprinkler systems for the wet treatment were shut off from mid-November to early May to avoid damage from frozen pipes. In an average year, BACE treatments approximately mimic precipitation in the driest (860 mm) and the wettest (1585 mm) years of the period on record (1960–2008). The ambient and wet sections of each greenhouse shelter were covered by deer fencing to reduce photosynthetically active radiation by ~5%, which approximates the reduction caused by the clear corrugated polycarbonate roof slats covering the drought sections. Within each precipitation section, we designated four 2 m by 2 m plots, which were subjected to one of four warming treatments: ambient temperature, low (target of +1.0 °C), medium (+2.7 °C), and high warming (+4.0 °C). Warming treatments were applied year-round with ceramic infrared heaters (200, 600, and 1000 W in the low, medium and high warming treatments, respectively), which were mounted 1 m above the ground at each corner of the 4 m<sup>2</sup> plots to provide an even spatial heating pattern across each plot. 'Dummy' heaters simulated shading from infrastructure in the ambient warming treatments. Infrared radiometers measured canopy temperatures in the center of the ambient and warmest plots along each group of four plots; these readings were used to provide active feedback control to regulate heater output to maintain the target 4 °C temperature difference. Heaters achieved the target temperatures much of the time, but during the day and during rainstorms temperatures tended to dip below targets. This tendency led to a decrease in the diurnal temperature range, similar to what is projected for the north-eastern United States (Meehl *et al.*, 2007).

The construction of the rainout shelters covering the drought treatments was completed in the spring of 2007, the extra watering of the wet sections started in June 2008, and the temperature treatments were initiated on July 1, 2008. Before the onset of the experiment, 0.6 m-deep trenches were dug around each plot and lined with two layers of polyethylene sheeting to prevent the lateral movement of water and nutrients between individual plots and their surroundings. We monitored volumetric water content (VWC) in the top 0–30 cm of soil in each plot using permanently installed time-domain reflectometry waveguides and a Campbell TDR-100 (Campbell Scientific, Logan, UT, USA). VWC measurements were taken weekly during the growing season and bi-weekly during the rest of the year. Seasonal soil moisture deficits developed at the onset of the growing season in May and were recharged to water holding capacity over winter (Fig. 2). As intended, the soil moisture content was highest in wet precipitation treatments, intermediate in ambient precipitation

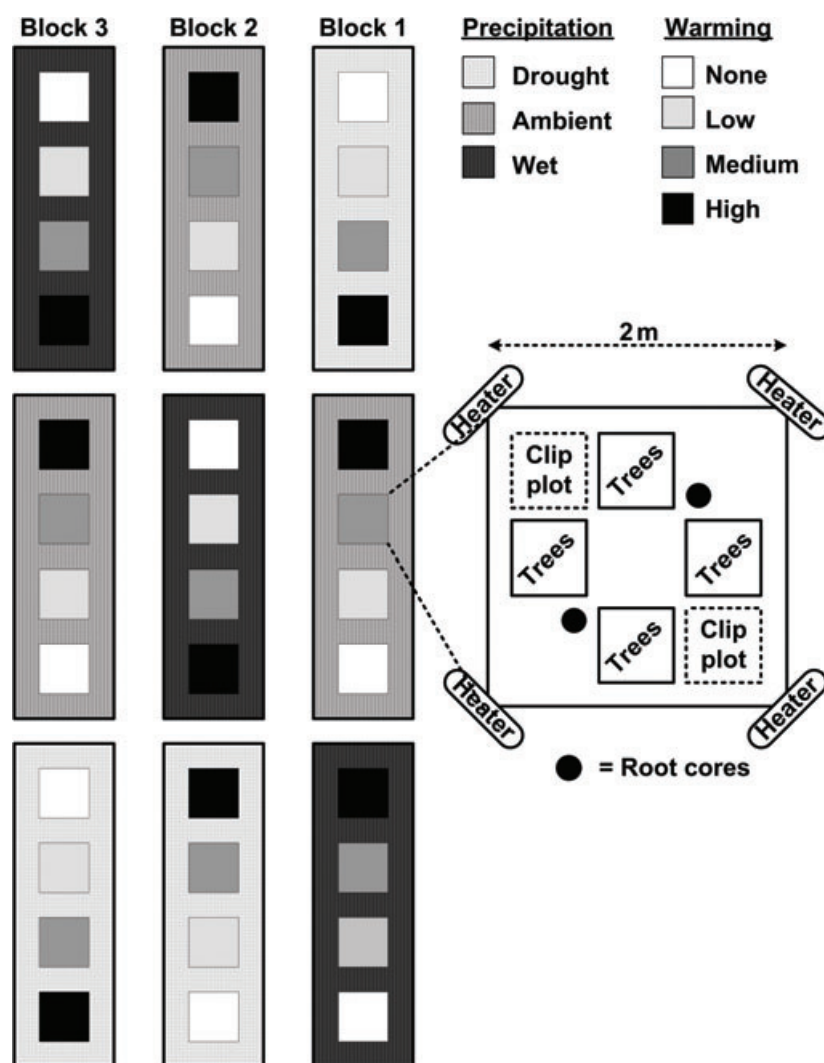


Fig. 1 The experimental layout of the Boston-area climate experiment (not drawn to scale). Individual plots are 2 m by 2 m.

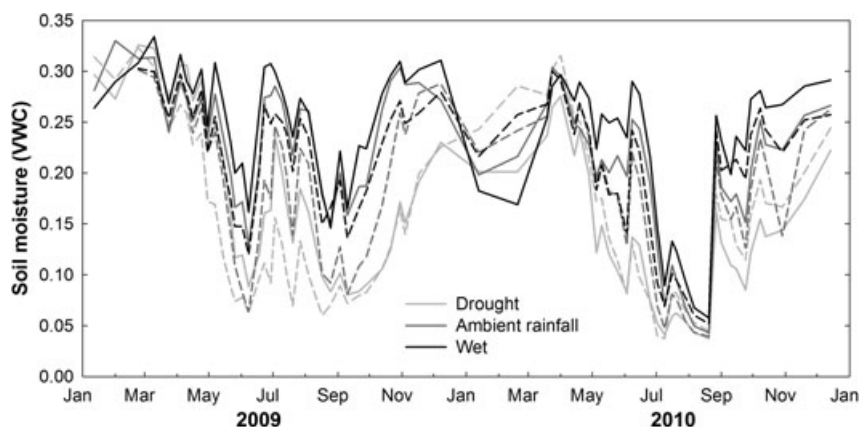
treatments and lowest in drought treatments, and warming treatments generally decreased soil moisture contents further (Fig. 2). The weather conditions at BACE were unusually cold and wet in the first full year of the experiment (2009) and unusually hot and dry in the second full year of the experiment (2010; Fig. 3).

Within each plot, we designated four evenly spaced 0.5 m by 0.5 m sub-plots for a tree seedling growth experiment. We planted one seedling (< 20 cm) of each of four tree species (*Acer rubrum*, *Betula lenta*, *Pinus strobus*, *Quercus rubra*) into each of these sub-plots in May–June 2008 and added a second seedling of each species in May 2009 for a maximum number of 32 tree seedlings per plot. To keep wildlife out of our study site, we fenced in the entire site with deer netting and reinforced it to a height of 1 m with chicken wire dug in to a depth of 30 cm. We continuously set traps to keep the local meadow vole (*Microtus pennsylvanicus*) population in check and to remove occasional cottontail rabbits (*Sylvilagus* spp.) from our site. We assessed animal disturbance in each plot at the end of

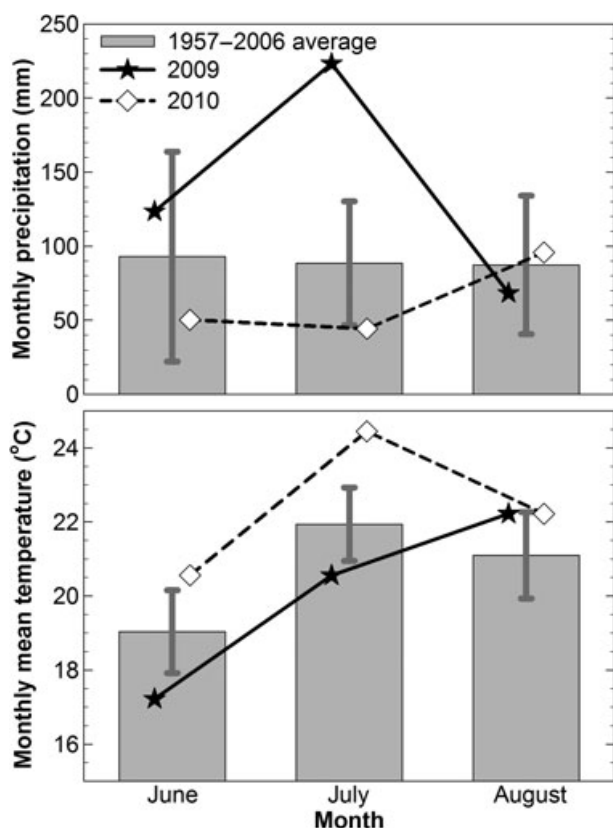
winter (~March 15th) by inspecting each plot and assigning it a score on an ordinal scale ranging from 0 (no visible disturbance) to 5 (> 75% of the plot surface disturbed).

#### Data collection

Herbaceous biomass samples were collected from two 0.25 m by 0.5 m sub-plots in each plot on two dates in 2008 and 2009 and from two 0.5 m by 0.5 m sub-plots on two dates in 2010. The first biomass sample each year was cut at a height of 10 cm in June, the second sample was cut at a height of 1–2 cm at the end of the growing season in mid-October. Biomass samples included live shoots as well as dead shoots and litter. In 2009, only plant species large enough to be sampled in each clip-plot were recorded in the field, while in 2010 all species that could be identified were recorded, even if their seedlings were too small to be clipped. Following each biomass harvest, the outer portion of each plot was cropped down to 10 cm (leaving a central 0.75 m by 0.75 m area intact)



**Fig. 2** Mean bi-weekly (summer) or monthly (winter) soil water content in the BACE drought treatments (light grey), ambient precipitation treatments (dark grey), and wet precipitation treatments (black) in either unheated (solid lines) or highest warming treatments (dashed lines) during 2009 and 2010. Soil moisture levels for plots at intermediate levels or warming are not shown, but generally fall between the unheated and warmest treatments. Soil moisture readings were taken bi-weekly during the growing season and monthly over winter. Note that summer soil moisture deficits usually started to develop in May, but that all soils generally recharged to the water holding capacity of approximately 30% volumetric water content over winter. Lower soil moisture readings between January and March indicate that some of the soil water was frozen and not detected by TDR.



**Fig. 3** Monthly total precipitation (top) and average temperature (bottom; based on daily averages) for the summer months at the BACE. Grey bars indicate the regional mean  $\pm$  SD from 1957 to 2006. The summer of 2009 was unusually wet and cool, while the summer of 2010 was unusually dry and hot.

to simulate hay cutting, which is a typical old-field management technique in New England. All harvests were conducted in the outer portions of the plots. The herbaceous biomass samples collected were sorted to species, dried to constant mass at 65 °C and weighed. Total biomass and the biomass components from different functional groups (i.e., grasses vs. forbs, perennials vs. annuals and biennials, and  $C_3$  vs.  $C_4$  plants) were determined for each sub-plot, averaged for each plot in each season, and then summed for an annual plot total. We used the biomass samples collected twice annually as an approximation of aboveground annual net primary production (ANPP). These biomass estimates likely underestimate actual ANPP by not accounting for losses to herbivores and volatile emissions, and by underestimating losses to mortality. Species richness ( $S$ ) and Shannon's evenness ( $E_H = H' / \ln S$ ), were calculated on a per-plot basis by combining the species information from both sub-plots and seasons in each year. Biomass and functional group biomass components from 2008 are available for qualitative comparisons of treatment effects to starting conditions (Supplementary Table S2), but these data were not included in the analyses, because 2008 was only a partial treatment year.

Root samples were collected annually from two root ingrowth cores per plot. The root ingrowth cores consisted of 7 cm wide, 40–60 cm deep cylindrical soil cores contained in 2.5 mm  $\times$  4 mm UV-protected polyethylene netting. The soil used in the root ingrowth cores was sieved with a 2 mm sieve to exclude roots and soil invertebrates and homogenized, so that all root ingrowth cores had the same soil moisture content when they were installed. The root ingrowth cores were first installed in early August 2008 and sampled every August thereafter. During sample collection, the top 10 cm section of each root ingrowth core was separated from the bottom 10–30 cm section, and both sample sub-sections were

processed separately. In 2010 only, we also sampled roots from the core sections remaining below 30 cm depth, which varied in length from 11 to 20 cm. We adjusted the root biomass from the core sections below 30 cm depth by dividing their root weights by the proportion of the actual core section length ( $\leq 20$  cm) to the maximum core section length (20 cm). The adjustment for root cores below 30 cm likely over-estimated the root weights of shorter cores at this depth, because root density declined quickly with depth at our site from 59.6% of the total root biomass in the top 10 cm, to 32.8% at depths of 10–30 cm, and 8.6% at depths below 30 cm (in ambient precipitation, unheated plots in 2010). All living roots were hand-picked from each sample, washed, dried to constant mass at 65 °C, and weighed. We used the root biomass estimated from the root ingrowth-cores collected once annually as a rough approximation of belowground annual net primary production (BNPP), recognizing that we did not account for root turnover or losses via root secretions.

### Statistical analyses

To analyse our data, we used mixed effects models with a randomized block, split-plot, repeated measures ANOVA design and restricted maximum likelihood (REML) estimation for all biomass and species diversity response variables. In our mixed models, precipitation treatments were whole-plot treatments and warming treatments were split-plot treatments within each precipitation section. Due to the different sub-plot sizes in 2009 and 2010, species richness and species evenness were analysed separately for both years in randomized block split-plot designs. We treated the 2 years as a fixed factor in biomass response models and specified it as a second split-plot treatment within plots. We used animal disturbance as a covariate in all models. Model assumptions were tested, and where appropriate, transformations were used to better meet model assumptions. Unless otherwise noted, data presented in the text and in graphs represent least square mean estimates from ANOVA models with standard errors. *P*-values used in the text are from ANOVA models unless specified as contrast *P*-values. We used contrasts to test specific treatment hypotheses as outlined above. To test hypotheses about different types of warming responses, we fit stepwise (no and low vs. medium and high warming), linear, and unimodal (no and high vs. low and medium warming) patterns to the overall warming treatment means as well as to warming treatment means by precipitation treatments. We calculated effect sizes with confidence intervals based on Hedges' *d* for total, aboveground and belowground biomass treatment estimates (raw means) according to Nakagawa & Cuthill (2007), correcting for small sample bias. We used SAS 9.2 PROC MIXED for all analyses (SAS Institute Inc., 2008, Cary, NC, USA).

## Results

### Aboveground, belowground and total NPP

Total net primary production (TNPP, the sum of total aboveground and belowground NPP) and above-

ground net primary production (ANPP) responded to interactions of warming and precipitation treatments. Although main effects and interactions of the treatments were not significant for either TNPP or ANPP ( $P = 0.1737$  and  $P = 0.1713$ , respectively), our *a priori* contrasts showed that warming altered plant growth in the drought treatment differently than in ambient and wet treatments (Table 1). In drought treatments, increasing levels of warming decreased annual ANPP, while in the ambient and wet treatments, increasing levels of warming had no to slightly positive effects on ANPP (Fig. 4), a pattern that was also reflected in TNPP (not shown). The interaction contrasts using linearly increasing effects of warming in drought vs. ambient and wet precipitation treatments ( $P = 0.0368$  and  $P = 0.0329$ ) fit slightly better than the contrasts hypothesizing stepwise (i.e., no and low vs. medium and high) responses to warming ( $P = 0.0492$  and  $P = 0.0411$ ) for TNPP and ANPP, respectively. A further contrast testing the differences between ambient and wet precipitation treatments at the highest levels of warming (medium vs. high warming) showed no further differences between these two precipitation treatments in either TNPP or ANPP ( $P = 0.4095$  and  $P = 0.3261$ , respectively).

The old-field community produced more total biomass in 2009 ( $656 \pm 34$  g m<sup>-2</sup> yr<sup>-1</sup>) than in 2010 ( $577 \pm 33$  g m<sup>-2</sup> yr<sup>-1</sup>), which was mostly due to inter-annual differences in belowground net primary production (BNPP). ANPP did not differ between 2009 ( $454 \pm 26$  g m<sup>-2</sup> yr<sup>-1</sup>) and 2010 ( $435 \pm 28$  g m<sup>-2</sup> yr<sup>-1</sup>),

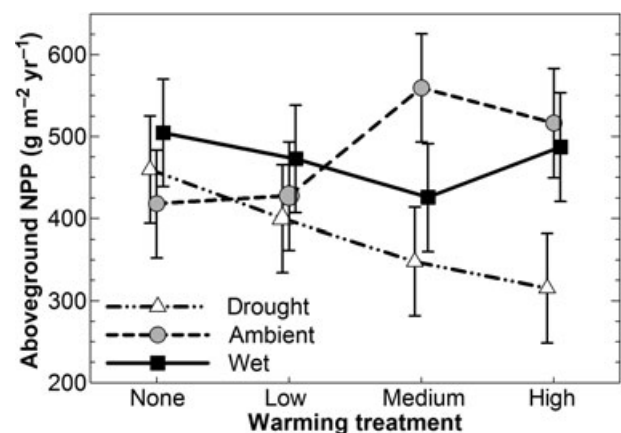


Fig. 4 Aboveground net primary production (mean  $\pm$  SE) along the warming gradient in each of the three precipitation treatments. Means are pooled across 2 years. In both treatment years, ANPP decreased along the warming gradient in drought treatments, but remained largely unaffected by warming in ambient and wet precipitation treatments. Treatment means are off-set to the left (drought treatments) or right (wet treatments) to facilitate the comparison of error bars.

**Table 1** Climate change effects on annual herbaceous plant biomass production: Repeated measures ANOVA effects and hypothesis-based planned comparisons. Plot estimates of biomass were based on two 0.25 × 0.5 m clip-plots in 2009 and on two 0.5 × 0.5 m clip-plots in 2010. Biomass was harvested in both June and October of each year

		TNPP	ANPP	Forb ANPP	Grass ANPP	Perennial ANPP	Non-perennial ANPP	C <sub>3</sub> ANPP	C <sub>4</sub> ANPP
ANOVA effects	df	P							
Precipitation (P)	2,4	0.7604	0.4236	0.4329	0.6629	0.4176	0.7703	0.7844	0.0807
Warming (W)	3,18	0.6998	0.9251	0.8250	0.9218	0.2590	0.3473	0.6957	0.2520
P × W	6,18	0.1737	0.1713	0.3051	0.4154	0.2216	0.8448	0.2411	0.5040
Year (Y)	1,24	0.0611	0.5893	0.4514	0.9583	<b>0.0105</b>	<b>0.0031</b>	<b>0.0336</b>	<b>0.0032</b>
P × Y	2,24	0.6946	0.6034	0.7607	0.3750	<b>0.0492</b>	0.1943	0.0557	<b>0.0250</b>
W × Y	3,24	0.7052	0.5121	0.6351	0.4072	0.3617	0.1746	0.5781	0.2590
P × W × Y	6,24	0.5641	0.7361	0.9305	0.8394	0.8317	0.6520	0.8111	0.6274
Disturbance	1,36	0.5609	0.5199	0.0668	<b>0.0288</b>	0.3003	0.5603	0.7756	0.6884
Hypothesis-based planned comparisons									
Precipitation main effects									
Precipitation: drought vs. other		0.4936	0.2168	0.3886	0.4641	0.2140	0.6155	0.5122	<b>0.0348</b>
Precipitation: ambient vs. wet		0.8987	0.9228	0.3211	0.6648	0.9190	0.5995	0.9589	0.6115
Warming main effects									
Warming: stepwise		0.5143	0.8696	0.8196	0.8163	0.3174	0.3041	0.5591	0.1878
Warming: unimodal		0.3623	0.7080	0.5325	0.6306	0.7262	0.9965	0.3993	0.1436
Warming: linear		0.4761	0.7480	0.6737	0.7261	0.1424	0.1523	0.4631	0.1749
P × W interactions									
P×W: drought vs. other, stepwise		<b>0.0492</b>	<b>0.0411</b>	0.5279	0.3581	0.3066	0.2147	0.1401	0.3181
P×W: drought vs. other, unimodal		0.6768	0.9563	0.5509	0.4254	0.8468	0.8627	0.7240	0.2441
P×W: drought vs. other, linear		<b>0.0368</b>	<b>0.0329</b>	0.7753	0.1618	0.2120	0.2781	0.1185	0.3162
P×W: ambient vs. wet, stepwise		0.0551	0.0612	<b>0.0456</b>	0.2850	0.0592	0.9013	0.0677	0.5332
P×W: ambient vs. wet×none vs. high		0.4402	0.2856	0.2803	0.5578	0.2874	0.9656	0.3681	0.9756
P×W: ambient vs. wet×med. vs. high		0.4095	0.3261	0.2762	0.6057	0.1438	0.4943	0.1737	0.1464
Precipitation by year interactions									
P×Y: drought vs. other by year		0.4673	0.3413	0.6553	0.1716	<b>0.0155</b>	0.0831	<b>0.0194</b>	<b>0.0070</b>
P×Y: ambient vs. wet by year		0.7111	0.8411	0.5318	0.6850	0.7807	0.5765	0.8341	0.9461
Warming by year interactions									
W×Y: stepwise by year		0.2770	0.2260	0.5212	0.7196	0.6375	<b>0.0476</b>	0.8257	0.1388
W×Y: linear by year		0.3550	0.3092	0.3876	0.9009	0.4105	<b>0.0318</b>	0.8578	0.2162

Effects significant at the  $\alpha = 0.05$  level are bold-faced for emphasis. Degrees of freedom (DF) presented are design-based and are listed in the order of numerator, denominator; actual model denominator DFs were calculated with the Kenward–Roger method and may be slightly lower in models with missing data.

despite pronounced inter-annual differences in summer precipitation and temperature (Fig. 3). BNPP in the top 30 cm, however, was greater in 2009 ( $201 \pm 23 \text{ g m}^{-2} \text{ yr}^{-1}$ ), the year characterized by a moister, cooler summer, than in 2010 ( $142 \pm 22 \text{ g m}^{-2} \text{ yr}^{-1}$ ), the year characterized by a hotter, drier summer ( $P = 0.0047$ ). The inter-annual variation we observed in TNPP and BNPP was substantial, but it was on a similar order of magnitude as interactive treatment effects. The TNPP effect sizes of years within each treatment combination (using 2009 as the control) ranged from  $-1.28$  to  $0.17$ , while the effect sizes of treatment combinations across both years (using ambient precipitation, unheated treat-

ments in each year as the controls) ranged from  $-0.47$  to  $0.78$ .

The BACE treatments affected deeper roots more than shallow roots and treatment effects varied significantly between years. Most of the roots of the old-field community at BACE were produced in the top 10 cm of the soil ( $64.3 \pm 2.5\%$  in 2009;  $56.0 \pm 2.6\%$  in 2010), and rapidly decreased with depth (10–30 cm roots:  $35.7 \pm 2.5\%$  in 2009;  $27.6 \pm 1.8\%$  in 2010; roots below 30 cm:  $16.4 \pm 1.4\%$ , 2010 data only). The interactive effect of precipitation by year on roots in the top 30 cm (Table 2) was driven by BNPP differences in the deeper soil layer ( $P = 0.0023$ ), as this interaction was not signif-

icant for roots in the top 10 cm ( $P = 0.2530$ ). The year by precipitation interaction for roots at 10–30 cm depth indicated that the decrease in BNPP from 2009 to 2010 affected roots in the drought treatments more strongly than roots in ambient or wet treatments (contrast  $P = 0.0032$ ; Fig. 5a). The particularly low root biomass in 2010 was likely due in part to the timing of collection of the root ingrowth cores; at the end of August, soil moisture levels had not yet recovered from the early-summer drought (Fig. 2) and root production was likely suppressed by the very low soil moisture preceding our sample collection. Precipitation treatments did not affect BNPP below 30 cm depth, and warming treatments did not significantly affect roots at any depth (Table 2). Overall, warming and precipitation affected aboveground biomass production more strongly than belowground biomass production (Supplementary Table S3).

#### Functional group biomass

Neither precipitation treatments nor warming treatments strongly affected forb or grass functional group contributions to ANPP (Table 1), but both groups were impacted by animal disturbance. Forb ANPP also showed a stepwise warming response that was opposite in ambient and wet precipitation treatments ( $P = 0.0456$ ): warming increased forb ANPP by 100% under ambient precipitation conditions, but decreased it by 53% in wet treatments (not shown). Animal disturbance reduced grass ANPP ( $P = 0.0288$ ) and tended to increase forb ANPP ( $P = 0.0668$ ), shifting the plant community closer to grass and forb co-dominance. From the onset of the experiment in 2008 until 2010, grass dominance across the site decreased from  $73 \pm 4\%$  to  $60 \pm 5\%$ .

Both perennial and non-perennial (i.e., the combination of annuals and biennials) ANPP changed between the two treatment years, with perennial ANPP responding more to precipitation and non-perennial ANPP responding more to warming treatments (Table 1). Perennial ANPP decreased from  $417 \pm 30 \text{ g m}^{-2} \text{ yr}^{-1}$  in 2009 to  $325 \pm 31 \text{ g m}^{-2} \text{ yr}^{-1}$  in 2010 ( $P = 0.0105$ ), while non-perennial ANPP increased from  $37 \pm 13 \text{ g m}^{-2} \text{ yr}^{-1}$  in 2009 to  $111 \pm 21 \text{ g m}^{-2} \text{ yr}^{-1}$  in 2010 ( $P = 0.0031$ ). The decrease in perennial ANPP from 2009 to 2010 occurred in ambient and wet precipitation treatments but not in drought treatments (contrast  $P = 0.0155$ ; Fig. 5b). A concurrent increase in non-perennials in the ambient and wet precipitation treatments was marginally significant (contrast  $P = 0.0831$ ). Non-perennial ANPP increased linearly along the warming gradient in 2010 (contrast  $P = 0.0318$ ; Fig. 6), offsetting a non-significant decreasing trend in peren-

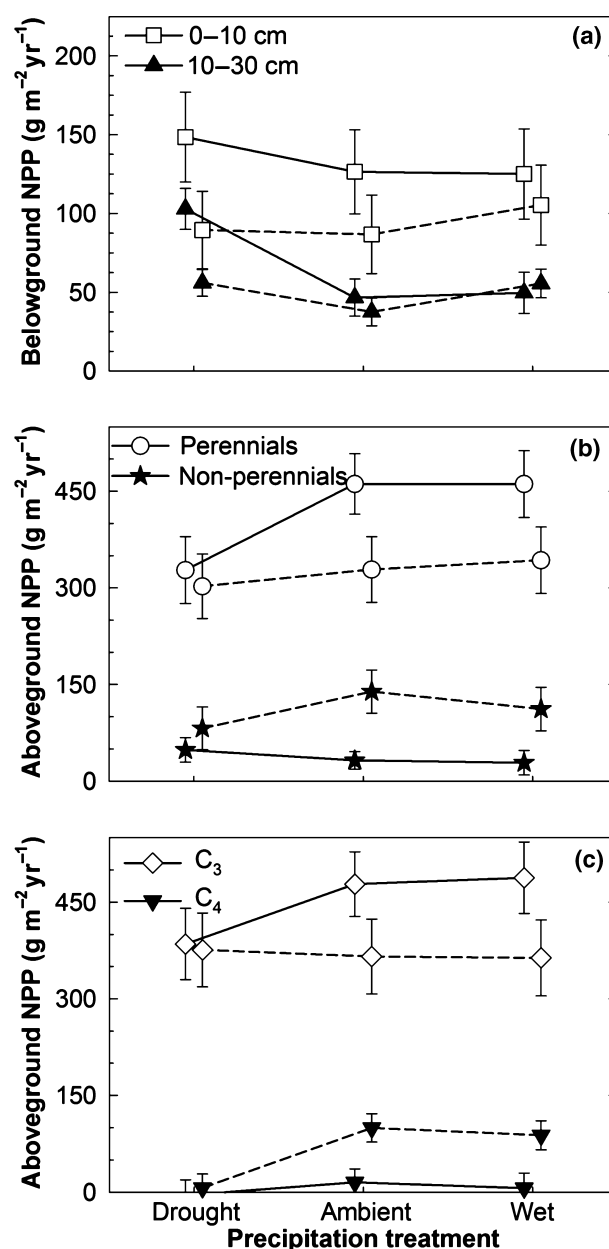


Fig. 5 The effect of precipitation treatments on BNPP at 10–30 cm depth and the ANPP contributions of perennial,  $C_3$ , and  $C_4$  functional groups (mean  $\pm$  SE) varied significantly between 2009 (solid lines) and 2010 (dashed lines). The treatment means are off-set to the left (2009) or right (2010) to facilitate the interpretation of year effects.

nial ANPP (contrast  $P = 0.4105$ ). Neither perennial nor non-perennial ANPP differed among warming treatments in 2009. Overall, the strong dominance of  $80 \pm 3\%$  perennials at the BACE in 2008 decreased to  $74 \pm 5\%$  by the end of 2010.

The functional group contributions of  $C_3$  and  $C_4$  plants to ANPP were both significantly affected by



**Table 2** Climate change effects on root growth based on root ingrowth cores collected annually in August in both 2009 and 2010: Repeated measures ANOVA effects and hypothesis-based planned comparisons

ANOVA effects	df	BNPP <sup>1</sup>	0–10 cm BNPP	10–30 cm BNPP	df	> 30 cm BNPP <sup>2</sup>
		<i>P</i>				<i>P</i>
Precipitation (P)	2,4	0.3731	0.9358	<b>0.0113</b>	2,3.1	0.0917
Warming (W)	3,18	0.5733	0.3094	0.3901	3,15.5	0.5611
P × W	6,18	0.7449	0.3399	0.3493	3,15.6	0.4933
Year (Y)	1,60	<b>0.0047</b>	<b>0.0234</b>	0.1039		
P × Y	2,60	<b>0.0060</b>	0.2530	<b>0.0023</b>		
W × Y	3,60	0.7988	0.2927	0.4826		
P × W × Y	6,60	0.5271	0.4518	0.4649		
Disturbance		0.9462	0.8210	0.3565	1,19	0.5790
Hypothesis-based planned comparisons						
Precipitation main effects						
Precipitation: drought vs. other		0.2102	0.7979	<b>0.0041</b>		0.1522
Precipitation: ambient vs. wet		0.5632	0.8134	0.2915		0.0690
Warming main effects						
Warming: stepwise		0.3506	0.6919	0.1954		0.2162
Warming: unimodal		0.3518	0.1302	0.4637		0.5446
Warming: linear		0.4468	0.9622	0.1316		0.2435
P × W interactions						
P×W: drought vs. other, stepwise		0.7609	0.2982	0.2302		0.1038
P×W: drought vs. other, unimodal		0.4891	0.0886	0.1278		0.6921
P×W: drought vs. other, linear		0.7999	0.2227	0.1104		0.0555
P×W: ambient vs. wet, stepwise		0.9689	0.9676	0.9500		0.7923
P×W: ambient vs. wet×none vs.high		0.4996	0.5943	0.7056		0.8651
P×W: ambient vs. wet×med. vs. high		0.7759	0.7611	0.9040		0.6371
Precipitation by year interactions						
P×Y: drought vs. other by year		<b>0.0032</b>	0.1600	<b>0.0010</b>		
P×Y: ambient vs. wet by year		0.3139	0.4538	0.3683		
Warming by year interactions						
W×Y: stepwise by year		0.8126	0.2046	0.1508		
W×Y: linear by year		0.8064	0.2406	0.2051		

<sup>1</sup>Total roots exclude roots below 40 cm depth;<sup>2</sup>Roots > 40 cm deep were only collected in 2010. Degrees of freedom (DF) presented are design-based and are listed in the order of numerator, denominator; actual model denominator DFs were calculated with the Kenward-Roger method and may be slightly lower in models with missing data.Effects significant at the  $\alpha = 0.05$  level are bold-faced for emphasis.

precipitation treatments, but the precipitation effects varied by year (Table 1). Across all treatments,  $C_3$  ANPP was higher in 2009 than in 2010 ( $P = 0.0336$ ), while  $C_4$  ANPP was higher in 2010 than in 2009 ( $P = 0.0032$ ).  $C_3$  ANPP in ambient and wet treatments was greater than in drought treatments in 2009, but precipitation treatments did not differ in 2010 (contrast  $P = 0.0194$ ) and overall remained at a level comparable to the 2009 drought treatment (Fig. 5c). The reverse was true for  $C_4$  ANPP, which was negligible in BACE plots in 2009, but which increased significantly in ambient and wet plots in 2010 (contrast  $P = 0.0070$ ). The ANPP contributions of  $C_3$  and  $C_4$  plants did not vary

significantly across the warming gradient or its interaction with precipitation treatments (Table 1). The near complete dominance of  $C_3$  plants at BACE in 2008 ( $99 \pm 1\%$ ) decreased to  $86 \pm 3\%$  by the end of 2010.

#### *Species richness and evenness*

Species richness in our old-field community was significantly affected by warming and precipitation treatments in both years, but the response patterns differed between 2009 and 2010. Species richness decreased along the warming gradient in 2009 (linear contrast  $P = 0.0041$ ), but the warming responses differed among

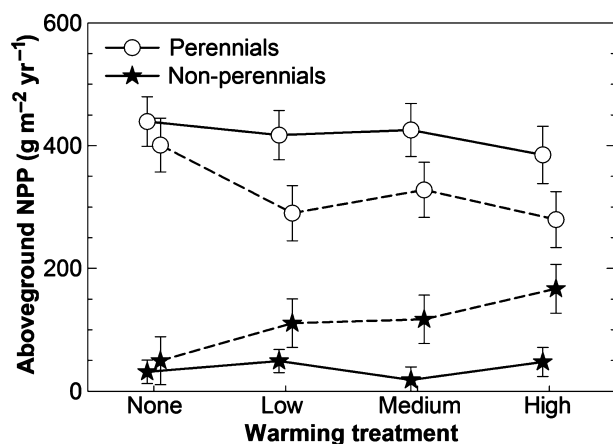


Fig. 6 Non-perennial (i.e., annual and biennial) ANPP (mean  $\pm$  SE) increased significantly along the warming gradient in 2010 (dashed lines), but not in 2009 (solid lines). The ANPP contribution of perennial plants did not change significantly along the warming gradient in either year. The treatment means are off-set to the left (2009) or right (2010) to facilitate the interpretation of year effects.

precipitation treatments: species richness declined linearly along the warming gradient in drought treatments, decreased only at higher levels of warming (stepwise) in wet treatments, and was highly variable in its response to warming in ambient precipitation treatments (Fig. 7). In 2010, drought treatments harboured

fewer species than ambient and wet treatments (contrast  $P = 0.0208$ ) and species richness in drought treatments decreased linearly along the warming gradient (contrast  $P = 0.0352$ ), whereas warming had no effect on species richness in the ambient and wet precipitation treatments (Fig. 7).

Effects of warming and precipitation on Shannon's evenness also interacted in 2009, but not in 2010. In 2009, species evenness decreased linearly along the warming gradient in drought treatments but not in ambient or wet precipitation treatments (contrast  $P = 0.0006$ ). The decrease in species evenness in warmed drought treatments in 2009 matched the observed trend of decreased species richness in these treatments in 2009 and 2010 (Fig. 7). Neither precipitation nor warming treatments or their interactions affected Shannon's evenness in 2010 (Table 3). Plot disturbance by animals increased species evenness in both years ( $P = 0.0032$  in 2009,  $P = 0.0260$  in 2010).

#### Animal disturbance

Most of the BACE plots were disturbed by rodents during the two treatment years, but the severity of animal disturbance varied by warming treatment and year. Digging and tunnelling by meadow voles caused the most extensive soil disturbance, and voles also ate plant shoots and roots. A few cottontail rabbits also ate

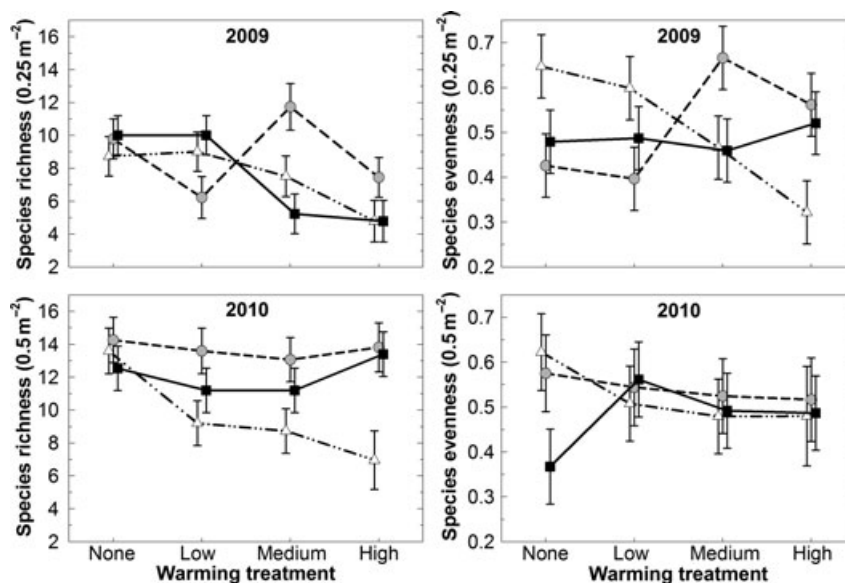


Fig. 7 The interactive effects of warming and precipitation treatments on species richness (mean  $\pm$  SE) and Shannon's evenness ( $E_{H'}$ , mean  $\pm$  SE) were year-specific. Species richness linearly decreased along the warming gradient regardless of precipitation treatments in 2009, but decreased along the warming gradient only in drought treatments in 2010 (significant interaction). Shannon's evenness decreased linearly along the warming gradient in drought treatments while increasing in a stepwise manner along the warming gradient in the ambient precipitation treatment in 2009, but did not differ between any warming or precipitation treatments in 2010. Note that species richness and Shannon's evenness were measured on smaller samples in 2009 than in 2010. Treatment means are off-set to the left (drought treatments) or right (wet treatments) to facilitate the comparison of error bars.

or damaged shoots. Animals disturbed plots more extensively in 2010 than in 2009 ( $P < 0.0001$ ; Fig. 8). In 2009, animal disturbance was highest in ambient temperature treatments and decreased linearly along the warming gradient, whereas in 2010 animal disturbance did not differ between warming treatments (linear warming by year contrast:  $P = 0.0182$ ). There was an indication that animal disturbance also varied significantly among precipitation treatments in different years (ANOVA precipitation by warming interaction:  $P = 0.0118$ ), but pairwise differences between all possible precipitation by year combinations were not significant. As a covariate, the animal disturbance index significantly predicted decreases in grass ANPP and increases in species evenness in plots.

## Discussion

### *Treatment effects and response patterns*

Results from the first two full years of treatments supported our initial hypotheses: ecosystem processes

responded interactively to warming and precipitation treatments, and at least some processes responded non-linearly. Neither large changes in precipitation amount nor moderate increases in temperature affected TNPP or ANPP by themselves; only the combination of drought and warming decreased TNPP across both years. The high sensitivity of grasslands to annual weather conditions often interacts with climate change treatments in experiments (Zavaleta *et al.*, 2003b; Engel *et al.*, 2009; Bloor *et al.*, 2010), sometimes eclipsing treatment effects in importance (e.g., Grime *et al.*, 2008). Our detection of interactive treatment effects despite large year-to-year variation in weather conditions shows that these ecosystem responses were robust and likely to occur across a variety of background conditions. These results, along with those from several other studies (e.g., De Boeck *et al.*, 2011; Kardol *et al.*, 2010b; Bai *et al.*, 2010; Andresen *et al.*, 2010; but see Bloor *et al.*, 2010; Niu *et al.*, 2008), indicate that single-factor climate change experiments with precipitation or warming can provide an incomplete picture of ecosystem responses.

**Table 3** Climate change effects on species richness and Shannon's evenness effects in 2009 and 2010: Repeated measures ANOVA effects and hypothesis-based planned comparisons. Plot estimates were based on two  $0.25 \times 0.5$  m clip-plots in 2009 and on two  $0.5 \times 0.5$  m clip-plots in 2010

		Species richness		Shannon's evenness	
		2009	2010	2009	2010
ANOVA effects	df	<i>P</i>			
Precipitation (P)	2,4	0.2264	<b>0.0421</b>	0.8945	0.5611
Warming (W)	3,18	<b>0.0148</b>	0.1948	0.7227	0.9282
P $\times$ W	6,18	<b>0.0292</b>	0.3830	<b>0.0112</b>	0.6843
Disturbance	1,18	0.6401	0.4316	0.0032	<b>0.0260</b>
Hypothesis-based planned comparisons					
Precipitation main effects					
Precipitation: drought vs. other		0.1716	<b>0.0208</b>	0.8679	0.8032
Precipitation: ambient vs. wet		0.0996	0.1508	0.6728	0.3329
Warming main effects					
Warming: stepwise		<b>0.0161</b>	0.1821	0.8764	0.5397
Warming: unimodal		0.3535	0.1582	0.6371	0.8490
Warming: linear		<b>0.0041</b>	0.1273	0.6365	0.5918
P $\times$ W interactions					
P $\times$ W: drought vs. other, stepwise		0.5204	0.0778	<b>0.0007</b>	0.5086
P $\times$ W: drought vs. other, unimodal		0.4407	0.9695	0.6308	0.4002
P $\times$ W: drought vs. other, linear		0.6588	<b>0.0352</b>	<b>0.0006</b>	0.3934
P $\times$ W: ambient vs. wet, stepwise		<b>0.0018</b>	0.6761	0.0625	0.6375
P $\times$ W: ambient vs. wet $\times$ none vs. high		0.2664	0.6803	0.5139	0.3696
P $\times$ W: ambient vs. wet $\times$ med. vs. high		0.1529	0.6398	0.2546	0.9894

Effects significant at the  $\alpha = 0.05$  level are bold-faced for emphasis. Degrees of freedom (DF) presented are design-based and are listed in the order of numerator, denominator; actual model denominator DFs were calculated with the Kenward-Roger method and may be slightly lower in models with missing data.

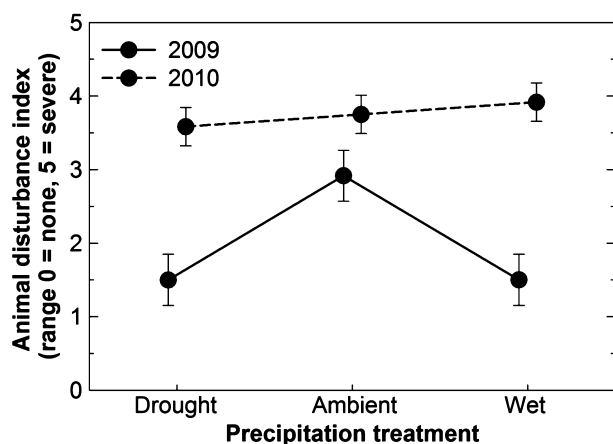


Fig. 8 Animal disturbance severity (mean  $\pm$  SE) by warming treatments in mid-March in 2009 (solid lines with black symbols) and 2010 (dashed lines with white symbols). Animal disturbance was visually assessed in each plot using an ordinal scale ranging from 0 (no visible disturbance) to 5 (> 75% of the plot surface disturbed).

Simple main effects of precipitation and warming treatments impacted species richness, annual root biomass production, and the ANPP contributions of specific plant functional groups, although these effects were sometimes specific to one of the two treatment years. Ecosystem responses to warming were often either linear or step-wise along the warming gradient, indicating that intermediate warming responses could be inferred from higher end-point warming treatments, but that effects of low levels of warming ( $\sim 1^\circ\text{C}$ ) may be subtle and hard to detect in field studies with small sample sizes. In our mesic old-field ecosystem, precipitation multi-level responses were generally due to differences between drought treatments and the wet and ambient treatments, and rarely identified any differences between ambient and wet precipitation treatments. Old fields and grasslands in which the mean soil water content is low and water deficits persist through the winter would likely show a stronger effect of precipitation additions (e.g., Chou *et al.*, 2008; Chimner *et al.*, 2010).

#### Belowground responses

As expected, drought increased BNPP, at least in the cool, wet summer of 2009. We were surprised to find that warming did not enhance BNPP in our study, as has been found in other studies (e.g., Hutchison & Henry, 2010; Niu *et al.*, 2010). Rates of root growth and mortality often increase with warming (Pregitzer *et al.*, 2000), although this response can be strongly mediated by water availability (Bai *et al.*, 2010). By collecting root ingrowth cores only once a year, we did not account for

seasonal root turnover and thus underestimated BNPP. It remains a possibility that if warming enhanced root growth in our old-field ecosystem, such increases in root growth were balanced by increased root mortality. Since aboveground and belowground responses can partially offset one another under dry conditions, both NPP components need to be considered in estimating changes in carbon inputs.

#### Functional group responses

The fairly rapid shifts in abundance of different functional groups in specific climate treatments despite largely stable TNPP suggest that the species composition of our old-field community was able to compensate quickly for changing temperature and soil moisture conditions. At the onset of the experiment, our old-field community was dominated by perennials and  $C_3$  plants. By 2010, the ANPP contribution of annuals and biennials had significantly increased in warmed plots and  $C_4$  grasses grew into ambient and wet precipitation treatments. The expansion of  $C_4$  species relative to  $C_3$  species during the hot and dry growing season of 2010 very closely mirrors similar  $C_4$  expansions in other grassland communities subjected to warming or extreme heating events (White *et al.*, 2000; Wan *et al.*, 2005; Sherry *et al.*, 2008). It is likely that soil moisture conditions in our drought treatments in 2010 presented unfavourable growing conditions even for drought-resistant  $C_4$  grasses. The concurrent increase of annuals and biennials in the ambient and wet treatments in 2010 is likely due to the fact that perennial  $C_3$  plants were replaced by annual  $C_4$  grasses, although it is not clear whether plants gained a competitive advantage due to their photosynthetic pathway, life history, or other differences correlated with these traits. The increased abundance of annuals and biennials along the warming gradient in 2010 could be due in part to increased early growth by spring annuals and in part to increased animal disturbance, as disturbance has been shown to increase the dominance of annuals and to reduce that of perennials in grassland communities (Wilson & Tilman, 1991). Such changes in plant functional group composition may alter the character of the ecosystem's response to climate going forwards (Smith *et al.*, 2009).

#### Biodiversity in a changing climate

Experimental warming generally decreased species richness in our old-field community plots, particularly under drought conditions. This finding supports observations of species losses made in other climate change experiments (e.g., Grime *et al.*, 2008; Arnone *et al.*, 2011;

Niinemets *et al.*, 2011), although experimental warming has not reduced species richness in all cases (e.g., Price & Waser, 2000; Zavaleta *et al.*, 2003a; Bloor *et al.*, 2010). Significant treatment effects on species evenness in 2009 preceded the treatment effects on species richness in 2010 (Fig. 7), suggesting that species evenness may function as a temporary early indicator of community change. Consistent with observations by Kardol *et al.* (2010a), we found a negative correlation between aboveground biomass and evenness (Supplementary Fig. S1). However, the relationship was quite weak, and we found no persistent differences in species evenness across our precipitation treatments. Greater plant species richness has been found to enhance the temporal stability of grassland annual aboveground plant production (Tilman *et al.*, 2006; Eisenhauer *et al.*, 2011). Species richness can also affect a plant community's ability to resist and to recover from disturbances such as droughts (e.g., Van Ruijven & Berendse, 2010). Depending on which species are lost, a decline in species richness in grassland ecosystems due to global warming could increase inter-annual variability in aboveground biomass production and could make them more vulnerable to droughts.

Species loss due to global warming or drought conditions might be less than is observed under abrupt experimental change; more gradual change would allow adaptation and immigration to occur. On the other hand, field experiments might under-estimate declines in species richness, because treatment plots are typically small and surrounded by unaffected habitat, which facilitates species recruitment from the surrounding species pool. However, even on ecosystem scales, small-scale topographical heterogeneity may be sufficient to provide plants with drought refugia that may play a key role in conserving biodiversity under climate change (Godfree *et al.*, 2011). Overall, the net effects of climate change on ecosystem plant diversity may be difficult to predict from field experiments, because ecological drivers of species diversity at smaller spatial and temporal scales are often different from ecological drivers at larger scales (Willis & Whittaker, 2002).

#### *Old fields, carbon uptake and climate change*

We extrapolate our results to the long term with caution, because inter-annual variation in the local climate can cause short-term ecosystem responses that may weaken, intensify, or even change direction over time. The different ecosystem responses over time likely express different phases of change due to ecosystem processes acting at different time scales (Shaver *et al.*, 2000; Luo *et al.*, 2011). Climate change effects on ecosystem functions may diminish over time as resources

are depleted (Kirschbaum, 2004; Beier *et al.*, 2008; Allison *et al.*, 2010) or plant community compositions shift (e.g., Harte *et al.*, 2006; Luo *et al.*, 2009; Langley & Magonigal, 2010). In some ecosystems, however, climate change effects may persist over relatively long time periods (e.g., Jackson *et al.*, 2009; Melillo *et al.*, 2011). Given the complex interactions between ecosystem processes operating on different time scales, longer term experiments and ecosystem models will be essential tools for forecasting the development and net effects of ecosystem responses and feedbacks to global environmental change.

In our old-field ecosystem, the combination of drought and warming decreased TNPP (by 10% in the warmest treatment) across both years, while TNPP remained relatively stable across single factor manipulations. Current regional climate change projections for the US Northeast project average temperature increases of 2.9–5.3 °C by 2070–2099 relative to 1961–1990, and a concurrent increase in the frequency of drought events despite unchanged summer rainfall totals (Hayhoe *et al.*, 2007). Our data suggest that old-field productivity and species diversity are likely to remain stable under these conditions in most years, but that both will become increasingly susceptible to droughts. Other results from this system suggest that decreases in carbon inputs under drought would be at least partially offset by decreases in carbon outputs, as drought and warming together increase the chemical recalcitrance of some litter (Tharayil *et al.*, 2011), and drought slows heterotrophic respiration (Suseela *et al.*, 2012). We expect that the increased presence of both annuals/biennials and C<sub>4</sub> species in our old-field ecosystem will primarily affect ecosystem carbon balance by buffering community TNPP from extreme weather events, although these shifts may impact litter quality. A recent study by Gentile *et al.* (2011), however, showed that long-term soil carbon dynamics are impacted more by litter quantity than quality, and increased dominance of C<sub>4</sub> grasses did not alter the net ecosystem carbon balance or soil carbon content of a tallgrass prairie over 7 years of warming (Luo *et al.*, 2009), despite causing an increase in litter mass. We found no indication that warming would dramatically increase carbon uptake of our old-field community under any precipitation conditions, suggesting that, unless the plant community is highly responsive to elevated CO<sub>2</sub>, the system is unlikely to offset climate change through accelerated carbon sequestration.

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## Supporting Information

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

**Figure S1.** Regression of species evenness by aboveground net primary production over 3 years (2008–2010).

**Table S1.** Names, growth forms, life history types, and photosynthetic pathways of all old-field species occurring at BACE

**Table S2.** Annual herbaceous biomass and functional group composition from the onset of the BACE experimental treatments in 2008–2010 ( $n = 3$  for each treatment mean)

**Table S3.** Climate change effects on annual herbaceous biomass ( $n = 6$  for each treatment mean: three treatment replicates in each of 2 years; means and standard deviations are based on raw data)

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