

Daily environmental conditions determine the competition–facilitation balance for plant water status

Alexandra Wright^{1,2*}, Stefan A. Schnitzer¹ and Peter B. Reich^{3,4}

¹Department of Biological Sciences, University of Wisconsin-Milwaukee, 3209 N Maryland Ave, Milwaukee, WI 53201, USA; ²Biological Sciences, Bard College, Annandale-on-Hudson, NY 12504; ³Department of Forest Resources, University of Minnesota, 1530 Cleveland Ave. N, St. Paul, MN, USA; and ⁴Hawkesbury Institute for the Environment, University of Western Sydney, Penrith, NSW 2751, Australia

Summary

1. Plants compete with their neighbours for a finite set of limiting resources, and this decreases individual plant performance, growth and survival. However, neighbouring plants also affect each other in positive ways.

2. Positive facilitative effects can occur when neighbouring plants ameliorate harsh abiotic conditions (temperature, wind and high irradiation). Thus, when environmental conditions are severe, the importance of facilitation may increase. The co-occurrence and masking effects of competition and facilitation among neighbouring plants have made it difficult to tease them apart in the past.

3. We planted bur oak acorns (*Quercus macrocarpa*) into an experimental diversity gradient in a central MN grassland that provided a gradient in plant biomass. We predicted that greater biomass of neighbours would increase both competition and facilitation as measured by impacts on the minimum leaf water potential reached on any given day. Under moderate conditions, competition should predominate, but under hot/dry conditions, facilitation should become more important. We measured temperature, humidity and soil moisture in these plots for two growing seasons, as well as oak seedling leaf water potential across a range of daily conditions.

4. On cool/humid days, plant interactions were dominated by competition for soil water: leaf water potentials of juvenile oaks were lower in plots with greater herbaceous biomass (and higher diversity). Conversely, on hot/dry days, facilitation of the microclimate determined the net effect of plants on their neighbours: leaf water potentials of juvenile oaks were higher in plots with higher herbaceous diversity and biomass.

5. *Synthesis.* In terms of plant water status, plant interactions among neighbours can flip from net negative (competition) to net positive (facilitation) depending on daily abiotic conditions. The relative importance of both positive and negative interactions for plant water status may affect the overall performance of plants over time.

Key-words: climate change, environmental severity, microclimate amelioration, plant–plant interactions, resource limitation, soil moisture, stress-gradient hypothesis, vapour pressure deficit

Introduction

Plants compete for limiting resources and the outcome of competitive plant interactions can help explain community composition, global plant distributions and species coexistence (Hardin 1960; Tilman 1977; Bond, Woodward & Midgley 2005). At the individual plant level, competition for shared resources can decrease growth (Ehleringer 1984; Gordon, Menke & Rice 1989) and increase mortality (Davis *et al.* 1999) of neighbouring plants. A key limiting resource in

many ecosystems is soil water. In the short term, when plants are water limited due to competition for soil water, they may close their stomates to reduce water loss at the leaf surface, thus leading to decreased photosynthetic rates, plant growth and survival (Gordon, Menke & Rice 1989). However, upon longer-term exposure to water stress, plants may make other morphological (leaf orientation) or physiological (osmotic potential) adjustments to retain higher carbon assimilation rates (Brown, Jordan & Thomas 1976).

Plants may also facilitate each other via amelioration of the local microclimate (Brooker *et al.* 2008). In fact, facilitation may be a component of many plant interactions, but it may be commonly obscured by competition (Stachowicz 2001;

*Correspondence author: E-mail: sashajwright@gmail.com

Bruno, Stachowicz & Bertness 2003). Plants can modify their local microclimate via shading and evaporative cooling. This environmental amelioration may be particularly important when environmental conditions are harsh and would otherwise cause plant physiological stress (the stress-gradient hypothesis, Bertness & Callaway 1994). For example, in hot arid ecosystems, environmental facilitation by neighbouring plants often results in increased germination, growth and survival of plants growing near neighbours, leading to clumped spatial distributions (Cuesta *et al.* 2010; Jia *et al.* 2010; Landero & Valiente-Banuet 2010; Armas, Rodríguez-Echeverría & Pugnaire 2011). In arid systems, physiological constraints related to microclimate may be more important than competition for resources. Plants may be negatively affected by high rates of water loss at the leaf surface, due to high vapour pressure deficit in the microclimate (Wright, Schnitzer & Reich 2014), or directly, due to photoinhibition at high light levels (Valladares & Pearcy 1997). Facilitation may be dominant in these stressful abiotic conditions, but weaken and become subordinate to competition as environmental stress lessens (Callaway *et al.* 2002).

In the past decade, theoretical and experimental work has suggested that both competition and facilitation may be ubiquitous in many plant communities, not solely the most arid ones (Bruno, Stachowicz & Bertness 2003; Montgomery, Reich & Palik 2010; Wright, Schnitzer & Reich 2014). We posit that, in terms of plant water status, the temporal dynamics of facilitation relative to competition may be a function of daily differences in abiotic conditions and therefore may change on a day-to-day basis. On cool humid days, plant water potential may be less negative overall, and the effects of facilitation due to microclimate amelioration from neighbours may be weak. In these cases, competition for soil water resources may dominate: plants growing in dense communities may experience more water stress due to more competition for water below-ground. Conversely, on hot/dry days, evaporative demand in the microclimate may be high, overall plant water status may be more negative, and the positive effects of facilitation due to microclimate amelioration from neighbours may dominate. Consequently, both processes may vary in relative strength over time and the total of competitive and facilitative interactions may affect longer-term performance (growth and survival).

We planted bur oak acorns (*Q. macrocarpa*) into 85 2 × 2 m experimental plant communities that varied in species richness from 1 to 16 species (Reich *et al.* 2012). Plant biomass increases strongly with species richness in this experiment; hence, the species richness gradient also serves as a gradient in neighbour density and biomass (Reich *et al.* 2012). We measured leaf water potential of oaks across a range of daily conditions at this site. We explored the relative importance of daily competition and facilitation in terms of plant water status, and how the balance of these two processes changes over the course of a growing season. We tested the specific hypothesis that both competition and facilitation are stronger in higher richness/higher biomass plant communities, but that these processes trade-off over short time periods depending on the relative severity of daily conditions. We predicted that:

- 1 On cool humid days, competition for soil water would be the dominant process driving plant water status, and thus, plant water status of juvenile oaks in higher diversity/higher biomass communities would be more negative than plant water status in lower diversity/lower biomass communities.
- 2 On hot/dry days, facilitation due to amelioration of the microclimate would be the dominant process driving plant water status, and thus, oak plant water status in higher diversity/higher biomass communities would be less negative than plant water status in lower diversity/lower biomass communities.

Materials and methods

SITE DESCRIPTION

We conducted this study from May 2010 to October 2012 in the ambient treatment plots of the BioCON plant diversity experiment at the Cedar Creek Ecosystem Science Reserve (CCESR) in central Minnesota, USA. Cedar Creek has a continental climate, with cold winters and warm summers and an average of 660 mm of rainfall per year (Reich *et al.* 2001b). Annual temperatures in 2012 were the warmest on record in the United States (NCDC 2012), and the CCESR field site was also drier than usual. Annual precipitation for 2012 (January–December) was 495 mm, which was 25% below the long-term average of 660 mm yr⁻¹ (Reich *et al.* 2001b; Fig. 1). Mean annual air temperature (January–December) at Cedar Creek, however, was 8.1 ± 1.3 °C (mean ± 95% confidence intervals), which was statistically consistent with the 24-year average (6.9 ± 1.2 °C, Cedar Creek Ecosystem Science Reserve hourly climate data), though July 2012 was the warmest month on record in the 24-year temperature data set (23.9 °C, Fig. 1).

Soils at this site are nutrient-poor glacial outwash sand plain with low water-holding capacity. The BioCON plots were established in 1997 by tilling and fumigating existing vegetation in six experimental blocks in an old field grassland at the site. Plots were then established as 2 × 2 m areas that were seeded with randomly assigned herbaceous species (all native or naturalized) from a pool of 16 total species from 4 functional groups – 4 C₃ grasses, 4 C₄ grasses, 4 legumes and 4 non-nitrogen fixing herbaceous plants (*Achillea millefolium*, *Agropyron repens*, *Amorpha canescens*, *Andropogon gerardi*, *Anemone cylindrica*, *Asclepias tuberosa*, *Bouteloua gracilis*, *Bromus inermis*, *Koeleria cristata*, *Lespedeza capitata*, *Lupinus perennis*, *Petalostemum villosum*, *Poa pratensis*, *Schizachyrium scoparium*, *Solidago rigida* and *Sorghastrum nutans*). There were 32 plots with 1 species (with every monoculture represented twice), 32 plots with 4 species, 9 plots with 9 species and 12 plots with 16 species. Since 1997, species mixes have been maintained by hand weeding to remove any species that migrated into the plot that were not planted in the original seed mix.

We conducted this experiment along a species richness gradient, which also provided a gradient in biomass production (Reich *et al.* 2001a, 2012), and both biomass and diversity may alter the relative strength of competition and facilitation intensity (Wright *et al.* 2013; Wright, Schnitzer & Reich 2014). Increased biomass production with greater plant diversity leads to reduced resource availability and increased intensity of competition for colonizing plants (Tilman, Wedin & Knops 1996; Fargione & Tilman 2005). Increased biomass production may also be associated with increased protection from environmental conditions. Consequently, increased biomass and

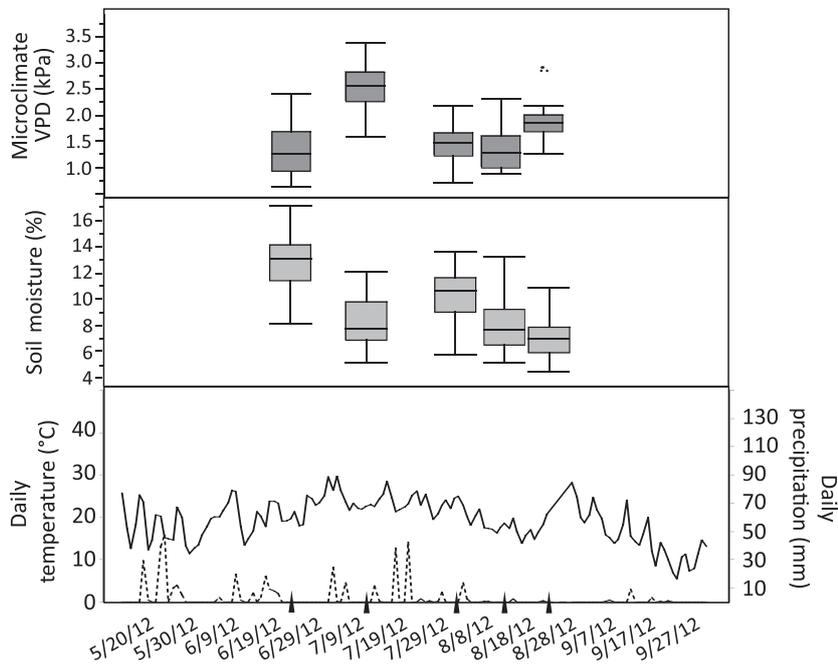


Fig. 1. We measured soil moisture in each plot on each day of measurement (a). We also measured relative humidity and air temperature in each plot, which we used to calculate vapour pressure deficit (b). We measured air temperature (solid line) and daily precipitation (dotted line) at the weather station at the site (c).

diversity may drive increased facilitation between plants due to amelioration of environmental extremes (Wright *et al.* 2013). In August 2012, we measured above-ground biomass of herbaceous species in all 85 plots. Above-ground biomass was clipped in 10×100 cm strips at the soil surface at least 15 cm from plot boundaries (to avoid edge effects).

We planted six bur oak (*Quercus macrocarpa*) acorns in all 85 plots in May 2010 and again in May 2012 to increase total seedlings available for the study. We planted acorns spaced at least 10 cm from any oak acorn neighbours. We measured leaf number, seedling height and seedling diameter of all seedlings at the beginning of the growing season (28 May 2012) and at the end of the growing season (27 August 2012) to assess relative growth rate (RGR) for each seedling. We also censused seedling emergence at the beginning of the first growing season (1 July 2010) and at the beginning of the 2012 growing season (28 May 2012). We recorded the survival of all seedlings that had emerged by 27 August 2012.

In May 2010, we planted a total of 37 bur oak acorns in a nearby harvest garden and harvested them periodically between July 2010 and July 2012, to derive allometric equations for biomass. We used these measurements to derive equations for both above-ground biomass ($AGB = 0.76 \times \text{diameter} - 0.02 \times \text{height} + 0.11 \times \text{leaves}$, $r^2 = 0.80$) and below-ground biomass ($BGB = 6.5 \times \text{diameter} - 0.12 \times \text{height} + 0.14 \times \text{leaves}$, $r^2 = 0.35$), though due to low r^2 values for BGB, we used AGB values to calculate RGR. We calculated above-ground RGR by taking $\ln(\text{final AGB}) - \ln(\text{initial AGB})/\text{duration}$ of study.

We measured pre-dawn and midday leaf water potential of a total of 151 randomly selected oak seedlings (stratified across the diversity gradient depending on seedling availability; see below), on five different sampling dates in 2012 (June 22, July 11, August 1, August 14 and August 24). For each sampling date, we also excluded seedlings that were at the largest (> 15 cm tall) or at the smallest (< 5 cm tall) ends of the seedling size spectrum. In other words, we restricted our sampling to seedlings that were relatively similar in size across the herbaceous biomass gradient. We chose sampling dates based on forecast data of maximum daily temperatures at the site, and we attempted to sample evenly across a range of temperatures

(20–30 °C). Oak leaves began to senesce the week of September 10, nearly 3 weeks after our final leaf water potential measurements (24 August 2012). The height of the herbaceous canopy varied depending on herbaceous biomass and species composition. Thus, oak leaves were more heavily shaded in the higher biomass plots. We sampled the youngest fully expanded leaf for all leaf water potential measurements. We measured leaf water potential using a Scholander pressure chamber (Soil Moisture Equipment Corp, Santa Barbara, CA, USA). Pre-dawn water potential (ψ_{pd}) was taken 2 h before dawn each day (0330–0530), and midday leaf water potential (ψ_{md}) was taken at solar noon (1200–1400). All measurements were taken by wrapping leaves in Ziploc bags, excising leaves using a razor blade and immediately transferring to the pressure chamber.

Plants were chosen for water potential measurements based on sampling at least five oak seedlings per age group per species richness level on each day, each oak having at least 3 leaves fully expanded at the time of sampling and not sampling from the same plant two sampling dates in a row. We sampled equally from the 2010 and 2012 age cohorts on each day of sampling. When more than one plot, or more than one plant within plot, met the above requirements, the plant was chosen randomly from the subset of available plants. Midday measurements were taken on the same plants as pre-dawn measurements to assess daily changes in plant water stress at the individual plant level. We used a comparison between pre-dawn (ψ_{pd}) and midday (ψ_{md}) leaf water potential ($\psi_{md} - \psi_{pd}$) to detect the daily change in plant water status at the individual plant level. A total of 27–37 plants were sampled on each sampling date depending on the constraints described above.

From May 2011 to October 2011 and May 2012 to October 2012, we measured plot-level air temperature and relative humidity (RH) on the north-western quadrant of the plot (> 50 cm from the plot edge) continuously using Maxim iButton dataloggers logging every 5 minutes (Maxim Integrated, San Jose, CA, USA). We then calculated vapour pressure deficit of the microclimate $VPD = \left(0.6108 \times e^{\frac{17.27 \times \text{Temp}}{\text{Temp} + 237.3}}\right) - \left(\frac{\text{RH}}{100} \times 0.6108 \times e^{\frac{17.27 \times \text{Temp}}{\text{Temp} + 237.3}}\right)$, (Anderson 1936). Dataloggers were installed on wooden tent stakes at approximately 20 cm above the ground surface and covered with plastic Dixie

cups. This ensured that the dataloggers were recording microclimate conditions 5–15 cm above the height of the oak seedlings (a conservative measure of average microclimate conditions under the herbaceous canopy). The covers were painted white to reflect direct sunlight and guard from direct saturation by rainwater. These dataloggers were installed in all plots where leaf water potential measurements were collected. We also measured shallow soil moisture (~6 cm depth) in these plots at 11:00 a.m. on the same day as leaf water potential measurements using an HH2 soil moisture meter with theta probe (Delta-T Devices Ltd, Cambridge, UK). Additional dataloggers were installed in a randomly assigned subset of plots throughout the rest of the experimental plots and moved every 2 weeks. We collected site-level temperature, humidity and vapour pressure deficit measurements from the Cedar Creek Ecosystem Science Reserve weather station.

ANALYSIS – MICROCLIMATE

We analysed the effects of herbaceous plant biomass on microclimate (air temperature and vapour pressure deficit) and whether the magnitude of these effects changed depending on site conditions. We calculated plot-level averages for microclimate temperature and VPD for each day 24-h period during the study ('plot level'). We also calculated daily averages for air temperature and vapour pressure deficit data collected from the Cedar Creek weather station ('site level'). We conducted separate mixed-effects ANCOVA to assess the effects of daily conditions (temperature and vapour pressure deficit), and the amelioration of these conditions by the plant community, on microclimate. To account for spatial autocorrelation associated with the blocked ring design and temporal autocorrelation associated with taking multiple measurements on the same plots over time, we included plot nested within block as a random effect. These two ANCOVAs assessed the following: (i) the fixed effects of daily average temperature at the site (taken from Cedar Creek weather station data), herbaceous biomass and their first-order interaction effects on microclimate temperatures and (ii) the fixed effects of daily average VPD at the site, herbaceous biomass and their first-order interaction effects on microclimate VPD.

ANALYSIS – SOIL MOISTURE

To assess the effects of daily changes in weather, seasonal trends in soil moisture at the site (the sandy soil rapidly becomes dry between major rain events and over the course of the season, Adair *et al.* 2011) and herbaceous biomass, on soil moisture, we conducted a mixed-effects ANCOVA with site-level VPD, herbaceous biomass and day of year as fixed effects. To account for spatial autocorrelation associated with the blocked ring design and temporal autocorrelation associated with taking multiple measurements on the same plots over time, we included plot nested within block as a random effect. Since we measured soil moisture in a 30-plot subset of the total plots used for this study, based on where leaf water potential measurements were taken on any given day, statistical power was too low across the gradient in herbaceous biomass on any single day to include random effects for ring and plot in this soil moisture statistical model.

ANALYSIS – LEAF WATER POTENTIAL

We conducted a mixed-effects ANCOVA to assess the effects of herbaceous biomass, site-level climatic conditions (VPD) and their interaction, on all measures of oak leaf water potential (Ψ_{pd} , Ψ_{md} , $\Psi_{md} - \Psi_{pd}$). For both pre-dawn and midday leaf water potential, we assessed the effects of site-level VPD for the 6-hr period prior to

water potential measurements. For the daily changes in leaf water potential ($\Psi_{md} - \Psi_{pd}$), we assessed the effects of site-level VPD integrated over this entire period (00:00–14:00). To account for spatial autocorrelation associated with the blocked ring design and temporal autocorrelation associated with taking multiple measurements on the same plots over time, we included plot nested within block as a random effect. To account for differences in the age of seedlings (the cohort from 2010 or 2012 described above), we included a random effect for seedling age in the statistical model as well.

We conducted a separate analysis to explore the plot-level mechanisms for competition and facilitation on all three measures of oak leaf water status (Ψ_{pd} , Ψ_{md} , $\Psi_{md} - \Psi_{pd}$). We directly assessed the effects of soil water and microclimate VPD on pre-dawn, midday and daily differences in oak water potential using a mixed-effects ANCOVA (microclimate VPD comparisons were made using the same time intervals as described above for site-level comparisons). To account for spatial autocorrelation of measurements taken within the same block and temporal autocorrelation associated with taking measurements on the same plot over time, we included plot nested within block as a random effect. We also included a random effect for seedling age to account for any differences between the two seedling ages in the statistical model.

ANALYSIS – GROWTH AND SURVIVAL

We analysed the effects of herbaceous biomass on above-ground oak seedling RGR and proportion survival (arcsine transformed) using a mixed-effects ANCOVA with plot nested in block and seedling age (cohort) included as random effects.

We also repeated all of the above analyses by replacing herbaceous biomass above with species richness. These results were qualitatively very similar and are included in Appendix S1 in Supporting Information.

Results

Over the course of our five leaf water potential measurement campaigns, daily temperature was the highest on July 11 and August 24 and lowest on June 22 and August 14. Relative humidity was highest in June and lowest in early July. This resulted in VPD being highest on July 11 and lowest on June 22 (Fig. 1a). There was no correlation between sampling date and temperature in the plots (Pearson product moment correlation coefficient, $r = 0.06$, $N = 168$) and no correlation between sampling date and relative humidity in the plots ($r = 0.16$, $N = 168$) – in other words, we did not sample all cool/humid days in the early summer and all hot/dry days in the late summer (Fig. 1a). There was a weak negative correlation between soil moisture and VPD at the site ($r = -0.24$, $N = 168$).

MICROCLIMATE AND SOIL MOISTURE

Under the herbaceous canopy layer (~20 cm above the soil surface), plots with higher species richness and more above-ground biomass were cooler, more humid and had lower vapour pressure deficit than plots with less biomass and fewer species (Table 1, Fig. 2, Appendix S1). The magnitude of the microclimate amelioration effect was stronger as daily weather conditions became hotter and drier (significant inter-

Table 1. Separate anovas for the effects of daily temperature on microclimate temperature and daily VPD on microclimate VPD. We also tested for interactions between these factors and above-ground biomass (AGB)

Fixed Effect	d.f. [†]	R^2	Plot Temp		Plot VPD	
			F	P	F	P
Herb AGB	1, 9306	0.96	66.24	< 0.0001*		
Site Temp	1, 9525		211 750	< 0.0001*		
Herb AGB × Site Temp	1, 9524		77.47	< 0.0001*		
Herb AGB	1, 8969	0.88			27.80	< 0.0001*
Site VPD	1, 9227				50 089	< 0.0001*
Herb AGB × Site VPD	1, 9230				482.5	< 0.0001*

[†]This analysis took into account spatial variation associated with the blocked design ('block' in the BioCON framework). In the linear mixed-effects model framework, denominator degrees of freedom 'float' based on the degree of variation attributed to random effects. Asterisks (*) indicate statistical significance.

action terms, Table 1, Fig. 2). In contrast, soil moisture was lower in diverse, higher biomass plots over the course of the season (except for early in the season) and progressively lower in the highest diversity and highest biomass plots during the growing season (Table S1, Fig. S1, Appendix S1). Thus, high-diversity plots with high biomass had lower residual soil water availability, but more mesic temperatures and less atmospheric evaporative demand.

WATER STRESS, GROWTH AND SURVIVAL

All oaks grew $\sim 2\times$ less in the highest diversity ($F_{1,57} = 13.6$, $P = 0.0005$) and $\sim 4\times$ less in the highest

biomass ($F_{1,60} = 30.2$, $P < 0.0001$) plots. Oak emergence was $\sim 1.5\times$ higher in the highest diversity compared with one-species plots ($F_{1,80} = 16.5$, $P = 0.0001$) and almost $3\times$ higher in the highest biomass plots ($F_{1,80} = 19.4$, $P < 0.0001$). Survival of emerged seedlings was equal across the diversity gradient ($F_{1,80} = 2.15$, $P = 0.15$) but almost $3\times$ higher in the highest biomass plots ($F_{1,80} = 5.33$, $P = 0.02$).

Pre-dawn leaf water potential values were unaffected by daily changes in site-level VPD or community biomass (Fig. S2, Table 2). The majority of the variation in pre-dawn leaf water potential values was due to soil moisture, where higher soil moisture led to less negative pre-dawn leaf water potential

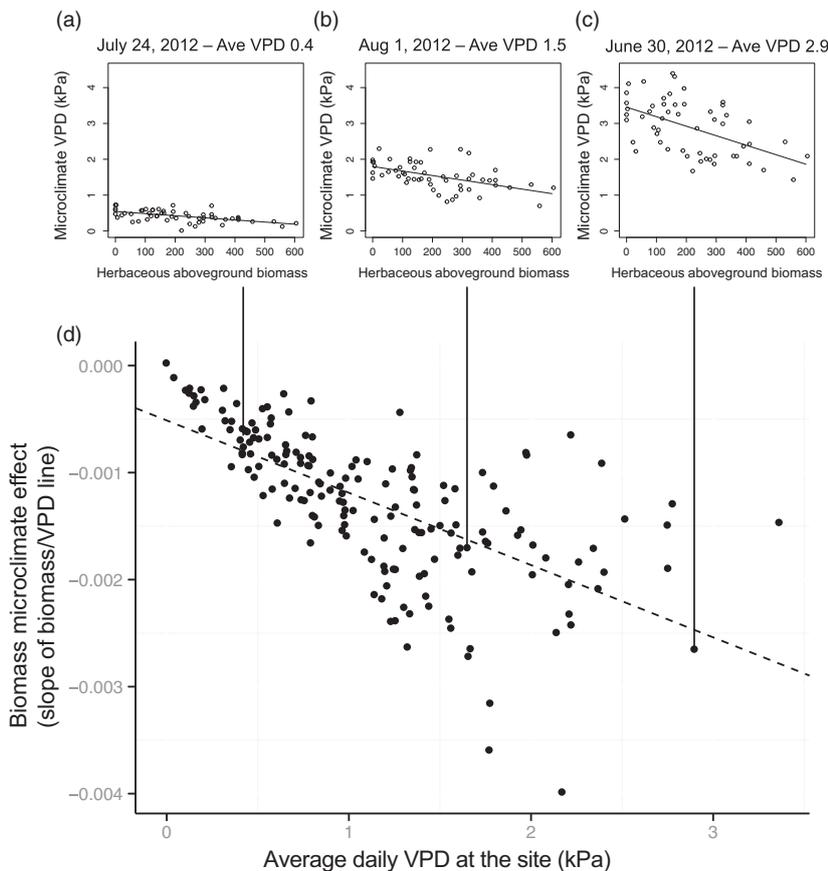


Fig. 2. We measured vapour pressure deficit in the microclimate of 55 randomly selected plots over the course of two years. We found that it was cooler in plots with more above-ground biomass (panels a–c). We also found that as average VPD at the site increased (on days when it was hotter and drier), plant biomass had an increasingly strong ameliorating effect on microclimate VPD (d). The dotted line below (d) represents the significant interaction between site-level VPD and community herbaceous biomass ($F_{1,9230} = 482.5$, $P < 0.0001^*$).

Table 2. The relationship between site conditions (vapour pressure deficit) and herbaceous biomass on oak leaf water potential. We conducted separate analyses for pre-dawn, midday and daily changes (diff). For pre-dawn leaf water potential, we compared with the 24-hr period prior to the pre-dawn measurement. For midday leaf water potential, we compared with the period between pre-dawn and midday. For the daily difference in leaf water potential, we compared with the 24-hour period prior to the midday measurement (to integrate over the whole day)

Fixed effect	Pre-dawn			Midday			Diff		
	d.f. [†]	<i>F</i>	<i>P</i>	d.f.	<i>F</i>	<i>P</i>	d.f.	<i>F</i>	<i>P</i>
Herb biomass	1, 25.2	1.50	0.23	1, 29.1	1.16	0.19	1, 29.7	0.73	0.40
Site VPD	1, 163	2.25	0.14	1, 162	30.9	<0.0001*	1, 160	16.5	<0.0001*
Herb biomass × Site VPD	1, 163	1.16	0.28	1, 162	0.01	0.94	1, 159	8.5	0.004*

[†]These analyses took into account spatial variation associated with the blocked design ('block' in the BioCON framework) and measurements taken on individuals in the same plots over time. In the linear mixed-effects model framework, denominator degrees of freedom 'float' based on the degree of variation attributed to block differences; this is why denominator degrees of freedom are different depending on the metric described in this table. Asterisks (*) indicate statistical significance.

and soil moisture declined over the course of the season (Fig. S3, Table S2).

Midday leaf water potential values were more negative on hot/dry days with a high VPD, but they did not vary overall with herbaceous biomass or species richness (Table 2, Appendix S1 – Table 3, Fig. S2). Midday leaf water potential was more negative when soil moisture was low and when plot VPD was high (Table S2, Fig. S3b,e).

The difference between pre-dawn leaf water potential (ψ_{pd}) and midday leaf water potential (ψ_{md}) reflects the change in water status of oak seedlings on a given day. By subtracting the pre-dawn leaf water potential from the leaf water potential during the time with the highest evaporative demand (midday), we can to some extent standardize (i.e. account for) differences in baseline (pre-dawn) water status and focus on daily responses. There was no main effect of

herbaceous biomass or species richness on the daily change in leaf water potential (Table 2, Appendix S1 – Table 3). There was an overall negative effect of VPD on daily change in leaf water potential, and a significant interaction between herbaceous biomass and daily VPD (Table 2, Appendix S1 – Table 3). This effect was not significantly driven by differences in oak seedling size (Fig. S4). Oak seedlings growing in higher diversity and higher biomass plots experienced net competitive effects on cool/humid days (more negative values of $\psi_{md} - \psi_{pd}$ in higher biomass plots). In contrast, plants growing in the same plots, experienced net facilitation from neighbours on hot/dry days (less negative values of $\psi_{md} - \psi_{pd}$ in higher biomass plots, Table 2, Fig. 3). The daily change in plant water status was driven by plot-level (microclimate) differences in vapour pressure deficit (Table S2, Fig. S3f).

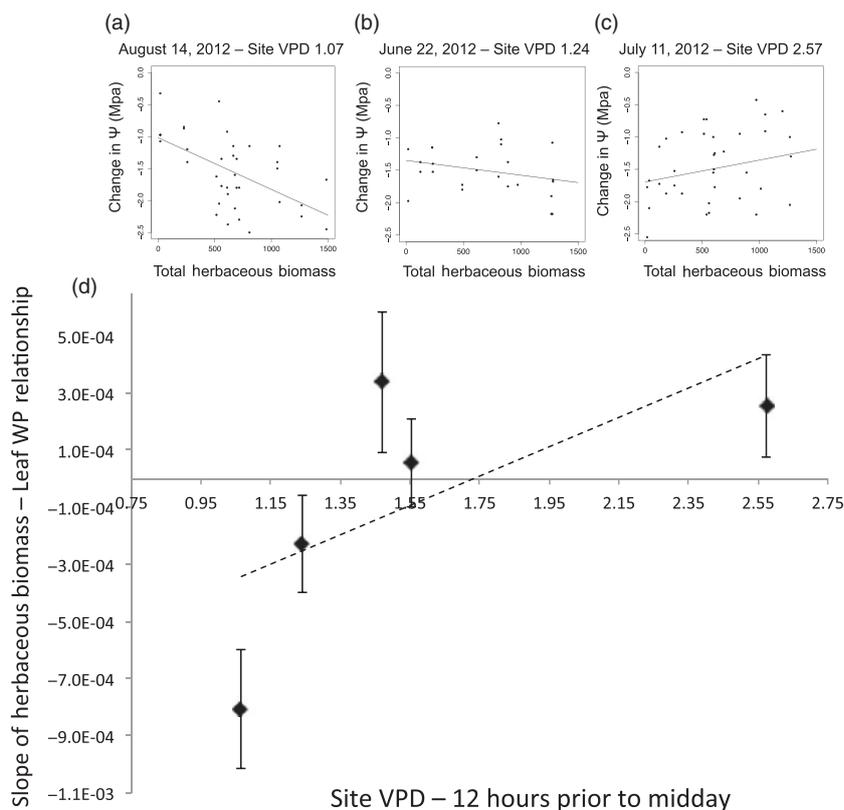


Fig. 3. We measured pre-dawn leaf water potential (ψ_{pd}) and midday leaf water potential (ψ_{md}) on five days that varied in VPD at the site. We then calculated the daily difference in leaf water potential ($\psi_{md} - \psi_{pd}$) on each of those days. We found that leaf water potential reflected net competition with higher biomass communities on cool humid days (a), but net facilitation with higher biomass communities on hot/dry days (c). The degree of this effect seemed to change nonlinearly with 24-h VPD (d). The dotted line below (d) indicates the two-way interaction between herbaceous biomass and 12-h site VPD ($F_{1,159} = 8.5$, $P = 0.004$).

Discussion

Measures of resource availability and juvenile oak seedling water status across days of differing evaporative demand showed differing strengths of facilitation and competition in this grassland experiment. Pre-dawn leaf water potential of bur oaks ranged from 0 to -1.0 MPa and was tightly linked with soil moisture. Similar to previous studies, mid-day leaf water potential ranged from -1.0 to -3.0 and was more tightly linked to vapour pressure deficit in the microclimate (Abrams & Knapp 1986). Increasing herbaceous biomass and species richness (in the seedlings neighbourhood) resulted in both greater resource limitation and in greater amelioration of environmental stress for the juvenile oaks. We found that the degree of amelioration of the microclimate became progressively stronger on hotter/drier days and this translated to the increasing importance of facilitation for plant water status. In particular, increasing herbaceous plant biomass reduced average VPD by 50% on the hottest/driest days (microclimate amelioration), compared to low biomass, low-diversity plots. Increasing plant herbaceous biomass reduced available soil moisture by as much as 50%, particularly late in the season (resource limitation). While reduced soil moisture in higher biomass, higher diversity plots likely drove increased competition for water when soil water was limiting (increasingly over the course of the season), in terms of oak seedling water status, this was outweighed by a facilitative advantage conferred in high-diversity and high-biomass plots when daily conditions (VPD) were arid.

Oak growth rates in our study were lowest in higher biomass higher diversity plots. We note that due to these differences in oak seedling size and structure, we selected oak seedlings for leaf water potential measurements that fell within a narrow size range across the biomass density gradient (a subset of the overall variation in oak sizes). We did this to reduce variation in leaf water potential measurements that were directly associated with differences in seedling size (Cavender-Bares & Bazzaz 2000). We also explored the direct effects of seedling size, leaf area and total number of leaves on daily leaf water potential measurements and found no evidence that oak size differences explained the trends in leaf water potential that we reported here (Fig. S4).

While oak growth was lowest in higher biomass and higher diversity plots in our study, we predict that the combined contribution of both competition and facilitation may underlie this trend. Decreasing oak growth with increasing community biomass does not necessarily mean that competition for resources was the only type of interaction happening between neighbours; it could mean there were just more competitive days than there were facilitative days. Specifically, competition for soil water between oak seedlings and the herbaceous plant community was strong below 12-h VPD values of ~ 1.75 kPa. Above 1.75 kPa, however, competition for soil water was outweighed by the facilitative effects of microclimate amelioration (VPD). Importantly, 2012 was one of the hottest and driest years on record. During the 2012 growing

season (May 15–September 31), there were 23 days with average daily VPD values greater than 1.75 kPa. While our current data set is necessarily limited in scope (due to the small number of days sampled), this implies that in terms of water status, oaks may have experienced predominant facilitative effects of the herbaceous community for $\sim 15\%$ of the growing season, whereas competition may have dominated for the remaining $\sim 85\%$ of the time. Though to be clear, this rough estimate does not account for competition for other limiting resources, such as nitrogen or light, which may further tip the competition/facilitation balance towards net competition.

Future work should explore these questions in terms of changes in stomatal conductance, osmotic adjustment, leaf positioning, carbon assimilation, water-use efficiency and the specific implications of these temporal variations in VPD for longer-term plant performance. Reduced VPD in the microclimate has the capacity to decrease water loss at the leaf surface (Ocheltree *et al.* 2013). While stomatal conductance was not measured in the current study, this change in VPD may help maintain higher levels of stomatal conductance and have a positive effect on overall photosynthetic rates (Ocheltree *et al.* 2013) in high-biomass plots on the hottest days. This pattern is consistent with the results we present here and would mean that an increased number of days with average VPD values greater than 1.75 kPa could potentially shift the plant–plant interaction balance in the future (He, Bertness & Altieri 2013; Stocker *et al.* 2013). It should be noted that differences in light levels between experimental plots in this system (Clark *et al.* 2012) may also affect stomatal conductance, but relative differences in light patterns among plots should not shift significantly over time and would not explain these day-to-day changes in plant interactions. Depending on local wind conditions, relative differences in CO_2 enrichment between experimental plots may also affect water-use efficiency and longer-term performance of seedlings, though CO_2 levels were not measured at the plot-level in this study. As a first estimation, these results suggest that plant performance over the course of a growing season may be influenced by simultaneously acting competition and facilitation for water and the potential for each process to mask the presence of the other (e.g. Montgomery, Reich & Palik 2010).

MICROCLIMATE AMELIORATION AND THE PLANT COMMUNITY

Our results also demonstrate the potential for a novel positive-feedback loop for microclimate amelioration. We found that plant communities with less biomass were up to 6°C hotter and had more than 2 times higher plot-level VPD on particularly hot and dry days. Conversely, on cool humid days, temperature and humidity varied little as a function of herbaceous biomass (Fig. 2). The gradient in herbaceous biomass from low- to high-diversity differed minimally across sampling dates in these comparisons, yet the effect that it had on the microclimate changed. When it is hotter and drier at the site, more herbaceous biomass may shade and intercept

irradiation that would otherwise heat the microclimate. Increased irradiation should also reduce relative humidity, driving the overall effect that we observed on vapour pressure deficit and plant performance. The greater the degree of environmental warming and drying, the greater the impact of the boundary layer provided by the herbaceous community. Furthermore, in higher biomass communities, there is more plot-level photosynthesis (indicated by greater overall productivity) in higher diversity plots (Reich *et al.* 2001a) and increased rates of plot-level transpiration; this may lead to increased evaporative cooling of the microclimate (due to a higher density of individuals and higher diversity of strategies for dealing with daily conditions). On cooler days (if cool because of cloud cover), there may be less irradiance overall, and the shading/evaporative cooling effect may not be happening. The stress-gradient hypothesis predicts that facilitation is more important for plants in environmentally severe conditions (Bertness & Callaway 1994). When environmental severity is strong, plants experience increased physiological stress, and this translates to stronger benefits of growing near neighbours. The stress-gradient hypothesis also predicts that when environmental conditions are mild, there may still be microclimate amelioration, but it may not translate to strong facilitation. We show that in terms of plant water status, as the stress-gradient hypothesis predicts, environmental amelioration may be most *important* for seedlings on hot and dry days (Fig. 3). We also show that the magnitude of the actual microclimate amelioration that is occurring is greater (Fig. 2).

SUMMARY

The majority of the past work on facilitation in plant communities has focused on documenting case studies where facilitation is dominant (deserts, tundra and salt marshes) with less focus on determining the relative roles (and masking effects) of both types of interactions among neighbouring plants. There is a large body of evidence documenting the shifting importance of competition and facilitation across systems depending on environmental severity. Specifically, the relative importance of facilitation tends to increase with increasing environmental severity at higher elevations (Callaway *et al.* 2002), in deserts (Holzapfel & Mahall 1999) and marine systems (Peterson & Heck 2001). Fewer studies have examined temporal variation in the drivers of such changes (but see Callaway 1992; Greenlee & Callaway 1996; Baumeister & Callaway 2006; Semchenko *et al.* 2011). Though predicted by theory (Bruno, Stachowicz & Bertness 2003), we know of no previous studies that have documented the shifting balance of competition and facilitation from 1 day to the next. Here, we demonstrate threshold environmental conditions (community biomass and diversity, soil moisture and vapour pressure deficit) that tip the balance temporally between competition and facilitation for plant water status and therefore demonstrate the underlying presence of both. Because both competition and facilitation are likely ubiquitous in many if not most plant communities, tests of competition may be missing underlying facilitation (e.g. 15% of days in our experiment) due to stronger overall

competitive effects (e.g. 85% of days). Conversely, in more severe environments, the role of competition may be overshadowed by facilitation, possibly leading to the flawed conclusion that competition is not important in severe environments (Bruno, Stachowicz & Bertness 2003). If taken to the next level, the mechanisms behind these day-to-day results may even scale from days to seasons to interannual variations. Only through careful examination of both the competitive and facilitative interactions between plants, can we understand plant community dynamics and make accurate predictions about how plant communities may change in the future.

Acknowledgements

This research was supported by the Department of Energy Program for Ecological Research Grant DE-FG02-96ER62291, the National Science Foundation Long-term Ecological Research Grant DEB-1234162, the NSF Long-term Research in Environmental Biology DEB-0716587 and DEB-1242531 and Ecosystem Sciences (NSF DEB-1120064) Programs, the University of Minnesota, the UWM Department of Biological Sciences and the NSF Graduate Research Fellowship Program. Special thanks to Tali Lee, Susan Barrott and Kally Worm for assistance in the execution and design of this experiment. We thank Joe Strini and Hannah Kruse for field assistance during the collection of leaf water potential data. We also thank Katie Barry and Laura Williams for extensive comments on previous versions of this manuscript. All authors confirm that there is no conflict of interest involved in the publication of this study.

Data accessibility

Oak leaf water potential, relative growth rates and plot VPD data: Cedar Creek Ecosystem Science Reserve Data Catalogue data set ID #adue141. Herbaceous community biomass data: Cedar Creek Ecosystem Science Reserve Data Catalogue data set ID #ple141.

Author contributions

AW designed the oak water potential study with assistance from SS and PR, AW collected the data pertaining to oak seedling growth and physiology, and PR conducted the BioCON experiment and organized its data acquisition, AW analysed the data and wrote the first draft of the manuscript, and all authors contributed substantially to revisions.

References

- Abrams, M.D. & Knapp, A.K. (1986) Seasonal water relations of three gallery forest hardwood species in northeast Kansas. *Forest science*, **32**, 687–696.
- Adair, E.C., Reich, P.B., Trost, J.J. & Hobbie, S.E. (2011) Elevated CO₂ stimulates grassland soil respiration by increasing carbon inputs rather than by enhancing soil moisture. *Global Change Biology*, **17**, 3546–3563.
- Anderson, D.B. (1936) Relative humidity or vapor pressure deficit. *Ecology*, **17**, 277–282.
- Armas, C., Rodríguez-Echeverría, S. & Pugnaire, F.I. (2011) A field test of the stress-gradient hypothesis along an aridity gradient. *Journal of Vegetation Science*, **22**, 818–827.
- Baumeister, D. & Callaway, R. (2006) Facilitation by *Pinus flexilis* during succession: a hierarchy of mechanisms benefits other plant species. *Ecology*, **87**, 1816–1830.
- Bertness, M. & Callaway, R. (1994) Positive interactions in communities. *Trends in Ecology & Evolution*, **9**, 191–193.
- Bond, W.J., Woodward, F.I. & Midgley, G.F. (2005) The global distribution of ecosystems in a world without fire. *New phytologist*, **165**, 525–537.
- Brooker, R.W., Maestre, F.T., Callaway, R.M., Lortie, C.L., Cavieres, L.A., Kunstler, G. *et al.* (2008) Facilitation in plant communities: the past, the present, and the future. *Journal of Ecology*, **96**, 18–34.
- Brown, K.W., Jordan, W.R. & Thomas, J.C. (1976) Water stress induced alterations of the stomatal response to decreases in leaf water potential. *Physiologia Plantarum*, **37**, 1–5.

- Bruno, J., Stachowicz, J. & Bertness, M. (2003) Inclusion of facilitation into ecological theory. *Trends in Ecology & Evolution*, **18**, 119–125.
- Callaway, R. (1992) Effect of shrubs on recruitment of *Quercus douglasii* and *Quercus lobata* in California. *Ecology*, **73**, 2118–2128.
- Callaway, R.M., Brooker, R.W., Choler, P., Kikvidze, Z., Lortie, C.J., Michalet, R., Paolini, L., Pugnaire, F.I., Newingham, B. & Aschehoug, E.T. (2002) Positive interactions among alpine plants increase with stress. *Nature*, **417**, 844–848.
- Cavender-Bares, J. & Bazzaz, F.A. (2000) Changes in drought response strategies with ontogeny in *Quercus rubra*: implications for scaling from seedlings to mature trees. *Oecologia*, **124**, 8–18.
- Clark, C.M., Flynn, D.F.B., Butterfield, B.J. & Reich, P.B. (2012) Testing the Link between Functional Diversity and Ecosystem Functioning in a Minnesota Grassland Experiment (ed. J.F. Cahill). *PLoS ONE*, **7**, e52821.
- Cuesta, B., Villar-Salvador, P., Puértolas, J., Rey Benayas, J.M. & Michalet, R. (2010) Facilitation of *Quercus ilex* in Mediterranean shrubland is explained by both direct and indirect interactions mediated by herbs. *Journal of Ecology*, **98**, 687–696.
- Davis, M., Wrage, K., Reich, P., Tjoelker, M., Schaeffer, T. & Muermann, C. (1999) Survival, growth, and photosynthesis of tree seedlings competing with herbaceous vegetation along a water-light-nitrogen gradient. *Plant Ecology*, **145**, 341–350.
- Ehleringer, J.R. (1984) Intraspecific competitive effects on water relations, growth and reproduction in *Encelia farinosa*. *Oecologia*, **63**, 153–158.
- Fargione, J.E. & Tilman, D. (2005) Diversity decreases invasion via both sampling and complementarity effects. *Ecology Letters*, **8**, 604–611.
- Gordon, D.R., Menke, J.M. & Rice, K.J. (1989) Competition for soil water between annual plants and blue oak (*Quercus douglasii*) seedlings. *Oecologia*, **79**, 533–541.
- Greenlee, J. & Callaway, R. (1996) Abiotic stress and the relative importance of interference and facilitation in montane bunchgrass communities in western Montana. *American Naturalist*, **148**, 386–396.
- Hardin, G. (1960) The competitive exclusion principle. *Science*, **131**, 1292–1297.
- He, Q., Bertness, M.D. & Altieri, A.H. (2013) Global shifts towards positive species interactions with increasing environmental stress (ed. M Vila). *Ecology Letters*, **16**, 695–706.
- Holzappel, C. & Mahall, B. (1999) Bidirectional facilitation and interference between shrubs and annuals in the Mojave Desert. *Ecology*, **80**, 1747–1761.
- Jia, X., Dai, X.-F., Shen, Z.-X., Zhang, J.-Y. & Wang, G.-X. (2010) Facilitation can maintain clustered spatial pattern of plant populations during density-dependent mortality: insights from a zone-of-influence model. *Oikos*, **120**, 472–480.
- Landero, J.P.C. & Valiente-Banuet, A. (2010) Species-specificity of nurse plants for the establishment, survivorship, and growth of a columnar cactus. *American Journal of Botany*, **97**, 1289–1295.
- Montgomery, R.A., Reich, P.B. & Palik, B.J. (2010) Untangling positive and negative biotic interactions: views from above and below ground in a forest ecosystem. *Ecology*, **91**, 3641–3655.
- NOAA National Climatic Data Center, State of the Climate: National Overview for Annual (2012) Available from <http://www.ncdc.noaa.gov/sotc/national/2012/13>. [accessed 14 March 2015].
- Ocheltree, T.W., Nippert, J.B. & Prasad, P.V.V. (2013) Stomatal Responses to Changes in Vapor Pressure Deficit Reflect Tissue-Specific Differences in Hydraulic Conductance. *Plant Cell and Environment*, **37**, 132–139.
- Peterson, B.J. & Heck, K.L. Jr (2001) Positive interactions between suspension-feeding bivalves and seagrass—a facultative mutualism. *Marine Ecology Progress Series*, **213**, 143–155.
- Reich, P., Knops, J., Tilman, D., Craine, J., Ellsworth, D., Tjoelker, M., Lee, T., Wedin, D., Naeem, S. & Bahaeddin, D. (2001a) Plant diversity enhances ecosystem responses to elevated CO₂ and nitrogen deposition. *Nature*, **410**, 809–810.
- Reich, P., Tilman, D., Craine, J., Ellsworth, D., Tjoelker, M., Knops, J., Wedin, D., Naeem, S., Bahaeddin, D. & Goth, J. (2001b) Do species and functional groups differ in acquisition and use of C, N and water under varying atmospheric CO₂ and N availability regimes? A field test with 16 grassland species. *New Phytologist*, **150**, 435–448.
- Reich, P.B., Tilman, D., Isbell, F., Mueller, K., Hobbie, S.E., Flynn, D.F.B. & Eisenhauer, N. (2012) Impacts of biodiversity loss escalate through time as redundancy fades. *Science*, **336**, 589–592.
- Semchenko, M., Lepik, M., Götzenberger, L. & Zobel, K. (2011) Positive effect of shade on plant growth: amelioration of stress or active regulation of growth rate? *Journal of Ecology*, **100**, 459–466.
- Stachowicz, J.J. (2001) Mutualism, Facilitation, and the Structure of Ecological Communities: positive interactions play a critical, but underappreciated, role in ecological communities by reducing physical or biotic stresses in existing habitats and by creating new habitats on which many species depend. *BioScience*, **51**, 235–246.
- Stocker, T.F., Qin, D., Plattner, G.K., Alexander, L.V., Allen, S.K., Bindoff, N.L. et al. (2013) Technical Summary. In: *Climate Change 2013: the Physical Science Basis. Contribution of Working Group I to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change* (eds T.F. Stocker, D. Qin, G.K. Plattner, M. Tignor, S.K. Allen, J. Boschung, A. Nauels, Y. Xia, V. Bex & P.M. Midgley). Cambridge University Press, Cambridge, United Kingdom and New York, NY, USA.
- Tilman, D. (1977) Resource competition between plankton algae: an experimental and theoretical approach. *Ecology*, **58**, 338–348.
- Tilman, D., Wedin, D. & Knops, J. (1996) Productivity and sustainability influenced by biodiversity in grassland ecosystems. *Nature*, **379**, 718–720.
- Valladares, F. & Pearcy, R. (1997) Interactions between water stress, sun-shade acclimation, heat tolerance and photoinhibition in the sclerophyll *Heteromeles arbutifolia*. *Plant Cell and Environment*, **20**, 25–36.
- Wright, A.J., Schnitzer, S.A. & Reich, P.B. (2014) Living close to your neighbors – the importance of both competition and facilitation in plant communities. *Ecology*, **95**, 2213–2223.
- Wright, A., Schnitzer, S.A., Dickie, I.A., Gunderson, A.R., Pinter, G.A., Mangan, S.A. & Reich, P.B. (2013) Complex facilitation and competition in a temperate grassland: loss of plant diversity and elevated CO₂ have divergent and opposite effects on oak establishment. *Oecologia*, **171**, 449–458.

Received 8 December 2014; accepted 9 March 2015

Handling Editor: Robert Jones

Supporting Information

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

Figure S1. Soil moisture in all plots according to date of measurement and environmental conditions.

Figure S2. Predawn, midday, and daily diff in leaf water potential measurements on all days of the study.

Figure S3. The effects of plot-level soil moisture and microclimate (VPD) on predawn, midday, and daily diff in leaf water potential.

Figure S4. The effect of oak seedling size on the daily change in leaf water potential experienced by oak seedlings across the community biomass gradient.

Table S1. Effects of DOY and environmental conditions on soil moisture (%).

Table S2. The effects of plot-level soil moisture and microclimate (VPD) on predawn, midday, and daily diff in leaf water potential.

Appendix S1. Analyses repeated using species richness of plots (instead of herbaceous biomass).