Adaptation to elevated CO$_2$ in different biodiversity contexts

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In the absence of migration, species persistence depends on adaptation to a changing environment, but whether and how adaptation to global change is altered by community diversity is not understood. Community diversity may prevent, enhance or alter how species adapt to changing conditions by influencing population sizes, genetic diversity and/or the fitness landscape experienced by focal species. We tested the impact of community diversity on adaptation by performing a reciprocal transplant experiment on grasses that evolved for 14 years under ambient and elevated CO$_2$, in communities of low or high species richness. Using biomass as a fitness proxy, we find evidence for local adaptation to elevated CO$_2$, but only for plants assayed in a community of similar diversity to the one experienced during the period of selection. Our results indicate that the biological community shapes the very nature of the fitness landscape within which species evolve in response to elevated CO$_2$. 

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Species do not evolve in isolation but within a community of interacting species. While some evidence exists for the impact of predator-prey or host-parasite interactions on adaptive evolution, we lack experimental data on the impact of competition on adaptation in natural systems. Laboratory and mesocosm studies have found contrasting results for how competition influences adaptation. Competition can inhibit adaptation, as found in algal cultures (Chlamydomonas reinhardtii) evolving to elevated CO₂ (ref. 5). Similarly, adaptive diversification to habitat heterogeneity in Pseudomonas fluorescens was prevented in the presence of interspecific competitors, as these competitors exclude intraspecific variants. Competition can also alter the nature of selection. For instance, increased water temperature caused the zooplankton Daphnia magna and D. pulex to evolve faster growth in the absence of competition but to evolve a larger size at maturity in the presence of competitors and predators. Similarly, bacterial species adapted differently to a novel environment when grown alone or with other bacteria species. In plants, the presence, composition, and diversity of competing species show tremendous spatial variation, can have a major impact on individual performance, and thus might have an important influence on species’ adaptation to environmental change. Yet, how biotic community context alters how species adapt to environmental change in natural field settings remains entirely unknown.

To address this knowledge gap, herein we report on an investigation of the impact of prairie grassland communities on the evolutionary response to elevated CO₂ over 14 years in the Biodiversity Carbon dioxide and Nitrogen experiment (BioCON) at the Cedar Creek Ecosystem Sciences Reserve (Minnesota, USA). To determine how the surrounding biological community influences a species’ ability to evolve in response to abiotic change, we focused on very different community structures: monoculture versus high diversity. By focusing on the presence or absence of interspecific competitors, we increased our power to detect the impacts of the surrounding species diversity on evolution. We tested four possible scenarios by which species diversity might affect the evolutionary responses of a focal species to abiotic environmental change.

The first scenario is that species diversity has no effect on adaptation to abiotic environmental change (Fig. 1a,b). If selective pressures exerted by changing abiotic conditions overwhelm those from the biotic community, species diversity should have no impact on local adaptation to abiotic change. Statistically, the response to selection (fitness of plants evolved under elevated CO₂ (eCO₂) minus fitness of plants evolved under ambient CO₂ (aCO₂)) should be predicted only by the change in CO₂ environment (ΔCO₂), regardless of species diversity (Fig. 1a,b).

The second scenario is that species diversity constrains adaptation to abiotic environmental change (Fig. 1c,d). When grown with more species, local adaptation might be reduced because competition for space and resources results in smaller effective population sizes of each competing species, reducing standing genetic variation and the rate at which new mutations arise. In addition, with more species in a community one species may, by chance, possess traits that pre-adapt it to the new niche(s) created by the changing environment. This pre-adapted species will increase in abundance, creating more competition and resulting in a further decline in abundance, and therefore ability to adapt, in the other species. Under this scenario, local adaptation to elevated CO₂ should be more evident for plants that experienced selection in a species-poor community (Fig. 1c) than in a species-rich community (Fig. 1d), regardless of the diversity of the community into which the plants were transplanted (the ‘assay’ community). Statistically, the response to selection should be predicted by a three-way interaction between the CO₂ selection environment (CO₂sel), the change in CO₂ environment (ΔCO₂), and diversity of the selection environment (divsel).

The third scenario is that species diversity promotes adaptation to abiotic environmental change (Fig. 1e,f). A more homogeneous environment caused by low species richness might select for and maintain fewer genotypes, reducing genetic diversity. Likewise, high species richness might increase environmental heterogeneity, thereby maintaining greater genetic variation and adaptive potential. This scenario predicts that a focal species should adapt faster to abiotic change (for example, eCO₂) when selection is experienced in a species-rich community. As with the second scenario, a significant CO₂sel × ΔCO₂ × divsel interaction should support this scenario, except with the opposite relationship to diversity (Fig. 1e,f).

The forth scenario is that species diversity changes the fitness landscape (Fig. 1g,h). The biological community may act to modify the selection environment created by eCO₂ thereby changing the shape of the fitness landscape. That is, the surrounding community may act like a prism, transforming an
applied selective pressure into the selective pressure actually experienced by a focal species\textsuperscript{2,7,19}. For example, increased CO\textsubscript{2} might select for faster growth, resulting in selection for more efficient nitrogen use in a species-poor community but not in a species-rich community that includes nitrogen-fixing plants\textsuperscript{20}. As another example, belowground microbial biomass has been found to decline with eCO\textsubscript{2} in species-poor communities but to rise in species-rich communities\textsuperscript{21}, thereby potentially altering the supply and abundance of various nutrients to plants\textsuperscript{22} in different biotic and abiotic contexts to which plants must locally adapt. Whatever the mechanism, this scenario predicts local adaptation to eCO\textsubscript{2} when fitness is assessed in a community with similar diversity as the community in which selection occurred (Fig. 1g,h). Statistically, this scenario should be supported by a significant interaction between CO\textsubscript{2} selection environment (CO\textsubscript{2sel}), change in CO\textsubscript{2} environment (ΔCO\textsubscript{2}), and change in diversity (Adv).

We tested these predictions using a reciprocal transplant experiment (Fig. 2) involving \textit{Poa pratensis} (Kentucky bluegrass), a species widespread and abundant across several continents, and one of the more common species in BioCON\textsuperscript{23}. We collected seeds from plots that had been exposed for 14 years to ambient or elevated (ambient + 180 p.p.m.) concentrations of CO\textsubscript{2} in species-poor (monoculture) or species-rich (16 species) grassland communities\textsuperscript{12}. We transplanted individuals with all four ‘histories’ into all four of these treatment combinations. Consistent with the fourth scenario, we find that the biological community alters the fitness landscape in elevated CO\textsubscript{2}, so that local adaptation is observed primarily when species are grown in a community similar to the one in which they were previously selected.

Results

Results for biomass production. We found that \textit{P. pratensis} locally adapted to the CO\textsubscript{2} environment, but only when the diversity of the community was the same in the past ‘selection’ and current ‘assay’ environment. That is, the term ΔCO\textsubscript{2} was statistically significant for aboveground (F\textsubscript{1, 306.6} = 7.8, \textit{P} < 0.006) and total biomass (F\textsubscript{1, 302.9} = 4.1, \textit{P} = 0.04) (Supplementary Table 1) when plants were selected and assayed in the same diversity treatment (species-poor to species-poor or species-rich to species-rich) (Supplementary Fig. 1). However, if adaptation to CO\textsubscript{2} was assessed by averaging the results from both species-rich and species-poor assay plots, local adaptation was not statistically detectable for any measure (Supplementary Tables 2 and 3; Supplementary Fig. 2). We thus find that the local adaptation does not depend solely on the community context during past selection but also depends on the current assay environment, leading us to reject scenarios one to three (Fig. 1).

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**Figure 2 | Steps involved in the transplant experiment.** (a) \textit{P. pratensis} seeds were collected from BioCON plots from different CO\textsubscript{2} and diversity treatments. (b) Seeds were germinated in a common garden greenhouse environment to reduce maternal environmental effects. Values in brackets indicate the total number of germinated seeds per BioCON selection environment (departures from 64 due to failed germination or additional sampling are specified in the data table). (c) Four daughter ramets were sampled per germinated seed (offspring) in the greenhouse and transported back to BioCON for the transplant experiment. (d) One daughter ramet per offspring was placed into each of the assay diversity and CO\textsubscript{2} environments (see text for additional details). 1 sp., \textit{P. pratensis} monoculture BioCON plots; 16 spp., 16 species BioCON plots; species-poor, \textit{P. pratensis} dominated species-poor plots; species-rich, species-rich plots (note that the latter two plots types describe the assay plots; Supplementary Methods 1).
Local adaptation was only seen when we took into account both the diversity of the selection environment and the assay environment. This indicates support for the fourth scenario in which species diversity alters the fitness landscape (Fig. 1g,h): plants exhibited adaptation to eCO2, but only when assayed in the community context in which selection occurred (Fig. 3a–c). For example, the response of aboveground biomass to selection in eCO2 (Fig. 3a) was positive in species-poor environments (left) but chiefly when assayed in species-poor environments (dark triangles). Similarly, plants from species-rich environments showed adaptation to eCO2 but only when assayed in species-rich environments (circles on the right). As a result, when performing a full statistical analysis including community diversity in both selection and assay environments, there were significant three-way CO2sel × ΔCO2 × Δdiv interactions for aboveground (F1,689.1 = 5.8, P = 0.016), belowground (F1,690.3 = 4.2, P = 0.041) and total biomass (F1,684.3 = 4.2, P = 0.039) (Supplementary Table 3), with increased adaptation to eCO2 when the community context remained the same between selection and assay environments (Fig. 3a–c). These significant interactions were consistent with plants demonstrating a ‘home’ plot advantage when assayed in a biotic and abiotic environment similar to the one in which they experienced selection (Supplementary Fig. 3a–c).

Results for survival and inflorescence production. The high survival of individuals across treatments (>80% survival) and the lack of flowering within species-rich plots (only 9% of individuals produced inflorescences) reduced our power to analyse these fitness components (Supplementary Fig. 3d). Although not significant, the direction of the results from an Aster analysis24 of survival and inflorescence production were consistent with the fitness landscape scenario (Fig. 3d; Supplementary Table 5).

Analysis with selection and assay environments. As an alternative statistical approach, we also analysed the data by treating the selection and assay environments (CO2ass and divass) (not the change in environments) as fixed factors (Supplementary Tables 4 and 6). This approach has reduced statistical power because the fourth scenario, the fitness landscape scenario, must be tested via a four-way interaction (CO2sel × CO2ass × divsel × divass), while the second and third scenarios are tested via three-way interactions (CO2sel × CO2ass × divsel). Nevertheless, these results were also consistent with the fitness landscape scenario (Supplementary Tables 4 and 6). Analysing the data in this alternative way aided in teasing apart immediate responses to the environment (that is, ‘plastic’ responses) from evolutionary changes. Indeed, we also detected strong plastic responses, with greater biomass for plants assayed in species-poor plots and for plants assayed in eCO2 (Supplementary Tables 4 and 6; Supplementary Note 1).

Discussion

Our study contrasted four scenarios for how the diversity of the surrounding neighbourhood community could impact evolutionary change in a focal species. Overall our results were most consistent with the scenario in which species diversity alters the
fitness landscape (Fig. 1g,h) and indicate that while adaptation to eCO2 confers a performance advantage in both community contexts, the advantage does not transfer directly across community contexts. Studies examining the effect of eCO2 on plants and communities have found that eCO2 typically accelerates plant growth, alters plant tissue chemistry42,53, increases plant biomass and reduces evapotranspiration53. These changes result in increased soil water content52, altered belowground microbial diversity28 and modified nutrient cycling. The nature of these changes, however, is likely to depend on the surrounding community. Indeed, increasing plant diversity also reduces soil moisture52, increases microbial community diversity51 and increases nutrient cycling52. These changes are expected to interact21 to shape the selective environment in which a plant grows. Since competition exerts strong selective pressures, we suggest that it is the combined effect of altered abiotic environment and changed competitive interactions shaping the fitness landscape that drive our result. A similar perspective is that, as mutations arise, their pleiotropic effects generate different selective trade-offs depending on the surrounding community, altering which mutations can spread (which is another way of saying that the community shapes the fitness landscape experienced by new mutations).

Our finding that adaptation to eCO2 does not transfer across community contexts may have important implications for understanding how species will respond at both small and large scales to rising global CO2 levels caused by fossil-fuel emissions. At small scales the composition of communities can vary tremendously due to aspect, slope, soil, and so on, and the evolutionary response of a metapopulation to a common selective pressure (for example, changing CO2) might be experienced in different ways at different sites, with the local community altering the selection experienced. With high gene flow between populations, adaptation to rising CO2 levels may be hampered as a consequence of maladaptation to the biotic environment. At larger scales, species are likely to shift their range boundaries in response to climate warming and therefore to encounter novel community contexts30,31. If range shifts alter the biotic environment, previous adaptations to the abiotic environment (for example, elevated CO2 or temperature) may no longer improve fitness when in a different community context.

To our knowledge, our study is the first experiment conducted in a natural field setting to test whether adaptation to an abiotic change in a macro-organism is impacted by community context. In plants, several studies have investigated the interaction between abiotic and biotic conditions on adaptation, but they either were not conducted in a natural field setting (that is, a pot experiment with a single competitor species6) or they focused on immediate phenotypic responses12–35. By performing a reciprocal transplant experiment of plants propagated under different biotic and abiotic conditions for 14 years, we can distinguish plastic from evolutionary responses in different community contexts. An additional compelling example of local adaptation at both a small spatial and temporal scale. Adaptation was observed in a perennial species in just 14 years to an important global change driver (eCO2) in a manner that depends on the community context at a microgeographic scale. This work thus contributes to the growing number of examples from field populations of local adaptation at a microgeographic scale56,57 or microgeographic58,59 scales.

There are some limitations to our study. First, the response to eCO2 was relatively modest, and responses to stronger agents of selection could be less or more contingent on biotic context. Second, there was limited scope for replication given the design of the original BioCON experiment; in particular, only two monoculture plots existed for P. pratensis under each CO2 treatment. Another consideration is that although P. pratensis reproduces predominantly asexually through rhizomatous growth or apomixis30,59, making selection within and among clones a likely mechanism of adaptation, on-going gene flow, through seed or pollen dispersal, from other BioCON plots or the surrounding prairie community (including P. pratensis) cannot be discounted (although this would reduce the likelihood of observing the selection responses that we did). We should also emphasize that we have compared only two types of communities, species-rich and species-poor. We do not know the extent to which our results are driven by diversity, per se, versus simply the presence of particular other species. However, an analysis of our P. pratensis aboveground biomass data versus percent cover of other species in the species-rich plots did not indicate that one species was driving our results (Supplementary Note 2; Supplementary Tables 7 and 8). Moreover, earlier studies in BioCON of effects of species-poor versus species-rich neighbourhoods on focal species show that resource competition and environmental stress amelioration (both via higher biomass related effects) are both enhanced by higher diversity41,42. Such effects would have been absent at work in the current study. Finally, as with most studies of this nature, we cannot be certain that maternal effects were completely eliminated by clonal growth for 6 months in a common greenhouse environment. Thus, it remains possible that some of the evolutionary responses we have documented are in fact transgenerational maternal effects. Importantly, these limitations point largely to reasons one might expect not to find adaptation to CO2 in these plots over the past 14 years, but we did indeed find evidence for adaptation that fell clearly in line with one or more priori prediction (and not the others).

Overall, our results indicate that the evolutionary response of a plant to elevated CO2 is manifested primarily when grown in the same type of biological community in which it evolved (species-rich or species-poor). This pattern supports the view that the selection imposed by a shift in the abiotic environment is experienced through the prism of surrounding species, altering the form of selection actually experienced by a focal species, consistent with the fitness landscape scenario. How often the biological community acts to change the selection experienced in altered environments can only be determined by additional empirical tests of these scenarios.

Methods

Sampling design. BioCON was initiated in 1997 and consists of six, 20 m-diameter circular rings, each with ~ 66 x 2 m plant communities (that is, plots)22. In three randomly selected rings, atmospheric CO2 is elevated by ~ 180 ppm above ambient, using free air carbon dioxide enrichment (FACE) technology, the other three rings are maintained at ambient conditions. In each plot, 1, 4, 9 or 16 grassland species were initially seeded (12 g m-2 of seed partitioned equally among all species planted in a plot). The 16 species planted into BioCON are four C4 grasses (Andropogon gerardii, Bouteloua gracilis, Schizachyrium scoparium, Sorghastrum nutans), four C3 grasses (Agropyron repens, Bromus sterilis, Lolium perenne, Poa pratensis), four nitrogen-fixing legumes (Amorpha canescens, Lespedeza capitata, Lupinus perennis, Petalostemum villosum), and four non-nitrogen-fixing forbs (Achillea millefolium, Anemone cylindrica, Asclepias syriaca, Solidago rigida). The plots are maintained through regular weeding. Although BioCON also manipulated nitrogen (ambient and elevated), only plots exposed to ambient nitrogen were included in the current study.

P. pratensis (Poaceae), the focal species in our study, is a perennial, facultatively apomictic grass. It reproduces largely via asexually produced seeds60,61 or via tillers (ramets). Although native to Europe, P. pratensis is extensively naturalized in North America due to its use as a fodder and turf grass43.

To assess the impact of community diversity on adaptation to CO2, we conducted a reciprocal transplant experiment, with an initial six-month period of plant growth and vegetative reproduction in a common greenhouse environment to reduce maternal effects. In June 2011, we collected P. pratensis seeds from species-poor (monoculture) and species-rich (16 species) plots in aCO2 and eCO2 conditions (Fig. 2a). Four of the six BioCON rings contain a single P. pratensis monoculture plot (two rings with aCO2 and two with eCO2). Within these four

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for root density (biomass per soil volume), rather than total root biomass, but for ease of reference we refer to this as ‘belowground biomass’. After the roots were extracted, they were washed free of soil and any roots from a different species were removed. The roots were then dried and weighed.

Previous work has shown that total vegetative weight (stems, leaves and roots) and reproductive weight (fruits, surrounding glumes and rachis) are highly correlated in *P. pratensis*. Similarly, amongst the plants that flowered, we find that the combined weight of the reproductive and root biomass is correlated with both aboveground biomass ($P < 0.0001$, adjusted $r^2 = 0.481$) and total biomass ($P < 0.0001$, adjusted $r^2 = 0.428$). We thus consider biomass to be a rough proxy for fitness.

**Statistical analysis.** Analysis of biomass data. We first tested for local adaptation to CO2 by holding the diversity environment constant; that is, by only including plants that were selected and assayed in the same diversity environment. Separate linear mixed effects models were performed on the logarithm (to meet assumptions of normality) of aboveground, belowground and total biomass. As fixed factors, we included all single and two-way interactions between the previous CO2 selection environment ($CO2_{sel}$), the change in CO2 environment ($ACO2$) relative to the CO2 selection environment, and the general diversity environment (that is, either species-rich to species-rich or species-poor to species-poor). All models also included the following random effects: selection plot, family (mother’s ID) nested within selection plot, assay ring and assay plot nested within assay ring. There was no statistical difference between the rings in which the plants were selected and much of this variance is likely absorbed by the term selection plot; thus we excluded selection ring from all models. We implemented all final models using the restricted maximum-likelihood method (REML) to estimate variance components. The signicance of all fixed effects were evaluated using type III estimable functions, and denominator degrees of freedom were determined using Satterthwaite’s approximation.

Testing scenario one: To determine whether local adaptation occurred regardless of diversity environment we performed separate linear mixed effects models on the logarithm of aboveground, belowground and total biomass while averaging over diversity environment. Analyses were performed as described above except that the performance of plants was averaged over the diversity environment (div$^{m}$), while selection diversity environment (div$^{sel}$) was included in the model instead of the general diversity environment.

Testing scenarios two, three and four: We performed separate linear mixed effects models on the logarithm of aboveground, belowground and total biomass. As fixed factors, we included the previous CO2 selection environment ($CO2_{p}$) and diversity selection environment (div$^{sel}$), which together define the BioCON plot from which each individual’s mother was sampled. We also included fixed-factor terms indicating whether the plant was exposed to a change in CO2, (ACO2), or in diversity (Adv), relative to the plots of their mothers. As an alternative statistical approach, we also analysed the data by testing the selective and assay (current environments) (CO2$^{p}$ and div$^{ass}$) (not the change in environments) as fixed factors. All models also included the following random effects: selection plot, family (mother’s ID) nested within selection plot, assay ring and assay plot nested within assay ring into which ramets were transplanted. For the reasons outlined above selection ring was excluded from all models.

To test for our hypothesized interactions we ran each full model and eliminated non-significant terms using likelihood ratio tests until the model contained the CO2$^{p}$, CO2$^{ass}$, diversity (div$^{ass}$), diversity environment (div$^{m}$) and their interactions. A model was considered to have converged when the inspection of the fitted and residual values, all models were found to meet assumptions of normality and homoscedasticity. In addition, a model was considered to have converged when the analysis was carried out in R version 3.0.2 (ref. 48) using the lmer and lmerTest packages. The analysis of biomass was carried out on all individuals that survived to the end of the experiment. However, data from 49 individuals that were growing in two plots that burned in a fire in May 2013 (three quarters of the individuals in one species-poor and one species-rich plot) were excluded from analysis, since these individuals were not relocated in spring 2013 were excluded. Thus in total, data for 765 individuals were included for the biomass analyses. In addition, 11 belowground biomass samples were excluded from the analysis as these cores were either burned in at an angle that missed most of the root mass during the core extraction process or half the soil fell out of the core during extraction. We implemented all final models using the restricted maximum-likelihood method to estimate variance components. The significance of all fixed effects was evaluated using type III estimable functions, and denominator degrees of freedom were determined using Satterthwaite’s approximation.

Analysis of survival and inflorescence production. To assess the importance of CO2$^{p}$, ACO2, div$^{ass}$ and Adv on survival and inflorescence production we used aster models with random effects implemented in R. Aster models facilitate the analysis of multiple life history stages as they can analyse survival and reproduction jointly. Furthermore, different life history traits can be modelled with different probability distributions, and account for the fact that later components of fitness (for example, flowering) depend on earlier components of fitness (for example, survival)24. In our Aster model we included (1) survival until the time of...
influence of competition on plant species richness. Ecol. Monogr. 60, 421–435 (2000).

28. Eisenhauer, N., Cesarz, S., Koller, R., Worm, K. & Reich, P. B. Global change belowground: impacts of elevated CO2, nitrogen, and summer drought on soil food webs and biodiversity. Glob. Change Biol. 18, 435–447 (2012).

29. Reich, P. B. et al. Impacts of biodiversity loss escalate through time owing to redundancy fading. Science 336, 589–592 (2012).

30. Parmesan, C. & Yohe, G. A globally coherent fingerprint of climate change impacts across natural systems. Nature 421, 37–42 (2003).

31. Walliser, G-.R. et al. Ecological responses to recent climate change. Nature 416, 389–395 (2002).

32. Bazzaz, F. A. & Garbutt, K. The response of annuals in competitive neighbourhoods: effects of elevated CO2. Ecology 69, 937–946 (1988).

33. Reznick, D. N. & Ghalambor, C. K. The population ecology of contemporary adaptations: what empirical studies reveal about the conditions that promote adaptive evolution. Genetica 112, 183–198 (2001).

34. Richardson, J. L., Urban, M. C., Bolnick, D. I. & Skelly, D. K. Microgeographic adaptation and the spatial scale of evolution. Trends Ecol. Evol. 29, 165–176 (2014).

35. Linhart, Y. B. & Grant, M. C. Evolutionary significance of local genetic differentiation in plants. Annu. Rev. Ecol. Syst. 27, 237–277 (1996).

36. Hendry, A. P. & Kinnison, M. T. Perspective: the pace of modern life: measuring rates of contemporary microevolution. Evolution 53, 1637–1653 (1999).

37. Reznick, D. N. & Ghalmador, C. K. The population ecology of contemporary adaptations: what empirical studies reveal about the conditions that promote adaptive evolution. Genetica 112, 183–198 (2001).

38. Richardson, J. L., Urban, M. C., Bolnick, D. I. & Skelly, D. K. Microgeographic adaptation and the spatial scale of evolution. Trends Ecol. Evol. 29, 165–176 (2014).

39. Linhart, Y. B. & Grant, M. C. Evolutionary significance of local genetic differentiation in plants. Annu. Rev. Ecol. Syst. 27, 237–277 (1996).

40. Wright, A. et al. 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 (2013).

41. Wright, A., Schnitzer, S. A. & Reich, P. B. Living close to your neighbors: The importance of both competition and facilitation in plant communities. Ecology 95, 2213–2223 (2014).

42. Akreberg, E. Apomictic and sexual seed formation in Poa pratensis. Hereditas 25, 359–370 (1939).

43. USDA & NRCS. Kentucky bluegrass (Poa pratensis L.). The Plants Database http://plants.usda.gov (2015).

44. Wagner, L. K. in Evolutionary Ecology of Plants (eds Bock, J. H. & Linhart, Y. B.). 273–284 (Westview Press, 1989).

45. Zuur, A. E., Leno, E. N., Walker, J. N., Saveliev, A. A. & Smith, G. M. in Mixed Effects Models and Extensions in Ecology with R (Springer Science-business Media, 2009).

46. Satterthwaite, F. E. An approximate distribution of estimates of variance components. Biometrics Bull. 2, 110–114 (1946).

47. R CoreTeam. R: A language and environment for statistical computing. (2013) http://www.r-project.org/.

48. Bates, D., Maechler, M., Bolker, B. & Walker, S. lme4: linear mixed-effects models using Eigen and S4. http://cran.r-project.org/package=lme4 (2014).

49. Kuznetsova, A., Brockhoff, P. B. & Christensen, R. H. B. lmerTest: tests for random and fixed effects for linear mixed effect models (lmer objects of lme4 package). http://cran.r-project.org/package=lme4 (2014).

50. Kuznetsova, A., Brockhoff, P. B. & Christensen, R. H. B. lmerTest: tests for random and fixed effects for linear mixed effect models (lmer objects of lme4 package). http://cran.r-project.org/package=lme4 (2014).

51. Geyer, C. J., Ridley, C. E., Latta, R. G., Ettersen, J. R. & Shaw, R. G. Local adaptation and genetic effects on nitrate calculations for exponential family models. Ann. Appl. Stat. 7, 1778–1795 (2013).

52. Kleyhans, E. J., Otto, S. P., Reich, P. B. & Vellend, M. Data from: adaptation to elevated CO2 in different biodiversity systems. Dryad Digital Repository (2016).

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