Climate change alters stoichiometry of phosphorus and nitrogen in a semiarid grassland

Feike A. Dijkstra1, Elise Pendall2, Jack A. Morgan3, Dana M. Blumenthal3, Yolima Carrillo1, Daniel R. LeCain3, Ronald F. Follett4 and David G. Williams5

1Department of Environmental Sciences, The University of Sydney, Eveleigh, NSW, 2015, Australia; 2Department of Botany and Program in Ecology, University of Wyoming, Laramie, WY, 82071, USA; 3Rangeland Resources Research Unit, USDA-ARS, Fort Collins, CO, 80526, USA; 4Soil, Plant, and Nutrient Research Unit, USDA-ARS, Fort Collins, CO, 80526, USA; 5Departments of Botany, Ecosystem Science and Management, Program in Ecology, University of Wyoming, Laramie, WY, 82071, USA

Author for correspondence: Feike A. Dijkstra
Tel: +61 2 8627 1122
Email: feike.dijkstra@sydney.edu.au
Received: 23 July 2012
Accepted: 22 August 2012


Key words: elevated carbon dioxide, grasslands, homeostasis, N : P stoichiometry, nutrient availability, PHACE, soil moisture, temperature.

Introduction

Ongoing increases in atmospheric CO2 and temperature have large effects on carbon (C) and nutrient cycles in terrestrial ecosystems, with potentially strong feedbacks to the global climate system (Heimann & Reichstein, 2008). Much attention has been given to CO2 and temperature effects on the nitrogen (N) cycle because of its importance for plant growth, C sequestration and biodiversity (An et al., 2005; Reich, 2009; Niü et al., 2010; Norby et al., 2010). Elevated CO2 often results in reduced N availability to plants, as increased plant growth stimulates plant N uptake and immobilization in long-lived plant biomass, and increased soil C stimulates microbial N immobilization in the soil (Luo et al., 2004). Reduced soil N availability may eventually dampen the stimulatory effect of elevated CO2 on plant growth (Luo et al., 2004; Norby et al., 2010). An increase in temperature, however, often increases N availability to plants, because increased microbial activity in the soil releases N from organic matter (Rustad et al., 2001; Melillo et al., 2002), which, in turn, can offset the immobilization effect of elevated CO2 (Hovenden et al., 2008).

Plant growth and microbial activity in terrestrial ecosystems are also often affected by phosphorus (P) availability (Güsewell, 2004; Cleveland & Liptzin, 2007; Elser et al., 2007). In most soils, N is supplied through biological N fixation and atmospheric deposition, whereas the primary source of P is rock weathering. Both nutrients are recycled through soil organic matter decomposition. N limitation often occurs in young soils with limited accumulation of atmospheric N input (Vitousek & Farrington, 1997), or in systems with high N loss through nitrification and denitrification processes, fires and leaching (Vitousek & Howarth, 1991). P limitation often occurs in old soils in which P is no longer supplied through weathering, or in soils with high P sorption/precipitation capacity (Walker & Syers, 1976; Vitousek & Howarth, 1991). Primary productivity in terrestrial ecosystems is therefore frequently limited by both N and P in soils of intermediate age (Vitousek & Farrington, 1997) or when the availability of both nutrients is low (Harpole et al., 2011). Indeed, the limitation of both N and P on primary productivity is widespread in terrestrial ecosystems (Güsewell, 2004; Elser et al., 2007; Craine et al., 2008; Craine & Jackson, 2010; Harpole et al., 2011).

The availability of P as well as N may be altered by elevated CO2 and warming, potentially causing shifts in N and P supplied to plants. Although the release of both nutrients is biologically controlled through the mineralization of soil organic matter by microbes, P availability is also controlled geochemically through sorption/desorption and dissolution/precipitation reactions (Achat et al., 2005; Reich, 2009).

Summary

• Nitrogen (N) and phosphorus (P) are essential nutrients for primary producers and decomposers in terrestrial ecosystems. Although climate change affects terrestrial N cycling with important feedbacks to plant productivity and carbon sequestration, the impacts of climate change on the relative availability of N with respect to P remain highly uncertain.

• In a semiarid grassland in Wyoming, USA, we studied the effects of atmospheric CO2 enrichment (to 600 ppmv) and warming (1.5/3.0°C above ambient temperature during the day/night) on plant, microbial and available soil pools of N and P.

• Elevated CO2 increased P availability to plants and microbes relative to that of N, whereas warming reduced P availability relative to N. Across years and treatments, plant N : P ratios varied between 5 and 18 and were inversely related to soil moisture.

• Our results indicate that soil moisture is important in controlling P supply from inorganic sources, causing reduced P relative to N availability during dry periods. Both wetter soil conditions under elevated CO2 and drier conditions with warming can further alter N : P. Although warming may alleviate N constraints under elevated CO2, warming and drought can exacerbate P constraints on plant growth and microbial activity in this semiarid grassland.
et al., 2009; Yang & Post, 2011). Elevated CO$_2$ and warming effects on these biotic and abiotic processes could alter the relative supply of N and P, and eventually cause changes in primary productivity, soil organic matter decomposition and biodiversity (Güsewell, 2004; Wassen et al., 2005; Güsewell & Gessner, 2009; Liu et al., 2010).

Plants and microbes require N and P in specific ratios for optimal growth and have limited flexibility to take up N and P under conditions of variable and often unbalanced N and P availability. Perturbations affecting the relative availability of N and P may have different impacts on plant growth and microbial activity because of their dissimilar requirements for N and P. For instance, microbial N : P ratios tend to be lower than plant N : P ratios (Cleveland & Liptzin, 2007), suggesting that the demand for P relative to N is greater for microbes than for plants. Furthermore, plant species differ in their stoichiometric homeostasis in the N : P ratio, or in their ability to maintain a constant N : P ratio under variable supply of both nutrients (Sterner & Elser, 2002; Güsewell, 2004). The maintenance of stoichiometric homeostasis in environments with variable nutrient supply is energetically expensive for plants (Sterner & Elser, 2002). Thus, environments with temporally variable nutrient supply may favour species that are flexible in their N : P stoichiometry (Sardans et al., 2012). The importance of stoichiometric N : P flexibility in plant growth responses to climate change is unknown.

In this study, we examined how elevated CO$_2$ and warming affect N and P cycling in a semiarid grassland in Wyoming, USA with low N and P availability. We included an irrigation treatment to study the direct effects of soil water availability on N and P cycling. Previously, we have demonstrated that elevated CO$_2$ increases significantly soil moisture as a result of reductions in plant stomatal conductance, and that warming decreases significantly soil moisture as a result of desiccation effects at this site (Morgan et al., 2011). Consequently, warming induces more extreme soil moisture conditions (i.e. faster and more severe soil drying after rainfall events), whereas elevated CO$_2$ dampens soil moisture extremes. The C3 grasses at this site respond most strongly to elevated CO$_2$, whereas the C4 grasses also respond to warming. Further, we have shown that soil N availability decreases with elevated CO$_2$ and increases with warming (Dijkstra et al., 2010; Carrillo et al., 2012). Here, we build on this work by examining P and N : P in plants, microbes and soils. The objectives of this study were to examine how elevated CO$_2$ and warming affect plant, microbial and available pools of N and P in the soil; how N and P dynamics are related to soil moisture; and how stoichiometric flexibility in plant N : P relates to changes in plant growth among individual species in response to elevated CO$_2$ and warming.

Materials and Methods

The Prairie Heating And CO$_2$ Enrichment (PHACE) experiment is located at the US Department of Agriculture Agricultural Research Service High Plains Grasslands Research Station, Wyoming, USA (latitude 41°11′N, longitude 104°54′W). The ecosystem is a northern mixed-grass prairie dominated by the cool-season C3 grasses _Pascopyrum smithii_ (Ryd.) A. Love and _Hesperostipa comata_ Trin and Rupr., and the warm-season C4 grass _Bouteloua gracilis_ (H.B.K) Lag. Other species include the sedge _Carex elaeocharis_ L. Bailey and the forb _Sphaeralcea coccinea_ (Nutt.) Rydb. These species comprise between 80 and 99% of the total aboveground biomass. The mean annual precipitation is 384 mm and the mean air temperatures are 17.5°C in July and −2.5°C in January. The soil is a fine loamy, mixed, mesic Aridic Argustoll with a pH of 7.0, bulk density of 1.2 g cm$^{-3}$, 62% sand, 23% silt and 15% clay. The calcareous soil contains substantial amounts of solid inorganic P (calcium phosphates), forming 42% of total soil P, whereas almost all soil N is in organic form (Table 1).

The core experiment consists of 20 circular plots (each 3.4 m in diameter) with two atmospheric CO$_2$ concentration levels (ambient and 600 ppmv) and two temperature levels (ambient and 1.5/3.0°C above ambient temperature during the day/night) in a full factorial design (five replicates for each treatment combination). The experiment uses Free-Air CO$_2$ Enrichment, or FACE, technology to raise the CO$_2$ concentration, and ceramic infrared heaters with a proportional-integral-derivative feed-back loop to raise the canopy temperature. The CO$_2$ treatment began in April 2006 and the warming treatment in April 2007. The experiment includes five extra plots that are kept under ambient CO$_2$ and temperature conditions, but with 60 mm yr$^{-1}$ of irrigation applied in three to four events during the growing season. This amount of added water increased the soil moisture content to levels approximating those observed in plots exposed to elevated CO$_2$. The volumetric soil moisture content was continuously monitored in all plots at a soil depth of 10 cm (Sentek Envirosmart sensors, Sentek Sensor Technologies, Stepney, SA, Australia). Detailed information about the site and experiment has been reported elsewhere (Dijkstra et al., 2010; Morgan et al., 2011).

Aboveground plant biomass was clipped in mid-July of 2007, 2008 and 2009 at peak biomass. A metal grid containing 24 quadrats (25 cm × 25 cm; total of 1.5 m$^2$) was placed inside the plots, and vegetation in every other quadrat (12 in total) was clipped to the crown. Aboveground biomass was separated by green and senesced biomass. Green biomass was further separated by species (senesced biomass could not be identified by species). We analysed N and P on senesced plant tissue and green tissue of the five species mentioned above. Samples were dried (60°C), weighed and ground, and then analysed for N concentration on an elemental analyser (Carlo Erba, Haake Buchler Instruments, Saddle Brook, NJ, USA). Subsamples were ashed before colorimetric P analysis using the ammonium molybdate–vanadate reagent (Jackson, 1958) on a spectrophotometer (Bausch and Lomb, Rochester, NY, USA). In 2007 and 2009, Plant Root Simulator (PRS) resin

<table>
<thead>
<tr>
<th>Pool</th>
<th>Total g m$^{-2}$</th>
<th>Organic form g m$^{-2}$</th>
<th>Organic form % of total</th>
<th>Inorganic form g m$^{-2}$</th>
<th>Inorganic form % of total</th>
</tr>
</thead>
<tbody>
<tr>
<td>C</td>
<td>2989 (168)</td>
<td>2945 (167)</td>
<td>98.5</td>
<td>44 (19)</td>
<td>1.5</td>
</tr>
<tr>
<td>N</td>
<td>256 (14)</td>
<td>256 (14)</td>
<td>99.8</td>
<td>0.4 (0.03)</td>
<td>0.2</td>
</tr>
<tr>
<td>P</td>
<td>64.4 (2.2)</td>
<td>38.3 (2.1)</td>
<td>59</td>
<td>26.1 (0.6)</td>
<td>41</td>
</tr>
</tbody>
</table>
Amino acids that are present in many soils (Hofmockel et al., 2010), it remains unclear how important organic N uptake is for plant nutrition (Na¨sholm et al., 2009). Even less clear is the importance of organic P uptake (Macklon et al., 1994). In 2005, before experimental treatments started, soils were sampled in each plot from 0 to 5 and 5 to 15 cm soil depth, and analysed for bulk density and inorganic C as carbonates (Sherrod et al., 2002).

In 2009, soils were sampled 1 wk after the aboveground biomass sampling in each plot at the same depths as in 2005. After inorganic C had been removed with 1 M H3PO4, soils were analysed for total organic C and N on an elemental analyser. Total organic and inorganic P were analysed using the ignition method. Soil subsamples were extracted with 0.05 M K2SO4 for C and N analyses on a total organic C analyser (Shimadzu Scientific Instruments, Wood Dale, IL, USA) and with 0.5 M NaHCO3 for colorimetric P analysis using the ammonium molybdate–ascorbic acid reagent (Olsen & Sommers, 1982). To correct for P sorption during fumigation and extraction, five off-plot samples were spiked with 100 µg P and similarly analysed for P (Brookes et al., 1982). We used 0.45, 0.54 and 0.4 extraction efficiency corrections for C, N and P, respectively, to calculate microbial C, N and P. Although these sorption and extraction efficiency correction factors are soil type dependent (e.g. soil texture), we do not expect them to be affected by the treatments of our experiment. All soil measurements were expressed per unit area using the bulk density measurements conducted in 2005 and by combining the two soil depths. Detailed methods for plant, PRS probe and soil N analyses are provided elsewhere (Dijkstra et al., 2010; Morgan et al., 2011).

The climate change treatments at PHACE altered the balance of available nutrients in soil and in plant and microbial biomass pools (Fig. 1). In general, elevated CO2 decreased N : P ratios in aboveground plant biomass (Fig. 1a,b). Warming increased N : P ratios, particularly in 2008 and 2009, although the warming effect was not as strong as the elevated CO2 effect. Elevated CO2 and warming had similar effects on the N : P ratios of the plant species B. gracilis, H. comata and P. smithii, but did not alter significantly the N : P ratios of the less abundant species C. eleocharis and S. coccinea (Supporting Information Table S1). The N : P ratios measured in green plant biomass were always smaller than in senesced plant biomass, indicating that the resorption of P was greater relative to that of N. Warming increased microbial N : P in 2009, particularly under ambient CO2 (Fig. 1c). The N : P ratio of available N and P in the soil, measured with the PRS resin probes for the 2007 and 2009 growing seasons, decreased with elevated CO2 (Fig. 1d).

Treatment effects on N : P stoichiometry were sometimes caused by changes in N, sometimes by changes in P, and were sometimes only expressed by simultaneous changes in N and P (Tables S2–S4). For example, although treatment effects on green plant N : P were caused by changes in N concentration in 2007 and 2008, they were caused by changes in P concentration in 2009 (Table S2). Warming increased the green plant N pool for the five dominant species in all 3 yr (P < 0.05 in 2008 and P < 0.1 in 2007 and 2009), as reported previously for all species combined (Dijkstra et al., 2010; Carrillo et al., 2012), but had no effect on the green plant P pool (Table S3). Elevated CO2 had no effect on the green plant N and P pools of the five dominant species. However, the highest P amounts were observed in the CT treatment (see Fig. 1 for definition), causing a marginally significant CO2 × temperature interactive effect in 2009. Elevated CO2 decreased the available N measured by the PRS probes in both 2007 and 2009, but had no effect on available P (Table S4). Warming increased available N
in both years, and also increased available P in 2007, but not to the same extent as N. Treatment effects on microbial N : P were caused by simultaneous changes in microbial N and P (Table S4). Elevated CO2 caused a marginally significant increase in the microbial N : C ratio (reflecting an increase in microbial N concentration), whereas warming reduced both the microbial P : C ratio (or microbial P concentration) and P amount.

The irrigation treatment decreased plant N : P ratios in 2007 and 2008, similar to that observed with elevated CO2, but not during the relatively wet year of 2009 when annual precipitation was 17% higher than the 132-yr mean for this site (Fig. 1e). In that relatively wet year, plant and PRS probe N : P ratios were lower than in the two previous years when precipitation was within 7% of the long-term mean. The PRS probe N : P ratios varied between 0.1 and 25, whereas the green plant N : P ratios varied between 5 and 18 across the CO2 and warming treatments and years. Further, PRS probe and green plant N : P ratios were negatively related to mean early-season soil moisture content measured at a soil depth of 10 cm (Fig. 2a,b). For the PRS probes, this negative relationship was caused by a decrease in N with increased soil moisture, whereas, for plants, this was caused by both a decrease in plant N concentration and an increase in plant P concentration with increased soil moisture (Fig. 2c–f). Green plant N content was not related to soil moisture, but green plant P content increased significantly with increased soil moisture (Fig. 2g,h).

We observed a significant positive relationship between microbial and green plant N : P and between microbial and PRS probe N : P measured in 2009 (Fig. 3). Microbial N : P ratios were much smaller than green plant N : P ratios, suggesting that P requirements relative to N were larger for microbes than for plants. However, the PRS probe N : P ratios measured in 2009 were similar to the microbial N : P ratios. Microbial N : P ratios were not related to total soil organic matter N : P ratios (P > 0.1, data not shown).

The N : P ratios of individual plant species were responsive to changes in the relative availability of N and P in the soil measured in 2007 and 2009 in the 20 core plots (Fig. 4). The five species measured showed a variable degree of stoichiometric flexibility or homeostasis, with the C4 grass B. gracilis showing the greatest flexibility (least homeostatic) to relative changes in available N and P in the soil, and the C3 grass P. smithii showing the least flexibility (most homeostatic). The slopes of the relationships shown in Fig. 3 between soil-available and plant N : P ratios were significantly different among the five species (interactive effect of species × log(PRS probe N : P) in ANCOVA, P = 0.03), and the H values, a measure of the degree of homeostasis (Sterner & Elser, 2002), ranged between 4.3 and 9.6. Despite this range, we did not observe consistent relationships between species-specific H values and plant growth responses to elevated CO2 and warming.

Discussion

In general, soil-available, plant and microbial N : P ratios decreased under elevated CO2 and increased with warming. We found strong evidence that the opposing effects of elevated CO2 and warming on available N : P to plants and microbes were driven by variations in soil moisture. Elevated CO2 increased soil moisture as a result of reductions in plant stomatal conductance, whereas the desiccating effect of warming decreased soil moisture (Morgan et al., 2011). Soil moisture control on soil-available and plant N : P ratios was supported by lower N : P ratios with irrigation, lower N : P ratios in
Fig. 2 Relationships between soil moisture and N : P ratios on the Plant Root Simulator (PRS) probes (a) and in green plants (b). N (c) and P (d) contents on PRS probes, N (e) and P (f) concentrations in green plant tissue, and green plant N (g) and P (h) pools. Relationships are shown for the species-weighted average and volumetric soil moisture at a soil depth of 10 cm, averaged over days of year (DOY) 100–200. Each point represents a specific plot measured in 2007, 2008 and 2009 (2007 and 2009 for PRS probes). Treatments: ct, ambient CO2 and temperature; cT, ambient CO2 and 1.5/3°C day/night warming; Ct, 600 ppmv CO2 and ambient temperature; CT, 600 ppmv CO2 and 1.5/3°C day/night warming.
Changes in soil moisture caused by elevated CO₂ or warming could also affect the proportional supply of P and N. Desorption and dissolution of inorganic P in the soil can function as an important source of P for plants and microbes, particularly in calcareous soils (Lajtha & Bloomer, 1988; Tunesi et al., 1999). The desorption and dissolution of inorganic P depend on inorganic equilibria with the P concentration in soil solution. An increase in soil moisture increases the diffusivity of P, thereby enhancing the uptake by plants and microbes (Lambers et al., 2006). Increased plant and microbial uptake of P would deplete soluble P rather rapidly if soluble P was not buffered by desorption and dissolution reactions (Fitter & Hay, 2002). An increase in soil moisture may then have increased the supply of P to plants and microbes through enhanced desorption and dissolution. At our site, a large fraction of the total P pool in the soil was in inorganic form (Table 1), suggesting that P desorption and dissolution may be especially important for P supply in this system. Increased P desorption and dissolution with increased soil moisture could explain why both green plant P concentrations and pools increased with increased soil moisture (Fig. 2f,h). However, the decrease in green plant N concentration with increased soil moisture (Fig. 2c) was most probably a result of a dilution effect caused by the increase in plant productivity, as the green plant N pool was unaffected by soil moisture (Fig. 2g). This also suggests that the decrease in available N with increased soil moisture (Fig. 2c) was caused by an increase in microbial N immobilization, as opposed to an increase in plant uptake. Previously, we have shown that irrigation and elevated CO₂-induced increases in soil moisture also increase microbial N immobilization (Dijkstra et al., 2010; Carrillo et al., 2012). Here, we propose that the soil moisture-induced increases in microbial N immobilization may also have been stimulated by increased microbial P supply through desorption and dissolution.

Further indirect support for the importance of inorganic reactions in controlling P supply at our site comes from the relationship between microbial and plant N : P. If plant N and P were predominantly supplied through organic matter decomposition, ecological stoichiometry theory would predict that the relationship between microbial and plant N : P ratios should be negative (Sterner & Elser, 2002). When microbes depend solely on soil organic matter as their source of N and P, a shift to greater uptake and storage of N relative to P during soil organic matter decomposition should result in greater release of P relative to N. In contrast, available and plant N : P ratios were both positively related to microbial N : P ratios in the soil (Fig. 3). This suggests that the availability of P was not primarily controlled by soil organic matter decomposition. It also suggests a decoupling of the supply of N (released mostly through mineralization) and P (mostly through desorption/dissolution) in this system. Similarly, decoupling of N and P supply from organic matter may also occur because N is released through biological mineralization (or oxidation of organic matter), whereas P can also be released through biochemical mineralization (release from P esters with the help of extracellular enzymes; McGill & Cole, 1981).

Under dry conditions, when geochemical reactions of P are limited by P diffusivity, the warming effect on N and P availability may have been largely driven by changes in soil organic matter
decomposition. For instance, available P measured with the PRS probes increased with warming in the relatively dry year of 2007 (Table S4), despite a potentially lower P supply from geochemical reactions as a result of warming-induced soil drying. This suggests that warming increased P mineralization in that year. Available N and plant N pools also increased with warming, further suggesting enhanced N mineralization with warming (Dijkstra et al., 2010). In contrast, the green plant P pool was not affected by warming in 2007 (Table S3), possibly because, under dry soil conditions, plant uptake of P was limited by the diffusion of P to the root, more so than N (Fitter & Hay, 2002). A direct warming effect on P mineralization may also help to explain why the available P did not relate to soil moisture (Fig. 2d). Overall, the supply of P at our site is likely to be controlled by a suite of mechanisms that are active under different environmental conditions, because of the nearly co-dominant inorganic and organic P sources.

We have no direct evidence for N and P limitation at our site. However, there are several reasons why we believe that both N and P were important for plant growth at our study site. First, soil available N and P concentrations at our site were low, increasing the likelihood that both nutrients affected plant growth (Harpole et al., 2011). The low N and P conditions are illustrated by the extremely low losses of N as N₂O at this site (<2 mg N m⁻² yr⁻¹, F. A. Dijkstra et al., unpublished results) and the low levels of available P (4–7 mg P kg⁻¹ soil, measured in nonfumigated soil extracts) relative to those in 98 North American grassland soils (2–100 mg P kg⁻¹ soil; Craine & Jackson, 2010). Second, species-weighted green plant N : P ratios in 2007 and 2008 were between 10 and 20, and, although critical plant N : P ratios for nutrient limitation are not always unambiguous, values between 10 and 20 may indicate limitation of both N and P for plant growth at the ecosystem level (Güsewell, 2004). In the relatively wet year 2009, when green plant N : P ratios were often below 10, plant growth may have become more N limited. Third, we found indirect evidence that N made available to plants through microbial mineralization may be mediated by P supply to microbes that have a relatively high P requirement. Indeed, the dependence of plant productivity on both N and P may be widespread in grasslands because of the nature of the interaction between P and N availability (Craine & Jackson, 2010).

Although the aboveground biomass of C3 and C4 grasses responded differently to elevated CO₂ and warming (Morgan et al., 2011) and the N : P ratio differed among plant species (Table S1), elevated CO₂ and warming effects on overall plant N : P ratios were most probably not caused by differential species responses and shifts in vegetation. The N : P ratios of the three dominant plant species B. gracilis, H. comata, and P. smithii all showed similar responses to elevated CO₂ and warming. In contrast, the N : P ratios of C. eleocharis and S. coccinea did not respond to elevated CO₂ and warming, but their contribution to the total aboveground biomass was relatively small (on average 10% for C. eleocharis and 3% for S. coccinea). All plant biomass samples were taken at peak biomass in mid-July, when plants were at a similar late stage of development, indicating that potential differences in plant N : P among species and years caused by developmental differences were most probably small.

The biomass N : P ratios of individual plant species were positively related to soil-available N : P ratios (none were strongly homeostatic), but species varied in their stoichiometric flexibility in response to changes in soil-available N : P (Fig. 4). It has been suggested that environments with a temporally variable nutrient supply may favour species that are flexible in their N : P
stoichiometry (Sardans et al., 2012), as the maintenance of stoichiometric homeostasis under these conditions is energetically expensive for plants (Sterner & Elser, 2002). In semiarid grasslands, flexible or nonhomeostatic nutrient uptake rates may also be advantageous for plants, so that N uptake is not impeded by low P availability during the long dry periods, and vice versa, and so that P uptake is not hindered by the relatively low N availability during the short wet periods. We found partial support for this. Warming induced more extreme soil moisture conditions (i.e. faster and more severe soil drying after rainfall events) and increased the above-ground biomass of B. gracilis (Morgan et al., 2011), which also showed the greatest flexibility in N : P. Similarily, water savings under elevated CO₂ reduced the extremes in soil moisture at our site, which could be relatively more advantageous to species with low N : P flexibility. Indeed, P. smithii showed the lowest N : P flexibility and was one of the C₃ grasses that responded most strongly to elevated CO₂ in 2007 and 2008. In contrast, H. comata and S. coccinea, which showed a similar high flexibility in N : P to B. gracilis, did not respond to warming or elevated CO₂. Possibly, differences in photosynthetic pathways and rooting depths (LeCain et al. 2006) confound the clear relationships between species-specific N : P flexibility and their responses to climate change. Stoichiometric homeostasis has also been linked to plant dominance in a similar semiarid grassland in Inner Mongolia (Yu et al., 2010), but we found no strong support for this. At our site, B. gracilis and P. smithii were two of the three dominant species, but showed the most and least N : P flexibility, respectively.

It has been argued that plant productivity may become increasingly constrained by N under elevated CO₂ alone (Luo et al., 2004; Norby et al., 2010). Our results suggest that an increase in P availability relative to N under elevated CO₂ may further exacerbate N constraints. However, warming could alleviate N constraints on plant growth under elevated CO₂, and may increase P constraints, particularly under dry conditions. As soils dried out more rapidly with warming, P became progressively less available to plants relative to N. It is unclear whether similar effects would occur in wetter environments and in noncalcareous soils that contain less inorganic P, and where P supply is largely controlled by organic matter decomposition. Enhanced precipitation increased plant P uptake, but elevated CO₂ and warming had no effect on plant P or N : P in an annual grassland in California (Menge & Field, 2007) or in other mesic ecosystems (Chapin et al., 1995; Niklaus et al., 1998; Finzi et al., 2001). Our results suggest that primary productivity and microbial activity in semiarid grasslands that occur widely on calcareous soils may become more reliant on P availability in a warmer and drier world. The enormous increase in anthropogenic N fixation and resultant increase in atmospheric N deposition (Galloway et al., 2008) could further exacerbate the increased P constraints on plant growth in a warmer and drier environment. Thus, the dynamics of P relative to N will be critical in predicting responses of semiarid grasslands to climate change.

Acknowledgements

We thank D. Smith for the installation and operation of the PHACE experiment, E. Hardy for assistance in installation, M. Parsons for assistance with data collection, F. Migletta for advice and help on the installation of the FACE system, B. Kimball for advice and help on the installation of the infrared heating system, and R. Phillips and three anonymous reviewers for comments on the manuscript. Research was supported by the US Department of Agriculture (USDA)-Agricultural Research Service Climate Change, Soils & Emissions Program, the US National Science Foundation (NSF) (DEB# 1021559), and the US Department of Energy’s Office of Science (Biological and Environmental Research). F.A.D. was supported by a fellowship from the Australian Research Council (FT100100779). The mention of commercial products is solely for the purpose of providing specific information and does not imply the recommendation or endorsement by the USDA. Any opinions, findings, and conclusions or recommendations expressed in this material are those of the authors and do not necessarily reflect the views of the US NSF.

References


Supporting Information

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

Table S1 Average green plant N : P ratios by species

Table S2 Average green and senesced plant N and P concentrations (%) averaged across all five species

Table S3 Average N and P pools in green plant biomass of the five species combined

Table S4 Average N and P pools on Plant Root Simulator (PRS) probes (2007 and 2009) and N : C, P : C and N and P pools in microbial biomass (2009)

Please note: Wiley-Blackwell are not responsible for the content or functionality of any supporting information supplied by the authors. Any queries (other than missing material) should be directed to the New Phytologist Central Office.