Grazing moderates increases in C₃ grass abundance over seven decades across a soil texture gradient in shortgrass steppe

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Abstract

Questions: How does long-term exclusion of cattle grazing influence plant community composition in a semi-arid grassland? Can spatial variation in the effects of cattle grazing exclusion be explained by variation in soil texture?

Location: Shortgrass steppe, northeast Colorado, USA, in the North American Great Plains.

Method: We used 16 long-term (72 yr) cattle exclosures to examine the effects of grazers on plant communities and evaluate whether grazer effects interact with soil texture.

Results: Although shortgrass steppe communities are relatively unaffected by grazing in the short-term (one to two decades), exclusion of cattle grazing for seven decades caused a compositional shift from dominance by a C₄ shortgrass (*Bouteloua gracilis*) to co-dominance by a C₃ midgrass (*Pascopyrum smithii*) and *B. gracilis*. The strength of this shift was highly variable across sites. Soil texture was correlated with the abundance of certain plant species, but did not explain variation in the magnitude of grazer effects. Grazed communities contained perennial and annual growth forms with a diversity of strategies to co-exist with grazers and *B. gracilis*. Ungrazed communities included increased abundance of annual, ruderal forbs and three woody plant species. Grazing effects occurred against a backdrop of changing plant communities: during the past seven decades, C₃ perennial graminoids and sub-shrubs have increased in relative abundance in both grazed and ungrazed communities.

Conclusions: Our long-term experiment shows that community responses to grazing in this semi-arid grassland occur very slowly, but are predictable, with C₄ shortgrasses eventually giving way to taller C₃ grasses and ruderal forbs. Spatial variation in grazing effects across sites (and lack of a relationship with soil texture) may reflect the importance of fine-scale heterogeneity in C₃ grass abundance, and the slow rate at which taller C₃ grasses can coalesce into mono-dominant patches that outcompete C₄ shortgrasses. Increased abundance of C₃ species over the past seven decades, both in the presence and absence of grazing, may be related to recovery from the severe drought and dust storms of the 1930s as well as enhanced growth of C₃ plants under increasing atmospheric [CO₂].

Introduction

Most semi-arid grasslands support native and/or domestic mammalian herbivores that consume a substantial proportion of above-ground net primary production (ANPP; McNaughton et al. 1991). How these grazers influence plant communities depends in part on ecosystem productivity and the evolutionary history of grazing (Milchunas & Lauenroth 1993). In semi-arid ecosystems with abundant herbivores, grazing and aridity can exert convergent selection pressures on the plant community, leading to the dominance of prostrate growth forms that allocate

significant resources below-ground, capitalize on short windows of moisture availability and regrow efficiently following defoliation (Milchunas & Lauenroth 1993; Quiroga et al. 2010). In the semi-arid shortgrass steppe of North America, the convergence of low precipitation, high growing season evaporative water loss, a long evolutionary history of grazing by large herbivores and the widespread replacement of native herbivores by domestic livestock following European settlement have generated a contemporary plant community dominated by C_4 shortgrasses (particularly blue grama [*Bouteloua gracilis*] and buffalograss [*Bouteloua dactyloides*]). Previous studies show this plant community is highly resistant to effects of grazers. Over time scales of 1–13 yr, grazing removal minimally affects shortgrass plant communities (Klipple & Costello 1960; Hyder et al. 1966; Holechek et al. 2006). After 23 yr of implementing grazing intensities that varied from light to heavy, Hyder et al. (1966) concluded that sustained heavy grazing reduced productivity of dominant grasses but minimally affected community composition. Following a half-century of grazer removal, C_3 midgrasses and some tall-statured annual forbs increased in abundance (Milchunas et al. 1989; Milchunas et al. 1990; Hart & Ashby 1998), but grazer effects on shortgrass communities were still modest compared to other grasslands around the world (Milchunas et al. 2008).

These past studies focused on flat to gently undulating loamy plains that typify the shortgrass steppe. Although travellers crossing the North American Great Plains often regard the region as a flat, homogenous landscape, topoedaphic heterogeneity contributes substantially to variation in plant community composition and productivity (Hyder et al. 1966; Liang et al. 1989; Milchunas et al. 1989). Soil texture influences communities through its influence on water infiltration during and following precipitation events, and subsequent retention during periods of high evaporative demand. The inverse texture hypothesis posits that in semi-arid regions where bare soil evaporation is significant, soils with higher sand content experience deeper water infiltration and less evaporative water loss, thereby enhancing annual net primary productivity (ANPP) and favouring species with higher moisture requirements relative to soils with larger clay and silt content (Noy-Meir 1973). In central North America, an east-to-west gradient of declining precipitation generates a semi-arid climate in much of the western Great Plains (Lauenroth et al. 1999). Across this precipitation gradient, ANPP patterns support the inverse texture hypothesis and indicate that for mean annual precipitation <370 mm, coarse-textured soils support larger ANPP than fine-textured soils (Sala et al. 1988; Liang et al. 1989).

We hypothesized that effects of grazing on plant communities in shortgrass steppe may have been underestimated by previous studies for two reasons. First, it may take many decades for community-level responses to grazing to become apparent (e.g. Fuhlendorf & Smeins 1999). Second, given the important influence of soil texture, which could in turn interact with the influence of grazing, we hypothesized that grazer exclusion would have modest effects on loamy soils (consistent with prior findings of Milchunas et al. 1989, 2008), but that divergence of grazed vs ungrazed communities would be positively associated with soil sand content. We used a network of livestock exclusures constructed in native shortgrass steppe in 1939 to examine whether long-term grazing effects on plant communities are contingent on soil texture. Here we (1) examine plant community divergence within 16 exclosures measured in 2011 (72 yr of grazing exclusion) as compared to shortgrass steppe grazed by livestock at moderate intensity, and (2) evaluate the degree to which divergence is contingent on soil texture. In addition, a portion of the sites we studied are concordant with sites for which Klipple & Costello (1960) report plant community composition measurements during 1940–1953, when the shortgrass steppe was recovering from the extreme drought and 'Dust Bowl' conditions of the 1930s. We use this subset of sites to examine temporal sifts in plant community composition of the shortgrass steppe over the past seven decades in relation to grazing.

Study area

We conducted this study at the USDA-Agricultural Research Service's Central Plains Experimental Range (CPER), a Long-Term Agro-ecosystem Research network site which encompasses ~6400 ha of shortgrass steppe in northeast Colorado, USA (40°50′ N, 104°43′ W). Mean annual precipitation is 340 mm and mean growing season precipitation (Apr–Aug) is 242 mm. During 2008–2011, annual precipitation was 330, 436, 360 and 356 mm and growing season precipitation was 240, 352, 285 and 245 mm, respectively. Thus, 2011 was a year of near-average precipitation. Most of the site is subdivided into 65–130-ha pastures that have been grazed by cattle at varying stocking rates since CPER was established in 1939. At that time, grazing exclosures (four-strand barbed wire; each ~90 m × 90 m) were constructed in >20 locations. Exclosures are on level terrain, with one built in most pastures that existed on CPER in 1939. Pronghorn antelope (*Antilocapra americana*) are present and can pass under exclosure fencing, but are rarely observed inside exclosures. Mammalian herbivores smaller than pronghorn (lagomorphs and rodents) are unimpeded by exclosures. Exclosures in pastures with winter cattle grazing or consistently high or low stocking rates were not

included in this study. The remaining 16 exclosures occurred in pastures consistently managed with season-long grazing during the growing season (May–Oct) at moderate stocking rates (~0.65 AUM·ha^{-1}) for the past 30 yr. Detailed stocking rate records are not available for every pasture prior to the 1980s, but the sites included in this study have experienced moderate, growing season grazing for either most or all of their 72-yr history, including the past 30 yr.

Parent materials of soils are primarily Holocene alluvial and aeolian deposits derived from local sources (Kelly et al. 2008). Alluvium is often covered by younger aeolian deposits, and both are discontinuous due to reworking by winds. Sandy soils are often associated with aeolian deposits, while some soils formed from mixed alluvium have higher clay and silt content (Kelly et al. 2008). Grazed plant communities are dominated by C$_4$ perennial short-grasses (blue grama [*B. gracilis*] and buffalograss [*B. dactyloides*]) which contribute >70% of ANPP (Lauenroth & Burke et al. 2008).

Methods

Field methods

We measured plant communities in paired 30 m × 30 m plots located inside and outside 16 exclosures (total of 32 plots) during Jul 2011. Grazed plots were located on the side of each exclosure that was most similar in terms of soil type (NRCS SSURGO database), slope and aspect, with 10-m spacing between the edge of plots and the exclosure fence. Within each plot, we established a 6 × 6 systematic grid of points with 5-m spacing. We visually estimated the foliar and basal cover of each plant species in a 50 cm × 20 cm quadrat centred over each point (36 total quadrats per plot), and also estimated the basal cover of bare soil and litter. Cover was recorded by classes consisting of 0–1%, 2–5%, 6–15%, 16–25%, 26–40%, 41–60% and >60%, and class midpoints were used in all statistical analyses (Hickman & Derner 2007). We conducted more intensive measurements of sparsely distributed shrubs (*Atriplex canescens* and *Ericameria nauseosa*) and sub-shrubs (*G. sarothrae*, *E. effusum* and *Krasheninnikovia lanata*). For these species, we subdivided the plot into 5-m wide belts and censused the entire plot to measure density per species. For these woody species, we measured canopy dimensions (height, length and width to the nearest cm) for up to the first 30 individuals encountered in the plot. We estimated canopy cover for each species at each site as the product of density and mean canopy area per individual. A fourth subshrub, *A. frigida*, was abundant and well distributed, so for this species we simply estimated canopy cover using the measurements from the 50 cm × 20 cm quadrats described previously.

At each plot, we collected 0–20-cm deep × 10-cm diameter soil cores. This depth encompasses 75% of the root biomass in shortgrass steppe (Liang et al. 1989). We divided each plot in half and collected cores at six systematic locations per half. The six cores per half were combined into two pooled samples per plot, passed through a 2-mm sieve and homogenized. A subsample from each pooled sample was analysed for sand, silt and clay fractions following Bouyoucous (1962).

Data analyses

We examined variation in plant species composition among plots for the data collected in 2011 with a NMS ordination of the species × site matrix based on plant basal cover. We used the Sørenson distance measure as implemented in PCORD (v 6.0; MjM Software Design, Gleneden Beach, OR, US). We used basal cover because it is not affected by current season grazing. We used NMS ordination because it does not make assumptions regarding normality of the data and performs well with simulated gradients. We excluded species that only occurred in ≤2 of the 32 plots. We used linear regression to examine the relationship between soil texture and ordination axis scores, and used paired *t*-tests to evaluate whether axis scores distinguished between grazed vs ungrazed sites.

We conducted univariate analyses of grazing effects on basal cover of 22 species using paired *t*-tests. These species were analysed either because they are abundant in shortgrass steppe and possess traits expected to confer grazing resistance or sensitivity, or they were among the 15 species most positively or negatively correlated with axis 1 of the NMS ordination and were present in ≥4 grazed/ungrazed pairs. One drawback to using basal cover is that prostrate grasses like *B. gracilis* are over-represented relative to their above-ground biomass as compared to more vertical growth forms such as *P. smithii* and most forbs and shrubs. This should be recognized when interpreting absolute values of basal cover presented here. Woody plants are especially poorly represented by basal cover, so we also conducted univariate analyses of canopy cover for the six most common woody species in the community, which for these species is relatively unaffected by current year grazing.

Two previous global-scale syntheses used an index of dissimilarity between grazed vs ungrazed sites to quantify grazing effects on grassland plant communities (Milchunas & Lauenroth 1993; Milchunas et al. 2008). We calculated this same index for our 16 exclosure sites, where Dissimilarity = 1 − (Whittakers index of community association) as defined by Whittaker (1952), and examined how this index relates to distances between paired sites in the NMS ordination.

We also sought to compare our 2011 measurements to historic studies of grazing effects at CPER in the 1940s and 1950s. In 1939, concurrent with the establishment of the exclosures at CPER, a grazing study was initiated that measured plant community composition in four replicate 65-ha pastures assigned to a moderate grazing treatment, and also reported on measurements from 12 exclosures (Klipple & Costello 1960). Our vegetation measurements in 2011 (see Methods above) included measurements in the same four moderately grazed pastures, as well as nine of the 12 exclosures that were included in their study. Klipple & Costello (1960) reported mean species composition of the four moderately grazed pastures (averaged across all replicates; no measure of variance) and mean species composition of the 12 exclosures (no measure of variance). Values reported include means of measurements collected 1–3 yr after grazing exclusion (1940–1942; mean annual precipitation = 392 mm), 7–9 yr after grazing exclusion (1946–1948; mean annual precipitation = 297 mm), and 12–13 yr after grazing exclusion (1952–1953; mean annual precipitation = 331 mm). Measurements in 2011 occurred with near-average precipitation (356 mm), which was intermediate between precipitation during 1940–1942 and 1952–1953. We examine temporal trends in relative species composition based on (a) the same four moderately grazed pastures sampled in the 1940s, 1950s and 2011, and (b) the same 12 exclosures sampled in the 1940s and 1950s and the corresponding subset of nine exclosures measured in 2011. Values for the 1940s and 1950s were based on 'percentage of the ground surface covered by vegetation of each species when viewed from directly above' (Klipple & Costello 1960, p. 14), expressed as a percentage relative to the maximum total percentage vegetation cover per plot. Therefore, we compare their data to our 2011 measurements of foliar cover by species, again expressed as a percentage relative to the maximum total percentage vegetation cover per plot. Our goal was to examine the magnitude of grazing effects on species in this ecosystem in the context of long-term compositional trends.

Results

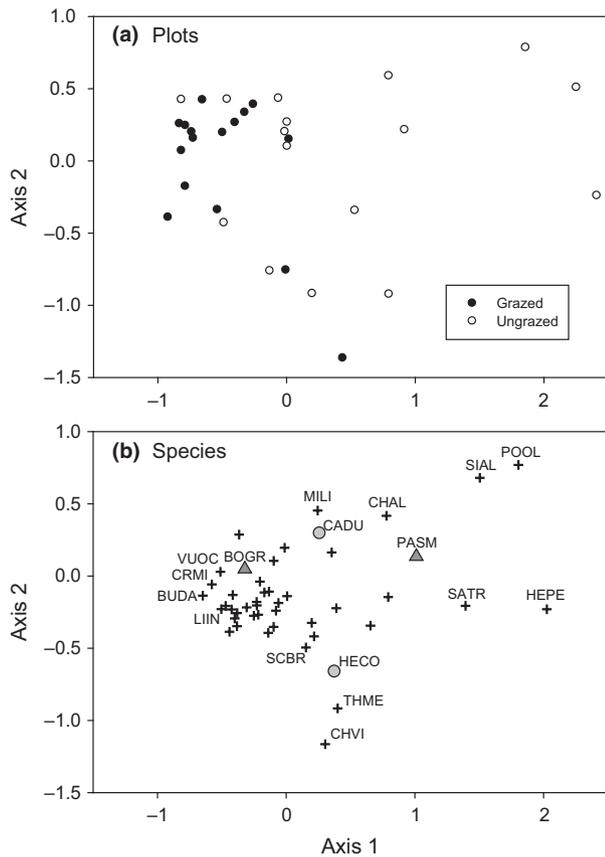### Plant community response to long-term grazing exclusion

The NMS ordination yielded a two-dimensional solution with a final stress of 11.3, and axes 1 and 2 cumulatively explained 94% of the variation in the original species matrix. Grazed vs ungrazed sites diverged significantly along axis 1 (Fig. 1; mean scores for grazed and ungrazed sites = 0.49 and −0.49 respectively; t_{15} = 4.29, P = 0.0006). Variation along axis 1 was not related to soil texture attributes (r < 0.14). Increasing axis 1

values were most strongly correlated with *B. gracilis* (r = −0.93) and *P. smithii* (r = 0.92). Thus, axis 1 represents a gradient of increasing replacement of the perennial C_4 bunchgrass (*B. gracilis*) by a perennial C_3 rhizomatous grass (*P. smithii*) under long-term grazing removal. In addition to *B. gracilis*, species with strong negative correlations with axis 1 (i.e. associated with moderate summer grazing) included a diversity of growth forms: two perennial C_4 grasses (*B. dactyloides* and *Aristida longiseta*), two perennial forbs (*Lithospermum incisum* and *Lathyrus polymorphus*), a biennial forb (*Thelosperma filifolia*), an annual grass (*Vulpia octoflora*) and an annual forb (*Cryptantha minima*; Fig. 1). In contrast, species with strong positive correlations with axis 1 (i.e. with long-term grazing exclusion) other than *P. smithii* were annual forbs considered to be ruderals (*Helianthus petiolaris*, *Chenopodium album*, *Chenopodium lepidium*, *Portulaca oleracea*, *Sisymbrium altissimum*, *Salsola tragus* and *Tragopogon dubius*; Fig. 1). Furthermore, the last four species in this list are exotics.

Axis 2 was moderately correlated with silt content (r = 0.45) but not clay or sand content (r = 0.10 and −0.32, respectively), and did not discriminate between grazing presence/absence (paired t = −0.27, P = 0.78). Axis 2 was most strongly associated with increasing abundance of *Carex duriuscula* (r = 0.62) and decreasing abundance of *Hesperostipa comata* (r = −0.80).

We used the difference in axis 1 scores (ΔAx1) between paired grazed and ungrazed plots as one measure of plant community divergence induced by grazing. In addition, we calculated the dissimilarity index (DI) for each grazed/ ungrazed pair. Variation in DI was significantly associated with variation in ΔAx1 (r^2 = 0.88, P < 0.0001), indicating that both are effective and largely equivalent measures of the magnitude of plant community divergence induced by long-term grazing removal in this ecosystem. DI varied from 0.12 to 0.85 (mean DI ± 95% CI = 0.40 ± 0.11). Most notably, ΔAx1 was not correlated with sand content ($F_{1,14}$ = 2.67, P = 0.12), silt content ($F_{1,14}$ = 1.60, P = 0.23) or clay content ($F_{1,14}$ = 2.00, P = 0.18). Similarly, DI was weakly correlated with sand content ($F_{1,14}$ = 3.43, P = 0.085; r^2 = 0.197; Fig. 2), and not with silt ($F_{1,14}$ = 2.73, P = 0.12) or clay content ($F_{1,14}$ = 1.92, P = 0.19). The relationship between DI and sand content was strongly influenced by the site with lowest sand content having low DI (Fig. 2), while sites with >50% sand exhibited a wide range of DI values (Fig. 2). When the low-sand site was excluded from analysis, there was no indication of a relationship between sand content and DI (P = 0.26).
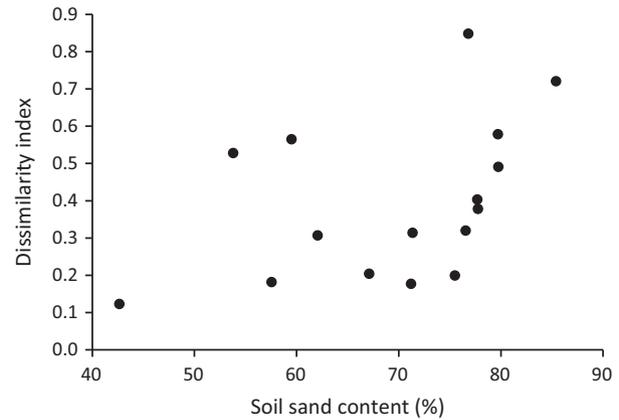
We also conducted univariate tests of grazing exclusion effects on basal cover of 22 individual species. Six species were significantly more abundant in grazed sites compared

Fig. 1. Non-metric multidimensional scaling ordination of grazed vs ungrazed plant communities in shortgrass steppe. Communities were sampled in 16 long-term (72 yr) grazing exclosures and 16 paired, moderately grazed plots in northeast Colorado, US. Panels show the distribution of grazed vs ungrazed plots (**a**) and species (**b**) along ordination axes 1 and 2. In panel (**b**), each four-letter code refers to the first two letters of the genus and species for a plant species, where VUOC = *Vulpia octoflora*, CRMI = *Cryptantha minima*, BOGR = *Bouteloua gracilis*, BUDA = *Bouteloua dactyloides*, LIIN = *Lithospermum incisum*, MILI = *Mirabilis linearis*, CADU = *Carex duriuscula*, SCBR = *Scutellaria brittonii*, HECO = *Hesperostipa comata*, CHAL = *Chenopodium album*, PASM = *Pascopyrum smithii*, SATR = *Salsola tragus*, POOL = *Portulaca oleracea*, HEPE = *Helianthus petiolaris*.

to exclosures ($P \leq 0.05$), and one species was marginally different ($P \leq 0.1$; Fig. 3, Appendix S1). Two species were significantly more abundant in exclosures ($P < 0.05$), and three species were marginally different ($P < 0.1$; Fig. 3, Appendix S1). Two species (*H. petiolaris* and *S. altissimum*) were only detected inside exclosures ($N = 4$ exclosures each) but their distributions were too skewed to conduct valid statistical tests. Two abundant C_3 perennial graminoids (*C. duriuscula* and *H. comata*) and the most abundant cactus (*Opuntia polyacantha*) were unaffected by long-term grazing exclusion (Appendix S1, Fig. 3).

We also analysed effects of grazing exclusion on the six most common woody plant species at the site. Here,
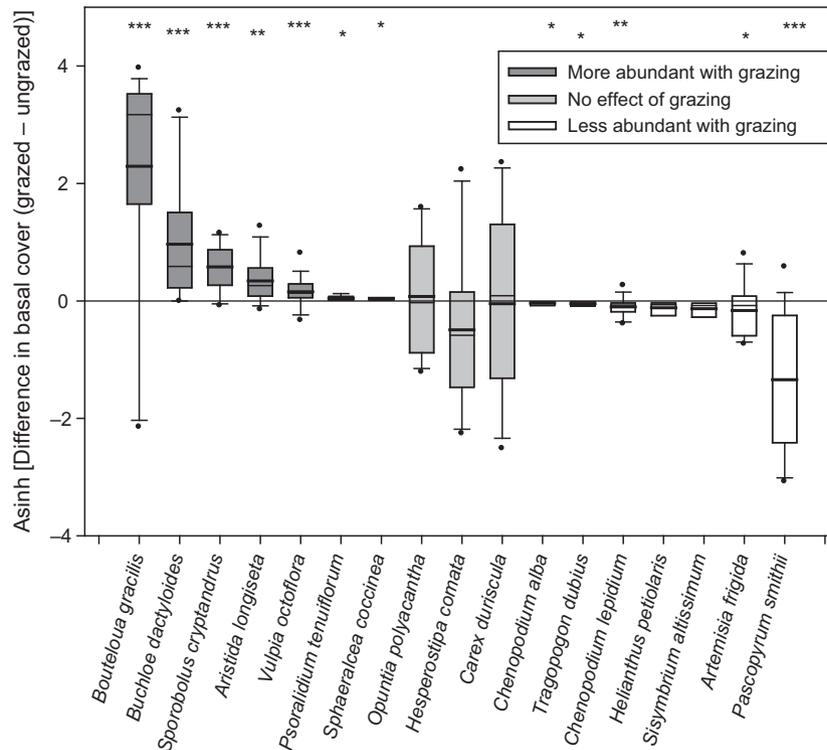

Fig. 2. Relationship between soil sand content and the degree of plant community dissimilarity between grazed vs ungrazed plots (DI). This relationship was strongly influenced by the site with the lowest sand content. When this site was excluded, we found no indication of a relationship between soil sand content and DI ($P = 0.26$).

we used canopy cover because it is a more effective measure of abundance than basal cover for woody species, and because canopy cover of woody species is minimally affected by current season grazing. One sub-shrub (*E. effusum*) was significantly more abundant in grazed plots, while two sub-shrubs (*K. lanata* and *A. frigida*) and one shrub species (*A. canescens*) were significantly more abundant inside exclosures (Appendix S2, Fig. 4). One sub-shrub (*G. sarothrae*) and one shrub (*E. nauseosa*) were unaffected by grazing (Appendix S2, Fig. 4).

Long-term trends in plant community composition

Comparisons of plant community composition during the 1940s and 1950s as reported by Klipple & Costello (1960) to comparable sites that we measured in 2011 provide insights to the influence of grazing on temporal trends over the past 72 yr. Most notably, C_3 perennial graminoids were rare in the 1940s, but increased substantially over time both in the presence and absence of grazing. *P. smithii* increased to a much larger degree without grazing, but the three most abundant C_3 graminoids all increased in the presence of moderate, growing season grazing over the past 72 yr (Tables 1 and 2). Conversely, two C_4 grasses declined by an order of magnitude since the 1940s (Table 1), but remained more abundant in grazed vs ungrazed sites (Fig. 3). *B. gracilis* has remained stable as the dominant species in the presence of grazing, and declined substantially without grazing. Surprisingly, some unpalatable species were stable over time regardless of grazing (the cactus *O. polyacantha*, and the subshrub *G. sarothrae*), or remained stable with grazing and

Fig. 3. Boxplots of differences in absolute basal cover between grazed vs ungrazed plots for 17 plant species. Values shown are on an inverse hyperbolic sine (Asinh) transformation scale, in order to enhance visibility of boxplots for species with both large and small grazing effects. Note the high degree of variability in the magnitude of the grazing effect, even for species that differed significantly between grazed and ungrazed sites. Within each boxplot, the thin black horizontal line shows the median and the thick black horizontal line shows the mean. *** $P < 0.01$, ** $P < 0.05$, * $P < 0.1$.
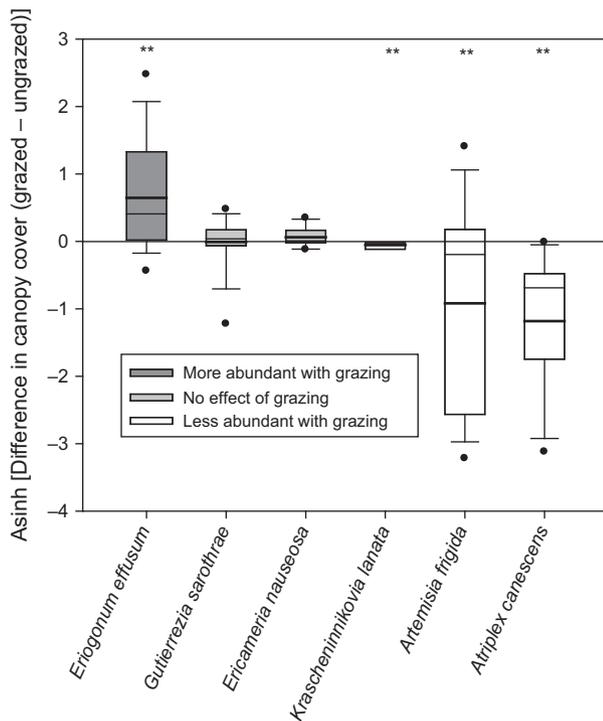
increased without grazing (the subshrub *A. frigida*; Tables 1, 2).

Discussion

The shortgrass steppe of North America is characterized by low and variable precipitation inputs, and a long evolutionary history of grazing by large herbivores (Lauenroth et al. 1999; Milchunas et al. 2008). Aridity and herbivory exert convergent selective forces that lead to dominance of C_4 shortgrasses across a broad range of soils and historic disturbances (reviewed by Lauenroth 2008), and plant communities that are insensitive to grazing exclusion over temporal scales of one to two decades (Klipple & Costello 1960; Hyder et al. 1966; Holechek et al. 2006). Five decades of grazing exclusion in shortgrass steppe induced only modest community shifts (Milchunas et al. 1989; Hart & Ashby 1998). However, after seven decades of grazing exclusion, we found substantial compositional dissimilarity between grazed vs ungrazed communities (mean of 40%) across a wide range of soil textures. High dissimilarity values reflect a switch from dominance by a C_4 shortgrass (*B. gracilis*) to co-dominance by a C_3 midgrass (*P. smithii*;

Figs 1 and 3). In more mesic and productive grasslands, compositional shifts following grazing removal are generally ascribed to a shift from below-ground competition to increasing competition for light (Milchunas & Lauenroth 1993). This same process may occur in shortgrass steppe (Appendix S3) but require a much longer time period, because even without grazing, periodic droughts allow standing dead tillers and litter to decompose and return light to the ground layer. Measurements of how light, soil moisture and soil N vary over time in grazed and ungrazed communities are needed to assess mechanisms driving long-term declines in *B. gracilis* in exclosures.

Species with higher abundance in the presence of grazing (and therefore *B. gracilis*) exhibited a diversity of growth forms, including an annual grass (*V. octoflora*), a stoloniferous, perennial, C_4 shortgrass (*B. dactyloides*), two perennial, C_4 midgrasses (*Sporobolus cryptandrus*, *A. longiseta*) and a perennial, leguminous forb (*Psoralidium tenuiflorum*). In addition, an annual forb (*C. minima*) and another perennial forb (*L. incisum*) were negatively correlated with axis 1 of the ordination (i.e. with grazed communities). Within this community, we see a diverse suite of strategies by which different species contend with

Fig. 4. Boxplots of differences in absolute canopy cover between grazed sites vs ungrazed exclosures for six woody plant species. Symbols and axes follow Fig. 3.

grazing. The two annuals have unique characteristics to avoid consumption by cattle. *V. octoflora*'s shallow root system results in the whole plant being uprooted when bitten, and cattle avoid the species so as to not consume soil. *C. minima* is palatable in early growth stage, but fine hairs on the leaves and stems harden into spines during flowering and deter grazing. *A. longiseta*'s thin leaves have high silica content (Holechek et al. 1989) and ungrazed plants retain old, dead tissues which reinforce grazer avoidance. *B. dactyloides* is frequently grazed, but its prostrate growth form creates an ungrazeable reserve of leaves and meristems near ground level. These findings are consistent with the generalization that grazing effects on plant community composition cannot be explained by a single co-varying set of plant traits (e.g. increases in species with tough, low-nutrient, unpalatable leaves), but rather that grazing facilitates co-existence of species employing diverse strategies related to grazing avoidance and tolerance (Díaz et al. 2001; Agrawal 2007; Diaz et al. 2007).

In contrast to the diverse growth forms that were more abundant with grazing, species that were more abundant in the absence of grazing (hence in communities co-dominated by *P. smithii*) were primarily tall, annual, ruderal forbs. A similar trend was noted by Milchunas et al. (1989, 1990). Our results, which include a different set of mostly

exotic ruderal species, provide additional evidence that grazing helps maintain invasion resistance in the short-grass steppe. Several mechanisms may underlie this pattern. First, persistence of tall, palatable ruderals may be directly inhibited by cattle consumption. In addition, reduced basal cover of *B. gracilis* crowns inside exclosures creates larger spacing between individual grass plants, which is not occupied as densely by roots and crowns of the rhizomatous *P. smithii*. This may create more microsites with reduced below-ground competition from *B. gracilis*, which facilitates germination and establishment of annual, C_3 forbs (Milchunas et al. 1992). We speculate that both reduced N uptake by the densely rooted *B. gracilis* (Liang et al. 1989) and enhanced soil moisture retention in spring beneath *P. smithii* canopies and dead tillers contributes to enhanced abundance of ruderals.

In addition to increased divergence between grazed vs ungrazed communities over the past 72 yr, the magnitude of divergence varied spatially (Fig. 2). We hypothesized that dissimilarity would increase with soil sand content, but this was not supported by any of our community-level analyses. Sites with >50% sand content varied substantially in dissimilarity values (Fig. 2b), and we conclude that grazing effects on community composition are unrelated to surface soil texture across a relatively broad gradient including sandy clay loams, sandy loams and loamy sands (50–85% sand; 10–28% clay).

The NMS ordination results show soil texture influences plant species composition, but again support the conclusion that soil-induced variation does not interact with grazing effects. Because the ordination is not constrained by any environmental variables, these two axes represent compositional variation across the landscape that could be influenced by factors other than grazing and soil texture. The finding that axis 1 effectively separated grazed vs ungrazed sites and axis 2 was correlated with soil texture indicates that grazing and texture are two primary drivers of compositional variability in this ecosystem. The finding that axis 1 was clearly unrelated to texture and axis 2 did not discriminate between grazed/ungrazed sites supports our conclusion that grazing effects were not contingent on soil texture. Furthermore, the two species that varied most with soil texture (*C. duriuscula* and *H. comata*) were not influenced by grazing exclusion. Persistence of *H. comata* at similar abundance with and without grazing may be related to the moderate cattle stocking rate, which allows cattle to select *P. smithii* over *H. comata*, hence having strong effects on *P. smithii* across the soil texture gradient, without affecting *H. comata*.

What then might explain the high degree of spatial variation in grazing effects on plant communities that we documented? Studies of the fine-scale distribution of *B. gracilis* and *P. smithii* where they co-exist may provide

Table 1. Changes over time in plant species composition of shortgrass steppe grazed by cattle at moderate stocking rates during the growing season and in ungrazed exclosures established in 1939 at the Central Plains Experimental Range in northeast Colorado. Values represent relative canopy cover of each species. Values for 1940–1953 are derived from Klipple & Costello (1960); estimates of among-pasture ($N = 4$) and among-exclosure ($N = 12$) variation are not reported. Values for 2011 include means $\pm$ 1SE based on among-pasture variation ($N = 4$) for grazed sites and among-exclosure variation ($N = 9$) for ungrazed sites.

Growth Form	Species	Moderate Summer Grazing				Ungrazed Exclosures			
		1940–42	1946–48	1952–53	2011 ($\pm$1 SE)	1940–42	1946–48	1952–53	2011 ($\pm$1 SE)
Graminoids	*Bouteloua gracilis*	55.0	65.3	63.8	60.2 (6.8)	68.56	76.38	72.13	29.30 (8.9)
	Bouteloua dactyloides	10.9	14.1	15.0	1.5 (0.9)	5.06	5.12	5.77	0.05 (0.05)
	Pascopyrum smithii	0.71	0.58	0.22	5.0 (4.9)	1.63	1.16	0.75	28.8 (6.2)
	Carex duriuscula	0.16	0.50	0.44	8.2 (2.6)	0.08	0.50	0.75	11.4 (3.4)
	Aristida longiseta	3.07	3.31	3.62	0.82 (0.56)	1.47	1.74	3.21	0.15 (0.13)
	Hesperostipa comata	0.24	0.50	0.33	3.69 (2.73)	0.08	0.83	1.60	7.96 (2.73)
	Muhlenbergii torreyi	4.25	2.15	2.30	0.00	2.37	0.74	1.18	0.07 (0.07)
	Sporobolus cryptandrus	0.32	0.17	0.55	2.31 (1.08)	0.24	0.17	0.43	0.76 (0.27)
	Elymus elymoides	0.08	0.17	0.11	0.66 (0.30)	0.03	0.50	0.43	0.30 (0.11)
	Vulpia octoflora	0.71	0.08	0.11	0.00	0.33	0.08	0.04	0.00
	Other grass	0.98	0.89	0.86	0.00	0.15	0.12	0.32	0.03 (0.03)
Forbs	*Sphaeralcea coccinea*	1.58	0.74	0.22	2.72 (0.71)	0.73	0.99	0.04	1.74 (0.49)
	Chenopodium leptophyllum	0.71	0.03	0.04	0.74 (0.25)	1.14	0.03	0.04	1.66 (0.65)
	Cryptantha minima	2.28	0.08	0.04	0.21 (0.12)	0.57	0.03	0.04	0.00
	Plantago patagonica	0.87	0.03	0.04	0.38 (0.3)	0.73	0.03	0.11	0.02 (0.02)
	Psoralidium tenuiflorum	0.47	0.25	0.04	0.76 (0.69)	0.24	0.17	0.04	0.08 (0.06)
	Astragalus spp.	0.55	0.25	0.11	0.08 (0.07)	0.41	0.17	0.04	0.00
	Oenothera spp.	0.03	0.03	0.04	0.08 (0.05)	0.65	0.08	0.04	0.44 (0.32)
	Helianthus petiolaris	0.24	0.03	0.04	0.00	0.24	0.03	0.04	0.70 (0.62)
	Salsola tragus	0.32	0.03	0.04	0.11 (0.09)	0.16	0.03	0.11	0.34 (0.18)
	Other forbs	5.29	0.98	1.38	1.43 (1.19)	1.89	1.04	1.30	2.72 (0.70)
Cactus	*Opuntia polyacantha*	4.10	2.48	4.39	6.89 (3)	7.92	4.05	6.63	5.85 (1.62)
Sub-shrubs	*Artemisia frigida*	0.16	0.83	1.54	0.29 (0.29)	1.55	1.90	2.56	4.90 (2.97)
	Eriogonum effusum	1.65	1.41	1.21	1.88 (0.66)	0.90	0.83	0.85	0.36 (0.17)
	Gutierrezia sarothrae	0.55	0.83	0.33	0.34 (0.23)	1.14	1.32	0.21	0.15 (0.14)
	Krascheninnikovia ceratoides	0.47	0.33	0.33	0.74 (0.74)	0.03	0.03	0.04	0.24 (0.24)
Shrubs	*Atriplex canescens*	0.87	0.66	0.88	0.61 (0.61)	0.33	0.25	0.43	1.66 (0.63)
	Ericameria nauseosa	3.47	3.23	1.86	0.33 (0.33)	1.31	1.65	0.75	0.00
	Other shrubs/cactus	0.03	0.03	0.04	0.04 (0.04)	0.03	0.03	0.11	0.00 (0.25)

Table 2. Summary of temporal trends in relative abundance of 12 of the most abundant plant species in the shortgrass steppe of northeast Colorado over a 72-yr period following the extreme drought of the 1930s, and how those temporal trends are modified by the presence vs absence of cattle grazing.

Temporal Trend	Growth Form	Species	Modification by Grazing
Increasing	C$_3$ grass	*Pascopyrum smithii*	Much larger increase without grazing
Increasing	C$_3$ grass	*Hesperostipa comata*	No effect
Increasing	C$_3$ sedge	*Carex duriuscula*	No effect
Increasing	Perennial Forb	*Sphaeralcea coccinea*	Slightly larger increase with grazing
Contingent on grazing	C$_4$ grass	*Bouteloua gracilis*	Stable with grazing; llarge decline without
Contingent on grazing	C$_4$ grass	*Sporobolus cryptandrus*	Increase with grazing; stable without
Contingent on grazing	Sub-shrub	*Artemisia frigida*	Stable with grazing; Increase without
Contingent on grazing	Sub-shrub	*Eriogonum effusum*	Stable with grazing; small decline without
Stable	Cactus	*Opuntia polyacantha*	No effect
Declining	C$_4$ grass	*Aristida longiseta*	Larger decline without grazing
Declining	C$_4$ grass	*Bouteloua dactyloides*	Larger decline without grazing
Declining	Shrub	*Ericameria nauseosa*	No effect

insight (Adler & Lauenroth 2000). Under long-term grazing exclusion, *P. smithii* increases in a patchy pattern at fine spatial scales, generating discrete *P. smithii*-dominated patches in the order of 3–5-m diameter interspersed with areas retaining *B. gracilis* dominance (Adler & Lauenroth 2000). These patterns arise in part because *P. smithii*'s

rhizomatous growth form constrains the spatial pattern in which established clumps expand. Similar spatial patterns occur in desert grassland where *B. gracilis* co-exists with rhizomatous *B. eriopoda* (Collins & Xia 2015). Variation in the degree of *P. smithii* dominance at the spatial scale we studied (30 m × 30 m plots) may be influenced by the historical pattern of *P. smithii* presence on the site, and subsequent variation in its expansion at fine spatial scales (e.g. Collins & Xia 2015). These spatial patterns combined with unpredictable droughts and deluges may limit the rate at which *P. smithii* can out-compete shortgrasses and coalesce into larger, mono-dominant patches. Viewing this in a probabilistic framework, each successive period of wet years may lead to a small portion of ungrazed sites reaching the stage where smaller *P. smithii* patches coalesce and suppress *B. gracilis* in the understorey, such that many decades are required for a majority of sites to attain this state.

Grasslands worldwide are experiencing woody plant increases, which can be influenced by altered grazing regimes, changes in precipitation patterns and soil moisture dynamics, and increased atmospheric $[CO_2]$ (Scholes & Archer 1997; Bond & Midgley 2012). The shortgrass steppe has been resistant to woody plant invasions over the past century, but both rising atmospheric $[CO_2]$ and increased precipitation intensity are altering soil moisture availability in ways that enhance woody plant encroachment (Morgan et al. 2001, 2007; Heisler-White et al. 2008). A key question is whether grazing also influences woody plants. Increased *A. canescens* and *K. lanata* cover inside exclosures can be explained by the direct effects of defoliation on these palatable species, as cattle consume both leaves and flowering stems, and late summer grazing suppresses growth and reproduction (Romo et al. 1995; Cibils et al. 2003). More surprising, the sub-shrub *A. frigida*, which benefits substantially from elevated atmospheric $[CO_2]$ (Morgan et al. 2007), was five times more abundant in exclosures than grazed plots. Given that this species is infrequently consumed by cattle (Klipple & Costello 1960; Milchunas et al. 2008), its increase with grazing exclusion may be related to indirect effects mediated through moisture and nutrient availability. Overall, our results suggest that livestock grazing enhances the dominance of C_4 graminoids in this ecosystem, and that efforts to predict how community composition will respond to changing climate drivers need to incorporate the effect of grazing.

Temporal trends in relation to grazing

Shortgrass steppe communities are recognized as being highly resistant to impacts of drought and grazing, but previous studies of temporal compositional dynamics focused historically on ploughed sites (i.e. secondary succession; Coffin et al. 1996). Our findings show that over the past 72 yr, trends in native (unploughed) shortgrass steppe are contingent on grazing for some species, but for others have occurred independent of grazing. Most notably, all of the C_3 perennial graminoids have increased more than five-fold in the presence of continuous, growing season cattle grazing at moderate stocking rates. One possible interpretation is that C_3 perennial graminoids were reduced to exceptionally low abundance by extreme drought conditions, dust storms and soil movement during the 1930s, and are slowly recovering back to pre-dust bowl conditions. C_3 graminoids and *A. frigida* may also be responding to increasing atmospheric $[CO_2]$, as suggested by manipulative experiments in shortgrass steppe (Morgan et al. 2001, 2007), an effect that could be accentuated by grazing removal. These explanations (drought recovery and elevated atmospheric $[CO_2]$) are not mutually exclusive. Increasing atmospheric N deposition is another potential driver of plant community change, but N deposition in the rural landscape of our region remains low (~5 $kg \cdot ha^{-1} \cdot yr^{-1}$; Burke et al. 2008) and N addition experiments have not enhanced C_3 perennial grasses (Milchunas et al. 2008).

Species that declined over time (Table 2) were mid-successional species that temporarily colonize sites with lower *B. gracilis* dominance, including a stoloniferous grass (*B. dactyloides*), a bunchgrass (*A. longiseta*) and a shrub (*E. nauseosa*; Costello 1944; Coffin et al. 1996; Grantz et al. 1998). Their decline is consistent with the idea that plant communities in the 1940s were impacted by drought and soil movement during the prior decade, from which they have recovered over the past 72 yr. Species that were stable over time and unaffected by grazing included one generally not eaten by cattle (the cactus *O. polyacantha*) and one that is highly palatable to cattle (the perennial forb, *Sphaeralcea coccinea*; Milchunas et al. 2008). These patterns differ from successional dynamics on ploughed fields (Coffin et al. 1996), and also show that variation among species in palatability to cattle is not sufficient to predict responses to grazing exclusion.

In contrast to short-term (one to two decade) studies in this semi-arid grassland, our findings show that long-term grazing exclusion induces substantial plant community change. Grazing exclusion increased abundance of *P. smithii*, ruderals and exotics, while communities subjected to grazing were dominated by *B. gracilis* with a diverse set of co-existing sub-dominants. Contrary to our predictions, grazing effects were not contingent on soil texture, which had independent effects on community composition. This study highlights the importance of long-term experiments to capture slow plant community shifts, both in response to sustained experimental treatments and in

response to external drivers such as historical disturbance events and increasing atmospheric [CO_2].

Acknowledgements

References

Adler, P. & Lauenroth, W. 2000. Livestock exclusion increases the spatial heterogeneity of vegetation in Colorado shortgrass steppe. *Applied Vegetation Science* 3: 213–222.

Agrawal, A.A. 2007. Macroevolution of plant defense strategies. *Trends in Ecology & Evolution* 22: 103–109.

Bond, W.J. & Midgley, G.F. 2012. Carbon dioxide and the uneasy interactions of trees and savannah grasses. *Philosophical Transactions of the Royal Society, Series B: Biological Sciences* 367: 601–612.

Bouyoucos, G.J. 1962. Hydrometer method improved for making particle size analyses of soils. *Agronomy Journal* 54: 464–465.

Burke, I.C., et al. 2008. Soil organic matter and nutrient dynamics of shortgrass steppe ecosystems. In: Lauenroth, W. & Burke, I.C. (eds.) *Ecology of the shortgrass steppe: a long-term perspective*, pp. 389–446. Oxford University Press, New York, NY, US.

Cibils, A.F., Swift, D.M. & Hart, R.H. 2003. Changes in shrub fecundity in fourwing saltbush browsed by cattle. *Journal of Range Management* 56: 39–46.

Coffin, D.P., Lauenroth, W.K. & Burke, I.C. 1996. Recovery of vegetation in a semiarid grassland 53 years after disturbance. *Ecological Applications* 6: 538–555.

Collins, S.L. & Xia, Y. 2015. Long-term dynamics and hotspots of change in a desert grassland plant community. *The American Naturalist* 185: E30–E45.

Costello, D.F. 1944. Important species of the major forage types in Colorado and Wyoming. *Ecological Monographs* 14: 107–134.

Diaz, S., Lavorel, S., McIntyre, S., Falczuk, V., Casanoves, F., Milchunas, D.G., Skarpe, C., Rusch, G., Sternberg, M., (...) & Campbell, B.D. 2007. Plant trait responses to grazing – A global synthesis. *Global Change Biology* 13: 313–341.

Díaz, S., Noy-Meir, I. & Cabido, M. 2001. Can grazing response of herbaceous plants be predicted from simple vegetative traits? *Journal of Applied Ecology* 38: 497–508.

Fuhlendorf, S. & Smeins, F. 1999. Scaling effects of grazing in a semi-arid grassland. *Journal of Vegetation Science* 10: 731–738.

Grantz, D.A., Vaughn, D.L., Farber, R., Kim, B., Zeldin, M., Van-Curen, T. & Campbell, R. 1998. Seeding native plants to restore desert farmland and mitigate fugitive dust and PM_{10}. *Journal of Environmental Quality* 27: 1209–1218.

Hart, R.H. & Ashby, M.M. 1998. Grazing intensities, vegetation, and heifer gains: 55 years on shortgrass. *Journal of Range Management* 51: 392–398.

Heisler-White, J.L., Knapp, A.K. & Kelly, E.F. 2008. Increasing precipitation event size increases aboveground net primary productivity in a semi-arid grassland. *Oecologia* 158: 129–140.

Hickman, K.R. & Derner, J.D. 2007. Blackland tallgrass prairie vegetation dynamics following cessation of herbicide application. *Rangeland Ecology and Management* 60: 186–190.

Holechek, J.L., Estell, R.E., Galyean, M.L. & Richards, W. 1989. Chemical composition, *in vitro* digestibility and *in vitro* VFA concentrations of New Mexico native forages. *Grass and Forage Science* 44: 101–105.

Holechek, J.L., Galt, D. & Khumalo, G. 2006. Grazing and grazing exclusion effects on New Mexico shortgrass prairie. *Rangeland Ecology and Management* 59: 655–659.

Hyder, D., Bement, R., Remmenga, E. & Terwilliger, C. 1966. Vegetation–soils and vegetation–grazing relations from frequency data. *Journal of Range Management* 19: 11–17.

Kelly, E.F., Yonkers, C.M., Blecker, S.W. & Olson, C.G. 2008. Soil development and distribution in the shortgrass steppe ecosystem. In: Lauenroth, W.K. & Burke, I.C. (eds.) *Ecology of the shortgrass steppe: a long-term perspective*, pp. 30–54. Oxford University Press, New York, NY, US.

Klipple, G.E. & Costello, D.F. 1960. *Vegetation and cattle responses to different intensities of grazing on short-grass ranges on the central Great Plains. Technical Bulletin No 1216.* US Government Printing Office, Washington, DC, US.

Lauenroth, W.K. 2008. Vegetation of the shortgrass steppe. In: Lauenroth, W.K. & Burke, I.C. (eds.) *Ecology of the shortgrass steppe: a long-term perspective*, pp. 70–83. Oxford University Press, New York, NY, US.

Lauenroth, W.K., Burke, I.C. & Gutmann, M.P. 1999. The structure and function of ecosystems in the central North American grassland region. *Great Plains Research* 9: 223–260.

Liang, Y., Hazlett, D. & Lauenroth, W.K. 1989. Biomass dynamics and water use efficiencies of five plant communities in the shortgrass steppe. *Oecologia* 80: 148–153.

McNaughton, S.J., Oesterheld, M., Frank, D.A. & Williams, K.J. 1991. Primary and secondary production in terrestrial ecosystems. In: Cole, J., Lovett, G. & Findlay, S. (eds.) *Comparative analyses of ecosystems*, pp. 121–139. Springer, New York, NY, US.

Milchunas, D.G. & Lauenroth, W.K. 1993. Quantitative effects of grazing on vegetation and soils over a global range of environments. *Ecological Monographs* 63: 327–366.

Milchunas, D.G., Lauenroth, W.K., Chapman, P.L. & Kazempour, M.K. 1989. Effects of grazing, topography and precipitation on the structure of a semiarid grassland. *Vegetatio* 80: 11–23.

Milchunas, D., Lauenroth, W., Chapman, P. & Kazempour, M. 1990. Community attributes along a perturbation gradient in a shortgrass steppe. *Journal of Vegetation Science* 1: 375–384.

Milchunas, D.G., Lauenroth, W.K. & Chapman, P.L. 1992. Plant competition, abiotic, and long- and short-term effects of large herbivores on demography of opportunistic species in a semiarid grassland. *Oecologia* 92: 520–531.

Milchunas, D.G., Lauenroth, W.K., Burke, I.C. & Detling, J. 2008. Effects of grazing on vegetation. In: Lauenroth, W.K. & Burke, I.C. (eds.) *Ecology of the shortgrass steppe: a long-term perspective*, pp. 389–446. Oxford University Press, New York, NY, US.

Morgan, J.A., Lecain, D.R., Mosier, A.R. & Milchunas, D.G. 2001. Elevated CO_2 enhances water relations and productivity and affects gas exchange in C3 and C4 grasses of the Colorado shortgrass steppe. *Global Change Biology* 7: 451–466.

Morgan, J.A., Milchunas, D.G., LeCain, D.R., West, M. & Mosier, A.R. 2007. Carbon dioxide enrichment alters plant community structure and accelerates shrub growth in the shortgrass steppe. *Proceedings of the National Academy of Sciences of the United States of America* 104: 14724–14729.

Noy-Meir, I. 1973. Desert ecosystems: environment and producers. *Annual Review of Ecology and Systematics* 4: 23–51.

Quiroga, R., Golluscio, R., Blanco, L. & Fernandez, R. 2010. Aridity and grazing as convergent selective forces: an experiment with an arid Chaco bunchgrass. *Ecological Applications* 20: 1876–1889.

Romo, J.T., Redmann, R.E., Kowalenko, B.L. & Nicholson, A.R. 1995. Growth of winterfat following defoliation in northern mixed prairie of Saskatchewan. *Journal of Range Management* 48: 240–245.

Sala, O., Parton, W., Joyce, L. & Lauenroth, W. 1988. Primary production of the central grassland region of the United States. *Ecology* 69: 40–45.

Scholes, R. & Archer, S. 1997. Tree-grass interactions in savannas. *Annual Review of Ecology and Systematics* 28: 517–544.

Whittaker, R.H. 1952. A study of summer foliage insect communities in the Great Smoky Mountains. *Ecological Monographs* 22: 1–44.

Supporting Information

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

Appendix S1. Effects of 72 yr of grazing exclusion on basal cover of 17 plant species measured in 2011 in shortgrass steppe of northeast Colorado.

Appendix S2. Effects of 72 yr of grazing exclusion on canopy cover of six woody plant species measured in 2011 in shortgrass steppe of northeast Colorado.

Appendix S3. Photographs of the contrast between a long-term ungrazed and long-term moderately grazed community in shortgrass steppe of northeast Colorado.