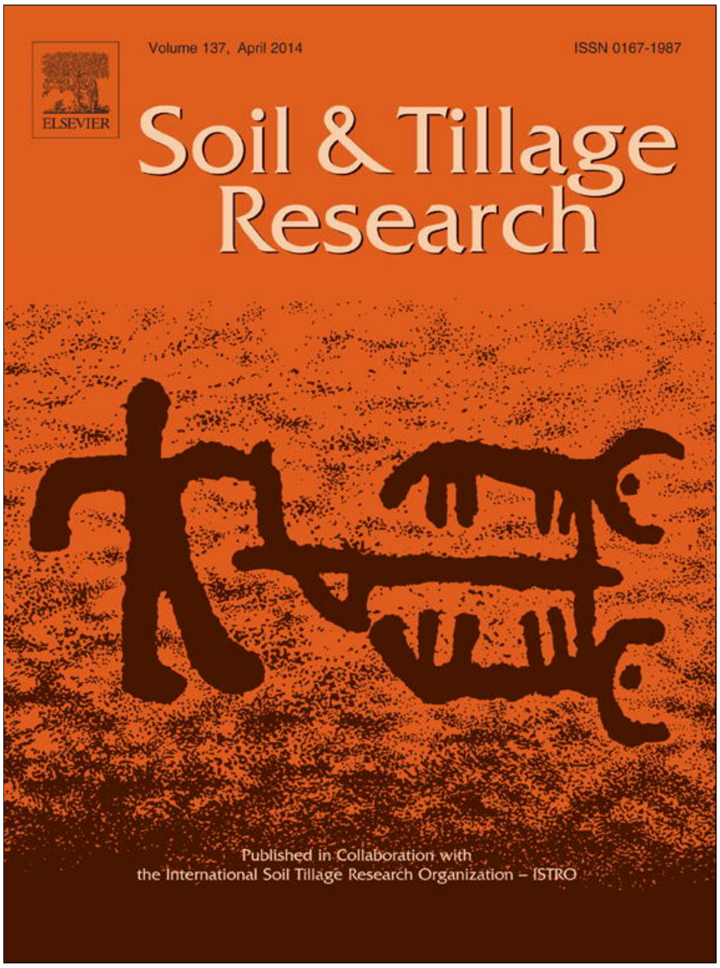
Volume 137, April 2014
ISSN 0167-1987
ELSEVIER
Soil & Tillage Research
Published in Collaboration with
the International Soil Tillage Research Organization – ISTRO

Soil & Tillage Research 137 (2014) 7–15

Contents lists available at ScienceDirect

Soil & Tillage Research

journal homepage: www.elsevier.com/locate/still

Long-term fencing effects on plant diversity and soil properties in China

Lei Deng, Zhinan Zhang, Zhouping Shangguan *

State Key Laboratory of Soil Erosion and Dryland Farming on the Loess Plateau, Northwest A&F University, Yangling, Shaanxi 712100, PR China

ARTICLE INFO

Article history:
Received 21 July 2013
Received in revised form 19 October 2013
Accepted 5 November 2013

Keywords:
Fencing
Grazing
Community
Soil
Temperate grassland

ABSTRACT

Overgrazing reduces plant species diversity, productivity and soil C and N storage due to degradation especially in arid and semi-arid ecosystems. We hypothesized that fencing could significantly reverse these trends in temperate grasslands. The effects of long-term (30 years) fencing on diversity and soil C and N storage were compared with areas where continuous grazing occurred on the Loess Plateau, China. Fencing increased vegetation coverage, height, plant diversity, biomass production and litter, resulting primarily from increases in the ratio of grass species as a percentage of the whole community and photosynthate allocation between above- and below-ground biomass indicated by differences in the root/shoot (R/S) ratios. Fencing significantly influenced soil bulk density (BD), moisture content (SW) and pH. Long-term fencing also led to marked increases in soil organic carbon (SOC), soil total nitrogen (TN), the carbon: phosphorus (C/P) and nitrogen: phosphorus (N/P) ratios, as well as soil C and N storage within 0–100 cm soil profile. The C/N ratio in the surface 0–5 cm fenced and grazed grasslands were also significantly different. Increases in soil C and N sequestration as a result of fencing occurred mainly at deeper soil depths (30–100 cm). These findings have important implications for both protecting and enhancing the resilience of ecosystems, which have been disturbed by grazing and for developing a more effective grasslands management strategy on the Loess Plateau.

1. Introduction

Globally, overgrazing by livestock is one of the most important human induced causes of arid and semiarid grasslands degradation (Su et al., 2005; Liang et al., 2009; Schönbach et al., 2011), lowering both the productivity and resilience of grasslands (Li et al., 2009; Zhou et al., 2011). The effect of overgrazing on the plant community and on soil resources are considered destructive because it reduces vegetation cover (Su et al., 2005; Wu et al., 2010a; Zhou et al., 2011; Louhaichi et al., 2012), increases undesirable species (Louhaichi et al., 2009), reduces species diversity (Li et al., 2006), destroys soil structure (Reynolds et al., 2003), and compacts soil as a direct result of trampling (Manzano and Návar, 2000). These processes increase soil crusting, reduce soil infiltration, and enhance susceptibility to soil erosion (Manzano and Návar, 2000). Therefore, developing ecosystem rehabilitation strategies for severely degraded grasslands is crucial (Li et al., 2009) at a global level.

Degraded grasslands have the capacity for self-recovery if the disturbance ceases for an extended length of time allowing for natural succession (Li et al., 2009). Recovery conditions offer the best overall strategy to restore diversity, ecosystem function and resilience (Palik et al., 2000; Wu et al., 2009, 2010a,b; Medina-Roldán et al., 2012; Bach et al., 2012; Deng et al., 2013a). Grassland management significantly influences plant density and composition, above- and belowground vegetation characteristics and soil properties (Deng et al., 2013a). Fencing is the most common management tool used to reverse grassland degradation throughout the world (Shrestha and Stahla, 2008; Wu et al., 2009, 2010a; Golodets et al., 2010).

Recent research has focused on the effects of fencing on vegetation succession, plant diversity, community structure and productivity (Smith et al., 2000; Gibson et al., 2001; Wu et al., 2009; Golodets et al., 2010; Deng et al., 2013a). Studies have also focused on the effects of fencing on soil nutrients (Mohr and Topp, 2005; Su et al., 2005; Miller et al., 2010; Wu et al., 2010b), soil microbial structure (Stark et al., 2000; Su et al., 2004; Huang et al., 2011), soil enzyme activities (Su et al., 2004), soil C and N cycling (Frank and Groffman, 1998; Stark et al., 2000) and storage

* Corresponding author at: State Key Laboratory of Soil Erosion and Dryland Farming on the Loess Plateau, Northwest A&F University, No. 26 Xinong Road, Yangling, Shaanxi 712100, China. Tel.: +86 29 87019107; fax: +86 29 87012210.
E-mail address: shangguan@ms.iswc.ac.cn (Z. Shangguan).

0167-1987/$ – see front matter © 2013 Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.still.2013.11.002

(Shrestha and Stahla, 2008; Zhou et al., 2011; Medina-Roldán et al., 2012). However, few studies have focused on the effects of long-term fencing on overall plant community composition, diversity and productivity, and other soil properties.

In China, the of the Loess Plateau (~40 million ha) is known for its complex terrain, extreme drought conditions and severe soil erosion (Liu et al., 2007), a result of a combination of overgrazing, the intensification of cultivation and other unsustainable land use practices (Zhou et al., 2006). Grazing exclusion through fencing is regarded as the most effective method available for restoring the ecology of the Loess Plateau (Wu et al., 2010a). Grassland restoration is a long-term and complex ecological process (Hastings et al., 2007).

The Yunwu Mountain reserve, excluded from grazing since 1982, is located in the only remaining grassland region of the Loess Plateau where long-term (30 years) fencing has made it possible to better understand the effect of grazing exclusion on both grassland vegetation and soil properties. Our objectives were to investigate the effects of long-term exclusion on: (1) plant community properties; (2) soil properties; and (3) soil C and N storage. This study contributes to our understanding of the restoration of plant production and soil C and N sequestration in degraded grasslands on the Loess Plateau.

2. Materials and methods

2.1. Study area

The study area is located in the Yunwu Mountain reserve for Vegetation Protection and Eco-environment, Ningxia Province, China (106°16′–106°24′E, 36°13′–36°19′N, 1700–2148 m a.s.l.) (Fig. 1). It is a hilly landscape in the middle of the Loess Plateau with deeply incised gullies and is characterized by a sub-arid climate within the mid-temperate zone. The Yunwu Mountain reserve is a century old protected grassland area. The 4000 ha reserve and most of the surrounding land lies between 1700 and 2000 m in altitude is closely dissected by steep and very steep gulleys. The vegetation is temperate grassland with the primary plant species being herbaceous plants (*i.e.*, *Androsace erecta*, *Artemisia capillaries*, *Artemisia frigid*, *Artemisia sacrorum*, *Heteropappus altaicus*, *Lespedeza davurica*, *Potentilla acaulis*, *Stipa bungeana*, *Stipa grandis*, *Thymus mongolicus*, *etc.*) of which the *Stipa bungeana* community has the most extensive distribution.

The reserve includes three areas: core, buffer, and experimental (Fig. 1), with comparatively similar geographical patterns. The core area has approximately 1000 ha that are totally enclosed and account for 25% of the total area. The buffer area covers 1200 ha and also accounts for approximately 25% of the total area. The role of buffer area is to prevent the effect of human activities on the core area. Outside the core and buffer areas is the experimental area, which encompasses approximately 1800 ha and accounts for 45% of the total area. In the experimental area, crop agriculture and animal husbandry are practiced. No fertilizer is applied as the three areas abut one another. The core area was fenced in 1982 and remains so today.

Before enclosure, the permanent grasslands were used as grazing land. Both the grazed and fencing grassland had similar initial conditions (slope degree, slope direction, topography and altitude) (Qiu et al., 2013), allowing the existing grazed grassland to be used as a control to compare the effects of exclusion on plant diversity and soil properties. In the core area the *Stipa bungeana* community is the most extensive; *Stipa grandis* and *Stipa bungeana* are the dominant grass species, and *Thymus mongolicus* and *Artemisia sacrorum* the dominant forb species. In contrast, freely grazed areas are characterized by a degraded *Stipa bungeana* community. In the grazed grasslands, *Stipa grandis* and *Stipa bungeana* are again the dominant grass species, but *Artemisia capillaries* and *Artemisia frigid* are the dominant forb species. The principle leguminous plant, *Lespedeza davurica*, occurs in both areas.

The study area's soil type is Aeolian soil (silt loam) with a soil pH ranging from 8.0 to 8.6. The area receives a mean annual precipitation of ~410 mm (1960–2010) of which a quarter falls during July and the remainder by September. The area's semi-arid temperate continental monsoon climate produces a mean annual temperature at 6.7 °C (1960–2010), a mean annual total of 2518 sunshine hours, a mean annual evaporation of 1600 mm, and 137 frost-free days per year on average.

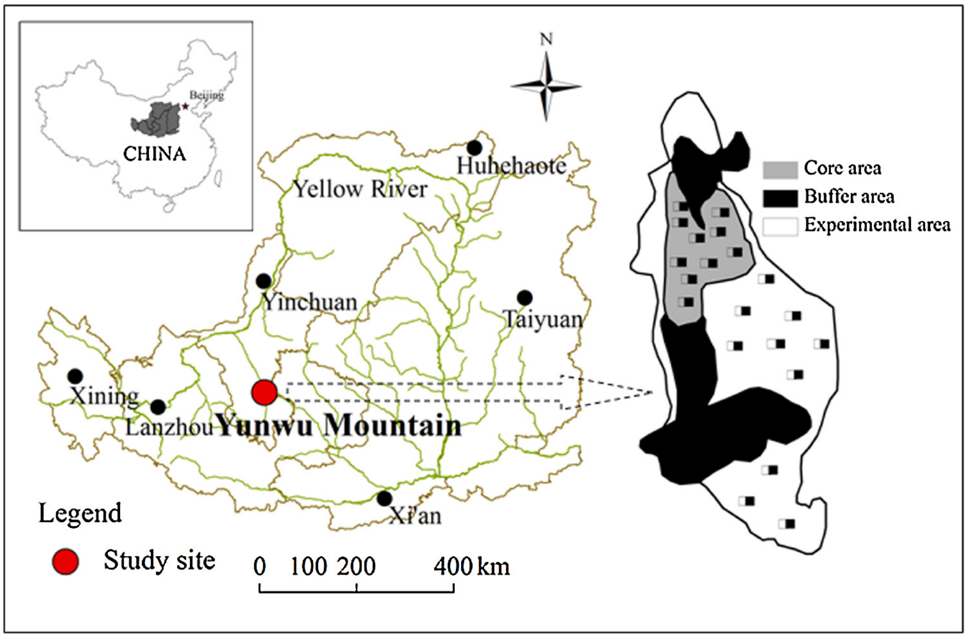

Fig. 1. Location of the Yunwu Observatory on the Loess Plateau. *Note*: the white quadrats in which individual species are divided, and the whole quadrats' species composition and height, density (number of individuals per square meter) and aboveground biomass of individual species are investigated; the black quadrats in which the individual species are undivided, and the whole quadrats' community above- and belowground biomass, canopy coverage and height are investigated.

2.2. Sampling and measurements

2.2.1. Experimental design

A temperate grassland dominated by *Stipa bungeana* and *Stipa grandis*, but including grasses and some forbs, was selected for this study in mid-August, 2011, when biomass had reached its peak. Using the line transect method we randomly selected ten 20 m × 20 m blocks in each of the grazed and fenced portions of the experimental and core areas giving us 20 blocks for the study (Fig. 1). Within the center of each block, we established two parallel 1 m × 1 m quadrats. One quadrat (the white blocks in Fig. 1) was divided (DQ) so that individual species composition, height, and density (number of individuals per square meter) in the aboveground biomass could be monitored. The other quadrat (the black quadrats in Fig. 1) was undivided (UQ) and investigated as a whole community for above- and belowground biomass, litter, canopy coverage and height, and soil properties.

2.2.2. Biomass measurement

In the DQ quadrats, all green, aboveground plant parts for each individual species, as well as all litter, were cut, collected, and put into envelops and tagged. For the UQ quadrats, the aboveground parts of all green plants and litter from the entire quadrat were sampled in the same way. To measure the belowground biomass, a 9 cm diameter root augur was used to take three soil samples for each depth of 0–5, 5–10, 10–20, 20–30, 30–50, 50–70, 70–100 cm. Samples taken at the same layer were then mixed to create a single sample. A 2 mm sieve was used to isolate the majority of plant roots from each sample. The remaining fine roots were collected by spreading the samples in shallow trays, overfilling them with water and allowing the outflow from the trays to pass through a 0.5 mm mesh sieve. No attempt was made to distinguish between living and dead roots. The isolated roots were oven-dried at 65 °C and weighed. Due to the large size of the aboveground biomass samples, each was weighed fresh and then a portion of each sample was dried and weighed. The total above-ground biomass of the samples was calculated by multiplying the ratio of the dry weight/fresh weight ratio within the sub-sample by the entire fresh weight.

2.2.3. Plant species identification, functional group and species diversity index

Plant species identification was done *in situ*. Unidentified specimens were collected and dried with a plant press and later identified by plant taxonomists. The Allen et al. (2011) method was used to divide the plants into one of three functional groups: grass (plant species of the *Poaceae* family), forb (any herbaceous, dicotyledonous broad-leaved plant) and leguminous (leguminous species group). The purpose behind forming functional groups was to represent the ecological structure of the flora, and then to use that structure to predict a general level of species assemblage (Santiago do Vale et al., 2010).

Species richness is the number of species in each quadrat (Stirling and Wilsey, 2001). The richness index (R), Shannon–Wiener diversity index (H) and Evenness index (E) of the grazed and fenced grassland communities were calculated as the:

Richness index (R):

$$R = S \tag{1}$$

Shannon–Wiener diversity index (H):

$$H = -\sum_{i=1}^{s} (Pi \ln Pi) \tag{2}$$

and

Evenness index (E):

$$E = \frac{H}{\ln S} \tag{3}$$

where S is the total species numbers of the grassland community, H is the Shannon–Wiener diversity index and Pi is the density proportion of i species.

2.2.4. Soil sampling and determination

In the UQ quadrats soil samples were taken at three points – the other two corners and the center, along that diagonal on which root samples had not been collected. The soil samples, collected and mixed in seven increments as described, were passed through a 2 mm screen to remove roots and other debris. Each sample was air-dried and stored at room temperature until soil physical and chemical properties could be determined. Soil BD (g cm^{-3}) of each soil layer (0–5, 10–20, 30–50, 70–100 cm) was measured using a soil bulk sampler with a 5 cm diameter and a 5 cm high stainless steel cutting ring (3 replicates) at points adjacent to where soil samples had been collected for chemical analysis. The original volume of each soil core and its dry mass after oven-drying at 105 °C were measured.

Soil water content was measured gravimetrically and expressed as a percentage of soil water to dry soil weight. Soil pH was determined using the method of acidity agent (soil–water ratio of 1:5) (PHS-3C pH acidometer, China). Soil BD was calculated depending on the inner diameter of the core sampler, sampling depth and the oven dried weight of the composite soil samples. Soil OC was assayed by dichromate oxidation (Nelson and Sommers, 1982), TN using the Kjeldahl method (Bremner, 1996), and TP after digestion of soil with $HClO_4$–H_2SO_4 (Parkinson and Allen, 1975). Each analysis was done in two replicates.

2.3. Calculation of soil C and N storage

There were no coarse soil fractions (*i.e.*, >2 mm) so coarse fragment values were not included in the formula used to calculate soil organic C storage (Cs) according to Eq. (4) (Guo and Gifford, 2002):

$$\text{Cs} = \frac{\text{BD} \times \text{SOC} \times D}{10} \tag{4}$$

where Cs is SOC storage (Mg ha^{-1}); BD is soil bulk density (g cm^{-3}); SOC is soil organic carbon concentration (g kg^{-1}); and D is soil thickness (cm).

Eq. (5) was used to calculate soil N storage (Ns) (Rytter, 2012):

$$\text{Ns} = \frac{\text{BD} \times \text{TN} \times D}{10} \tag{5}$$

where, Ns is soil N storage (Mg ha^{-1}); BD is soil bulk density (g cm^{-3}); TN is soil TN concentration (g kg^{-1}); and D is soil thickness (cm).

2.4. Statistical analysis

All data were expressed as mean ± standard error (SE) of mean in ten blocks. One-way ANOVA was performed to test for differences in biomass, functional group composition, plant diversity, soil properties, soil C and N storage, soil organic carbon storage between fenced and grazed meadows to assess the effects of long-term exclusion on above- and belowground properties and soil C and N storage. Significant differences were evaluated at the 0.05 level. All statistical analyses were performed using the software program SPSS, ver. 17.0 (SPSS Inc., Chicago, IL, USA).

3. Results and discussion

3.1. Grassland community coverage, height and density

In this study, fencing had a positive effect on above- and belowground, litter, and total vegetation biomass. ANOVA showed that fenced grasslands had greater total plant coverage ($P < 0.001$) and height ($P < 0.001$) when compared to grazed grassland (Fig. 2a and b). Excluding grazing for 30 years increased total coverage and height by 6.9 and 168%, respectively. Plant density showed no significant difference between the fenced grassland and grazed areas ($P > 0.05$) (Fig. 2c).

Palatable grasses have greater competitive ability than unpalatable grasses and show a marked increase in abundance in grasslands where livestock are excluded for ten or more years (Gallego et al., 2004; Wu et al., 2009). Fenced grasslands had higher species richness (R) ($P < 0.05$) than grazed areas (Fig. 2d), but the Shannon–Wiener diversity index (S) showed no significant difference between the two ($P > 0.05$) (Fig. 2e). The evenness index (E) in fenced grassland was also lower ($P < 0.01$) (Fig. 2f) confirming that excluding grazing simultaneously increases species richness and decreases community evenness.

3.2. Grassland community biomass

The fenced grasslands had greater aboveground biomass ($P < 0.001$), belowground biomass ($P < 0.05$), total biomass ($P < 0.001$), and litter biomass ($P < 0.01$) than did grazed areas, but the root/shoot (R/S) ratios were lower ($P < 0.001$) (Fig. 3) confirming that grazing increases the biomass allocation ratio of the belowground vegetation (Wang et al., 2010). Excluding grazing increased grass density ($P < 0.01$) but reduced that of forbs ($P < 0.01$) (Fig. 4a and b). Although there was no significant difference between the sites (Fig. 4c) the larger sporadic presence of legumes in the fenced grasslands may indicate the occurrence of an ecological shift or some type of ecological adaptation.

Overall, fencing increased aboveground biomass, belowground biomass, total biomass, and litter by 172, 39, 81 and 47%, respectively but decreased the R/S ratio by 47.1%. The fraction of grass biomass increased from 47 to 81%, while that of forbs decreased from 53 to 19% (Fig. 4a and b).

3.3. Soil physical and chemical properties

Long-term fencing has significant affects on pH value, soil bulk density, and soil moisture (Wu et al., 2010a). In this study

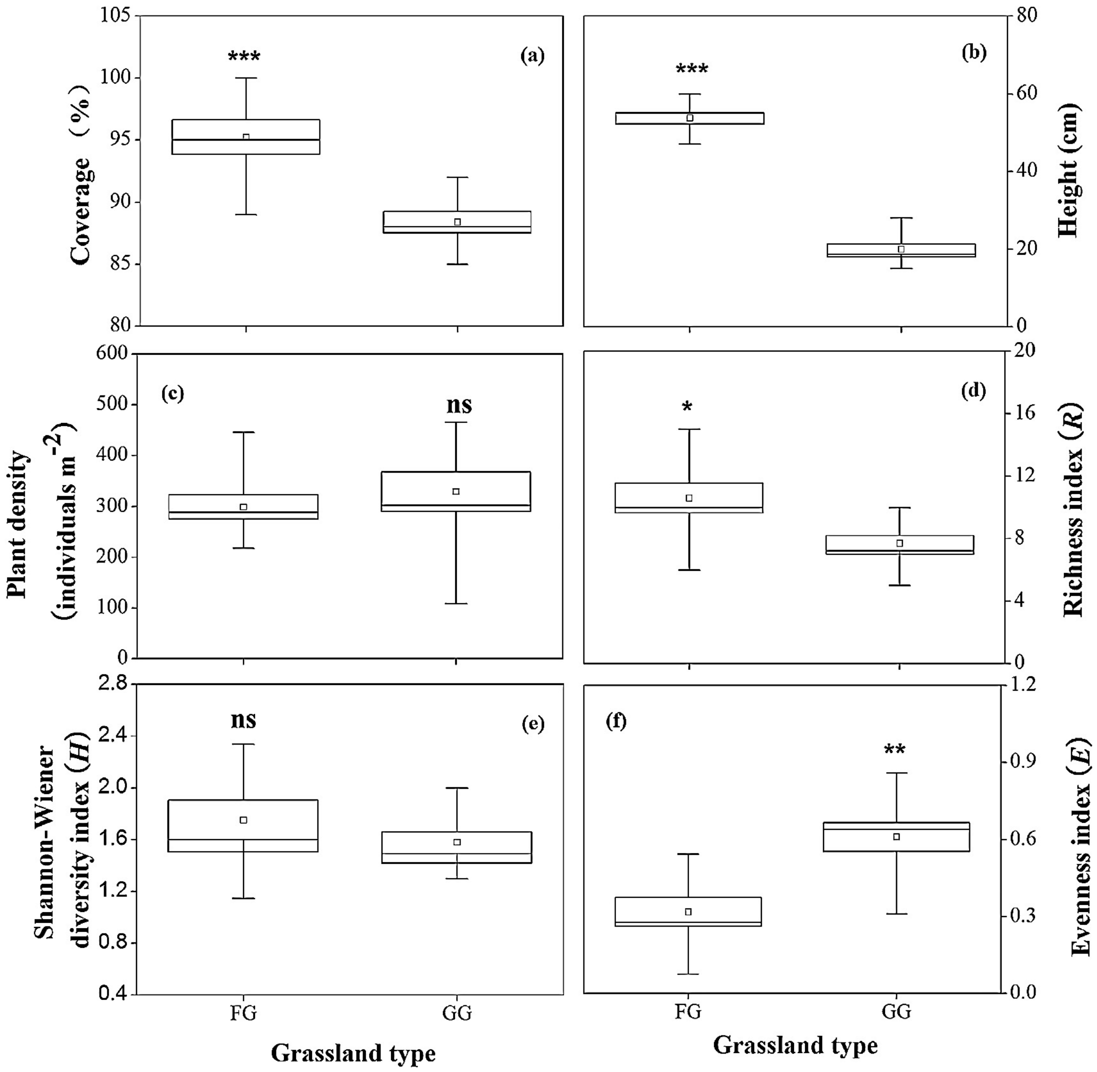

Fig. 2. Effect of fencing (FG) and grazing (GG) on coverage (a), height (b), plant density (c), richness index (d), Shannon–Wiener diversity index (e) and evenness index (f) of the temperate grassland community. The box plot introduction: the box is ±SE, the whisker is min–max, the small square is mean values, and the horizontal line is the median. Significant difference between fenced and grazed meadows are indicated by symbols, ***$P < 0.001$, **$P < 0.01$, *$P < 0.05$; ns, no significant difference.

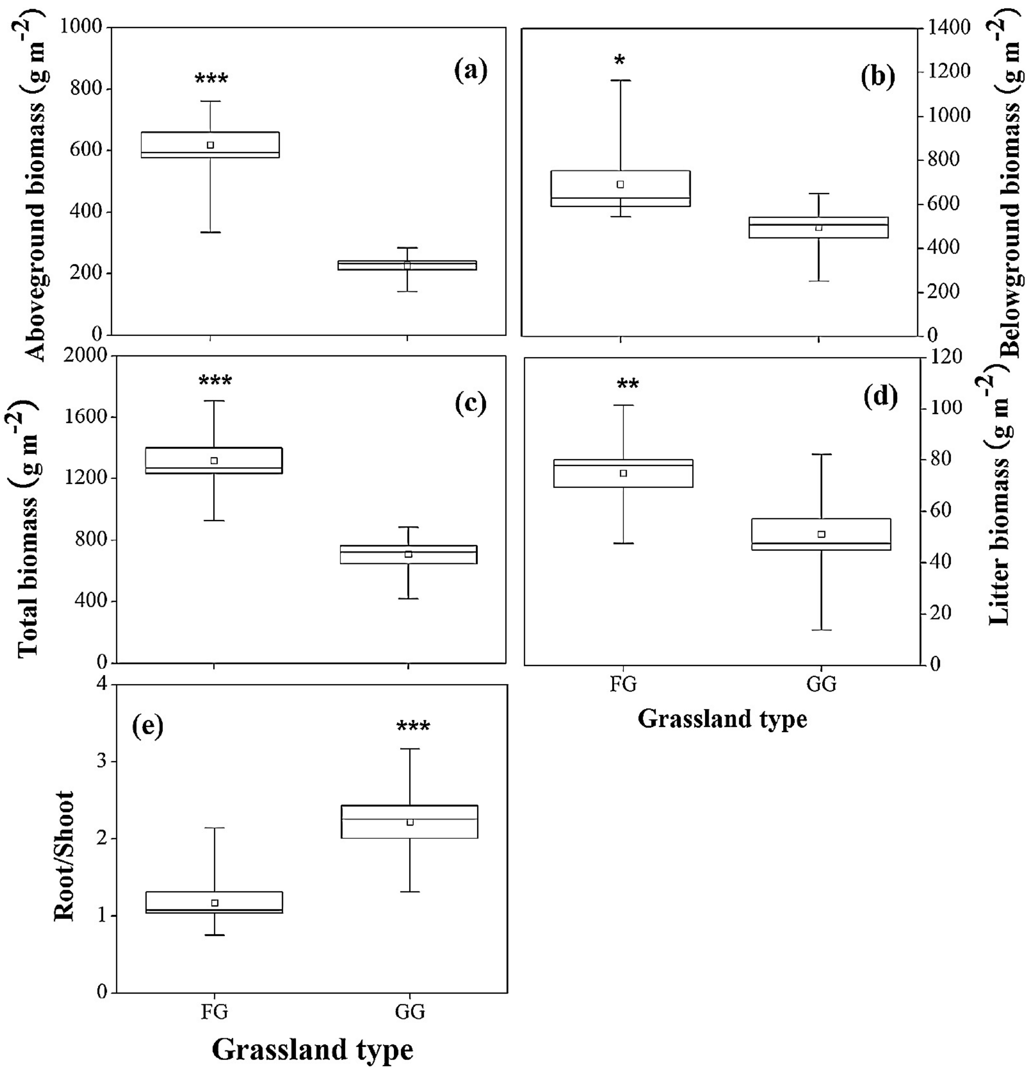

Fig. 3. Effect of fencing (FG) and grazing (GG) on aboveground (a), belowground (b), total (c), litter biomass (d), and root/shoot ratio (e) of the temperate grassland community. The box plot introduction: the box is ±SE, the whisker is min–max, the small square is mean values, and the horizontal line is the median. Significant difference between fenced and grazed meadows are indicated by symbols, ***$P < 0.001$, **$P < 0.01$, *$P < 0.05$.

long-term fencing significantly altered soil properties. Soil BD ($P < 0.01$) and pH ($P < 0.05$) were both reduced compared with grazed areas (Fig. 5a and c). Reduced trampling by fencing may have resulted in a reduction in bulk density. Grassland restoration has improved the content of phospholipid fatty acid (PLFA) in soils (Bach et al., 2012), and PLFA had significant negative correlation with soil pH ($P < 0.05$) (Huang et al., 2011), meaning that vegetation recovery reduced soil pH. Long-term fencing did increase soil water content, but the difference was only significant at the 70–100 cm soil depth ($P < 0.05$) (Fig. 5b).

Fig. 5 There was no difference in surface (0–10 cm) C and N storage between the fenced and grazed areas ($P > 0.05$), but both were greater in the underlying soil depths (10–100 cm) ($P < 0.001$) (Fig. 6ab). In addition, fencing significantly increased soil C and N storage in soil depths of 20 to 100 cm ($P < 0.05$) (Fig. 6c and d). According to the amount of soil C and N sequestration over 30 years, we estimated that the annual mean sequestration rates for C and N were 3.00 and 0.32 Mg ha^{-1} year^{-1} in the 0–100 cm soil depth under grazing exclusion. Soil C and N sequestration values increased in increments of 5 cm in the 70–100 cm soil depth which had, as a whole, the highest values within the entire 0–100 cm soil profile (Fig. 7).

Many studies have found that vegetation recovery improves SOC and TN content (Su et al., 2004, 2005; Mohr and Topp, 2005; Shrestha and Stahla, 2008; Miller et al., 2010; Wu et al., 2010a, 2010b; Zhou et al., 2011; Medina-Roldán et al., 2012). SOC and TN show similar trends under fencing (Fig. 5d and e), indicating that SOC dynamics are closely coupled with TN dynamics which may be a reason why there was no difference in the C/N ratios of the underlying soils when comparing fenced and grazed grasslands (Fig. 5g). Fencing significantly increased SOC ($P < 0.001$) and TN ($P < 0.01$) in the 0–100 cm soil depth (Fig. 5c and d). Topsoil P (TP) was significant ($P > 0.05$), in the top 50 cm on fenced grassland, but had lower TP at deeper depths (50–100 cm) ($P < 0.05$). Apart from the 0–5 cm surface layer ($P < 0.05$), soil C/N showed no significant difference between fenced and grazed grasslands (5–100 cm) ($P > 0.05$) (Fig. 5g) mainly because fencing had more effects on soil carbon accumulation ($P < 0.001$) than soil TN ($P < 0.01$) in the surface soil (Fig. 5g). In addition, due to SOC and TN significantly increased (Fig. 5d and e) long-term fencing had greater soil C/P and N/P in comparison to the grazed grasslands ($P < 0.01$). Fenced areas did have greater soil C/P and N/P ratios than grazed areas ($P < 0.01$) (Fig. 5h and i).

If the plant community has an affect on soil processes, then soil processes should correlate with plant dynamics (Li et al., 2009).

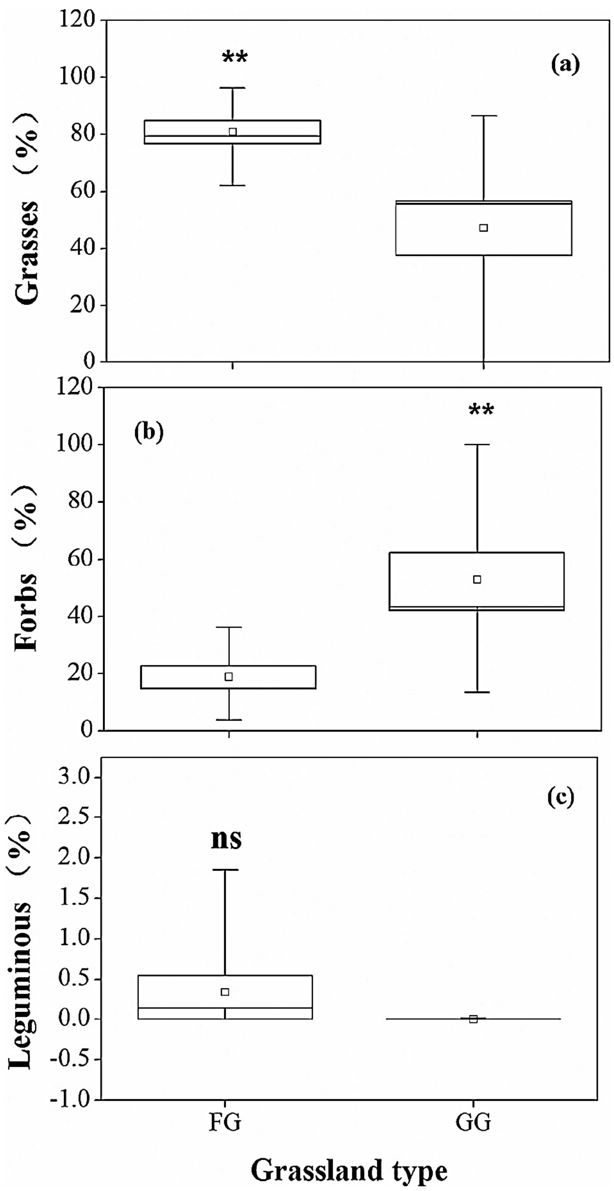

Fig. 4. Effect of fencing (FG) and grazing (GG) on grass species group (a), forb species group (b) and leguminous species group (c) of the temperate grassland community. The values are the ratios of aboveground biomass in each functional group. The box plot introduction: the box is ±SE, the whisker is min–max, the small square is mean values, and the horizontal line is the median. Significant difference between fenced and grazed meadows are indicated by symbols, $^{**}P < 0.01$, $^{*}P < 0.05$; ns, no significant difference.

Plants affect soil structure and function through both the quantity and quality of organic matter inputs that promote microbial activity and soil aggregation (Bach et al., 2012). Grassland restoration increased soil microbial biomass ($P < 0.05$), specifically fungi ($P < 0.001$), and restored soils exhibited higher rates of C and N mineralization ($P < 0.01$) (Bach et al., 2012), and the contents of microbial biomass C and N also increased significantly ($P < 0.05$) (An et al., 2009; Huang et al., 2011; Medina-Roldán et al., 2012). Zhou et al. (2011) reported vegetation recovery reduced C and N losses from wind erosion due to increase plant cover and productivity. In addition, more organic matter (litter, dead roots, mycorrhizae, and exudates) input to the soil leads to SOC and TN increases through vegetation recovery (Prietzel and Bachmann, 2012), and more above- and belowground biomass in fencing grassland (Figs. 3 and 8) which also accelerates more root input into the soil. However, in our study, fenced grassland had lower TP than grazed grassland in the deeper soils (50–100 cm) ($P < 0.05$). This may be due to the P deficit of the Loess Plateau. On grazed grassland, soil P eluviation is greater than the uptake of P by plants, and in turn, plant uptake is greater than soil P eluviation in fenced grassland.

Wu et al. (2010a) reported that long-term fencing can improve soil C storage of 0–30 cm soil depths in an alpine swamp meadow. Zhou et al. (2011) reported that soil C and N storages in the top 30 cm of soil measured in 2006 were 13.6 and 5.4 times greater, respectively, than those measured in 1981 under 26 years grazing exclusion in a desert shrubland. Our study also found that long-term fencing increased soil C and N storage in 0–30 cm soil depths compared to grazed grassland. However, our study also found that there had no detectable impact on C and N storage in surface soil (0–10 cm) following 30 years of grazing exclusion ($P > 0.05$), a finding which agrees with the result of Medina-Roldán et al. (2012). Animal manure input and trampling leading to higher soil BD may be the main reasons for soil C and N storage in surface soil showing no significant difference.

When estimating the rate of soil sequestration, the soil thickness reported in different studies has varied, leading to incomparable results in the carbon sequestration rate. For example, Nelson et al. (2008) reported that the soil C sequestration rate was 1.4–2.9 Mg ha^{-1} year^{-1} in 0–60 cm soil layer after grassland restoration cultivated land south-central Saskatchewan, Canada. According to the results of Wu et al.'s (2010a) study, we estimated that the soil C sequestration rate was nearly 0.6 Mg ha^{-1} year^{-1} in the 0–30 cm soil layer following 9 years of fencing in the alpine meadow of the Qinghai-Tibetan Plateau. The rate in Yunwu Mountain on the Loess Plateau is relative higher than global C sequestration rate of grassland (Lal, 2004). In our study, we estimated that the annual mean soil C sequestration rate was 3.00 Mg ha^{-1} year^{-1} in the 0–100 cm soil depth after fencing for 30 years compared to long-term grazing grassland, which is similar to Deng et al.'s results (2013b). Using a meta-analysis they reported that following vegetation restoration (<23 year) in the Yunwu Mountain reserve of the Loess Plateau, the soil carbon sequestration rate had reached 2.77 Mg ha^{-1} year^{-1}. Qiu et al. (2013) reported an accumulation of 1.68–4.40 Mg C ha^{-1} year^{-1} in 0–80 cm soil depths, compared to grazed grasslands.

Wang et al. (2012) reported a 20–37% decrease in SOC and a 5–37% decrease in TN after light and heavy grazing on the northern Tibetan Plateau, corresponding to losses of 0.14– 0.29 Mg C ha^{-1} 10 cm^{-1} year^{-1}. Ingram et al. (2008) reported that 21 years of heavy grazing resulted in losses of 0.60 Mg C ha^{-1} year^{-1} in the 0–60 cm soil layer in a mixed-grass ecosystem. We also estimated that the annual mean soil N sequestration were 0.32 Mg ha^{-1} year^{-1} of 0–100 cm soil depths under fencing. Qiu et al. (2013) have also reported that grazing exclusion resulted in an accumulation of 0.16–0.40 Mg N ha^{-1} year^{-1} in 0–80 cm soil depths, compared to grazed grasslands. The increase in N stock will reduce N limitation and support long-term C sequestration (Li et al., 2012). However, since the rate of relative N stock change was found to be lower than that of relative C stock change, the C/N ratio gradually increased, we also found the C/N were higher in fenced grasslands compared to grazed grasslands, although the difference was not significant (Fig. 5g). This may result in the occurrence of progressive N limitation in the long-term, reducing the rate of C sequestration (Li et al., 2012). The accumulation of C and N in grassland directly due to increased input of C and N into soils by litter and roots (Figs. 3 and 8), may be indirectly attributed to the decreased loss of vegetation C and N by virtue of grazing exclusion, and the decreased loss of soil C and N by reducing soil respiration, and ammonium volatilization and N_2O emission resulting from nitrification (Polley et al., 2008; Wu et al., 2012).

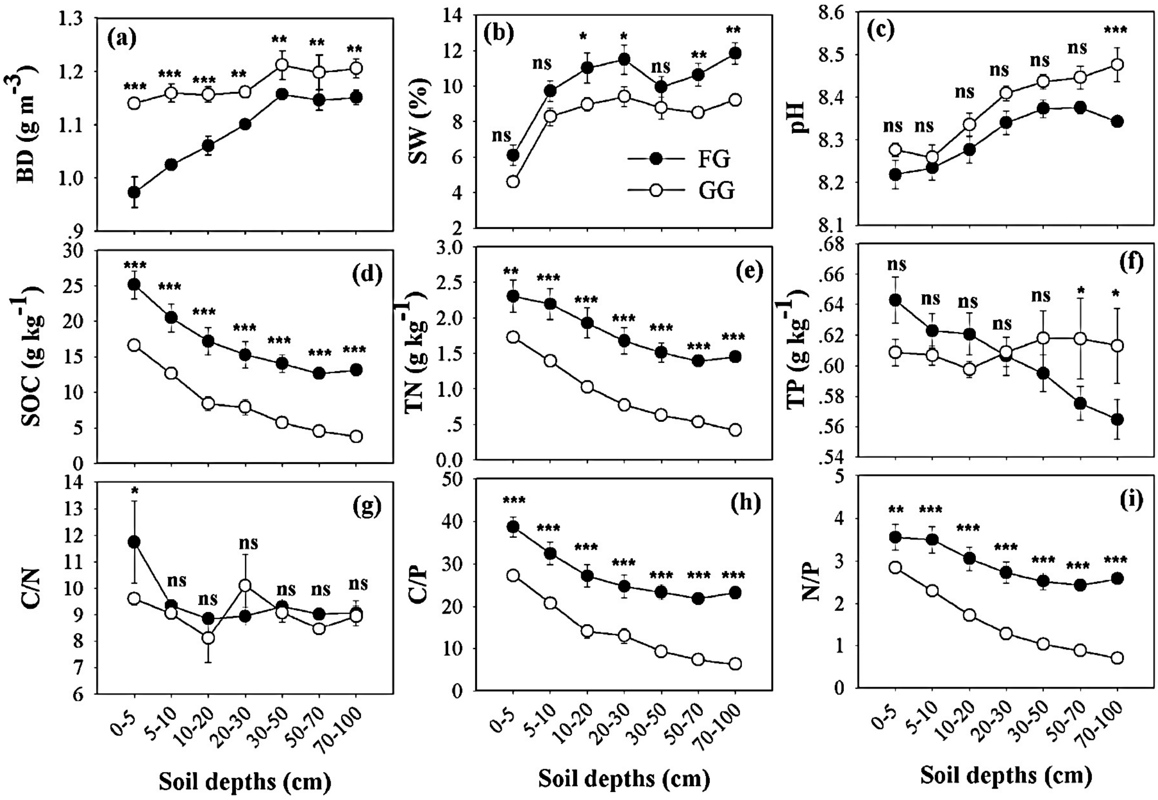

Fig. 5. Effect of fencing (FG) and grazing (GG) on soil bulk density (BD) (a), soil water content (b), soil pH (c), SOC (d), TN (e), TP (f), C/N (g), C/P (h) and N/P (f) of the temperate grassland community. The values are mean ± SE. Significant difference between fenced and grazed meadows are indicated by symbols, ***$P < 0.001$, **$P < 0.01$, *$P < 0.05$; ns, no significant difference.

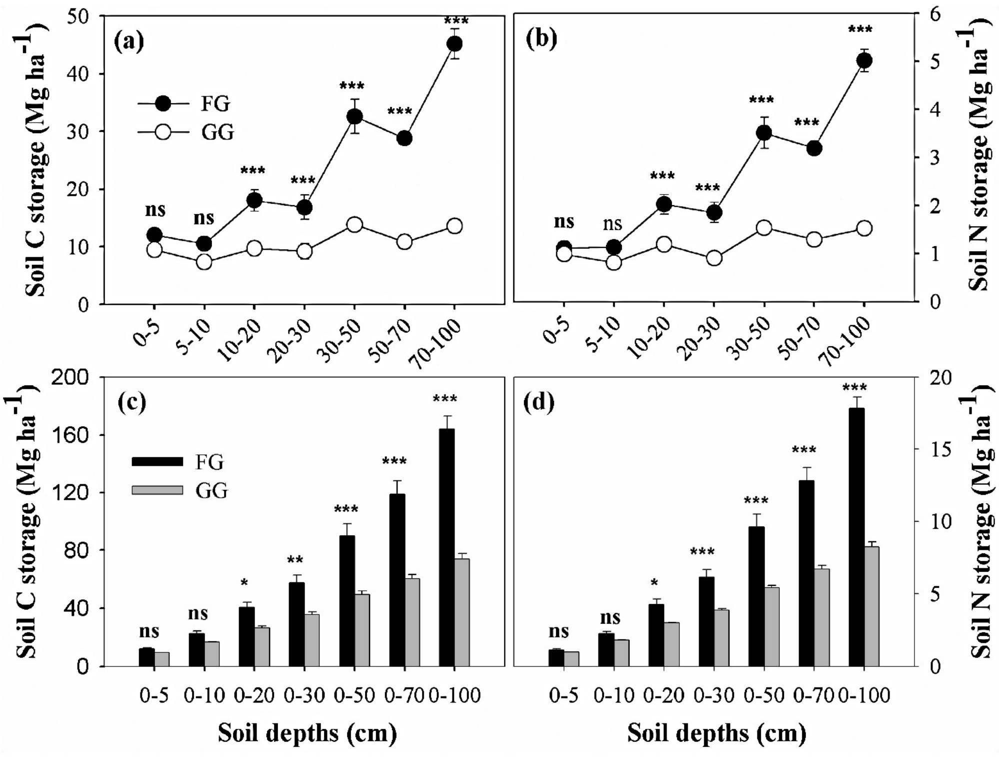

Fig. 6. Effect of fencing (FG) and grazing (GG) on soil OC storage (a), soil N storage (b), soil cumulative C storage (c) and soil cumulative N storage (d) of the temperate grassland community. The values are mean ± SE. Significant difference between fenced and grazed meadows are indicated by symbols, ***$P < 0.001$, **$P < 0.01$, *$P < 0.05$; ns, no significant difference.

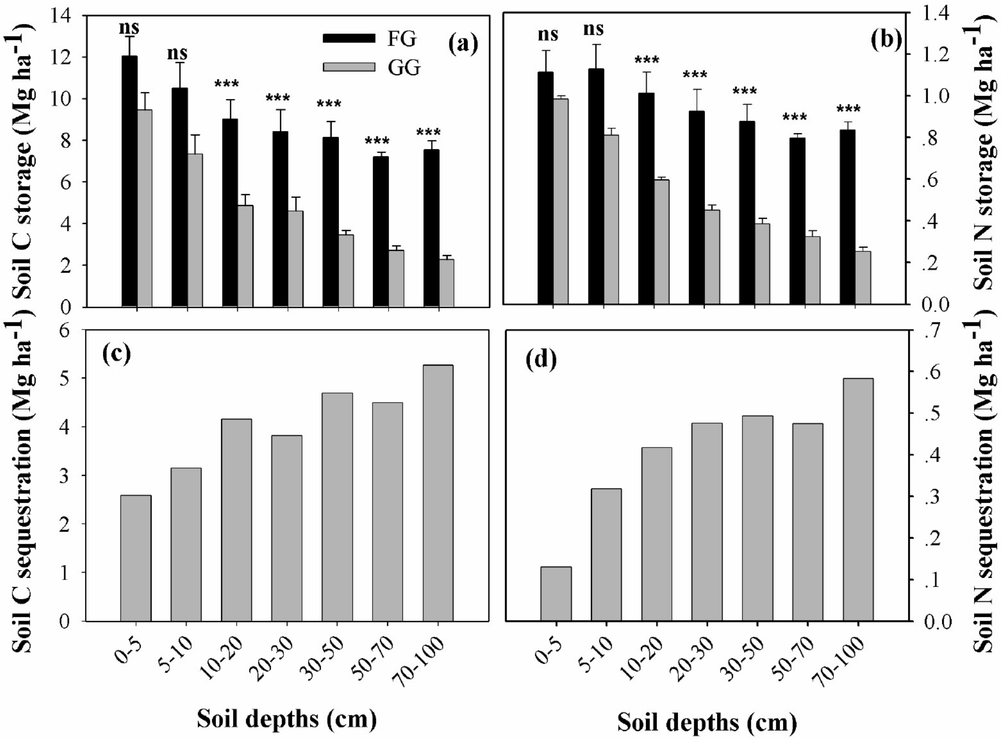

Fig. 7. Effect of fencing (FG) and grazing (GG) on soil OC storage (a), soil N storage (b), soil C sequestration (c) and soil N sequestration of the temperate grassland community at each soil depth for every 5 cm. The values are mean ± SE. Significant difference between fenced and grazed meadows are indicated by symbols, $^{***}P < 0.001$; ns, no significant difference.

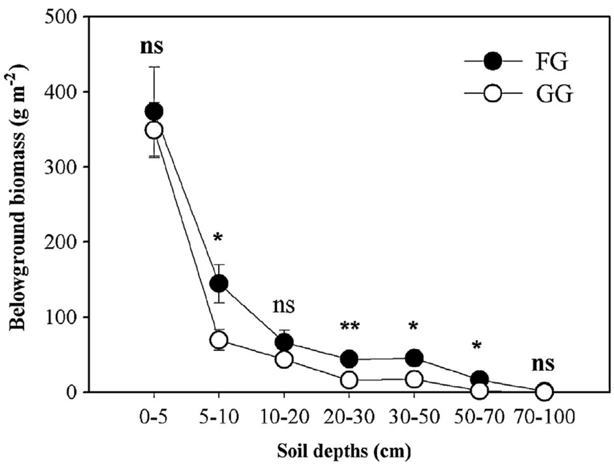

Fig. 8. Effect of fencing (FG) and grazing (GG) on the belowground biomass distribution of the temperate grassland community. The values are mean ± SE. Significant difference between fenced and grazed meadows are indicated by symbols, $^{**}P < 0.01$, $^{*}P < 0.05$; ns, no significant difference.

4. Conclusions

Fencing increases vegetation coverage, height, plant diversity, biomass production and litter, resulting primarily from an increase in the ratio of grass functional group species, and increases in the allocation ratio of aboveground biomass owing to a reduced R/S ratio. Fencing has significant effects on soil BD, soil moisture and pH. Long-term grazing exclusion also led to marked increases in SOC, soil TN, C/P, N/P and soil C and N storage within the 0 to 100 cm soil profile. Future studies should place more emphasis on changes deeper in the soil profile. The findings are important for evaluating the resilience of grassland ecosystems disturbed by grazing. The overall conclusion is simple: fencing is an effective restoration strategy for the recovery of degraded grasslands.

Acknowledgements

The study was funded by the Strategic Priority Research Program of the Chinese Academy of Sciences (XDA05050403) and the Scholarship Award for Excellent Doctoral Student granted by the Ministry of Education, PRC.

References

Allen, V.D., Batello, C., Berretta, E.J., Hodgson, J., Kothmann, M., Li, X., McIvor, J., Milne, J., Morris, C., Peeters, A., Sanderson, M., 2011. An international terminology for grazing lands and grazing animals. Grass Forage Sci. 66, 2–28.

An, S.S., Huang, Y.M., Zheng, F.L., 2009. Evaluation of soil microbial indices along a revegetation chronosequence in grassland soils on the Loess Plateau, Northwest China. Appl. Soil Ecol. 41, 286–292.

Bach, E.M., Baer, S.G., Six, J., 2012. Plant and soil responses to high and low diversity grassland restoration practices. Environ. Manage. 49, 412–424.

Bremner, J.M., 1996. Nitrogen-total. In: Sparks, D.L. (Ed.), Methods of Soil Analysis, Part 3. America Society of Agronomy, Madison, SSSA Book Series: 5, pp. 1085–1121.

Deng, L., Sweeney, S., Shangguan, Z.P., 2013a. Grassland responses to grazing disturbance: plant diversity changes with grazing intensity in a desert steppe. Grass Forage Sci., doi:10.1111/gfs.12065.

Deng, L., Sweeney, S., Shangguan, Z.P., 2013b. Long-term effects of natural enclosure: carbon stocks, sequestration rates and potential for grassland ecosystems in the Loess Plateau. Clean-Soil Air Water, http://dx.doi.org/10.1002/clen.201300176.

Frank, D.A., Groffman, P.M., 1998. Ungulate vs. landscape control of soil C and N processes in grasslands of Yellowstone National Park. Ecology 79, 2229–2241.

Gallego, L., Distel, R.A., Camina, R., Rodríguez Iglesias, R.M., 2004. Soil phytoliths as evidence for species replacement in grazed rangelands of central Argentina. Ecography 27, 725–732.

Gibson, R.S., Hewitt, A., Sparling, G., Bosch, O.J.H., 2001. Vegetation change and soil quality in central Otago Tussock grasslands, New Zealand. Range J. 22, 90–204.

Golodets, C., Kigel, J., Sternberg, M., 2010. Recovery of plant species composition and ecosystem function after cessation of grazing in a Mediterranean grassland. Plant Soil 329, 365–378.

Guo, L.B., Gifford, R.M., 2002. Soil carbon storage and land use change: a meta analysis. Global Change Biol. 8, 345–360.

Hastings, A., Byers, J.E., Crooks, J.A., Cuddington, K., Jones, C.G., Lambrinos, J.G., Talley, T.S., Wilson, W.G., 2007. Ecosystem engineering in space and time. Ecol. Lett. 10, 153–164.
Huang, Y.M., Michel, K., An, S.S., Zechmeuster-Boltenstern, S., 2011. Changes in microbial-community structure with depth and time in a chronosequence of restored grassland soils on the Loess Plateau in northwest China. J. Plant Nutr. Soil Sci. 174, 765–774.
Ingram, L.J., Schuman, G.E., Buyer, J.S., Vance, G.F., Ganjegunte, G.K., Welker, J.M., Derner, J.D., 2008. Grazing impacts on soil carbon and microbial communities in a mixed-grass ecosystem. Soil Sci. Soc. Am. J. 72, 939–948.
Lal, R., 2004. Soil carbon sequestration impacts on global climate change and food security. Science 304, 1623–1627.
Li, D.J., Niu, S.L., Luo, Y.Q., 2012. Global patterns of the dynamics of soil carbon and nitrogen stocks following afforestation: a meta-analysis. New Phytol. 195, 172–181.
Li, W.J., Li, J.H., Knops, J.M.H., Wang, G., Jia, J.J., Qin, Y.Y., 2009. Plant communities, soil carbon, and soil nitrogen properties in a successional gradient of sub-alpine meadows on the eastern Tibetan Plateau of China. Environ. Manage. 44, 755–765.
Li, X.R., Jia, X.H., Dong, G.R., 2006. Influence of desertification on vegetation pattern variations in the cold semi-arid grasslands of the Qinghai-Tibet plateau, North-west China. J. Arid Environ. 64, 505–522.
Liang, Y., Han, G.D., Zhou, H., Zhao, M.L., Snyman, H.A., Shan, D., Havstad, K.M., 2009. Grazing intensity on vegetation dynamics of a typical steppe in northeast Inner Mongolia. Range Ecol. Manage. 62, 328–336.
Liu, S.L., Guo, X.D., Fu, B.J., Lian, G., Wang, J., 2007. The effect of environmental variables on soil characteristics at different scales in the transition zone of the Loess Plateau in China. Soil Use Manage. 23, 92–99.
Louhaichi, M., Ghassali, F., Salkini, A.K., Petersen, S.L., 2012. Effect of sheep grazing on rangeland plant communities: case study of landscape depressions within Syrian arid steppes. J. Arid Environ. 79, 101–106.
Louhaichi, M., Salkini, A.K., Petersen, S.L., 2009. Effect of small ruminant grazing on the plant community characteristics of semi-arid Mediterranean ecosystems. Int. J. Agric. Biol. 1, 681–689.
Manzano, M.G., Návar, J., 2000. Processes of desertification by goats overgrazing in the Tamaulipan thornscrub (matorral) in north-eastern Mexico. J. Arid Environ. 44, 1–17.
Medina-Roldán, E., Paz-Ferreiro, J., Bardgett, R.D., 2012. Grazing exclusion affects soil and plant communities, but has no impact on soil carbon storage in an upland grassland. Agr. Ecosyst. Environ. 149, 118–123.
Miller, J.J., Chanasyk, D.S., Curtis, T., Willms, W.D., 2010. Influence of streambank fencing on the environmental quality of cattle-excluded pastures. J. Environ. Qual. 39, 991–1000.
Mohr, D., Topp, W., 2005. Influence of deer exclusion on soil nutrients in oak forests of a Central European low mountain. Land Degrad. Dev. 16, 303–309.
Nelson, D.W., Sommers, L.E., 1982. Total carbon, organic carbon, and organic matter. In: Page, A.L., Miller, R.H., Keeney, D.R. (Eds.), Methods of soil analysis. American Society of Agronomy and Soil Science Society of American, Madison, pp. 1–129.
Nelson, J.D.J., Schoenau, J.J., Malhi, S.S., 2008. Soil organic carbon changes and distribution in cultivated and restored grassland soils in Saskatchewan. Nutr. Cycl. Agroecosyst. 82, 137–148.
Palik, B.J., Goelbel, P.C., Kirkman, L.K., West, L., 2000. Using landscape hierarchies to guide restoration of disturbed ecosystems. Ecol. Appl. 10, 189–202.
Parkinson, J.A., Allen, S.E., 1975. A wet oxidation procedure suitable for the determination of nitrogen and mineral nutrients in biological material. Commun. Soil Sci. Plant Anal. 6, 1–11.
Prietzel, J., Bachmann, S., 2012. Changes in soil organic C and N stocks after forest transformation from Norway spruce and Scots pine into Douglas fir, Douglas fir/spruce, or European beech stands at different sites in Southern Germany. Forest Ecol. Manage. 269, 134–148.
Polley, H.W., Frank, A.B., Sanabria, J., Phillips, R.L., 2008. Interannual variability in carbon dioxide fluxes and flux-climate relationships on grazed and ungrazed northern mixed-grass prairie. Global Change Biol. 4, 1620–1632.
Qiu, L.P., Wei, X.R., Zhang, X.C., Cheng, J.M., 2013. Ecosystem carbon and nitrogen accumulation after grazing exclusion in semiarid grassland. PLoS ONE 8, e55433.
Reynolds, H.L., Packer, A., Bever, J.D., Clay, K., 2003. Grassroots ecology: plant-microbe-soil interactions as drivers of plant community structure and dynamics. Ecology 84, 2281–2291.
Rytter, R.M., 2012. Stone and gravel contents of arable soils influence estimates of C and N stocks. Catena 95, 153–159.
Santiago do Vale, V., Schiavini, I., de Oliveira, A.P., Gusson, A.E., 2010. When ecological functions are more important than richness: a conservation approach. J. Ecol. Nat. Environ. 2, 270–280.
Schönbach, P., Wan, H.W., Gierus, M., Bai, Y.F., Müller, K., Lin, L.J., Susenbeth, A., Taube, F., 2011. Grassland responses to grazing: effects of grazing intensity and management system in an Inner Mongolian steppe ecosystem. Plant Soil 340, 103–115.
Shrestha, G., Stahla, P.D., 2008. Carbon accumulation and storage in semi-arid sagebrush steppe: effects of long-term grazing exclusion. Agr. Ecosyst. Environ. 125, 173–181.
Smith, R.S., Shiel, R.S., Millward, D., Corkhill, P., 2000. The interactive effects of management on the productivity and plant community structure of an upland meadow: an 8-year field trial. J. Appl. Ecol. 37, 1029–1043.
Stark, S., Wardle, D.A., Ohtonen, R., Helle, T., Yeates, G.W., 2000. The effects of reindeer grazing on decomposition, mineralization and soil biota in a dry oligotrophic Scots pine forest. Oikos 90, 301–310.
Stirling, G., Wilsey, B., 2001. Empirical relationships between species richness, evenness and proportional diversity. Am. Nat. 158, 286–300.
Su, Y.Z., Li, Y.L., Cui, J.Y., Zhao, W.Z., 2005. Influences of continuous grazing and livestock exclusion on soil properties in a degraded sandy grassland, Inner Mongolia, northern China. Catena 59, 267–278.
Su, Y.Z., Zhao, H.L., Zhang, T.H., Zhao, X.Y., 2004. Soil properties following cultivation and non-grazing of a semi-arid sandy grassland in northern China. Soil Till. Res. 75, 27–36.
Wang, L., Niu, K.C., Yang, Y.H., Zhou, P., 2010. Patterns of above-and belowground biomass allocation in China's grasslands: evidence from individual-level observations. Sci. Chin. Ser. C: Life Sci. 53, 851–857.
Wang, X., Yan, Y., Cao, Y., 2012. Impact of historic grazing on steppe soils on the northern Tibetan Plateau. Plant Soil 354, 173–183.
Wu, G.L., Du, G.Z., Liu, Z.H., Thirgood, S., 2009. Effect of fencing and grazing on a Kobresia-dominated meadow in the Qinghai-Tibetan Plateau. Plant Soil 319, 115–126.
Wu, G.L., Liu, Z.H., Zhang, L., Chen, J.M., Hu, T.M., 2010a. Long-term fencing improved soil properties and soil organic carbon storage in an alpine swamp meadow. Plant Soil 332, 331–337.
Wu, G.L., Liu, Z.H., Zhang, L., Hu, T.M., Chen, J.M., 2010b. Effects of artificial grassland establishment on soil nutrients and carbon properties in a black-soil-type degraded grassland. Plant Soil 333, 469–479.
Wu, H.H., Wiesmeier, M., Yu, Q., Steffens, M., Han, X.G., Kögel-Knabner, I., 2012. Labile organic C and N mineralization of soil aggregate size classes in semiarid grasslands as affected by grazing management. Biol. Fert. Soils 48, 305–313.
Zhou, Z.C., Shangguan, Z.P., Zhao, D., 2006. Modeling vegetation coverage and soil erosion in the Loess Plateau Area of China. Ecol. Model. 198, 263–268.
Zhou, Z.Y., Li, F.R., Chen, S.K., Zhang, H.R., Li, G.D., 2011. Dynamics of vegetation and soil carbon and nitrogen accumulation over 26 years under controlled grazing in a desert shrubland. Plant Soil 341, 257–268.