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Grazing exclusion effects on above- and below-ground C and N pools of typical grassland on the Loess Plateau (China)

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article info abstract

Article history: Received 12 April 2014 Received in revised form 21 July 2014 Accepted 25 July 2014 Available online 12 August 2014

Keywords: Grazing exclusion Carbon pool Nitrogen pool Plant–soil interface Semi-arid grassland

Fencing with grazing exclusion is an effective grassland restoration and management practice used to achieve sustainability of grassland ecosystems worldwide. However, how the fencing with grazing exclusion affects ecosystem services related to carbon and nitrogen dynamics in grassland ecosystems has remained controversial over the past two decades. We investigated plant biomass, diversity and function groups, as well as soil bulk density, pH, soil carbon and nitrogen contents and the carbon/nitrogen ratio within the upper 0–1 m soil layer in fenced grassland with grazing exclusion and in grazed grassland on the Loess Plateau (China) in 2012 and 2013. We estimated the carbon and nitrogen pools of the plants and soils to determine how the grazing exclusion affected them. Results showed that soil carbon content in the topsoil, plant biomass and diversity, and grasses increased, while bulk density, pH and forbs decreased after grazing exclusion. The increases in soil carbon content, the cumulative organic carbon pool and the rate of change in the cumulative organic carbon pool mainly occurred in the upper 20 cm soil layer after 8 years of grazing exclusion. Our study suggested that the 8-year grazing exclusion had a great influence on the carbon pools, but there were no changes in the soil nitrogen pool. Identifying the main factors that affect the carbon and nitrogen dynamics after grazing exclusion among the soil and plant properties should be given more attention in future studies. From Dryland Farming on the Loss Plateral Ristinue of Soil and Woter Conservation, Northwest A6P Unher

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1. Introduction

Carbon (C) and nitrogen (N) are key nutrients for all living organisms on earth, and play an important role in regulating both the structure and function of terrestrial ecosystems [\(Elser et al., 2010](#page-6-0)). Grassland is a widely distributed biome type in northwest China and covered a total area of 4 million km^2 in 2001 (Bai et al., 2012; Jiang [et al., 2011](#page-6-0)). Grasslands have inherently high soil carbon pools (Cp) and soil nitrogen pools (Np) that supply plant nutrients, increase soil aggregation and the cation exchange and water holding capacities, and limit soil erosion [\(Conant et al., 2001; Kool et al., 2007\)](#page-6-0). A small percentage change in Cp or Np could have a great impact on atmospheric carbon oxide ($CO₂$) or nitrous oxide ($N₂O$) concentrations because Cp and Np are much greater than the atmospheric C and N pools [\(Chapuis-Lardy et al., 2007; Schlesinger, 1990; Yang et al., 2007](#page-6-0)). Thus, maintenance of Cp and Np is a key factor in achieving the sustainability of grassland ecosystems.

However, intensive livestock grazing or other agricultural uses of grasslands have resulted in widespread vegetation and soil degradation, such as a reduction of plant species diversity, net primary productivity, and soil nutrient contents ([Fu et al., 2000; He et al., 2012; Jiang et al.,](#page-6-0) [2011; Li et al., 2009; Mcsherry and Ritchie, 2013; Zhou et al., 2011](#page-6-0)). Nearly 100% of uncultivated grasslands are grazed by large mammals, and grazing has become a key factor controlling Cp and Np ([Mcsherry](#page-6-0) [and Ritchie, 2013](#page-6-0)). Previous studies on grazing in grasslands have shown that grazing can cause a number of changes that affect the possible mechanisms involved in the losses of C and N from grazed grasslands (GG). These include changes in plant community composition [\(Jiang et al., 2011\)](#page-6-0), primary production [\(Conant et al., 2001](#page-6-0)) and belowground biomass (BGB) productivity [\(Garcia-Pausas et al., 2011](#page-6-0)), as well as the associated changes in the soil microbial community [\(Klumpp](#page-6-0) [et al., 2009; Shang et al., 2014](#page-6-0)). It is critical to obtain a better understanding of how grazing influences the key properties of ecosystem function and sustainability and, thereby, to provide guidelines for improving grassland management practices.

The major factors that influence C and N pools in grassland are related to the following variables: soil properties, including soil type

Abbreviations: C, carbon; N, nitrogen; BD, soil bulk density; AGB, aboveground biomass; BGB, belowground biomass; SL, semi-decomposition layer mass; Cp, soil organic carbon pool; Np, soil nitrogen pool; SOC, soil organic carbon content; TN, soil total nitrogen content; FG, fenced grassland with grazing exclusion; GG, grazed grassland; S, Richness index; H, Shannon–Wiener diversity index; E, Evenness index.

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[\(Mcsherry and Ritchie, 2013](#page-6-0)), soil sampling depth ([Deng et al., 2013a\)](#page-6-0), soil bulk density (BD) [\(Xie et al., 2007\)](#page-7-0), and soil pH [\(Liu et al., 2013\)](#page-6-0); biotic variables, such as grassland species composition ([Dybzinski](#page-6-0) [et al., 2008; Oelmann et al., 2011; Wedin and Tilman, 1996\)](#page-6-0), plant biomass [\(Piñeiro et al., 2010\)](#page-7-0), and plant traits [\(De Deyn et al., 2008;](#page-6-0) [Oelmann et al., 2011\)](#page-6-0); and management practices, such as grazing and grazing exclusion [\(Jones and Donnelly, 2004; Mcsherry and Ritchie,](#page-6-0) [2013](#page-6-0)). Both abiotic and biotic factors influence the net accumulation rates of soil organic carbon and total nitrogen by affecting the ratio of organic matter inputs to mineralization ([O'Brien et al., 2010; Shang](#page-6-0) [et al., 2012, 2014\)](#page-6-0).

Grazing exclusion was one of the effective management practices that aimed to increase soil C and N contents in rangelands, which have been highly relevant as potential strategies for C sequestration. However, grazing exclusion effects on Cp and Np have not been conclusive [\(Tilman et al., 2006; Yang et al., 2007; Zhou et al., 2011](#page-7-0)). In some cases, Cp and Np were increased ([He et al., 2012; Qiu et al., 2013;](#page-6-0) [Shang et al., 2014; Wu et al., 2010a](#page-6-0)), while in others there were either decreases ([Hafner et al., 2012; Shi et al., 2013](#page-6-0)) or no significant effects [\(Medina-Roldán et al., 2012; Yang et al., 2010\)](#page-6-0). Further studies on C and N dynamics following grazing exclusion by fencing in semiarid grazed grasslands, especially short-term manipulative experiments, are needed to examine the direct effects on Cp and Np in northern China's grasslands (Golluscio et al., 2009; Piñeiro et al., 2009; Qiu [et al., 2013; Yang et al., 2010](#page-6-0)).

In this study, we assessed the effect of an 8-year (short-term) grazing exclusion on the vegetation aboveground biomass (AGB) and belowground biomass (BGB), the litter and the semi-decomposing layer mass (SL), soil properties, and the Cp and Np of the plant-soil interface using two-years of data collected from a semi-arid grassland on the Loess Plateau, China. Specifically, we addressed the following questions: (i) how does the relatively short-term grazing exclusion affect the contents and pools of soil C and N by changing grassland plant properties (i.e., species richness, above- and below-ground biomass, plant function groups and litter biomass) and soil BD and pH; and (ii) how do the above- and below-ground C and N pools respond to grazing exclusion. incre[a](#page-6-0)sed (He et al., 2012; Our et al., 2013; condition-

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2012; Shi et al., 2013) or no signif

2. Method and material

2.1. Experimental site and design

The study site was located in Yuzhong County, Gansu Province (104°09′ E, 35°57′ N, 1966 m). It was in the semi-arid continental temperate monsoon climate zone. According to data available for the period 2005–2013 at the study site from the National Meteorological Information Center of China, the annual mean temperature is 7.39 °C and the annual cumulative precipitation is 373.33 mm, of which approximately 80% falls between May and September (Fig. 1). The site was within an area where the topography was characteristic of the Loess Plateau, consisting of plains, ridges, mounds, etc., with an elevation ranging between 1714 and 2089 m. The soil was classified as a Sierozem, a calcareous soil that is characteristic of the Chinese loess region and that is fertile but extremely susceptible to erosion ([Li et al.,](#page-6-0) [2010](#page-6-0)). The experiments were conducted in a fenced area containing the grazing exclusion grassland (FG) and in a grazed grassland (GG), where both treatments were on gentle slopes and had the same slope aspect. The dominant grass species were Stipa bungeana, Artemisia frigida and Leymus secalinus in both the FG and GG areas. The FG was established in 2005 and covered a total area of about 8 ha. The grazing intensity of the GG was 2–3.5 sheep ha^{-1} during the summer months from May to September, and 1–2 sheep ha^{-1} during the winter months from October to the following April. The study used data collected for the two years of 2012 and 2013.

In mid August of 2012 and 2013, when the grassland community biomass peaked, ten 20 m \times 20 m blocks were randomly selected in both the GG and FG areas [\(Wu et al., 2010a](#page-7-0)). In each block, five quadrats $(1 m \times 1 m)$ were examined, one located at each of the four corners and one at the center of the block [\(Deng et al., 2013a](#page-6-0)). Hence, 20 blocks were investigated in this study, and we surveyed ten blocks with fifty quadrats in both the FG and GG areas, i.e., a total of one hundred quadrats. In each quadrat, the coverage and plant height, above- and belowground biomass, and litter biomass were determined, and soil samples from the 0–100 cm soil layer were collected.

2.2. Aboveground plant sampling

In each quadrat, all green, aboveground plant parts of each individual species, as well as the entire litter layer, were cut, collected, and put into separate labeled envelops. To measure the belowground biomass, soil samples were collected from five soil layers (0–5, 5–10, 10–20, 20–30, and 30–50 cm) in three places within each quadrat using a 9 cm diameter root auger. The roots collected within the soil samples were isolated by spreading the samples in shallow trays, overfilling the trays with water and allowing the outflow from the trays to pass through a 0.5-mm mesh sieve [\(Deng et al., 2013a\)](#page-6-0). The root tissues, litter and aboveground plant parts were dried at 65 °C for 24 h and weighed to determine the dry mass. Plant organic carbon was assayed by dichromate oxidation ([Nelson and Sommers, 1982](#page-6-0)) and plant total nitrogen was determined by using the modified Kjeldahl method (Bremner, 1996). Each analysis was carried out in two replicates.

The Richness Index, Shannon–Wiener diversity index and Evenness index of the fenced and grazed grassland communities were calculated using the following functions ([Wu et al., 2009](#page-7-0)):

Richness index (R) : $R = S$ Shannon–Wiener diversity index (H):

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

Evenness index (E) :

$$
E=\frac{H}{\ln S},
$$

where S is the total number of species in the grassland community and Pi is the mass ratio of the ith species to the total biomass.

Fig. 1. Annual mean temperature and annual cumulative precipitation at the study site (2005–2013). Note: The straight lines were fitted to the annual mean temperature or annual mean precipitation data over time.

2.3. Soil sampling and determination

Soil samples were taken at three points in the quadrats of each block near the belowground biomass sampling points as described above. Undisturbed soil cores and disturbed soil samples from seven soil layers (0–5, 5–10, 10–20, 20–30, 30–50, 50–70, 70–100 cm) were collected using a soil corer and a soil drilling sampler (9-cm inner diameter), respectively. The undisturbed samples from the same soil layers within one quadrat were then mixed together to make one composite sample. The soil samples were air-dried and were then passed through a 0.14-mm sieve, and roots and other debris were removed. Soil pH was determined for a soil–water ratio of 1:5. The soil BD (g cm^{−3}) of the different soil layers was measured using the soil cores (volume, 100 cm³) by the volumetric ring method ([Wu et al., 2010a\)](#page-7-0). Soil organic carbon was assayed by dichromate oxidation ([Nelson and Sommers,](#page-6-0) [1982\)](#page-6-0), and total soil nitrogen was determined using the modified Kjeldahl method ([Bremner, 1996\)](#page-6-0). Each analysis was carried out in two replicates. We used the following equation to calculate the soil organic carbon pool for each layer (Deng et al., 2013a):

$Cp = BD \times SOC \times D$

where, Cp is the soil organic carbon pool (kg m^{-2}); BD is the soil bulk density (g cm^{−3}); SOC is the soil organic carbon content (g kg^{−1}); and D is the thickness of the sampled soil layer (m). The total Cp for the 0–100 cm soil layer was obtained by totaling the Cp of each layer within it.

The following equation was used to calculate the soil nitrogen pool in each layer [\(Deng et al., 2013a](#page-6-0)):

$Np = BD \times TN \times D$

where, Np is the soil nitrogen pool (kg m⁻²); BD is soil bulk density (g cm⁻³); TN is the total soil nitrogen content (g kg^{-1}); and D is the thickness of the sampled soil layer (m). The total Np for the 0–100 cm soil layer was obtained by totaling the Np of each layer within it.

2.4. Statistical analyses

The differences among each of the above- and below-ground properties for a given grazing treatment were examined by t-tests following ANOVA that compared the results obtained from all the blocks in either 2012 or 2013, but no significant differences were found among the blocks or between the two years (Table 1). Therefore, all data for 2012 and 2013 were expressed as the mean \pm standard error (SE) of the ten samples collected for each treatment during that particular year of the two-year study. Plant community structure and soil characteristics were analyzed to assess the effects of grazing exclusion. Differences in AGB, BGB, Litter, SL, plant density, richness index, SOC, TN, Cp and Np between FG and GG treatments were assessed using analysis of variance (ANOVA). Significant differences were evaluated at the 0.05, 0.01, and 0.001 levels. Figures were created using SigmaPlot version 8.0 (Systat software Inc., San Jose, CA, USA) and all of the statistical tests were carried out using SPSS version 12.0 (SPSS Inc., Chicago, IL, USA).

3. Results

3.1. Grazing exclusion effects on plant properties

The AGB was 113.50 \pm 4.38 g m⁻² in FG, and was significantly greater than that in GG (62.53 \pm 3.38 g m⁻²) at the peak growing season harvest ($P < 0.001$; [Table 2\)](#page-3-0). The BGB (to 50-cm soil depth) increased under grazing exclusion, from 313.20 \pm 22.48 to 371.00 \pm 9.85 g m^{-2} (P < 0.01; [Table 2](#page-3-0)). The litter and SL amounts were also notably higher under FG than those under GG. Accordingly, the root/shoot ratio of the total plant biomass decreased from 5.11 ± 0.42 in GG to 3.31 \pm 0.13 in FG (P < 0.001, [Table 2\)](#page-3-0) during the study period. The mean S values were 6.3 for FG and 6 for GG. However, both the H and E indexes of the grassland community were significantly higher for GG than those for FG [\(Table 2\)](#page-3-0). The FG sites had a significantly larger grass species group ($P < 0.001$) and a smaller forbs species group than the GG sites ($P < 0.001$, [Fig. 2\)](#page-3-0). The fraction of grass biomass increased from 33.1% to 71.1%, while that of forbs decreased from 60.4% to 25.0% after grazing exclusion (Fig. 2). The leguminous species groups at the FG and GG sites exhibited no significant differences ($P > 0.05$).

Grazing exclusion significantly increased C and N stored in AGB, litter and SL (Fig. 3). The C pools in AGB, BGB, SL and litter were on average 76.5% ($P < 0.001$), 25.42% ($P < 0.01$), 76.5% ($P < 0.001$) and 421.7% ($P < 0.001$) higher for FG than for GG, respectively, while the corresponding N pools were $71.9% (P < 0.001)$, 2.8% (P > 0.05), 123.7% $(P < 0. 01)$ and 324.0% higher $(P < 0.001)$. The total plant C and N were significantly increased by about 51.5% and 29.5% in FG than in GG (Fig. 3). These results suggested that grazing exclusion had the potential to enhance the accumulation of C and N in biomass and litter in semiarid grasslands.

3.2. Grazing exclusion effects on soil physical and chemical properties

In total, 700 soil samples (100 quadrats with 7 soil depths) were analyzed for soil BD, pH, C content (SOC) and N content (TN) in the two grassland treatments. An ANOVA proved that soils under FG had significantly lower pH values in the 0–10 cm soil layer ($P < 0.05$), but higher values in the 10–50 cm soil layer ($P < 0.01$), when compared with those under GG. Short-term grazing exclusion resulted in notable reductions in BD in the upper 20 cm soil layer ($P < 0.05$) [\(Fig. 4](#page-4-0)). The SOC was significantly increased in the upper 50 cm soil layer ($P < 0.001$), while little change occurred in the 50–100 cm layer, after the shortterm grazing exclusion. The TN was not significantly changed after grazing exclusion at any depth of the 0–100 cm soil layer [\(Fig. 5b](#page-4-0)). Accordingly, the soil organic carbon content/soil total nitrogen content (C/N) ratios significantly increased ($P < 0.01$) after grazing exclusion ([Fig. 5](#page-4-0)c). The SOC and TN decreased with soil depth under both GG and FG [\(Fig. 5](#page-4-0)). **html** the C6 is the parti[c](#page-3-0)ular the redistricts of $V \approx 0.01$, ine Fraction and the redistricts of the sample station (Fig. 2). The leads

3.3. Grazing exclusion effects on soil C and N pools

Grazing exclusion resulted in significant increases in the soil C pool within the 0–5, 10–20 and 30–50 cm soil layers, although there were

Table 1

F-statistic and probability values of one-way ANOVA tests of the effect of the year of sampling on plant properties in grassland communities in fenced and grazed grasslands (n = 10).^a

Treatment	AGB $(g m^{-2})$	BGB $(g \; m^{-2})$	Litter $(g m^{-2})$	SL. $(g m^{-2})$	Root/shoot ratio			
Grazed	0.207 0.661	0.024 0.88	0.055 0.821	0.999 0.347	0.189 0.675	0.276 0.614	2.067 0.188	0.252 0.629
Fenced	0.414 0.538	0.202 0.665	0.065 0.805	1.058 0.334	0.097 0.764	0.783 0.402	0.321 0.587	0.779 0.403

^a Abbreviations for the plant properties are listed in the Abbreviations.

Table 2

Effects on plant properties in grassland communities of either fencing without grazing or grazing in 2012 (n = 10), 2013 (n = 10) and the total for the two years combined (n = 20).^a

^a Abbreviations for the plant properties are listed in the Abbreviations.

^b Values (mean \pm SE) are means of ten squares; significant difference between fenced and grazed grasslands are indicated by symbols: ***P < 0.001, *P < 0.01, *P < 0.05; ns, no significant difference.

no changes in the other soil layers ([Fig. 6](#page-5-0)a). Short-term grazing exclusion did not result in increases in the soil N pool in any of the investigated soil layers (Fig. 6b). The cumulative soil C pool in the upper 20 cm soil layer increased by about 0.18 kg m⁻² (from 1.57 to 1.76 kg m⁻², $P < 0.001$), which represented about 50% of the total increase within the 0–100 cm soil layer (from 4.68 to 5.04 kg m[−] , $P < 0.001$) after eight-years of grazing exclusion (Fig. 7a). Thus, the main increase in the soil C pool occurred near the soil surface and the annual mean soil C sequestration rate was 0.05 kg m⁻² yr⁻¹ within the 0–100 cm soil layer. In contrast, no trend was identified in the cumulative soil nitrogen pool (Np) and Np remained almost constant in all the soil layers during this 8-year period (Fig. 7b).

4. Discussion

Grazing was generally expected to lead to greater SOC losses and to reduce potential C inputs to soil organic matter due to the removal of photosynthetic tissue and subsequent respiration of assimilated C by the grazing animals ([Derner and Schuman, 2007; Klumpp et al., 2009;](#page-6-0) Lettens et al., 2005). The study showed that aboveground C and N increased by 107% and 91%, respectively, while the belowground biomass C and N only increased by about 25% and 2% after 8 years of grazing exclusion (Fig. 3). Qiu et al. (2013) observed that 76%–80% and 50%– 75% increases in C and 62%–81% and 55%–106% increases in N occurred in AGB and BGB (0–80 cm soil depth), respectively, after 17 years of grazing exclusion on the Loess Plateau. The C and N increases in the

Fig. 2. Effect of grazing exclusion after fencing (FG) and grazing (GG) on function groups (grasses, forbs and leguminous species) during the two year study, and the effect of the different sampling years on the function groups under FG or GG. Note: The bar values of mean $+$ SE (n $=$ 5) are the ratios of aboveground biomass in each functional group. Significant differences between FG and GG are indicated by symbols above the bars: ***P < 0.001; **P < 0.01; *P < 0.1; ns, no significant difference. NS, means no significant difference between 2012 and 2013 in each function group.

Fig. 3. Effect of grazing exclusion after fencing (FG) and grazing (GG) on (a) the plant carbon (C) pool, and (b) the plant nitrogen (N) pool, in the aboveground biomass (AGB), belowground biomass (BGB), litter, and semi-decomposition layer (SL). Note: Significant differences in C or N pools from different sources between FG and GG treatments are indicated by symbols next to the bars: ***P $<$ 0.001, **P $<$ 0.01, *P $<$ 0.05, no symbol, no significant difference. Different letters above the bars indicate significant differences in the vegetation C or N pools between FG and GG treatments.

Fig. 4. Effect of grazing exclusion after fencing (FG) and grazing (GG) on (a) soil pH and (b) bulk density. Note: The values are mean \pm SE, Significant differences between FG and GG at different soil depths are indicated by symbols: ***P < 0.001, *P < 0.01, *P < 0.05, no symbol, no significant difference.

BGB were lower in this study than those in other studies, which was mainly due to the shorter period following grazing exclusion and to the different species compositions (Deng et al., 2013b; Qiu et al., 2013).

Although C and N pools in belowground plant tissues increased after the grazing exclusion was implemented, the increases were smaller than those found in other studies, but plant roots also significantly influenced soil C and N levels (Mi et al., 2008; Wen et al., 2013). Mokany et al. [\(2006\)](#page-6-0) found that a temperate grassland had a mean root/shoot ratio of 4.2, which was similar to our findings that the root/shoot ratios of the total plant biomass in FG and GG were 3.31 and 5.11, respectively. The C and N pools in the aboveground vegetation (including the SL and litter) have been found to be smaller than those belowground (King [et al., 1997](#page-6-0)). This has been ascribed to the higher belowground carbon allocation, the virtual absence of persistent woody structures aboveground, and the generally higher decomposability of the shoot, as compared with the root, tissues (Aerts and Chapin, 1999; Dodd and [Mackay, 2011; Frank et al., 1995\)](#page-6-0). Thus, roots have been identified as the major C and N sinks within the BGB (Wu et al., 2010b), which was in agreement with the findings of our study that the C and N sinks in the BGB were about 60% and 61%, respectively, of the total C and N in the plants. **lsio[n](image of Fig.�4)** after fencing (FC) and grazing (GC) on (a) soil pH and (b) bulk density. Note: The values are mean \pm 5. Significated by symbols: " P = 0.001, " P = 0.01, " P = 0.01, P = 0.05, no symbol, no significant di

Our results showed that the FG had higher plant diversity than GG, with more grass species but fewer forbs species [\(Table 2](#page-3-0), Fig. 2). Higher plant diversity mitigated soil carbon losses in deeper soil layers [\(Steinbeiss et al., 2008\)](#page-7-0), and might also lead to greater soil carbon pools in the long-term; therefore, conservation of biodiversity might play a role in greenhouse gas mitigation ([Steinbeiss et al., 2008;](#page-7-0) Tilman et al., 2006). The C pool was increased during our study by the increasing plant diversity after grazing exclusion. Grasses had predominantly negative responses whereas forbs species mostly had positive responses to grazing, and no significant responses were observed among the herbaceous legumes [\(Díaz et al., 2007](#page-6-0)). Plant trait composition may influence the soil decomposer diversity through the diversity of substrates and habitats they provide ([Porazinska et al., 2003](#page-7-0)), and decomposer diversity can in turn affect soil carbon cycling through functional complementarities [\(De Deyn et al., 2008; Hättenschwiler et al.,](#page-6-0) 2005; Wardle, 2006). Therefore, the changes in the function groups can also account for the changes in SOC after fencing with grazing exclusion.

The present study indicates that the grazing exclusion affected the SOC and TN contents differently. Grazing exclusion of the grassland resulted in significant increases in SOC in the upper 50 cm soil layer but no changes were observed in TN in any of the investigated soil layers, which reflected similar changes that occurred in the C and N levels in the belowground plant tissues [\(Figs. 3 and 5](#page-3-0)). There were fewer differences among the soil TN contents at the grazing excluded site than among those at the grazed site because, after grazing, the biomass allocation ratio of the belowground vegetation increased and the N inputs counteracted the N outputs for a short period ([Medina-Roldán](#page-6-0) [et al., 2012; Shang et al., 2012; Wen et al., 2013\)](#page-6-0). Thus, C and N pools

Fig. 5. Effect of grazing exclusion after fencing (FG) and grazing (GG) on (a) soil organic carbon content, (b) soil nitrogen content and (c) soil organic carbon content/soil nitrogen content (C/N) . Note: The values are mean \pm SE. Significant differences between FG and GG are indicated by symbols: ***P < 0.001, **P < 0.01, *P < 0.05; no symbol, no significant difference.

Fig. 6. Effect of grazing exclusion after fencing (FG) and grazing (GG) on (a) the soil organic carbon pool and (b) the soil nitrogen pool. Note: The values are mean \pm SE. Significant differences between FG and GG are indicated by symbols: ***P < 0.001, **P < 0.01, *P < 0.05; no symbol, no significant difference.

in the plant roots demonstrated significant positive relationships with soil C and N concentrations (Chen et al., 2007).

Fenced grassland with grazing exclusion had a lower soil pH value and BD in the topsoil, and both of these affected the soil C and N (Wu [et al., 2010a](#page-7-0)). Grazing might lead to soil compaction, reduction in soil porosity, increased soil BD, and limited oxygen concentrations and, thus, could restrict the activity of edaphon in grassland soil (Shi et al., [2013; Wu et al., 2010a](#page-7-0)). This may have contributed to the lower SOC in the grazed areas in our study. Soil pH has a significant negative relationship with soil carbon content, which was also one of our findings in that pH decreased and SOC increased in the topsoil after grazing exclusion ([Liu et al., 2013](#page-6-0)). The C/N ratios increased under grazing exclusion conditions, which suggest potential N limitations for SOC accumulation after the implementation of grazing exclusion (Piñeiro [et al., 2006\)](#page-7-0). Soil depth also influences SOC and TN whereby they decrease as soil depth increases, which was consistent with the findings of [Deng et al. \(2013a\)](#page-6-0) [\(Fig. 5](#page-4-0)). The effect of the grazing exclusion on SOC was mainly observed in the upper 50 cm soil layer, where nutrient and soil organic matter accumulation results from complex interactions between plant and soil biotic processes (Deng et al., 2013b). Soil pH, type and BD, plant biomass, diversity and function groups, and grassland management practices were all factors that could have affected the C and N dynamics in our study. **[h](image of Fig.�7)[n](image of Fig.�6)** [a](#page-6-0)nd there (FG) and grazing (GG) on (a) the soil organic carbon pool and (b) the soil nitrogen pool. Notes The relationted by symbols: ""P< 0.001, "P< 0.01, "P< 0.01, "P< 0.01, "P< 0.01, "P< 0.01, "P< 0.01, "P < 0.0

The cumulative Cp in the upper 100 cm soil layer in our study was about 5.04 and 4.67 kg m−² under FG and GG, respectively, of which about 36% and 32% was in the upper 20 cm layer. These values were lower than those obtained in recent studies [\(Deng et al., 2013a,](#page-6-0) 2013b; Mi et al., 2008). [Yang et al. \(2010\)](#page-7-0) estimated that the Cp was 6.36 kg m−² in a northern typical steppe in Inner Mongolia, China. [Mi](#page-6-0) et al. (2008) measured the Cp in the upper 1 m soil layer within different precipitation ranges across China. They reported estimated Cp values of 14.4 and 16.4 kg m⁻² for precipitation ranges of 400–500 and 500–800 mm, respectively, which both had mean annual precipitation amounts that were higher than the 373.33 mm in our study region. The effects of precipitation on the grazing exclusion effect on Cp may also be driven by grassland types associated with different precipitation levels (Mcsherry and Ritchie, 2013). The Np was not significantly different after the implementation of the grazing exclusion in any of the seven studied soil layers. In general, for a given soil depth, the Np was determined by the soil nitrogen content and the soil BD. After grazing exclusion, the soil nitrogen content was not significantly changed, and the soil BD significantly changed only in the 0–5 and 10–20 cm soil layers. Thus, the Np was not significantly changed after 8 years of grazing exclusion.

The cumulative Cp in the upper 20 cm soil layer increased by about 0.18 kg m⁻², which was about 50% of the increase in the 0–100 cm soil

Fig. 7. Effect of grazing exclusion after fencing (FG) and grazing (GG) on (a) the cumulative soil organic carbon pool and (b) the cumulative soil nitrogen pool. Note: The values are mean \pm SE. Significant differences between FG and GG are indicated by symbols: ***P < 0.001, **P < 0.01, *P < 0.05; no symbol, no significant difference.

layer after 8 years of grazing exclusion. The soil C pool increased at a rate of 22.5 g m⁻² year⁻¹ in the 0–20 cm layer and at about 45 g m⁻² year⁻¹ in the 0–100 cm layer over the 8 years of grazing exclusion. The rate of C accumulation in our study was much lower than the mean rate of 89.9 g m^{-2} year^{-1} occurring over 26 years that was reported by [Zhou et al. \(2011\)](#page-7-0) for an arid area in northwest China. However, our rate was similar to the rate of C accumulation found in the upper 20 cm soil layer (28.3 g m $^{-2}$ year $^{-1}$) that occurred over 12 years under a N limited grassland in Cedar Creek, Minnesota (Fornara and Tilman, 2008). Furthermore, the highest C accumulation rates occurred 9–18 years after grazing pressure was reduced in the 26 year study of [Zhou et al. \(2011\)](#page-7-0), suggesting that the effects of fencing with grazing exclusion on the soil C and N pools need long-term monitoring to more fully account for the changes in the dynamics of C and N.

5. Conclusion

Our findings suggest that excluding grazing from grassland is likely to reap benefits for both above- and below-ground C sequestration in the short term on the semiarid Loess Plateau. Soil pH, type and BD, plant biomass, diversity and function groups, grassland management practices, and precipitation are all factors that could have affected the C and N dynamics in this study. The increases in SOC, cumulative Cp and the rate of change in cumulative Cp mainly occurred in the upper 20 cm soil layer after the eight-year grazing exclusion in the studied grassland. Our study suggested that 8 years of fencing with grazing exclusion had a great influence on the carbon pools but there were no changes in the soil nitrogen pools. These findings are particularly important when assessing the resilience of this livestock-disturbed grassland in a semiarid region and to understanding the complex interactions between soil C accumulation and plant species number, functional group composition and productivity, which can help to develop effective long-term management strategies. Identifying the main factors that influence C and N dynamics and how C and N levels respond to longterm grazing exclusion should be studied in the future. Frank A, Tanak D, Heath May Frank A, Tanak D, Holman, L, Folos 914

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Acknowledgment

We thank Hua Fu, Decao Niu, and Xudong Li for their help with the field experiment monitoring and thank Dr. Elizabeth M Baggs and two anonymous reviewers for their valuable suggestions. This research was funded by Projects of the Natural Science Foundation of China (NSFC41390463, NSFC41371282, NSFC41371242), the Strategic Priority Research Program — Climate Change: Carbon Budget and Related Issues of the Chinese Academy of Sciences (CAS) (Grant No. XDA05050403), the Action Plan for West Development Project of Chinese Academy of Sciences (KZCX2-XB3-13), Project of Natural Science Foundation of Shaanxi Province (2011JQ5003, 2014KJXX-15), and the '100-Talent Program' of CAS.

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