

Is grazing exclusion effective in restoring vegetation in degraded alpine grasslands in Tibet, China?

Yan Yan and Xuyang Lu

Key Laboratory of Mountain Surface Processes and Ecological Regulation, Institute of Mountain Hazards & Environment, Chinese Academy of Sciences, Chengdu, China

ABSTRACT

Overgrazing is considered one of the key disturbance factors that results in alpine grassland degradation in Tibet. Grazing exclusion by fencing has been widely used as an approach to restore degraded grasslands in Tibet since 2004. Is the grazing exclusion management strategy effective for the vegetation restoration of degraded alpine grasslands? Three alpine grassland types were selected in Tibet to investigate the effect of grazing exclusion on plant community structure and biomass. Our results showed that species biodiversity indicators, including the Pielou evenness index, the Shannon-Wiener diversity index, and the Simpson dominance index, did not significantly change under grazing exclusion conditions. In contrast, the total vegetation cover, the mean vegetation height of the community, and the aboveground biomass were significantly higher in the grazing exclusion grasslands than in the free grazed grasslands. These results indicated that grazing exclusion is an effective measure for maintaining community stability and improving aboveground vegetation growth in alpine grasslands. However, the statistical analysis showed that the growing season precipitation (GSP) plays a more important role than grazing exclusion in which influence on vegetation in alpine grasslands. In addition, because the results of the present study come from short term (6–8 years) grazing exclusion, it is still uncertain whether these improvements will be continuable if grazing exclusion is continuously implemented. Therefore, the assessments of the ecological effects of the grazing exclusion management strategy on degraded alpine grasslands in Tibet still need long term continued research.

Subjects Biodiversity, Ecology, Ecosystem Science, Plant Science **Keywords** Grazing exclusion, Alpine grassland, Grassland degradation, Community characteristics, Biomass, Tibet

Submitted 26 March 2015 Accepted 24 May 2015 Published 16 June 2015

Corresponding author Xuyang Lu, xylu@imde.ac.cn

Academic editor Harpinder Sandhu

Additional Information and Declarations can be found on page 12

DOI 10.7717/peerj.1020

© Copyright 2015 Yan and Lu

Distributed under Creative Commons CC-BY 4.0

OPEN ACCESS

INTRODUCTION

Tibet is an important ecological security shelter zone that acts as an important reservoir for water, regulating climate change and water resources in China and eastern Asia (*Sun et al.*, 2012). Alpine grasslands are the most dominant ecosystems over all of Tibet, covering more than 70% of the whole plateau's area and representing much of the land area on the Eurasian continent (*Wang et al.*, 2002). Alpine grasslands in this area are grazed by indigenous herbivores, such as yak and Tibetan sheep. These ecosystems have traditionally served as the principal pastures for Tibetan communities and are regarded as one of the

major pastoral production bases in China (*Wen et al., 2013a*). Alpine grasslands also provide ecosystem functions and services, such as carbon sequestration, biodiversity conservation, and soil and water protection, and are also of great importance for Tibetan culture and the maintenance of Tibetan traditions (*Wen et al., 2013b*; *Shang et al., 2014*).

Alpine grasslands in Tibet have been regionally degrading, even desertifying since the 1980s. For instance, from 1981 to 2004 in northern Tibet, which is the main extent of alpine grassland distribution and an important livestock production centre in Tibet, degraded alpine grasslands accounted for 50.8% of the total grassland area, and severely and extremely severely degraded grasslands accounted for 8.0% and 1.7%, respectively (Gao et al., 2010). Grassland degradation may be due to a combination of global climate change, rapidly increasing grazing pressure, rodent damage and other factors (Chen et al., 2014). Nevertheless, overgrazing, which caused by an increase in the population of humans and domestic livestock in Tibet, is widely considered the primary cause of grassland degradation. Overgrazing may result in significant changes to the composition and structure of the plant community including significant decreases in the regenerative ability of the grasslands, decreases in biomass, decreases in the amount of nutrients returned to the soil as litter, and eventually cause grassland degradation (Zhou et al., 2005). Additionally, overgrazing causes an increase in potential evapotranspiration, thereby promoting the warming of local climate and further accelerating alpine grassland degradation processes (Du et al., 2004). Under conditions of overgrazing by livestock, the succession of degraded grasslands can become a vicious circle: overgrazing causes grassland degradation, which facilitates rodent infestation, which further degrades grasslands (Kang et al., 2007).

In an attempt to alleviate the problem of grassland degradation in Tibet, China's state and local authorities initiated a program in 2004 called 'retire livestock and restore pastures' (Fig. 1). As part of this campaign, grazing exclusion by fencing has been widely used as an approach to the restoration grasslands (*Wei et al., 2012*). Grazing exclusion is an effective grassland management practice that aimed to prevent grassland degradation and retain grassland ecosystem function throughout the world in recent decades (*Mata-González et al., 2007*; *Mofidi et al., 2013*). This management strategy is expected to restore vegetation and enhance rangeland health in overgrazed and degraded grasslands characterized by low productivity, low vegetation cover and low biomass in Tibet. This campaign has been in progress for more than ten years, which brings to light the question: is this program successful in the restoration of degraded alpine grasslands? This question has attracted great attention in recent years and has inspired a large number of studies on the effect of grazing exclusion on the alpine grasslands (*Wei et al., 2012*; *Wu et al., 2012*; *Shi et al., 2013*).

Nevertheless, research results with regard to the effect of grazing exclusion on plant biomass and biodiversity were not consistent. For instance, grazing exclusion could result in the improvement of grass cover, species biodiversity and biomass due to the absence of grazing in some degraded grassland ecosystems (*Mata-González et al.*, 2007; *Mofidi et al.*, 2013). However, grazing exclusion may induce a decrease of species richness and

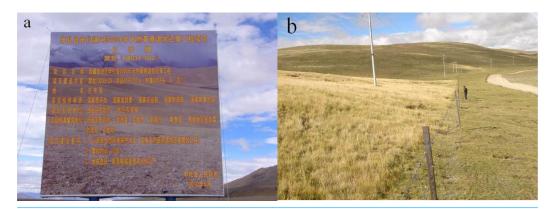


Figure 1 The 'retire livestock and restore pastures' program in Tibet. (A) A grazing exclusion sign set by the government, (B) the fence-line contrast between the fenced and grazed grassland.

biodiversity by the replacement of species that are highly adapted to grazing by strongly dominant competitors that increase in abundance due to grazing cessation, such as certain graminoids (*Mayer et al.*, 2009; *Shi et al.*, 2013). The lack of a consistent response of vegetation to grazing exclusion has been attributed to a broad range of factors that determine whether and how herbivores affect plant communities, include the time of grazing exclusion (*Mayer et al.*, 2009), growing season precipitation (*Wu et al.*, 2012), productivity (*Schultz, Morgan & Lunt, 2011*), climatic conditions (*Jing, Cheng & Chen, 2013*) and so on. Therefore, specific studies are crucial to that ecosystems are properly managed and that conservation goals are achieved. In the Tibetan Plateau, although numerous studies exploring the effect of grazing exclusion on alpine grassland ecosystem structure and function have been published in recent years (*Wu et al.*, 2009; *Shang et al.*, 2013; *Li et al.*, 2014; *Luan et al.*, 2014; *Zhang et al.*, 2015), the majority of studies focus on a single alpine grassland ecosystem type and in one experimental site. Little studies focus on the assessment of the effects of grazing exclusion on alpine grassland ecosystem on regional scale (*Wu et al.*, 2013; *Wu et al.*, 2014).

To gain a better understanding of the restoration and management of degraded grasslands in Tibet, studies are needed to investigate alpine grassland vegetation growth and community composition dynamics. Thus, the aim of this study was to investigate the effects of excluding grazing herbivores through fencing on high-altitude alpine grasslands in Tibet, and to assess whether fencing can be used as an effective grassland management tool to restore vegetation in degraded alpine grasslands. Three alpine grassland types and nine counties, which represent the main natural alpine grassland distribution in Tibet, were selected as sampled sites according to the time and range of grazing exclusion. We hypothesized that in the absence of grazing, the vegetation cover, height, the above- and below-ground biomass, species richness, diversity would improve due to the absence of disturbance from herbivorous livestock. In addition, based on different plant species diversity and community structure, vegetation productivity and cover, and environmental conditions, we further hypothesized that vegetation biomass and biodiversity responses to the absence of grazing would differ among different alpine grassland types.

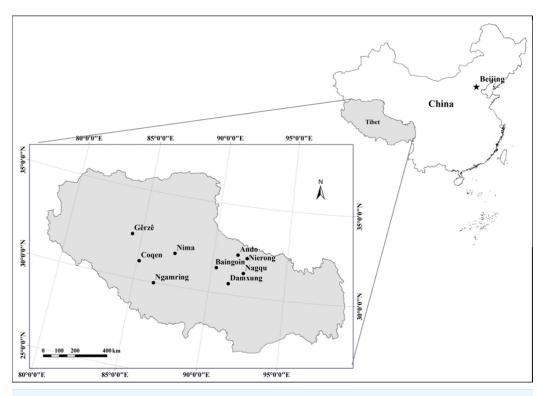


Figure 2 Location of sampling sites of alpine grasslands in Tibet.

METHODS

Study area

Tibet is located between $26^{\circ}50'$ and $36^{\circ}29'$ N and $78^{\circ}15'$ and $99^{\circ}07'$ E and covers a total area of more than 1.2 million km², which is approximately one-eighth of the total area of China (Fig. 2). The main portion of the Qinghai-Tibetan Plateau lies at an average altitude of 4,500 m; it is geomorphologically unique in the world. Because of its extensive territory and highly dissected topography, the region has a diverse range of climate and vegetation zones. The solar annual radiation is strong and varies between 140 and 190 kcal cm⁻² in different parts of the region. Annual sunshine tends to increase from the east to the west and ranges from 1,800 to 3,200 h. The average annual temperature is rather low, with a large diurnal range, and varies from $18 \,^{\circ}\text{C}$ to $-4 \,^{\circ}\text{C}$; the average temperature in January varies from $10 \,^{\circ}\text{C}$ to $-16 \,^{\circ}\text{C}$; the average temperature in July varies from $24 \,^{\circ}\text{C}$ to $8 \,^{\circ}\text{C}$, and decreases gradually from the southeast to the northwest. The average annual precipitation is less than 1,000 mm in most areas of Tibet, and reaching up to 2,817 mm in the east and dropping down to approximately 70 mm in the west (*Zou et al.*, 2002; *Dai et al.*, 2011).

According to the first national survey of Chinese grassland resources, Tibet ranks first among all Chinese provinces and autonomous regions in the diversity of its grassland ecosystems, comprising 17 types of grassland based on the classification system used for the whole country (*Gai et al.*, 2009). Among all grassland types, alpine steppe is the most common grassland type in Tibet; it is composed of drought tolerant perennial herb

m 11 1 5	C.1	1		m•1 .
lable Descri	ption of the san	ipling sites of al	lpine grasslands in	Tibet.

Location	Longitude (E)	Latitude (N)	Altitude (m)	Grassland type	Dominant species
Damxung	91°14′56″	30°36′08″	4,407	Alpine meadow	Kobresia pygmaea C. B. Clarke
Nagqu	92°09′11″	31°16′30″	4,458	Alpine meadow	Kobresia humilis
Nierong	92°16′49″	32°07′48″	4,614	Alpine meadow	Kobresia littledalei C. B. Clarke
Ando	91°38′28″	32°15′37″	4,696	Alpine meadow	Kobresia pygmaea C. B. Clarke
Baingoin	92°09′11″	31°16′30″	4,632	Alpine steppe	Carex moorcroftii Falc. Ex Boott
Nima	87°24′57″	31°48′27″	4,550	Alpine steppe	Stipa purpurea
Coqen	85°09′09″	31°01′58″	4,687	Alpine steppe	Stipa purpurea
Ngamring	86°37′52″	29°38′38″	4,583	Alpine steppe	Carex moorcroftii Falc. Ex Boott
Gêrzê	84°49′34″	31°59′25″	4,591	Alpine desert steppe	Stipa purpurea

or small shrubs under cold and arid and semiarid climate conditions, and represents approximately 38.9% of the total Tibetan grassland area. Alpine meadow is the second largest grassland type, and is composed of perennial mesic and mesoxeric herbs under cold and wet climate conditions, occupying approximately 31.3% of the total grassland area of Tibet. Alpine desert steppe occupies approximately 10.7% of the total grassland area, and is composed by xeric small shrubs and small grasses under cold and arid climate conditions; it is a transitional type of alpine grassland from the steppe to the desert in Tibet (*Land Management Bureau of Tibet*, 1994).

Survey design, sampling, and data collection

Since the 'retire livestock and restore pastures' ecological engineering program started in 2004, more than 2.4×10^6 ha of alpine grasslands in Tibet have been fenced to exclude livestock grazing. Nine counties in Tibet, in which the extent of fenced area was relatively larger, were selected to investigate the effect of grazing exclusion on plant community composition and biomass in alpine grasslands (Fig. 2). These nine counties represented three of the main natural grassland vegetation types in Tibet, including alpine meadow, alpine steppe and alpine desert steppe (Table 1). In these counties, areas which were fenced over the years of 2005–2007 were chosen as sampling sites in the present study. Since fencing establishment, the fenced grassland has completely been excluded from livestock grazing, while the surrounding grassland continued conventional grazing by yak and sheep around the year. No accurate information, such as timing, intensity, and frequency, on grazing activities and pasture management in the open free grazing grassland, but the actual averaged stocking rate ranges were approximately from 0.16 sheep units hm⁻² of the westernmost county to 2.05 sheep units hm⁻² in the easternmost county for the study region (Wu et al., 2014). In addition, no specific permits were required for the described field studies and the field studies did not involve protected animals or plants. The enclosed areas were defined as grazing exclusion (GE) plot and the areas outside of fencing were defined as free grazing (FG) plot. Field surveys were conducted during late July to mid-August in 2013; three pairs (fenced versus free grazed) of plots in each site were chosen and surveyed.

At each sample plot, three pairs of 0.5 m \times 0.5 m quadrats at each GE and FG treatment sample plots were laid out collinearly at intervals of approximately 20 m. All species within each quadrat were identified and their coverage, density, frequency and natural height were measured. The frequency counts were made by dividing the 0.5 m \times 0.5 m frame into 10 cm \times 10 cm cells. Within each cell, presence/absence species data were recorded. These data were summed up to calculate frequencies per quadrat (1–100%). The geographical coordinates, elevation and vegetation types for each site were also recorded, and the picture of each quadrat was taken using a digital camera to calculate community cover. Aboveground and belowground plant components were harvested. Aboveground plant parts in the sample quadrat were clipped to the soil surface with scissors and belowground plant parts in the sample quadrat were directly acquired by excavation, including both live roots and dead roots of all plant species. After sun-drying of plant samples in the field, they were brought to laboratory and oven-dried at 65 °C for 72 h to determine biomass.

Monthly meteorological datasets with spatial resolutions of 0.5° from 2005 to 2013, which generated by Thin Plate Spline (TPS) method using ANUSPLIN software (ERSI, Redlands, California, USA) and the data sources include monthly mean temperature and monthly precipitation data from more than 2,400 well distributed climate stations across China, were derived from the China Meteorological Data Sharing Service System (http://cdc.nmic.cn). The average growing season (from May to September) temperature (GST) and growing season precipitation (GSP) from 2005 to 2013 matched with nine sites' locations were extracted from these meteorological raster surfaces in ArcGIS 10.0 (ERSI, Redlands, California, USA) for further analyses.

Plant community characteristics

Total cover, community vegetation height, and the Simpson index, Shannon index, and Pielou index were used to describe the plant community characteristics of alpine grassland ecosystems. The vegetation total cover was acquired from pictures of each quadrat by using CAN-EYE software (INRA-UAPV, France) and the vegetation height of the community was directly measured as the height of the dominant vegetation within each quadrat. To reveal the variation in community composition characteristics in grazing exclusion process, the Pielou evenness index (E), Shannon diversity index (H), and Simpson dominance index (D) were used to indicate plant community biodiversity changes. The Pielou evenness index reflects allocation information and species composition. The Shannon diversity index, which ranging in theory from 0 to infinity and incorporating both species richness and evenness aspects together, increases as the number of species increases and as individuals are evenly distributed among species. The Simpson dominance index gives the probability of two randomly chosen individuals drawn from a population belonging to the same species, a higher value also indicates a higher diversity.

The following formulas were used to calculate Pielou evenness index (E), Shannon–Wiener diversity index (H), and Simpson dominance index (D):

$$E = \left(-\sum P_i \ln P_i\right) / \ln S$$

$$H = -\sum P_i \ln P_i$$

$$D = 1 - \sum P_i^2$$

where $P_i = n_i/T$, n_i is the count of each plant species i in a quadrat, T is the total count of all plant species in a quadrat, in that context, P_i is the relative probability of finding the species i in a quadrat. S is the total observed number of species in a quadrat.

Statistical analysis

A paired difference t-test was conducted to test differences in the examined parameters between fenced and grazed plots within each grassland type. Analysis of covariance (ANCOVA) by the general linear model (GLM) was employed to evaluate the effects of grazing exclusion treatment and climatic factors on the plant community and biomass indices in Tibet. In the ANCOVA analysis, fixed factor was alpine grassland grazing treatments (FG and GE), while the covariates were GST and GSP. The two covariates GST and GSP that were used to fit the linear ANCOVA models were not highly interacted with the fixed factor (P > 0.05). Pearson correlation analysis was used to test the relationships between different plant community composition and biomass indices. The least significant difference test was used to compare the means at P < 0.05. All statistical analyses were performed using IBM SPSS Statistics 19 software (SPSS/IBM, Chicago, IL, USA).

RESULTS

Plant community characteristics

Changes in the selected plant community characteristics are shown in Table 2. Compared to the FG plots, vegetation total cover of alpine grassland (alpine meadow + alpine steppe + alpine desert steppe) was 8.83% higher (P < 0.05) and the community vegetation height was 2.65 cm higher (P < 0.05) in the GE plots. However, grazing exclusion did not significantly affect the biodiversity of alpine grassland (P > 0.05), although the Simpson index, Shannon index, and Pielou index in GE plots were 0.03, 0.07, and 0.01 lower than those indices in FG plots, respectively. Among three alpine grassland types, there was no significant difference in the Simpson index, Shannon index, or Pielou index between FG and GE plots in alpine meadow, alpine steppe or alpine desert steppe. Nevertheless, significant difference in total cover was found in alpine steppe and in community vegetation height was found in both alpine meadow and alpine steppe (P < 0.05). The results from ANCOVA analysis demonstrated that grazing exclusion had a significant effect on vegetation cover and vegetation height, but did not affect biodiversity indices, including D, H, and E. For growing season climate factors, GST had a significant effect on D and H, whereas GSP had a significant effect on most plant community characteristic indices (Table 3).

Aboveground and belowground biomass

Grazing exclusion had a significant effect on aboveground biomass of alpine grasslands, the mean value of aboveground biomass of the GE plots were 15.43 g cm⁻², higher than that of

Table 2 Plant community characteristics and biomass. Statistical comparison of overall mean values of plant community characteristics and biomass indices \pm standard error (S.E.) in alpine grassland by using paired difference t-test ($\alpha = 0.05$) between free grazing (FG) plots and grazing exclusion (GE) plots. P-values below 0.05 are in bold.

Indices	Site	Alpine meadow	Alpine steppe	Alpine desert steppe	Alpine grassland
Total cover (TC)	FG (%)	61.32 ± 5.29	$\textbf{14.96} \pm \textbf{1.36}$	6.74 ± 1.43	$\textbf{34.65} \pm \textbf{5.27}$
	GE (%)	70.07 ± 3.54	$\textbf{25.59} \pm \textbf{3.46}$	8.74 ± 0.46	$\textbf{43.48} \pm \textbf{5.23}$
Vegetation height (VH)	FG (cm)	$\textbf{2.62} \pm \textbf{0.56}$	$\textbf{5.39} \pm \textbf{0.30}$	5.77 ± 0.29	$\textbf{4.20} \pm \textbf{0.39}$
vegetation neight (VII)	GE (cm)	$\textbf{5.63} \pm \textbf{1.02}$	$\textbf{8.13} \pm \textbf{1.11}$	6.66 ± 0.19	$\textbf{6.85} \pm \textbf{0.70}$
Simpson index (D)	FG	0.37 ± 0.05	0.67 ± 0.02	0.43 ± 0.05	0.51 ± 0.04
Simpson fidex (D)	GE	0.35 ± 0.07	0.66 ± 0.02	0.23 ± 0.07	0.48 ± 0.05
Shannon index (<i>H</i>)	FG	0.80 ± 0.11	1.32 ± 0.08	0.71 ± 0.10	1.02 ± 0.08
Shannon maex (11)	GE	0.74 ± 0.13	1.31 ± 0.05	0.38 ± 0.10	0.95 ± 0.09
Pielou index (E)	FG	0.42 ± 0.05	0.76 ± 0.02	0.65 ± 0.09	0.60 ± 0.04
	GE	0.43 ± 0.07	0.76 ± 0.04	0.55 ± 0.14	0.59 ± 0.05
Aboveground biomass (AB)	$FG (g m^{-2})$	70.86 ± 10.82	$\textbf{33.95} \pm \textbf{4.86}$	$\textbf{17.15} \pm \textbf{5.39}$	$\textbf{48.49} \pm \textbf{6.56}$
	$GE (g m^{-2})$	80.62 ± 10.94	$\textbf{55.39} \pm \textbf{5.78}$	$\textbf{31.23} \pm \textbf{3.69}$	$\textbf{63.92} \pm \textbf{6.28}$
Belowground biomass (BB)	$FG (g m^{-2})$	628.27 ± 240.67	$\textbf{203.30} \pm \textbf{26.49}$	218.33 ± 46.71	393.85 ± 112.84
	$GE (g m^{-2})$	$1,010.86 \pm 265.88$	$\textbf{272.80} \pm \textbf{33.02}$	232.67 ± 36.18	596.37 ± 137.11
Total biomass (TB)	$FG (g m^{-2})$	699.13 ± 241.52	$\textbf{237.25} \pm \textbf{28.49}$	235.48 ± 51.96	442.34 ± 114.77
	GE $(g m^{-2})$	$1,091.48 \pm 263.27$	$\textbf{328.19} \pm \textbf{37.24}$	263.89 ± 35.96	660.28 ± 137.98

Table 3 Effects of grazing exclusion and climate factors. Results from analysis of covariance (ANCOVA) by the general linear model (GLM) showing *F* values and *P* values of plant community characteristics and biomass indices, which the fixed factor was grazing treatments (free grazing and grazing exclusion) and the covariates were growing season temperature (GST) and growing season precipitation (GSP). *P*-values below 0.05 are in bold.

Indices	Grazing exclusion		GST		GSP	
	F-value	P-value	F-value	P-value	F-value	P-value
Total cover (TC)	22.82	< 0.001	0.60	0.443	52.27	< 0.001
Vegetation height (VH)	11.35	0.001	2.44	0.124	0.32	0.572
Simpson index (D)	0.61	0.440	8.24	0.006	10.27	0.002
Shannon index (H)	0.62	0.435	13.81	0.001	6.99	0.011
Pielou index (E)	0.09	0.771	2.51	0.120	20.86	< 0.001
Aboveground biomass (AB)	4.07	0.049	0.01	0.950	20.69	< 0.001
Belowground biomass (BB)	1.58	0.215	0.00	0.982	11.66	0.001
Total biomass (TB)	1.84	0.181	0.00	0.980	13.28	0.001

FG plots (P < 0.05). However, grazing exclusion had no significant effect on belowground biomass and total biomass (aboveground biomass + belowground biomass) (P > 0.05, Table 2). Among the three alpine grassland types, for alpine meadow, there were no significant changes in biomass features 6–8 years after fencing, including aboveground, belowground, and total biomass. For alpine steppe, the aboveground, belowground, and total biomasses were all significantly increased due to grazing exclusion (P < 0.05). Moreover, grazing exclusion led to a significant increase in the aboveground biomass in

Table 4 Correlation relationships among plant community characteristics and biomass indices. Pearson's correlation coefficients among plant community characteristics and biomass indices of alpine grasslands in Tibet, and their significance levels.

Indices	TC	VH	D	Н	E	AB	BB
VH	-0.07						
D	-0.49^{**}	0.29*					
Н	-0.40^{**}	0.23	0.97**				
E	-0.64^{**}	0.32*	0.92**	0.84**			
AB	0.72**	0.18	-0.21	-0.17	-0.34^{*}		
BB	0.47**	-0.05	-0.27^{*}	-0.24	-0.29^{*}	0.21	
TB	0.50**	-0.04	-0.28^{*}	-0.24	-0.30^{*}	0.26	0.99**

Notes.

TC, Total coverage; VH, Vegetation height; D, Simpson index; H, Shannon index; E, Pielou index; AB, Aboveground biomass; BB, Belowground biomass; TB, Total biomass.

alpine desert steppe (P < 0.05, Table 2). Statistical analyses from ANCOVA showed that grazing exclusion had a significant effect on aboveground biomass of alpine grassland ecosystems in Tibet (P < 0.01, Table 3). The effects of climate factors on biomass were differ between GST and GSP, which GSP had a significant effect on all biomass indices, but GST had no any effect on biomass of alpine grasslands (Table 3).

Relationship among community characteristics and biomass indices

Correlation analyses showed that total cover was negatively correlated with D, H and E (P < 0.01), and significant positively correlated with aboveground, belowground, and total biomass (P < 0.01). However, community vegetation height was positively correlated with D and E (P < 0.01), and no correlations were found between the community vegetation height and any of the biomass parameters (P > 0.05). The community biodiversity indices D, H, and E were all positively correlated with each other (P < 0.01). In addition, the total biomass was positively correlated with the belowground biomass (P < 0.01; Table 4).

DISCUSSION

Overgrazing due to sharp growth of the human population and of food demand in recent years is a major cause of grassland degradation on the Tibetan Plateau (*Wei et al.*, 2012; *Shang et al.*, 2014). Grassland degradation has significantly altered species composition and decreased productivity in the region (*Zhou et al.*, 2006; *Ma, Zhou & Du*, 2013). The exclusion of livestock through the use of mesh fencing to create large-scale enclosures has become a common grassland management strategy for restoring degraded grasslands of the Tibetan Plateau in recent decades (*Wu et al.*, 2009; *Shi et al.*, 2013). Is grazing exclusion an effective policy to restore vegetation in degraded alpine grassland in Tibet? In the present study, three alpine grassland types and nine counties were selected as sampled sites according to the time and range of grazing exclusion to investigate the effects of grazing exclusion by fencing on plant community characteristics and biomass in degraded alpine grasslands.

 $^{^{*}} P < 0.05.$

^{**} *P* < 0.01.

Impacts of grazing exclusion on community characteristics

Vegetation cover is an important index for measuring the protective function of vegetation to the ground. Our study shows that continuous grazing exclusion resulted in a significant increase in the total vegetation cover of alpine grasslands (Table 2). This result is consistent with previous reports, supporting the conclusion that the exclusion of grazing livestock in the degraded alpine grasslands of the Tibetan Plateau exerts a strong effect on ecosystem dynamics by increasing vegetative cover (*Wu et al.*, 2009; *Shang et al.*, 2013). The mean vegetation height of community in GE plots was 6.85 cm, which was approximately 1.63 times that in the FG plots (Table 2). Similar results have also been reported in other studies of alpine grasslands in Tibet (*Deléglise*, *Loucougaray & Alard*, 2011; *Shang et al.*, 2013). Increased vegetation cover and height in the Tibetan Plateau after fencing has been reported due to the colonization capacity of the vegetation (*Shang et al.*, 2013) and the prevention of livestock herbivory on forage grasses, especially for graminoids and sedgy species that are palatable to livestock (*Wu et al.*, 2009).

Species diversity, indicated by the Pielou evenness index, Shannon-Wiener diversity index, and Simpson dominance index, showed no statistically significant difference between GE plots and FG plots (Table 2). Similar results had also been reported in the steppe rangelands of the Central Anatolian Region in Turkey (Firincioğlu, Seefeldt & Şahin, 2007) and in the temperate semidesert rangelands of Nevada in North America (Courtois, Perryman & Hussein, 2004). However, the negative consequences for biodiversity after long-term grazing exclusion have also been found in many types of grassland ecosystems (Schultz, Morgan & Lunt, 2011; Maccherini & Santi, 2012). Therefore, there is no general agreement about the species diversity response to grazing exclusion in grassland ecosystems. On one hand, changes in plant species diversity due to grazing or grazing exclusion depend on resource partitioning and competitive patterns in vegetation; for instance, some species with lower competitive ability are reduced in density or disappear from the plant community entirely because of competition, light resources or nutrient availability (Grime, 1998; Van der Wal et al., 2004). On the other hand, the biodiversity response also depends on regional variation in major habitat characteristics, such as soil fertility, soil water availability, and growing-season precipitation (Olff & Ritchie, 1998; Wu et al., 2012; Wu et al., 2014).

A comparison of the community characteristics among the three alpine grassland types showed that total cover, Simpson index, Shannon index, and Pielou index were not significantly different between FG plots and GE plots in all three grassland types, except for that grazing exclusion resulted in the community vegetation heights increasing by 3.01 cm and 2.74 cm in alpine meadow and alpine steppe (P < 0.05), respectively (Table 2). Furthermore, statistical analyses showed that grazing exclusion had a significant effect on vegetation total cover and vegetation height, but grazing exclusion did not affect biodiversity indices (Table 3). This result indicated that short-term grazing exclusion resulted in increase of vegetation growth, but did not lead to obvious change in community composition in degraded alpine grassland ecosystems. The main differences of the plant community characteristics mainly come from the growing season climate differences of alpine grasslands (Table 3).

Impacts of grazing exclusion on biomass

Biomass is often considered a good approximation of productivity, especially in grassland communities (Chiarucci et al., 1999). The aboveground biomass of the GE plots was 31.82% higher than those of the FG plots (P < 0.05, Table 2). Therefore, the grazing exclusion resulted in obvious improvements in community aboveground biomass of degraded alpine grassland. Previous studies found that grazing exclusion significantly increased the total above-ground biomass of alpine meadows in the Tibetan Plateau, and in the fenced meadow, four functional groups, including the grass species group, the sedge species group, the leguminous species group and the noxious species group showed an increase in biomass, whereas only the forbs species group showed a decrease (Zhou et al., 2006; Wu et al., 2009). Wu et al. (2013) found that grazing exclusion increased total aboveground biomass by 27.09% in the Changtang region of Tibet. Their results are strongly consistent with the results of this study. The distinct and positive effect of grazing exclusion on biomass is mainly attributed to the absence of disturbance from herbivorous livestock (Mata-González et al., 2007; Wu et al., 2009); it may secondarily be attributed to the improvement of soil conditions (soil organic carbon and nitrogen storage, water infiltration rate, basal soil respiration, temperature, and moisture) after grazing exclusion, which favours the regeneration and the development of herbaceous species (*Zhao et al.*, 2011; Mofidi et al., 2013).

Among three alpine grassland types, for alpine meadows, the biomass indices, including the mean values of the aboveground, belowground, and total biomass, tended to be higher in GE plots compared to FG plots, but the difference between then were not statistical significant. Nevertheless, for alpine steppe, the aboveground, belowground, and total biomass were all significantly higher due to grazing exclusion; For alpine desert steppe, only the aboveground biomass was significantly higher in fenced plots. *Wu et al.* (2013) also investigated the effect of grazing exclusion on alpine grasslands in the same region in Tibet, and found that grazing exclusion tended to increase aboveground biomass 17.80% in alpine meadow, 34.78% in alpine steppe, and 12.99% in alpine desert steppe, respectively; although these biomass values were also not statistical significant different from those of free grazed grasslands. However, grazing exclusion resulted in the improvement of aboveground biomass in whole alpine grasslands (alpine meadow + alpine steppe + alpine desert steppe) across regional scale in Tibet through the results from both *Wu et al.* (2013) and our study (Table 2).

There are increasing evidences show that precipitation plays a key role in the spatial distribution of species richness and diversity, primary production, and carbon and water cycles of alpine grassland ecosystems in this region (*Hu et al.*, 2010; *Yang et al.*, 2010; *Wu et al.*, 2012; *Wu et al.*, 2013). The precipitation gradients distributions control vegetation growth and community composition were also found in the present study which GSP had significant effect on all biomass indices of alpine grasslands, as well as relative community characteristic indices (Table 3). In fact, the similar results were also reported by *Wu et al.* (2012) and *Wu et al.* (2014)) in this region, therefore, these potential shift of GSP in

Tibet should be considered when recommending any policies designed for the vegetation restoration of degraded alpine grasslands in the future.

The values of the aboveground, belowground, and total biomass were positively correlated with total vegetation cover in the alpine grasslands of the Tibet (Table 4). In addition, the total vegetation cover of alpine grasslands increased after continuous grazing exclusion (Table 2). Therefore, it is suggested that the higher biomass in GE plots was due to the increased vegetation cover. Other studies demonstrated that the grassland biomass and vegetation cover could simultaneously decrease or increase with grazing or not (Gao et al., 2009; Li et al., 2011). The biomass and vegetation cover simultaneously increased due to grazing exclusion was because of the absence of disturbance from herbivorous livestock (Jeddi & Chaieb, 2010), and also because of changes in plant competition and reproduction (Jing, Cheng & Chen, 2013). Moreover, the higher values of aboveground biomass and coverage of certain dominant species in communities under grazing exclusion would result in changes in species' dominance and community composition (Wu et al., 2013). These results were partially validated and expanded upon in our study which the species diversity slightly declined in GE plots with the increasing of grassland biomass and vegetation cover (Table 2). Furthermore, the vegetation cover was negatively correlated with plant biodiversity indicators, D, H and E(P < 0.01) and the aboveground biomass of alpine grassland was negatively correlated with E(P < 0.01) (Table 4).

CONCLUSIONS

The restoration of degraded grassland ecosystem is a complex and long-term ecological process (Gao et al., 2014; Jing et al., 2014). Six to eight years of grazing exclusion in Tibet has not changed species diversity as indicated by the Pielou evenness index, Shannon-Wiener diversity index, and Simpson dominance index, but has significantly improved total vegetation cover, the vegetation height of community and the aboveground biomass of degraded alpine grasslands. These results demonstrate that grazing exclusion is an effective measure for maintaining community stability and improving aboveground vegetation growth in alpine grasslands. Nevertheless, it is worth mentioning that from the ANCOVA analysis, the growing season precipitation (GSP) had a significant effect on all vegetation indicators, except for vegetation height; but grazing exclusion only significantly affected vegetation cover, vegetation height and aboveground biomass (Table 3). Therefore, the GSP plays a more important role than grazing exclusion in which influence on plant community characteristics and biomass in alpine grasslands. In addition, the improvement of the vegetation cover, height and aboveground biomass due to the absence of disturbance from herbivorous livestock in the present study come from the examination short-term (6–8 years) effects of grazing exclusion, so it is questionable whether these improvements will be continuable if grazing exclusion is continuously implemented. Long term observations may be necessary to assess the ecological effects of the grazing exclusion management strategy on degraded alpine grasslands in Tibet. Thus, there is a need for continued research on the role of fencing on grassland restoration, management, and utilization in future.

ADDITIONAL INFORMATION AND DECLARATIONS

Funding

This study was funded by the National Natural Science Foundation of China (41201053), the 135 Strategic Programs of the Institute of Mountain Hazards and Environment (sds-135-1203-03), and the Action Plan of the Chinese Academy of Sciences (CAS) for Western Development (KZCX2-XB3-08). The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Grant Disclosures

The following grant information was disclosed by the authors:

National Natural Science Foundation of China: 41201053.

135 Strategic Programs of the Institute of Mountain Hazards and Environment: sds-135-1203-03.

Action Plan of the Chinese Academy of Sciences for Western Development: KZCX2-XB3-08.

Competing Interests

The authors declare there are no competing interests.

Author Contributions

- Yan Yan performed the experiments, wrote the paper, reviewed drafts of the paper.
- Xuyang Lu conceived and designed the experiments, analyzed the data, contributed reagents/materials/analysis tools, prepared figures and/or tables, reviewed drafts of the paper.

Supplemental Information

Supplemental information for this article can be found online at http://dx.doi.org/10.7717/peerj.1020#supplemental-information.

REFERENCES

- Chen B, Zhang X, Tao J, Wu J, Wang J, Shi P, Zhang Y, Yu C. 2014. The impact of climate change and anthropogenic activities on alpine grassland over the Qinghai-Tibet Plateau. *Agricultural and Forest Meteorology* 189–190:11–18 DOI 10.1016/j.agrformet.2014.01.002.
- **Chiarucci A, Wilson JB, Anderson BJ, De Dominicis V. 1999.** Cover *versus* biomass as an estimate of species abundance: does it make a difference to the conclusions? *Journal of Vegetation Science* **10**:35–42 DOI 10.2307/3237158.
- **Courtois DR, Perryman BL, Hussein HS. 2004.** Vegetation change after 65 years of grazing and grazing exclusion. *Journal of Range Management* **57**:574–582 DOI 10.2307/4004011.
- Dai F, Su Z, Liu S, Liu G. 2011. Temporal variation of soil organic matter content and potential determinants in Tibet, China. *Catena* 85:288–294 DOI 10.1016/j.catena.2011.01.015.

- **Deléglise C, Loucougaray G, Alard D. 2011.** Spatial patterns of species and plant traits in response to 20 years of grazing exclusion in subalpine grassland communities. *Journal of Vegetation Science* **22**:402–413 DOI 10.1111/j.1654-1103.2011.01277.x.
- **Du M, Kawashima S, Yonemura S, Zhang X, Chen S. 2004.** Mutual influence between human activities and climate change in the Tibetan Plateau during recent years. *Global and Planetary Change* **41**:241–249 DOI 10.1016/j.gloplacha.2004.01.010.
- **Firincioğlu HK, Seefeldt SS, Şahin B. 2007.** The effects of long-term grazing exclosures on range plants in the Central Anatolian Region of Turkey. *Environmental Management* **39**:326–337 DOI 10.1007/s00267-005-0392-y.
- Gai JP, Christie P, Cai XB, Fan JQ, Zhang JL, Feng G, Li XL. 2009. Occurrence and distribution of arbuscular mycorrhizal fungal species in three types of grassland community of the Tibetan Plateau. *Ecological Research* 24:1345–1350 DOI 10.1007/s11284-009-0618-1.
- **Gao QZ, Li Y, Xu HM, Wan YF, Jiangcun WZ. 2014.** Adaptation strategies of climate variability impacts on alpine grassland ecosystems in Tibetan Plateau. *Mitigation and Adaptation Strategies for Global Change* 19:199–209 DOI 10.1007/s11027-012-9434-y.
- **Gao YH, Schuman M, Chen H, Wu N, Luo P. 2009.** Impacts of grazing intensity on soil carbon and nitrogen in an alpine meadow on the eastern Tibetan Plateau. *Journal of Food Agriculture and Environment* **7**:749–754.
- Gao Q, Wan Y, Xu H, Li Y, Jiangcun W, Borjigidai A. 2010. Alpine grassland degradation index and its response to recent climate variability in Northern Tibet, China. *Quaternary International* 226:143–150 DOI 10.1016/j.quaint.2009.10.035.
- **Grime JP. 1998.** Benefits of plant diversity to ecosystems: immediate, filter and founder effects. *Journal of Ecology* **86**:902–910 DOI 10.1046/j.1365-2745.1998.00306.x.
- **Hu ZM, Yu GR, Fan JW, Zhong HP, Wang SQ, Li SG. 2010.** Precipitation-use efficiency along a 4500-km grassland transect. *Global Ecology and Biogeography* **19**:842–851 DOI 10.1111/j.1466-8238.2010.00564.x.
- **Jeddi K, Chaieb M. 2010.** Changes in soil properties and vegetation following livestock grazing exclusion in degraded arid environments of South Tunisia. *Flora* **205**:184–189 DOI 10.1016/j.flora.2009.03.002.
- Jing Z, Cheng J, Chen A. 2013. Assessment of vegetative ecological characteristics and the succession process during three decades of grazing exclusion in a continental steppe grassland. *Ecological Engineering* 57:162–169 DOI 10.1016/j.ecoleng.2013.04.035.
- Jing Z, Cheng J, Su J, Bai Y, Jin J. 2014. Changes in plant community composition and soil properties under 3-decade grazing exclusion in semiarid grassland. *Ecological Engineering* 64:171–178 DOI 10.1016/j.ecoleng.2013.12.023.
- Kang L, Han X, Zhang Z, Sun OJ. 2007. Grassland ecosystems in China: review of current knowledge and research advancement. *Proceedings of the Royal Society B: Biological Sciences* 362:997–1008 DOI 10.1098/rstb.2007.2029.
- **Land Management Bureau of Tibet. 1994.** *Grassland resources in Tibet autonomous region.* Beijing: Sciences Press. (in Chinese).
- Li W, Huang HZ, Zhang ZN, Wu GL. 2011. Effects of grazing on the soil properties and C and N storage in relation to biomass allocation in an alpine meadow. *Journal of Plant Nutrition and Soil Science* 11:27–39 DOI 10.4067/S0718-95162011000400003.
- Li YY, Dong SK, Wen L, Wang XX, Wu Y. 2014. Soil carbon and nitrogen pools and their relationship to plant and soil dynamics of degraded and artificially restored grasslands of the Qinghai–Tibetan Plateau. *Geoderma* 213:178–184 DOI 10.1016/j.geoderma.2013.08.022.

- Luan J, Cui L, Xiang C, Wu J, Song H, Ma Q, Hu Z. 2014. Different grazing removal exclosures effects on soil C stocks among alpine ecosystems in east Qinghai–Tibet Plateau. *Ecological Engineering* 64:262–268 DOI 10.1016/j.ecoleng.2013.12.057.
- Maccherini S, Santi E. 2012. Long-term experimental restoration in a calcareous grassland: Identifying the most effective restoration strategies. *Biological Conservation* 146:123–135 DOI 10.1016/j.biocon.2011.11.032.
- Ma M, Zhou X, Du G. 2013. Effects of disturbance intensity on seasonal dynamics of alpine meadow soil seed banks on the Tibetan plateau. *Plant and Soil* 369:283–295 DOI 10.1007/s11104-012-1560-5.
- Mayer R, Kaufmann R, Vorhauser K, Erschbamer B. 2009. Effects of grazing exclusion on species composition in high-altitude grasslands of the Central Alps. *Basic and Applied Ecology* **10**:447–455 DOI 10.1016/j.baae.2008.10.004.
- Mata-González R, Figueroa-Sandoval B, Clemente F, Manzano M. 2007. Vegetation changes after livestock grazing exclusion and shrub control in the southern Chihuahuan Desert. *Western North American Naturalist* 67:63–70 DOI 10.3398/1527-0904(2007)67[63:VCALGE]2.0.CO;2.
- Mofidi M, Jafari M, Tavili A, Rashtbari M, Alijanpour A. 2013. Grazing exclusion effect on soil and vegetation properties in Imam Kandi Rangelands, Iran. *Arid Land Research and Management* 27:32–40 DOI 10.1080/15324982.2012.719575.
- Olff H, Ritchie ME. 1998. Effects of herbivores on grassland plant diversity. *Trends in Ecology & Evolution* 13:261–265 DOI 10.1016/S0169-5347(98)01364-0.
- Schultz NL, Morgan JW, Lunt ID. 2011. Effects of grazing exclusion on plant species richness and phytomass accumulation vary across a regional productivity gradient. *Journal of Vegetation Science* 22:130–142 DOI 10.1111/j.1654-1103.2010.01235.x.
- Shang Z, Deng B, Ding L, Ren G, Xin G, Liu Z, Wang YL, Long RJ. 2013. The effects of three years of fencing enclosure on soil seed banks and the relationship with above-ground vegetation of degraded alpine grasslands of the Tibetan plateau. *Plant and Soil* 364:229–244 DOI 10.1007/s11104-012-1362-9.
- Shang ZH, Gibb MJ, Leiber F, Ismail M, Ding LM, Guo XS, Long RJ. 2014. The sustainable development of grassland-livestock systems on the Tibetan plateau: problems, strategies and prospects. *The Rangeland Journal* 36:267–296 DOI 10.1071/RJ14008.
- Shi X, Li XG, Li CT, Zhao Y, Shang Z, Ma Q. 2013. Grazing exclusion decreases soil organic C storage at an alpine grassland of the Qinghai–Tibetan Plateau. *Ecological Engineering* 57:183–187 DOI 10.1016/j.ecoleng.2013.04.032.
- Sun H, Zheng D, Yao T, Zhang Y. 2012. Protection and construction of the national ecological security shelter zone on Tibetan Plateau. *Acta Oceanologica Sinica* 67:1–12.
- Van der Wal R, Bardgett RD, Harrison KA, Stien A. 2004. Vertebrate herbivores and ecosystem control: cascading effects on tundra ecosystems. *Ecography* 27:242–252 DOI 10.1111/j.0906-7590.2004.03688.x.
- Wang G, Qian J, Cheng G, Lai Y. 2002. Soil organic carbon pool of grassland soils on the Qinghai–Tibetan Plateau and its global implication. *Science of the Total Environment* 291:207–217 DOI 10.1016/S0048-9697(01)01100-7.
- Wen L, Dong SK, Li YY, Sherman R, Shi JJ, Liu DM, Wang YL, Ma YS, Zhu L. 2013a. The effects of biotic and abiotic factors on the spatial heterogeneity of alpine grassland vegetation at a small scale on the Qinghai–Tibet Plateau (QTP), China. *Environmental Monitoring and Assessment* 185:8051–8064 DOI 10.1007/s10661-013-3154-y.

- Wen L, Dong S, Li Y, Li X, Shi J, Wang Y, Liu D, Ma Y. 2013b. Effect of degradation intensity on grassland ecosystem services in the Alpine region of Qinghai-Tibetan Plateau, China. *PLoS ONE* 8:e58432 DOI 10.1371/journal.pone.0058432.
- Wei D, Xu R, Wang Y, Wang Y, Liu Y, Yao T. 2012. Responses of CO₂, CH₄ and N₂O fluxes to livestock exclosure in an alpine steppe on the Tibetan Plateau, China. *Plant and Soil* 359:45–55 DOI 10.1007/s11104-011-1105-3.
- Wu G, Du G, Liu Z, Thirgood S. 2009. Effect of fencing and grazing on a Kobresia-dominated meadow in the Qinghai-Tibetan Plateau. *Plant and Soil* 319:115–126 DOI 10.1007/s11104-008-9854-3.
- Wu J, Zhang X, Shen Z, Shi P, Yu C, Song M, Li X. 2012. Species richness and diversity of alpine grasslands on the Northern Tibetan Plateau: effects of grazing exclusion and growing season precipitation. *Journal of Resources and Ecology* 3:236–242.
- Wu J, Zhang X, Shen Z, Shi P, Xu X, Li X. 2013. Grazing-exclusion effects on aboveground biomass and water-use efficiency of alpine grasslands on the northern Tibetan Plateau. Rangeland Ecology & Management 66:454–461 DOI 10.2111/REM-D-12-00051.1.
- Wu J, Shen Z, Shi P, Zhou Y, Zhang X. 2014. Effects of grazing exclusion on plant functional group diversity of alpine grasslands along a precipitation gradient on the northern Tibetan Plateau. *Arctic Antarctic and Alpine Research* 46:419–429 DOI 10.1657/1938-4246-46.2.419.
- Yang YH, Fang JY, Fay PA, Bell JE, Ji CJ. 2010. Rain use efficiency across a precipitation gradient on the Tibetan Plateau. *Geophysical Research Letters* 37:Article L15702 DOI 10.1029/2010GL043920.
- Zhang WN, Ganjurjav H, Liang Y, Gao QZ, Wan YF, Li Y, Baima YZ, Xirao ZM. 2015. Effect of a grazing ban on restoring the degraded alpine meadows of Northern Tibet, China. *The Rangeland Journal* 37:89–95 DOI 10.1071/RJ14092.
- **Zhao LP, Su JS, Wu GL, Gillet F. 2011.** Long-term effects of grazing exclusion on aboveground and belowground plant species diversity in a steppe of the Loess Plateau, China. *Plant Ecology and Evolution* **144**:313–320 DOI 10.5091/plecevo.2011.617.
- **Zhou H, Tang Y, Zhao X, Zhou L. 2006.** Long-term grazing alters species composition and biomass of a shrub meadow on the Qinghai -Tibet Plateau. *Pakistan Journal of Botany* **38**:1055–1069.
- Zhou H, Zhao X, Tang Y, Gu S, Zhou L. 2005. Alpine grassland degradation and its control in the source region of the Yangtze and Yellow Rivers, China. *Grassland Science* 51:191–203 DOI 10.1111/j.1744-697X.2005.00028.x.
- Zou XY, Li S, Zhang CL, Dong GR, Dong YX, Yan P. 2002. Desertification and control plan in the Tibet Autonomous Region of China. *Journal of Arid Environments* 51:183–198 DOI 10.1006/jare.2001.0943.