

The effects of fencing on biomass and carbon storage in the grasslands of various counties at Liaoning, northeastern China

Efectos de clausurar áreas al acceso de herbívoros domésticos sobre el almacenamiento de biomasa y carbono en los pastizales naturales de varias localidades en la Provincia de Liaoning, al noreste de China

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Abstract

Grasslands play a major role in carbon (C) storage (i.e., sequestration). The objectives of this study were to determine (1) the effects of grassland fencing on aboveground, root and total plant (aboveground + litter + belowground) biomasses, and aboveground, root, total plant, total soil and grassland C storages in various counties of the Liaoning Province, China, and (2) the relationship between total plant biomass and total plant C storage on fenced and unfenced areas in various counties at Liaoning. Results showed that fenced areas had a greater (p<0.05) total plant biomass (> 60%) and C storage (> 64%), and total soil C storage (> 47%), and a 50% greater (p<0.05) total grassland C storage than unfenced areas in all studied counties. Mean carbon storages for all studied counties decreased > 37% on roots and > 6% in the soil as soil depth increased. There was a linear, positive relationship between total plant biomass and total plant C storage in all studied counties (R2> 0.95, p<0.05). Our results indicate that fencing may be an important management strategy for restoring lightly or moderately degraded grassland in Liaoning.

Resumen

Los pastizales naturales cumplen un rol importante en el almacenaje (es decir, secuestro) de carbono (C). Los objetivos de este estudio fueron determinar: (1) los efectos de clausurar pastizales naturales al acceso de herbívoros domésticos sobre las biomasas aéreas, radicales y de toda la planta (aérea + broza + raíces), y sobre los almacenamientos de C en las partes aérea y radical, en toda la planta y el suelo, y en todo el pastizal en varias localidades de la Provincia de Liaoning, China, y (2) la

MULTEQUINA 30(1): 67-98, 2021



relación entre la biomasa versus el almacenamiento de C de toda la planta en áreas clausuradas o no al acceso de los herbívoros domésticos en varias localidades en Liaoning. Las áreas clausuradas tuvieron una mayor (p<0.05) biomasa (> 60%) y almacenamiento de C (> 64%) de toda la planta, un mayor almacenaje de C en todo el suelo estudiado (> 47%), y un 50% más (p<0.05) de almacenaje de C en todo el pastizal que las áreas no clausuradas en todas las localidades estudiadas. En todas las localidades estudiadas, el almacenamiento promedio de C disminuyó > 37% en el sistema radical y > 6% en el suelo a medida que se incrementó la profundidad del mismo. Hubo una relación lineal, positiva entre la biomasa y el almacenamiento de C en toda la planta en todas las localidades (R2> 0.95, p<0.05). Nuestros resultados indican que clausurar áreas al pastoreo puede ser una importante estrategia de manejo para restaurar pastizales ligera o moderadamente degradados en Liaoning.

Keywords: biomass, C storage, fencing, grassland, Liaoning

INTRODUCTION

Grasslands play an important role in global carbon (C) storage, and the potential to sequester C is quite different among different world's ecosystems (López-Fando & Pardo 2011). Grasslands, which are one of the most widely distributed ecosystems on Earth (Fang et al., 2010), contain a large part of the terrestrial biosphere's C (Cheng et al., 2011). Indeed, incorrect grazing management of grasslands may decrease the global C storage both directly and indirectly (Li et al., 2013). Minor alterations in grasslands may have outstanding effects on C storage (Deng et al., 2016). In China, the total national land is covered approximately 44% by grasslands, accounting for about 9-16% of the world's grassland C storage (Ni, 2002). As alpine grasslands of the Qinghai-Tibetan Plateau play a key role in the terrestrial carbon cycle (Zhao et al., 2006), montane meadows in the Liaoning Province, China, might also play an important role in C storage. In recent decades, part of the grasslands of Liaoning have been se**Palabras clave** : biomasa, almacenaje de C, clausura de áreas al acceso de herbívoros domésticos, pastizal natural, Liaoning

riously degraded by some environmental changes and human activities (Harris, 2010). Grassland degradation has seriously led to a loss of soil organic C (SOC) in northwest China (Li et al., 2019). The loss of C has disadvantageous effects on soil nutrients and soil structure, leading to global warming by increasing the C dioxide concentration in the atmosphere (Xiong et al., 2016). It is critical to restore the degraded grasslands to decrease the environmental problems such as the soil C loss to the atmosphere and subsequent climatic warming (Paustian et al., 2016). Therefore, large scale fencings to livestock grazing based on the "Self-design (Natural Restoration)" theory have turned into a significant way to abate grassland degradation (Li et al., 2019).

Soil C storage is a function of the balance between the inputs from aboveground and belowground biomasses and the losses because of the decomposition of these biomasses (Pendall et al., 2011). Carbon contained in the grassland biomass in the Liaoning Province is an important component of the total regional C budget, and changes in the

species composition influence not only the type but also the availability of aboveground C (Steinbeiss et al., 2008). Carbon storage in the aboveground parts of vegetation is a time-integrated expression of processes ranging from photosynthesis and nutrient cycling to disturbance and climate change (Asner et al., 2012). Belowground biomass C is also a necessary component of the C cycle in grassland ecosystems and an important element in soil C sequestration (Li et al., 2019). Soil microorganisms can also affect various soil properties, and are responsible for the transformation and cycling of plant nutrients and soil organic matter, thus also affecting C sequestration (Li et al., 2013). Soil microbial biomass carbon can also be significantly affected by land-use practices (Li et al., 2013). However, the effects of grassland degradation and management strategies are inconsistent and variable on soil C storage, depending on the system. Herbivores, for example, may facilitate or reduce C sequestration rates (He et al., 2011; Silva et al., 2019).

Some researchers have studied soil organic C contents on fenced versus unfenced grasslands in Inner Mongolia, China (Li et al., 2013). However, relatively few studies have clearly described the effects of fenced versus unfenced grasslands on C sequestration in the Liaoning Province. We assume that restoration processes occurring on grassland enclosures (i.e., fencing) to domestic livestock may affect the stocks of C in those grasslands by affecting the carbon contents in the aboveground, belowground and litter biomasses, and in the soil. We hypothesize that total plant biomass (i.e., aboveground + root + litter), and total plant and soil C storages are greater on fenced than unfenced areas, and differ between counties in the Liaoning Province. The objectives of this study were to determine (1) the effects of grassland fencing on aboveground, root and total plant biomasses; and aboveground, root, total plant and soil C storages at various counties of the Liaoning Province, and (2) determine the relationship between total plant biomass and total plant C storage on fenced and unfenced areas in various counties at Liaoning.

MATERIALS AND METHODS

Site description

This study was conducted in the Desertification Governance Base of six counties of Liaoning province in China. The six counties were Yixian (YX), Lingyuan (LY), Kazuo (KZ), Jianchang (JC), Beipiao (BP) and Jianping (JP) (41°02'N-42°16' N, 119°36' E-121°23' E; Figure 1). The climate is characterized by cool summers and long, cold winters. The mean daily temperature is 20 °C, ranging from 15 °C to 30 °C in August. Mean annual temperatures were 8.5 °C in Yixian, 7.1 °C in Lingyuan, 7.2 °C in Kazuo, 7.3 °C in Jianchang, 7.5 °C in Beipiao, and 6.7 °C in Jianping. Mean annual precipitations were 562 mm in Yixian, 601 mm in Lingyuan, 561 mm in Kazuo, 610 mm in Jianchang, 475 mm in Beipiao, and 531 mm in Jianping.

The experimental area had been exposed to long-term heavy grazing before 2011. As a result, grasslands were severely degraded, as indicated by their low vegetation cover. Since early 2011, exclosures were established to prevent herbivory grazing at each study county (i.e., fencing). Unfenced areas were used as a control. Further characteristics of the study site are described in **Tables 1** and **2**.



Figure 1. Diagrammatic representation illustrating the location of each of the six studied counties in the Liaoning Province, China. L1= Yixian; L2= Lingyuan; L3= Kazuo; L4=Jian-chang; L5= Beipiao; L6= Jianping

Figura 1. Diagrama que ilustra las 6 localidades estudiadas en la Provincia de Liaoning, China (L1= Yixian; L2= Lingyuan; L3= Kazuo; L4=Jianchang; L5= Beipiao; L6= Jianping)

Experimental procedures

At each county, samples were taken within the exclosures, and were compared to those exposed to all-year-round grazing outside those exclosures. The vegetation was sampled in August (late summer) 2011. At this time of the year, grassland plants reached their peak growth accumulation since the beginning of the growing season. In each study area of the same county (i.e., treatments), five $1 \times 1 \text{ m}^2$ randomly distributed plots were established. Immediately after plot establishment, the soil surface within each plot was cleaned from any residue. Afterwards, the number of plants was counted, and plant species composition and cover determined within each quadrat during August following Mueller-Dombois &

 Table 1. Longitude, Latitude, Altitude, Dominant soil characteristic, Vegetation type and Utilization method (fenced, unfenced) at each of the studied counties

Tabla 1. Longitud, Latitud, Altitud, Característica dominante del suelo, tipo de vegetación y
método de utilización (áreas clausuradas o no al acceso de herbívoros domésticos) en cada una de
las localidades estudiadas

Site	Longitude	Latitude	Altitude (m)	Dominant soil characteristic	Vegetation type	Utilization method
Yixian	121°05' E	41°41' N	194	DB	VF and Weed	Fenced
	120°90' E	41°32' N	136	DB	Weed	Unfenced
Beipiao	120°51' E	41°82' N	407	MASS	VF and Weed	Fenced
	121°23' E	41°81' N	384	CS	Weed	Unfenced
Jianchang	119°92' E	40°89' N	352	MASS	VF and Weed	Fenced
	119°72' E	40°56' N	301	MASS	VF and Weed	Unfenced
Kazuo	119°64' E	41°25' N	533	MASS	VF and S	Fenced
	119°65' E	41°20' N	457	MASS	Weed	Unfenced
Jianping	119°44' E	41°42' N	617	MS	S and Weed	Fenced
	119°45' E	42°16' N	799	MS	LCT and SG	Unfenced
Lingyuan	119°43' E	41°03' N	479	DB	S	Fenced
	119°36' E	41°02' N	621	DB	CV and Weed	Unfenced

DB is dark brown, MASS is meadow aeolian sandy soil, CS is chestnut soil, MS is meadow soil VF is Vervain Family, CV is *Chloris virgata*, S is Sedge, LCT is *Leymus chinensis* Tzvel., SG is *Stipa grandis*

Ellenberg (1974). After this measurement, we cut all plants to 5 cm height in each quadrat using scissors, and collected the plant litter on the soil surface, to measure aboveground biomass following Ma et al. (2002). To measure the below-ground biomass, a 9-cm-diameter root auger was used to take one soil sample for each depth range of 0-10 cm, 10-20 cm, and 20-30 cm in each 1 m x 1 m quadrat. The roots found in the soil samples were isolated using a sieve (2 mm, 0,5 mm). All isolated roots as well as the soil were oven dried at 65 °C for 24 h and weighed.

A soil sampling drill of a 5.046-cm-inner-diameter (S1 Canada) was used to obtain an undisturbed soil core from 0 to 30 cm on each of the five 1 x 1 m quadrats; soil subsamples from 0 to 30 cm in depth were taken every 10 cm. The roots were separated from the soil samples by sieving through a 2 mm mesh screen. The soil samples without roots and debris were air dried and stored for analyzing soil physico-chemical characteristics.

For the soil bulk density, a 5.046-cm inner-diameter soil sampling drill (S1 Canada) was used to obtain an undisturbed soil core from 0 to 30 cm. Thereafter, the soil bulk density (g cm³) in the undisturbed soil cores from the fenced and unfenced quadrats was measured by the soil bulk sampler method (Pennock et al., 2008). After sieving the soil through a 2-mm-mesh screen to remove roots and other residues, it was oven dried to constant weight at 105 °C. **Table 2**. Species composition and total mean cover (%) for each species on fenced and unfenced areas in six counties of the Liaoning Province, China, in early 2011. Each value of total cover for each species was the mean of n=5

Tabla 2. Composición de especies y cobertura total promedio (%) para cada especie en áreas clausuradas o no al acceso de herbívoros domésticos en seis localidades de la Provincia de Liaoning, China, a principios de 2011. Cada valor de cobertura total para cada especie fue el promedio de n=5

Site	Utilization method	Species	Total coverage (%)
Yixian	Fenced	Vitex negundo var. heterophylla	85.00
		Carex neurocarpa	90.00
		Portulaca oleracea	82.00
		Chloris virgata	83.00
	Unfenced	Digitaria sanguinalis	78.33
		Artemisia sacrorum Ledeb	85.00
Jianping	Fenced	Carex humilis var. nana	93.33
		Astragalus dahuricus	92.50
		Artemisia mongolica	95.00
	Unfenced	Lespedeza juncea	94.00
		Artemisia mongolica	95.00
		Cleistogenes squarrosa	94.00
		Thymus mongolicus	94.00
Lingyuan	Fenced	Carex callitrichos	92.33
		Lespedeza juncea	93.50
		Vitex negundo var. heterophylla	92.00
		Artemisia gmelinii	92.00
		Cleistogenes caespitosa	90.00
	Unfenced	Potentilla discolor	75.00
		Chloris virgata	76.67
		Artemisia gmelinii	77.50
Kazuo	Fenced	Carex callitrichos	97.33
		Lespedeza juncea	97.33
	Unfenced	Salsola collina	77.50
		Setaria viridis	73.33
		Poa annua	70.00
		Artemisia gmelinii	65.00

Site	Utilization method	Species	Total coverage (%)
Jianchang	Fenced	Lespedeza juncea	80.00
		Artemisia gmelinii	87.00
		Cleistogenes caespitosa	85.00
		Carex neurocarpa	88.75
		Vitex negundo var. heterophylla	90.00
		Amphicarpaea edgeworthii	85.00
		Sanguisorba officinalis	88.00
	Unfenced	Chloris virgata	82.50
		Cleistogenes caespitosa	80.00
		Artemisia gmelinii	82.80
		Carex neurocarpa	83.00
		Lespedeza juncea	85.00
		Vitex negundo var. heterophylla	83.00
		Sanguisorba officinalis	84.00
		Salsola collina	84.50
		Cleistogenes squarrosa	85.00
Beipiao	Fenced	Lespedeza juncea	78.50
		Artemisia scoparia	83.00
		Setaria viridis	81.67
		Vitex negundo var. heterophylla	83.25
		Miscanthus sacchariflorus	88.00
		Puccinellia tenuiflora	88.00
		Bothriochloa ischaemum	82.00
	Unfenced	Leymus chinensis	92.80
		Artemisia scoparia	92.80
		Lespedeza juncea	94.00
		Potentilla discolor	92.00

Soil pH was measured by an acidity agent (PHS-3C pH acidometer, China). Total soil and plant N concentrations were evaluated by the Kjeldahl method (Bremner, 1996).

Determination of organic C concentrations

After measuring aboveground and root biomasses, plant samples were pulverized using a ball mill. Concentrations of aboveground plant C (APC), litter C (LC), root C (BRC), and organic soil C (OSC) were all analyzed by the potassium dichromate oxidation external heating method (GB9834-1988).

Formulas

We calculated the Aboveground Plant C Storage (APCS; Mg/ha); Litter C Storage (LCS; Mg/ha); Total Aboveground C Storage (TACS; Mg/ha); Total Root C Storage (TRCS; Mg/ha); Total Plant C Storage (TPCS; Mg/ha); Total Soil C Storage (TSCS; g/m⁻²); Total Grassland C Storage (TGCS;Mg/ha) on a per unit area basis up to 30 cm soil depth, and Total Grassland C Storage(TGCS;Tg) using the following formulas:

TAB=SCB+LB TRB= Σ RBi (from hi = 1 to 3) APCS=TAB *APC LCS =LB * LC TACS=APCS+LCS RCSi= RBi * RCi TRCS= Σ RCSi (from hi = 1 to 3) TPCS=TACS+TRCS OSCSi= OSCi * SBSi * hi (from hi = 1 to 3) TSCS= Σ OSCSi (from hi = 1 to 3) TGCS= TSCS+TPCS TGCS= TGCS *S/108 where TAB is total aboveground biomass (g/m²); SCB is total standing crop biomass (g/m^2) ; LB is litter biomass (g/m^2) ; TRB is total root biomass (g/m²); hi is soil depth (i.e., 1=0-10, 2=10-20, 3=20-30 cm); **RBi** is the root biomass of the ith soil layer (g/m⁻³); APC is aboveground plant C concentration (%); APCS is aboveground plant C storage (Mg/ha); LC is litter C concentration (%); LCS is litter C storage (g/m²); TACS is total aboveground C storage; RCi is root C concentration of ith soil layer (%); RCSi is root C storage of ith soil layer (Mg/ ha), TRCS is total root C storage (Mg/ ha); TPCS is total plant C storage (Mg/ ha); OSCi is organic soil C concentration of ith soil layer (%); SBSi is soil bulk storage of the ith soil layer (g/m^{-3}) ; OSCSi is organic soil C storage of ith soil laver (Mg/ha); TSCS is total soil C storage (Mg/ha); TGCD is total grassland C storage per unit surface area (Mg/ha); S is fenced area (ha); TGCS is total grassland C storage (Tg).

Statistical analysis

Treatments were analyzed following a completely randomized design using two-way ANOVA for comparing fencing level (i.e., fenced and unfenced areas) versus counties for various variables, and three-way ANOVA for comparing variable values at each of the three studied soil depths. When F tests were significant (p < 0.05), differences among means were determined using the LSD test of Fisher (Snedecor and Cochran, 1980). All statistical analyses were performed using SPSS21.

RESULTS

Soil physicochemical characteristics

There was a significant (p<0.05) interaction between Utilization Method x County for soil bulk density, soil pH, and soil N concentrations. At the same time, the interaction between Utilization Method x County was not significant (p>0.05) for soil C concentrations.

Soil bulk density

Soil bulk density was lower (p<0.05) on fenced than unfenced areas in 3 out of the 6 counties (i.e., Lingyuan, Kazuo and Jianping; **Table 3**). In 2 (i.e., Yixian and Jianchang) out of the 6 counties, soil bulk density was similar (p>0.05) on fenced and unfenced areas. On fenced areas, Beipiao showed the greatest (p<0.05) soil bulk density in comparison to the other 5 counties. These last counties showed a similar (p>0.05) soil bulk density. The

Table 3. Soil bulk density (g/cm3), soil pH and soil N concentrations on fenced and unfenced areas at each of the studied counties. There was interaction between Utilization method x Counties for each of the shown soil variables. Each value is the mean of n=5. Different letters to the left of the comma indicate significant differences (p<0.05) between utilization methods within each county, and those to the right of it indicate significant differences (p<0.05) among counties within each utilization method

Tabla 3. Densidad de volumen de suelo (g/cm3), pH y concentraciones de N del suelo en áreas clausuradas o no al acceso de herbívoros domésticos en cada una de las localidades estudiadas. Hubo una interacción entre Método de utilización x Localidades para cada una de las variables de suelo mostradas. Cada valor es el promedio de n=5. Letras diferentes a la izquierda de la coma indican diferencias significativas (p<0.05) entre métodos de utilización dentro de cada localidad, y aquellas a la derecha de la coma indican diferencias significativas (p<0.05) entre localidades dentro de cada método de utilización

Counties	Utilization method	Soil bulk density (g/cm ³)	pН	Soil nitrogen (%)
Yixian	Fenced	1.265±0.049 a,b	7.312±0-089 a,b	0.087±0.002 a,c
	Unfenced	1.246±0.069 a,bc	7.384±0.095 a,d	0.045±0.001 b,ab
Lingyuan	Fenced	1.211±0.039 a,b	7.186±0.048 a,b	0.053±0.008 a,e
	Unfenced	1.362±0.077 b,ab	7.628±0.103 b,c	0.045±0.002 a,ab
Kazuo	Fenced	1.150±0.106 a,b	7.840±0.022 a,a	0.076±0.019 a,cd
	Unfenced	1.357±0.079 b,ab	7.856±0.038 a,b	0.042±0.004 b,ab
Jianchang	Fenced	1.102±0.056 a,b	7.176±0.071 a,b	0.061±0.008 a,de
	Unfenced	1.239±0.069 a,bc	7.368±0.052 b,d	0.044±0.003 a,ab
Beipiao	Fenced	1.446±0.073 a,a	7.328±0.097 a,b	0.117±0.002 a,b
	Unfenced	1.186±0.064 b,c	6.584±0.040 b,e	0.036±0.001 b,b
Jianping	Fenced	1.237±0.051 a,b	7.874±0.063 a,a	0.167±0.017 a,a
	Unfenced	1.499±0.080 b,a	8.192±0.026 b,a	0.057±0.005 b,a

greatest and lowest (p<0.05) soil bulk densities were shown at Jianping and Beipiao, respectively, on unfenced areas.

Soil pH

Jianping and Kazuo showed a similar (P>0.05) but greater (p<0.05) soil pH than the other 4 counties on fenced areas (**Table 3**). These last counties had a similar (p>0.05) soil pH on those areas. On unfenced areas, soil pH followed the following order from greatest to lowest (p<0.05): Jianping > Kazuo > Lingyuan > Yixian > Jianchang > Beipiao.

Soil N concentrations

Soil N concentrations were greater (p<0.05) on fenced than unfenced areas on four (i.e., Yixian, Kazuo, Beipiao, Jianping) out of the six counties (**Table 3**). These last counties showed a similar (p>0.05) soil N concentration. The

greatest and lowest (p<0<05) soil N concentrations were found in Jianping and Lingyuan, respectively, on fenced areas. At the same time, the greatest and lowest (p<0.05) soil N concentrations were shown in Jianping and Beipiao, respectively, on unfenced areas.

Soil C concentrations

Soil C concentrations were almost twice as great (p<0.05) on fenced than unfenced areas (**Table 4**). In turn, the greatest and lowest (p<0.05) soil C concentrations were found at Yixian and Jianping, respectively.

Aboveground, root and total plant biomasses

Aboveground biomass

Standing crop biomass

Fencing level and county showed a significant interaction (p=0.001) for stand-

Table 4. Soil C concentrations on fenced versus unfenced areas, and among counties in the Liaoning Province. There was not an interaction (p>0.05) between Utilization method x Counties for the studied variable. Each value is the mean of n=5. Different letters between Utilization methods indicate significant differences (p<0.05) between fenced and unfenced areas. Different letters among counties indicate significant differences (p<0.05) among them. *Tabla 4. Concentraciones de C del suelo en áreas clausuradas versus no clausuradas al acceso de herbívoros domésticos, y entre localidades en la Provincia de Liaoning. No hubo interacción (p>0.05) entre Método de utilización x Localidades para la variable estudiada. Cada valor es el promedio de n=5. Letras diferentes entre métodos de utilización indican diferencias significativas (p<0.05) entre áreas clausuradas o no al acceso de herbívoros domésticos. Letras diferentes entre localidades indican diferencias significativas (p<0.05) entre ellas*

Soil carbon concentrations (%)						
Utilization Method						
		Fenced	2.030 a			
		Unfenced	1.112 b			
Counties						
Yixian	Lingyuan	Kazuo	Jianchang	Beipiao	Jianping	
1.997 a	1.566 b	1.592 b	1.735 ab	1.482 b	1.053 c	

ing crop biomass. Standing crop biomass was at least 57% greater on fenced than unfenced areas in all counties (**Figure 2**). The greatest (p<0.05) standing crop biomasses on fenced areas were found at Yixian and Lingyuan, while those lowest (p<0.05) were shown at Beipiao, Jianchang and Jianping. There were not significant differences (p>0.05) among counties on standing crop biomasses on unfenced areas.

Aboveground litter biomass [3]

The interaction between fencing level and county was not significant (p=0.229) for litter biomass. Litter biomass was 66% greater (p<0.05) at fenced (116.7 g/m²) than unfenced (39.1 g/m²) areas. However, all counties had a similar (p=0.130) litter biomass.

Total aboveground biomass

The interaction between fencing level and county was significant (p=0.013) for total aboveground (standing crop + litter) biomass. Total aboveground biomasses were more than 59% greater (p<0.05) on fenced than unfenced areas at all counties (**Figure 2**). The greatest and lowest (p<0.05) total aboveground biomasses were found at Lingyuan, and Beipiao and Jianping, respectively, on fenced areas. On unfenced sites, total aboveground biomasses were greater (p<0.05) at Xixian, Lingyuan and Kazuo than at Beipiao and Jiangping.

Root biomass

All interactions between fencing level and locality, soil depth x county and soil depth x fencing level were significant at p=0.000. Root biomasses were at least 77% greater (p<0.05) on fenced than unfenced sites at all locations but Lingyuan (**Figure 3**). Root biomasses were similar (p>0.05) at Yixian, Kazuo and Jianchang on fenced areas. These biomasses, however, were greater (p<0.05) than those found at Lingyuan, Beipiao and Jianping, which did not differ (p>0.05) among them at those sites. There were no differences (p>0.05) on root biomasses among counties on unfenced areas.

Root biomasses were greater (p<0.05) at 0-10 than 20-30 cm soil depth at all locations but Lingvuan and Jianpiang. At these locations, there were not significant differences (p>0.05) among soil depths in root biomass. The greatest (p<0.05) root biomass at 0-10 cm soil depth was found at Jianchang. At this soil depth, the lowest (p<0.05) root biomasses were found at Lingyuan, Beipiao and Jianping. Yixian, Lingyuan, Kazuo, Jianchang and Beipiao showed a similar (p>0.05) root biomass at 10-20 cm soil depth. However, root biomass at this soil depth was greater (p<0.05) at Yixian than at Jianping. At 20-30 cm soil depth, root biomass was greater (p<0.05) at Yixian, Kazuo, Jianchiang and Beipiao than at Lingyuan and Jianping.

On fenced sites, the highest (p<0.05) and lowest (p<0.05) root biomasses occurred at 0-10 and 20-30 cm soil depths, respectively. Unfenced sites, however, showed a similar (p>0.05) root biomass at all study soil depths. At 0-10, 10-20 and 20-30 cm soil depths, root biomasses were greater (p<0.05) on fenced than unfenced sites. While root biomasses were above 100 g/m² on fenced sites at all study soil depths, they were below 64 g/m² on unfenced sites at these same soil depths.



Figure 2. Standing crop and total aboveground (standing crop + litter) plant biomasses (g/ m^2) at six counties (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao and L6= Jianping) in the Liaoning Province, northeastern China. Each histogram is the mean of n=5. Different letters to the left above the histograms indicate significant differences (p<0.05) between fenced and unfenced sites within each county, and those to the right of them indicate significant differences (p<0.05) among the studied counties within each fencing level *Figura 2. Biomasas de las plantas en pie y total aéreas (biomasa en pie + broza) (g/m²) en seis localidades (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao y L6= Jianping) en la Provincia de Liaoning al noreste de China. Cada histograma es el promedio de n=5. Letras diferentes a la izquierda encima de los histogramas indican diferencias significativas (p<0.05) entre áreas clausuradas o no al acceso de herbívoros domésticos dentro de cada localidades estudiadas dentro de cada nivel de clausura*



Figure 3. Root biomass (g/m²) at (1) fenced and unfenced sites on each of the studied counties in the Liaoning Province, northeastern China (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao and L6= Jianping); (2) 0-10, 10-20 or 20-30 cm soil depth from the soil surface within each of the studied counties; (3) 0-10, 10-20 or 20-30 cm soil depth from the soil surface within each fencing level. Each histogram is the mean of n=5. Different letters to the left above the histograms indicate significant differences (p<0.05) between (1) fenced and unfenced sites within each county, and those to the right of them indicate significant differences (p<0.05) among the studied counties within each fencing level; (2) different soil depths (0-10, 10-20 or 20-30 cm from the soil surface) within each studied county, and those to the right indicate significant differences (p<0.05) among the counties within each studied soil depth; (3) between soil depths (0-10, 10-20 or 20-30 cm from the soil surface) within each fencing

level, and those to the right of them indicate significant differences (p<0.05) between fenced and unfenced sites within each study soil depth

Figura 3. Biomasa radical (g/m^2) en (1) áreas clausuradas o no al acceso de herbívoros domésticos en seis localidades de la Provincia de Liaoning, al noreste de China (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao y L6= Jianping); (2) 0-10, 10-20 o 20-30 cm de profundidad del suelo desde la superficie del mismo en cada una de las localidades estudiadas; (3) 0-10, 10-20 o 20-30 cm de profundidad del suelo desde la superficie del mismo dentro de cada nivel de clausura. Cada histograma es el promedio de n=5. Letras diferentes a la izquierda de la coma encima de los histogramas indican diferencias significativas (p<0,05) entre (1) áreas clausuradas o no dentro de cada localidad, y aquellas a la derecha de la coma indican diferencias significativas (p<0.05) entre las localidades estudiadas dentro de cada nivel de clausura; (2) diferentes profundidades del suelo (0-10, 10-20 o 20-30 cm desde la superficie del mismo) dentro de cada localidad, suquellas a la derecha de la coma indican diferencias significativas (p<0.05) entre las localidades del suelo (0-10, 10-20 o 20-30 cm desde la superficie del mismo) dentro de cada localidad estudiada, y aquellas a la derecha de la coma indican diferencias significativas (p<0.05) entre las localidades dentro de cada profundidad del suelo estudiada; (3) entre las profundidades del suelo (0-10, 10-20 o 20-30 cm desde la superficie del suelo) dentro de cada nivel de clausura, y aquellas a la derecha de la coma indican diferencias significativas (p<0.05) entre áreas clausuradas o no dentro de cada profundidad del suelo estudiadas significativas (p<0.05) entre áreas clausuradas o no dentro de cada profundidad del suelo estudiada

Total root biomass

There was an interaction (p=0-000) between county and fencing level for total root biomass. Total root biomass was more than 76% greater (p<0.05) on fenced than unfences sites at all counties, but Lingyuan (**Figure 4**). While total root biomass was greater (p<0.05) at Yixian, Kazuo and Jianchang than at Lingyuan, Beipiao and Jianping on fenced sites, all counties showed a similar (p>0.05) total root biomass on unfenced ones.

Total plant biomass

The interaction between fencing and county was significant (p=0.000). Total plant biomass was greater (p<0.05) on fenced than unfenced sites at all coun-



Figure 4. Total root biomass (g/m^2) on fenced and unfenced sites at the various studied counties (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao and L6= Jianping) in the Province of Liagoning, China. Each histogram is the mean of n=5. Different letters to the left above the histograms indicate significant differences (p<0.05) between fenced and unfenced sites within each county, and those to the right of them indicate significant differences (p<0.05) among the studied counties within each fencing level

Figura 4. Biomasa radical total (g/m^2) en áreas clausuradas o no al acceso de herbívoros domésticos en seis localidades de la Provincia de Liaoning, China (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao y L6= Jianping) Cada histograma es el promedio de n =5. Letras diferentes a la izquierda encima de los histogramas indican diferencias significativas (p<0.05) entre áreas clausuradas o no dentro de cada localidad, y aquellas a la derecha de la coma indican diferencias siganificativas (p<0.05) entre las localidades estudiadas dentro de cada nivel de clausura ties (**Figure 5**). Yixian, Kazuo and Jianchang had a similar (p>0.05) total plant biomass on fenced sites. These biomasses were greater (p<0.05) than those at Lingyuan, Beipiao and Jianping at these sites. On unfenced sites, however, there were no significant differences on total plant biomass among counties on unfenced sites.

Aboveground, root, total plant, soil and grassland C storage

Aboveground C storage

Standing crop C storage

There was an interaction between counties and fencing level for standing crop C storage (p=0.004). Fenced sites showed a greater (p<0.05) standing crop C stor-



Figure 5. Total plant biomasses (g/m²) on fenced and unfenced sites at each of the studied counties. (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao and L6= Jianping) in the Province of Liagoning, China. Different letters at the left above of the histograms indicate significant differences (p<0.05) between fencing levels within each of the studied counties, and those to the right of them indicate significant differences (p<0.05) among the studied counties within each fencing level

Figura 5. Biomasas totales de las plantas (g/m^2) en áreas clausuradas o no al acceso de herbívoros domésticos en cada una de las seis localidades de la Provincia de Liaoning, China (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao y L6= Jianping). Letras diferentes a la izquierda encima de los histogramas indican diferencias significativas (p<0.05) entre áreas clausuradas o no al acceso de herbívoros domésticos dentro de cada una de las localidades estudiadas, y aquellas a la derecha de la coma indican diferencias significativas (p<0.05) entre las localidades estudiadas dentro de cada nivel de clausura age than unfenced ones at all counties (**Figure 6**). Standing crop C storage was similar (p>0.05) at Yixian, Jianchang and Jianping on fenced sites. These standing crop C storage on fenced sites were greater (p<0.05) than those at Lingyuan, Kazuo and Beipiao, which did not show significant differences among them. Unfenced sites, however, did not show significant differences (p>0.05) among counties.

Aboveground litter C storage

There was not a significant interaction (p=0.203) between counties and fencing levels for litter C storage. However, litter C storage was more than 75% greater (p<0.05) at fenced than unfenced sites (data not shown). On the other hand, litter C storage was similar (p>0.05) at all counties (mean= 0.28 Mg/ha; data not shown).

Total aboveground C storage

The interaction between locations and fencing levels was significant (p=0.008) for total aboveground C storage (**Figure 6**). Total aboveground C storage was greater (p<0.05) on fenced than unfenced sites at all counties. On fenced sites, the greatest and lowest (p<0.05) total aboveground C storage were found at Jianping, and Lingyuan and Beipiao, respectively. Yixian, Kazuo and Jianchang, however, showed a similar total aboveground C storage on fenced sites. Unfenced sites showed a similar (p>0.05) total aboveground C storage at all counties.

Root C storage

There was an interaction (p<0.05) between counties and fencing levels, counties and soil depths, and fencing levels and soil depths for root C storage (**Figure 7**). Root C storage was greater (p<0.05) on fenced than unfenced sites at all counties. On fenced sites, root C storage was similar (p>0.05) at Yixian, Kazuo and Jianchang. These values, however, were greater (p<0.05) than those found at Lingyuan, Beipiao and Jianping on fenced sites. At these sites, Lingyuan, Beipiao and Jianping showed a similar (p>0.05) root C storage. Root C storage was similar (p>0.05) at all locations on unfenced sites.

There was a decreasing (p<0.05) root C storage with increasing soil depth at Yixian. At Lingyuan, root C storage was similar (p>0.05) at 0-10 and 10-20 cm soil depth. These values, however, were greater (p<0.05) than those found at 20-30 cm soil depth at this location. At Kazuo and Jianchang, root C storage was greater (p<0.05) at 0-10 than at 10-20 and 20-30 cm soil depths. Finally, there were no significant differences (p>0.05) in root C storage among soil depths at Beipiao and Jianping.

While root C storage decreased (p<0.05) with increasing soil depth on fenced sites, there were no significant differences (p>0.05) in root C storage among soil depths on unfenced sites. At all soil depths, root C storage was greater (p<0.05) on fenced than unfenced sites.

The interaction between fencing level and county was significant (p=0,000) for total root C storage (**Figure 8**). Total root C storage was greater (p<0.05) on fenced than unfenced sites on all locations. Yixian, Kazuo and Jianchang had a similar (p>0.05) total root C storage on fenced sites. These values were greater (p<0.05) than those found at Lingyuan, Beipiao and Jianping at these sites. These later locations, however, showed a similar



Figure 6. (1) Standing crop C storage (Mg/ha) and (2) Total aboveground C storage (Mg/ha) on fenced and unfenced sites on each of the studied counties (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao and L6= Jianping) in the Province of Liagoning, China. Each histogram is the mean of n=5. Different letters to the left above the histograms indicate significant differences (p<0.05) between fenced and unfenced sites within each county, and those to the right of them indicate significant differences(p<0.05) among counties within each fencing level

Figura 6. (1) Almacenamiento de C en las plantas en pie (Mg/ha) y (2) Almacenamiento de C en la parte áera total (Mg/ha) en áreas clausuradas o no al acceso de herbívoros domésticos en cada una de las seis localidades estudiadas en la Provincia de Liaoning, China (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao y L6= Jianping). Cada histograma es el promedio de n=5. Letras diferentes a la izquierda encima de los histogramas indican diferencias significativas (p<0.05) entre áreas clausuradas o no al acceso de herbívoros domésticos dentro de cada localidad, y aquellas a la derecha de la coma indican diferencias significativas (p<0.05) entre localidades dentro de cada nival de clausura



Figure 7. Root carbon storage (Mg/ha) at (1) fenced and unfenced sites on each of the studied counties in the Liaoning Province, northeastern China (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao and L6= Jianping); (2) 0-10, 10-20 or 20-30 cm soil depth from the soil surface within each of the studied counties: (3) 0-10, 10-20 or 20-30 cm soil depth from the soil surface within each fencing level. Each histogram is the mean of n=5. Different letters to the left above the histograms indicate significant differences (p<0.05) between (1) fenced and unfenced sites within each county. and those to the right of them indicate significant differences (p<0.05) among the studied counties within each fencing level; (2) different soil depths (0-10, 10-20 or 20-30 cm from the soil surface) within each studied county, and those to the right of them indicate significant differences (p<0.05) among the counties within each studied soil depth; (3) different soil depths (0-

10, 10-20 or 20-30 cm from the soil surface) within each fencing level, and those to the right of them indicate significant differences (p<0.05) between fenced and unfenced sites within each study soil depth

Figura 7. Almacenamiento de C en el sistema radical (Mg/ha) en (1) áreas clausuradas o no al acceso de herbívoros domésticos en cada una de las localidades estudiadas en la Provincia de Liaoning al noreste de China (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao y L6= Jianping); (2) 0-10, 10-20 o 20-30 cm de profundidad del suelo desde la superficie del mismo dentro de cada una de las localidades estudiadas; (3) 0-10, 10-20 o 20-30 cm de profundidad del suelo desde la superficie del mismo dentro de cada nivel de clausura. Cada histograma es el promedio de n=5. Letras diferentes a la izquierda encima de los histogramas indican diferencias significativas (p<0.05) entre (1) áreas clausuradas o no al acceso de herbívoros domésticos dentro de cada localidad, y aquellas a la derecha de la coma indican diferencias significativas (p<0.05) entre las localidades estudiadas dentro de cada nivel de clausura; (2) diferentes profundidades del suelo desde la superficie del mismo (0-10, 10-20 o 20-30 cm) dentro de cada localidad estudiada, y aquellas a la derecha de la coma indican diferencias significativas (p<0.05) entre las localidades dentro de cada una de las profundidades del suelo estudiadas; (3) diferentes profundidades del suelo (0-10, 10-20 o 20-30 cm desde la superficie del mismo) dentro de cada nivel de clausura, y aquellas a la derecha de la coma indican diferencias significativas (p<0.05) entre áreas clausuradas o no al acceso de herbívoros domésticos dentro de cada profundidad del suelo estudiada



Figure 8. Total root C storage (Mg/ha) on fenced and unfenced sites at the various studied counties (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao and L6= Jianping) in the Province of Liagoning, China. Each histogram is the mean of n=5. Different letters to the left above the histograms indicate significant differences (p<0.05) between fenced and unfenced sites within each county, and those to the right of them indicate significant differences (p<0.05) among the studied counties within each fencing level

Figura 8. Almacenamiento de C en el sistema radical total (Mg/ha) en áreas clausuradas o no al acceso de herbívoros domésticos en seis localidades de la Provincia de Liaoning, China (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao y L6= Jianping). Cada histograma es el promedio de n=5. Letras diferentes a la izquierda encima de los histogramas indican diferencias significativas (p<0.05) entre sitios clausurados o no al acceso de herbívoros domésticos dentro de cada localidad, y aquellos a la derecha de la coma indican diferencias significativas (p<0.05) entre las localidades estudiadas dentro de cada nivel de clausura

(p>0.05) total root C storage on fenced sites. Total root C storage was similar (p>0.05) at all locations on unfenced sites.

Total plant C storage

Fenced sites showed a greater (p<0.05) total plant C storage (i.e., total aboveground + root C storage) than unfenced ones (**Figure 9**). While total plant C storage was similar (p>0.05) at Yixian, Kazuo and Jianchang, these values were greater (p<0.05) than those shown at Lingyuan, Beipiao and Jianping. However, these latter locations showed a similar (p<0.05) total plant C storage. On the other hand, unfenced sites showed a similar (p>0.05) total plant C storage at all locations.



Figure 9. Total plant C storage (Mg/ha) on fenced and unfenced sites at each of the studied counties (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao and L6= Jianping) in the Province of Liagoning, China. Different letters at the left above of the histograms indicate significant differences (p<0.05) between fencing levels within each of the studied counties, and those to the right of them indicate significant differences (p<0.05) among the studied counties within each fencing level

Figura 9. Almacenamiento de C en toda la planta (Mg/ha) en áreas clausuradas o no al acceso de herbívoros domésticos en cada una de las localidades estudiadas (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao y L6= Jianping) en la Provincia de Liaoning, China. Letras diferentes a la izquierda encima de los histogramas indican diferencias significativas (p<0.05) entre áreas clausuradas o no al acceso de herbívoros domésticos dentro de cada una de las localidades estudiadas estudiadas, y aquellas a la derecha de la coma indican diferencias significativas (p<0.05) entre las localidades estudiadas dentro de cada nivel de clausura

Soil C storage

The interactions between county and fencing level (p=0.033) and county and soil depth (p=0.006) were significant for soil C storage. However, the interaction between fencing level and soil depth was not significant (p=0.448). Soil C storage was greater on fenced than unfenced sites at all counties (**Figure 10**). Soil C

storage was greater (p<0.05) at Yixian than at Kazuo, Jianchang, and Jianping on fenced sites. Lingyuan and Beipiao showed similar (p>0.05) soil C storage on fenced sites. The lowest (p<0.05) soil C storage on fenced sites was found at Jianping. Yixian, Lingyuan, Kazuo and Beipiao showed similar (p>0.05) soil C storage at unfenced sites. Even more,



Figure 10. Soil carbon storage (Mg/ha) at (1) fenced and unfenced sites on each of the studied counties in the Liaoning Province, northeastern China (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao and L6= Jianping); (2) 0-10, 10-20 or 20-30 cm soil depth from the soil surface within each of the studied counties; (3) Each histogram is the mean of n=5. Different letters to the left above the histograms indicate significant differences (p<0.05) between (1) fenced and unfenced sites within each county, and those to the right of them indicate significant differences (p<0.05) among the studied counties within each fencing level; (2) different soil depths (0-10, 10-20 or 20-30 cm from the soil surface) within each studied county, and those to the right indicate significant differences (p<0.05) among the counties within each studied soil depth; (3) fenced and unfenced sites within each study soil depth (0-10, 10-20 or 20-30 cm from the soil surface)

Figura 10. Almacenamiento de C en el suelo (Mg/ha) en (1) áreas clausuradas o no al acceso de herbívoros domésticos en cada una de las seis localidades de la Provincia de Liaoning al noreste de China (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao y L6= Jianping); (2) 0-10, 10-20 o 20-30 cm de profundidad del suelo desde la superficie del mismo dentro de cada una de las localidades estudiadas; (3) entre áreas clausuradas o no al acceso de herbívoros domésticos dentro de cada profundidad del suelo estudiada (0-10, 10-20 o 20-30 cm desde la superficie del suelo). Cada histograma es el promedio de n=5. Letras diferentes a la izquierda encima de los histogramas indican diferencias significativas (p<0.05) entre (1) áreas clausuradas o no al acceso de herbívoros domésticos dentro de cada localidades del suelo estudiada, y aquellas a la derecha de la coma indican diferencias significativas (p<0.05) entre las localidades del suelo desde la superficie del mismo) dentro de cada localidade sel suelo (0-10, 10-20 o 20-30 cm desde la superficie del mismo) dentro de cada localidades del suelo (0-10, 10-20 o 20-30 cm desde la coma indican diferencias significativas (p<0.05) entre las localidades estudiadas dentro de cada nivel de clausura; (2) diferentes profundidades del suelo (0-10, 10-20 o 20-30 cm desde la superficie del mismo) dentro de cada localidad estudiada, y aquellas a la derecha de la coma indican diferencias significativas (p<0.05) entre las localidades dentro de cada profundidad del suelo estudiada; (3) entre áreas clausuradas o no al acceso de herbívoros domésticos dentro de cada localidades dentro de cada profundidad del suelo estudiada (0-10, 10-20 o 20-30 cm desde la superficie del mismo)

Yixian, Lingyuan, Jianchang and Beipiao did not differ (p>0.05) in soil C storage on unfenced sites. Finally, Yixian, Jianchang, Beipiao and Jianping also did not show significant differences (p>0.05) on unfenced sites in soil C storage.

Kazuo and Beipiao did not show significant differences (p>0.05) in soil C storage among soil depths. However, Yixian, Lingyuan, Jianchang and Jianping showed a greater (p<0.05) soil C storage at 0-10 than 20-30 cm soil depths. Nevertheless, soil C storage was similar (p>0.05) at 10-20 and 20-30 cm soil depths at Yixian, Lingyuan, Jianchang and Jianping. The greatest and lowest (p<0.05) soil C storageat 0-10 cm soil depth occurred at Yixian and Lingvuan, and Jianping, respectively. At the same time, Lingyuan, Kazuo, Jianchang and Beipiao showed a similar (p>0.05) soil C storage at 0-10 cm soil depth. Yixian, Lingyuan, Kazuo, Jianchang and Beipiao showed a similar (p>0.05) soil C storage at 10-20 cm soil depth. At this depth, however, soil C storage was greater (p<0.05) at Kazuo than at Jianping. At 20-30 cm soil depth, soil C storage was greater (p<0.05) at Beipiao than at Jianchang, and it was also greater (p<0.05) at Jianchang than at Jianping. However, soil C storage was similar (p>0.05) at Yixiao, Lingyuan, Kazuo and Jianchang at 20-30 cm soil depth (Figure 10). Finally, Yixian and Jianping did not differ (p>0.05) in the soil C storage at 20-30 cm soil depth.

There were no differences (p>0.05) in soil C storage among the three studied soil depths on fenced and unfenced sites. Also, soil C storage was similar (p>0.05)between fenced and unfenced sites at each of the studied soil depths.

The interaction between fencing level and county was not significant (p=0.170) for total soil C storage. However, there were significant differences for each of the main effects (fencing level, p=0.000; counties, p=0.010). Fenced sites had a greater (p<0.05) total soil C storage than unfenced ones (**Figure 11**). Yixian, Lingyuan, Kazuo and Jianchang showed a greater (p<0.05) total soil C storage than Jianchang and Jianping. At the same time, these two later counties showed a similar (p>0.05) total soil Cstorage.

Total grassland C storage

The interaction between fencing level and county was not significant (p>0.05). Nevertheless, there were significant differences between fencing levels and among counties (p<0.05). Total grassland C storage was similar (p>0.05) at Yixian, Lingyuan, Kazuo, Jianchang and Beipiao (**Figure 12**). Jianping showed a lower (p<0.05) grassland C storage than the remaining counties. However, this county had a similar grassland C storage as Jianchang.

Plant/soil ratio of carbon storage

The proportion of total plant C storage (aboveground + litter + roots) ranged from 8.63% to 18.61% on fenced areas, and there was a mean of 4.76% on those unfenced in the six counties. Nearly 95% of the grassland C in all counties was stored in the unfenced soil (**Figure 13**).

The relationship between total plant biomass and total plant C storage

There was a close and positive relationship (p<0.05) between total plant biomass and total plant C storage at all of the 6 studied counties on fenced as well as on unfenced areas (**Figure 14**). The R2



Figure 11. Total soil C storage (Mg/ha) (1) among counties in the Liaoning Province, northeastern China (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao and L6= Jianping), and (2) between fenced (F) and unfenced sites (UF) on average for all study counties *Figura 11. Almacenamento de C total en el suelo (Mg/ha) (1) entre localidades en la Provincia de Liaoning, al noreste de China (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao y L6= Jianping), y (2) entre áreas clausuradas (F) o no (UF) en promedio para todas las localidades estudiadas*



Figure 12. Total grassland C storage (Mg/ha) among counties in the Liaoning Province, northeastern China (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao and L6= Jianping) or fencing levels on average for all counties. Each histogram is the mean of n=5. Different letters above the histograms indicate significant differences (p<0.05) (1) among counties on average for fenced and unfenced sites, and (2) between fenced (F) and unfenced (UF) sites on average for all counties

Figura 12. Almacenamiento de C total en el pastizal natural (Mg/ha) entre localidades en la Provincia de Liaoning, al noreste de China (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao y L6= Jianping) o niveles de clausura en promedio para todas las localidades. Cada histograma es el promedio de n=5. Letras diferentes encima de los histogramas indican diferencias significativas (p<0.05) (1) entre localidades en promedio para áreas clausuradas o no al acceso de herbívoros domésticos, y (2) entre sitios clausurados (F) ó no (UF) en promedio para todas las localidades



Figure 13. Percentage ratio of carbon storage in the plant versus the soil within each study county in the Liaoning Province, China (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao and L6= Jianping), on fenced or unfenced sites at each of the studied counties. Each histogram is the mean of n=5

Figura 13. Relación porcentual de almacenaje de C en la planta versus el suelo dentro de cada localidad estudiada en la Provincia de Liaoning, China (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao y L6= Jianping), en áreas clausuradas o no al acceso de herbívoros domésticos en cada una de las localidades estudiadas. Cada histograma es el promedio de n=5

between those two variables ranged from 0.5771 (Yixian, unfenced areas) to 0.9968 (Kazuo, fenced areas) among all of the studied relationships.

DISCUSSION

Various studies have determined the effects of soil bulk density, soil pH, and total soil N and C concentrations on plant biomass and C storage, and on soil C storage in various grassland ecosystems worldwide (e.g., Li et al., 2019). Houlbrooke et al. (1997) reported the detrimental effects of increasing soil bulk density on shoot growth of various ryegrass lines. This has been attributed to the detrimental effects of increasing soil

bulk density on root and shoot growth, root surface area /g shoot, and penetration depth of roots into the soil (Hallmark & Barber, 1981; Place et al., 2008). Binkly and Vitousek (1989) discussed the importance of soil pH in determining the soil nutrient availability for plant roots, and the occurrence and activity of soil microorganisms. They reported that the activity of several soil microorganisms is favoured by more neutral pH conditions in the soil (i.e., closer to 7 on the pH scale), and that the uptake rate for cations seems to be highest in the more neutral pH range. Yang et al. (2009) showed that as total soil N increased, both aboveground and root biomasses



Figure 14. Simple linear regressions between total plant biomass (TPB; g/m²) and total plant carbon storage (TPCS; Mg/ha) on fenced (black circles) or unfenced sites (black squares) on each of the studied counties in the Liaoning Province, China (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao and L6= Jianping). All symbols are individual data *Figura 14.* Regresiones lineales simples entre la biomasa total de las plantas (TPB; g/m²) y el almacenaje de C en toda la planta (TPCS; Mg/ha) en áreas clausuradas (círculos negros) o no (cuadrados negros) en cada una de las localidades estudiadas en la Provincia de Liaoning, China (L1= Yixian, L2= Lingyuan, L3= Kazuo, L4= Jianchang, L5= Beipiao y L6= Jianping). Todos los símbolos son datos individuales

also increased in Tibetan grasslands. Li et al. (2019) showed that total soil N and organic C contents increased on longterm fenced areas on temperate grasslands in northwest China. In turn, plant functional composition has influenced the rates of soil carbon and nitrogen accumulation through time in herbaceous grassland communities (Fornara & Tilman, 2008). Yang et al. (2009) and Li et al. (2019) reported that greater total soil N and organic C contents led to greater plant biomasses and C storage, and also to greater soil organic C storage in various old fields in numerous herbaceous grassland communities. In agreement to results of Wang et al. (2014) on grasslands of the Loess Plateau, China, fenced areas had mostly lower soil bulk densities, pH values closer to 7, and more N and C soil concentrations than unfenced areas. This may partially contribute to explain the higher values for all studied variables on fenced than unfenced areas in all counties.

We determined a greater root biomass and C storage in the top soil layer (i.e., 0-10 cm from the soil surface) than deeper in the soil profile in 4 out of the 6 counties. This agrees with findings of Ni et al. (2015) who reported that more root biomass was observed in the top soil layer in various vegetation functional types in karst terrain (i.e., a highly special geomorphology mainly consisting of limestone and dolomite) of southwestern China.

In general, the highest total aboveground biomass and C storage were obtained in Jianping, and the lowest values for this variable were found in Lingyuan and Beipiao in the fenced areas. In these areas, the highest total root biomasses and C storage, however, were

reached in Yixian, Kazuo and Jianchang, and the lowest values for these parameters were shown in Lingyuan, Beipiao and Jianping. The greatest total root biomasses and C storages were higher than values found for the total aboveground biomasses and C storages in the fenced areas. Greater biomasses belowground than aboveground have been reported for various authors in different world's grasslands (WenHong et al., 2008; Li et al., 2019). As a result, the greatest total plant biomasses and C storages were also reached in Yixian, Kazuo and Jianchang, and the lowest values for this parameter were shown in Lingyuan, Beipiao and Jianping on the fenced areas. The low total plant biomasses in Beipiao and Jianping might be partially attributed to their lower annual precipitations (Beipiao= 475 mm, Jianping= 531 mm) in comparison to the other counties (> 560 mm). WenHong et al. (2008) reported that total aboveground and belowground biomasses significantly decreased as annual precipitation also decreased in desert, typical and meadow steppes of Inner Mongolia. In the unfenced areas, however, there were mostly no significant differences among locations for the standing crop and total aboveground biomasses and C storages, and also for the root, total root and total plant biomasses and C storages. Finally, root biomasses and C storages were greater at 0-10 than at 10-20 or 20-30 cm soil depth at four out of the six studied locations (Yixian, Kazuo, Jianchiang, and Beipiao for root biomasses, and Yixian, Lingyuan, Kazuo and Jianchang for root carbon storages). WenHong et al. (2008) also found that belowground biomass decreased as soil depth increased in various, different steppes of Inner Mongolia.

In all studied locations, standing crop and total aboveground biomasses and C storages, root, total root and total plant biomasses and C storages were much greater on fenced than unfenced areas. In other studies, fencing has also increased the grassland ecosystem biomass and C storage (Akiyama & Kawamura, 2007). In the past years, avoiding livestock grazing from the grasslands by establishing fences has become a widespread management method for conserving and restoring grassland plants (Golodest et al., 2010; Chen & Tang, 2016). Generally, fencing increases plant C storage by increasing plant biomass (Wang et al., 2014). These findings agree with our study, where fencing increased both plant aboveground and root biomasses. Total plant biomass directly influences the amount of C contained in the vegetation, which will be subsequently incorporated into the soil as litter (Ni, 2001). There is a buildup of litter in the fenced treatments due to the presence of more grass species, as livestock grazing has been excluded (Li et al., 2019). Moreover, long-term fencing can have some changes in soil loosening, soil infiltration rates, soil bulk density, soil porosity and increases in oxygen concentrations, which make a change in the soil organic C levels as well (Li et al., 2019). The decreases of soil bulk density in the fenced areas may be attributed to the fact that trampling may have increased the bulk density in the unfenced but not in the fenced areas. Also, an increase in plant roots and soil microorganisms may have decreased the bulk densities in the fenced areas (Deng et al., 2014; Aldezabal et al., 2015). Other studies, however, reported that soil C storage significantly increased with increased grazing pressure; these increases had a significant positive correlation with the increase of root biomass allocation (Li et al., 2011).

Although aboveground living plant biomass plays an important role in fixing C, it is clear from these results that fencing also had a great impact in dramatically increasing root biomass (Zeng et al., 2017; Li et al., 2019). Fencing increased twice as much total soil C storage up to 30 cm soil depth in comparison to unfenced sites. Unfencing has resulted in C losses from this grassland ecosystem in the past (Guo et al., 2008). There may be a range of potential mechanisms through which soil C increased in the fenced meadows (Holt, 1997). Firstly, the return of C from increased aboveground biomass and litter. Secondly, root biomass is an important component of soil C sequestration. Some researchers have supported that lightly and moderately degraded grasslands can be restored to their initial state within 10-20 years by reducing the grazing intensity and establishing fencing (Li et al., 2013). However, for heavily and extremely degraded grasslands, we should not depend on fencing alone, as other measures will be necessary (Fang et al., 2010). Similar to the results of Li et al. (2019), our results showed that the fenced grasslands had a much higher C storage than the unfenced ones. All these indicate that fencing is an effective restoration method, conducive to increasing C storage in grasslands (Mcsherry & Ritchie, 2013; Chen & Tang, 2016). Silva et al. (2019), however, found that moderate grazing did not reduce aboveground C stocks significantly in various shrub-grassland habitat types in the European Union. Even more, these authors reported that moderate grazing is a management practice that effectively contributes to the conservation of a network of sites in the European Union. These sites were established to halt biodiversity loss and ensure the long-term conservation of threatened species and habitat types. Even more, the effects of grazing intensity on belowground C cycling should be highlighted since they may need to be incorporated into regional and global models for predicting effects of human disturbance on global grasslands, and assessing the climate biosphere feedbacks.

Some researchers have confirmed that total plant C storage is often determined by total plant biomass (Kemper, 2015). Total carbon storage is also important at an organ plant level. For example, when a photosynthetic surface area is not available, plant regrowth is dependent from the C stored in plant crowns, stem bases and roots (Briske & Richards, 1995).

Total plant biomass correlated linearly and positively with total plant C storage at the studied grassland ecosystems on either fenced or unfenced areas in all counties. This agrees with the results reported by Liao & Boutton (2008). The slopes of the linear regression equations were greater on fenced than unfenced sites in all counties, but Yixian (Bardgett et al., 1998). These findings indicate that any given change in total plant biomass will cause a greater change in total plant C storage at the fenced than the unfenced areas in all counties, except Yixian. Thus, fencing might contribute to grassland restoration (Golodets et al., 2010).

CONCLUSIONS

Our results confirmed that fencing increased total plant biomass (aboveground + litter + roots), C storage, and total soil C storage in the studied grasslands at the various counties in the Liaoning Province. Fencing proved to be an important management tool for improving degraded grasslands in the desertification governance base of the Liaoning province. After fencing was established there was a higher grassland biomass and C storage because of the increase in plant aboveground, root, and litter biomasses.

ACKNOWLEDGEMENTS

Chan Zhou acknowledges financial support from the National Science Foundation of China (Grant 32071860), Natural Science Foundation of Liaoning Science and Technology Department (Grant 2019-MS-155), Serving Local Project of Liaoning Provincial Education Department (LFW202001) and cooperation project of national forestry science and technology demonstration forest farm in Fuxin Mongolian Autonomous County. C.A. Busso thanks (1) the sabbatical leave given by Universidad Nacional del Sur and the Consejo Nacional de Investigaciones Científicas y Técnicas de la República Argentina (CON-ICET), (2) the Associateship awarded by the Third World Academy of Sciences (TWAS)-UNESCO, and (3) housing, facilities and financial support from the Institute of Applied Ecology, Chinese Academy of Sciences, Shenyang, China.

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Recibido: 01/2021 Aceptado: 03/2021