

Article

Mowing Increases Root-to-Shoot Ratio but Decreases Soil Organic Carbon Storage and Microbial Biomass C in a Semiarid Grassland of North China

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Abstract: Quantifying the long-term effects of mowing on soil organic carbon (SOC) is of great importance for understanding the changes in the carbon cycle of the grassland ecosystem and for managing the grassland strategies for both production and soil nutrients. We investigated SOC content and storage within the 0–30 cm soil layer in the grasslands following the application of different mowing regimes—i.e., mowing once every 2 years ($M_{1/2}$), mowing twice every 3 years ($M_{2/3}$), mowing once a year ($M_{1/1}$), mowing twice a year ($M_{2/1}$), and no mowing (CK)—in the semiarid steppe of northern China. The results indicated that SOC storage and microbial biomass C (MBC) decreased significantly with soil depth. Different mowing frequencies all declined SOC storage and MBC of the grassland ecosystem; however, the root-to-shoot ratio (R:S) was increased. The SOC storage was greatest under CK and had the following order: $CK > M_{1/2} > M_{2/3} > M_{1/1} > M_{2/1}$ at 0–20 cm, while no significant difference existed in the five mowing frequencies at the soil 20–30 cm layers. Our findings elucidate that different mowing regimes influence soil carbon storage by altering the productivity of vegetation, litter, plant community composition, soil microbial biomass, and resource allocation between aboveground plants and belowground roots, which need to be considered in the sustainable utilization of grasslands in the future. The results of this study support the view that mowing once every 2 years may be an effective mowing management regime for semiarid grasslands, as it conserves both above and belowground parts and maintains the healthy development of ecosystem functions in semiarid grasslands.



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Keywords: different mowing regimes; soil organic carbon storage; microbial biomass C; root-to-shoot ratio

1. Introduction

Due to increasing concerns about anthropogenic climate change, actions to reduce greenhouse gas emissions and to stabilize their concentration in the atmosphere are being taken [1]. The sequestration of soil organic carbon (SOC) by improved soil management is a promising contribution to the mitigation of climate change [2]. In this respect, grassland ecosystems account for 40% of terrestrial land cover and store about 34% of the terrestrial carbon stock [3], with about 90% of their carbon stored belowground as root biomass and SOC, thus playing an important role in soil carbon sequestration [3,4]. However, grassland ecosystems have been increasingly impacted by human disturbance (e.g., overgrazing, unreasonable mowing, and land-use conversion to agriculture) and climate change. Worldwide, grasslands have undergone severe decreases in biodiversity and grassland ecosystem functions, leading to a reduction in SOC storage with a significant impact on soil C, N, and P cycling [5,6]. All of these factors are actually triggered by the complex interactions between vegetation and biotic/abiotic factors [7]. Empirical and experimental studies have

shown that improving grassland management can increase SOC storage, thus mitigating carbon losses due to climate change, long-term overgrazing, unreasonable mowing, and grassland degradation [4]. Therefore, it is critical to obtain a better understanding of how these interactions influence the direction and magnitude of changes in soil C storage, which is important for predicting the effects of human initiatives on soil fertility and revealing the underlying mechanisms of biogeochemical cycling [8,9]. In particular, we need to develop appropriate management measures to achieve both ecological and economic sustainability and profitability.

The change in SOC content is an indicator of the evolution of soil health and quality in the context of land-use change, while the decrease in SOC storage is an important manifestation of the degradation of ecosystem function [10]. The impacts of SOC, total nitrogen (TN), total phosphorus (TP), and their stoichiometry on modulating ecosystem function are further modulated by ecosystem management strategies [11,12]. The impact of these management strategies on processes impacting soil biogeochemical cycling via plant–soil interactions is poorly understood. Traditionally, mowing is one of the most important methods of land-use management in arid and semiarid natural grasslands, regulating the imbalance among forage seasons, regions, and years of good harvest. It is normal practice to mow grassland year to year without any fertilizer application by local herders in the semiarid region of northern China [13]. Experimentally, mowing affects plant growth by removing and destroying photosynthetic tissues directly and by affecting biologically regulated processes in the soil through its influence on soil organisms indirectly [14]. These direct and indirect pathways can have different effects on SOC levels, which can influence plant productivity and ecosystem function in turn. Recently, several studies have focused on how different management strategies influence the soil C cycle. For instance, Yang [9] showed that ecological restoration measures in the semiarid steppe of Inner Mongolia affected SOC content and C/nutrient ratios. Hou et al. [15] highlighted that moderate mowing can facilitate soil C and N concentration and further change the relative allocation of nutrients. However, the long-term effects of mowing regimes (frequency) on the distribution of SOC storage and microbial biomass C (MBC) have hardly been evaluated for the grasslands in the typical steppe region of Inner Mongolia. How different mowing management practices affect SOC storage through soil–plant interactions remains unknown. Few studies have focused on the contribution of these differences to driving changes in SOC levels in soil.

Soil microorganisms are a critical link between aboveground and underground parts of terrestrial ecosystems, which participate in many ecological processes [16]. They can transform complex organic substrates into simpler ones and significantly alter the ratio of C, N, and P in the soil [17], which is an important regulator driving the release or sequestration of soil C, N, and P nutrients [18,19]. Soil microbial biomass is the active part of soil organic matter and the key driver of ecosystem C dynamics, which is closely related to mineralization processes of soil nutrients and regulates vegetation productivity [20]. Microbial biomass C, N, and P stoichiometry determines the direction of nutrient sequestration or mineralization, affecting soil nutrient availability [21]. Shifts in plant community composition or nutrient allocation between above and belowground are the main mechanisms driving the response of soil nutrient stoichiometry to grassland management practices [22,23]. Therefore, there is a lack of research results on whether changes in MBC and its stoichiometry under different mowing management affect plant community composition and the interaction of plant communities with soil elements, which in turn affects soil C storage.

A long-term mowing intensity experiment was conducted in a semiarid grassland of central Inner Mongolia to examine the effects of different mowing measures on grassland succession processes. In this present study, we summarized the direct (mainly via alteration of soil properties) and indirect (mainly via mediation of plant community composition, above- and belowground biomass) effects of four mowing regimes on SOC when compared with no mowing. We aimed to address the following three questions: (1) How do different

mowing regimes affect the root-to-shoot ratio (R:S), SOC storage, and MBC in different soil layers? (2) Which mowing management regime is favorable to maintain soil SOC storage? (3) Specifically, how do plant production and microbial biomass C regulate the SOC storage in response to different mowing regimes?

2. Materials and Methods

2.1. Study Site

The study was conducted at the Baiyinxile Ranch, located in the Xilingol region of Inner Mongolia, China (43°26′–44°08′ N, 116°04′–117°05′ E) (Figure A1a). The region experiences a temperate semiarid climate where the growing season usually lasts from early May to late September. The annual mean precipitation is 315 mm, 80% of which falls between May and September, and the annual mean temperature is -0.5 °C, with the lowest monthly mean temperature in January (-19.6 °C) and the highest in July (21.8 °C). The perennial plants grow from April/May to September/October, about 150 days. The natural vegetation is dominated by *Stipa grandis* P.A.Smirn, *Leymus chinensis* (Trin.) Tzvel, and *Cleistogenes squarrosa* (Trin. ex Ledeb.) Keng, the three species accounting for 58–82% of the total aboveground biomass of the grassland community. The soil is a sandy loam chestnut soil (or Calci-Orthic Aridisol in the US soil taxonomy classification system) with a mean pH of 8.07 in the plant root layer (0–20cm).

2.2. Experimental Design and Different Mowing Regimes

The experimental site was established in 2001, containing grassland subjected to a long-term continuous mowing exclusion (unmown) and four intensities of mowing in a randomized block design (Figure A1b). The experiment was located on a flatland area on the side of a 1.96 ha permanent enclosure, where livestock grazing was prevented. The area was divided into 20 plots with 5 m buffers. Four replicates of each of five mowing treatments (i.e., mowing frequencies) were randomly assigned to these 20 permanent plots that were 20 m × 30 m in size. The four treatments were as follows: (1) CK, no mowing; (2) $M_{1/2}$, mowing once every 2 years; (3) $M_{2/3}$, mowing twice every 3 years; (4) $M_{1/1}$, mowing once a year; (5) $M_{2/1}$, mowing twice a year. At the time of harvest, a small hand-pushed lawnmower was used to cut the grass to a 6 cm stubble height over the entire plot (6 cm has consistently been considered to be a reasonable height for sustainable use of the studied grassland [13]), and the cut plant material was removed. The mowing was conducted each year on 15 August (or 1–3 days later depending on the weather) for the $M_{1/2}$, $M_{2/3}$, and $M_{1/1}$ treatments and on 15 June and 15 September for the $M_{2/1}$ treatment.

2.3. Plant Sampling and Analysis

We sampled plant and soil communities in the plots under the five mowing treatments in mid-August 2019. First, within each of the 20 plots (5 treatments × 4 replicates), we surveyed plant communities using three 1 m × 1 m quadrats in a diagonal line of each plot to measure the plant aboveground biomass by clipping standing plants by species and weighing them after 48 h of oven-drying at 65 °C. Total plant aboveground biomass (AB) was calculated as the sum of all species biomass. We also estimated plant belowground biomass (BB) by collecting three soil cores from each of three quadrats in each plot, which were used to generate a composite sample (0–100 cm depth, 7 cm diameter). Roots within the core were rinsed using sieves (mesh size 0.25 mm) on the same day, and then dried at 65 °C for 48 h until constant weight using an analytical balance. The root-to-shoot ratio (R:S) was calculated as the ratio of BB to AB.

2.4. Soil Sampling and Analysis

After removal of plant material in the three quadrats in each plot, the soil samples were collected from three layers (0–10 cm, 10–20 cm, and 20–30 cm) using an auger (5 cm in diameter) from the quadrats, because of significant plant–soil interaction effects occurring in the topsoil layer [24]. Three soil cores were randomly selected from each plot and combined into one composite sample for each layer. Each fresh composite soil sample was first sieved through a 2 mm mesh size to remove stones and roots, and then divided into two subsamples. One subsample was immediately frozen on dry ice for soil microbial biomass (stored at $-80\text{ }^{\circ}\text{C}$), while the other was air-dried for the physicochemical analysis.

The SOC content was analyzed using the potassium dichromate oxidation spectrophotometric method [25].

2.5. Soil Microbial Biomass

Microbial biomass C and N concentrations (MBC and MBN) were determined using the chloroform fumigation/extraction method [26,27]. Simply put, for each soil subsample, preincubated fresh soil (10 g weight) was used for chloroform fumigation for 24 h at $25\text{ }^{\circ}\text{C}$ and the same weight of non-fumigated soil was used for the control. Then, 50 mL of 0.5 M K_2SO_4 was used for the extraction of MBC and MBN. Concentrations of extracted C and N were determined using a TOC/analyzer.

2.6. Data Calculation and Statistical Analysis

The storage of SOC ($\text{SOC}_{\text{storage}}$, $\text{kg}\cdot\text{m}^{-2}$) in selected layers was calculated as follows [28]:

$$\text{SOC}_{\text{storage}} = \text{BD} \times \text{SOC} \times \text{D} \quad (1)$$

where SOC represents the SOC contents ($\text{g}\cdot\text{kg}^{-1}$), BD is the soil bulk density ($\text{g}\cdot\text{cm}^{-3}$), and D is the thickness of the sampled soil layer (m).

Treatment effects on BB, R:S, SOC content, SOC storage, MBC, and MBC/MBN were analyzed using two-way ANOVA with different mowing regimes and soil depths as the main factors, along with their interactions. One-way ANOVA was employed to evaluate the changes in plant biomass (AB, BB, and R:S), content, and storage of SOC across different mowing treatments at various soil depths, followed by the Tukey's range test at $p < 0.05$. Pearson correlation coefficients were determined to depict the correlations among plant biomass (AB, BB, R:S), MBC, MBC/MBN, and SOC storage for each soil depth. Statistical analyses were conducted using version 26.0 of SPSS software (IBM/SPSS, Chicago, IL, USA). A random forest analysis method was used to obtain important indicators using the randomForest R package [29] with 999 trees to explore the relative contributions of different mowing regimes, plant biomass (AB, BB, and R:S), litter MBC, and MBC/MBN to the vertical distribution of SOC storage.

3. Results

3.1. Changes in Plant Production under Different Mowing Regimes

The plant production gradually decreased with increasing mowing intensity. Mowing treatment ($M_{2/1}$, $M_{1/1}$, and $M_{2/3}$) significantly decreased AB ($p < 0.05$) when compared with the no mowing control (CK), whereas $M_{1/2}$ did not induce significant changes in AB when compared to CK ($p > 0.05$; Figure 1a). The relative biomass of *Leymus chinensis* was lower in the $M_{1/2}$ and $M_{2/3}$ treatments than in the CK ($p < 0.05$), and then gradually increased with increasing mowing intensity. In contrast, the light mowing disturbance according to both $M_{1/2}$ and $M_{2/3}$ regimes significantly increased the relative biomass of *Stipa grandis* ($p < 0.05$; Figure 1b). The relative biomass of *Cleistogenes squarrosa* was higher in $M_{2/1}$ than in the other mowing regimes. Mowing significantly reduced the litter biomass when compared with the control group (CK), while increasing the mowing intensity gradually decreased litter biomass ($p < 0.05$; Figure 1c). In addition, different mowing regimes and soil layers had significant effects on BB and R:S ($p < 0.05$; Table 1).

BB in the $M_{2/1}$ plot was higher than that in other measures across the 0–20 cm soil depth range, whereas it gradually decreased with increasing mowing intensity (Figure 1d). Moreover, R:S at the 0–20 cm depth was higher in the mowing treatments than in CK; it was always greatest in the $M_{2/1}$ plot (Figure 1e). Different mowing regimes had no significant effects on BB and R:S in the 20–30 cm layer ($p > 0.05$; Figure 1d,e).

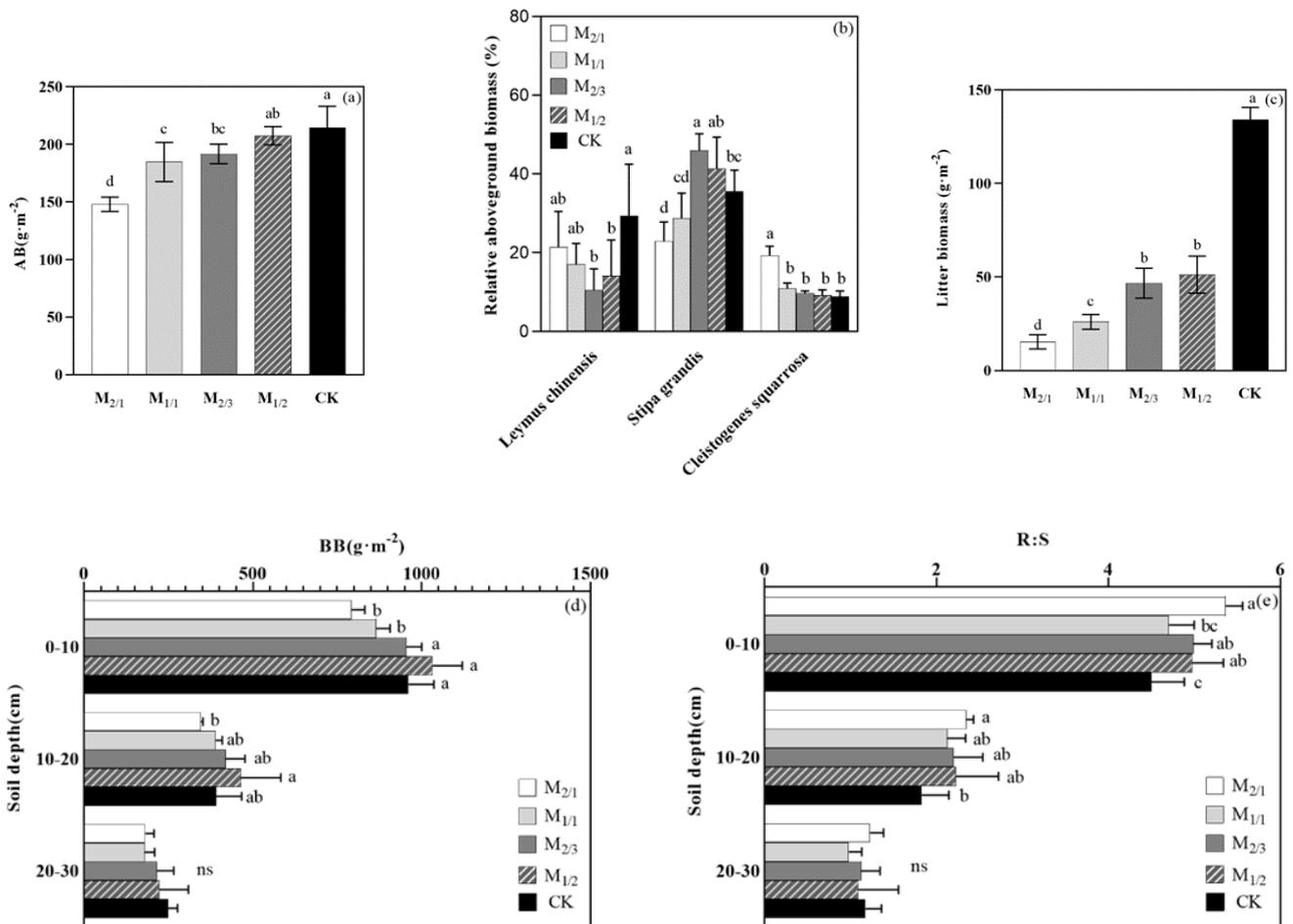


Figure 1. Effects of different mowing regimes on AB (a), relative aboveground biomass of dominant plant species (b), litter biomass (c), BB (d) and R:S (e) in a semiarid grassland of Inner Mongolia. Values are means (\pm SE) of four replicate plots. The different letters denote significant differences between treatments ($p < 0.05$). The same letters denote no significant differences between treatments ($p > 0.05$). AB: plant aboveground biomass; BB: belowground biomass; R:S: root-to-shoot ratio; CK: no mowing; $M_{1/2}$: mowing once every 2 years; $M_{2/3}$: mowing twice every 3 years; $M_{1/1}$: mowing once a year; $M_{2/1}$: mowing twice a year.

Table 1. Results of two-way ANOVA for the effects of different mowing treatments or soil depth and their interaction on belowground biomass (BB), root-to-shoot ratio (R:S), content and storage of SOC ($\text{SOC}_{\text{storage}}$), microbial biomass C (MBC), and soil MBC to soil MBN ratio (MBC/MBN).

	Soil Depths		Treatments		Soil Depths \times Treatments	
	F	<i>p</i>	F	<i>p</i>	F	<i>p</i>
BB	754.622	<0.001	9.044	<0.001	1.835	0.095
R:S	877.333	<0.001	4.598	0.003	1.212	0.314
SOC	125.953	<0.001	2.712	0.042	0.703	0.687
$\text{SOC}_{\text{storage}}$	198.937	<0.001	4.799	0.003	1.209	0.315
MBC	453.009	<0.001	56.742	<0.001	3.16	0.006
MBC/MBN	29.484	<0.001	6.1	0.001	2.337	0.034

3.2. Changes in Soil SOC Content and Storage with Depth and Different Mowing Regimes

Significant differences in SOC content and storage were found between samples taken at various soil depths and in various treatments (Table 1). SOC content and storage decreased significantly along with increasing depth (Table 1 and Figure 2a,b). The SOC content in the high-frequency mowing treatments $M_{1/1}$ and $M_{2/1}$ was significantly lower than that in CK in the 0–10 cm layer ($p < 0.05$), while no significant difference was detected in the 0–10 cm layer among $M_{1/2}$, $M_{2/3}$, and CK treatments ($p > 0.05$; Figure 2a).

SOC storage exhibited a consistent order, i.e., $CK > M_{1/2} > M_{2/3} > M_{1/1} > M_{2/1}$, in the soil 0–20 cm layer, while no significant difference existed across the five mowing frequencies in the soil 20–30 cm layer (Figure 2b). SOC storage in $M_{1/1}$ ($2.46 \text{ kg}\cdot\text{m}^{-2}$) and $M_{2/1}$ ($2.20 \text{ kg}\cdot\text{m}^{-2}$) plots was significantly lower than that in CK ($2.79 \text{ kg}\cdot\text{m}^{-2}$), which was consistent with the SOC content ($p < 0.05$; Figure 2b). However, the difference is that the storage in $M_{2/3}$ ($2.61 \text{ kg}\cdot\text{m}^{-2}$) was significantly lower than that in CK ($p < 0.05$). Compared with $M_{1/2}$, $M_{2/1}$ significantly decreased the SOC content in the 10–20 cm layer, while it decreased the SOC storage at 10–20 cm depth when compared with CK ($p < 0.05$; Figure 2b).

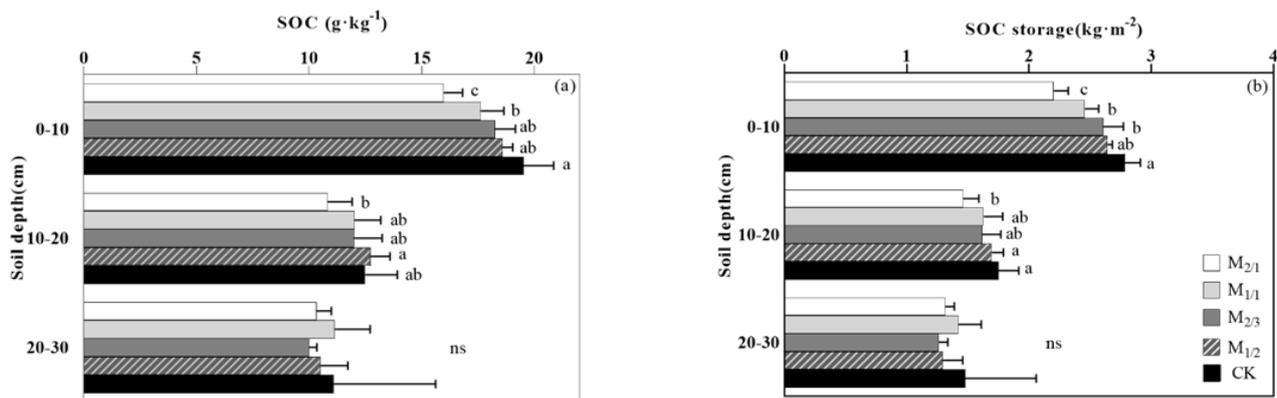


Figure 2. Vertical distribution of SOC content (a) and SOC storage (b) in the 0–30 cm soil layer under different mowing regimes in a semiarid grassland of North China. The different letters denote significant differences between treatments ($p < 0.05$). The same letters denote no significant differences between treatments ($p > 0.05$). CK: no mowing; $M_{1/2}$: mowing once every 2 years; $M_{2/3}$: mowing twice every 3 years; $M_{1/1}$: mowing once a year; $M_{2/1}$: mowing twice a year.

3.3. Changes in MBC and MBC/MBN with Depth and Different Mowing Regimes

Along with increasing depth, microbial biomass C (MBC) declined and showed significant differences at various soil depths among treatments ($p < 0.05$; Table 1 and Figure 3a). With increasing mowing frequencies, the MBC decreased sequentially at 0–10 cm and 20–30 cm depths. $M_{2/3}$, $M_{1/1}$, and $M_{2/1}$ treatments significantly reduced MBC at various depths when compared with CK ($p < 0.05$; Figure 3a). MBC in the $M_{1/2}$ plot was signifi-

cantly lower than CK at 10–20 cm ($p < 0.05$), while no significant difference was detected at the 0–10 cm and 20–30 cm layers (Figure 3a).

Significant differences in the soil microbial biomass MBC/MBN ratio were observed at various depths among treatments (Table 1 and Figure 3b). The MBC/MBN ratio at layers other than 10–20 cm of soil depth declined with increasing mowing frequencies. The MBC/MBN ratio in $M_{2/1}$ was significantly lower than that in CK in the 0–10 cm and 20–30 layers ($p < 0.05$), while no significant difference was detected among CK, $M_{1/2}$, and $M_{2/3}$ at the same depths ($p > 0.05$; Figure 3b).

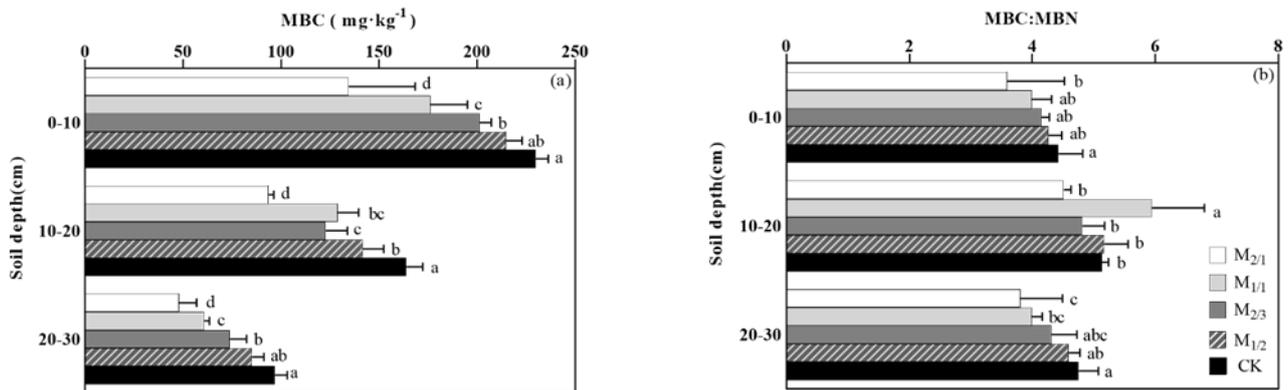


Figure 3. Vertical distribution of MBC (a) and MBC/MBN storage (b) in the 0–30 cm soil layer under different mowing regimes in a semiarid grassland of North China. The different letters denote significant differences between treatments ($p < 0.05$). The same letters denote no significant differences between treatments ($p > 0.05$). CK: no mowing; $M_{1/2}$: mowing once every two years; $M_{2/3}$: mowing twice every three years; $M_{1/1}$: mowing once a year; $M_{2/1}$: mowing twice a year.

3.4. Driving Factors of Soil C Storage Change

The results of Pearson correlation analysis indicated that the SOC storage was negatively correlated with *Cleistogenes squarrosa* biomass and R:S but positively correlated with AB, BB, *Stipa grandis* biomass, litter, MBC, and MBC/MBN ratio in the 0–10 cm layer ($p < 0.05$; Figure 4), while SOC storage positively correlated with AB, BB, litter, and MBC in the 10–20 cm layer ($p < 0.05$; Figure A2a), but not in the 20–30 cm layer (Figure A2b). The relative contributions of litter, *Cleistogenes squarrosa* biomass, MBC, and AB to SOC storage rate were greater than those of the other factors in the topsoil layer (0–10 cm; Figure 5a). For the 10–20 cm layer, the relative contributions of MBC/MBN, MBC, BB, and *Leymus chinensis* biomass to the SOC storage rate were greater than those of the other factors (Figure 5b). The relative contributions of three dominant plant species to SOC storage rate were greater than those of the other factors across the 20–30 cm topsoil layer (Figure 5c).

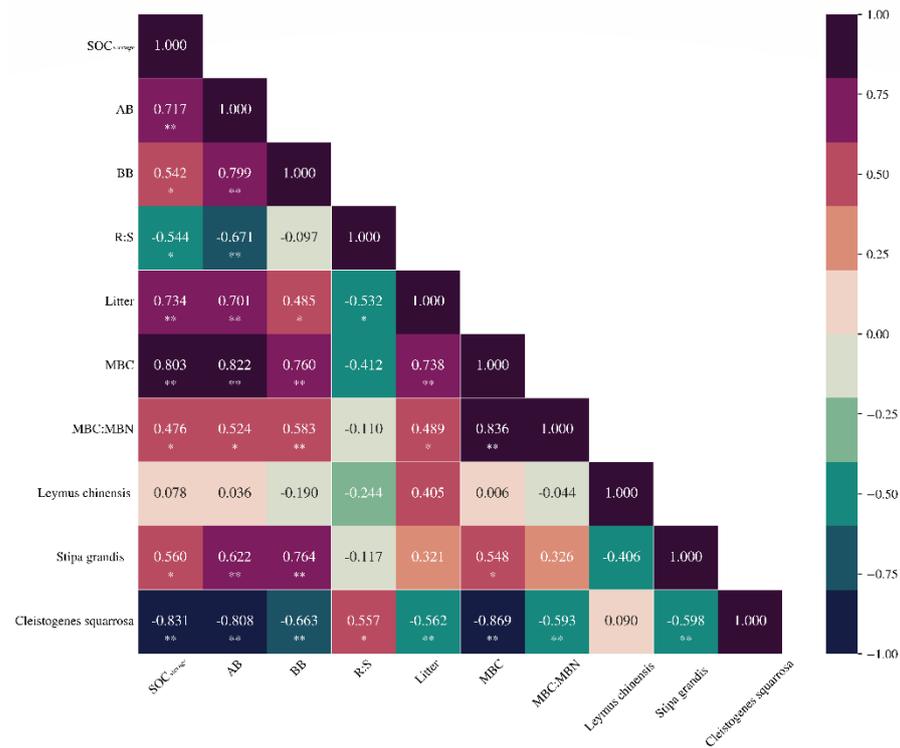


Figure 4. Pearson correlation coefficients between the plants and soil properties in the 0–10 cm layer. Positive correlations are labeled as “+”, and negative correlations are labeled as “–”. Significance levels are denoted as follows: ** $p < 0.01$, * $p < 0.05$. AB: plant aboveground biomass; BB: plant belowground biomass; R:S: root-to-shoot ratio; SOC_{storage}: soil organic carbon storage; MBC: microbial biomass C; MBC/MBN: soil MBC to soil MBN ratio; relative aboveground biomass of *Stipa grandis*, *leymus chinensis*, and *Cleistogenes squarrosa*.

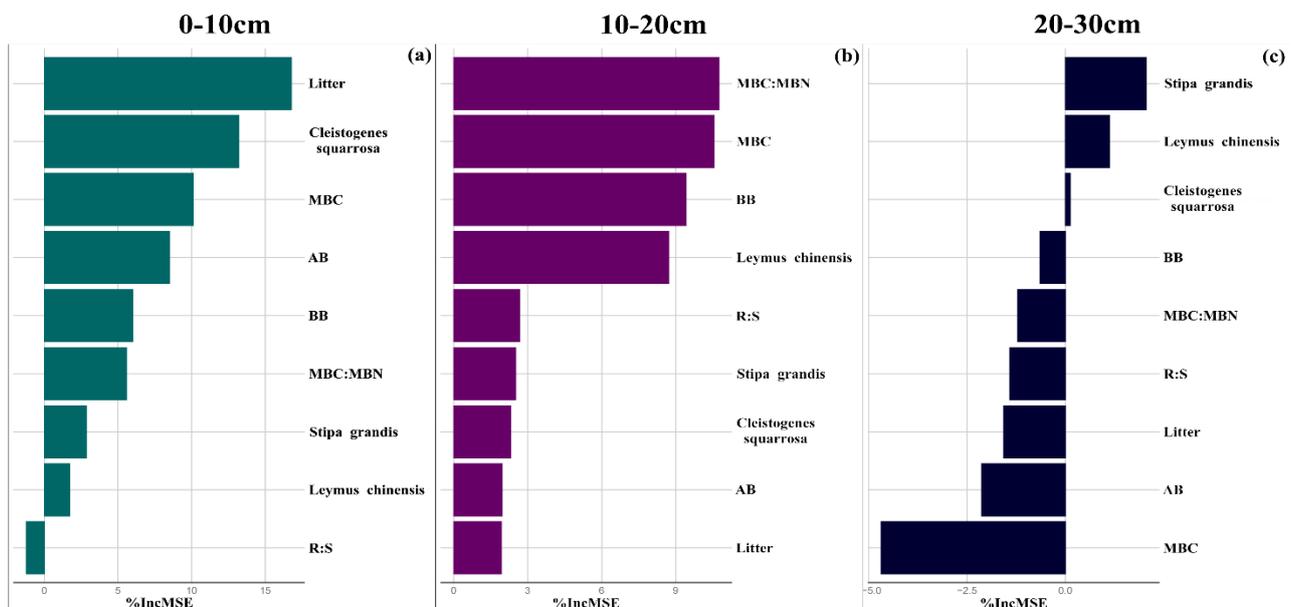


Figure 5. Relative contribution of changes in plant aboveground biomass (AB), belowground biomass (BB), litter, root-to-shoot ratio (R:S), microbial biomass C (MBC), soil MBC to soil MBN ratio (MBC/MBN), and relative aboveground biomass of *Stipa grandis*, *leymus chinensis*, and *Cleistogenes squarrosa* on soil SOC storage in the 0–10 cm (a), 10–20 cm (b), and 20–30 cm (c) layers.

4. Discussion

4.1. Effects of Mowing Regimes on Plant Communities

As one of the most important grassland ecosystems in the typical grassland area of Inner Mongolia, the *Stipa grandis* steppe is a main site of mowing activity. Appropriate mowing management represents one effective way to utilize grassland resources, which helps to maintain plant productivity and soil quality [30]. Research has shown that the aboveground biomass and litter decreased with increasing mowing intensity according to $M_{1/2}$, $M_{2/3}$, $M_{1/1}$, and $M_{2/1}$ measures compared to CK (Figure 1a,c). These results are in line with the findings of Yankai Zhong et al. [31], who reported that light mowing disturbances are conducive to the improvement of vegetation production and proportion of fine herbage compared to high-intensity mowing disturbances. The experiment on the underground biomass of the community in different mowing regimes showed that mowing twice a year and mowing once a year exerted a negative impact on BB in the surface soil layer (0–20 cm), consistent with earlier reports [32]. The reason for this is most probably that high-frequency mowing treatment led to a continuous reduction in standing litter and litter in the grassland plant community. In addition, without the input of live storage excreta, it is impossible to return materials to the grassland ecosystem, which will cause reduced soil organic matter input and soil nutrients to become increasingly barren. These results show that high-frequency mowing has a negative effect on plant root growth, which would affect the return of belowground C resources to the soil. However, too much litter was accumulated on the surface over 19 years without mowing, which had an interception effect on the rainwater, inevitably affecting its leakage to the deep soil and, thus, affecting the water absorption of roots. This effect is more pronounced when precipitation is low. In addition, when the ground is covered with thick litter, sunlight cannot directly reach the soil surface, resulting in a slow increase in soil temperature during early spring and a negative impact on the growth and development of plants, as well as the growth of roots. In contrast, light mowing reduced the ground cover, playing a role in promoting root growth with no adverse effect. Therefore, it is speculated that moderate mowing regimes can promote root growth. Moreover, we also found that mowing increased the R:S at 0–20 cm depth, with no direct effects in deeper layers. The main reason may be that the belowground biomass was also reduced by mowing (in terms of net plant productivity), while the aboveground biomass was affected by mowing to a greater extent. This result is consistent with previous findings with respect to the distribution of biomass between belowground and aboveground under different mowing intensities in the typical steppe grasslands of Inner Mongolia [33]. This also indirectly explains that soil carbon reduction is affected by the reduced contribution of plant roots by mowing. Therefore, we recommend that plant communities regulate R:S by adapting to different mowing regimes, consequently affecting C inputs and cycling.

Different mowing regimes affected plant productivity by altering plant community composition. After the 19-year mowing experiment, we discovered a sharp decline in *Leymus chinensis* biomass when mowing once every 2 years, whereas mowing treatments twice every 3 years led to an increase in *Stipa grandis* biomass. These increases were mainly attributed to the compensation effects of plant functional groups [34]. These changes compensated for the biomass loss caused by the decline in *Leymus chinensis* when mowing once every 2 years and mowing twice every 3 years; however, with the increase in mowing intensity, the plant community undergoes retrogressive succession, whereby the proportions of the dominant plant species in the original community, *Stipa grandis* and *Leymus chinensis*, decreased gradually, while the proportion of short grass, *Cleistogenes squarrosa*, increased. Ultimately, this led to a significant decrease in aboveground community biomass due to *Cleistogenes squarrosa* having a much lower biomass compared to *Stipa grandis* and *Leymus chinensis*. We also speculate that changes in plant community composition might drive the biomass allocation between shallow and deep layers, resulting in the biomass being more distributed in the 0–10 cm root layer. Due to the growth and decline in plant populations caused by mowing, the proportion of *Cleistogenes squarrosa* with a shallow

root distribution in the community increased; accordingly, the distribution range of plant roots also naturally became shallower. Our observations in this regard are in line with results from previous studies [35], but additional research is needed to more precisely determine how different mowing regimes affect plant community composition, as well as their contribution to allocation between AB and BB or between shallow and deep layers.

4.2. Effects of Different Mowing Gradients on SOC

The results of this study showed that light mowing maintained SOC storage, while moderate- and high-frequency mowing significantly decreased SOC storage in the topsoil layer (0–10 cm) compared to enclosure control. With increasing depth, only mowing twice a year significantly decreased the SOC storage in the 10–20 cm soil layer with no significant effects on SOC storage across different mowing treatments in the deeper layer (20–30 cm). This indicates that soil C storage is less active in the deep soil layer than in the topsoil layer. Our findings also showed that soil C storage is strongly influenced by AB, BB, litter, and R:S in the surface soil. Liang et al. [36] investigated the grazing effects on soil organic carbon in Qinghai–Tibetan Plateau grasslands and revealed the significant negative effects of grazing on AB, litter, SOC, and TN, which compares well with our study. It is noteworthy that our findings are consistent with a recent study showing that, with increased mowing frequency, the SOC and TN content showed a decreasing trend in the topsoil. There are multiple potential reasons for mowing decelerating soil C storage in the surface soil. Firstly, mowing leads to a reduction in surface litter, which in turn reduces the carbon input to the soil while also reducing the ability to conserve the topsoil with high soil organic carbon. Secondly, the decrease in AB reduces the potential of grassland to capture atmospheric CO₂ via plant photosynthesis, thereby decreasing the source of SOC input in the soil [37]. Thirdly, mowing accelerates the release of C from soil to the atmosphere by promoting soil respiration [38].

From the results, we found that the different mowing gradients indirectly influenced SOC storage by altering plant community composition, which is highly consistent with the grazing influence on soil C pools through selective foraging by causing changes in plant community composition, as presented by Liang et al. [36]. Mowing can regulate plant community composition and lead to the interaction of stress-tolerant and competing species, thereby affecting AB, BB, R:S, and C input and cycling.

4.3. Effects of Mowing Regimes on MBC and MBC/MBN

This study demonstrated that the soil MBC significantly decreased under moderate- and high-frequency mowing compared to CK according to soil depth, indicating that long-term mowing is deleterious for microbial growth. This result is in agreement with studies conducted in semiarid grassland, which indicated that grazing remarkably suppressed soil MBC [39]. In our study, MBC in the 0–20 cm topsoil was positively and significantly correlated with SOC storage (Figures 4 and A2a). These patterns indicate that soil C and nutrient contents may determine the distribution of microbial biomass C. Both above-ground and belowground factors, such as photosynthetic C supply, litter quantity [40], root exudates [41], and soil microbial community structure, can affect microbial biomass. Firstly, the demand for C of soil microbes is met by the available C originating from the translocation of photosynthates to the rhizosphere [42]. However, by cutting off plant photosynthesis C supply and removing a large number of nutrients through leaf tissue damage, mowing may also directly lead to microbial substrate limitation [43], reducing the consequent microbial biomass under moderate- to high-frequency mowing. Secondly, the decrease in soil water content after mowing may have negative effects on soil MBC by inhibiting root growth and exudation (especially fine roots) [44]. Thirdly, our results show that the soil MBC/MBN decreased under mowing disturbance at 0–10 cm and 20–30 cm depths, indicating an alteration of the soil microbial community structure, e.g., a decrease in fungi [45]. Fungi are the key drivers of recalcitrant SOC decomposition, with a greater

carbon use efficiency than bacteria [4]; hence, their decrease may contribute to soil microbial C limitation and microbial biomass decline [46].

4.4. Implications for Typical Steppe Sustainable Management in Inner Mongolia

The 19-year experiment with different mowing regimes had different impacts on SOC stock in the semiarid grassland of North China. This could be attributed to the different types of nutrient resources in the grassland ecosystem [12]. Soil C storage in the high-frequency mowing plot was more affected compared to the low-frequency mowing plot. High-frequency mowing reduced plant photosynthetic capacity and led to lower AB and litter biomass. This, in turn, affected the supply of nutrients to the root system, indirectly leading to microbial substrate limitation. The low-frequency mowing plot allowed more soil water and nutrient resources to be obtained for root growth compared with the high-frequency mowing plot, which in turn led to higher BB allocation, increasing SOC accumulation. On the other hand, high-frequency mowing caused a sharp decline in *Stipa grandis* biomass and led to retrogressive succession in the plant community. Compared to high-frequency mowing, low-frequency mowing maintained a stable plant community composition and higher plant biomass, thereby supporting soil SOC storage [47]. However, such ecosystem processes will eventually limit plant productivity when the mowing management time is extended. Therefore, it is considered that low-frequency mowing $M_{1/2}$, followed by $M_{2/3}$, might be the appropriate sustainable mowing management regime for the semiarid grassland evaluated in this study, which may lead to considerable positive feedback on plant production mediated by soil nutrient cycling.

5. Conclusions

In the present work, the effects of five grassland mowing regimes (no mowing, mowing once every 2 years, mowing twice every 3 years, mowing once a year, and mowing twice a year) on SOC storage in the 0–30 cm soil depth range were compared in a semiarid grassland of North China. The pattern of variation in soil SOC storage in the 0–20 cm layer was consistent across different mowing regimes, always remaining greatest in CK, with the order $CK > M_{1/2} > M_{2/3} > M_{1/1} > M_{2/1}$; no significant difference existed in the soil 20–30 cm layer. At the same time, all treatments led to a significant decrease in SOC storage with depth. Mowing management regulated SOC storage in the 0–20 cm layer through the interaction of several interrelated mechanisms (plant community composition, productivity of vegetation, litter biomass, soil microbial biomass, and resource allocation between aboveground plants and belowground roots), while the plant community composition regulated SOC storage in the 20–30 cm layer. We, therefore, consider that, from the perspective of maintaining C storage in grassland ecosystems and economic sustainability and profitability, mowing once every 2 years is likely the optimal mowing management strategy. In addition, mowing should be combined with proper N fertilization management, which helps to promote plant recovery, increase nutrient content in the soil, promote nitrogen fixation, and maintain a dynamic balance of nutrients. Research in the future will be required to understand the effects of different mowing regimes on the vertical distribution of soil C, N, and P, as well as their sequestration at lower soil depths, thus increasing existing knowledge of the grassland ecosystem stability mechanism.

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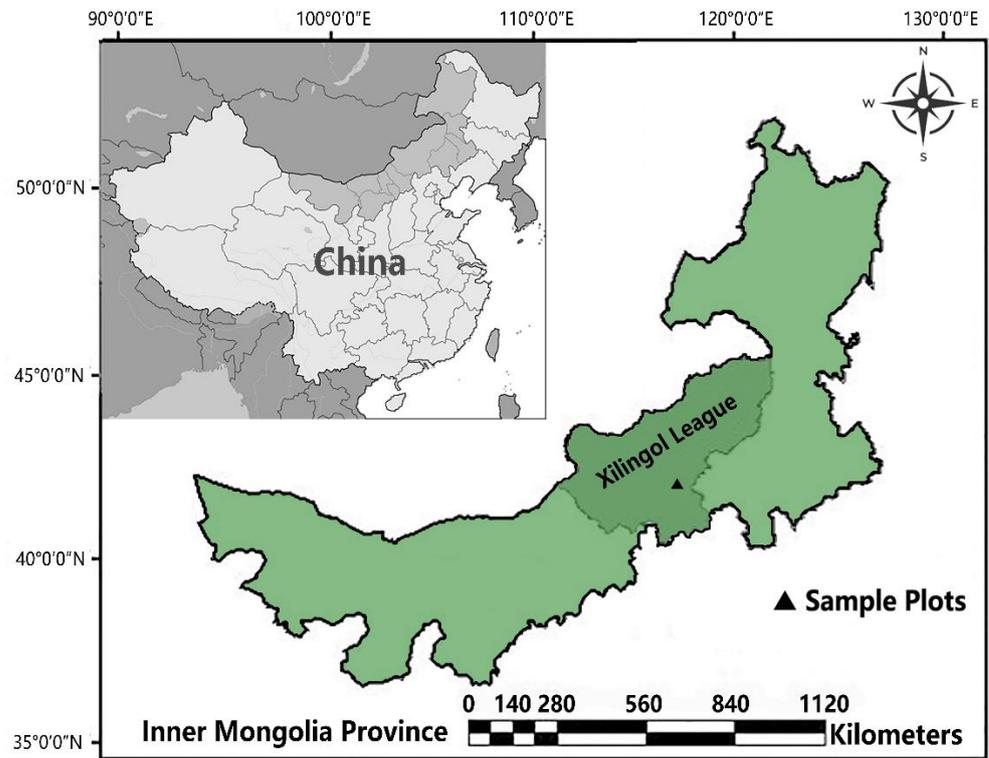
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Data Availability Statement: Not applicable.

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Conflicts of Interest: The authors declare no conflict of interest.

Appendix A

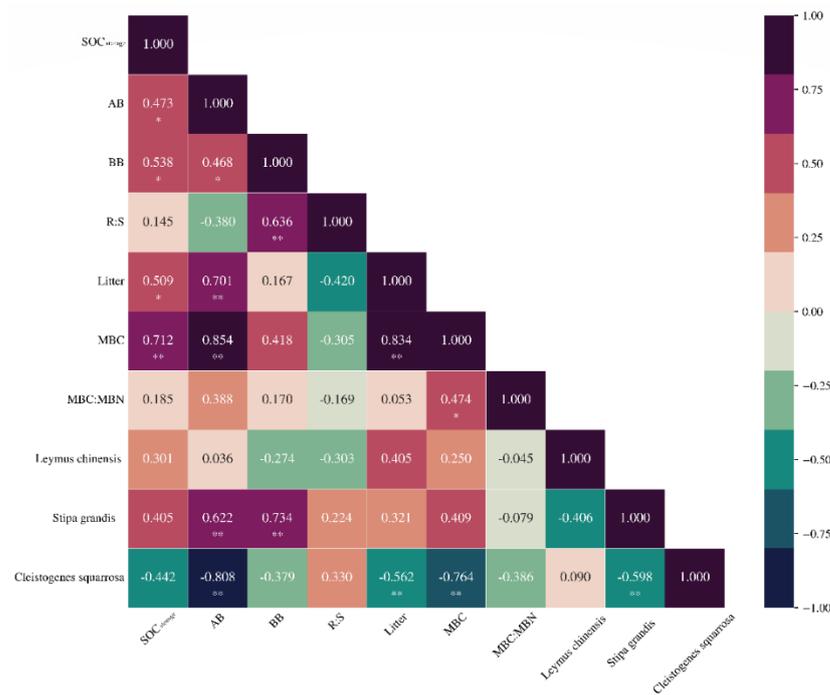


(a)

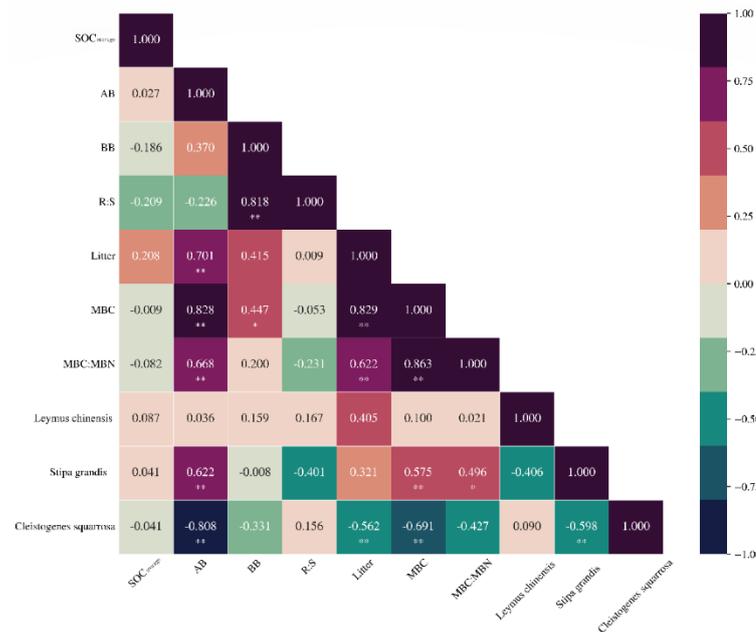


(b)

Figure A1. (a). Location map of the study area and sampling site. (b). Layout of mowing experimental plots on *Stipa grandis* grassland. The four replicate plots for each of the four mowing frequencies (cuts per year) were randomly assigned to 24 permanent plots that were 20 m × 30 m in size. M_{1/2}: mowing once every 2 years (mowing on 16 August); M_{2/3}: mowing twice every 3 years (mowing on 16 August); M_{1/1}: mowing once a year (mowing on 16 August); M_{2/1}: mowing twice a year (mowing on 25 June and 12 September). The control plots were sampled at CK_a.



(a)



(b)

Figure A2. (a). Pearson correlation coefficients between plant and soil properties in the 10–20 cm layer. Positive correlations are labeled as “+”, and negative correlations are labeled as “–”. Significance levels are as follows: ** $p < 0.01$, * $p < 0.05$. AB: plant aboveground biomass; BB: plant belowground biomass; R:S: root–to–shoot ratio; SOC_{storage}: soil organic carbon storage; MBC: microbial biomass C; MBC/MBN: soil MBC to soil MBN ratio; relative aboveground biomass of *Stipa grandis*, *Leymus chinensis*, and *Cleistogenes squarrosa*. (b). Pearson correlation coefficients between plant and soil properties in the 20–30 cm layer. Positive correlations are labeled as “+”, and negative correlations are labeled as “–”. Significance levels are as follows: ** $p < 0.01$, * $p < 0.05$. AB: plant aboveground biomass; BB: plant belowground biomass; R:S: root–to–shoot ratio; SOC_{storage}: soil organic carbon storage; MBC: microbial biomass C; MBC/MBN: soil MBC to soil MBN ratio; relative aboveground biomass of *Stipa grandis*, *Leymus chinensis*, and *Cleistogenes squarrosa*.

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