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Impacts of Grazing Disturbance on Soil Nitrogen Component Contents and Storages in a *Leymus chinensis* Meadow Steppe

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Abstract: Long-term grazing leads to soil degradation in Inner Mongolia grassland. Based on the Hulunbeier meadow steppe, the variation characteristics of soil nitrogen content and storage in soil layers between 0–40 cm, under six different grazing intensities, and the response of vegetation and other physical and chemical properties of soil to grazing were studied. The main results were as follows: (1) Moderate grazing increased soil total nitrogen (TN), soluble total nitrogen (STN) and microbial biomass nitrogen (MBN) contents, while heavy grazing decreased MBN content. In the year with more rain, heavy grazing increased nitrate nitrogen (NO_3^--N) content and storage, while less rain increased ammonium nitrogen (NH_4^+-N) content. (2) The proportion of 0–40 cm nitrogen components showed an upward trend in the year with more rain, and the opposite in the years with less rainfall with the increase of grazing intensity. Soil soluble organic nitrogen (SON) and NO_3^--N storages decreased and MBN storage increased in rainy years. (3) Soil nitrogen component contents and storages were correlated with plant growth status, soil moisture (SM) and soil bulk density (SBD), and were significantly negatively correlated with soil temperature (ST) and pH ($p < 0.05$). The content and storage of soil nitrogen were affected by grazing, soil, vegetation, meteorological and other environmental factors. Moderate grazing was more conducive to the improvement of soil nitrogen storage capacity and the healthy development of grassland.

Keywords: grazing intensity; meadow steppe; soil nitrogen components; nitrogen storage; total soil nitrogen

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1. Introduction

Nitrogen is a key element in determining an ecosystem's function, plant growth, limiting community primary and secondary productivity, and also regulating the structure and function of grassland ecosystems to some extent [1–5]. Grassland covers 20–40% of the Earth's land surface and supplies approximately 30% of the meat [6]. Grasslands are important nitrogen reservoirs in terrestrial ecosystems, providing nutrients for plants and maintaining species diversity, carbon and nitrogen sequestration, and soil and water conservation [7]. Grazing is one of the main utilization methods of using grassland [8], and is a key factor in controlling the nitrogen pool [9]. Although much research has been carried out previously, there is still uncertainty about the processes by which grazing disturbances affect the soil nitrogen cycle, because the response of the soil nitrogen pool to grazing is complex. It is generally believed that the deposition of livestock manure, urine and trampling behavior, after grazing, affect soil nitrogen mineralization and fixation, increasing the speed of nitrogen cycling; thus, the soil mineral nitrogen of grassland would subsequently increase [10]. Moreover, intensive grazing would lead to a reduction in soil nutrients, species numbers and aboveground biomass [3,11–14], and causing grassland degradation. However, these results do not adequately explain the mechanisms of interaction between grazing and grassland soil nitrogen spatial heterogeneity and vegetation.

Soil TN plays an important role in grassland productivity and is a key indicator of soil fertility [15]. Soil soluble nitrogen components limit the mineralization of organic matter, and their high mobility affects nitrogen sequestration and nutrient loss in deep soil [16,17]. Although less abundant than TN, soluble nitrogen is the most readily depleted and limits plant growth forms [18]. Soil microorganisms drive and regulate the cycling of soil nutrients and their interactions, playing an especially important role in nitrogen transformation [19,20]. Soil microbial biomass is the fastest nutrient that plants can absorb, and it can reflect the adequacy of the soil nutritional status and biological activity [21]. Soil MBN only accounts for 0.5–15.3% of soil TN, but it plays an important role in the conversion process between the organic and inorganic soil nitrogen pools, and it is highly sensitive to soil environmental changes and soil nitrogen changes [22,23]. Grazing exclusion has positive effects on plant productivity, soil nitrogen content and storage [24,25], while under proper grazing, soil fertility and vegetation productivity significantly improved [26]. Overgrazing is the main cause of grassland degradation, which leads to a decrease in soil nitrogen content [27].

This research mainly studied the spatial distribution characteristics of soil nitrogen content and storage, and the changes in soil–vegetation environmental factors after different grazing disturbances in the Hulunbuir meadow steppe. The main objectives were to: (1) study the allocation of soil nitrogen component content and storage in the Hulunbuir meadow steppe under different grazing intensities, and provide basic data for meadow steppe soil quality management; (2) reveal the influence of soil nitrogen component distribution and supply on vegetation growth to guide the implementation of vegetation restoration measures; and (3) predict the potential nitrogen storage capacity in grassland ecosystems under grazing disturbance to provide a basis for the scientific management and utilization of grassland soils.

2. Materials and Methods

2.1. Overview of the Experimental Site

This subject is based on the long-term grazing platform called National Field Scientific Observation and Research Station in the Hulunbuir Grassland, in Inner Mongolia (49°20′ N, 119°57′ E), China, with an average elevation of 666–680 m. The climate is temperate and semiarid continental. The average annual precipitation can reach 400 mm, and the average annual air temperature ranges from −5 °C to −2 °C. The maximum daily temperature is 36 °C in July, and the minimum daily temperature is −48 °C in January. The soil type is mainly black calcium soil or chestnut calcium soil. The grassland type is *Leymus chinensis* meadow steppe, in which the main established species is *L. chinensis*, the dominant species are *Stipa baicalensis* and *Cleistogenes squarrosa*, and the associated species are *Vicia amoena* and *Poa pratensis*.

The experimental platform was established in 2009 with 18 plots randomly distributed in a homogeneous area of 90 hectares. The experiment was divided into 6 grazing intensities with 3 replications, and the animal rates were 0, 0.23, 0.34, 0.46, 0.69 and 0.92 cow.AU/ha (1 standard animal unit (AU) equivalent to 500 kg of cattle). The different grazing intensities were implemented using 250 to 300 kg of grazing cattle with 6 grazing intensities of 0, 2, 3, 4, 6 or 8 cattle, for a total of 69 cattle, and these categories were denoted as no grazing ($G_{0.00}$), very light grazing ($G_{0.23}$), light grazing ($G_{0.34}$), moderate grazing ($G_{0.46}$), heavy grazing ($G_{0.69}$) and very heavy grazing ($G_{0.92}$), respectively. During the plant growing seasons in 2020 and 2021, cattle were grazed for 120 consecutive days from June to September. During this period, grazing cattle were present in the test area both day and night without supplemental feed, but with an adequate water and salt supply. Samples for this study were collected in August 2020 and 2021 (Figure 1).

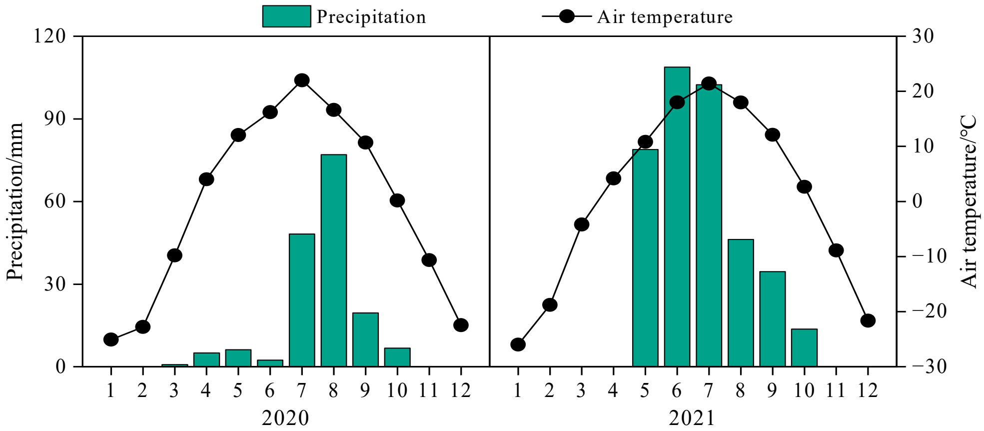

Figure 1. Map of meteorological variation at sample sites.

2.2. Sample Collection and Measurement Method

The samples were collected in August 2020 and 2021. Five points were selected at the corresponding positions of the community sampling, and a soil drill with a diameter of 5 cm was used to repeat three times of the sampling at each point. Soil samples of 0–40 cm were drilled and generally divided into four soil layers: 0–10 cm, 10–20 cm, 20–30 cm and 30–40 cm. Then, the soil samples collected from various points at the same level were mixed and screened in equal proportions, and taken to the laboratory for analysis.

The Kjeldahl method was used for the determination of soil TN. SON was calculated as the difference between STN and inorganic nitrogen. STN was determined by the potassium sulphate oxidation method, and soluble inorganic nitrogen was determined by flow cytometry. NO_3^--N and NH_4^+-N were extracted by potassium chloride and measured by flow cytometry. MBN was extracted by potassium sulphate and determined by flow cytometry. The drying method was used to determine the SM. Soil temperature was measured by a pen-probe thermometer. SBD was measured by the ring knife method. Soil pH was extracted by water without carbon dioxide and measured by a pH meter. The aboveground biomass was obtained by cutting, and the sample size was 1 m × 1 m.

2.3. Calculation and Statistics

Total nitrogen storage was calculated from the soil nitrogen content (*Bj*), soil bulk density (*Cj*), soil depth (*Dj*) and soil layer (*j*). The nitrogen component storage formula is as follows:

$$RN = \sum_{j=1}^{n} (Bj \times Cj \times Dj) \times 10 \quad (1)$$

where *Bj* is in g/kg, *Cj* is in g·cm^{-3}, *Dj* is in cm, and 10 is the unit conversion coefficient.

In this study, data sorting and statistical analysis were performed using IBM SPSS, version 22.0 (IBM Corporation, Armonk, NY, USA) and Microsoft Excel 2021 (Microsoft, Seattle, WA, USA), and figures were drawn using Origin 2022 (OriginLab Corporation, Northampton, MA, USA). The variance analysis of soil nitrogen components content and storage under different grazing intensities was carried out by multifactor analysis, and multiple comparisons were made by the LSD and Duncan methods. Pearson correlation analysis was performed, and the significance level was set as $p < 0.05$, while $p < 0.01$ was considered extremely significant.

3. Results

3.1. Changes in Soil Nitrogen Components Content under Different Grazing Intensities

In 2020, the content of nitrogen components in 0–40 cm soil layer did not change significantly compared with no grazing. In 2021, compared with no grazing, SON content showed a trend of decreasing first and then increasing, NH_4^+-N content increased in $G_{0.92}$, while MBN content decreased significantly in $G_{0.92}$. The response of soil TN to grazing was mainly concentrated in the upper soil layer. The soil TN content (2020) in $G_{0.34}$ was significantly higher than that in $G_{0.23}$ and $G_{0.69}$ in the 0–10 cm soil layer and soil surface ($p < 0.05$), with a concentration up to 3.93 g/kg. However, compared to no grazing, the soil TN content in $G_{0.00}$ and $G_{0.46}$ was significantly higher than that in other grazing intensities in the 10–20 cm soil layer ($p < 0.05$), with a decrease of 2.58% to 11.30%. Soil TN (2021) in $G_{0.34}$, $G_{0.69}$ and $G_{0.92}$ was significantly higher than that in $G_{0.46}$ at the soil surface.

The soil STN content (2020) in $G_{0.34}$ was significantly higher than that in $G_{0.46}$, $G_{0.69}$ and $G_{0.92}$ ($p < 0.05$) in the 30–40 cm soil layer, decreasing by 16.55%, 23.19% and 18.62%, respectively. Compared with no grazing, the SON content (2021) in the 30–40 cm soil layer increased significantly in $G_{0.23}$ and $G_{0.92}$. In the 10–20 cm soil layer, the soil NO_3^--N content (2020) in $G_{0.92}$ was significantly higher than that in the other grazing intensities, with values up to 7.57 mg/kg. In 2021, the NH_4^+-N content of $G_{0.69}$ was significantly higher than that of $G_{0.00}$–$G_{0.46}$ in the 10–20 cm soil layer ($p < 0.05$), with a decrease of 15.09–18.28%. In the 20–30 cm soil layer, the MBN content (2021) of $G_{0.92}$ was significantly lower than that of no grazing (Figure 2).

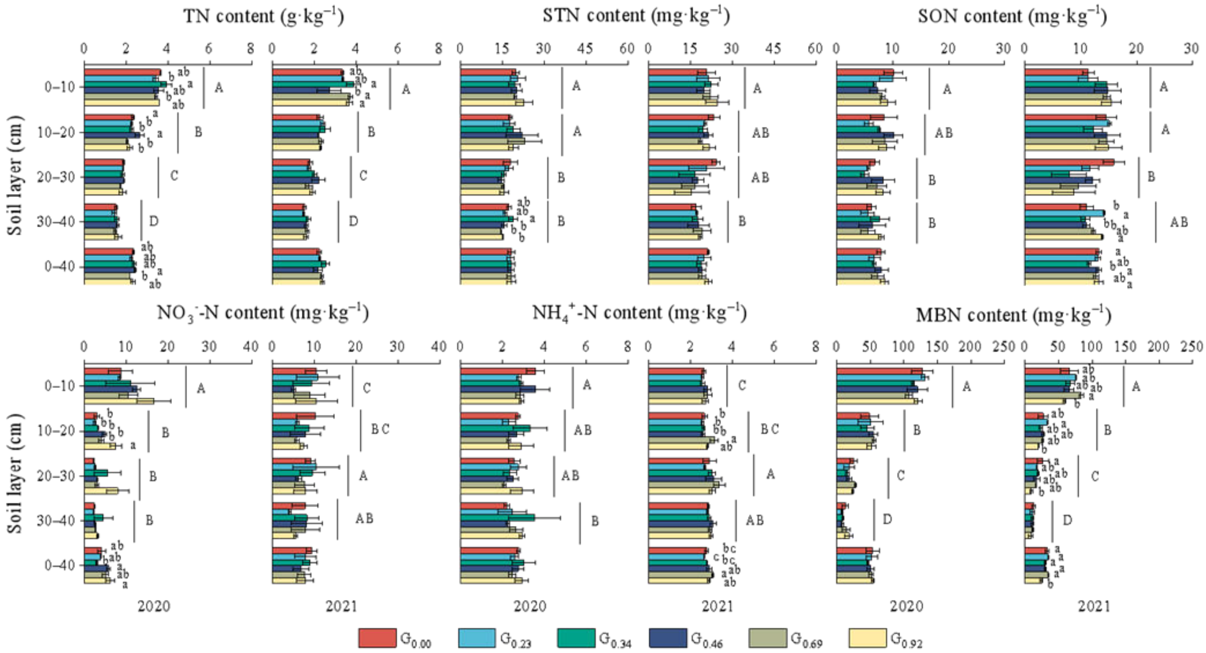

Figure 2. Soil nitrogen content dynamics at different grazing intensities over two years. Note: Different lowercase letters indicate significant differences between different grazing intensities ($p < 0.05$), different capital letters indicate significant differences between different soil layers ($p < 0.05$), and no letters indicate no significant difference.

3.2. Interaction Analysis of Soil Nitrogen Components

Soil nitrogen components changed significantly under interannual, soil layer and the interaction of interannual and soil layer (Table 1). The SON, NO_3^--N, MBN contents and storages, ST, SM, and SBD showed significant differences among the different years ($p < 0.01$). The STN content, TNS storage and pH changed significantly in different years ($p < 0.05$). The TN, NO_3^--N, MBN contents and storages, and the contents of STN, SON were significant differences among the different soil layers ($p < 0.01$). The interaction between grazing intensity and soil layer significantly affected SM and pH ($p < 0.01$). Under the interaction of interannual and soil layer, the contents and storages of NO_3^--N and MBN, and SM changed significantly ($p < 0.01$). The combined effects of interannual, grazing intensity and soil layer had no significant effect on soil nitrogen content, storage and soil characteristics ($p > 0.05$).

Table 1. Multifactor analysis of soil nitrogen components in each layer under grazing disturbance over two years.

Source of Variation	**IN**		**GI**		**SL**		**IN × GI**		**IN × SL**		**GI × SL**		**IN × GI × SL**	
	F	***p***	**F**	***p***	**F**	***p***	**F**	***p***	**F**	***p***	**F**	***p***	**F**	***p***
TN content	0.02	0.88	2.01	0.08	366.28	<0.01	2.14	0.07	1.33	0.27	1.79	<0.05	1.15	0.32
STN content	4.14	<0.05	0.23	0.95	6.47	<0.01	0.20	0.96	0.14	0.94	0.62	0.85	0.45	0.96
SON content	117.67	<0.01	1.21	0.31	6.33	<0.01	0.28	0.93	0.76	0.52	0.76	0.72	1.43	0.15
NO_3^--N content	10.26	<0.01	0.99	0.43	9.49	<0.01	1.42	0.22	4.18	<0.01	0.37	0.98	0.35	0.99
NH_4^+-N content	0.70	0.40	0.84	0.53	0.52	0.67	1.43	0.22	3.86	<0.05	0.64	0.83	0.85	0.63
MBN content	111.63	<0.01	1.01	0.42	400.17	<0.01	1.18	0.32	37.46	<0.01	0.46	0.96	0.83	0.64
TN storage	4.77	<0.05	0.57	0.72	133.71	<0.01	0.85	0.52	0.94	0.43	0.51	0.93	0.60	0.87
STN storage	0.25	0.62	0.52	0.76	1.49	0.22	0.59	0.71	0.46	0.71	0.78	0.70	0.65	0.83
SON storage	81.26	<0.01	1.26	0.29	2.08	0.11	0.80	0.55	0.46	0.71	0.91	0.56	1.48	0.13
NO_3^--N storage	8.87	<0.01	0.96	0.45	5.76	<0.01	1.83	0.11	4.46	<0.01	0.39	0.98	0.38	0.98
NH_4^+-N storage	0.38	0.54	0.33	0.90	2.59	0.06	0.94	0.46	2.24	0.09	0.66	0.81	0.75	0.73
MBN storage	102.81	<0.01	1.33	0.26	260.77	<0.01	1.26	0.29	28.27	<0.01	0.55	0.90	0.35	0.99
ST	72.23	<0.01	0.50	0.77	0.00	1.00	0.02	1.00	0.00	1.00	0.00	1.00	0.00	1.00
SM	305.15	<0.01	3.42	<0.01	280.58	<0.01	3.23	<0.05	35.72	<0.01	0.47	0.95	0.65	0.82
SBD	20.49	<0.01	1.37	0.24	31.07	<0.01	1.55	0.18	2.21	0.09	0.91	0.56	1.00	0.47
pH	4.76	<0.05	4.08	<0.01	6.80	<0.01	5.59	<0.01	0.88	0.45	0.51	0.93	1.23	0.27

Note: IN: interannual, GI: grazing intensity, SL: soil layer, TN: total nitrogen, STN: soluble total nitrogen, SON: soluble organic nitrogen, NO_3^--N: nitrate nitrogen, NH_4^+-N: ammonium nitrogen, MBN: microbial biomass nitrogen, ST: soil temperature, SM: soil moisture, SBD: soil bulk density.

3.3. The Proportion of Nitrogen Components to Total Nitrogen

Grazing increased the proportion of 0–40 cm nitrogen components in 2020, but decreased in 2021, except for the proportion of NH_4^+-N. With increasing soil depth, the proportions of STN, SON and NH_4^+-N increased ($p < 0.05$), the proportion of MBN decreased ($p < 0.05$), and the proportion of NO_3^--N showed no significant difference ($p > 0.05$). In 2021, the proportion of SON tended to increase in the 0–20 cm soil layer as grazing disturbance increased, while it decreased in the 20–40 cm layer. The proportion of SON in $G_{0.46}$ was significantly higher than in $G_{0.00}$~$G_{0.34}$ and $G_{0.69}$ in soil 0–10 cm layer ($p < 0.05$). The proportion of soil MBN in $G_{0.23}$ was significantly higher than that in $G_{0.34}$ and $G_{0.69}$ in the 10–20 cm soil layer. In the 20–30 cm soil layer, $G_{0.00}$ was significantly higher than in $G_{0.92}$ ($p < 0.05$), which decreased by 0.99% (Figure 3).

3.4. Changes in Soil Nitrogen Storage

Soil TN and STN storage changed little in different years, compared with 2020, SON and NO_3^--N storage increased, and MBN storage decreased in 2021. In 2020, the NO_3^--N storage in $G_{0.92}$ was significantly higher than that in the other grazing intensities in the 10–20 cm soil layer ($p < 0.05$); compared with no grazing, it increased by 71.27%. In 2021, the SON storage in $G_{0.00}$ was significantly higher than that in $G_{0.34}$ in the 20–30 cm soil layer ($p < 0.05$) with a 56.30% reduction. The storage of MBN in the 0–10 cm and 10–20 cm soil layers in $G_{0.23}$ was significantly higher than that in $G_{0.92}$ ($p < 0.05$), increasing by 34.22% and 45.34%, respectively (Figure 4).

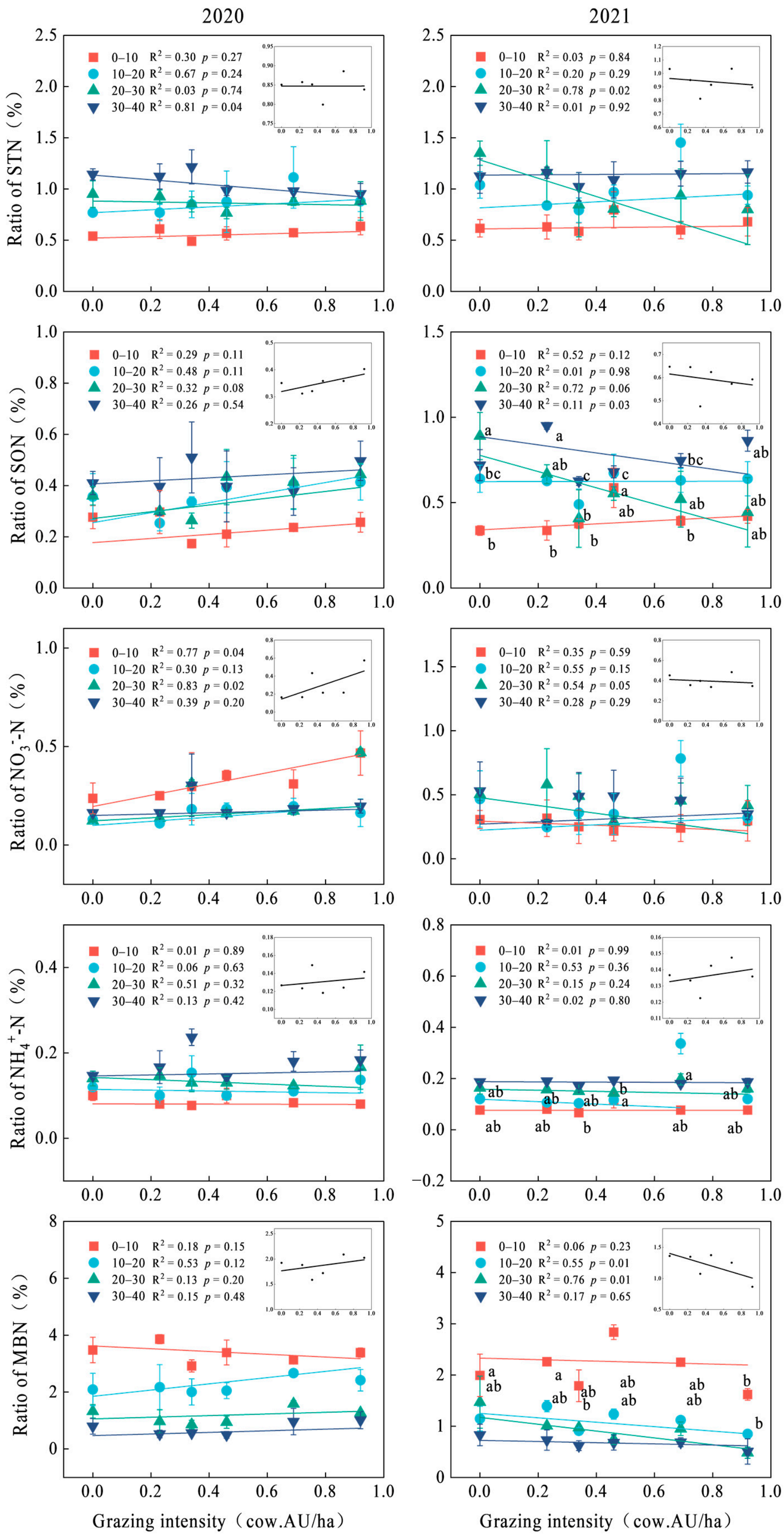

Figure 3. Dynamics of soil nitrogen components under different grazing intensities over two years. Note: The small figure on the upper right is the trend of 0–40 nitrogen component ratio. Different lowercase letters indicate significant differences between different grazing intensities ($p < 0.05$), and no letters indicate no significant difference.

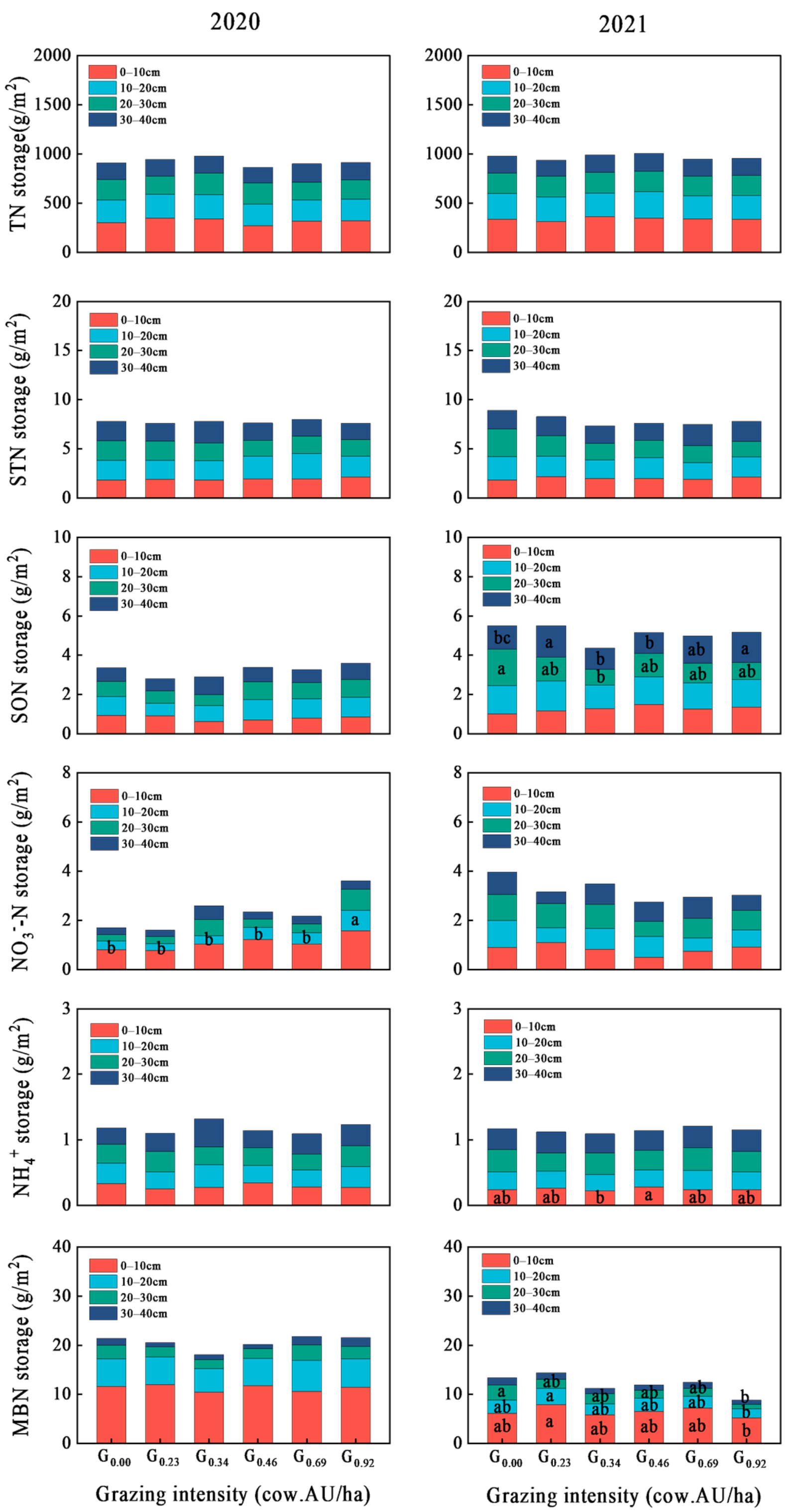

Figure 4. Dynamics of soil nitrogen stocks under different grazing intensities over two years. Note: Different lowercase letters indicate significant differences between grazing intensities ($p < 0.05$), and no letters indicate no significant difference.

3.5. Correlation between Nitrogen Components and Environmental Factors

The SON content and storage showed a highly significant positive correlation with height, coverage and aboveground biomass ($p < 0.01$). The MBN content showed a highly significant negative correlation with coverage and aboveground biomass ($p < 0.01$), and a significantly negative correlation with height ($p < 0.05$). The MBN storage showed a highly significant negative correlation with coverage ($p < 0.01$), and a significantly negative correlation with height and aboveground biomass ($p < 0.05$). The ST showed a highly significant positive correlation with MBN content and storage; a highly significant negative correlation with SON content and storage ($p < 0.01$); and a significantly negative correlation with NO_3^--N content and storage ($p < 0.05$). The SM showed a highly significant positive correlation with SON content and storage and NO_3^--N content; a highly significant negative correlation with MBN content and storage ($p < 0.01$); and a significantly positive correlation with STN content and NO_3^--N storage ($p < 0.05$). The SBD showed a highly significant positive correlation with MBN content and storage and STN storage ($p < 0.01$); a significantly positive correlation with STN content and NH_4^+-N storage; and a significant negative correlation with SON and NO_3^--N contents ($p < 0.05$). The TN content showed a significantly negative correlation with pH ($p < 0.05$). The air temperature and precipitation showed a highly significant positive correlation with SON content and storage and NO_3^--N content; a highly significant negative correlation with MBN content and storage ($p < 0.01$); and a significantly positive correlation with STN content and NO_3^--N storage ($p < 0.05$).

Grazing intensity showed a highly significant positive correlation with density; a highly significant negative correlation with height, cover, and AGB ($p < 0.01$); and was significantly negative with SM ($p < 0.05$). The contents of soil nitrogen components and their corresponding storage showed a highly significant positive correlation ($p < 0.01$). The STN content showed a highly significant positive correlation with NO_3^--N content and storage ($p < 0.01$), and a significantly positive correlation with SON content and storage ($p < 0.05$) (Figure 5).

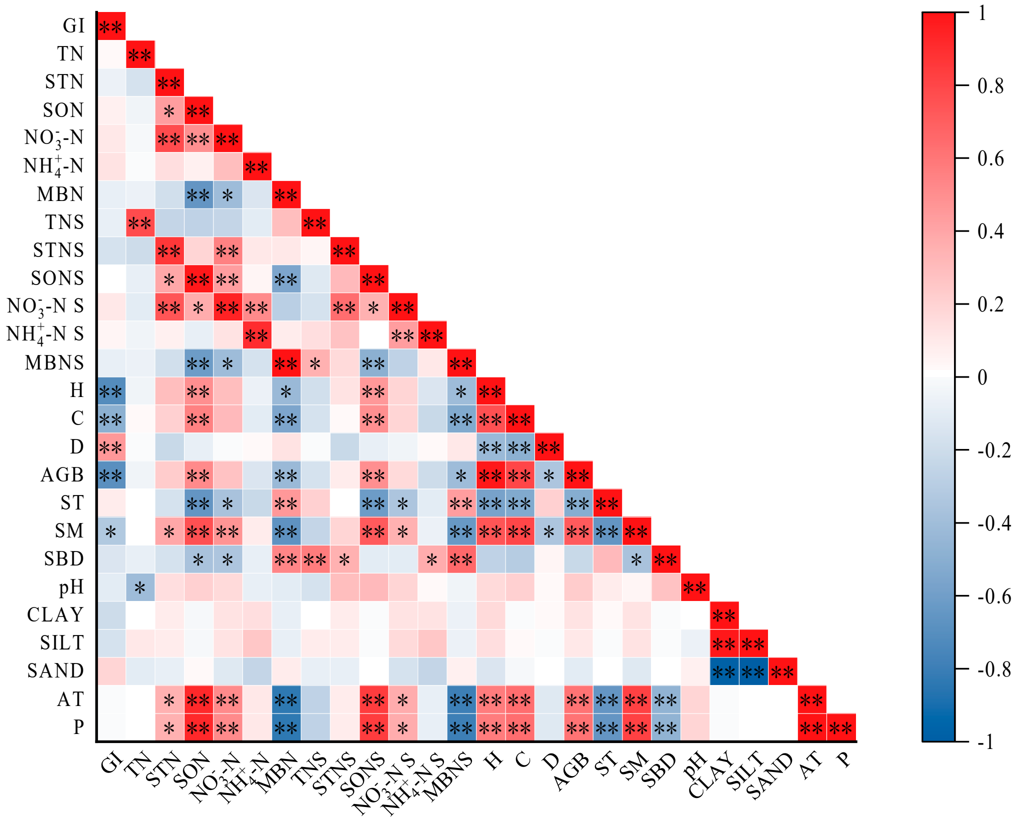

Figure 5. Correlation of different grazing intensities over two years. * $0.01 < p \leq 0.05$, ** $0.001 < p \leq 0.01$; GI: grazing intensity, TN: total nitrogen, STN: soluble total nitrogen, SON: soluble organic nitrogen, NN: NO_3^--N, AN: NH_4^+-N, MBN: microbial biomass nitrogen, TNS: total nitrogen storage, STNS: soluble total nitrogen storage, SONS: soluble organic nitrogen storage, NNS: NO_3^--N storage, ANS: NH_4^+-N storage, MBNS: microbial biomass nitrogen storage, H: height, C: coverage, D: density, AGB: aboveground biomass, ST: soil temperature, SM: soil moisture, SBD: soil bulk density.

4. Discussion

4.1. Effect of Grazing on the Contents of Different Nitrogen Components in Soil

The effects of grazing livestock on soil nitrogen content were mainly divided into two aspects: On the one hand, they directly affected plant photosynthesis through feeding behavior, reduced the aboveground productivity of grassland, and limited the input of nitrogen; on the other hand, livestock urine and feces returned some nitrogen to the soil through volatilization and leaching, and were mainly concentrated in the soil surface, resulting in an increase in nitrogen content in the surface layer. In this study, due to the relatively stable soil total nitrogen, the soil total nitrogen content changed little in two years. The effects of grazing on soil TN were mainly concentrated in the range of 0–20 cm, and with increasing grazing intensity, the soil TN content showed a trend of first increasing and then decreasing. Compared with no grazing, the soil TN content increased under moderate grazing, while overgrazing reduced the soil nitrogen content. This result is because the feeding and trampling of moderate grazing livestock stimulated plant growth and microbial activity. Due to the increase in surface coverage, SM increased, and root exudates and root biomass increased, which promoted soil respiration and soil mineralization. In contrast, overgrazing inhibited plant biomass accumulation and microbial activity, significantly reduced SM, destroyed soil aggregates, reduced soil permeability, and ultimately reduced nitrogen input [4,4,28].

The soil soluble nitrogen content varied in this study, but both the STN and the SON contents decreased significantly with increasing soil depth. The response to grazing was mainly concentrated in the 30–40 cm soil layer, where STN showed a trend of increasing and then decreasing as grazing intensity increased, while the SON content increased significantly in $G_{0.23}$ and $G_{0.92}$, compared with no grazing. The SON in grazing soil was the main output of nitrogen leaching, so soluble nitrogen changed significantly in deeper soil [29]. Light grazing stimulated plant growth and microbial activity, increased root exudates and root biomass, and promoted soil mineralization. In contrast, the soluble organic nitrogen contained in manure of overgrazed livestock increased nitrogen input [30,31]. In 2021, the temperature is higher than that in 2020, the precipitation is less, the leaching effect in the soil is less, and the microbial activity is reduced. The precipitation in 2020 is more than that in 2021, and the soil moisture is sufficient, which accelerates the activity of nitrifying bacteria. Nitrification leads to an increase in soil nitrate nitrogen. In 2021, on the contrary, ammonium nitrogen increases. NO_3^--N and NH_4^+-N were concentrated in the 10–20 cm soil layer, and their contents increased in $G_{0.92}$. The NO_3^--N and NH_4^+-N content are two forms of nitrogen required by plants; plant roots were mostly distributed at 10–20 cm, and livestock manure and urine substances in the overgrazing treatment indirectly increased the fertility of grassland soil [32,33]. The MBN content decreased significantly with increasing soil depth, and in $G_{0.92}$, it decreased in each soil layer. Overgrazing will break the balance of the ecosystem, change the spatial pattern and size of soil pores, and change the soil oxygen content, thereby reducing soil microbial activity [34,35].

4.2. Effects of Grazing on the Proportion and Storage of Different Nitrogen Components in Soil

The proportion of soil nitrogen components to TN was MBN > STN > SON > NO_3^--N > NH_4^+-N. Under grazing disturbance, the proportion and storage of different nitrogen components in soil did not change in the 2020 with more precipitation, but showed different changes in 2021, which indicated that the changes in soil nitrogen components were likely to be closely related to hydrothermal conditions. The changes in nitrogen fractions affected by grazing in 2021 were as follows: as the depth of the soil layer increased, the proportions of STN, SON and AN increased, while the proportion of MBN decreased. This result indicates that the response of MBN to soil depth change was the most obvious. The decrease in soil organic matter content led to a decrease in the soil MBN source and a decrease in soil porosity and bulk density, thus affecting microbial activities and enzyme activities [36–38]. In this research, we found that the proportion of SON increased under moderate grazing, but the change in SON storage was different. This difference was because most of the soluble organic nitrogen in the soil comes from root exudates, litter decomposition, etc., and it is easily leached and reduced under overgrazing, resulting in a decrease in nitrogen mineralization [39–41]. Soil NH_4^+-N storage increased under overgrazing due to the increased ability of microorganisms to capture urine nitrogen, thereby reducing nitrogen loss [42]. The proportion of MBN decreased under overgrazing, and MBN storage increased under moderate grazing, but decreased under overgrazing [43].

4.3. Effects of Grazing on Environmental Factors and Nitrogen Components

The impact of livestock on grasslands is mainly focused plant community composition, productivity and soil physicochemical properties, which in turn affect soil fertility and even lead to soil degradation. In this study, the reason why the community density increased with an increase in grazing intensity was that the intake of cattle reduced palatable plants such as *L. chinensis*, and the presence of *Artemisia frigida* and *Potentilla acaulis* increased. The increase in grazing intensity resulted in decreases in community height and cover, aboveground biomass, and soil-water-holding capacity, thus limiting the input of soil nitrogen sources to the meadow, which was consistent with the results of other studies [44], leading to a decrease in soil TN content. In this study, plant growth status was positively correlated with SON content and reserves, and negatively correlated with microbial nitrogen content and reserves. This is because moderate grazing stimulates plant growth through compensatory responses and returns soil nitrogen in the form of manure, thereby reducing the impact of livestock on the ecosystem [45]. However, when the grazing intensity is too substantial, the nitrogen returned from plant residues to the soil is greatly reduced, the plant community structure changes, the soil nutrient cycle is changed, and the soil microbial activity is reduced [46,47]. The results of this study show that the ST and SBD will reduce SON and NO_3^--N, increase MBN, and SBD will increase the soil STN and NH_4^+-N. The SM will increase STN, SON and NO_3^--N, but reduce MBN. Increased trampling behavior of livestock will cause soil compaction, thereby reducing soil porosity, increasing SBD and ST, increasing water evaporation rate, and affecting soil nitrogen pool [48]. The effects of grazing on soil pH were both negative and positive. After grazing, aboveground biomass decreases, soil evaporation and organic matter decomposition rate increases, and soil salinization are accelerated. Grazing may also lead to soil acidification through urination, thus affecting the nitrogen cycle [49]; these theories rationalize the different performance of soil TN under different grazing intensities.

5. Conclusions

The contents of nitrogen components showed different trends under different grazing intensities. In rainy years, moderate grazing increased STN content, while overgrazing decreased STN and increased NO_3^--N content. On the contrary, moderate grazing reduced SON content, and heavy grazing reduced MBN and NH_4^+-N contents. Under different grazing intensities, the proportion of soil nitrogen components was ordered as MBN > STN > SON > NO_3^--N > NH_4^+-N. In the year with more rainfall, moderate grazing decreased the proportion of NH_4^+-N and decreased SON storage, heavy grazing decreased the proportion of MBN and increased NO_3^--N storage and decreased MBN storage.

The soil nitrogen components changed significantly under interannual, soil layer and the interaction of interannual and soil layer, while grazing had little effect on soil nitrogen fractions. Changes in soil nitrogen components and storages were affected by grazing, but mainly by hydrothermal conditions. The plant growth status and SM were significantly negatively correlated with grazing intensity, SON, NO_3^--N content and storage, and significantly positively correlated with MBN content and storage. The contents and storages of SON and NO_3^--N were significantly negatively correlated with ST and SBD, but the contents and storages of MBN were significantly positively correlated.

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