

Climatic Controls on Stable Carbon and Nitrogen Isotope Compositions of Temperate Grasslands in Northern China

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Abstract

Aims The natural abundances of stable carbon (C) and nitrogen (N) isotopes ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) are extensively used to indicate the C and N biogeochemical cycles at large spatial scales. However, the spatial patterns of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ in plant-soil system of grasslands in northern China and their main driving factors are still not well understood.

Methods We conducted sampling campaigns during 2016-2018 in grasslands of northern China and measured plant and soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ compositions to determine effects soil physicochemical properties and climatic factors on spatial distribution of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$.

Results Generally, plant and soil $\delta^{13}\text{C}$ values increased with the decrease of mean annual precipitation (MAP). The interactions between mean annual temperature (MAT) and soil organic carbon have significant impact on soil $\delta^{13}\text{C}$. However, plant and soil $\delta^{15}\text{N}$ decreased with the increase of MAT. Within all factors, the interactions between MAT and MAP on soil $\delta^{15}\text{N}$ were significant.

Conclusions Our results suggest that C cycling in grasslands of northern China is strongly mediated by plant community and MAT, because C4 species were more prevalent in arid regions. Meanwhile, N cycling is mainly directly regulated by MAT and plant community composition via its effect on the plant $\delta^{15}\text{N}$. All of these will provide scientific references for future research on the C and N biogeochemical cycles of temperate grassland ecosystems in northern China.

1. Introduction

Grasslands, comprising open grassland, grassy shrublands and savannas, cover nearly 40% of the world's land area and provide a wide range of ecosystem services to humans (O'Mara 2012; Bardgett et al. 2021). They store approximately one-third of the total carbon (C) in terrestrial ecosystems and most of the C is stored within 1 m soil layers, which contributes significantly to the mitigation of global climate change (Wang et al. 2016; Xu et al. 2015). In addition, grasslands also play an important role in the global nitrogen (N) biogeochemical cycle (Chen et al. 2021; Risch et al. 2019). Grasslands are vulnerable terrestrial ecosystems due to overexploitation (Liu et al. 2020). C and N cycles in global grassland ecosystems are sensitive to global climate change and land-use change, especially extreme precipitation and global warming in the temperate zone (Wang et al. 2016). However, it is difficult to explore the C and N dynamics by directly measuring the change of C and N stocks because of their relatively slow change processes. Recently, with the rapid development of stable isotope ratio mass spectrometry, stable C ($\delta^{13}\text{C}$) and N ($\delta^{15}\text{N}$) isotope compositions, reflecting C and N transformation processes in plant-soil systems, have become an important tool to study the C and N biogeochemical cycles in terrestrial ecosystems (Dong et al. 2018; Han et al. 2020; Xia et al. 2021).

Nowadays, stable $\delta^{13}\text{C}$ isotope has emerged as useful tool to assess the magnitude and distribution of plant productivity, water use efficiency and soil C turnover rate (Mcdowell et al. 2010; Wu et al. 2018). Previous studies have indicated that plant $\delta^{13}\text{C}$ composition is mainly controlled by plant's photosynthetic pathway, and soil $\delta^{13}\text{C}$ composition mainly depends on the plant-derived organic C and SOC decomposition (An and Li 2015; Dixon et al. 2010). Plant community composition has a distinct influence on the plant $\delta^{13}\text{C}$ (Chen et al. 2021; Luo et al. 2018). For example, the plant community composition with more C4 species will lead to higher stable $\delta^{13}\text{C}$ values (Wu et al. 2019). In addition, forbs with higher water use efficiency also have relatively higher $\delta^{13}\text{C}$ values compared to graminoids and sedges (Liu et al. 2018). Besides, the soil stable $\delta^{13}\text{C}$ composition has become an important integrative measure of soil organic carbon (SOC) input and output (Bird et al. 1996; Wang et al. 2017). Soil stable $\delta^{13}\text{C}$ composition depends not only on that of plant residuals, but also on synthetic action of abiotic and biotic factors (e.g., SOC decomposition, microbial mobilization and immobilization) (Wu et al. 2019; Yang et al. 2015). Besides, the stable $\delta^{13}\text{C}$ composition can differ significantly among various layers within the same soil profiles (Brunn et al. 2014; Carvalhais et al. 2014; Wang et al. 2017). Up-to-date, studies in tropical, temperate and tundra regions have demonstrated that soil physicochemical properties (e.g., pH, C/N ratio and soil moisture) and climatic factors (e.g., mean annual temperature and mean annual precipitation) regulate biogeochemical processes in soil and influence interactions between soil and plants, and can shape the spatial and temporal distribution of stable $\delta^{13}\text{C}$ composition in the terrestrial ecosystem (Nel et al. 2018; Yang et al. 2015).

Compared to the C cycle, the N cycle is more complex due to the various influencing factors along different environment gradients (Craine et al. 2015). Numerous studies have demonstrated that stable $\delta^{15}\text{N}$ values in terrestrial ecosystems are positively correlated with mean annual temperature (MAT) but negatively related to mean annual precipitation (MAP) (Craine et al. 2015; Nel et al. 2018). Besides, stable $\delta^{15}\text{N}$ composition is also influenced by soil C and N contents and other soil physicochemical properties (Craine et al. 2015; Yang et al. 2013). For example, ammonia (NH_3) volatilization will accelerate when soil pH is high, which leads to an abiotic gaseous N loss and higher soil $\delta^{15}\text{N}$ values (Booth et al. 2005; Chen et al. 2021; Yang et al. 2013). Generally, stable $\delta^{15}\text{N}$ signals the openness of N biogeochemical cycle in terrestrial ecosystems (Boeckx et al. 2005). The N input in terrestrial ecosystems by livestock manure, biological N fixation and N deposition could alter the stable $\delta^{15}\text{N}$ composition in plant-soil system (Fang et al. 2011). The stable $\delta^{15}\text{N}$ composition of plant also depends on the various preferences of species to the available N forms and the fractionation during plant mycorrhiza transfer process (Chen et al. 2021; Wu et al. 2019; Xu et al. 2011). The soil stable $\delta^{15}\text{N}$ is mainly controlled by plant N uptake and microbial mediated N-cycling processes (Golluscio et al. 2009).

Being one of the most widely-distributed terrestrial ecosystems, grasslands play a crucial role in the global terrestrial biogeochemical cycles of C and N (Yan et al. 2017; Yao et al. 2018). Grasslands in China are mainly distributed in arid and semi-arid regions, covering an area of approximately 4×10^8 ha and accounting for 41.7% of the country's territory, and contain different grassland types adapted to various climatic conditions and altitudes. Previous studies have shown that the stable $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ compositions of plant and soil are mainly controlled by climatic variables and soil characteristics (Wu et al. 2018) and increased our understanding of C and N cycles at both regional and global scales. However, there are few studies on the role of climatic factors in

regulating the stable $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ compositions in temperate grassland ecosystems. Therefore, this study focuses on the $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ of plants and soils in grasslands of northern China. We aimed to explore the spatial patterns of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ in plant–soil system of grasslands in northern China and their driving factors. The results will provide scientific references for future research on the C and N biogeochemical cycles of grassland ecosystem.

2. Materials And Methods

2.1 Study area

This study was conducted in the Inner Mongolian Plateau along a precipitation gradient (41.06°–49.48°N, 114.26°–120.94° E) in northern China (Figure 1). The plateau is characterized by a dry and cold climate and belongs to the continental semi-arid grasslands of the Central Asian steppe ecosystem, with MAT ranging from -0.2 to 9 °C, and MAP ranging from 152 to 502 mm. Over 66% of the plateau is covered by temperate desert steppe (TDS), temperate typical steppe (TTS) and temperate meadow steppe (TMS). The three grassland types are distributed along the precipitation gradient across the plateau. Temperate meadow steppe is located at the wet end of the precipitation gradient, and the dominating species are *Stipa baicalensis*, *Filifolium sibiricum*, and *Carex pediformis* (Shen et al. 2016). Both plant species richness (25 species per square meter) and ANPP ($> 200 \text{ g m}^{-2} \text{ yr}^{-1}$) are the highest along the transect. By contrast, temperate desert steppe is distributed at the dry end of the precipitation gradient, and consists of *Cleistogenes squarrosa*, *Agropyron mongolicum* and *C. duriuscula*, which has the minimum ANPP ($< 60 \text{ g m}^{-2} \text{ yr}^{-1}$) and plant species richness (3 species per square meter). In the middle of the precipitation gradient, temperate typical steppe is dominated by *Stipa grandis*, *S. krylovii* and *Artemisia sacrorum*, which has a medium ANPP and plant species richness. The growing season of vegetation is mainly from May to September, during which the precipitation accounts for about 80% of the whole year. Based on the Chinese soil classification system, the soil types in temperate meadow steppe, temperate typical steppe and temperate desert steppe are Chernozem, Kastanozems and Calcisols, respectively.

2.2 Field sampling

We conducted three consecutive sampling campaigns during the summer (from June to August) of 2016–2018, and sampled 255 biomass plots and soil profiles from 85 sites covering all major grassland types in northern China. To avoid human disturbance, all sampling sites were located about 1 km away from the major roads. At each site, a 30×30 m quadrat was randomly selected. Soil samples were collected from three pits at a depth of 0–10, 10–20, 20–40, and 40–60 cm. Within each quadrat, aboveground parts of all plants in three 1×1 m plots were harvested. All the plant samples were rinsed with pure water (18.2 M Ω cm) to remove dust particles and then oven-dried for 60 h at 65 °C. Soil samples were naturally air-dried and then passed through 2 mm mesh sieve after removing big roots and rocks. Both the plant and soil samples were ground into a fine powder for determination of physicochemical properties and isotope values.

2.3 Chemical and isotope measurement

Soil pH was measured by immersing an electronic meter in a 1:2.5 mixture of the homogenized soil material and pure water, with a precision of ± 0.5 (Han et al., 2020). Soil samples passed through a 2 mm sieve were used to determine soil particle sizes by Mastersizer 3000 (Malvern Instruments, Malvern, England) and the error was less than 1%. Soil samples were finely ground ($< 149 \mu\text{m}$), treated with 0.5 mol L $^{-1}$ HCl solution at room temperature for 24 h and then washed to neutrality, dried and ground for determining soil organic carbon (SOC) content. The values of SOC, soil total nitrogen (STN) and the C and N contents of plant were measured by an elemental analyzer (Elementar analyser vario Max CN, Germany), with a precision of $\leq 0.01\%$. Quality assurance of methodologies was checked with a standard soil reference sample (GBW07405) and the precision was better than 5%. Actual SOC contents in the original soil samples should be calibrated because the removal of inorganic C reduces sample mass (Liu et al., 2021).

The weight of soil and plant samples used for isotope analysis depends on the C and N contents of the samples. The $^{13}\text{C}/^{12}\text{C}$ and $^{15}\text{N}/^{14}\text{N}$ ratios were determined by a stable isotope ratio mass spectrometer (MAT-253 plus, USA) coupled to an elemental analyzer (Li et al. 2021; Wu et al. 2019; Yang et al. 2015). The results were normalized based on the measured values of standards (Vienna Pee Dee Belemnite standard for $^{13}\text{C}/^{12}\text{C}$, atmospheric N_2 standard for $^{15}\text{N}/^{14}\text{N}$) and the $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values were calculated as follows:

$$\delta^{13}\text{C} (\text{‰}) = (R_{\text{sample}}/R_{\text{std}} - 1) \times 10^3 \quad (1)$$

$$\delta^{15}\text{N} (\text{‰}) = (R_{\text{sample}}/R_{\text{std}} - 1) \times 10^3 \quad (2)$$

where R_{sample} is the ratio of $^{13}\text{C}/^{12}\text{C}$ (or $^{15}\text{N}/^{14}\text{N}$) of the sample and R_{std} is the $^{13}\text{C}/^{12}\text{C}$ of Vienna Pee Dee Belemnite standard or the $^{15}\text{N}/^{14}\text{N}$ of atmospheric N_2 standard. The precision for the $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ measurements is 0.1‰ and 0.2‰, respectively.

2.4 Statistical analyses

One-way ANOVA analysis with least significant difference (LSD) test was performed to determine the significance of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values in different grassland types at the level of $p < 0.05$. The correlations between the $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values and different climate factors and soil properties were determined by linear regression analysis. A general linear model (GLM) was used to evaluate the combined effects of climate factors (MAT and MAP), soil properties (pH, EC, Clay, SOC, STN and the C/N ratio) and plant community composition on $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values in vegetation and surface soil (0–10 cm). Meanwhile, the structural equation model (SEM) was used to test direct and indirect effects of soil properties, climate factors and plant community composition on $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values in vegetation and surface soil. This statistical method can use a multiple regression approach to explain the interactions and nonlinearities.

3. Results

3.1 Soil physicochemical properties

The grasslands of northern China displayed significant differences in soil physicochemical properties (Table 2). Soil pH in temperate meadow steppe was lower than that in temperate typical steppe and temperate desert steppe, and there was no significant difference among various soil layers of each grassland type. However, soil EC in temperate meadow steppe was significantly higher than that in temperate typical steppe and temperate desert steppe, especially in the surface soils (0-10 cm). SOC and TN contents showed similar distribution trends and decreased with soil depth in each grassland type. At the same time, the values of SOC and TN in temperate meadow steppe showed a decreasing trend along the precipitation gradient in the grasslands of northern China, and were highest in the temperate meadow steppe (Table 2). Meanwhile, the C/N ratio showed the opposite trend and slightly increased with increasing precipitation, especially in surface soil. Besides, soil clay contents in temperate meadow were significantly higher than those in temperate typical steppe and temperate desert steppe.

Table 1
Plant C and N contents and their isotope composition in different grasslands.

Grassland type	C content	N content	C/N ratio	$\delta^{13}\text{C}$	$\delta^{15}\text{N}$
	(mg g^{-1})	(mg g^{-1})		(‰)	(‰)
TMS (n=9)	437.88±6.18 ^a	11.68±0.83 ^a	38.16±2.49 ^a	-27.55±0.49 ^a	3.81±1.32 ^a
TTS (n=56)	441.29±2.06 ^{ab}	20.88±0.84 ^b	23.10±1.05 ^b	-26.39±0.31 ^a	-0.78±0.34 ^b
TDS (n=20)	424.62±5.29 ^b	24.17±2.02 ^b	18.96±1.44 ^b	-23.52±0.96 ^b	1.22±0.85 ^a

Note: TMS, Temperate meadow steppe; TTS, Temperate typical steppe; TDS, Temperate desert steppe. Different letters indicate significant difference among the three grassland types (LSD tests, $p < 0.05$).

Table 2
Soil pH, EC, clay, SOC and TN contents and their isotope composition, and C/N ratio in different grasslands.

Type	Depth (cm)	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)	SOC (mg g^{-1})	TN (mg g^{-1})	C/N ratio	pH	EC (us/cm)	Clay (%)
TMS (n=9)	0-10	-25.60±0.66 ^{Aa}	5.28±0.47 ^{Aa}	24.63±3.31 ^{Aa}	2.66±0.39 ^{Aa}	9.43±0.54 ^{Aa}	6.80±0.41 ^{Aa}	195.63±55.89	13.39±1.71 ^{Aa}
	10-20	-25.63±0.56 ^{Aa}	5.94±0.45 ^{Aa}	24.35±6.77 ^{Aab}	2.13±0.43 ^{Aab}	10.98±0.96 ^{Aa}	6.98±0.41 ^{Aa}	140.47±48.10	11.22±1.39 ^{Aa}
	20-40	-25.19±0.44 ^{Aa}	6.47±0.55 ^{Aa}	11.78±1.75 ^{Abc}	1.24±0.20 ^{Abc}	9.74±0.44 ^{Aa}	7.30±0.46 ^{Aa}	146.11±55.31	15.23±3.07 ^{Aa}
	40-60	-25.04±0.39 ^{aa}	6.59±0.50 ^{Aa}	8.73±1.68 ^{Ac}	0.83±0.11 ^{Ac}	8.72±1.03 ^{Aa}	7.33±0.45 ^{Aa}	140.82±65.14	15.89±3.61 ^{Aa}
TTS (n=56)	0-10	-24.27±0.18 ^{Ba}	3.84±0.40 ^{Aa}	9.25±1.01 ^{Ba}	1.04±0.10 ^{Ba}	11.09±1.49 ^{Aa}	8.02±0.12 ^{Ba}	119.35±15.23	6.01±0.64 ^{Ba}
	10-20	-23.77±0.19 ^{Bb}	5.03±0.39 ^{Ab}	7.85±0.88 ^{Bab}	0.97±0.12 ^{Bab}	7.77±0.32 ^{Ab}	8.23±0.12 ^{Bab}	124.75±25.95	4.44±0.60 ^{Ba}
	20-40	-23.85±0.16 ^{Bab}	5.61±0.34 ^{Ab}	6.24±0.73 ^{Bbc}	0.79±0.10 ^{ABab}	7.63±0.54 ^{Ab}	8.34±0.11 ^{Bab}	105.84±8.09	4.56±0.73 ^{Ba}
	40-60	-23.92±0.15 ^{Bab}	5.95±0.29 ^{Ab}	5.07±0.66 ^{Bc}	0.65±0.09 ^{Ab}	7.92±0.63 ^{Ab}	8.37±0.11 ^{Bb}	151.02±42.37	4.48±0.71 ^{Ba}
TDS (n=20)	0-10	-22.60±0.28 ^{Ba}	4.56±0.80 ^{Aa}	3.99±0.56 ^{Ca}	0.55±0.15 ^{Ca}	15.47±4.47 ^{Aa}	8.27±0.16 ^{Ba}	119.68±29.13	5.22±1.71 ^{Ba}
	10-20	-22.45±0.31 ^{Ca}	5.68±0.79 ^{Aa}	3.66±0.46 ^{Cab}	0.62±0.09 ^{Ba}	8.95±2.17 ^{Aab}	8.18±0.16 ^{Ba}	88.51±10.36	3.38±0.61 ^{Ba}
	20-40	-22.15±0.31 ^{Ca}	5.64±0.63 ^{Aa}	3.32±0.44 ^{Cab}	0.56±0.08 ^{Ba}	8.41±1.42 ^{Aab}	8.30±0.15 ^{Ba}	108.95±19.08	3.81±0.70 ^{Ba}
	40-60	-22.58±0.26 ^{Ca}	5.98±0.83 ^{Aa}	2.69±0.29 ^{Cb}	0.53±0.05 ^{Aa}	6.04±0.65 ^{Aab}	8.35±0.15 ^{Ba}	105.28±15.19	3.67±0.63 ^{Ba}

Note: TMS, Temperate meadow steppe; TTS, Temperate typical steppe; TDS, Temperate desert steppe. Capital letters represent significant difference in the same layer between different grassland types based on LSD tests ($p < 0.05$). Lowercase letters represent significant difference in the same grassland type between different depths based on LSD tests ($p < 0.05$).

3.2 Changes of $\delta^{13}\text{C}$ in different grasslands

As shown in Table 1, mean plant $\delta^{13}\text{C}$ values increased with the decrease of precipitation and increased from -27.55‰ in temperate meadow steppe to -23.52‰ in temperate desert steppe. Different soil layers of each grassland showed significant difference in soil $\delta^{13}\text{C}$ values (Table 2). Soil $\delta^{13}\text{C}$ was lowest in temperate meadow steppe (from -25.60‰ to -25.04‰) (Table 2). Compared to temperate meadow steppe, soil $\delta^{13}\text{C}$ increased significantly by 5.2% and 11.7% in temperate typical steppe and temperate desert steppe in 0-10 cm layer, respectively. Climatic factors, soil physicochemical properties, and their interactions significantly affected soil $\delta^{13}\text{C}$ (Figure 2). The interactions between MAT and SOC on soil $\delta^{13}\text{C}$ were significant (Table 3).

Table 3
Summary of the results of the general linear models (GLMs) for temperate grassland of northern China, showing the effects of mean annual temperature (MAT), mean annual precipitation (MAP), soil pH, EC, SOC, TN, the soil C/N ratio, clay, plant C content and plant N content on plant and soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ (at 0-10 cm depth).

Variables	Plant $\delta^{13}\text{C}$			Soil $\delta^{13}\text{C}$			Plant $\delta^{15}\text{N}$			Soil $\delta^{15}\text{N}$		
	MS	SS(%)	<i>p</i> value	MS	SS(%)	<i>p</i> value	MS	SS(%)	<i>p</i> value	MS	SS(%)	<i>p</i> value
MAT	5.13	1.09	0.42	48.37	29.29	<0.01	76.68	17.33	<0.01	84.58	32.72	<0.01
MAP	18.73	3.99	0.13	4.17	2.52	0.12	113.68	25.70	<0.01	41.83	16.18	<0.01
pH	/	/	/	1.63	0.99	0.32	13.00	2.94	0.10	3.91	1.51	0.23
EC	8.19	1.74	0.31	/	/	/	1.89	0.43	0.52	0.55	0.21	0.65
SOC	/	/	/	15.88	9.62	<0.01	/	/	/	8.17	3.16	0.08
TN	6.97	1.49	0.35	1.68	1.02	0.32	/	/	/	5.89	2.28	0.14
C/N ratio	9.75	2.08	0.27	0.27	0.16	0.69	0.45	0.10	0.75	0.43	0.17	0.69
Clay	10.99	2.34	0.24	0.73	0.44	0.51	2.47	0.56	0.46	0.27	0.10	0.75
Plant C	10.50	2.24	0.25	/	/	/	19.44	4.40	0.04	/	/	/
Plant N	5.41	1.15	0.41	/	/	/	0.90	0.20	0.66	/	/	/

Note: Bold values are statistically significant ($p < 0.05$).

Abbreviations: MS: mean squares; SS: the proportion of the total variance explained by the variable.

3.3 Changes of $\delta^{15}\text{N}$ in different grasslands

There was significant difference in plant $\delta^{15}\text{N}$ among various grasslands in northern China. Plant $\delta^{15}\text{N}$ was highest in temperate meadow steppe (mean 3.81‰), followed by temperate desert (1.22‰) and temperate typical steppe (-0.78‰) (Table 1). For each grassland, soil $\delta^{15}\text{N}$ slightly increased with soil depth. In surface soil, soil $\delta^{15}\text{N}$ showed similar pattern trend with plant $\delta^{15}\text{N}$. Compared to temperate meadow steppe, soil $\delta^{15}\text{N}$ significantly decreased by 13.6% and 27.3% in temperate desert steppe and temperate typical steppe in 0-10 cm layer, respectively (Table 2). Climatic factors, soil texture, and their interactions significantly affected soil $\delta^{15}\text{N}$ (Figure 2). The interactions between MAT and MAP on soil $\delta^{15}\text{N}$ were significant (Table 3).

4. Discussion

4.1 Factors controlling plant and soil $\delta^{13}\text{C}$ in grasslands of northern China

Previous studies have revealed strong impacts of land+use change, especially grassland degradation, on the plant and soil $\delta^{13}\text{C}$ (Li et al. 2021; Yang et al. 2015). However, the patterns and controls of plant and soil $\delta^{13}\text{C}$ in grasslands of northern China are less well studied. In this study, the responses of plant $\delta^{13}\text{C}$ values to MAP (Table 1) are consistent with the previous studies under wet climates (Bai et al. 2012; Feng et al. 2020; Peri et al. 2012). These insensitive responses could be attributed to the absence of aridity for stomatal conductance during photosynthesis (Cooper 1988; Wang et al. 2013). The plant $\delta^{13}\text{C}$ could be affected by different photosynthetic pathways. C_4 plants have higher water use efficiency than C_3 plants and are commonly the most dominant species at sites with strong water stress (Feng et al. 2020). Plants with the C_3 photosynthetic pathway commonly have relatively lower $\delta^{13}\text{C}$ values than those that use the C_4 pathway (Dong et al. 2018; Li et al. 2021). Compared to temperate meadow steppe, more C_4 species (e.g., *Artemisia dracunculus* and *A. anethifolia*) appear in the temperate typical steppe and temperate desert steppe due to the low precipitation, and its relative abundance in the community will be higher and lead to the increase of plant $\delta^{13}\text{C}$ values in arid regions, especially in temperate desert steppe. Therefore, the declining trend of plant $\delta^{13}\text{C}$ values at community-level in our study area mainly results from the shift in the dominant plant functional group from C_3 to C_4 .

Plant $\delta^{13}\text{C}$ values are also different among various plant species (Zheng and Shangguan 2007). Previous studies indicated that the $\delta^{13}\text{C}$ values of forbs were significantly higher than those of sedges and herbages (Li et al. 2021; Zheng and Shangguan 2007). Some forbs species are competitive due to their higher water use efficiency under limited soil water availability and the decrease in stomatal conductance (Gebauer et al. 2002). Meanwhile, insufficient CO_2

supply due to the reduction in stomatal conductance leads to higher $\delta^{13}\text{C}$ values in forbs. Therefore, the decreased percentage of graminoids and sedges along with the decrease water availability from temperate meadow steppe to temperate desert steppe might be other causes for the increase plant $\delta^{13}\text{C}$ values in temperate desert steppe.

In natural ecosystems, soil $\delta^{13}\text{C}$ values are generally determined by plant litter $\delta^{13}\text{C}$ signature, which is further supported by the positive relationship between soil and plant community level $\delta^{13}\text{C}$ values in our study area (Figure 2). The variances for vegetation $\delta^{13}\text{C}$ and soil $\delta^{13}\text{C}$ were largely explained by the aboveground different plant community composition, suggesting that changes of the plant community composition are important to ecosystem C cycling among different grasslands in northern China. Besides, consistent with results of the Zoige Plateau, there was a distinct negative relationship between soil $\delta^{13}\text{C}$ and MAP in our study, indicating that soil $\delta^{13}\text{C}$ values in the study area are also controlled by soil moisture. At the same time, the soil $\delta^{13}\text{C}$ values tend to increase in the remained SOC, because microbes prefer the ^{12}C during SOC decomposition (An and Li 2015). Thus, the accumulation of old SOC with low turnover rates can result in a relatively high soil $\delta^{13}\text{C}$ value. The removal of surface soil layers due to the water and wind erosion in arid regions (e.g., temperate desert steppe) will make the remaining SOC enriched in soil $\delta^{13}\text{C}$ composition.

The GLM analysis showed that MAP and SOC explained 29.29% and 9.62% of the soil $\delta^{13}\text{C}$ variance, respectively (Table 3), indicating that MAP is an important factor controlling the soil $\delta^{13}\text{C}$ variation in grasslands of northern China. The SEM analysis revealed that MAT, plant $\delta^{13}\text{C}$ and the combination of SOC and STN had overall stronger effects on soil $\delta^{13}\text{C}$ values than the other environmental variables (Figure 3). In addition, MAP could also indirectly affect soil $\delta^{13}\text{C}$ values via influencing plant N contents.

4.2 Factors controlling plant and soil $\delta^{15}\text{N}$ in grasslands of northern China

Soil $\delta^{15}\text{N}$ composition typically ranges from -6‰ to 16‰, providing important information about N dynamics in different natural ecosystems, and (Kahmen and Buchmann 2008; Shan et al. 2019). In our study, soil $\delta^{15}\text{N}$ overall increased with depth, which was consistent with the previous studies (Dong et al. 2018). This phenomenon was mainly attributed to the biogeochemical processes such as mineralization, denitrification, plant uptake and microbial immobilization, etc., which usually preferred lighter isotope (^{14}N) and made ^{15}N enrichment with soil depth in N cycling (An and Li 2015; Li et al. 2021).

In this study, plant $\delta^{15}\text{N}$ values are negatively correlated with MAP (Figure 2), which is consistent with those in global ecosystems (Craine et al. 2010). At the same time, although the relationship between soil $\delta^{15}\text{N}$ and MAP was not significant in northern China, the soil $\delta^{15}\text{N}$ values showed a decreasing trend with increasing MAP. This trend is mainly attributed to a relatively more 'open' N biogeochemical cycle (that is, both N inputs and outputs are large relative to internal N cycling) in arid environments (Chen et al. 2021; Cheng et al. 2009; Yang et al. 2013). Besides, temperate desert steppe with low water availability would constrain N biogeochemical cycle processes such as denitrification and leaching, inhibiting the loss of N from the ecosystem in the form of denitrification products and leachates (Yang et al. 2013). Meanwhile, such losses in temperate meadow steppe with high water availability usually occur. Therefore, these various N cycle processes in different grasslands of northern China have systematic effects on soil $\delta^{15}\text{N}$ composition.

The GLM analysis showed that MAT and MAP explained 32.72% and 16.18% of the soil $\delta^{15}\text{N}$ variance, respectively (Table 3), indicating that climatic factors are the major factors controlling the soil $\delta^{15}\text{N}$ variation in grasslands of northern China. Indeed, the SEM analysis revealed that MAT and MAP, along with plant $\delta^{15}\text{N}$, had overall strongest effects on soil $\delta^{15}\text{N}$ values (Figure 3). In addition, MAT and MAP could also indirectly affect soil $\delta^{15}\text{N}$ values via its effects on plant $\delta^{15}\text{N}$ composition.

4.3 Implications for C and N biogeochemical cycles

Our results have three important implications for understanding the C and N biogeochemical cycles in temperate grasslands. Firstly, plant community composition influences C cycling via changes in the relative abundance of different functional groups (An and Li 2015; Dong et al. 2018). The appearance of more forbs and C_4 species in temperate desert steppe could increase plant and soil $\delta^{13}\text{C}$, plant N content and C/N ratio (Chen et al. 2021; Nel et al. 2018; Xia et al. 2021). All of these processes could influence on soil C and N turnover, via changing N mineralization rate and soil organic matter decomposition rate. Secondly, heavier C tends to accumulate in the remaining SOC, because microbes favor the ^{12}C during SOC decomposition (Bai et al. 2012; Cooper 1988; Golluscio et al. 2009). Thus, SOC with a slow turnover rate will result in a relatively high soil $\delta^{13}\text{C}$ (Bird et al. 1996). In addition, the water and wind erosion of surface soils, especially in arid regions (e.g., temperate desert steppe), will make the remaining SOC enriched in soil $\delta^{13}\text{C}$ (Chen et al. 2021; Feng et al. 2020), and the soil $\delta^{13}\text{C}$ in our study area is mainly controlled by MAP. Thirdly, a relatively more 'open' N biogeochemical cycle (that is, both N inputs and outputs are large relative to internal N cycling) in arid environments (Chen et al. 2021; Cheng et al. 2009; Yang et al. 2013), and the plant and soil $\delta^{15}\text{N}$ in grasslands of northern China were mainly controlled by MAT and MAP. Taken together, the C and N isotope patterns in our study area provide insights into the effects of plant community composition and climate on C and N biogeochemical cycles in the grasslands of northern China.

5. Conclusions

Grasslands in northern China play a crucial role in the terrestrial biogeochemical cycles of C and N. In this study, different grassland ecosystems have significant different patterns of plant and soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values. Plant community composition, especially the increase in percentage biomass of forbs contributes most to the change in plant and soil $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values. The SOC with a low turnover rate in combination with strong soil erosion by water and wind in arid regions (e.g., temperate desert steppe) could result in relatively high soil $\delta^{13}\text{C}$ values. Soil stable $\delta^{15}\text{N}$ composition in our study area is mainly controlled by MAT and MAP, indicating that climatic factors (MAT and MAP) play important role in regulating the stable $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ compositions. The results will provide scientific references for future research on C and N biogeochemical cycles in temperate grasslands of northern China.

Declarations

Acknowledgments

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Figures

Figure 1

Distribution of study sites in temperate grassland of northern China.

Figure 2

Correlation between $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ in plant and soil (at 0-10 cm depth) and climate factors, plant and soil parameters.

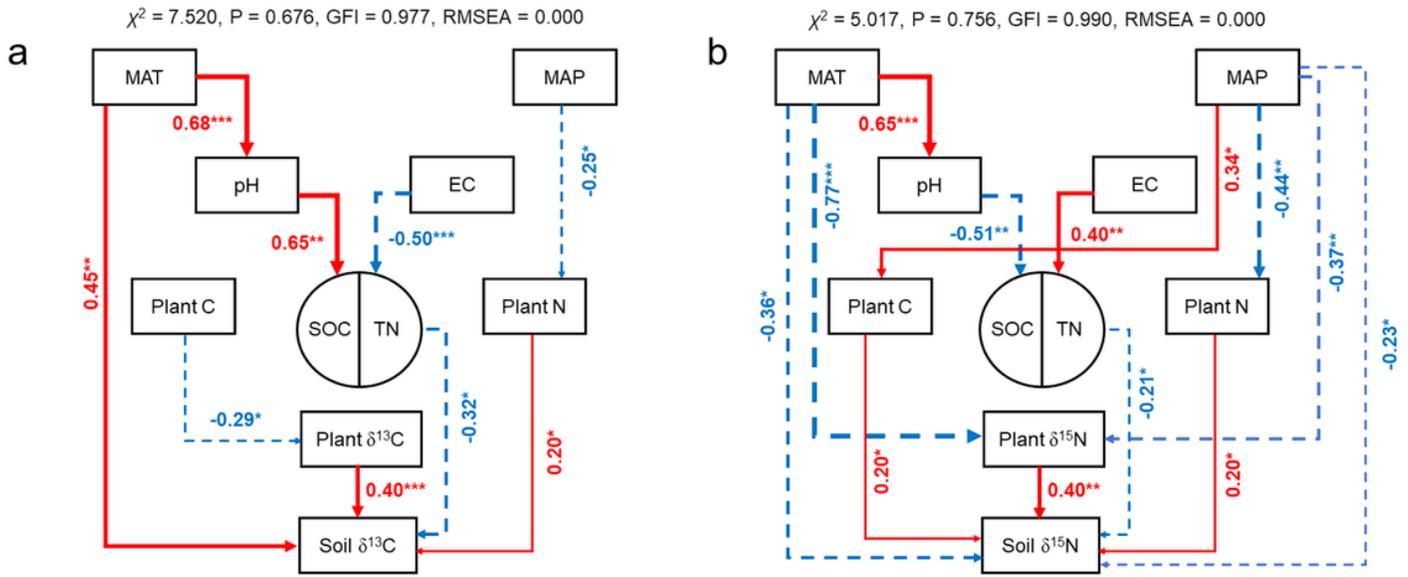


Figure 3

Structural equation model (SEM) of the effects of environmental variables on ecosystem $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values in temperate grasslands of northern China.