Shrub leaf stoichiometry and its driving factors in the grasslands of the Altay Mountains, Northwest China Postprint

Authors: MA Xuexi, JING Wei, LIANG Yuanye, ZHANG Wenbo, LIU Haoyu, YAN Meng, MAO Jiefei, FAN Lianlian, LI Yaoming, CHEN Xi, Georgy A LAZKOV, GAO Yingzhi, Gao Yingzhi

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Abstract

Grassland shrub invasion is a widespread phenomenon in global arid and semi-arid regions, affecting grassland ecosystems in multiple ways. Against the backdrop of intensifying climate change and human activities, studying the nutrient and stoichiometric characteristics of Spiraea shrubs in grassland ecosystems and their relationships with environmental factors can provide valuable insights into the nutrient utilization and survival strategies of these shrubs, which in turn provides a scientific basis for formulating future conservation measures. This study was conducted in July 2023 in the Altai Mountains of northwestern China, where Spiraea shrubs thrive in five grassland types: temperate steppe desert, temperate desert steppe, temperate grassland, temperate meadow steppe, and mountain meadow. Leaf and soil samples were collected from each grassland type to analyze the concentrations of carbon (C), nitrogen (N), and phosphorus (P), as well as the stoichiometric characteristics of leaves and soils. Subsequently, correlation analysis and redundancy analysis (RDA) were performed to investigate the variations in leaf C, N, P concentrations and leaf stoichiometry of Spiraea shrubs and their influencing factors. The results showed that there were significant or highly significant differences ($P<0.050$) in leaf C, N, P concentrations and leaf stoichiometry (C:N, C:P, and N:P ratios) of Spiraea shrubs among the five grassland types. The N:P ratio of Spiraea shrub leaves across the five grassland types ranged from 7.37 to 11.77, indicating that nitrogen availability generally limited the growth of Spiraea shrubs. RDA results showed that the factors with the greatest influence on leaf C, N, P concentrations and stoichiometric characteristics of Spiraea shrubs were, in order: soil total nitrogen > mean annual precipitation > altitude > soil pH > soil organic carbon > mean annual temperature. The contribution rates of these factors were 35.32%, 13.19%, 10.20%, 8.82%, 8.34%, and 6.48%, respectively. The study determined that, compared with climatic factors, soil nutrients had a greater impact on the growth and nutrient accumulation of Spiraea shrubs. This study provides an important theoretical foundation and data support for deeply understanding the response mechanisms of shrub species to climate change in the grassland ecosystems of the Altai Mountains.

Full Text

1 Introduction

Ecological stoichiometry is a scientific field that investigates the balance of energy and various chemical elements within ecosystems (Fang et al., 2025). It has been extensively utilized to examine factors limiting ecosystem productivity and the cycling of materials, particularly carbon (C), nitrogen (N), and phosphorus (P) (Shen et al., 2024). Employing ecological stoichiometry as a research tool to investigate the distribution, cycling, and nutrient limitation indicators of C, N, and P in ecosystems is crucial for unveiling the factors and mechanisms influencing ecosystem processes (Zeng and Chen, 2005). Leaves, as basic structural and functional units of plants, serve as the primary sites for energy absorption and nutrient utilization (Xu et al., 2021; Yang et al., 2025). Leaf stoichiometry can reflect the environmental adaptation capabilities of plants (Wang et al., 2022; Ma et al., 2024). The stoichiometric coupling of C, N, and P in plant leaves is closely linked to ecological functions such as plant growth, photosynthesis, and biogeochemical cycling (Yang et al., 2015; Wei et al., 2021). Specifically, the C:N and C:P ratios reflect the efficiency of nutrient use and the growth rate of plants (Chang et al., 2024), while the N:P ratio is widely utilized in identifying nutrient limitations in plants (Güsewell, 2004; Fang et al., 2025).

Foliar ecological stoichiometry is an essential diagnostic tool that serves as an indicator of plant nutrient cycling and ecosystem productivity (Li et al., 2019; Zhang et al., 2024). Numerous studies have determined that the stoichiometric characteristics of various plant functional groups are substantially influenced by several environmental factors, including soil, geography, climate, and water status (Chen et al., 2019; Tian et al., 2021; Meng et al., 2024; Pan et al., 2024). In fact, soil not only impacts plant growth and distribution (Bui and Henderson, 2013) but also plays a critical role in determining the ecological stoichiometry within plant bodies (Wang et al., 2019). Moreover, geographical factors such as elevation, latitude, and longitude markedly influence the ecological stoichiometry of plants. For instance, Tian et al. (2018) compiled an extensive dataset comprising 12,055 paired measurements of leaf N, P, and their stoichiometric ratios, encompassing global, regional, and site-specific records. Their findings indicated a statistically significant latitudinal gradient within these parameters, exhibiting a progressive decline from tropical to temperate and then to boreal regions. Mao et al. (2024) investigated the distribution of plants in alpine meadows across various elevation gradients; they observed that with rising elevation, plants in distinct functional zones implemented different strategies to adapt to local ecology by either increasing, decreasing, or maintaining their C, N, and P concentrations along with their stoichiometric ratios. Furthermore, climatic factors, particularly precipitation, have a direct impact on plant C, N, and P concentrations and their stoichiometry. In another study of different plant communities at a global scale, Tian et al. (2019) discovered that the N and P concentrations and N:P ratios in the leaves of approximately one-third to one-half of the plants decreased with increasing mean annual temperature (MAT) or mean annual precipitation (MAP). However, the leaf N and P concentrations and N:P ratios of at least one-third of plants did not conform to this global trend. Niu et al. (2020) explored how slope aspect significantly influenced the stoichiometric characteristics of plant leaves, with concentrations of C as well as C:P and N:P ratios being substantially higher on sunny slopes compared to shady slopes. Du et al. (2023) reported that on a regional scale, the stoichiometric characteristics of C, N, and P in plant leaves were collectively influenced by elevation, climate, and soil nutrients. Li et al. (2024) identified variations in the distribution of N and P concentrations among plant organs, specifically noting that their concentrations were greater in leaves compared to stems and roots, and these concentrations were thoroughly influenced by elevation, MAT, and MAP. In summation, the patterns and stoichiometric characteristics of C, N, and P across various plant species in different regions exhibit inconsistencies.

Grassland shrub encroachment is defined as the increase in shrub density, cover, and biomass within grassland communities, a phenomenon that is prevalent in arid and semi-arid regions worldwide (Gao and Liu, 2015). This encroachment impacts grassland ecosystems in several ways, influencing their productivity, genetic diversity, species diversity, and ecosystem diversity (Ding et al., 2023; Ma and Gao, 2025). The Altay Mountains, situated at the juncture of Russia, China, Mongolia, and Kazakhstan (Ni, 2004), are one of Central Asia’ s major mountain ranges, characterized by significant climatic variations in recent years (Chen et al., 2012). The Altay Mountains’ unique climatic features render vegetation productivity particularly vulnerable to climate change and human disturbances (Aili et al., 2022). Due to the frequency of extreme climate events and the impact of human activities, many grasslands in the Altay Mountains are gradually transforming into shrublands (Ding et al., 2023). Currently, shrub encroachment in the grasslands of the Altay Mountains is primarily characterized by the dominance of *Spiraea* and *Caragana* (Zhang et al., 2022b; Ma et al., 2025a).

At present, research on ecological stoichiometry within the Altay Mountains has predominantly focused on soils and wetlands, with a scarcity of studies regarding the leaf C, N, and P concentrations as well as leaf stoichiometry of *Spiraea* shrubs (Hao et al., 2021; Li et al., 2024). Therefore, this study focuses on the leaves of *Spiraea* shrubs from five distinct grassland types in the Altay Mountains of Northwest China, with the objective of analyzing (1) the relationships between leaf C, N, and P concentrations and leaf stoichiometry of *Spiraea* shrubs in relation to soil C, N, and P nutrients across different grassland types; and (2) the impact of climatic and soil factors on the stoichiometric characteristics of *Spiraea* shrub leaves. In light of the interplay between soil properties and plant leaf stoichiometry, this study proposes two hypotheses. First, significant variations exist in the concentrations of C, N, and P and stoichiometric characteristics of *Spiraea* shrub leaves across the selected grassland types. Second, soil nutrients play a more imperative role in influencing the leaf C, N, and P concentrations and leaf stoichiometry of *Spiraea* shrubs compared to climatic factors. This study aims to provide a theoretical basis for comprehending the nutrient supply and constraints faced by shrub plants in the Altay Mountains, while also serving as a reference for the conservation of grassland ecosystems against the backdrop of escalating climate change and human activities.

2.1 Study area

The study area (46°24 00 -49°10 12 N, 86°10 48 -91°04 48 E; Fig. 1 [Figure 1: see original paper]) is located in the Altay Mountains situated in the northern part of Xinjiang Uygur Autonomous Region, Northwest China. This northwest-southeast-trending mountain range stretches roughly 380 km from west to east and 290 km from north to south, with elevations between 1000 and 3500 m and a total area of approximately 1.08$×10^{4}$ km^2 (Zhang et al., 2022a). Located at the intersection of arid desert and semi-arid desert regions, this area is characterized by a continental climate, with warm yet short summers and cold, long winters. The region’ s annual mean temperature is approximately -2.0°C, with a maximum recorded temperature of 33.3°C (Xie et al., 2024). However, in the middle to high mountainous regions (1400–2600 m), the annual mean temperature drops below -9.0°C, and July, the warmest month, experiences an average temperature of only around 15.0°C. In the lower hilly areas, the annual mean temperature falls below 4.0°C. Additionally, there is a general trend of decreasing precipitation from both north to south and west to east. Due to its distinct topography and climate, the region is home to a variety of grassland types, which exhibit distinct vertical zonation. Ascending from the desert lowlands to the alpine summits, the grasslands form a distinct elevational sequence: lowland meadow, temperate steppe desert (TSD), temperate desert steppe (TDS), temperate steppe (TS), temperate meadow steppe (TMS), mountain meadow (MM), alpine meadow, and alpine steppe. Additionally, the main soil types in this region include brown calcic soil, chestnut calcic soil, and black calcic soil (Zhang et al., 2019a; Fan et al., 2023).

Fig. 1 Overview of the study area based on the elevation and locations of field sampling plots in the five grassland types of the Altay Mountains (a), as well as the photos showing the landscape of the five grassland types (b–f). (b), temperate steppe desert (TSD); (c), temperate desert steppe (TDS); (d), temperate steppe (TS); (e), temperate meadow steppe (TMS); (f), mountain meadows (MM).

2.2 Field sampling

Five grassland types were identified and selected in this study: TSD, TDS, TS, TMS, and MM (Fig. 1). These areas experience MAT ranging from -2.3°C to 5.3°C and MAP between 166 and 354 mm. *Spiraea* shrubs are prevalent across all these five grassland types (Ma et al., 2025b), where shrubs are predominantly distributed between 736 and 1855 m in elevation. Regarding the dominance of herbaceous plants, the TSD is characterized by the presence of *Seriphidium gracilescens* and *Festuca ovina*, whereas the TDS is dominated by *Seriphidium gracilescens*, *Artemisia frigida*, and *Festuca ovina*. Both the TS and MM are dominated by *Carex tristachya*, while *Carex tristachya* and *Festuca ovina* dominate the TMS.

In July 2023, field sampling was conducted across the five grassland types. For each grassland type, three 100 m$×100*mplotswereconstructed*, *resultinginatotalof*15*plots*(*Table*1).*Withineac*
m subplots were established along the diagonal and at the center to measure shrub characteristics and gather leaf and soil samples.

Specific procedures were implemented within each subplot. First, we quantified shrub coverage (%), crown width (cm), and plant height (cm) in the field, and then calculated patch size (m^2) from the crown-width measurements (Table 1). Second, we collected five healthy shrubs using an S-shaped transect sampling method, focusing on intact, sun-exposed leaves of the current year located at the mid-canopy height. The leaf samples were taken back to the laboratory, oven-blanched at 105.0°C for half an hour, and then dried at 65.0°C until a constant weight was achieved. Subsequently, the leaf samples were ground and sieved through a 0.2-mm sieve to identify the plant nutrients (C, N, and P). Third, we collected four soil cores (0–40 cm depth) around the stem base of one selected shrub at an equal distance in the four cardinal directions (east, south, west, and north) and pooled them into one composite sample. Each plot contained three replicate composite samples, yielding a total of 45 soil samples. These samples were then sieved through a 2.0-mm sieve to remove debris and stored in sterile bags. Additionally, a global positioning system (GPS) was used to record each sampling plot' s elevation and geographical coordinates (Table 1).

Table 1 Details of the sampling plots for the five grassland types

Grassland type	Longitude	Latitude	Elevation (m)	Coverage (%)	Patch size (m^2)	Plant height (cm)
TSD	87°43 25 E	47°51 16 N	30$±1.32	0.08±0.03	14.62±4.53	20.30±1.60

||*TDS*|86°21′52″*E*|48°12′56″*N*|0.2

Note: TSD, temperate steppe desert; TDS, temperate desert steppe; TS, temperate steppe; TMS, temperate meadow steppe; MM, mountain meadows. Data are expressed as mean±SE (standard error).

2.3 Determination of leaf and soil properties

The bulk density (BD) of the soil was determined in the field through the core method (Bretas et al., 2025). The proportions of clay (<2 m), silt (2-20 m), and sand (20-2000 m) in the soil were confirmed using a laser particle size analyzer (Mastersizer 2000, Malvern Panalytical Ltd., Malvern, UK). The soil water content (SWC) was measured using the oven-drying method (Almási et al., 2025), the soil pH was assessed using a pH meter, and the electrical conductivity (EC) was evaluated using an EC meter (SevenExcellence S470, Mettler Toledo, Greifensee, Switzerland). High-temperature combustion using an elemental analyzer (Enviro TOC cube, Elementar Analysensysteme GmbH, Langenfeld, Germany) was utilized to determine the soil organic carbon (SOC) and total C of the plant leaves. Additionally, the Kjeldahl method was used to determine the soil total nitrogen (STN) and total N of the plant leaves, whereas the $HClO_4$-H_2SO_4 digestion, followed by molybdenum-antimony anti spectrophotometry, was used to measure the soil total phosphorus (STP) and total P of the plant leaves. Furthermore, the soil available nitrogen (SAN) was appraised by the alkaline hydrolysis diffusion method, and the soil available phosphorus (SAP) was determined by the $NaHCO_3$ extraction-molybdenum-antimony anti colorimetric method. These properties were all determined by referring to the methods outlined in Bao (2008).

2.4 Data analysis

The MAT and MAP data (2000-2020) were obtained from the TerraClimate dataset (https://www.climatologylab.org/terraclimate.html) to estimate the regional climatic patterns. An analysis of variance (ANOVA; Duncan' s multiple range test) was conducted using SPSS 11.5 (SPSS Inc., Chicago, IL, USA) to analyze the significance of variations in the C, N, and P concentrations and stoichiometric characteristics of *Spiraea* shrub leaves among the different grassland types. Origin 2018 (OriginLab, Northampton, MA, USA) was utilized to plot the leaf stoichiometric diagrams. Redundancy analysis (RDA) was conducted using CANOCO 5.0 (Microcomputer Power, Ithaca, New York, USA) to investigate the relationships between leaf and soil C, N, and P concentrations and their stoichiometric traits, as well as associations between soil properties and climatic factors.

3.1 Soil properties

The BD of the soil ranged from 1.05 to 1.43 g/cm^3, with considerable differences ($P<0.050$) noted among the five grassland types (Table 2). It is important to note that all grassland types exhibited a neutral to slightly acidic pH ($pH<7.00$), although the pH variations among them were not statistically significant ($P>0.050$). The MM type exhibited the highest SWC and SAN, while the TSD recorded the lowest of these values. Conversely, the trend for SAP was reversed, with the TSD having the highest concentration (12.57 g/kg) and the

TS demonstrating the lowest (4.77 g/kg). Based on the size measurements of the soil particles, two grassland types—TSD and TDS—were classified as sandy loam, while the remaining three types were categorized as silty loam.

Table 2 Soil physical and chemical properties of the five grassland types

Grassland type	BD (g/cm^3)	pH	SWC (%)	EC (S/cm)	SAN (mg/kg)	SAP (mg/kg)	Clay (%)	Silt (%)	Sand (%)
TSD	1.43$±0.06*a*	6.82±0.35*a*	2.10±0.72*c*	30.42±8.95*b*	61.99±18.56*b*	12.57±4.42*a*	3.96±2.05*ab*	41.70±20.1	

Note: BD, bulk density; SWC, soil water content; EC, electrical conductivity; SAN, soil available nitrogen; SAP, soil available phosphorus. Different lowercase letters within the same column indicate significant differences of soil properties among the grassland types (P<0.050) based on Duncan' s multiple range test. Mean±SE.

As illustrated in Figure 2 [Figure 2: see original paper], the SOC, STN, ratio of SOC to STP (soil C:P ratio), and ratio of STN to STP (soil N:P ratio) exhibited significant variations (P<0.010) across the five grassland types, following the order of MM>TS>TMS>TDS>TSD. There were no major differences (P>0.050) in STP among the five grassland types, but the overall trend was TSD>MM>TDS>TMS>TS. Substantial differences (P<0.050) were noted in the ratio of SOC to STN (soil C:N ratio), with the order being TDS>MM>TS>TSD>TMS.

3.2 Leaf C, N, and P concentrations and stoichiometric characteristics

The average concentrations of C, N, and P in the leaves of *Spiraea* species across all the examined grassland types were 461.36, 16.66, and 1.93 g/kg, respectively (Fig. 3 [Figure 3: see original paper]). The C concentrations in the leaves exhibited variability among the five grassland types, with the hierarchy being TDS>TS>MM>TMS>TSD. With the exception of TDS and TSD, no significant differences in C concentrations were observed among the other three grassland types. The N concentrations of *Spiraea* shrub leaves across the five grassland types spanned from 16.13 to 18.81 g/kg, with the order being TSD>TDS>TMS>MM>TS; with the exception of TSD, no significant differences were noted among the remaining grassland types. The P concentrations of *Spiraea* shrub leaves varied from 1.41 to 2.38 g/kg across the five grassland types, with the order being TDS>TSD>MM>TS>TMS.

The C:N ratios of *Spiraea* shrub leaves across the five grassland types ranged from 24.07 to 30.57, with the ratio in TSD being significantly lower than those in the other grassland types (P<0.050), following the order TS>MM>TMS>TDS>TSD. The C:P ratios of *Spiraea* shrub leaves fluctuated between 194.86 and 365.19, with the ratio in TMS being significantly higher than those in the other grassland types (excluding TS; P<0.050), forming the order TMS>TS>MM>TDS>TSD. The N:P ratio of *Spiraea* shrub leaves in TMS was significantly higher than those in the other grassland types (excluding TS; P<0.050), following the order TMS>TS>MM>TSD>TDS.

3.3 Correlations of leaf C, N, and P concentrations and stoichiometric characteristics with environmental factors

The concentration of C in *Spiraea* shrub leaves did not exhibit significant correlations with soil characteristics, MAT, MAP, and elevation (P>0.050; Fig. 4 [Figure 4: see original paper]). In contrast, the N concentration of *Spiraea* shrub leaves exhibited negative correlations with SOC, STN, and STP, as well as with soil stoichiometric variables (excluding soil C:N ratio). However, it displayed positive correlations with soil BD, pH, SAP, and MAT. The P concentration of *Spiraea* shrub leaves demonstrated significant negative correlations with STN, soil C:P ratio, soil N:P ratio, SWC, and MAP, while it was significantly positively correlated with soil BD, SAP, and MAT. The C:N and C:P ratios of *Spiraea* shrub leaves were significantly correlated with the fundamental properties of the soil. Specifically, the C:N ratio of *Spiraea* shrub leaves exhibited considerable positive correlations with SOC, STN, STP, SWC, SAN, soil stoichiometric variables (excluding soil C:N ratio), and elevation, but it was significantly negatively correlated with soil BD, pH, and MAT. The C:P ratio of *Spiraea* shrub leaves was significantly positively correlated with STN, soil C:P ratio, soil N:P ratio, and MAP, and negatively correlated with BD, SAP, and MAT. Moreover, the N:P ratio of *Spiraea* shrub leaves was significantly positively correlated with MAP and negatively correlated with SAP.

3.4 Factors influencing leaf C, N, and P concentrations and stoichiometric characteristics

Through RDA, we delved into the environmental factors influencing leaf C, N, and P concentrations and stoichiometric characteristics, as shown in Figure 5 [Figure 5: see original paper]. The findings indicated that the explanation rates of axis 1 and axis 2 were 39.36% and 21.52%, respectively, resulting in a total explanation rate of 60.88% for the first two axes. The concentration of STN demonstrated the highest explanatory power for the variations in C, N, and P concentrations of *Spiraea* shrub leaves, as well as the leaf stoichiometry, accounting for 23.20%. This was followed by MAP with an explanatory power of 8.70% and elevation of 6.70%. In terms of each factor' s contribution, the effects of STN, elevation, pH, SOC, and MAT were extremely significant, indicating that these factors could effectively delineate the differences in the leaf C, N, and P concentrations and stoichiometric characteristics of *Spiraea* shrubs. The order of importance of the various environmental factors influencing the leaf C, N, and P concentrations and leaf stoichiometry of *Spiraea* shrubs was determined as follows: STN>MAP>elevation>pH>SOC>MAT>soil C:N ratio>SAP>soil N:P ratio>SWC>EC>STP>SAN>BD>soil C:P ratio (Table 3).

4 Discussion

4.1 Leaf C, N, and P concentrations and leaf stoichiometry of *Spiraea* shrubs across five grassland types

In this research, the average concentrations of leaf C, N, and P were found to be 461.36, 16.66, and 1.93 g/kg, respectively. Additionally, the average leaf C:N, C:P, and N:P ratios were 28.29, 258.70, and 9.24, respectively. Notably, both the leaf C:N and C:P ratios were lower than the global averages for terrestrial plants (35.90 and 805.00, respectively) (Elser et al., 2000).

Our research revealed significant variations in the concentrations of C, N, and P, along with their stoichiometric characteristics, in the leaves of *Spiraea* shrubs across five grassland types in the Altay Mountains. Specifically, the leaves of *Spiraea* shrubs in TDS exhibited the highest C concentration (467.39 g/kg), while the lowest C concentration was observed in the leaves of *Spiraea* shrubs in TSD (452.73 g/kg). However, the correlation analysis did not identify any environmental factors influencing the C concentration of *Spiraea* shrub leaves. Although existing research indicated that a warmer climate and increased precipitation could increase leaf C concentration through enhanced photosynthesis in the short term, several factors such as elevated respiratory consumption (Atkin et al., 2015), dilution effects from plant growth, and adjustments in C allocation (Wright et al., 2004; Poorter et al., 2012) substantially constrain this effect.

Moreover, our study found that the N concentration of *Spiraea* shrub leaves in TSD (18.81 g/kg) was significantly higher than those in the other grassland types, with no substantial differences noted among the latter. This phenomenon may be attributed to the significantly lower SWC in TSD compared to the other types of grasslands, aligning with the findings of Lu et al. (2023) in desert grasslands, where SWC emerged as a crucial factor impacting leaf N concentration in arid and semi-arid regions. In a similar vein, the investigation conducted by Zhang et al. (2019b) demonstrated that drought stress considerably decreases aboveground biomass (including stems and leaves) as plants allocate resources toward survival over growth. Limited water availability not only restricts photosynthesis and cell expansion but also leads to stunted plant development. Consequently, this may cause an increase in N concentration within the remaining plant tissues. It is important to note that the N concentration in *Spiraea* shrub leaves exhibited negative correlations with STN and SAN, which may be caused by the accelerated plant growth rate and significant increase in biomass accumulation when N supply in the soil was sufficient, resulting in the "dilution" of N concentration per unit mass of leaves. Additionally, the leaves of *Spiraea* shrubs in TDS had the highest P concentration (2.38 g/kg), while the leaves of *Spiraea* shrubs in TMS had the lowest P concentration (1.41 g/kg). This phenomenon may be attributed to the plants' ability to maintain a high level of internal chemical stability through the regulation of nutrient concentrations and their ratios, thus resisting the adverse effects of external environmental changes (Wu et al., 2024). According to the correlation analysis results shown in Figure

4, there was no relationship between leaf P and STP, but leaf P exhibited a significant positive correlation with SAP.

In plants, ecological stoichiometry is a discipline that investigates the balance of energy and chemical elements (primarily C, N, and P) within ecosystems (He and Han, 2010). The leaf C:N and C:P ratios in plant leaves not only represent the plant’ s ability to assimilate C during nutrient uptake but also reflect its nutrient utilization efficiency (Wang et al., 2024). In contrast, the leaf N:P ratio indicates the structure and function of the community as well as the nutritional limitations of the plant (Liang et al., 2023). Generally, an N:P ratio of the leaves greater than 16.00 indicates that P deficiency is a significant factor limiting plant growth. Similarly, an N:P ratio of the leaves between 14.00 and 16.00 suggests that both N and P affect plant growth, and an N:P ratio of the leaves less than 14.00 indicates that N deficiency is an important factor limiting plant growth (Reich and Oleksyn, 2004; Shi et al., 2017). The N:P ratio of *Spiraea* shrub leaves in the grasslands of the Altay Mountains (9.24) was significantly lower than the national average of 14.40 (Han et al., 2005) and the global average of 11.00 (Yin, 2024). Although the overall C assimilation capacity of *Spiraea* shrubs in the study area was comparable to the global average, their utilization of N and P was weak. Furthermore, the leaf N:P ratios across the five grassland types in this study ranged from 7.37 to 11.77, confirming that leaf N concentration is a crucial factor restraining plant growth (Wang et al., 2021).

In summary, considerable differences were observed in the C, N, and P concentrations and stoichiometric characteristics of *Spiraea* shrub leaves across the five grassland types, which aligns with the first scientific hypothesis proposed in this study. Nevertheless, compared to global terrestrial plants, the *Spiraea* shrubs examined in the arid study area exhibited a weak assimilation capacity for C but strong N and P utilization ability. Although the N concentration in the *Spiraea* shrub leaves within the study area (16.66 g/kg) was lower than the national average (18.60 g/kg), it was still a crucial factor limiting plant growth. The study area’ s arid environment may have enhanced the shrubs’ utilization of P, potentially explaining why the P concentration of *Spiraea* shrub leaves was higher than that of plant leaves from other regions. This suggests that plants adjust their C, N, and P concentrations in response to the environment during the growth process to efficiently improve water use and effectively cope with drought stress. However, through an ecological stoichiometric analysis of *Spiraea* shrub leaves, the current study discovered that the growth of *Spiraea* shrubs in the five grassland types of the Altay Mountains was generally constrained by N.

4.2 Influencing factors on the C, N, and P concentrations and stoichiometric characteristics of *Spiraea* shrub leaves

Soil serves as the primary source of nutrients aiding plant growth and development, and its nutrient characteristics play a crucial role in managing the N and P concentrations of plant leaves (Luo et al., 2022; Wu et al., 2023). The concentrations of C, N, and P in the soil, along with their stoichiometric characteristics, SWC, temperature, precipitation, and other environmental factors, significantly impact the C, N, and P concentrations and stoichiometric characteristics of plant leaves (Reich and Oleksyn, 2004; Zhang et al., 2012; Feng et al., 2024). Nevertheless, the aforementioned factors did not have a substantial impact on the C concentration of *Spiraea* shrub leaves within this study (Fig. 4). This investigation revealed that soil and climatic factors influence the element concentrations of *Spiraea* shrub leaves in the grasslands of the Altay Mountains, with leaf N concentration presenting positive correlations with soil properties such as pH and BD. According to Jia et al. (2024), who explored the impact of soil pH on 15 elements (including C, N, P, manganese (Mn), aluminum (Al), rubidium (Rb), etc.) in plant leaves, soil pH heavily influenced the enrichment coefficient of elements in plants that exceed 10.00% for 7 elements. Among these, the enrichment coefficients of N, C, and P were recorded at 100.00%, 39.22%, and 27.66%, respectively. This enrichment effect likely resulted in the positive correlation between soil pH and leaf N concentration. Min et al. (2014) pointed out that an increase in soil pH could lead to reduced enzyme activity, which may lower mineralization rates, thereby affecting the availability of C and N in the soil and indirectly altering the N concentrations in plant leaves (Fig. 4). The BD of the soil is a key physical parameter that characterizes soil porosity and compaction status. The positive correlation between soil BD and leaf P concentration may stem from the low permeability of the compact soil, the difficulty of N leaching during precipitation events, the relatively high N concentration in the surface soil layer, and the easier access to plant roots. The P concentration of *Spiraea* shrub leaves was primarily positively correlated with SAP, which is in alignment with the observations made by Liu et al. (2021) in cotton leaves. The C:N ratio of *Spiraea* shrub leaves exhibited negative correlations with soil BD and pH, while showcasing positive correlations with other soil properties including SOC, STN, STP, soil C:P ratio, soil N:P ratio, SWC, and SAN (Fig. 4). Additionally, a significant correlation was observed between the N:P ratio of *Spiraea* shrub leaves and SAP. Besides the factors listed in Figure 4, existing literature also confirms that leaf stoichiometric characteristics are influenced collectively by multiple environmental factors, with climatic and geographical factors exerting a greater influence than soil factors (Tao et al., 2021).

The current study found that the N and P concentrations of *Spiraea* shrub leaves were all positively correlated with MAT (Figs. 4 and 5). The observed increase in N levels with rising temperature (Fig. 5) could be attributed to metabolic processes and microbial activity being affected by the warmer conditions. This accelerates the decomposition and mineralization of organic matter in the soil and water, which in turn increases N availability. Conversely, low temperatures can inhibit N movement and plant N uptake (Xia et al., 2014). According to the temperature-plant physiology hypothesis proposed by Reich and Oleksyn (2004), the N and P concentrations of plant leaves decrease with the rise in average temperature. However, the pattern of leaf N and P concentrations in this study does not seem to confirm this hypothesis, possibly due to the MAT range in the study area (2.9°C–5.3°C) being comparatively narrower than the global scale (12.8°C–28.0°C). Further, MAP was positively correlated with the leaf N:P and C:P ratios but negatively correlated with the leaf P concentration (Figs. 4 and 5).

Overall, considering the various contributing factors to the C, N, and P concentrations and stoichiometric characteristics of *Spiraea* shrub leaves (Table 3), STN underwent the most significant impact, followed by MAP and elevation. Feng et al. (2024) highlighted that climatic factors and soil properties exert distinct influences on the spatial variations of plant C, N, and P stoichiometric characteristics across different ecosystems. Specifically, climatic factors, soil pH, SWC, and soil texture primarily govern the spatial variations of soil stoichiometry (C, N, and P stoichiometric characteristics) in regions experiencing pronounced climatic gradients. In contrast, in regions where climatic variations are less pronounced, soil factors and topography emerge as the main influencing factors. While this study effectively summarizes the effects of environmental factors (including soil properties, elevation, and climatic factors) on C, N, and P concentrations and stoichiometric characteristics of *Spiraea* shrub leaves, it is important to note that other environmental factors may also exert their influences, such as microbial biomass and abundance, soil aggregates, etc. Therefore, future research should ensure the incorporation of these additional environmental factors, such as soil microbial biomass, soil aggregates, soil texture, and grazing pressure, to more comprehensively investigate the structural and functional characteristics of *Spiraea* shrubs in the grasslands of the Altay Mountains.

5 Conclusions

This study investigated the differences in C, N, and P concentrations and stoichiometric characteristics of *Spiraea* shrub leaves across five grassland types in the Altay Mountains. It delved into the relationships of C, N, and P concentrations and stoichiometric characteristics of *Spiraea* shrub leaves with environmental factors (including soil properties, elevation, and climatic factors). The findings revealed that significant or highly significant differences ($P<0.050$) existed in the leaf C, N, and P concentrations and leaf stoichiometry (C:N, C:P, and N:P ratios) in *Spiraea* shrubs across the five grassland types. Positive correlations were observed between leaf N concentration and environmental factors such as soil pH, BD, MAT, and SAP. In contrast, leaf P concentration exhibited positive correlations with SAP, MAT, and soil BD. Given the substantial impact of climatic factors and soil properties on the C, N, and P concentrations and stoichiometric characteristics of *Spiraea* shrub leaves, the key factors were STN>MAP>elevation>soil pH>SOC>MAT. Notably, the comprehensive analysis of leaf stoichiometry and RDA unveiled that leaf N concentration was a limiting factor for shrub growth across the five grassland types in the Altay Mountains. This implies that *Spiraea* shrubs play a crucial role in constraining the availability of N and, consequently, restricting the growth of plants within the ecosystem. Therefore, future studies should integrate key ecosystem drivers to holistically assess the structural and functional dynamics of *Spiraea* shrub communities in the grasslands of the Altay Mountains.

Conflict of interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Author contributions

Conceptualization: MA Xuexi, JING Wei, LI Yaoming, GAO Yingzhi; Data curation: MA Xuexi, JING Wei, LIANG Yuanye; Formal analysis: MA Xuexi, ZHANG Wenbo, LIU Haoyu; Investigation: MA Xuexi, LIANG Yuanye, ZHANG Wenbo, LIU Haoyu, YAN Meng, MAO Jiefei, FAN Lianlian; Methodology: MA Xuexi, MAO Jiefei, FAN Lianlian, GAO Yingzhi; Resources: MAO Jiefei, CHEN Xi, Georgy A LAZKOV; Software: MA Xuexi, JING Wei, LIANG Yuanye, YAN Meng; Validation: MA Xuexi, JING Wei, ZHANG Wenbo, LIU Haoyu; Visualization: MA Xuexi, JING Wei, YAN Meng; Writing - original draft: MA Xuexi, JING Wei; Writing - review and editing: MA Xuexi, JING Wei, LI Yaoming, CHEN Xi, Georgy A LAZKOV, GAO Yingzhi. All authors approved the manuscript.

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