Effects of grazing on CO₂, CH₄, and N₂O fluxes in three temperate steppe ecosystems

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Abstract. Terrestrial ecosystems play a critical role in regulating the emission and uptake of the most important greenhouse gases (GHGs) such as CO₂, CH₄, and N₂O. However, the effects of grazing on these GHG fluxes in different steppe types remain unclear. Here, we compared the effects of grazing on seasonal CO₂, CH₄, and N₂O fluxes in the meadow (MS), typical (TS), and desert (DS) temperate steppe ecosystems in northern China. CO₂ emission rates increased from 311.4 ± 73.2 to 349.6 ± 55.4 mg m⁻² h⁻¹ in MS, but decreased in TS (from 341.3 ± 93.0 to 239.5 ± 81.9 mg m⁻² h⁻¹) and DS (from 212.1 ± 53.7 to 163.0 ± 83.4 mg m⁻² h⁻¹) in response to summer grazing (SG). N₂O emission rates increased in MS from 4.7 ± 2.2 to 8.1 ± 3.4 l g m⁻² h⁻¹, but not significantly changed in TS (9.2 ± 4.2 vs. 8.4 ± 2.4 l g m⁻² h⁻¹) and DS (6.3 ± 1.5 vs. 5.7 ± 1.6 l g m⁻² h⁻¹) by SG. CH₄ uptake rates increased in MS from 33.0 ± 11.7 to 47.1 ± 10.4 l g m⁻² h⁻¹ and decreased from 64.4 ± 7.6 to 56.2 ± 5.9 l g m⁻² h⁻¹ in TS in response to SG. In MS and DS, N₂O emissions were positively related to seasonal CO₂ emissions and negatively related to CH₄ uptakes. No significant relationships were found between GHG fluxes in TS. Summer grazing did not affect the relationship between CO₂ and N₂O emissions in MS, but reduced the relationship by enhancing the effect of aboveground biomass (AGB) on N₂O emission in DS. The significant negative relationship between CH₄ uptake and N₂O emission in MS and DS could be attributed to the significant relationship between soil temperature (ST) and AGB in MS and to the significant effects of soil moisture on both CH₄ uptake and N₂O emission in DS. The decrease in the magnitude of the correlation coefficients between CH₄ uptake and N₂O emission by SG was due to the negative relationship between ST and AGB simultaneously in MS and DS. Our results suggest that effects of SG on GHG fluxes varied in different steppes and the relationship among GHGs was steppe-dependent and SG also changed the relationship by affecting GHG fluxes induced by varied soil and environmental factors.

Key words: grassland; grazing; greenhouse gases; relationship.

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INTRODUCTION

CO₂, CH₄, and N₂O are the three most important greenhouse gases (GHGs) contributing 64%, 17%, and 6%, respectively, to the total global warming potential of all GHGs (IPCC 2013). Terrestrial ecosystems play a key role in regulating the emission and uptake of GHGs (Han et al. 1999), and fluxes are biogeochemically coupled with the soil and the atmosphere (Han et al. 1999). CO₂ emissions from soil to atmosphere (i.e., soil respiration) are the sum of microbial respiration, root respiration, and bulk soil respiration (Xu et al. 2008a). N₂O fluxes from soils are derived from microbial-mediated nitrification and denitrification under aerobic and anaerobic
conditions, respectively (Holst et al. 2008, Zhong et al. 2014, Li et al. 2015). CH4 fluxes are the function of a balance between the production by methanogenic microbes and the consumption by methanotrophic microbes (Hou et al. 2012, Li et al. 2015). CO2, CH4, and N2O are the most biogeochmically mobile forms of C and N. Emission and uptake of these gases determine the turnover rate between terrestrial ecosystems and the atmosphere (Schlesinger and Bernhardt 2013). Understanding relationships between GHGs and the underlying mechanisms driving those relationships is very important in improving our understanding of global C and N cycling (Xu et al. 2008a, Zhang et al. 2014a, b).

Few studies have assessed the relationships between multiple GHG fluxes in terrestrial ecosystems (Dong et al. 2000, Xu et al. 2008a, Yao et al. 2010, Zhang et al. 2014a, b). Positive and linear correlations between CO2 and N2O fluxes have been found in temperate grasslands (Dong et al. 2000, Xu et al. 2008a, Yao et al. 2010, Zhang et al. 2014a, b), tropical forests (Garcia-Montiel et al. 2002, 2004), and croplands (Zhou et al. 2004). Negative linear relationships between CH4 and CO2 fluxes have been found in grasslands (Wu et al. 2010), and a trade-off relationship between N2O and CH4 fluxes was found in croplands (Zhou et al. 2004). However, there is no consensus on the relationships between multiple GHG fluxes and the mechanisms driving these relationships are largely unknown.

Previous studies indicate that soil characteristics such as texture, structure, water-holding capacity, pH, and organic C content strongly influence the metabolic pathways in the C and N cycles (Garcia-Montiel et al. 2004, Xu et al. 2008a, Yao et al. 2010). The magnitude, controls, and associations between GHG fluxes differ considerably among different grassland types (Frank et al. 2002, Garcia-Montiel et al. 2004). Grasslands account for approximately 25% of the earth’s land surface and have a significant effect on global GHG exchange (Batjes 1998) due to their capacity to produce, store, and cycle C and N substrates (Wang et al. 2005). Livestock grazing is the most important human practice in grasslands and has been shown to be an important factor regulating the emission and uptake of GHGs (Schönbach et al. 2012, Skiba et al. 2013, Tang et al. 2013, Wang et al. 2014). The biophysical and chemical controls of GHG fluxes in response to grazing vary across steppe types due to site-specific grazing history (Frank et al. 2002, Tang et al. 2013).

To assess the effects of grazing on GHG fluxes in meadow (MS), typical (TS), and desert steppes (DS) in Inner Mongolia, China, a three-year field study measuring fluxes (CH4, CO2, and N2O) as well as soil, vegetation, and environmental factors was carried out. We hypothesize that (1) grazing effects changed with different GHGs for special mechanisms of production and consumption, (2) relationships among multiple GHGs fluxes are steppe-dependent for different soil characteristics and environmental conditions, and (3) summer grazing (SG) affects the relationships by changing the soil and environmental factors.

**Materials and Methods**

**Study site**

In 2011, along east–west transect in the Inner Mongolia grasslands, MS, TS, and DS were assigned, respectively. The growing season starts in early May and ends in late September. The mean annual temperature and precipitation (1971–2000) are 2.1°C and 395 mm for the MS site (120.3° N, 45.1° E, 656 m asl), –0.3°C and 293 mm for the TS site (116.7° N, 43.6° E; 1268 m asl), and 3.1°C and 175 mm for the DS site (111.9° N, 41.8° E; 1428 m asl). The soils are Typical Kastanozem, Calcic Chernozem, and light-colored Chernozem for the MS, TS, and DS, respectively (The Food and Agriculture Organization of the United Nations soil classification). The major plant species are Leymus chinensis, Festuca sibiricum, and Carex spp. in the MS; Stipa grandis, Agropyron michnii, Cleistogenes squarrosa, and other bunchgrasses in the TS; Stipa breviflora, Artemisia frigida, Thymus serpyllum, and Caragana shrubs in the DS (Hou et al. 2016).

**Experimental design and field measurements**

Randomly selected paired plots of SG and ungrazed (NG) areas of 100 × 100 m² were established at each of the three sites. Summer grazing plots had strictly restricted stocking rates and time (0.5 sheep unit per hm² at growth season). The NG areas were enclosed to prohibit grazing. Three flux chamber bases (length × width × height = 0.5 × 0.5 × 0.1 m) were installed randomly
10 cm into soil 10–15 d prior to measurements in each plot. Canopy biomass in the frame of bases was clipped to ground level one day before gas sampling. Gas samples were collected in the morning (9:00–10:00) and the chamber bases were moved to neighboring locations within each plot after gas sampling was completed. The sampling frequency was once every 10–15 d during the growing season of each year (from May to October 2011–2013).

An in situ static chamber technique was used for measurements of GHG fluxes. Stainless steel chambers, 25 cm high and shaded with a reflective plastic sheet, were placed onto the bases sealed with water. Chamber headspace gas samples were collected at 0-, 10-, 20-, and 30-min time intervals using 100-mL polypropylene syringes. The gases were then injected into 100-mL sealed airbags. The airbags were transported to the laboratory within two days. Gas measurements were done with a Hewlett-Packard 5890 series II gas chromatograph fitted with an electron detector (for N$_2$O) and flame ionization detector (for CH$_4$ and CO$_2$). Certified CH$_4$, CO$_2$, and N$_2$O standards of 1.92, 348, and 0.338 µL/L, respectively, were used for calibration. In situ daily emission/uptake rates of GHGs were determined according to the concentration trend in chamber headspace and recorded as µg·m$^{-2}$·h$^{-1}$ for CH$_4$ and N$_2$O, and mg·m$^{-2}$·h$^{-1}$ for CO$_2$. Mean seasonal fluxes were calculated as the arithmetic mean value.

Soil temperature (ST) was measured concurrently with gas samples using a portable digital thermometer (902C; Shengtong Instrument Factory, Hebei, China). Soil moisture (SM) was also measured concurrently from soil samples ($n = 3$) collected just outside the bases using a stainless steel corer (3.5 cm in diameter) and oven-dried at 105°C for about 20 h to a constant weight. Aboveground biomass (AGB) was measured monthly by clipping canopy biomass to the ground level in three to six quadrats (1 × 1 m) at each plot each year. Corresponding belowground biomass (BGB) was also sampled in 2012 and 2013 using a stainless steel corer (7.0 cm in diameter) in each quadrat and put in root bags. After being rinsed, BGB and AGB were oven-dried at 70°C to constant weight (about 48 h).

A baseline survey of vegetation and soil characteristics was carried out in May 2012. Soils were sampled using a stainless steel corer (3.5 cm in diameter) with three replications for each plot (SG vs. NG) at each steppe site. The soil samples were sieved through 2-mm mesh and separated into two parts. One part was stored at 4°C for the measurement of NH$_4^+$, NO$_3^–$, and microbial biomass C and N (MBC and MBN). MBC and MBN were determined using the chloroform fumigation-extraction method (Liu et al. 2007). Soil NH$_4^+$ and NO$_3^–$ concentrations were measured using Continuous-Flowing Analyzer (AutoAnalyzer 3, SEAL Analytical, Norderstedt, Germany). The second part was air-dried for measurements of soil pH, texture, soil total C and N, and available phosphorus and potassium. Soil pH was measured using pH meter in soil water suspension (soil:water = 1:2.5). Soil texture was measured using laser particle size analyzer (Mastersizer 2000, Malvern Instruments Ltd., Worcestershire, UK). Air-dried soil subsamples were ground to fine powder (mesh number 100) with a mortar and pestle, and total C and total N were assayed using an elemental analyzer (vario EL III; Elementar, Hanau, Germany). Available phosphorus and available potassium were assayed using 0.5 mol/L sodium bicarbonate method and flame atomic absorption spectrophotometry. Soil bulk density was determined using the soils sampled by a stainless steel cylinder of 98.2 cm$^{-3}$ (5 cm in diameter, 5 cm in height).

**Statistical analyses**

One-way ANOVAs and Duncan’s multiple range tests were used to compare differences in soil physicochemical properties, environmental factors, and vegetation between no gazing and SG and steppe types. A $t$ test was used to examine SG effects on the mean seasonal flux of CH$_4$, CO$_2$, and N$_2$O in each year. The path analysis of structural equation model (SEM) was performed to analyze pathways that may explain environmental and vegetation effects on CH$_4$, CO$_2$, N$_2$O fluxes and their relations under NG and SG in each steppe. Amos version 17.0.2 (Amos Development Corporation, Chicago, Illinois, USA) was used to parameterize the model. SigmaPlot 10.0 (Systat Software Inc., San Jose, California, USA) was employed to create figures.

**RESULTS**

**Soil and vegetation characteristics**

The DS and TS soils were significantly sandier than the meadow (Table 1). Soil bulk density was...
Table 1. Soil properties and vegetation conditions in the three steppe types.

| Properties and conditions | Meadow steppe | Typical steppe | Desert steppe |
|---------------------------|--------------|----------------|--------------|
|                          | NG           | SG             | NG           | SG             |
| Clay (%)                  | 5.5 ± 0.2a   | ...            | 3.8 ± 0.8b   | ...            |
| Silt (%)                  | 57.9 ± 0.2a  | ...            | 46.2 ± 0.2b  | ...            |
| Sandy (%)                 | 36.6 ± 0.3b  | ...            | 50.0 ± 1.3a  | ...            |
| BD (g/cm³)                | 0.99 ± 0.03b | 1.12 ± 0.05ab  | 1.11 ± 0.06ab | 1.09 ± 0.02ab  |
| TC (%)                    | 2.26 ± 0.19a | 2.33 ± 0.19a   | 1.63 ± 0.15b | 1.46 ± 0.11b   |
| TN (%)                    | 0.23 ± 0.01a | 0.23 ± 0.02a   | 0.19 ± 0.02ab | 0.18 ± 0.01ab  |
| C/N                       | 9.83 ± 0.32a | 9.96 ± 0.04a   | 8.70 ± 0.06b | 8.33 ± 0.28b   |
| pH                        | 7.85 ± 0.21b | 6.95 ± 0.20c   | 6.96 ± 0.12c | 7.58 ± 0.15b   |
| NH₄⁺-N                   | 0.21b        | 0.20c          | 0.15b        | 0.15b          |
| NO₃⁻-N                   | 8.73 ± 0.06b | 11.47 ± 0.61a  | 9.03 ± 0.26b | 7.17 ± 0.23c   |
| AK (mg/kg)                | 413 ± 50a    | 224 ± 33b      | 282 ± 22b    | 251 ± 20b      |
| AP (mg/kg)                | 6.33 ± 2.67a | 3.43 ± 0.33a   | 3.56 ± 0.11a | 3.22 ± 0.47a   |
| MBC (mg/kg)               | 116 ± 9a     | 92 ± 1a        | 141 ± 17a    | 118 ± 9a       |
| MBN (mg/kg)               | 13.1 ± 1.3b  | 10.7 ± 0.8b    | 17.3 ± 1.2a  | 12.5 ± 1.7b    |
| ST (°C)                   | 22.25 ± 1.04a| 22.55 ± 0.73a  | 17.84 ± 1.59b| 18.25 ± 1.06b  |
| SM (g/g%)                 | 16.02 ± 1.02a| 12.97 ± 1.65b  | 7.18 ± 1.01c | 7.24 ± 0.55c   |
| AGB (g/m²)                | 313 ± 57a    | 196 ± 39c      | 265 ± 47b    | 149 ± 75d      |
| BGB (g/m²)                | 474 ± 91e    | 543 ± 66d      | 779 ± 68a    | 603 ± 54c      |

Notes: NG, no grazing; SG, summer grazing; BD, bulk density; TC, total carbon; TN, total nitrogen; C/N, total carbon/total nitrogen ratio; AP, available phosphorus; AK, available potassium; MBC, microbial biomass carbon; MBN, microbial biomass nitrogen; ST, soil temperature; SM, soil moisture; AGB, aboveground biomass; BGB, belowground biomass. Different lowercase letters in the same row indicate significant differences (P < 0.05) based on Duncan’s multiple range tests.

Greatest in the desert site and lowest in the meadow site. Grazing did not affect soil bulk density.

Soil pH was close to neutral at the TS and slightly alkaline in the meadow and desert sites (Table 1). Grazing significantly decreased soil pH in the MS and significantly increased soil pH in the TS (Table 1). Soil total C and N were significantly higher in the MS (Table 1). Significant differences in the soil C/N ratio were detected between sites, and values were highest in the MS and lowest in the DS (Table 1). Again, no significant grazing effects on C/N ratio were found. Soil available N was significantly lower in the DS than in the MS and the TS (Table 1). Grazing significantly decreased available N in the TS and DS, but significantly increased available N in the MS. Grazing significantly decreased available potassium in the MS and DS (Table 1).

The MS and TS showed comparable MBC and MBN, while the DS had significantly lower values (Table 1). Grazing only affected soil MBN in the TS (Table 1).

Aboveground biomass in the MS was significantly greater than in other two sites. Grazing significantly reduced AGB in all sites (Table 1). Summer grazing increased BGB in the MS and significantly decreased BGB in the other two sites (Table 1).

Soil temperature was significantly lower in the TS compared to the MS and DS due to its higher altitude (Table 1). Grazing had no significant effects on ST. The TS and DS had similar SM values, which were significantly (P < 0.05) lower than that in the MS (Table 1). Grazing significantly decreased mean SM in the MS, but did not affect the other sites (Table 1).

Effects of grazing on CH₄, CO₂, N₂O fluxes

Under NG, mean seasonal CH₄ uptake of the three years was greatest in the DS (75.8 ± 7.2 μg·m⁻²·h⁻¹) followed by the typical (64.4 ± 7.6 μg·m⁻²·h⁻¹) and finally the MS (47.1 ± 10.4 μg·m⁻²·h⁻¹; Fig. 1A–C). Summer grazing significantly decreased the mean seasonal CH₄ uptake rate by 43% in the MS, significantly increased it by 13% in the TS, and had little effects in the DS (Fig. 1A–C).

Under NG, mean seasonal CO₂ emissions of the three years in the MS (311.4 ± 73.2 mg·m⁻²·h⁻¹) and TS (341.3 ± 93.0 mg·m⁻²·h⁻¹) were significantly higher than in the DS (212.1 ± 53.7 mg·m⁻²·h⁻¹; Fig. 1D–F). Summer grazing
significantly enhanced the mean seasonal CO₂ emission rate by 12% in the MS and significantly reduced it by 30% in the TS and by 23% in DS (Fig. 1D–F).

Under NG, mean seasonal N₂O emissions in the TS (9.2 ± 4.2 µg·m⁻²·h⁻¹) were slightly higher than those in the MS (4.7 ± 2.2 µg·m⁻²·h⁻¹) and the DS (6.3 ± 1.5 µg·m⁻²·h⁻¹; Fig. 1H–J). Summer grazing significantly enhanced the mean seasonal N₂O emission rate by 79% in the MS, whereas no significant effects were detected in the TS and DS (Fig. 1H–J).

**Relations among CH₄, CO₂, and N₂O fluxes and their changes by grazing**

Under NG, the main driving factors of seasonal CH₄ uptake were ST, AGB, and SM in the MS, TS, and DS, respectively (Fig. 2A–C). Summer grazing changed the driving factor from AGB to ST in the TS (Fig. 2E). No change was
observed in the other two steppes (Fig. 2D, F). Soil temperature, SM, and AGB had significant effects on seasonal CO₂ emissions in the three steppes under NG and their total contribution to seasonal CO₂ emission decreased from the MS ($R^2 = 0.84$), to the TS ($R^2 = 0.77$), to the DS ($R^2 = 0.31$; Fig. 2A–C). Summer grazing also altered the driving environmental factors on N₂O flux in all three sites (Fig. 2D–F). Seasonal N₂O emission was significantly related to SM and AGB in the MS, ST and AGB in the TS, and SM in the DS (Fig. 2A–C). Summer grazing also altered the driving environmental factors on N₂O flux in all three sites (Fig. 2D–F).

A significant positive correlation under NG was found between seasonal CO₂ and N₂O emissions in the MS ($r = 0.59$, $P < 0.01$) and the DS ($r = 0.44$, $P < 0.01$), but not in the TS (Fig. 2A–C).
Summer grazing significantly decreased the magnitude of the correlation coefficients between seasonal CO$_2$ and N$_2$O emissions in the DS, not in the MS and TS (Fig. 2A–C). A significant negative relationship between seasonal CH$_4$ uptakes and N$_2$O emissions was found in the MS ($r = 0.32$, $P < 0.05$) and the DS ($r = 0.46$, $P < 0.01$); SG significantly reduced the magnitude of the correlation coefficients in the both steppes (Fig. 2A, C, D, F). In contrast, no significant relationship was detected between seasonal CH$_4$ uptake and N$_2$O emission in the TS, regardless of grazing (Fig. 2B, E). No significant relationship between seasonal CH$_4$ uptake and CO$_2$ emission was detected in all three steppes under NG and SG plots (Fig. 2).

**DISCUSSION**

**Effects of grazing on soil and vegetation traits**

Grazing significantly reduced soil water content in the MS likely as a result of the combined effects of enhanced soil surface evaporation induced by decreased vegetation cover and reduced infiltration caused by soil compaction (Wang et al. 2002, Zhong et al. 2014). Increases in soil pH in response to grazing at the typical and desert sites could be related to decreased litter accumulation and root biomass as well as urine and dung input (Table 1; Cui et al. 2005). In contrast, decreased soil pH value at the MS site could be caused by an increased root biomass and leguminous species leading to the formation of organic acids (Table 1; Brady and Weil 2002). Increased available soil N in the MS likely resulted from urine and dung inputs, an apparent increase in leguminous species, and significantly decreased SM which inhibits N absorption by plants (Frank and Evans 1997, Hamilton and Frank 2001). Decreases in soil available N in the TS and DS could be caused by grazing-enhanced N absorption by plants and decreased leguminous species. N absorption by plants has been shown to increase in response to herbivory (Xu et al. 2008b). In addition, N in urine and dung at these steppes is more prone to volatilization under a dry and hot climate (Zhang et al. 2013). Significant decreases in soil inorganic N under heavy livestock grazing are well documented at TS sites (Wu et al. 2011) and DS (Su et al. 2005), whereas significant increases have been reported at MS (Han et al. 2008). Decreases in available soil potassium by grazing in all three steppe types could be attributable to increased K output via canopy biomass consumption by livestock.

**Effects of grazing on CH$_4$, CO$_2$, and N$_2$O flux**

The mean seasonal CH$_4$ uptake rates (47.1–75.8 µg m$^{-2}$ h$^{-1}$) of the temperate steppe presented here fall within the range of 2–105 µg m$^{-2}$ h$^{-1}$ and keep in pace with a previous summary by Wang et al. (2014) who calculated a mean CH$_4$ uptake rate (standard deviation) of 59.62 ± 56.96 µg m$^{-2}$ h$^{-1}$ by obtained 465 sets from 50 articles in grasslands. For each steppe type in our study, the mean seasonal CH$_4$ uptake rate in the MS (47.1 µg m$^{-2}$ h$^{-1}$) was slightly higher than CH$_4$ mean value (28.45 ± 24.32 µg m$^{-2}$ h$^{-1}$) of 128 published data in meadow in China (Wang et al. 2014), while in the TS, it (64.4 µg m$^{-2}$ h$^{-1}$) was slightly lower than than mean value (71.45 ± 61.21 µg m$^{-2}$ h$^{-1}$) of published 337 data in China (Wang et al. 2014), and in the DS, it (75.8 µg m$^{-2}$ h$^{-1}$) was largely higher than that mean value (41.19 ± 26.01 µg m$^{-2}$ h$^{-1}$) of published 21 data in semiarid DS in China (Wang et al. 2014). The mean seasonal CH$_4$ uptake was greatest in the DS followed by typical and finally the meadow site, which is consistent with previous studies (Tang et al. 2013, Wang et al. 2014). This can be largely attributed to the differences in soil water content and organic matter and partly to sand content, topsoil NH$_4^+$, and pH value (Wu et al. 2010, Fang et al. 2014). Higher SM is favorable for CH$_4$ production in soil but not for CH$_4$ oxidation and diffusion of atmospheric CH$_4$ into the soil (Koschorreck and Conrad 1993). High availability of NH$_4^+$ can enhance ammonium-oxidizing bacteria at the expense of the CH$_4$-oxidizing bacteria (Brady and Weil 2002); thus, higher SM and NH$_4^+$ lead to lower CH$_4$ uptake in the MS (Table 1). Lower organic matter restricts CH$_4$ production and higher sand content is more conducive to CH$_4$ oxidation, which leads to a relatively higher magnitude of CH$_4$ uptake in the TS and DS soils (Wang and Han 2005).

The observed decrease in mean seasonal CH$_4$ uptake in the grazed MS resulted from decreased O$_2$ and CH$_4$ diffusion due to soil compaction by grazing and trampling. The significantly increased soil NH$_4^+$ concentration, and significantly lowered
pH value, suppressed CH$_4$ oxidation and uptake (Table 1; Wang and Han 2005, Fang et al. 2014). Significant increases in CH$_4$ uptake in the other two grazed steppes were more likely related to increased pH value and decreased soil NH$_4^+$ concentration that favored CH$_4$ oxidation and uptake (Table 1; Fang et al. 2014).

The mean seasonal CO$_2$ emission rate (163–349 mg$\cdot$m$^{-2}\cdot$h$^{-1}$) was similar to previous studies in Inner Mongolia grassland (Dong et al. 2005, Jia et al. 2007). For each steppe type, the mean seasonal fluxes of the three years in the MS (349 mg$\cdot$m$^{-2}\cdot$h$^{-1}$) were similar to previous studies (352.3 mg$\cdot$m$^{-2}\cdot$h$^{-1}$) by Dong et al. (2005). The mean seasonal CO$_2$ emission rate (341.3 mg$\cdot$m$^{-2}\cdot$h$^{-1}$) in the TS was greater than those found in previous studies in other adjacent experiment sites with range of 206.7–267.7 mg$\cdot$m$^{-2}\cdot$h$^{-1}$ (Li et al. 2000, Chen et al. 2003, Jia et al. 2007). In the DS, the mean seasonal CO$_2$ emission rate (163 mg$m^{-2}h^{-1}$) fall within the range of 150–253 mg$m^{-2}h^{-1}$ in the similar experiment sites in Inner Mongolia grassland (Zhang et al. 2014a, b).

Differences in mean seasonal CO$_2$ emissions between steppe types were largely attributable to the differences in soil organic matter, soil water content, and live root biomass (Table 1; Li et al. 2000, Zhang et al. 2003). Significantly reduced mean seasonal CO$_2$ emissions by SG in the TS and DS could be the result of significant decreases in litter accumulation and root biomass as previous studies have suggested (Wan and Luo 2003, Bahn et al. 2008, Chen et al. 2008). In contrast, the mechanism for grazing-enhanced CO$_2$ emission in the MS involves increased root biomass, N availability, and lowered pH values (Table 1).

The mean seasonal N$_2$O emission rates (4.7–9.2 $\mu$g$\cdot$m$^{-2}\cdot$h$^{-1}$) were in accordance with previous studies in Inner Mongolia grassland (Du and Chen 1997, Chen et al. 2000, Liu et al. 2015). In the MS, the mean seasonal value of N$_2$O flux was similar to previous studies by Lu et al. (2015). In the TS, previous studies indicated that the mean seasonal values in L. chinensis were 6.9 and 7.9 $\mu$g$\cdot$m$^{-2}\cdot$h$^{-1}$ (Du and Chen 1997, Chen et al. 2000), slightly lower than that in our studies (9.2 ± 4.2 $\mu$g$\cdot$m$^{-2}\cdot$h$^{-1}$).

The significantly higher mean seasonal N$_2$O emission in the TS coincided with much higher contents of MBC, MBN, and available ammonium and nitrate (Table 1). The significantly lower soil pH and the mean seasonal temperature in the TS also played an important role, as supported by the significant negative relationship between seasonal N$_2$O emission and ST observed (Table 1, Fig. 2B, E).

The significantly enhanced mean seasonal N$_2$O emissions with SG in the MS can be explained by the following three soil water status-dependent mechanisms (Schrama et al. 2013). First, due to the wetter soil at the meadow site, input of dung and urine by livestock may provide more substrates for nitrification and denitrification (Velthof and Oenema 1995, Oenema et al. 1997). Second, soil compaction by livestock can enhance N$_2$O emission due to decreased aeration of the soil (Clayton et al. 1997). Third, grazing significantly reduced SM leading to decreased N absorption by plants in favor of microbial denitrification (Brady and Weil 2002). This mechanism could also explain the lack of grazing effects on N$_2$O emissions in the TS and DS. As a rule, N in dung and urine is more prone to ammonia volatilization in hot and semiarid climates and compaction has minor effects on aeration in sandy soils. In addition, competitive N absorption between plants and microbes at these N-limited sites may have constrained N$_2$O-producing processes such as nitrification and/or denitrification (Brady and Weil 2002).

Our study indicated that GHG fluxes varied among steppe types, which were due to not only large differences in annual precipitation, annual temperature, but also differences in vegetation composition and soil type. Some previous studies also indicated that vegetation and soil type can play important roles in GHG fluxes for certain types of steppe (Du and Chen 1997, Wang et al. 1997, Chen et al. 2000, Dong et al. 2000). Vegetation and soil type can affect soil microbial composition, diffusion of O$_2$ and GHGs, and ST and SM (Du and Chen 1997, Dong et al. 2005). The effects of vegetation and soil type on GHGs will be considered in next studies.

**Correlations among GHG fluxes by steppe type and grazing**

A significant positive correlation was found between seasonal CO$_2$ and N$_2$O emissions in the MS and DS (Fig. 2A, C), which is previously reported trend in several MS communities.
(Dong et al. 2000, Holst et al. 2008, Yao et al. 2010). This can be explained by SM and AGB, which are driving factors for the fluxes of both gases (Fig. 2A, C; Yao et al. 2010). Mechanistically, CO₂ production derives from root respiration and microbial respiration via the aerobic microbial decomposition of the soil organic matter (Xu et al. 2008a, Zhang et al. 2014a, b). N₂O fluxes are produced directly by the anaerobic denitrification of nitrates derived from mineralization, whereas aerobic organisms decompose organic N (95–99% of total soil N) largely in protein form (Brady and Weil 2002, Zhang et al. 2014). N₂O is emitted from soils by microbial-mediated nitrification and denitrification under aerobic and anaerobic conditions, respectively (Zhang et al. 2014b, Li et al. 2015); ST had significant effects on microbial activities and metabolism processes (Dunfield et al. 1993). In addition, ST affects root activity and their ability to absorb nutrients, which provides nutrients to the shoot biomass. The increase in NH₄⁺ and NO₃⁻ contents by SG (Table 1), increase in N₂O emissions and decrease in CH₄ uptake (Fig. 1A, D), via inhibition of CH₄ oxidation by CH₄-consuming bacteria, and increase in N₂O production were observed (Zhang et al. 2014a, b). In the DS, SM was the main factor regulating relationship between CH₄ uptake and N₂O emissions and SG decreased the magnitude of the correlation coefficients by strengthening the effect of AGB on N₂O emissions in DS (Fig. 2C, F). In drought conditions, SM has a significant effect on microbial activities and nutrition movements related to CH₄ oxidation and N₂O production (Wang et al. 2014). Thus, the mechanisms that regulate the relationship between CH₄ uptake and N₂O emission were different among steppes and SG effects were also different.

A significant correlation between ST and AGB was the main reason for the significant negative relationship between CH₄ uptake and N₂O emissions. Summer grazing decreased the magnitude of the correlation coefficients by decreasing the relationship between ST and AGB. However, SG improved the effect of SM on N₂O emission in the MS (Fig. 2A, D). Mechanistically, CH₄ flux is the function of balance between the consumption by methanotrophic microbes and the production by methanogenic microbes (Hou et al. 2012, Li et al. 2015). N₂O is emitted from soils by microbial-mediated nitrification and denitrification under aerobic and anaerobic conditions, respectively (Zhang et al. 2014b, Li et al. 2015); ST had significant effects on microbial activities and metabolism processes (Dunfield et al. 1993). In addition, ST affects root activity and their ability to absorb nutrients, which provides nutrients to the shoot biomass. The increase in NH₄⁺ and NO₃⁻ contents by SG (Table 1), increase in N₂O emissions and decrease in CH₄ uptake (Fig. 1A, D), via inhibition of CH₄ oxidation by CH₄-consuming bacteria, and increase in N₂O production were observed (Zhang et al. 2014a, b). In the DS, SM was the main factor regulating relationship between CH₄ uptake and N₂O emissions and SG decreased the magnitude of the correlation coefficients by strengthening the effect of AGB on N₂O emissions in DS (Fig. 2C, F). In drought conditions, SM has a significant effect on microbial activities and nutrition movements related to CH₄ oxidation and N₂O production (Wang et al. 2014). Thus, the mechanisms that regulate the relationship between CH₄ uptake and N₂O emission were different among steppes and SG effects were also different.

The lack of significant seasonal correlations between CO₂ emission and CH₄ uptake in all steppes is consistent with previous study (Zhang et al. 2014b). The driving factors were different for both gas fluxes among steppes and grazing (Fig. 2). All ST, SM, and AGB had significant effects on CO₂ emission, but only ST had a significant effect on CH₄ uptake in the MS. Limited SM, a major controlling factor for the seasonal dynamics of soil respiration but not of CH₄ uptake, can explain this observation (Zhang et al. 2014b). Grazing had minor impacts on this correlation and the seasonal pattern of SM and relevant relations to CO₂ emission and/or CH₄ uptake.
CONCLUSIONS

The mean seasonal values of the three years for CO$_2$ emissions were 349.6 ± 55.4, 341.3 ± 93.0, and 212.1 ± 53.7 mg m$^{-2}$ h$^{-1}$; for CH$_4$ uptakes were 33.0 ± 11.7, 64.4 ± 7.6, and 75.8 ± 7.2 µg m$^{-2}$ h$^{-1}$; and for N$_2$O emissions were 4.7 ± 2.2, 9.2 ± 4.2, and 6.3 ± 1.5 µg m$^{-2}$ h$^{-1}$ in the MS, TS, and DS, respectively. Summer grazing decreased CH$_4$ uptake by 43%, increased CO$_2$ emission by 13%, and N$_2$O emission by 79% in the MS. In the TS, SG increased CH$_4$ uptake by 13% and decreased CO$_2$ emission by 30%. In the DS, SG decreased CO$_2$ emission by 23%.

We found a significant positive relationship between seasonal CO$_2$ and N$_2$O emissions, and a significant negative relationship between CH$_4$ uptake and N$_2$O emission in the MS and DS. No significant relationships were found between CH$_4$ uptake and CO$_2$ emission. Summer grazing did not change the relationship between CO$_2$ and N$_2$O emissions in the MS, but decreased the magnitude of the correlation coefficients in the DS. The negative relationship between CH$_4$ uptake and N$_2$O emission was due to the significant relationship between ST and AGB in the MS, and to the significant effects of SM on both GHG fluxes in the DS. Summer grazing decreased the magnitude of the correlation coefficients between CH$_4$ uptake and N$_2$O emission in the MS by decreasing the significant relationship between ST and AGB. Grazing enhanced SM effects on N$_2$O emissions and enhanced AGB effects on N$_2$O emission in the DS.

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