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Soil organic matter on arid saline-alkali land drives greenhouse gas emissions from artificial and natural grasslands in different directions

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Reducing greenhouse gas (GHG) emissions and mitigating the pace of global warming are crucial to achieving a balance between economic development and ecological protection. However, research on GHG emissions from different types of artificial grasslands is limited. This study aimed to elucidate the dynamics of GHG emission fluxes in three types of artificial grasslands and natural saline meadow grassland (NG). Meteorological and soil data were collected to determine the correlations between GHG fluxes and environmental variables. All grasslands were sources of CO₂ and N₂O. Artificial grasslands were sinks of CH₄, whereas NG was a CH₄ source. FCO₂ was the main source of GHGs in grasslands. The average emissions of FCO₂ and FN₂O in artificial grasslands were higher than those in NG. Soil temperature had a positive effect on GHG emissions in all grasslands and soil organic matter content was the main factor affecting all grassland GHG fluxes.

KEYWORDS

natural grassland, artificial grassland, greenhouse gases, carbon cycle, emission reduction and carbon sequestration, saline-alkali land

1 Introduction

Reducing greenhouse gas (GHG) emissions, protecting food security, and maintaining ecosystem stability are critical concerns for society (De Deyn et al., 2008; Ericksen et al., 2009; Istudor et al., 2019). The main greenhouse gas is carbon dioxide (CO₂); methane (CH₄) and nitrous oxide (N₂O) are also important gases that lead to temperature increase (Su et al., 2013; Licite and Lupikis, 2020). The global warming potentials (GWPs) of CH₄ and N₂O are 25 and 298 times those of CO₂, respectively (Yamulki et al., 2013). In recent years, the global surface warming trend has slowed, but long-term change remains ongoing (Tang et al., 2018). From 1961 to 2014, the annual average temperature in China increased by 0.28°C per decade, which is much higher than the global average level; however, the warming has slowed down in recent years (Jingzhi et al., 2016). Reducing GHG emissions and mitigating the pace of global warming are crucial to achieving a balance between economic development and ecological protection. This is significant for sustainable development strategies. To mitigate climate change and achieve carbon neutrality, it is essential to utilize the capacity of terrestrial ecosystems to sequester carbon and reduce emissions.

Grasslands are important components of terrestrial ecosystems, that play critical roles in the global carbon cycle. Approximately 26% of the Earth's land area is covered by grassland; the associated soil carbon content accounts for 34.0%–37.1% of the global carbon stock (Robin et al., 2000). China's grasslands account for 12.5% of the world's grasslands and cover 41.7% of the country's land area. The carbon content of grasslands constitutes 32.1% of China's total terrestrial ecosystem carbon stock (Tang et al., 2018). However, the average biomass carbon density of Chinese grasslands is significantly lower than the global average, indicating a substantial carbon sequestration potential (Zijing et al., 2023). With the long-term and rapid development of animal husbandry in China, grassland resources are being excessively consumed. Artificial grasslands have become the primary means for alleviating grassland pressure and addressing livestock feed shortages. In 2013, the area of artificial grasslands in China reached ~209,000 km². It has been expanding continuously. This expansion highlights the increasingly important role of artificial grasslands in carbon sequestration and emission reduction efforts (Su et al., 2013; Rigan et al., 2016).

Although artificial grasslands occupy an increasingly important position, current research on artificial grasslands mostly focuses on the production benefits or soil properties and less attention is paid to the ecological benefits of GHG emissions of different types of artificial grasslands. More attention is paid to the GHG emissions of different management modes of grasslands under human interference. The results of previous studies have showed that grazing reduces the CH₄ absorption and N₂O emissions of grasslands and increases CO₂ emissions by affecting the physical properties of soil moisture, temperature, and porosity (Wang et al., 2003; Wang et al., 2013; Wang et al., 2015). Chatskikh et al. reported that the N₂O emission coefficient of grazing grassland is higher than that of mowing grassland (Chatskikh et al., 2005). Guan et al. showed that the main factors affecting soil respiratory carbon emissions differ depending on the utilization mode, and the soil respiration rate of grassland under mowed utilization is higher than that of grazing. Some researchers showed that intensive management and fertilization will lead to higher GHG emissions from grassland (Leahy et al., 2004; Barneze et al., 2022). Considering the limited research on GHG emissions from different types of artificial grasslands, the effects of GHG emissions from different grassland types, including monoculture and mixed grasslands, were clarified in this study.

The main objectives of this study: 1) elucidation of the emission dynamics and greenhouse effects of different GHGs emitted by various grasslands; and 2) investigation of influencing factors and emission mechanisms of different GHGs in different grassland types. To this ends, GHG emissions from different vegetation types in artificial and natural grasslands were measured to understand the driving force of the greenhouse effect, which was helpful for human intervention to achieve the ecological benefits of carbon sequestration and emission reduction.

2 Materials and methods

2.1 Site description

This study was conducted at the Linze Grassland Agriculture Experimental Station at Lanzhou University (99°51'–100°30'E, 38°57'–39°42'N; Figure 1), which is situated in a continental desert

grassland characterized by an average annual temperature of 7.6°C, with a minimum temperature of –28°C, and a maximum temperature of 38°C. The region experiences relatively limited precipitation, mainly concentrated in late summer and early autumn seasons, accounting for over 60% of the total annual rainfall of 118.4 mm. Evaporation levels are high, with an average of 1,830.4 mm. The station is located at an elevation ranging from 1,380 m to 2,278 m, with an average elevation of 1,390 m. The average frost-free period spans 176 days. In 2016, the annual average temperature was 9.56°C with an annual precipitation of 184.64 mm. The average temperature during the growing season was 20.68°C with a precipitation of 144.51 mm. In 2017, the annual average temperature increased to 9.75°C with an annual precipitation of 201.74 mm. The average temperature during the growing season was 20.83°C with a precipitation of 149.69 mm. The soil in this area exhibits alkaline properties. The dominant grassland type is saline meadow grassland, primarily composed of halophytic species including *Phragmites communis*, *Agropyron cristatum* (L.) Gaertn, *Suaeda glauca* Bunge, *Kalidium foliatum* (Pall.) Moq.

2.2 Experimental design

The experiment was conducted in Natural Grassland (NG) and three artificial grassland types: Perennial monoculture Bromegrass (Bro), perennial monoculture Alfalfa (Alf), and perennial mixed Alfalfa*Bromegrass (A*B) (The planting ratio of Alfalfa to Bromegrass was 1:1). The artificial grasslands were seeded and applied with 75 kg hm⁻² urea and 225 kg hm⁻² diammonium phosphate as the base fertilizer only in 2015, and the planting area of each artificial grassland was 0.16 hm⁻². For each type of grassland, three plots (0.5 m × 0.5 m) with similar growth characteristics were selected as replicate.

2.3 Sampling and measurement

2.3.1 Greenhouse gas flux

Greenhouse gas flux samplings were carried out by static soil chambers (30 cm × 30 cm × 30 cm) every half month from May to September during the 2016–2017 growing season. The day before gas extraction, the above-ground plants were mowed to the ground, and the concave bottom frame of the box was smashed into the soil to form a good sealing environment. Collect gas from 9 am to 11 am the next day (Hou et al., 2016), and measure the soil temperature (ST) at 5 cm soil depth. The concentration of CH₄ and CO₂ was determined by LGR methane/carbon dioxide laser analyzer, and the concentration of N₂O was determined by LGR nitrous oxide analyzer.

Gas sampling occurred at intervals of 10 min, with measurements taken at four time points: 0, 10, 20, and 30 min after the static chamber was sealed. The greenhouse gas flux was calculated using the following formula (Chen et al., 2017):

$$F = \rho \frac{V}{A} \frac{P}{P_0} \frac{T_0}{T} \frac{dCt}{dt}$$

where F is the measured gas release flux (ug·m⁻²h⁻¹ or mg·m⁻²h⁻¹), V is the box volume, A is the bottom area of the box, Ct is the concentration of the measured gas mixture volume in the box at time t, t is the time required for sampling, ρ is the density of the measured gas in the

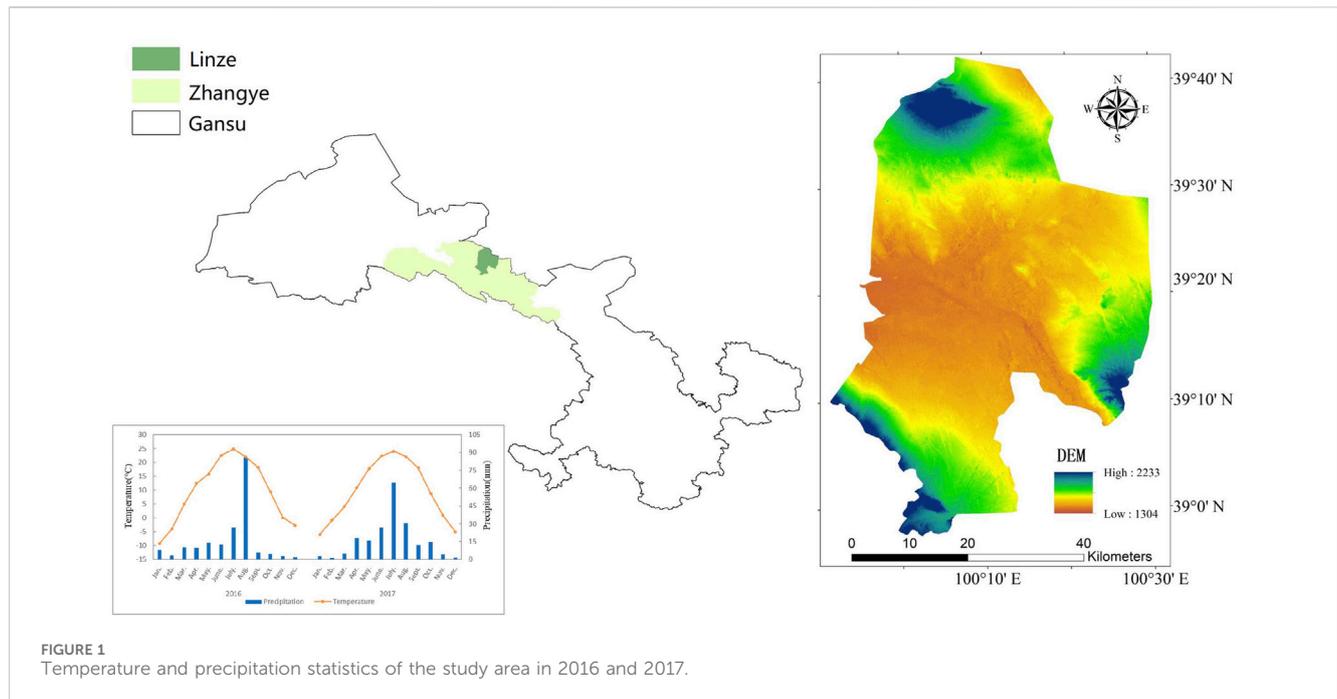


FIGURE 1 Temperature and precipitation statistics of the study area in 2016 and 2017.

TABLE 1 The initial values of soil physical and chemical indexes of different grasslands in growing season.

		PH	Salinity	TN (g/kg)	SOM (g/kg)	NH ₄ ⁺ (mg/kg)	NO ₃ ⁻ (mg/kg)
2016	NG	7.51 ± 0.03	10.31 ± 0.23	1.49 ± 0.06	16.18 ± 0.19	5.25 ± 0.74	14.32 ± 1.16
	Bro	7.22 ± 0.01	2.22 ± 0.05	1.28 ± 0.16	7.71 ± 0.20	2.78 ± 0.11	12.53 ± 0.78
	Alf	7.21 ± 0.03	2.37 ± 0.13	1.18 ± 0.29	7.48 ± 0.13	3.85 ± 0.78	11.77 ± 0.58
	A*B	7.24 ± 0.04	2.67 ± 0.03	1.62 ± 0.07	11.14 ± 0.18	1.24 ± 0.08	13.29 ± 0.25
2017	NG	7.61 ± 0.07	3.71 ± 0.40	1.39 ± 0.13	8.32 ± 0.10	7.39 ± 0.25	18.49 ± 0.55
	Bro	7.07 ± 0.06	2.10 ± 0.04	1.52 ± 0.07	8.67 ± 0.16	8.07 ± 0.41	20.24 ± 0.33
	Alf	7.24 ± 0.20	2.04 ± 0.01	1.45 ± 0.04	9.34 ± 0.28	6.29 ± 0.18	17.88 ± 0.73
	A*B	7.13 ± 0.01	2.17 ± 0.13	1.63 ± 0.28	9.22 ± 0.21	7.58 ± 0.21	19.71 ± 0.42

The number indicates the mean value ±SE.

standard state, T₀ and P₀ are the absolute temperature and pressure of the air under the standard condition respectively, P is the air pressure at the sampling site, T is the absolute temperature at the time of sampling.

Using GWP₍₁₀₀₎ to measure the overall situation of GHG emissions from different grasslands:

$$GWP = FCH_4 \times 25 + FCO_2 + FN_2O \times 298$$

2.3.2 Climate and soil

Climate data was obtained from the records of the local meteorological station, which included temperature (T) and precipitation (P) measurements.

Soil samples from 0 to 30 cm within the sample plots were collected after the first GHG measurement of each month and repeated three times for each grassland.

Soil water content (SWC) was determined using the drying method, soil organic matter (SOM) using the potassium dichromate

external heating method, total nitrogen content in soil (Michaud et al.) using the Kjeldahl method, nitrate (NO₃⁻) and ammonium (NH₄⁺) ions using UV spectrophotometry, pH using the 1:1 soil-to-water suspension method, and salinity using the soil solution electrical conductivity method. The initial soil properties were shown in Table 1.

2.3.3 Statistical procedures

The gas flux data was analyzed using SPSS for both the analysis of variance (ANOVA) to compare the differences in gas emission flux between different grasslands, as well as regression analysis to investigate the relationship between gas flux and environmental factors. Additionally, the differences in gas emissions between 2 years were compared. The regression analysis aimed to select the model with the highest fitting coefficient. The R package “lavaan” was utilized to explore the mechanisms of greenhouse gas emissions from different grasslands and to calculate the direct

TABLE 2 Regression fitting relationship between greenhouse gas flux and environmental factors.

		T	P15	ST	SWC	PH	Salinity	SOM	TN	NH ₄ ⁺	NO ₃ ⁻
FCH ₄	NG	\	0.11*	\	-0.28*	\	\	\	\	\	\
	Bro	\	-0.16*	\	-0.33**	\	\	\	\	-0.26*	\
	Alf	\	\	0.15*	-0.48***	\	\	\	\	\	\
	A*B	\	\	\	-0.37**	-0.31***	\	\	\	-0.25**	\
FCO ₂	NG	0.24**	\	0.30***	\	\	0.30**	-0.34***	\	\	0.19*
	Bro	0.27***	\	0.33***	\	\	\	0.59***	\	\	\
	Alf	\	-0.24***	0.52***	\	\	\	0.48***	\	\	\
	A*B	0.22**	-0.29***	0.44***	\	\	-0.41***	0.58***	0.62***	0.34**	-0.26**
FN ₂ O	NG	0.11*	\	0.14*	-0.25*	0.18*	\	\	\	-0.52***	\
	Bro	0.24**	\	\	\	\	\	0.15*	\	-0.52***	-0.17*
	Alf	\	\	\	0.33***	\	\	\	\	-0.30**	\
	A*B	0.21**	\	0.12*	-0.65***	-0.19*	0.37**	\	0.22*	0.46***	-0.25*

The numbers represent the fitting R², the underline represents exponential fitting, italics represent quadratic fitting, and the other numbers represent linear fitting. ‘\’ indicates that the factor fitting coefficient is less than 0. T, temperature; P₁₅, precipitation 15 days before measurement; ST, soil temperature; SWC, soil water content; SOM, soil organic matter; TN, soil total nitrogen. Significance levels: **p* < 0.05, ***p* < 0.01, ****p* < 0.001.

and indirect effects of various factors. The research findings revealed that grassland GHG emissions were influenced more significantly by soil temperature (ST) and soil water content (SWC) compared to air temperature (T) and precipitation (P15) (Table 2). When constructing the structural equation model, only ST and SWC were retained as substitutes for climate factor.

3 Results

3.1 Greenhouse gas fluxes

3.1.1 Response of FCH₄ to different grassland types

During the growing season, NG was the overall methane source, whereas artificial grassland acted as a methane sinks. With respect to methane emissions/uptake, except for the Bro and Alf monoculture grasslands in 2016, all grasslands showed distinct unimodal fluctuation patterns, with higher emission and uptake rates observed from July to August. NG demonstrated low emissions and uptake of CH₄ during the early and late growth stages while exhibiting higher emission rates during the peak growth period. Bro and Alf monoculture grasslands showed no significant fluctuations in FCH₄ in 2016 and had higher uptake peaks in 2017. The A*B mixed grassland showed high uptake peaks in both years. No significant difference between the 2 years was observed in any of the grasslands (*p* > 0.05; Figures 2A, B).

3.1.2 Response of FCO₂ to different grassland types

FCO₂ initially increased and then decreased as the growing season progressed. In 2016, the FCO₂ of the monoculture grasslands tended to be higher than that of NG and that of the mixed-sowing grassland was significantly higher than that of NG (*p* < 0.05; Figure 2D). No significant differences were observed among artificial grasslands. The time and duration of the emission peaks

were different for each grassland and the dynamic monthly emission rates were significantly different. In 2017, the monthly dynamics of grasslands were similar. Except for Alf, the other grasslands showed the highest FCO₂ at the end of June and a low FCO₂ at the end of July, which could be related to rainfall occurring on those days (Figure 2C). Although the emission dynamics between the grasslands differed over the 2 years, the overall emission rates were not significantly different (Figure 2D).

3.1.3 Response of F N₂O to different grassland types

FN₂O showed a noticeable downward trend from July to August 2016; Bro and A*B showed higher FN₂O (38.76 ± 9.33 μg m⁻²h⁻¹, 58.80 ± 23.63 μg m⁻²h⁻¹) from June to July in 2017, while the emission changes of other grasslands were less fluctuating. Except for the fact that NG FN₂O in 2016 was significantly lower than Alf, FN₂O in the whole growing season and 2 years of other grasslands were not significantly different (Figures 2E, F).

3.1.4 GWP

By converting the emissions of different GHGs to CO₂ equivalents, the GWP for each grassland can be calculated, with CO₂ being the primary contributor. N₂O and CH₄ contribute less. The CH₄ contribution rate of NG ranged from 0.1% to 0.4%, the N₂O contribution rate ranged from 0.6% to 1%, and the CO₂ contribution rate ranged from 98% to 99%. For artificial grasslands, methane is in an absorptive state with a contribution ranging from -0.5% to -0.2%. The contribution of carbon dioxide and nitrous oxide ranged from 97% to 99% and 1%–3%, respectively. The GWPs of NG and Bro in 2017 were higher than those in 2017, whereas those of Alf and A*B exhibited the opposite trend. Specifically, in 2016, the GWP ranking was AB > Alf > Bro > NG, whereas it was GWP ranking is Bro > A*B > NG > Alf in 2017 (Figure 3) 4.1.

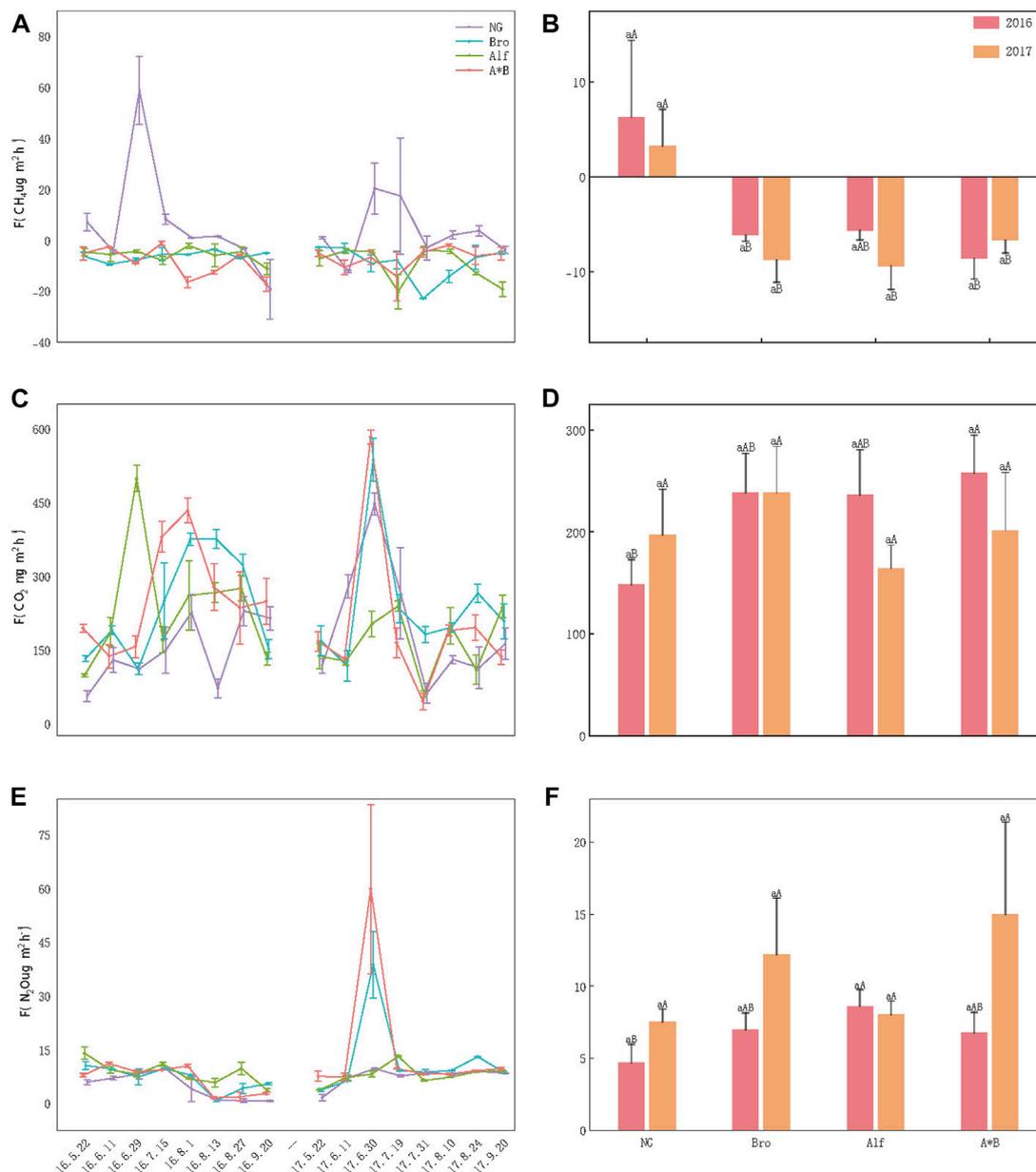


FIGURE 2 Dynamic and difference analysis of greenhouse gas emissions. **(A, C, E)** Monthly dynamics of greenhouse gases in different grasslands; **(B, D, F)** Annual dynamics of greenhouse gases in different grasslands. The error bar in the figure represents the average standard error (SE). Lowercase letters indicate the difference between the 2 years for grasslands and the uppercase letters indicate the difference between different grasslands in the same year. NG, Natural grassland; Bro, Bromegrass; Alf, Alfalfa; A*B, Alfalfa and Bromegrass mixed grassland. Significance levels $p < 0.05$.

3.2 Soil variables

No significant differences were observed between groups and years. However, in the case of NG, ST was higher in 2017 than in 2016. For artificial grasslands, ST decreased over 2 years (Figure 4A). In 2016, NG had a significantly higher SWC than Alf. In 2017, Alf had a significantly higher SWC than in 2016 (Figure 4B). In 2016, NG had a significantly higher soil pH than A*B. NG also had a significantly higher salinity than the other grasslands in both years.

However, in 2017, the salinity was significantly lower than 2016 for all grasslands (Figures 4C, D).

The SOM content of NG and A*B significantly differed in the 2 years (Figure 4E). The TN contents of the groups did not significantly differ. However, the TN content of Alf in 2017 was significantly higher than that in 2016 (Figure 4F). NG had the highest NH_4^+ concentration in both years, but no significant differences were observed between the grassland groups (Figure 4G). A significant difference in the NO_3^- content was

recorded between the groups. In 2017, the NO_3^- concentration was significantly higher than that in 2016 (Figure 4H).

3.3 Correlation between environment variables and GWP effects of different grassland types

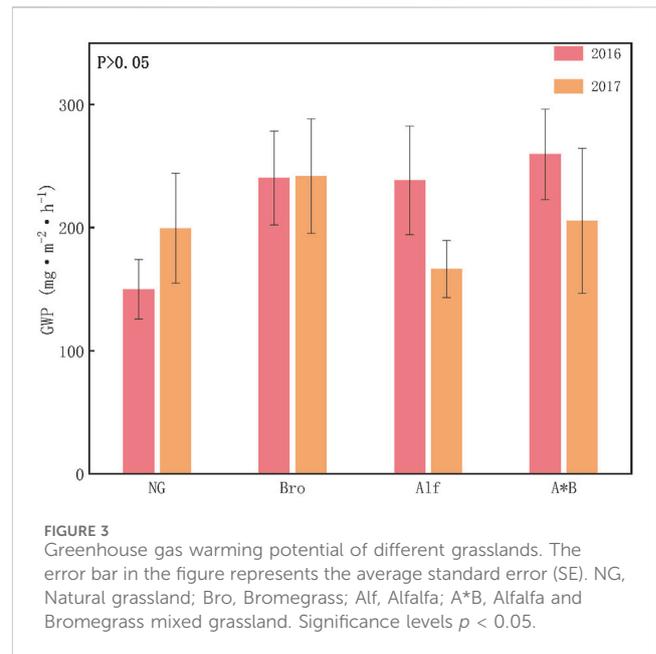
The SEM revealed that SOM played a crucial role as the primary driving factor of GWP in various grassland types. Nitrogen had a significant effect on GWP in different directions, except for the lack of substantial regulation of GWP_{Alf} (Figure 5). In particular, SOM had a negative effect on GWP_{NG} and a positive effect on artificial grasslands. Although SOM had a negative indirect effect on $\text{GWP}_{\text{A*B}}$ the effect was smaller than its positive effect. The soil pH had a negative effect on GWP across all grasslands (Figure 6). In addition, ST exerted a strong positive effect on GWP through both indirect and direct pathways (Figures 5, 6). Among them, NO_3^- had a stronger indirect positive effect on GWP_{NG} by negatively influencing SOM and the soil environmental factors accounted for 58.2% of the GWP_{NG} effect. NO_3^- had a higher indirect or direct negative effect on GWP_{Bro} , whereas NH_4^+ had a stronger direct positive effect. Soil factors collectively explained 76.2% of GWP_{Bro} . Salinity and pH negatively regulate soil nutrition, resulting in an indirect negative effect on GWP_{Alf} . SWC, NO_3^- , and NH_4^+ had direct or indirect effects and the salinity and pH together accounted for 75.8% of the GWP_{Alf} . TN, NO_3^- , and salinity had direct and indirect negative effects on $\text{GWP}_{\text{A*B}}$ and these soil factors collectively explained 80.4% of the $\text{GWP}_{\text{A*B}}$ effect (Figure 5; Figure 6).

4 Discussion

4.1 FCH_4 response to different grassland types

Our results indicate that NGs produce minor methane emissions and exhibit absorption during the early and late stages of growth while demonstrating relatively significant methane emissions during the mid-stage of the growing season. In contrast, artificial grasslands consistently serve as weak methane sinks throughout the growing season, with a peak in the methane absorption during the mid-stage (Figure 2). These results are consistent with those of previous research in this field (Chen et al., 2017; Wang et al., 2023).

Fluctuations in FCH_4 during the growing season in different grassland types are influenced by both grassland characteristics and climatic factors. Generally, anaerobic metabolism and CO_2 reduction by biological processes are the main sources of soil CH_4 (McEwing et al., 2015; McNicol and Silver, 2015; Michaud et al., 2017). In the early stages of the growing season, FCH_4 approaching zero can be attributed to lower soil temperatures and limited organic matter inputs that constrain soil metabolic activity (Table 1; Figure 2A) (Yavitt et al., 1997; Whalen, 2005; Song et al., 2011). As the growing season progresses with increasing temperatures and vegetation growth, the emission rates of CH_4 driven by carbon input tend to increase (Sutton-Grier and Megonigal, 2011; Cao et al., 2020). However, note that the concurrent diffusion of oxygen produced by photosynthesis into the rhizosphere before being fully consumed through respiration may inhibit both methane production and consumption (Frenzel and



Karofeld, 2000). The varying conditions of FCH_4 in different grasslands can be attributed to several underlying factors: (1) NG is a saline-alkali marsh and the dominant species is reed. Reeds have well-developed aeration tissues and the results of previous studies showed that their root tips play an important role in controlling the diffusion of CH_4 to their aerial parts. Leaf transport accounts for 45.34% of the total atmospheric emissions (Duan et al., 2009), facilitating gas exchange between the soil and atmosphere as well as diffusion of methane from soil and sediments to the atmosphere (Chapuis-Lardy et al., 2007; Ma et al., 2013; Bao et al., 2020). In addition, research has indicated that FCH_4 in flooded reeds is higher than that under non-flooded conditions, in line with the results of our study. We observed a significant positive correlation between NG and P_{15} (Table 2), indicating that the higher SWC in 2016 resulted in a higher FCH_4 (Figures 2B, 4B). (2) NG has a diverse vegetation species composition, whereas artificial grasslands have a relatively low diversity of soil microorganisms. This leads to increased competition for oxygen in NG soils, favoring methane production in anaerobic environments (Lalke-Porczyk and Donderski, 2005; Duan et al., 2009; Schultz and Pett, 2018). (3) Our study revealed that FCH_4 is primarily driven by the SWC, with an initial increase followed by a decrease as the moisture content increases (Table 2), which is consistent with previous research findings (Chen et al., 2017; Wang et al., 2020). Furthermore, on some artificial grassland, we observed that FCH_4 increased first and then decreased with the increase of P_{15} (Table 2). Precipitation during the growing season promotes microbial activity and methane production, but at the same time reduces soil aeration, and CH_4 may be oxidized and adsorbed during transport in water, inhibiting methane emissions (Luis Marin-Muniz et al., 2015; Cao et al., 2020). In addition, as the growing season progresses, increased carbon input and temperature stimulate the overall heterotrophic microbial respiration, leading to the consumption of competitive electron acceptors by non-methanogenic and methanogenic bacteria (Strom et al., 2003; Morin et al., 2014), resulting in greater methane absorption by artificial grasslands during periods of high precipitation (Figure 2A). This may explain the different

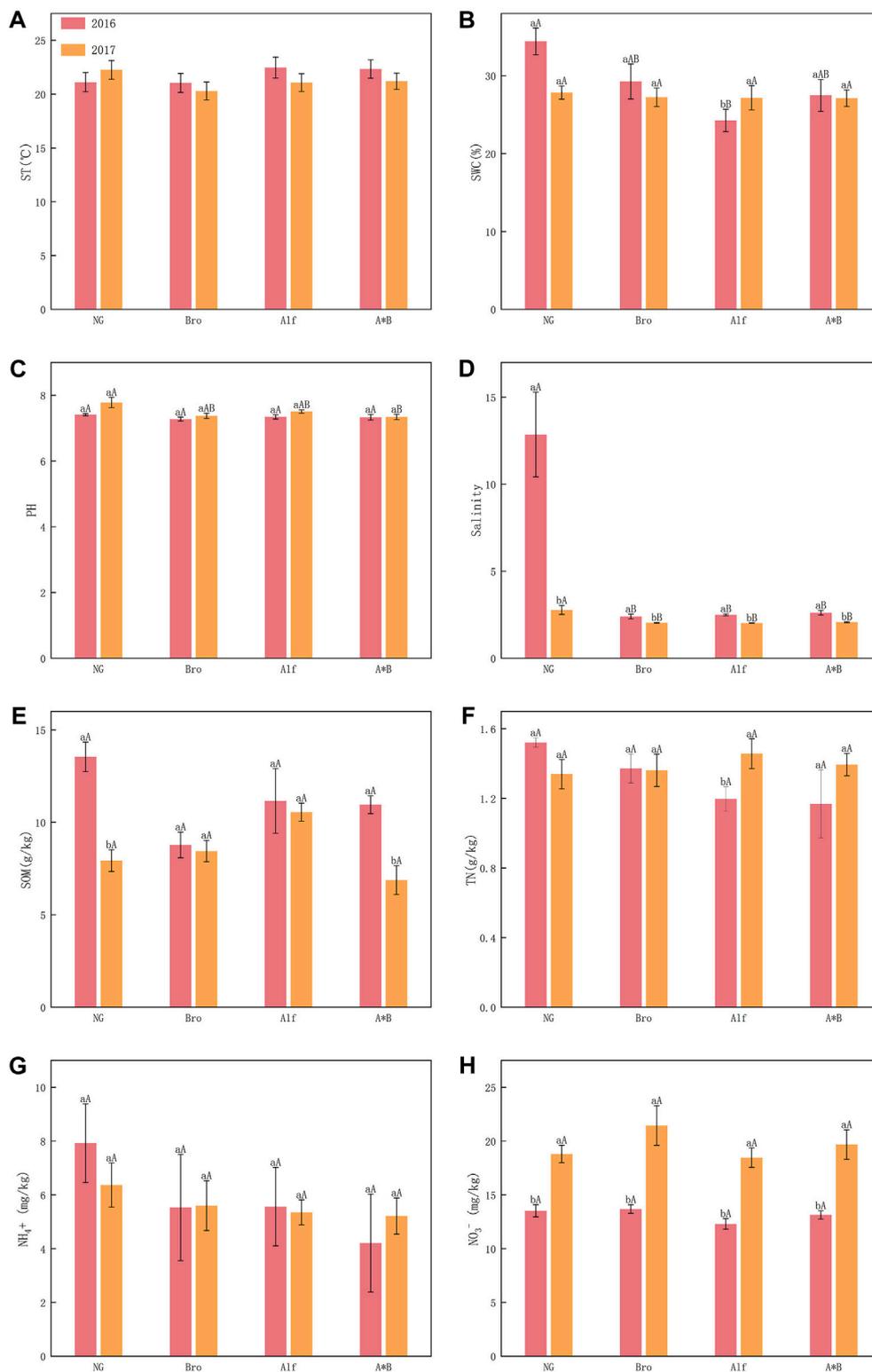
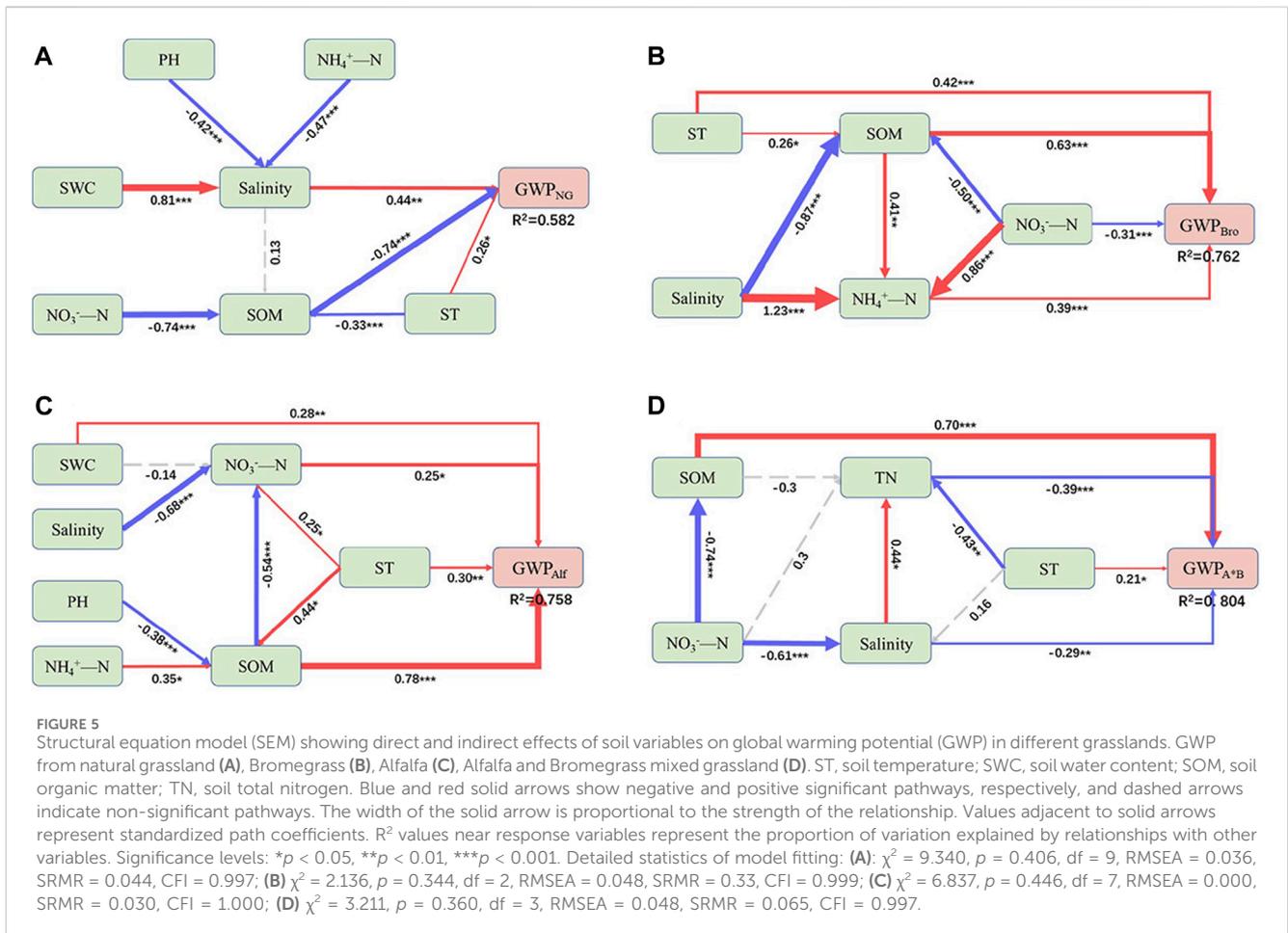


FIGURE 4 Interannual variation of soil indexes in different grasslands (A-H). The error bar in the figure represents the average standard error (SE). The lowercase letters indicate the difference between the 2 years of grassland, and the uppercase letters indicate the difference between different grasslands in the same year. NG, Natural grassland; Bro, Bromegrass; Alf, Alfalfa; A*B, Alfalfa and Bromegrass mixed grassland. ST, soil temperature; SWC, soil water content; SOM, Soil contains organic matter; TN, soil total nitrogen. Significance levels $p < 0.05$.



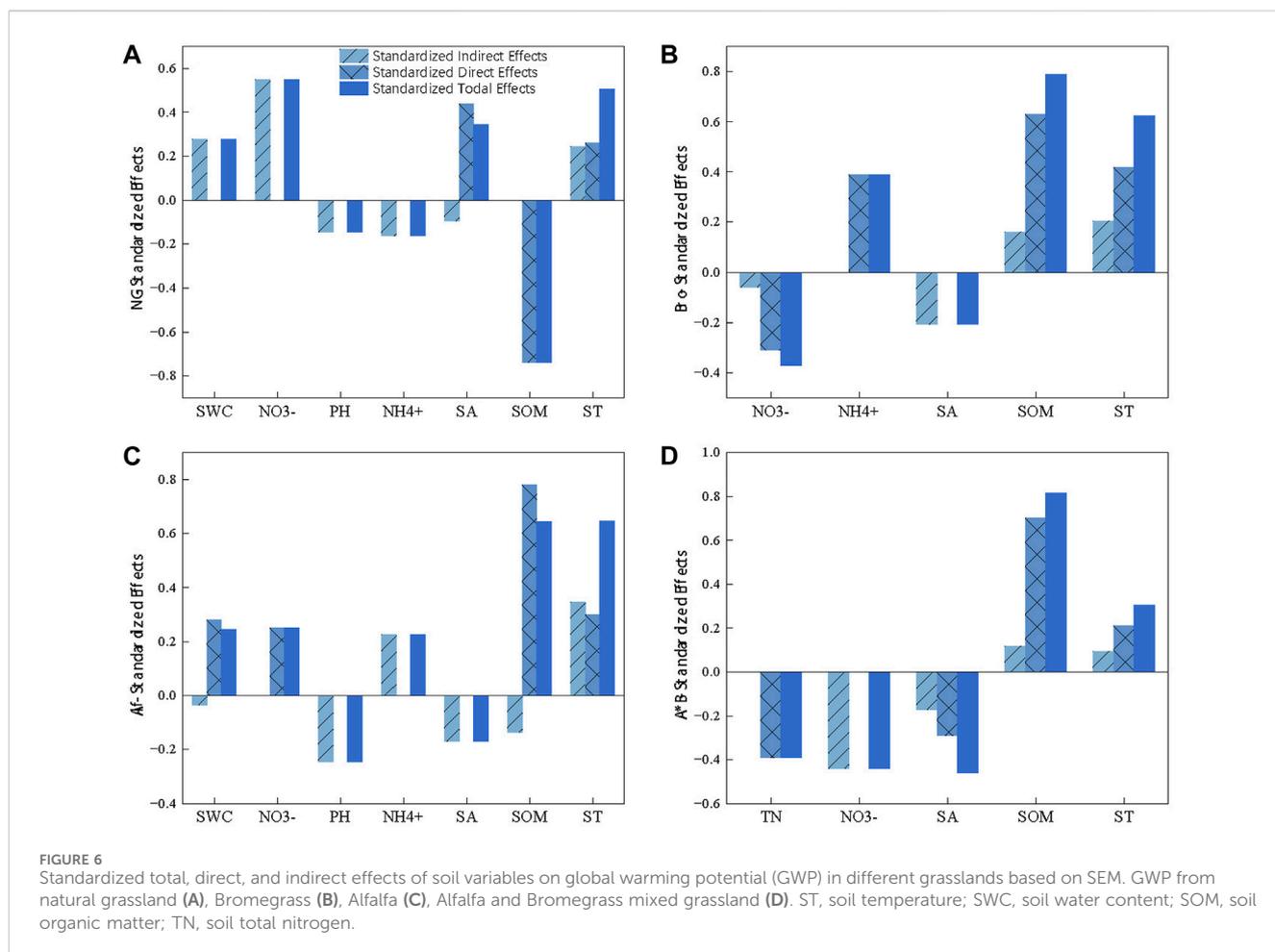
responses of NG and artificial grassland to P₁₅, as NG has abundant ventilatory organization.

4.2 FCO₂ response to different grassland types

We found that FCO₂ peaked during the mid-growing season and remained low at the beginning and end of the growing season (Figure 2). Higher temperatures, precipitation, and biomass during the midseason contribute to the input of substrates for soil microbial respiration and create a suitable environment for soil respiration, resulting in an increased FCO₂ (Sutton-Grier and Megonigal, 2011; Sun et al., 2013). However, a low FCO₂ value was observed at the end of July 2017 (Figure 2), which could be associated with precipitation during the measurement period because it hinders the diffusion of carbon dioxide. Furthermore, no significant correlation was observed between Bro FCO₂ and P₁₅ or SWC (Table 2), which may explain the relatively smaller fluctuations in Bro FCO₂ compared with other grasslands (Figures 2C, D).

The hydrothermal conditions in 2017 were better than those in 2016, indicating that the FCO₂ was higher. However, only NG had a higher ST in 2017 than in 2016, whereas artificial grasslands showed a decrease in ST in the second year (Figure 4A). In addition, NG exhibited a significant positive correlation with P₁₅, whereas Alf and A*B showed significant negative

correlations with P₁₅ (Table 2). Variations in precipitation and soil temperature explain the interannual changes in FCO₂ among the different grasslands (Sutton-Grier and Megonigal, 2011). Furthermore, our study indicates that NG FCO₂ strongly correlated with the salinity, SOM, and nitrate content, in addition to the ST. Monoculture grasslands only strongly correlated with SOM, whereas mixed-species grasslands exhibited strong correlations with multiple soil factors (Table 2). Bro and Alf have well-developed root systems that can alter the structure of soil aggregates, thereby influencing the distribution and flow of organic carbon (Six et al., 2002; Smucker et al., 2003; Li et al., 2020). The increased secretion of root exudates can enhance the soil microbial activity (Cheng et al., 1994; Kuz'yakov and Larionova, 2006; de Vries et al., 2019), leading to higher FCO₂ and lower SOM levels (Figures 2D, 4E). In contrast, non-legume wheatgrass exhibits better drought and salinity tolerance than purple prairie clover (Gong et al., 2015; Jia et al., 2018), which may explain why Alf had lower FCO₂ and higher SOM levels (Li et al., 2012). The higher emissions and lower carbon storage in mixed-species grasslands could be attributed to the complementary competition among different species (Figures 2D, 4E) (Lucero et al., 2000; Zhang et al., 2017). The mixture of legumes and grasses promotes better nutrient supply, and competition among plants stimulates rhizosphere effects by producing more types of root exudates, consequently jointly promoting respiration intensity in the soil layer.



4.3 FN₂O response to different grassland types

FN₂O exhibited minimal fluctuations and a gradual, non-discernible pattern over time regardless of the grassland type. These results are consistent with those of previous studies. Several researchers have indicated that N₂O emissions are significantly influenced by soil freeze–thaw cycles (Cao et al., 2020). The steeper warming observed during the early growing season in 2016 (Figure 1) may account for the higher initial N₂O emissions and the overall downward trend in 2016, as well as the subsequent upward trend in 2017 (Figures 2E, F). The formation of N₂O is a complex process involving the combined effects of nitrification and denitrification, which are regulated by environmental factors such as the soil temperature and moisture (Ding et al., 2013; Xia et al., 2022). N₂O is produced during nitrification and denitrification processes, with different dominant contributors and responses in various systems. Castaldi (2000) found that cropland N₂O mainly originated from denitrification and was more sensitive to temperature compared to forests (Castaldi, 2000). For grasslands, soil rewetting and water-holding capacity influenced the source and yield of N₂O, with soil moisture having a greater impact than temperature during the growing season (Du et al., 2006; Liu et al., 2012; Liu et al., 2018; Guo et al., 2022; Yu et al., 2022; Guo et al., 2023). Our results showed similar patterns.

We observed a strong negative correlation between FN₂O emissions and NH₄⁺ concentrations in all grasslands (Table 2). Some grasslands showed correlations with other soil physicochemical properties and were influenced by climatic factors, although a consistent trend was not observed. Further investigations into the mechanisms of soil microbial nitrification and denitrification processes may be necessary to better understand the mechanisms of soil N₂O emissions.

4.4 Drivers of GWP effects in different types of grassland

GWP generated from NG, Bro, Alf, and A*B were mainly regulated by different factors. In NG and Bro, GWP were mainly regulated by ST, SOM, and inorganic nitrogen. In contrast, those of Alf were mainly regulated by ST and SOM. For A*B, GWP were affected by SOM, NO₃⁻, TN, and salinity. This is consistent with previous reports that showed that Bro has a higher salt and alkali tolerance; therefore, it is mainly regulated by soil nutrients, whereas Alf has less nutrient limitation due to nitrogen fixation, and A*B is regulated by nutrient competition and salinity (Figure 5) (Lucero et al., 2000; Zhang et al., 2017; Jia et al., 2018). Regarding NG and artificial grasslands, SOM had different effects on GWP (Figure 5). This may be due to the following reasons: (1) Compared with NGs,

artificial grasslands adapt poorly to salt and alkaline environments. The input of fresh organic matter stimulates the activity of relevant organisms in artificial grasslands, thereby increasing GHG emissions and raising GWP. Thus, artificial grasslands use more fresh organic matter to complete the carbon cycle process and maintain or increase the soil organic carbon storage (Smith and Fang, 2010; Keiluweit et al., 2015). (2) NG is directly and negatively regulated by SOM and directly and positively regulated by salinity. Previous studies have reported that plant growth under saline conditions is inhibited, external organic matter input is reduced, and the SOC mineralization rate increases (Wong et al., 2010). In addition, under saline stress, soil microorganisms affect organic matter secretion and the number of dead bodies to maintain survival, thereby reducing the accumulation of SOM (Schimel et al., 2007). Therefore, NG may rely more on the old carbon pool for carbon cycling. Although A*B is also negatively regulated by the salinity, the input of fresh organic matter under legume–grass intercropping stimulated the positive regulation of GHG emissions by SOM. Furthermore, we observed a negative correlation between SOM and nitrate in all grassland systems. This may be due to microbial nitrification, which is inhibited under salt and alkaline conditions, reducing the availability of nitrate (McClung and Frankenberger, 1985; Badia, 2000; Fagodiya et al., 2022). Most plants are nitrophilous, and consume nitrate. All these imply a negative correlation between nitrate and SOM. NG, as a salt and alkali marsh grassland, exacerbates this change under wetland conditions (Kronzucker et al., 1997). Therefore, nitrate in NG affected GWP through negative regulation SOM production, while Bro and A*B produced opposite regulatory effects based on the same pathway. With respect to Alf, GHG emissions were positively regulated by NH_4^+ and NO_3^- , but the effect was not strong. This is closely related to the symbiosis between legumes and rhizobia. Rhizobia can convert nitrogen gas into ammonium ions for plant use, which greatly reduces the demand for exogenous nitrogen by leguminous plants and provides nitrogen sources for soil microorganisms, which reduces the nutrient limitation of the life activities in the soil layer, so that none of the environmental factors in Alf has a prominent influence on GWP (Figure 6C).

5 Conclusion

The results of this study demonstrated that fluxes varied among different grassland types. The FCH_4 of NG exhibited both sink and source functions over the growing season but a net emission over the entire season, whereas artificial grasslands served as CH_4 sinks throughout the growing season. The trend of FCO_2 in different grassland types was similar, with an initial increase followed by a decrease over the growing season. FN_2O showed a flat dynamic pattern during the growing season, without any noticeable regularity. Overall, the GHG emissions of different grasslands were dominated by CO_2 , with lower fluxes of N_2O and CH_4 . In addition, both FCO_2 and FCH_4 showed clear peaks during the growing season.

Our research indicates that the SWC was the primary driver of FCH_4 , whereas SOM and ST strongly correlated with FCO_2 . In addition, NH_4^+ significantly affected FN_2O . ST had a positive effect on GWP from all grasslands. With the exception of NG, SOM had a negative effect. Conversely, artificial grasslands were positively

affected by SOM. Furthermore, the nitrogen availability had a significant effect on NG and Bro. Finally, we observed that A*B was negatively affected by both the nitrogen availability and salinity. Together, these results suggest that artificial grasslands are sensitive to fresh organic input. Modifying nitrogen inputs in artificial grasslands could potentially alter the SOM and GHG emissions, leading to benefits in carbon sequestration and emission reduction. In our future work, we will explore the roles of different components of SOM in the carbon cycle in relation to microbial activities, and reveal the GHG emission mechanism and carbon stabilization effect more clearly.

Data availability statement

The original contributions presented in the study are included in the article/Supplementary Material, further inquiries can be directed to the corresponding author.

Author contributions

ZW: Writing–original draft. YC: Writing–review and editing. FH: Writing–review and editing.

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Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Supplementary material

The Supplementary Material for this article can be found online at: <https://www.frontiersin.org/articles/10.3389/fenvs.2024.1338180/full#supplementary-material>

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