Science of the Total Environment 829 (2022) 154628

Contents lists available at ScienceDirect

Science of the Total Environment

journal homepage: www.elsevier.com/locate/scitotenv

Appropriate N fertilizer addition mitigates N_2O emissions from forage crop fields

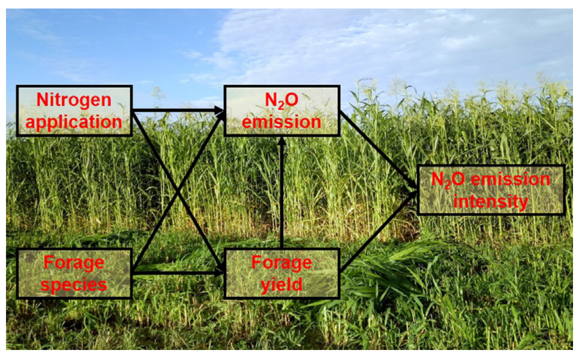
Jiao Ning, Shanning Lou, Yarong Guo, Shenghua Chang, Cheng Zhang, Wanhe Zhu, Fujiang Hou *

State Key Laboratory of Grassland Agro-Ecosystems, Ministry lab, College of Pastoral Agriculture Science and Technology, Lanzhou University, Lanzhou, Gansu 730020, China

HIGHLIGHTS

- N rate of ~200 kg ha^{-1} is more appropriate for forage to reduce N emissions.
- Barley and rye (lower CP) showed higher N_2O emission intensities.
- CP yield had a negative correlation with N_2O emission at 200 N and 250 N.
- N_2O emissions was explained better by the crude protein yield than the hay yield at 200 N.
- N rates of 150–200 kg ha^{-1} balance the competition between microbes and crops for N.

GRAPHICAL ABSTRACT



ARTICLE INFO

Article history:
Received 18 January 2022
Received in revised form 12 March 2022
Accepted 13 March 2022
Available online 16 March 2022

Editor: Elena Paoletti

Keywords:
Forage crop
Crude protein yield
N_2O emission intensity
Nitrogen application rate
Emission factor

ABSTRACT

Forage crops are widely cultivated as livestock feed to relieve grazing pressure in agro-pastoral regions with arid climates. However, gaseous losses of soil nitrogen (N) following N fertilizer application have been considerable in response to the pursuit of increased crop yield. A two-year experiment was carried out in a typical saline field under a temperate continental arid climate to investigate the effect of N application rate on N_2O emissions from barley (*Hordeum vulgare L.*), corngrass (*Zea mays* × *Zea Mexicana*), rye (*Secale cereale L.*), and sorghum-sudangrass hybrid (*Sorghum bicolor* × *Sorghum sudanense*). The dynamics of N_2O emissions, hay yield, and crude protein (CP) yield were measured under four N application rates (0, 150, 200, and 250 kg ha^{-1}) in 2016 and 2017. An N_2O emission peak was observed for all crop species five days after each N application. Cumulative N_2O fluxes in the growing season ranged from 0.66 to 2.40 kg ha^{-1} and responded exponentially to N application rate. Emission factors of N_2O showed a linear increase with N application rate for all crop species, but the linear slopes significantly differed between barley or rye and corngrass and sorghum-sudangrass hybrid. The hay and CP yields of all forage grasses significantly increased with the increase of N application rate from 0 to 200 kg ha^{-1}. Barley and rye with lower hay and CP yields showed higher N_2O emission intensities. The increased level of N_2O emission intensity was higher from 200 to 250 kg ha^{-1} than from 150 to 200 kg ha^{-1}. At N application rates of 200 and 250 kg ha^{-1}, CP yield had a significantly negative correlation with cumulative N_2O emission and explained 50.5% and 62.9% of the variation, respectively. In conclusion, ~200 kg ha^{-1} is the optimal N rate for forage crops to minimize N_2O emission while maintaining yield in continental arid regions.

1. Introduction

Nitrous oxide (N_2O) is a long-lived greenhouse gas (GHG) with 296 times greater global warming potential that of carbon dioxide (CO_2) over a 100-year period (IPCC, 2013) and is an atmospheric contaminant of major environmental concern. Croplands are an essential component of

* Corresponding author.
E-mail address: cyhoufj@lzu.edu.cn (F. Hou).

http://dx.doi.org/10.1016/j.scitotenv.2022.154628

terrestrial ecosystems and are responsible for more than 66% of global anthropogenic N_2O emissions (Portmann et al., 2012). In China, more than 60% of croplands are located in semi-arid and arid regions (China Agriculture Yearbook Editorial Board, 2011), where water shortage and soil salinization usually occur alongside low soil water retention capacity and poor nutrient contents (Wong et al., 2009). Therefore, as a major nutrient, nitrogen (N) fertilizers are extensively applied with the aim of enhancing crop yield, and economic profitability in these areas.

Nitrogen input is one of the key drivers of N_2O emissions and their high application rates incredibly increase N_2O emissions from croplands. The increase is estimated to account for 54% of the increase of global N_2O emissions in recent decades (Tian et al., 2019). A meta-analysis concluded that N application can increase N_2O emissions by up to 216% in agricultural ecosystems (Liu and Greaver, 2009). Broad-scale annual N emission budgets with a range of 4.4–9.5 Tg indicate great uncertainty in regional N_2O emissions (Hashimoto, 2012; Xu et al., 2012). The IPCC has recommended a default N_2O emission factor (EF) of 1% for applied inorganic N fertilizer lost as N_2O (IPCC, 2006), which implies a linear increase of N_2O emissions with increasing N application rates. Previous studies have shown a linear correlation between N_2O emissions and N application rate in a Chinese vegetable field (Yi et al., 2017) and a Canadian corn field (Roy et al., 2014). However, a meta-analysis from 78 papers presented that N_2O emissions exponentially increase with increasing the N application rates (Shcherbak et al., 2014). Increasingly, evidence suggests an exponential or non-linear relationship between N application rates and N_2O emission and non-constant N_2O EFs in the soil with different N application rates (Cardenas et al., 2010; Rahman et al., 2021). However, the response curves of both N_2O emissions and EFs to N application rates remain uncertain in many cropping systems. Investigation of the responses in specific soils can help to minimize the under-or over-estimation of N_2O emissions based on default EF values.

The magnitude of N_2O emissions in croplands not only depends on the N application rate but also on crop type (Van Groenigen et al., 2015). Studies regarding N fertilizer application increasing N emissions in crop fields mainly focus on food crops. Forage crops support more than 70% of sheep and goats and 50% of livestock meat globally (Hou et al., 2008). In arid and semi-arid areas, stall-feeding is prevalent because of the serious seasonal drought, and therefore forage crops have been widely planted in recent years to meet the increasing demand for meat products and relieve grazing pressure (Venuto and Kindiger, 2008; Lithourgidis and Dordas, 2010; Miller et al., 2018). Increasing N application rates significantly increases the forage yield within a certain range, afterward there is an inflection point where additional N application would have no significant effects or can even reduce e forage yields. Thus, adjusting N application rates according to crop needs has the potential to decrease soil N interceptions and mitigate N_2O emissions with little or no yield penalty (Davidson and Kanter, 2014). To this end, N_2O emission intensity (NEI), a measure of yield-scale N_2O emissions, has been used as an index to evaluate N_2O emissions from agricultural ecosystems (Van Groenigen et al., 2010; Pittelkow et al., 2014).

Few studies have investigated the response of N_2O emissions to N fertilizer application for annual forage crops, especially in saline soil. The area of saline land accounts for more than 50% of global arable land (Shrivastava and Kumar, 2015). Soluble salts in soil decrease the rates of mineralization and nitrification, which can increase (Reddy and Crohn, 2014; Kong, 2015) or decrease (Azam and Müller, 2003; Adviento-Borbe et al., 2006) soil N_2O emissions. Therefore, in this study, soil N_2O emissions from four species of a temperate forage crop system grown on saline soil were monitored under four N fertilizer application rates over two complete growing periods. The main objectives were to: (1) identify which forage crops emit more N_2O under different N application rates; (2) fit the relationship between N_2O emissions or EF and N application rate; and (3) determine the appropriate N application rate for forage crops in arid regions. The following two hypotheses are proposed: (1) the crop species with higher N uptake from soil (i.e., CP yield) emits lower N_2O by decreasing the soil N content; and (2) the relationship between N_2O, EF and N application rates show differences between species as a result of the variation in N demand among different crops. These results would help to improve agricultural GHG reduction in arid regions.

2. Materials and methods

2.1. Study site

The field study was carried out at the Linze Grassland Agricultural Trial Station of Lanzhou University (39°15′N, 100°02′E), Zhangye, Gansu Province, China in the growing seasons of 2016 and 2017. The site is located in the northwest inland arid area and has a temperate continental climate. The mean annual temperature was 8.8 °C and the average precipitation was 88.2 mm in 2016 and 2017. The soil type at the research site is Aquisalids (Ning et al., 2020). The study site is flat and initial soil properties were measured before the experimental manipulation. The basic soil properties at 20 cm soil depth were as follows: soil organic carbon content, 9.78 g kg^{-1}; total N content, 1.32 g kg; pH, 8.6; salt content, 0.6%–0.9%; and bulk density, 0.93 g cm^{-3}.

2.2. Experimental design

The experimental field has been managed under a conventionally plowed system for 5 years with a wheat–corn rotation. Four annual crop species, barley (*Hordeum vulgare L.*), corngrass (*Zea mays* × *Zea Mexicana*), rye (*Secale cereale L.*), and sorghum-sudangrass hybrid (*Sorghum bicolor* × *Sorghum sudanense*) were chosen for measuring N_2O emissions and forage productivity under different N fertilizer rates. Four N fertilizer rates including 0 kg ha^{-1} (0 N), 150 kg ha^{-1} (150 N), 200 kg ha^{-1} (200N), and 250 kg ha^{-1} (250 N) were used according to the demand of these forage crops. The experiment was organized in a randomized complete block design with three replications. Each plot was 15 m × 6 m (area = 90 m^2), and an isolation belt of 1.5 m wide and a ridge were designed between each plot to prevent water and N fertilizer leakage. Traditional flat planting (50 cm in width) was used for the corngrass and sorghum-sudangrass hybrid, their seeding spacings respectively were 25 and 20 cm, and evaluated densities were 82,500 and 105,000 plants ha^{-1}, respectively. Broadcast sowing was used for barley and rye and the seeding rates were 375 and 300 kg ha^{-1}, respectively.

All crop species were sown at the end of May. A hole-sowing machine with a seeding depth of 4–5 cm was used for corngrass and sorghum-sudangrass hybrid, and a manual broadcasting sower was used for barley and rye. Nitrogen fertilizer (urea) was applied in two equal portions on 28 June and 10 August in 2016 and 30 June and 12 August in 2017 before supplemental irrigation of 120 mm. Each year, the other two irrigations were provided to forage crops. The first irrigation (120 mm) was providedin late April (before sowing) while the second irrigation (200 mm) in late October (winter irrigation). During the growing season, weeds were pulled manually once a fortnight. The corngrass and sorghum-sudangrass hybrid were cut three times per year at the stubble height of 15 cm on 26 July, 24 August, and 26 September in 2016 and 28 July, 25 August, and 28 September in 2017. Crop average heights of the corngrass and sorghum-sudangrass hybrid were about 1.3 and 1.6 m, respectively. Barley and rye were also cut three times per year at the stubble height of 5 cm on 20 July, 17 August, and 20 September in 2016 and 23 July, 20 August, and 22 September in 2017. Crop average heights were about 30 cm.

2.3. Nitrous oxide sampling and analysis

The sampling of N_2O gas in the field was carried out during the growing season (from before sowing to after harvesting) in 2016 and 2017. Since more than 90% of cumulative N_2O emissions occurred in 15 days after nitrogen application in conditions similar of the present study (Lin et al., 2020; Jahangir et al., 2021), gas samplings were conducted on 0, 1, 3, 5, 7, 10, and 15 days after N application, and once a fortnight for all other times during the growing season.

The N_2O flux was measured at 9:00–11:00 of each chosen day. Static opaque chambers (30 cm × 30 cm × 30 cm) were used to sample gas in each plot. The structure of the static chamber was the same as Ning et al. (2020). Gas samples were taken using a 50-ml plastic syringe and then transferred into a 300-mL vacuum aluminum foil gas-collection bag. For each sampling event during 30 min, four gas samples of approximately 200 mL were collected at time intervals of 10 min. The temperature inside the chamber was measured with an electronic thermometer during gas sampling.

The N_2O concentration was analyzed within 24 h by an N_2O Spectrum Analyzer (Model No. 908–0015-0000, Los Gatos Research, USA). The exchange flux of N_2O was used to describe the concentration change of gas per unit time in the chamber, which was calculated according to Liu et al. (2017):

$$Flux = \rho H \times \frac{P}{P_0} \times \frac{273.15}{T} \times \frac{dCt}{dt}$$

where ρ is the standard gas density, H is chamber height, P is the atmospheric pressure of the sampling sites (85.48 kPa), P_0 is standard atmosphere pressure, T is the temperature in Kelvin inside the chamber, and dCt/dt is the mean rate of N_2O concentration change with time. Cumulative N_2O emissions during the growing season were the sum of daily fluxes. Undetermined daily fluxes were gap-filled using linear interpolation of the arithmetical means of N_2O fluxes for the two close days (Chen et al., 2013).

The EF of N_2O was calculated according to IPCC (2006):

$$EF\ (\%) = \frac{N_2O_F - N_2O_{0F}}{N_F} \times 100$$

where N_2O_F is the cumulative N emissions from a fertilizer plot, N_2O_{0F} is the cumulative N emission from the plots without N application, and N_F is the corresponding N application rate.

2.4. Forage yield and yield-scaled N_2O fluxes

To determine the hay and crude protein (CP) yields, three quadrats (50 cm × 50 cm) were set up in each plot for barley and rye, and eight typical plants of corngrass and sorghum-sudangrass hybrid were randomly selected in each plot. Sampled fresh grasses were oven-dried at 65 °C for a minimum of 48 h. After weighing, the forage samples were ground and passed through a 0.25-mm sieve, then the CP was measured by the Kjeldahl method (Schuman et al., 1973).

The N_2O emission intensity (NEI) was calculated by dividing the cumulative N_2O emissions by the hay and CP yields (Dyer et al., 2010). Hay yield-scaled and CP yield-scaled N_2O fluxes were represented by NEI_{hay} and NEI_{CP}, respectively.

2.5. Statistical analyses

All statistical analyses were conducted using SAS 9.4 (SAS Institute Inc., Cary, NC, USA). Results of a Shapiro–Wilk test (UNIVARIATE Procedure) indicated that data collected from this study were normally distributed. The ANOVA (GLM) was applied to examine the independent and interaction effects of fixed factors on dependent variables. A least significant difference test was used to compare the mean differences in cumulative N_2O emissions, hay and CP yields, EF, and NEIs between species, N application rates, and years. The exponential correlation of N application rate and cumulative N_2O emissions for each species was determined using nonlinear regression. The N application rates in relation to EFs were determined using linear regression (GLM Procedure), and the regression slopes were compared using an analysis of covariance (ANCOVA) with a CONTRAST statement for pairwise comparison.

3. Results

3.1. Temporal dynamics of N_2O fluxes

The mean air temperature and cumulative precipitation in the growing season in 2016 and 2017 were 19.5 °C and 46.0 mm, and 21.1 °C and 105.1 mm, respectively.

Repeated-measures ANOVA for the effects of N application rate and crop species showed that N_2O dynamics over the two-year sampling were not significantly affected by crop species but were significantly affected by the N application rate ($F = 153.91$, $P < 0.001$) (Fig. 1B). There were N_2O emission peaks after each N application to the soils, with the highest N_2O emission peak observed on Day 5 after N application for all crop species (Fig. 1B).

3.2. Cumulative emission and emission factor of N_2O

The ANOVA for cumulative N_2O emissions using N application rate, crop species, and year as fixed effects showed that all factors and all interactions of each two factors had significant effects on the cumulative N_2O flux (Fig. 2).

Cumulative N_2O fluxes ranged from 0.66 to 2.40 kg ha^{-1} and significantly increased with N application rate for all crop species ($P < 0.05$) (Fig. 2). The mean cumulative N_2O fluxes in 2016 and 2017 under 0 N, 150 N, and 200 N for barley were 83.1%, 137.5%, and 225.2% greater than that under 0 N, respectively. For corngrass, they were 80.9%, 137.9%, and 196.4% greater than that under 0 N, respectively. For rye, they were 93.0%, 149.8%, and 243.1% greater than that under 0 N, respectively. In the sorghum-sudangrass hybrid, they were 78.9%, 126.7%, and 188.7% greater than that under 0 N, respectively. The cumulative N_2O emissions responded exponentially to the N application rates from 0 to 250 kg ha^{-1} (Fig. 3A).

The ANOVA for EF based on N application rate, crop species, and year as fixed effects showed that N application rate, species, and their interaction had significant effects on EF ($P < 0.05$). The EF ranged from 0.23% to 0.27%, from 0.28% to 0.33%, and from 0.33% to 0.43% for 150 N, 200 N, and 250 N, respectively (Table 1). The EFs did not differ among crop species at N application rates under 150 N; however, under 200 N, the EFs were greater for rye and corngrass than for sorghum-sudangrass hybrid and under 250 N, the EFs were greater for rye and barley than for corngrass and sorghum-sudangrass hybrid (Table 1). The EFs of 250 N were significantly greater than those of 200 N for all species, and EFs of 200 N were significantly greater than those of 150 N for corngrass and sorghum-sudangrass hybrid ($P < 0.05$). The EFs of N_2O responded linearly to the N application rates from 150 N to 250 N ($P < 0.05$) (Fig. 3B), and the linear slopes significantly differed between barley or rye and corngrass and sorghum-sudangrass hybrid (the P values of barley vs corngrass, barley vs sorghum-sudangrass hybrid, rye vs corngrass, and rye vs sorghum-sudangrass hybrid were 0.018, 0.009, 0.032, and 0.017, respectively).

3.3. Forage production and N_2O emission intensity

The ANOVA for hay and CP yields using N application rate, crop species, and year as fixed effects showed that N application rate, species, and the interaction of year and species had significant effects on the CP yield ($P < 0.05$). Nitrogen fertilizer application significantly improved the hay and CP yield (Fig. 4). Compared with 0 N, the 150 N, 200 N, and 250 N treatments respectively increased the mean hay yield by 14.5%, 33.5%, and 33.6% for barley, by 12.5%, 30.0%, and 30.7% for corngrass, by 15.9%, 30.6%, and 28.9% for rye, and by 10.9%, 23.3%, and 22.9% for sorghum-sudangrass hybrid. Moreover, they increased the mean CP yield by 18.5%, 36.4%, and 38.5% for barley, by 13.9%, 29.6%, and 30.6% for corngrass, by 15.5%, 29.2%, and 28.6% for rye, and by 11.4%, 22.4%, and 22.4% for sorghum-sudangrass hybrid. There were no significantly differences in both yields between 200 N and 250 N. Hay and CP yields were greater for corngrass and sorghum-sudangrass hybrid than barley and rye regardless of the N application level (Fig. 4).

The ANOVA for NEI using N application rate, crop species, and year as fixed effects showed that N application rate, species, year, and the

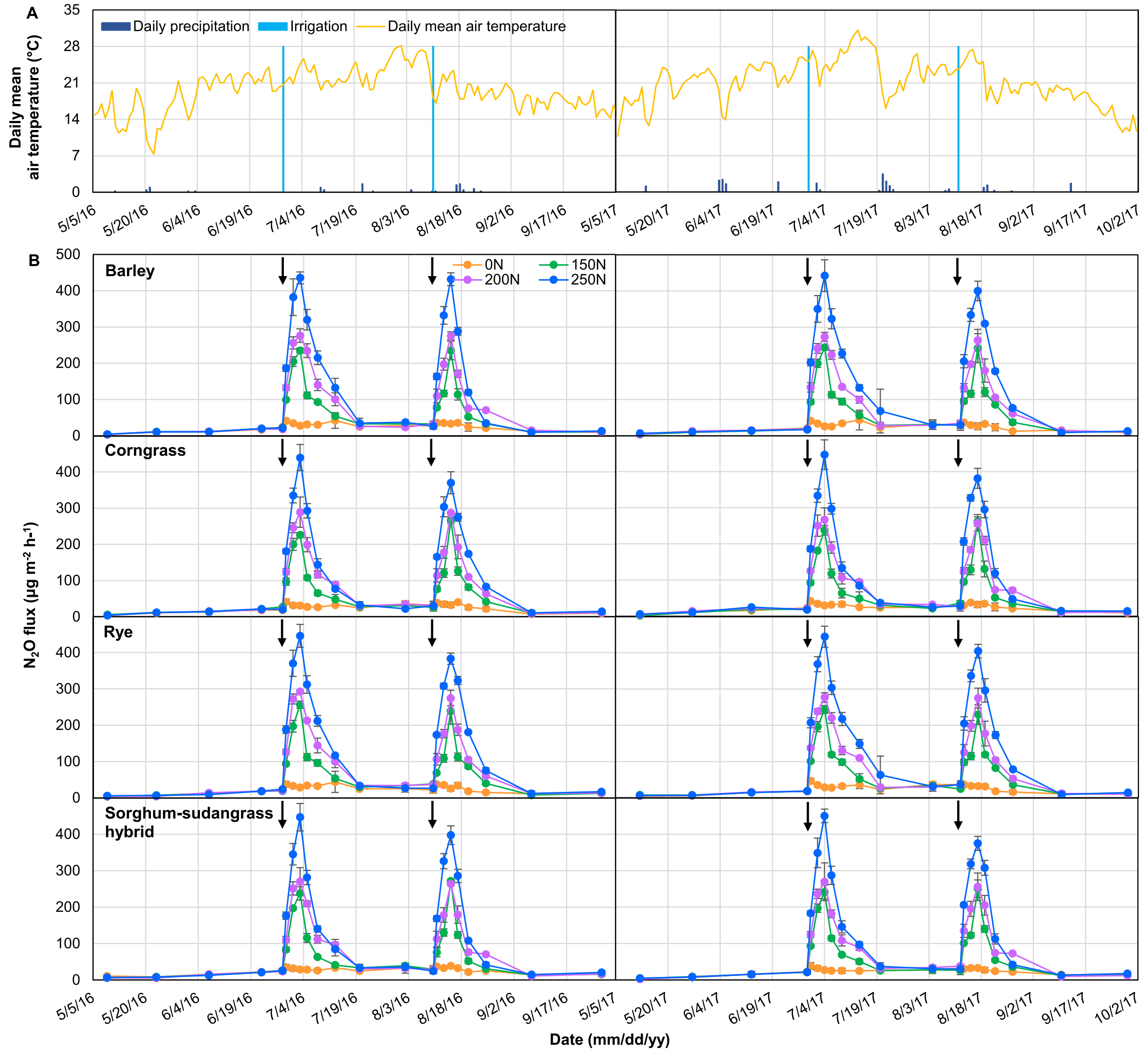


Fig. 1. Daily air temperature, precipitation, and irrigation of study site (A) and N_2O flux (mean $\pm$ SE; $n = 3$) for four N application rates over time in barley, corngrass, rye, and sorghum-sudangrass hybrid fields (B) at the growing seasons in 2016 and 2017. The arrow shows the day of N application.

interactions of every pair of factors had significant effects on both NEIs ($P <$ 0.05). The N_2O intensities for hay and CP yields of all species significantly increased with the increase of N application rate, and thus they were greatest under 250 N (Table 2). The N_2O intensities for hay and CP yields were significantly greater in barley and rye than in corngrass and sorghum-sudangrass hybrid, and they were significantly greater in corngrass than in the sorghum-sudangrass hybrid.

The cumulative N_2O emissions were not significantly correlated with hay yields under 0 N, 150 N, and 200 N, but exhibited a negative relationship with hay yield under 250 N (Fig. 5A). In addition, the cumulative N_2O emissions showed a significant negative correlation with with CP yield under 200 N and 250 N (Fig. 5B).

4. Discussion

4.1. Nitrous oxide emissions in different crop species

At no fertilizer application (0 N), contrary to our first hypothesis, the sorghum-sudangrass hybrid (with greater hay and CP yields) emitted more N_2O than the rye (with lower hay and CP yields) (Fig. 2 and Fig. 4). Crops in agricultural ecosystems directly regulate N cycling by taking up N from the soil and storing it in their biomass (Fowler et al., 2013), and indirectly by regulating the biotic factors driving N_2O emissions (Abalos et al., 2014). Many ecologists have found several links between plant growth traits and biogeochemical processes and have also revealed that plants may influence soil N cycling in a manner consistent with the acquisitive–conservative gradient (Baxendale et al., 2014; Grassein et al., 2015; De Vries and Bardgett, 2016). In comparison with conservative plant species (those with a low ability to acquire soil available N), acquisitive species (those with a high ability to acquire soil available

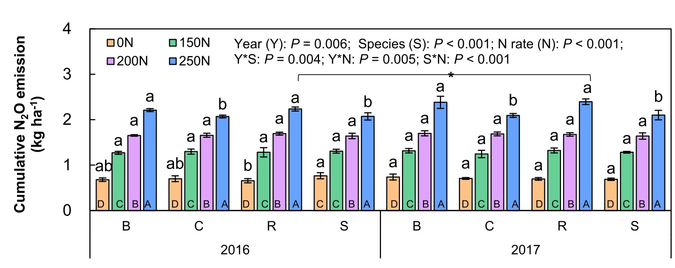


Fig. 2. Cumulative N_2O emissions (mean $\pm$ SE) for four N application rates in barley (B), corngrass (C), rye (R), and sorghum-sudangrass hybrid (S) fields. The means with the same lowercase letters among different species and with the uppercase letters among different N fertilizer rates in the same year are not significantly different at $P < 0.05$; asterisks (*) indicate a significant difference of soil N_2O emissions between different species at $P < 0.05$.

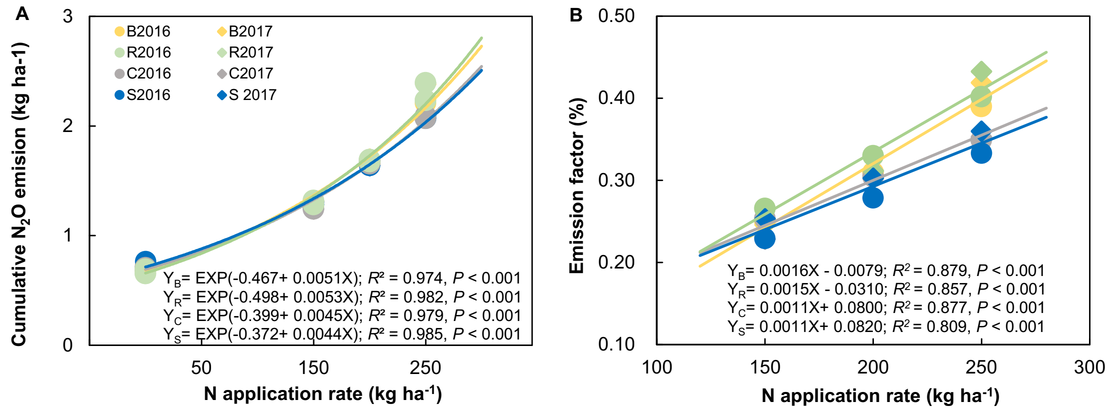


Fig. 3. Relationship between N application rate and cumulative N_2O emissions (A), and N_2O emission factors (B) in the four crop fields.

Table 1
Emission factors (% of applied N) from different N application rates in the four crop fields.

Year	Crop	N_2O emission factor (% of N applied)						$F_{2,6}$	P
		150 kg ha^{-1}		200 kg ha^{-1}		250 kg ha^{-1}			
2016	B	0.251 ± 0.015	Ac	0.310 ± 0.005	ABb	0.390 ± 0.004	Ca	57.00	<0.001
	C	0.254 ± 0.005	Ac	0.305 ± 0.010	ABb	0.349 ± 0.011	DEa	30.17	<0.001
	R	0.266 ± 0.029	Ab	0.330 ± 0.010	Ab	0.402 ± 0.011	BCa	13.08	0.007
	S	0.229 ± 0.017	Ac	0.279 ± 0.011	Bb	0.333 ± 0.007	Ea	16.82	0.004
2017	B	0.246 ± 0.023	Ab	0.307 ± 0.018	ABb	0.419 ± 0.011	ABa	23.40	0.002
	C	0.229 ± 0.016	Ac	0.312 ± 0.010	Ab	0.353 ± 0.003	DEa	34.00	<0.001
	R	0.266 ± 0.013	Ac	0.312 ± 0.009	Ab	0.433 ± 0.004	Aa	91.94	<0.001
	S	0.253 ± 0.009	Ac	0.303 ± 0.009	ABb	0.360 ± 0.014	Da	23.88	0.001
	$F_{7,16}$	0.67		1.71		16.98			
	P	0.697		0.177		<0.001			

The means (±SE) with the same uppercase letters in column and lowercase in row are not significantly different at P < 0.05.

N) can destabilize the soil environment and increase the abundance and activity of soil microorganisms involved in the N cycle (Grigulis et al., 2013; Baxendale et al., 2014), and thus they may increase N_2O emissions. However, these results have mainly been demonstrated in soil with low mineral N contents (Butterbach-Bahl et al., 2013; Abalos et al., 2016). Therefore, we deduced that sorghum-sudangrass hybrid play a role of acquisitive plant in the soil without N application. This may explain our finding that the sorghum-sudangrass hybrid emitted

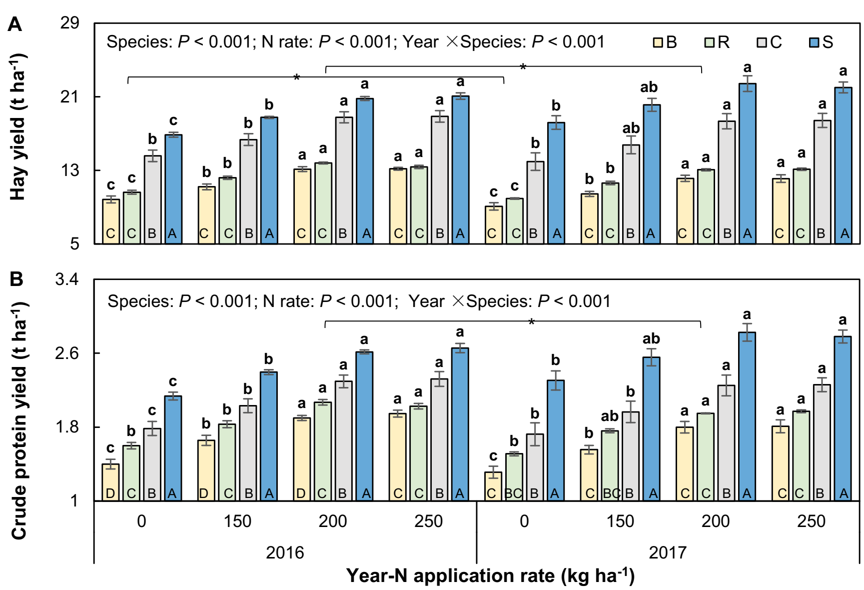


Fig. 4. Hay yield (mean ± SE) (A) and CP yield (mean ± SE) (B) under four N application rates in 2016 and 2017. In the same year, the means with the same lowercase letters between different N fertilizers and with the same uppercase letters between different species are not significantly different at P < 0.05; asterisks (*) imply significant difference between different years at P < 0.05.

Table 2
NO_2 emission intensity for hay yield (NEI_{hay}) and for CP yield (NEI_{CP}) under four N application rates in 2016 and 2017.

Year	Crop	N_2O intensity (kg N_2O t hay/CP^{-1})								$F_{3,8}$	P
		0 kg ha^{-1}		150 kg ha^{-1}		200 kg ha^{-1}		250 kg ha^{-1}			
NEI_{hay} (kg N_2O t hay^{-1})											
2016	B	0.069 ± 0.002	Ad	0.113 ± 0.005	Ac	0.126 ± 0.003	Ab	0.168 ± 0.003	Aa	149.46	<0.001
	C	0.048 ± 0.002	Bd	0.080 ± 0.002	Bc	0.088 ± 0.002	Bb	0.110 ± 0.004	Ba	106.25	<.001
	R	0.062 ± 0.003	Ad	0.105 ± 0.004	Ac	0.123 ± 0.002	Ab	0.167 ± 0.001	Aa	290.02	<0.001
	S	0.045 ± 0.002	Bd	0.069 ± 0.001	Cc	0.079 ± 0.002	Cb	0.099 ± 0.001	Ca	155.12	<0.001
$F_{3,8}$		21.75		43.36		112.38		218.15			
P		<0.001		<0.001		<0.001		<0.001			
2017	B	0.081 ± 0.003	Ad	0.126 ± 0.004	Ac	0.140 ± 0.005	Ab	0.197 ± 0.001	Aa	157.06	<0.001
	C	0.051 ± 0.004	Cc	0.080 ± 0.005	Cb	0.092 ± 0.003	Cb	0.114 ± 0.005	Ca	34.25	0.001
	R	0.070 ± 0.001	Bd	0.113 ± 0.001	Bc	0.128 ± 0.002	Bb	0.182 ± 0.001	Ba	1024.1	<0.001
	S	0.038 ± 0.001	Dc	0.064 ± 0.003	Db	0.073 ± 0.002	Db	0.096 ± 0.005	Da	68.61	<0.010
$F_{3,8}$		51.06		60.88		86.49		198.81			
P		<0.001		<0.001		<0.001		<0.001			
NEI_{CP} (kg N_2O t CP^{-1})											
2016	B	0.485 ± 0.014	Ad	0.768 ± 0.034	Ac	0.870 ± 0.017	Ab	1.136 ± 0.027	Aa	121.09	<0.001
	C	0.392 ± 0.014	Bd	0.639 ± 0.012	Bc	0.722 ± 0.013	Bb	0.894 ± 0.030	Ba	124.19	<0.001
	R	0.411 ± 0.022	Ad	0.700 ± 0.026	ABc	0.818 ± 0.017	Ab	1.103 ± 0.008	Aa	219.57	<0.001
	S	0.358 ± 0.015	Bd	0.545 ± 0.015	Cc	0.628 ± 0.016	Cb	0.781 ± 0.007	Ca	158.40	<0.001
$F_{3,8}$		10.5		16.2		45.53		66.5			
P		<0.001		<0.001		<0.001		<0.001			
2017	B	0.561 ± 0.023	Ad	0.847 ± 0.029	Ac	0.947 ± 0.044	Ab	1.319 ± 0.014	Aa	111.11	<0.001
	C	0.414 ± 0.035	Cc	0.637 ± 0.040	Cb	0.751 ± 0.030	Bb	0.928 ± 0.034	Ca	37.45	<0.001
	R	0.459 ± 0.009	Bd	0.750 ± 0.009	Bc	0.859 ± 0.013	Ab	1.215 ± 0.009	Ba	988.12	<0.001
	S	0.299 ± 0.009	Dd	0.504 ± 0.020	Dc	0.580 ± 0.013	Cb	0.757 ± 0.036	Da	73.11	<0.001
$F_{3,8}$		24.85		29.46		31.05		96.19			
P		<0.001		<0.001		<0.001		<0.001			

The means (±SE) with the same uppercase letters in column and lowercase in row are not significantly different at $P < 0.05$.

more N_2O than the rye. In addition, at lower levels of available N, the plants with lower N uptake have a weaker impact on N mineralization (De Vries and Bardgett, 2016), and soil microorganisms compete more effectively than crops for N resources at low N soils (Kuzyakov and Xu, 2013; Thebault et al., 2014). Therefore, the N_2O emission may not be affected by crop yields in the soil with low N contents and/or no N application.

When soil available N is sufficient after N fertilizer application, acquisitive species often achieve high N uptake rates, and biomass and CP productions (De Vries and Bardgett, 2016). A high N capture by the plants may lead to lower soil mineral N levels, thereby reducing the substrate concentration for nitrification and denitrification, and thus decreasing N_2O emissions (Abalos et al., 2018). We found that cumulative N_2O emissions of corngrass and sorghum-sudangrass hybrid (with greater CP and hay yields) were significantly lower than those of barley and rye (with low CP and hay yields) only under 250 N (Fig. 2). Moreau et al. (2015) showed that increasing plant N uptake negatively affected the abundance of nitrate-reducing microorganisms. Therefore, it is likely that forage crops under a high N application rate regulate N_2O emissions by controlling N uptake (crude protein yield) then adjusting the abundance of denitrifying communities in soils, and the competition of soil microbial communities and crops for N source may be balanced by an N application rate of around 150–200 kg ha^{-1}.

4.2. Nitrous oxide emission under different N application rates

The results of the present study support the findings of previous studies that annual cumulative N_2O emissions are predominantly driven by peak fluxes of short duration triggered by N fertilization events, and the magnitude of these peak fluxes increases with N application rate (Bell et al., 2016; Roche et al., 2016; Cardenas et al., 2019). Nitrous oxide emissions in fields are primarily regulated by the competition between crops and microorganisms for N sources (Kim et al., 2013). At N application rates below crop demand, N_2O emissions linearly increase with the increase of N application rate because of the limited substrate for denitrification (Roy et al., 2014; Yi et al., 2017; Jahangir et al., 2021). However, once N application rates exceed crop demand, the N available to soil microbial metabolisms increases sharply with the N application rate, resulting in an exponential increase of N_2O emissions (McSwiney and Robertson, 2005; Davidson and Kanter, 2014). Exponential models had a better fit to the responses of N_2O emissions to N application rates in the present study than linear models, which suggests that N application was likely to exceed crop demand and points out the potential opportunities of reducing the N application rate while maintaining or improving productivity (Fig. 3A). Exponential responses of N_2O emission to N application rates also were demonstrated in various similar cropping systems such as maize (Song et al., 2018) and wheat (Millar et al., 2018).

Different soil and climate types lead to a wide variation of N_2O emissions stimulated by N fertilization (Smith et al., 2012; Rees et al., 2013; Rahman et al., 2021), and thus variation trends of EFs with N application rates can show increases, decreases, or remain constant in agricultural soils (Dai et al., 2013; Hinton et al., 2015; Kuang et al., 2021;). In addition, the non-significant correlations between EF and N application rates have also been reported in spring barley fields (Roche et al., 2016) and intensive grassland (Bell et al., 2016). Consistent with our second hypothesis, positive linear relationships were detected between the EF and N application rates, and the linear slopes were greater in barley and rye than in corngrass and sorghum-sudangrass hybrid (Fig. 3B). The change of N uptake caused by the difference in crop N demand and yield under N application is likely to be the main reason for the difference in EFs among different crops (Aguilera et al., 2013; Forrestal et al., 2017; Rahman et al., 2021). Therefore, our results suggest that forage grasses in the same soil environment may not change the positive linear relationship between N application rate and EF, but change the linear slope.

The default N_2O EF is 1% according to IPCC, however, an increasing number of regional studies have shown incredibly wide ranges of EFs in different agricultural systems. Rahman et al. (2021) reported that the EFs ranged from 0.04% to 4.68% because of the different temperature, precipitation, and management practices between fields. A meta-analysis (Stehfest and Bouwman, 2006) showed that N_2O emissions from agricultural fields were lower in temperate climates than tropical and subtropical climates. Takeda et al. (2021) reported that EFs ranged from 0.69% to 1.11% in a sugarcane cropping system, partly exceeding the IPCC default

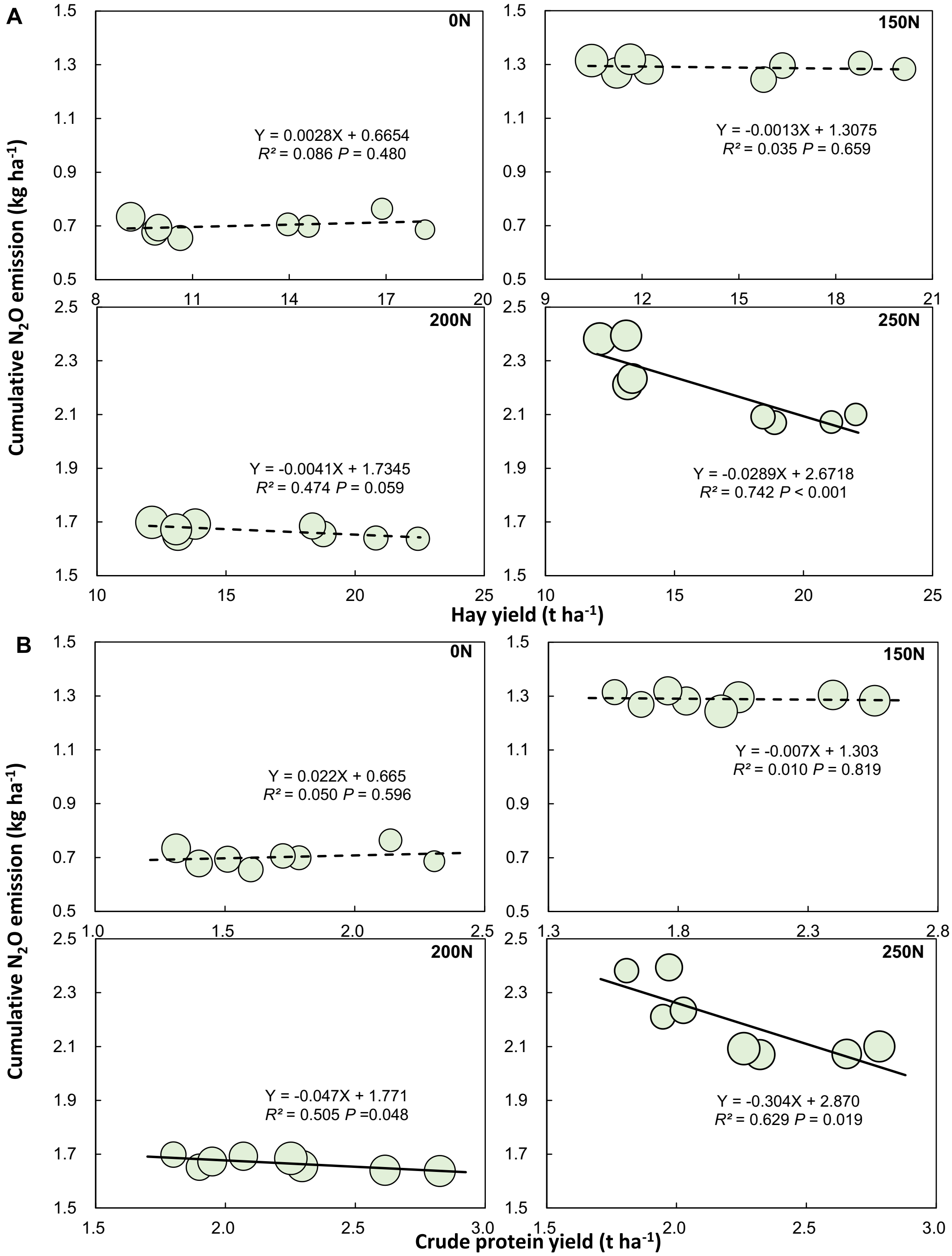


Fig. 5. Relationships between cumulative N_2O emissions and hay yield (A) and CP yield (B) under different N application rates. The size of each bubble represents the ratio of mean cumulative N_2O emissions to mean hay or CP yield.

of 1% at 250 kg N ha^{-1}. A large EF of 2.3% at the global scale (Thompson et al., 2019) implies that measuring regional N_2O emissions rather then correcting the current assumption of fixed EFs is important. In the present study, the EFs for 150 N, 200 N, and 250 N were 0.25%, 0.31%, and 0.38%, respectively, remaining far below the IPCC default EF of 1% (Table 1). The low EFs may be due to N consumption in other forms, for example, plant absorption and N leaching. In the present study, the capacities of arid and saline soil to retain water and fertilizer are low, and flood irrigation may further increase soil N leaching. Azam and Müller (2003) reported that N_2O emissions can be restrained in soil with high salt contents in arid regions. In addition, the time of N application may have facilitated relatively lower N_2O emissions. Since fertilizer was applied about one month after grass planting, N demand by all the crops was likely already pretty high when fertilizer was applied. Therefore, we conclude that extensive irrigation, saline soil, and N application time may reduce the N_2O emissions caused by N fertilizer.

4.3. Yield-scaled N_2O emissions

The calculation of yield-scaled N_2O emissions under different N application rates provides a method to balance the benefit between crop production and environmental sustainability (Van Groenigen et al., 2010;

Pittelkow et al., 2014). Using this metric, we found that overall mean N_2O emissions per unit hay/CP yield of corngrass and sorghum-sudangrass hybrid under 0 N, 150 N, 200 N, 250 N were 35.1%/23.7%, 36.1%/24.2, 35.6%/23.2, and 31,3%/29.6%, respectively, lower than that of barley and rye (Table 2). In arid regions, forage crops are often used to relieve grazing pressure in recent years (Lithourgidis and Dordas, 2010; Miller et al., 2018). We found that in the current context of widespread forage crop use, C4 species, such as corngrass and sorghum-sudangrass hybrid, should be promoted to develop productive agro-pastoral regions that efficiently retain N and contribute to mitigating climate change. Although yield-scaled N_2O emissions significantly increased with the N application rate, the level of increase was higher from 200 N to 250 N than from 150 N to 200 N. This suggests that ~200 kg ha^{-1} is the appropriate N application rate for these forage crops to reduce N emissions while maintaining yield in arid regions.

In addition, the significant negative correlations between cumulative N_2O emissions and CP yield were detected under 200 N and 250 N (Fig. 5B), and that between cumulative N_2O emissions and hay yields was detected only under 250 N (Fig. 5A). This confirms the importance of CP and biomass productivities as a key mechanism regulating crop–soil interactions in terms of N_2O emissions when soil available N is high (Grassein et al., 2015; De Vries and Bardgett, 2016; Abalos et al., 2018). Notably, CP yield was more efficient than hay yield for explaining the change of cumulative N_2O emissions under a wide range of high N applications, which may indicate that CP yield can directly reflect crop N uptake.

5. Conclusion

Results from the present study indicated the forage crops with higher yields emitted more soil N_2O receiving no N application but less N_2O from soils under high N application rates. This implies that the competition between soil microbial communities and crops for N sources achieve balance at N application rates of around 150–200 kg ha^{-1} in arid saline regions. We found that forage grasses in the same soil environment might not change the positive linear relationship between N application rate and EF, but change the linear slope. In addition, extensive irrigation, saline soil, and N application time may reduce the N_2O emissions caused by N fertilizer by increasing leaching. We suggest that an N application rate of ~200 kg ha^{-1} is appropriate for these forage crops to reduce N emissions while maintaining yield in continental arid regions. Barley and rye had lower hay and CP yields and higher N_2O emission intensities. Cumulative N_2O emission had a negative correlation with CP yield at N rates of 200 and 250 kg ha^{-1} and with hay yield at at N rates of 250 kg ha^{-1}. Future studies on N application in crop fields should include more N application levels to identify the N application rate that best balances the competition between soil microbial communities and crops for the N source to minimize N_2O emissions.

CRediT authorship contribution statement

Jiao Ning: Conceptualization, Methodology, Validation, Formal analysis, Investigation, Writing – original draft, Writing – review & editing. **Shanning Lou:** Validation. **Yarong Guo:** Investigation. **Shenghua Chang:** Resources, Data curation. **Cheng Zhang:** Investigation, Supervision. **Wanhe Zhu:** Supervision. **Fujiang Hou:** Conceptualization, Writing – review & editing, Project administration, Funding acquisition.

Declaration of competing 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.

Acknowledgments

This research was supported by The National Natural Science Foundation of China (32161143028, U21A20242), the National Key Research and Development Program of China (2021YFD1300504), the Program of National Science and Technology Assistance (KY202002011), Innovative Research Team of Ministry of Education (IRT_17R50), and the Technological Support for Grassland Ecological Management and 'Lanzhou City's Scientific Research Funding Subsidy to Lanzhou University'.

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