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Effects of precipitation exclusion on N_2O emissions in a savanna ecosystem in SW China

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ABSTRACT

Savanna ecosystems play a crucial role in global N_2O emissions. However, our understanding of N_2O emissions under limiting precipitation conditions is lacking. This study evaluates the effects of precipitation reduction on soil N_2O fluxes from a woody savanna ecosystem in Yunnan Province, Southwest China. Precipitation exclusion shelters were installed above the tree canopy, and four total treatments were established as follows: a control (CK) and precipitation exclusions of 30% (PE3), 50% (PE5), and 70% (PE7). Two years (2015–2016) of N_2O fluxes, soil temperature and soil water content data were collected. The N_2O fluxes were generally low, ranging from 0.039 to 0.245 mg N m^{-2} day^{-1}, and they were strongly linked to precipitation events. Additionally, the N_2O fluxes during the rainy season were significantly greater than those during the dry season. The maximum N_2O flux was observed in August, and the minimum flux occurred in December. Precipitation exclusion had a significant negative influence on the N_2O fluxes. The N_2O emissions of CK, PE3, PE5, and PE7 were 0.20, 0.17, 0.13, and 0.12 kg N ha^{-1} yr^{-1}, respectively. With the exacerbation of precipitation exclusion, the decrease rate of precipitation exclusion on the N_2O emissions increased over the entire year (eventually reaching 41.8% in PE7), but the decrease rate of precipitation exclusion on the soil N_2O emission during the dry season was stronger than that during the rainy season. Additionally, the proportion of dry season N_2O emissions to total annual emissions decreased (from 45% to 41%), and that of rainy season N_2O emissions to total annual emissions increased (from 55% to 59%) over the year, whereas they exhibited a stable trend from PE5. The data show that the Yuanjiang savanna is a net source of N_2O; precipitation reduction decreases the N_2O emissions in the savanna regions, indicating that precipitation reduction can only slow the increase in the N_2O concentration in the atmosphere and can therefore slow global warming. In addition, the N_2O emissions during the dry season may play a significant role in total N_2O emissions and be more sensitive to precipitation reduction than those during rainy season. These possibilities should be considered in future studies, especially in those ecosystems that experience substantial inter-annual climatic fluctuations.

1. Introduction

The increasing global temperature and changing precipitation regimes associated with global climate change are expected to cause and exacerbate regional drought events, especially in mid-latitude and subtropical dry regions (IPCC, 2013). These events will likely alter soil nitrogen cycling processes and consequently affect the emissions of N_2O from soil (Davidson et al., 2008; Wieder et al., 2011). N_2O is one of the most important anthropogenic greenhouse gases, with a net greenhouse effect per unit mass that is approximately 320 times stronger than that of CO_2 over a 100-year span, and it contributes to approximately 6% of the currently observed global warming (IPCC, 2013; Rodhe, 1990). The atmospheric N_2O concentration is 322 ppb, which is 19% higher than preindustrial levels, and atmospheric N_2O has increased at a mean rate of 0.7 ppb yr^{-1} over the past 30 years (IPCC, 2013; Spahni et al., 2005). The increasing N_2O concentration may lead to feedback to climate change.

Natural N_2O sources are predominantly terrestrial ecosystems

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(Bouwman et al., 1993; Hirsch et al., 2006). Some studies have confirmed the importance of N_2O sources between the equator and 30°N and suggested possible emissions increase from this region over time (Galloway et al., 2008; Montzka et al., 2011). In soils, N_2O is a chemical by-product of nitrification and an obligatory intermediate by-product of denitrification (Davidson et al., 1986; Wrage et al., 2005). Under aerobic conditions, autotrophic and/or heterotrophic nitrifying organisms oxidize NH_4^+ via NH_2OH and NO_2 to NO_3^-. Under anaerobic conditions in the soil or in micro-sites, denitrifying organisms use NO_3^-/NO_2 as alternative electron acceptors. According to soil or micro-site environmental conditions, the NO_3^- is reduced stepwise via NO_2, NO and N_2O to N_2. The underlying environmental conditions determine the rates of nitrification and denitrification and thus the N_2O emissions from soils. Because the heterogeneity of soils as well as gas diffusion and metabolic activity can create anaerobic micro-sites even in well-aerated soil, nitrification and denitrification processes can occur simultaneously in the soil. Soil water status has been found to act as the primary driver of N_2O production and emissions, which primarily regulate the soil aeration conditions and the availability of NH_4^+ and NO_3^- as well as the contents of decomposable organic substrate, whereas soil temperature and pH are secondary controls (Baggs, 2008; Butterbach-Bahl et al., 2013). The precipitation magnitude and distribution patterns deeply affect the soil water status. Numerous studies have examined the response of soil N_2O emissions to precipitation reduction, but the results are inconsistent. Some studies have found that a lack of precipitation reduces N_2O emissions by decreasing the soil water content and net nitrification and nitrogen mineralization rates as well as the dissolved organic carbon (Davidson et al., 2004; Geng et al., 2017; Wood and Silver, 2012), whereas other studies have shown that precipitation reduction increases the soil content of dissolved organic carbon (as delivered from the litter to the soil under drier conditions) or the availability of nitrogen (due to root mortality and reduced plant uptake of nitrogen), thereby increasing N_2O emissions (Cattânio et al., 2002; Wieder et al., 2011). However, those studies primarily focused on tropical and subtropical forests and had one treatment without precipitation, which might cause uncertainty and confuse the effect of the precipitation reduction on the details. Additionally, the mechanism of the precipitation exclusion effects on N_2O emissions remains unclear. Therefore, soil N_2O emissions should be studied in detail under future climate change scenarios in various ecosystems.

Savanna ecosystems in tropical and subtropical regions cover approximately 16 million km^2, or an eighth of the total land surface (Scholes and Hall, 1996). Savannas are mixed ecosystems characterized by a grass understory with a tree/shrub overstory and distinct wet and dry seasons (Anderson et al., 2007; Grace et al., 2006). Savannas are thought to contribute to approximately 17% of the global N_2O production from terrestrial systems (Zhuang et al., 2012). However, relevant studies have been primarily conducted during the rainy season, and whether savanna soil is a source or sink of N_2O during the dry season remains uncertain. Some studies have found that the N_2O fluxes during the dry season in savanna soil are extremely low and often below the minimum detection limit. However even under extended dry and hot conditions, in nitrogen poor soils, soil microbial denitrifiers may exploit N_2O as a nitrogen substrate in the absence of NO_2^- and NO_3^-, leading to N_2O uptake (Cruvinel et al., 2011; Grover et al., 2012; Livesley et al., 2011; Verchot et al., 1999). Other studies have shown that N_2O fluxes in savanna soil are high and positive (from the soil to the atmosphere) during the dry season (Mapanda et al., 2012; Poth et al., 1995; Sanhueza et al., 1990). Considering the conflicting results and the reality of precipitation reductions, a precipitation exclusion experiment should be conducted in a savanna area and include the dry season to better understand the N_2O fluxes in savanna areas and the associated response to reduced precipitation.

Yuanjiang is a savanna ecosystem and located in a hot-dry valley. In this area, both the precipitation amount and frequency have decreased over the past thirty years (Chen et al., 2015; Fei et al., 2017). We established a precipitation-excluding roof above the canopy of a woodland savanna and set up three exclusion treatments to evaluate the effects of precipitation exclusion on N_2O emissions from the soil. Additionally, we determined whether the Yuanjiang savanna is an N_2O source during both the rainy and dry seasons. We hypothesized that precipitation exclusion would decrease N_2O emissions and that as the precipitation exclusion worsened, the N_2O emissions would stabilize and the soil would not change from an N_2O source to an N_2O sink.

2. Materials and methods

2.1. Experimental site

The study site is located at the Yuanjiang Savanna Ecosystem Research Station (YSERS; 23°27′N, 102°10′E; 551 m above sea level) of the Xishuangbanna Tropical Botanical Garden, Chinese Academy of Sciences, in Yunnan Province, Southwest China. The climate is dry and hot, the long-term (from 1980 to 2014) mean annual temperature at the site is 24.0 ± 0.5 °C, and the mean annual precipitation is approximately 795 mm, with 81% of the precipitation falling from May to October (rainy season). The yearly total number of sunshine hours is 2261.7, and the annual average pan evaporation is 2750 mm. The slope of the study plot terrain is ~15° and the soil is classified as torrid red earth (dry red soil) according to Chinese soil classification, which is equivalent to the ferralic cambisol (FAO classification) (Fei et al., 2017). The soil layer is shallow, not exceeding 20 cm depth in most areas.

The primary ecosystem in this area is woodland savanna, which is the most typical savanna ecosystem in China, and is dominated by the trees *Lannea coromandelica* and *Polyalthia cerasoides*, with frequent succulents such as *Euphorbia royleana* and ground cover of the grass *Heteropogon contortus*. The crown canopy is 5–6 m high (Editorial Committee for Vegetation of Yunnan, 1987; Jin and Ou, 2000).

2.2. Design of the experiment

The experimental conditions were established in March of 2014 and included control plots (hereafter CK) and precipitation exclusion plots, which were fenced to exclude all vertebrate herbivores. The precipitation exclusion design involved three treatments to: exclude 30%, 50%, and 70% of the precipitation, hereafter known as PE3, PE5, and PE7, respectively. Each treatment had three repetitions, and each plot measured 10 m × 10 m. Trenches were excavated around the perimeter of the treatment plots to reduce the potential lateral movement of soil water from the surrounding forest into the plots. Because the soil layer is shallow, the trenches were excavated to the bedrock. All the measurements reported here were collected at least 2 m from the trench edge to prevent edge effects. In each PE plot, 10 m × 10 m roofs were constructed from transparent polyethylene sheets above the tree canopy and supported by frames made of galvanized steel pipes. The polyethylene sheets transmitted over 85% of the photosynthetically active radiation. The roofs were sloped such that water drained into the gutters attached to their lower lips and was transported via a PVC pipe system (20 cm in diameter) to a ground well away from the experimental area (Jin et al., 2018).

2.3. N_2O flux measurements

N_2O fluxes were measured twice per month from January 2015 to December 2016 using the static opaque chamber method. A static chamber was placed in the middle of each of the 12 plots. This chamber was made of polyethylene plastic, had dimensions of 60 cm long by 32 cm wide by 25 cm high and was embedded 5 cm deep into the soil. A lid with a rubber O-ring was fastened to the chamber during sampling. All the chambers and lids were covered with reflecting film on the outside surface. Five 60 ml gas samples were collected and stored from each chamber at 0, 10, 20, 30, and 40 min using 100 ml plastic syringes. Before each sampling, 20 ml of gas was used to flush the gas pipeline (Yao et al., 2009; Zhou et al., 2016). The interval time between any two measurements was greater than ten days. The sampling time was 08:00–11:00 a.m. local time. Small plants, insects, and grasses were regularly and carefully removed from each chamber, but the litters were left in their natural state.

Samples of gas from the chamber that were collected using a syringe were analyzed immediately in the laboratory with a gas chromatograph (GC, Agilent 7890A, Agilent Technologies, CA, USA), which was equipped with an electron capture detector (ECD) at 330 °C according to the DN-Ascarite method with N_2 as the carrier gas and an ascarite filter to remove CO_2 from the samples. The N_2O samples were isothermally separated in two stainless steel columns (both with an inner diameter of 2 mm, and they were either 1 or 2 m long) packed with Porapak Q, 80–100 mesh, at 55 °C. Four standard N_2O samples (National Center for Standard Matters, Beijing, China) with concentrations of 350 ppbv were analyzed every 10 samples to assess the accuracy of the sample analysis (Zhou et al., 2016). Within this manual measurement system, the detection limit for N_2O was 4.5×10^{-3} mg N m^{-2} day^{-1}.

The N_2O flux was calculated using the following formula:

$$F = \frac{M}{V_0}\frac{P}{P_0}\frac{T_0}{T}H\frac{dc}{dt} \tag{1}$$

where F is the N_2O flux ($mg \cdot m^{-2} \cdot h^{-1}$); M is the molar mass of N_2O ($g \cdot mol^{-1}$); P_0 and T_0 are the air pressure and temperature of an ideal under standard conditions, respectively (1013.25 hPa and 273.15 K); V_0 is the molar volume of N_2O under standard conditions ($22.41\ l \cdot mol^{-1}$); H is the chamber height (m); P and T are the actual air pressure and temperature in the chamber, respectively (hPa and °C); and dc/dt is the initial slope of the relationship between the N_2O concentration and time, which was a linear relationship.

The N_2O emissions, or N in units of $kg \cdot ha^{-1} \cdot yr^{-1}$, were calculated as follows:

$$N = \sum_{m=1}^{12} F*d*k \tag{2}$$

where F is the N_2O flux ($mg \cdot m^{-2} \cdot day^{-1}$), m is the month, d is the day number of the month, and k is the conversion factor.

2.4. Measurements of the soil temperature and the soil water content

The soil temperature (ST, at 10 cm depth) was recorded using a digital thermometer (6310; Spectrum, IL, USA), and the soil water content (volumetric water content, v/v%) at 10 cm depth was measured using time-domain reflectometry probes (CS616, Campbell Scientific, Inc., Logan, UT, USA) and recorded with a data logger (CR800, Campbell Scientific, Inc., Logan, UT, USA).

2.5. Soil sampling and analysis

During the observation of soil N_2O fluxes, approximately 200 g soil samples were collected from a point close to the static chambers at a depth of 0–10 cm using a stainless-steel auger (5 cm in diameter) every three months starting from January 2015. After they were sampled, the soils were brought back to the laboratory immediately and passed through a 2 mm sieve to remove roots, gravel, and stones. All the soil analyses were completed within 1 week. The ammonia nitrogen (NH_4^+-N) and nitrate nitrogen (NO_3^--N) concentrations were determined from the 2 M KCl extraction liquid using a continuous flow autoanalyzer (AutoAnalyzer 3; Germany). Portions of each soil sample from under the same treatment were pooled to create a composite sample for analyzing microbial biomass nitrogen (MBN), microbial biomass carbon (MBC), dissolved organic carbon (DOC), and total dissolved nitrogen (TDN). The soil MBC and MBN were determined by the chloroform fumigation-extraction method (Vance et al., 1987). In detail, for each sample, three 7.0 g replicate samples were not fumigated, and three 7.0 g replicate samples were fumigated with ethanol-free chloroform for 24 h at 25 °C in a sealed incubator in the dark. After this fumigation, the chloroform was removed from the soils completely. Both the fumigated and unfumigated samples were extracted with 35 ml of freshly prepared 0.05 M K_2SO_4 and then capped and shaken at 300 rpm for 1 h. The suspensions were then centrifuged for 10 min at 5000 × g, and the supernatants were filtered through 0.45 μm nitrocellulose membrane filters (Pall Life Science Company, Beijing, China). These filtered samples were analyzed for their DOC and TDN concentrations using Pt-catalyzed high-temperature combustion (680 °C) and a total organic carbon/total nitrogen analyzer (LiquiTOC II, Elementar Analyzer System, Germany). The DOC and TDN were determined on the unfumigated filters, and the differences in DOC and TDN between the unfumigated and fumigated filters were used as the MBC and MBN, respectively (Zhou et al., 2016).

2.6. Data analyses

A two-way analysis of variance (ANOVA) was performed followed by a post-hoc Tukey test to determine the differences in gas fluxes, soil temperature and soil water content when considering the treatments and seasons as sources of variations. When the seasonal difference was significant ($P < 0.05$), a comparison was made using a two-independent-sample t-test. Stepwise regression analyses were conducted to determine the effects of the soil water content and the soil temperature on the N_2O flux.

The decrease rate of precipitation exclusion was calculated using the following function:

$$DR = \frac{N_{CK} - N_{PE}}{N_{CK}}*100\% \tag{3}$$

where DR is the decrease rate of precipitation exclusion, N_{CK} is the N_2O emission (kg N ha^{-1} yr^{-1}) in CK plots, and N_{PE} is the N_2O emission (kg N ha^{-1} yr^{-1}) in PE plots.

3. Results

3.1. Precipitation and the effect of exclusion

The average annual precipitation between 2015 and 2016 was 777 mm, of which 68.8% occurred during the rainy season. The average wettest month was July (117 mm), but rain fell during every month. Several large rain events occurred, including during the dry season; for example, from January 9, 2015 to January 11, 2015, it rained 66 mm (Fig. 1a).

The soil water content exhibited remarkable seasonal variations and was strongly linked to precipitation (Fig. 1b). Notably, the average soil water content in the CK plots was 16.4%. The precipitation exclusion decreased the soil water content during both the rainy and dry seasons, but the difference in the soil water content between PE5 and PE7 was not significant (Table 1).

The soil temperatures at 10 cm ranged from 11.9 to 34.9 °C, with a mean value of 25.2 °C, and remarkable seasonal variations were observed. Precipitation exclusion generally increased the soil temperature. The mean soil temperature measured in CK was comparable to the temperature measured in PE3 and slightly lower than the mean soil temperatures measured in PE5 and PE7 (Table 1).

Precipitation exclusion decreased the NH_4^+-N, DOC, TDN, MBC, and MBN during both the rainy and dry seasons, but significant differences were only found in NH_4^+-N and DOC during the rainy season (Table 2).

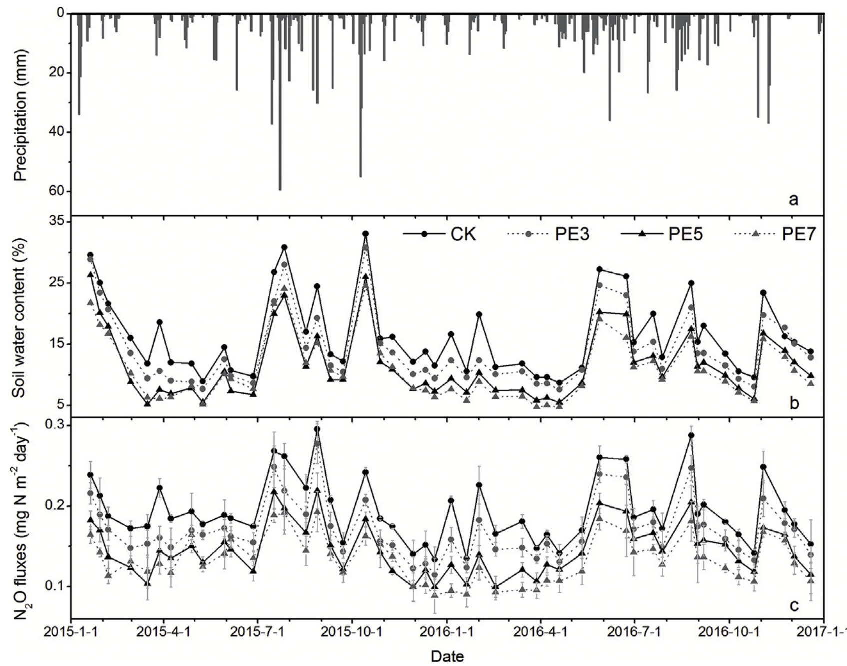

Fig. 1. Seasonal dynamics of ecological factors and N_2O fluxes in the Yuanjiang savanna ecosystem: a) precipitation, b) soil water content at 10 cm, and c) N_2O fluxes. The bars represent one standard error.

3.2. The dynamics of N_2O

The N_2O flux ranged from 0.039 to 0.245 mg N m^{-2} day^{-1}. The variation in the N_2O flux was like the variation in the soil water content and lagged changes in precipitation (Fig. 1).

Significant seasonal differences were observed in all the treatments ($P < 0.05$) (Fig. 2). In the CK plots, the N_2O fluxes during the rainy season (average flux: 0.157 ± 0.010 mg N m^{-2} day^{-1}) were higher than those of the dry season (average flux: 0.130 ± 0.006 mg N m^{-2} day^{-1}); the maximum monthly average N_2O flux (0.199 ± 0.010 mg N m^{-2} day^{-1}) was observed in August, and the minimum monthly average N_2O flux (0.104 ± 0.011 mg N m^{-2} day^{-1}) was observed in December.

The soil of the Yuanjiang savanna emitted 0.20 kg N ha^{-1} yr^{-1} as N_2O. The N_2O emissions during the rainy season totaled 0.11 kg N ha^{-1} yr^{-1} and accounted for 54% of the total emissions. The N_2O emissions during the dry season totaled 0.09 kg N ha^{-1} yr^{-1} and accounted for 46% of the total emissions (Fig. 3).

3.3. The effect of precipitation exclusion on the N_2O emissions

Precipitation exclusion had a significant negative influence on the N_2O fluxes in both the rainy season and dry season. The mean annual N_2O flux measured in the CK was significantly higher than the mean annual fluxes measured in the PE plots. Additionally, the fluxes measured in PE5 and PE7 were not significantly different (Fig. 2). However, no N_2O absorption was observed, even in PE7, during the dry season (Fig. 1c).

As the precipitation exclusion continued, the decrease rate of precipitation exclusion increased over the entire year (eventually reaching 41.8% in PE7), but that during the dry season had a stronger effect than that during the rainy season (Table 3). Additionally, with the exacerbation of precipitation exclusion, the dry season proportion of N_2O emissions decreased, and the rainy season proportion of N_2O emissions increased, and they remained stable in PE5 (Fig. 3).

3.4. Factors influencing N_2O fluxes

A pearson correlation analysis showed that the N_2O fluxes were significantly affected by the following soil factors: the NH_4^+-N ($R = 0.75$, $P < 0.01$), DOC ($R = 0.64$, $P < 0.01$), TDN ($R = 0.64$, $P < 0.01$), SW ($R = 0.54$, $P < 0.05$), MBC ($R = 41$, $P < 0.05$) and MBN ($R = 0.51$, $P < 0.05$). However, the correlations between N_2O fluxes and each of ST ($R = 0.33$, $P > 0.05$) and NO_3^--N ($R = 0.08$,

Table 1
Results of two-way analysis of variance of soil temperature and soil water content of the four treatments in the rainy and dry seasons.

Factors	Soil temperature (°C)		Soil water content (%)	
	Significance	Mean ± SE	Significance	Mean ± SE
Treatment	$P < 0.05$		$P < 0.05$	
CK		24.7 ± 0.15 a		16.4 ± 0.24 a
PE3		24.8 ± 0.14 a		13.9 ± 0.21 b
PE5		25.4 ± 0.16 b		11.2 ± 0.20 c
PE7		25.9 ± 0.15 b		10.5 ± 0.18 c
Season	$P < 0.05$		$P < 0.05$	
Rainy		28.1 ± 0.11 a		13.4 ± 0.26 a
Dry		22.3 ± 0.18 b		11.9 ± 0.22 b
Treatment × Season	$P > 0.05$		$P > 0.05$	

Different letters denote significant differences at the 0.05 level between the treatments or seasons.

Table 2
The effect of precipitation exclusion on soil nutrients during the dry and rainy seasons (Mean ± SE).

Season	Treatment	NH_4^+-N	NO_3^--N	DOC	TDN	MBC	MBN
Dry	CK	7.0 ± 1.0	1.9 ± 0.7	128.6 ± 22.5	34.9 ± 6.1	1149.8 ± 81.5	227.23 ± 32.9
	PE3	6.6 ± 1.1	2.0 ± 0.6	116.4 ± 15.9	28.3 ± 5.5	1088.3 ± 78.0	212.3 ± 23.5
	PE5	5.0 ± 1.4	2.0 ± 0.5	91.9 ± 8.8	23.3 ± 4.4	1050.3 ± 72.4	193.37 ± 13.2
	PE7	3.8 ± 1.5	2.1 ± 0.4	90.7 ± 12.9	21.5 ± 3.1	953.3 ± 37.0	170.89 ± 14.3
Rainy	CK	12.9 ± 0.9a	2.5 ± 1.1	204.4 ± 15.8a	47.3 ± 13.1	1114.9 ± 84.6	270.9 ± 21.7
	PE3	11.3 ± 0.7 ab	2.5 ± 0.5	191.5 ± 19.8 ab	44.1 ± 10.7	1041.5 ± 58.1	254.5 ± 20.5
	PE5	8.2 ± 0.6bc	2.5 ± 0.6	151.3 ± 4.0 ab	35.4 ± 5.2	1016.2 ± 70.5	219.32 ± 12.4
	PE7	7.5 ± 0.7c	3.3 ± 0.7	133.9 ± 7.5b	32.2 ± 2.5	881.22 ± 63.7	213.48 ± 21.4

NH_4^+-N is the ammonia nitrogen; NO_3^--N is the nitrate nitrogen; DOC is the dissolved organic carbon; TDN is the total dissolved nitrogen; MBC is the microbial biomass carbon; and MBN is the microbial biomass nitrogen.

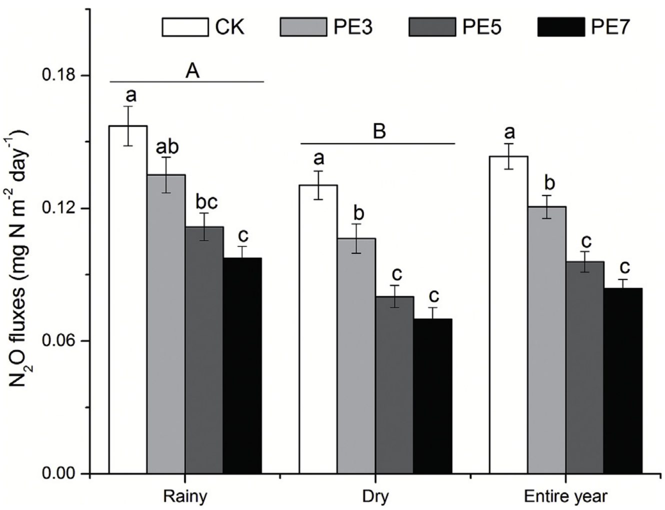

Fig. 2. Seasonal and treatment differences in mean daily nitrous oxide (N_2O) fluxes measured in the four treatments during the rainy season, dry season and entire year. The bars represent one standard error. Different capital letters denote a significant difference between seasons at the 0.05 level, and different lowercase letters denote a significant difference among treatments at the 0.05 level.

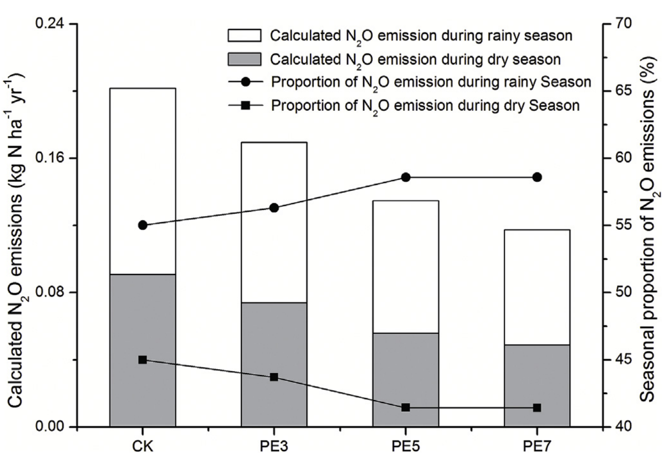

Fig. 3. N_2O emissions in the rainy season, dry season and entire year as well as the proportions of N_2O emissions in the rainy season and dry season that account for the total annual emissions.

Table 3
The decrease rate of precipitation exclusion on N_2O emission in the rainy season, dry season, and entire year.

	Dry season	Rainy season	Entire year
CK	–	–	–
PE3	18.52%	14.10%	16.09%
PE5	38.53%	28.97%	33.27%
PE7	46.38%	38.04%	41.79%

Decrease rate = $(N_{CK}–N_{PE})/N_{CK} \times 100\%$, N_{CK} is the N_2O emission in the CK plots, and N_{PE} is the N_2O emission in the PE plots.

Table 4
Pearson correlations between N_2O fluxes and soil factors.

	ST	SW	NH_4^+-N	NO_3^--N	DOC	MBC	MBN	N_2O
ST	1							
SW	−0.19	1						
NH_4^+-N	0.33	0.83**	1					
NO_3^--N	0.02	0.14	0.31	1				
DOC	0.63**	0.24	0.64**	0.10	1			
MBC	0.63**	0.52*	0.78**	0.07	0.92**	1		
MBN	0.56*	0.53*	0.84**	0.26	0.90**	0.93**	1	
N_2O	0.25	0.55*	0.75**	−0.21	0.64**	0.73**	0.75**	1

*Correlation is significant at the 0.05 level; **. Correlation is significant at the 0.01 level. ST is the soil temperature at 10 cm; SW is the soil water content at 10 cm; NH_4^+-N is the ammonia nitrogen; NO_3^--N is the nitrate nitrogen; DOC is the dissolved organic carbon; TDN is the total dissolved nitrogen; MBC is the microbial biomass carbon; and MBN is the microbial biomass nitrogen.

$P > 0.05$) were not significant (Table 4).

4. Discussion

4.1. N_2O fluxes from savanna ecosystems

The annual average N_2O flux in the Yuanjiang savanna was 0.143 ± 0.005 mg N m^{-2} day^{-1}, which is generally low compared with the values from areas of tropical rainforest (0.677 ± 0.084 mg N m^{-2} day^{-1}) and subtropical forest (0.957 ± 0.289 mg N m^{-2} day^{-1}) of similar latitude (Tang et al., 2006; Yan et al., 2008). This trend might have occurred because of the low precipitation and the high pan evaporation of Yuanjiang, which made the soil water content low in Yuanjiang, thereby prejudicing the production of N_2O.

Compared with the N_2O fluxes of other savannas, those of the Yuanjiang savanna were relatively high, potentially because Yuanjiang is an undisturbed woodland savanna located in a valley with a relatively high capacity for soil water retention and sufficient nutrient accumulation (Mapanda et al., 2012; Sanhueza et al., 1990). For example, compared with the soil NH_4^+-N of other savannas (Table S1), that of the Yuanjiang savanna was relatively high; moreover, a global data analysis showed that the N_2O fluxes exhibited a linear correlation with

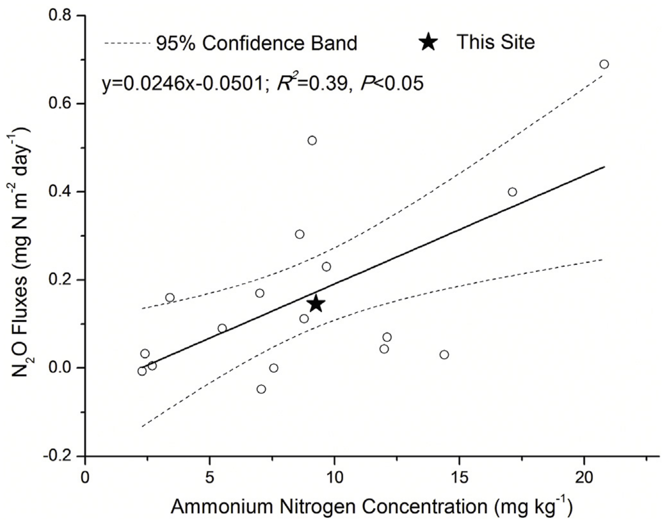

Fig. 4. Relationship between N_2O fluxes and ammonium nitrogen concentrations. The data are from 18 savanna ecosystems across the world.

the soil ammonia content ($P < 0.01$, $R^2 = 0.39$, Fig. 4). However, vegetation was regularly and carefully removed from each chamber during the experiment, which caused a reduction in canopy cover and an increase in ultraviolet radiation in the chambers. Lower canopy cover and higher ultraviolet radiation are conducive to litter decomposition (Baker and Allison, 2015; Gliksman et al., 2017). With the decomposition of litter, more nutrients enter the soil, which contribute to N_2O emission (Marhan et al., 2015). However, the ranges of Yuanjiang N_2O fluxes were below the range of -0.003–0.690 mg N m^{-2} day^{-1} observed for other savannas (Table S1). During our experiments, the high N_2O fluxes usually occurred after precipitation because the high soil water content was conducive to the formation of anaerobic micro-environments, and then it favored N_2O production by denitrification.

In other savannas during the dry season, some researchers have found the N_2O fluxes to be below the detection limit (Levine et al., 1996; Pinto et al., 2002) or even negative (Donoso et al., 1993; Grover et al., 2012; Livesley et al., 2011; Verchot et al., 1999). However, the average flux in the dry season was 0.130 ± 0.006 mg N m^{-2} day^{-1} in the Yuanjiang savanna, which is much higher than the values reported in other studies of savannas (Livesley et al., 2011; Poth et al., 1995; Verchot et al., 1999). The higher N_2O fluxes indicated that the Yuanjiang savanna might make a greater contribution to global warming than other savannas. The reason for this difference might be the increase in dry season precipitation. In our study, it rained every month during the dry season, and the precipitation during the dry season totaled 242 mm, accounting for 31% of the total precipitation. Additionally, this total was approximately 60% greater than the long-term average (from 1965 to 2014, accounting for 19% of the total precipitation); therefore, the soil water content remained at a relatively high level and benefited N_2O production. Additionally, the stimulatory effect of precipitation after the long rain-free period was marked and caused an N_2O pulse (Hao et al., 1988; Scholes et al., 1997; Werner et al., 2014). Those factors caused the N_2O emissions during the dry season to total 0.09 kg N ha^{-1} yr^{-1} and accounted for 46% of the total emissions.

Savanna systems experience substantial inter-annual climatic fluctuations. However, during the period of our experiment, the precipitation during the dry season was high. From 1965 to 2014, the number of years in which the dry season precipitation was equal to or greater than 242 mm (during our experiment) was 14, representing approximately 28% of the total number of years. Years in which the proportion of dry season precipitation relative to the entire year of precipitation was equal to or greater than 31% (during our experiment) occurred 12 times, or approximately 24% of the total number of years. Moreover, precipitation changes, including the frequency and intensity of precipitation events as well as the seasonal precipitation changes, have been predicted to become increasingly severe over the coming century in some regions of the world (IPCC, 2013). In view of the natural precipitation changes, N_2O emissions during the dry season should be considered when estimating the total N_2O emissions in savanna ecosystems, especially under changing precipitation patterns. In addition, we emphasize that long-term measurements should continue, with special emphasis on the dry season.

4.2. *Effects of precipitation exclusion*

Only a few precipitation exclusion experiments have been conducted, but they were not focused on savanna ecosystems (Table S2). In the Yuanjiang savanna, precipitation exclusion had a significant negative effect on N_2O emissions, which is similar to the effect of precipitation exclusion observed in some other ecosystems (Geng et al., 2017; Wood and Silver, 2012); however, this finding contradicts some studies that showed positive effects (Cattânio et al., 2002; Wieder et al., 2011).

Precipitation exclusion decreases the soil water content and increases the soil temperature. Unlike other precipitation exclusion experiments, our study was conducted in a savanna ecosystem characterized by low precipitation, intense radiation, and shallow soil layers, which led to a relatively high annual average soil temperature (24.7 °C at 10 cm) and a relatively low soil water content (16.4% in CK) (Table 1). Additionally, under normal conditions, both the soil water content and the soil temperature during the rainy season were higher than those during the dry season. We found that the soil water content and soil temperature had a common influence on the production of N_2O, according to the stepwise regression analysis (Table 5). Notably, the N_2O flux decreased with the decreasing soil water content and soil temperature. With the exacerbation of precipitation exclusion, the contribution of the soil water content to the N_2O flux decreased, and the contribution of the soil temperature to the N_2O flux increased, then remained stable in PE5. The higher temperature increased the size of most of the soil microbial colony-forming units, and it was conducive to the accumulation of NH_4^+-N and NO_3^--N (Shi et al., 2012). At 25 °C both the nitrification and microbial biomass reached their maximums, which are conducive to N_2O fluxes in savanna ecosystems (Castaldi et al., 2010). However, the contribution of the temperature was subordinate and only reached 20% (Table 5). The decrease in N_2O caused by the decrease in the soil water content was much larger than the increase in N_2O caused by the increase in the soil temperature; therefore, the N_2O flux decreased with continued precipitation exclusion.

Precipitation exclusion can influence the availability of nitrogen and DOC as well as the growth and activity of soil microorganisms. Because of precipitation exclusion, the nitrogen deposition decreased. Decreased nitrogen deposition from the atmosphere can lead to low N_2O emissions (Dalal and Allen, 2008; Werner et al., 2014). In

Table 5
Stepwise regression analysis results of the soil water content and soil temperature for N_2O fluxes.

Treatment	P	Vars. entered.	Delta R^2	Contribution (%)
CK	$P < 0.01$	SW	0.704	79.02
		ST	0.083	10.52
PE3	$P < 0.01$	SW	0.609	70.44
		ST	0.106	14.85
PE5	$P < 0.01$	SW	0.644	57.38
		ST	0.174	21.25
PE7	$P < 0.01$	SW	0.626	61.05
		ST	0.151	19.41

savannas, the combined effects of soil water availability, soil nitrogen and soil organic carbon content on microbial activity regulate the nitrification and denitrification processes, which are the major pathways of N_2O production (Brümmer et al., 2008; Castaldi et al., 2006, 2010; Grover et al., 2012; Rees et al., 2006). Yuanjiang is a savanna ecosystem that receives relatively little precipitation (795 mm yr^{-1}). Precipitation exclusion decreased the soil NH_4^+-N (Table 2), and a global data analysis showed that N_2O fluxes exhibited a linear correlation with the soil ammonia content ($P < 0.01$, $R^2 = 0.39$, Fig. 4). The decrease in the net nitrification and nitrogen mineralization rates inhibited the growth and activity of nitrifying bacteria and decreased the production of N_2O (Chen et al., 2017; Geng et al., 2017; Hartmann and Niklaus, 2012; Shi et al., 2012). In the meantime, precipitation exclusion decreased the DOC (Table 2). The DOC combined with the soil water content regulated the rate of denitrification (Wieder et al., 2011). The decrease in the DOC and soil water content might limit the production of N_2O from denitrification. Furthermore, precipitation exclusion also influenced the growth and activity of soil microorganisms. With the exacerbation of precipitation exclusion, the MBC and MBN decreased (Table 2), which indicated that the total amount of soil microorganisms decreased. In other ecosystems, a water-filled pore space (WFPS) of 60% has been considered the approximate 'tipping point' between nitrification (< 60%) and denitrification (> 60%) dominating the production of N_2O (Davidson, 1991; Müller and Sherlock, 2004). However, the strong correlation between N_2O flux and soil NH_4^+-N and the weak correlation between N_2O flux and soil NO_3^--N suggest that nitrification was predominantly responsible for N_2O production (Panek et al., 2000; Rees et al., 2006). During our observations, the WFPS values were less than 50%; moreover, the N_2O fluxes showed a significant correlation with the NH_4^+-N and no significant correlation with the NO_3^--N (Table 4), which indicated that nitrification might be the most important process regulating N_2O emissions in the Yuanjiang savanna ecosystem.

Precipitation exclusion may alter the growth of plants and the vegetation composition. In other experiments, short-term precipitation exclusion had no or insignificant effect on plant productivity or root biomass but resulted in a substantial reduction of net emissions of N_2O (Hartmann and Niklaus, 2012; Shi et al., 2012). In our experiment, it is possible that the two years of data were insufficient perhaps after long-term precipitation exclusion, the plant roots will die and decay, increasing the inputs of organic carbon to the soil and thereby increasing the production and emission of N_2O (Cattânio et al., 2002). However, in savanna ecosystems, precipitation reduction might serve to increase the competitive pressure exerted by trees on grasses (February et al., 2013), Compared with herbaceous savannas, tree savannas produce lower N_2O emissions (Castaldi et al., 2004), which causes the N_2O emission decrease.

With the exacerbation of precipitation exclusion, the decrease rate of precipitation exclusion during the dry season increased and the proportion of dry season N_2O emission decreased, which indicated that the dry season might be more sensitive to precipitation reduction than the rainy season. Compared with the rainy season, the dry season precipitation was lower. When the precipitation was reduced during the dry season, the soil water content was more easily decreased, limiting the activity of soil microorganisms for N_2O production. Under extended dry and hot conditions, soil microbial denitrifiers might exploit N_2O as a nitrogen substrate in the absence of NO_2^- and NO_3^- in nitrogen-poor soils. During our experiments, no negative N_2O fluxes were observed, even in the PE7 treatment in the dry season. This finding may have occurred because precipitation occurred during every month (Fig. 1a), which prevented the formation of extended dry and hot conditions. In addition, the soil NH_4^+-N in Yuanjiang was higher than the values reported was in other savannas (Fig. 4). Moreover, soil NH_4^+-N showed no significant difference among the different precipitation exclusion treatments during the dry season (Table 2), which provided the nitrogen substrate for N_2O production. However, the lack of negative N_2O indicated that Yuanjiang savanna soil could not absorb the N_2O.

N_2O is an important greenhouse gas and plays a significant role in global warming. Precipitation (or soil water content) is an important ecological factor regulating N_2O emission. Savanna is sensitive to precipitation change and produces more than 17% of the land-source N_2O. However, no studies on the effect of precipitation reduction on N_2O emissions in savanna ecosystems have been conducted. The Yuanjiang savanna represents the most typical type savanna in China and is located in a region where precipitation is decreasing according to the last thirty years of meteorological data. Our study provides insight into the N_2O emission of savanna ecosystem under precipitation reduction based on the actual measurement data and can serve as a reference for predicting N_2O emissions under precipitation reduction in other savanna ecosystems. The data show that precipitation reduction led to decreases in N_2O emission, which can slow the increase rate of N_2O concentration in the atmosphere. Considering the strong warming effect of N_2O, such decreases will slow global warming.

5. Conclusions

Effects of precipitation reduction on N_2O emissions were studied for two continuous years in Yuanjiang savanna ecosystem, the most typical savanna ecosystem of China, which was the first time in savanna ecosystem across the word. We found that Yuanjiang savanna emitted N_2O, precipitation reduction decreased N_2O emissions. The N_2O fluxes of this study were higher during the dry season than the values of other savanna ecosystems. The decrease rate of precipitation exclusion on N_2O emissions during the dry season was stronger than during the rainy season. Additionally, the N_2O emissions decreased as precipitation exclusion continued, but it remained stable from PE5, and no negative N_2O fluxes were observed. We conclude that the savanna may be a source of N_2O; the N_2O emissions during the dry season may play a vital role in total N_2O emissions, a possibility that should be considered in the future studies, especially in those ecosystems that experience substantial inter-annual climatic fluctuations. Additionally, precipitation reduction decreased N_2O emission from soil but did not change the soil from an N_2O source to an N_2O sink, which indicates that precipitation reduction slowed the increase in N_2O concentration in the atmosphere and can thereby help to slow the global warming.

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Appendix A. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.atmosenv.2018.05.035.

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