



## Nitrogen deposition induced significant increase of N<sub>2</sub>O emissions in an dry alpine meadow on the central Qinghai–Tibetan Plateau

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### ABSTRACT

Atmospheric concentrations of nitrous oxide (N<sub>2</sub>O) have increased over the last 150 years due to human activities. Soils are important source of N<sub>2</sub>O where its production is largely regulated by biological processes. Nitrogen (N) deposition can alter the processes of autotrophic nitrification and denitrification, thus affecting the rate and direction of the soil N<sub>2</sub>O exchange with the atmosphere. It is obvious that atmospheric N deposition has influenced a dry alpine meadow in the Qinghai–Tibetan Plateau (QTP). Therefore, we investigated the effects of N deposition on the annual N<sub>2</sub>O emissions in an alpine meadow of the QTP. The N<sub>2</sub>O flux was measured for 2 years using static chambers and gas chromatography methods at four treatments (N0, background level; N7, add 7 kg N ha<sup>-1</sup> yr<sup>-1</sup>; N20, add 20 kg N ha<sup>-1</sup> yr<sup>-1</sup>; N40, add 40 kg N ha<sup>-1</sup> yr<sup>-1</sup>). We found that high N deposition increased the N<sub>2</sub>O flux, not only in the growing season but also in winter and the spring thaw period. The average annual N<sub>2</sub>O fluxes at N0 and N40 plots were 3.1 and 6.1 μg m<sup>-2</sup> h<sup>-1</sup>, respectively. Compared with N0, the average annual N<sub>2</sub>O fluxes increased at N7, N20, and N40 plots by 13.7%, 47.6%, and 98.7%, respectively, the N<sub>2</sub>O fluxes in N40 plots increased by 113.6%, 41.6%, and 78.7% during the growing season, winter and spring thaw period, respectively. The emission of N<sub>2</sub>O during the growing season, winter and spring thaw period accounted for 63.2%, 29.9% and 6.9% of annual total emission of N<sub>2</sub>O, respectively. The N<sub>2</sub>O flux correlated significantly with air temperature, soil temperature, soil water content, total nitrogen, total organic carbon, and soil NH<sub>4</sub><sup>+</sup>-N content in the alpine meadow.

### 1. Introduction

Nitrous oxide (N<sub>2</sub>O) is one of the major greenhouse gases (IPCC, 2013). It is long-lasting, with an atmospheric residence time of 150 years and accounts for 7% of the current anthropogenic greenhouse effect (Peng et al., 2011). The atmospheric concentration of N<sub>2</sub>O has increased significantly since preindustrial times, with the anthropogenic perturbation of the global N cycle (Wolf et al., 2010). From a value of about 270 ppb at the beginning of the industrial era to 319 ppb in 2005, it is still increasing by 0.2%–0.3% per year (IPCC, 2013). At the same time, N deposition dramatically increased in the terrestrial ecosystem, and continues to increase because of the economy in the following years (Jiang et al., 2010). The average annual N deposition increased by approximately 8 kg of N per hectare between the 1980s and the 2000s in China (Liu et al., 2013a). Grassland soils cover approximately 25% of the total global terrestrial area, and therefore play

an important role in global N<sub>2</sub>O emissions (Du et al., 2006). The N<sub>2</sub>O emissions from grassland soils are affected by the atmospheric N deposition (Jiang et al., 2010; Peng et al., 2011; Li et al., 2012).

Several studies have investigated the effects of N deposition on N<sub>2</sub>O emissions in different grassland types, including semiarid temperate steppes (Peng et al., 2011), alpine meadows (Jiang et al., 2010; Zhu et al., 2015; Zhao et al., 2017), and alpine steppes (Wei et al., 2014; Zhao et al., 2017), the conclusions drawn differed for different grassland types. In many studies, N deposition generally increases the emission of N<sub>2</sub>O (Peng et al., 2011; Li et al., 2012; Wei et al., 2014). Peng et al. (2011) reported that the N<sub>2</sub>O flux increased with increasing N input on semiarid temperate steppes in Inner Mongolia, but did not increase after the N input reached a certain level. Soil moisture and the availability of soil carbon regulated the microbial processes of autotrophic nitrification and denitrification when the N supply was sufficient for the two processes in this area (Chen et al., 2013). However, the

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study by Li et al. (2012) in an alpine grassland in the southern Tianshan Mountains showed that the addition of N increased N<sub>2</sub>O emissions, although there were no significant differences in N<sub>2</sub>O emissions among the different N input treatments, and Zhu et al. (2015) suggested that N deposition had no effect on the N<sub>2</sub>O flux in sub-humid upland soils in northern China, because nitrification and denitrification were limited by low temperatures and low soil moisture in these two ecosystems (Ju et al., 2009). Therefore, the differences between studies are thought to arise from the different site-specific hydrothermal conditions, N deposition rates, and soil nutrients, because they control the dominant microbial processes leading to N<sub>2</sub>O emissions from soils (Hellebrand et al., 2003; Simona et al., 2004; Peng et al., 2011).

Although N<sub>2</sub>O emissions from natural grasslands have been investigated worldwide (Schulze et al., 2010; Vilain et al., 2010), most measurements have been made using short-term studies and only during the growing season (Jiang et al., 2010; Dijkstra et al., 2013; Wei et al., 2014; Li et al., 2015; Zhu et al., 2015). Only a few studies of N<sub>2</sub>O emissions have observed natural grasslands throughout the year, including the winter and spring thaw period (Soussana et al., 2007; Wolf et al., 2010; Li et al., 2012). Wolf et al. (2010) reported that N<sub>2</sub>O emissions during the spring thaw period dominated the total annual N<sub>2</sub>O emissions, which accounting for 72% of the annual emissions on continental steppes. However, some studies have reported that the contribution of the N<sub>2</sub>O emissions during winter and spring thaw period to the total annual N<sub>2</sub>O budget was small (Peng et al., 2011; Li et al., 2012), although N deposition increased N<sub>2</sub>O emissions during the spring thaw period (Peng et al., 2011). Measurements made during the winter and in the spring thaw period in grassland ecosystems have shown that the N<sub>2</sub>O flux can be very important in the calculation of the annual budget (Wolf et al., 2010; Yao et al., 2010; Li et al., 2012).

The QTP, a 2.5 million km<sup>2</sup> region dominated by alpine grassland ecosystems (Gao et al., 2014), covers nearly one-quarter of the area of China and represents one of the largest alpine grasslands in the world (He et al., 2006; Ganjurjav et al., 2016). The QTP characterized as high altitude, low temperature (Qiu, 2008; Zhang et al., 2012), and low soil N (Dong et al., 2004). Dry alpine meadow is the dominant ecosystem on the QTP, and it is very sensitive to the global change (Wang et al., 2014). The alpine meadow is an important base of animal husbandry and supports domestic grazing by yak (*Bos grunniens*), as well as endangered ungulates including Tibetan antelope (*Pantholops hodgsoni*) and wild yak (Ganjurjav et al., 2016). Alpine meadow ecosystem is a main N<sub>2</sub>O source on the QTP, and significantly affected by environmental changes (Wei et al., 2014; Li et al., 2015; Zhao et al., 2017). The plateau is experiencing the increase of atmospheric N deposition rates because of industrial and traffic sources in south Asia transported by Indian monsoon (Liu et al., 2013a, 2015).

However, the N<sub>2</sub>O emissions during the winter and spring thaw period in the alpine meadows on the QTP have not been reported. The response of N<sub>2</sub>O emissions to N deposition in the spring thaw period and winter and the contributions of these periods to the total annual N<sub>2</sub>O budget are unclear. To better understand the effects of N deposition on N<sub>2</sub>O emissions, and especially the role of the alpine meadow ecosystem in the global N<sub>2</sub>O budget during future increase in N deposition, four N treatments were established in the field between May 2015 and April 2017 in an alpine meadow on the central QTP. In this study, we focused on the following two scientific questions: (1) Based on consecutive measurements, how does nitrogen deposition affect the N<sub>2</sub>O flux? (2) How do the growing season, winter, and the spring thaw period contribute to the total annual N<sub>2</sub>O budget during nitrogen deposition in alpine meadows?

## 2. Materials and methods

### 2.1. Study site and experimental design

The research area is located in Nagqu County, Nagqu Prefecture,

Tibet Autonomous Region, China (31.441°N, 92.017°E; 4500 m above sea level). During the last sixty years, the mean annual temperature is −1.2 °C. The annual precipitation is 431.7 mm with more than 90% of the annual rainfall occurring during the growing season (May–September). The annual sunshine (total hours) is 2789.9 h (Ganjurjav et al., 2015). Dry alpine meadow is the main grassland type in this area. The sedges *Kobresia pygmaea* and the grass *stipa purpurea* are the main graminoids, and *Potentilla acaulis* and *Oxytropis ochrocephala* are the main forbs. The soil bulk density at 5 cm depth was 1.01 g cm<sup>−3</sup>. The experimental field was grazed by yak before the experiment and fenced in August 2010. The site was not grazed or mowed during the whole experimental period.

The local ambient N deposition is estimated to be range from 6.96 to 7.55 kg N ha<sup>−1</sup> yr<sup>−1</sup> (Lü and Tian, 2007) with the main forms being NH<sub>4</sub>-N and NO<sub>3</sub>-N (Liu et al., 2013a). We setup 1, 3 and 6 times amount of N deposition to simulated future climate change scenario, and initiated the N deposition experiment in May 2014 with four replicates of each of four treatments: N0 plots (normal; background level), N7 plots (add 7 kg ha<sup>−1</sup> yr<sup>−1</sup>; Low-N level), N20 plots (add 20 kg ha<sup>−1</sup> yr<sup>−1</sup>; Medium-N level) and N40 plots (add 40 kg ha<sup>−1</sup> yr<sup>−1</sup>; High N-level), with a total of 16 plots (each 3 × 3 m with an 1 m wide buffer zone). NH<sub>4</sub>NO<sub>3</sub> was used to simulate N deposition during the growing seasons. The added N was sprayed onto the N deposition treatment plots monthly (May–August), while N0 plots received the same amount of water (about 0.5 mm per month). General characteristics of the different treatments are shown in Table 1.

### 2.2. Environmental variables

Meteorological data were recorded at a weather station in Nagqu County. We used the EM50 data collection systems (Decagon Devices, Inc., NE, USA) with temperature sensor (ECT) and moisture sensor (EC-5) to measure soil temperature and soil moisture at depth of 5 cm. Data were collected at 30-min intervals from May 2015 to April 2017. On the basis of soil volumetric moisture content as well as soil bulk density that we calculated percent water filled pore space (WFPS) as follows Eq. (1) (Peng et al., 2011):

$$WFPS = \text{volumetric moisture content} \times \frac{100}{(1 - \text{bulk density}/2.65)} \quad (1)$$

### 2.3. Plant and soil measurements

We measured the aboveground biomass at peak biomass in mid-August in 2015 and 2016. We set 30 calibration plots (0.5 × 0.5 m<sup>2</sup>) adjacent to the experimental plots. We recorded the cover and height of each plant species in both the experimental plots and the calibration plots. The plants in the calibration plots were cut, sorted, and placed in envelopes. The envelopes were then placed in a drying oven for 30 min at 105 °C, after which the temperature was maintained at 70 °C until a constant weight was reached, then weighed. We calculated the total cover and mean height of graminoids and forbs. Then we applied a nondestructive method, based on linear regression between aboveground biomass and total cover and mean height of graminoids and forbs in calibration plots to estimate the total aboveground biomass in the experimental plots (Xia et al., 2009), and aboveground biomass was calculated as follows:

$$AB_{2015} = \frac{0.41C_g + 0.82C_f + 4.77H_g + 9.06H_f - 18.77}{10^3}$$

$$AB_{2016} = \frac{0.25C_g + 1.77C_f + 9.95H_g + 3.05H_f - 33.26}{10^3}$$

Where  $AB_{2015}$  and  $AB_{2016}$  were aboveground biomass in 2015 and 2016, respectively (Kg m<sup>−2</sup>).  $C_g$  and  $C_f$  were covers of graminoids and forbs, respectively (%).  $H_g$  and  $H_f$  were mean height of graminoids and

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