

The role of maize plants in regulating soil profile dynamics and surface emissions of nitrous oxide in a semiarid environment

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Abstract To study the effects of maize plants on nitrous oxide (N₂O) fluxes from a dryland experimental farm, in situ soil profiles of N₂O concentrations and surface emissions were investigated in a field experiment from 2014 to 2015 in the semiarid areas of northwestern China. The experiment included four treatments: unplanted and N-unfertilized soil (C0), unplanted soil fertilized with 225 kg N ha⁻¹ (CN), maize-planted and N-unfertilized soil (P0), and maize-planted soil fertilized with 225 kg N ha⁻¹ (PN). Surface N₂O emissions and soil N₂O concentrations at depths of 0, 10, 20, 30, 40, and 50 cm were measured weekly. Nitrogen fertilization significantly increased the soil N₂O concentrations and surface emissions. Compared to the unplanted soil, the presence of maize plants significantly decreased the N₂O concentrations at depths of 10–40 cm during the maize growing season. The modeled N₂O fluxes at a depth of 10 cm presented a similar pattern to the chamber measurements. However, there was a discrepancy between the concentration gradient and chamber methods when the fluxes were high, mainly because the

gradient method could not detect N₂O production and consumption process above the uppermost gas sampler (0–10 cm). Soil moisture and temperature were critical factors affecting the N₂O concentrations and surface emissions. The respective cumulative surface emissions and effluxes at a depth of 10 cm during the maize growing season (PN treatment) were decreased by 8.9 and 17.9% in 2014 and by 14.7 and 17.5% in 2015 compared to values of the CN treatment. This was mainly due to the decrease in soil moisture caused by the growth of the maize plants, which resulted in a soil condition less suitable for N₂O production.

Keywords Greenhouse gas · Rainfed agriculture · Soil profile · N₂O diffusion · Gradient method

Introduction

Nitrous oxide (N₂O) is a greenhouse gas that plays a key role in global warming and climate change. The global warming potential of N₂O over 100 years is 263 times higher than that of carbon dioxide (CO₂) (Neubauer and Megonigal 2015). Moreover, N₂O has also become the single most important ozone-depleting substance in the twenty-first century (Ravishankara et al. 2009).

Nitrous oxide is produced in soil during biological nitrification and denitrification processes as well as during coupled nitrification and denitrification (Baggs 2011; Bremner 1997), which are strongly affected by interdependent controlling factors: soil moisture, temperature, oxygen (O₂) content, nitrogen (N) availability, and carbon (C) availability (Ju et al. 2011; Liu et al. 2017; Philippot et al. 2009). The primary source of increasing N₂O in the atmosphere is the use of N fertilizers on agricultural soil worldwide (Smith 2017). Soil surface N₂O flux represents the integrated production, consumption, and

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transport of N_2O within the soil as well as the transfer across the soil surface to the atmosphere rather than a gross estimate of N_2O production (Goldberg et al. 2008). However, soil surface N_2O fluxes provide little information about belowground N_2O dynamics. Because N_2O consumption in soil is usually masked by the higher rates of N_2O production (Chapuis-Lardy et al. 2007), investing the production and consumption of N_2O is helpful to quantitatively understand the relative importance of process of N_2O production and consumption in soils. The soil concentration gradient method, which assumes that gas transport in the subsoil and between the soil and atmosphere is primarily driven by diffusive transport in the soil, has been widely applied in studies on in situ subsoil N_2O flux (Nan et al. 2016; Wolf et al. 2011; Yoh et al. 1997; Zhou et al. 2016). Because the N_2O produced in subsoil could be reduced to dinitrogen (N_2) when it diffuses upwards (Chapuis-Lardy et al. 2007; Gao et al. 2014; Kellman and Kavanaugh 2008), some studies have indicated that there was a disagreement between the estimated flux derived by the concentration gradient method and the measured N_2O emission (Pihlatie et al. 2007; Wolf et al. 2011). Therefore, combining the subsoil N_2O fluxes and surface emissions could yield more information on the depth of the N_2O production and consumption and the mechanisms responsible for these processes (Kellman and Kavanaugh 2008; Pihlatie et al. 2007).

Plants are critical determinants of microbial N_2O production as a result of their impact on soil moisture, temperature, aeration, labile organic C, and mineral N content (Hayashi et al. 2015; Meier et al. 2016; Philippot et al. 2009). However, there is no consensus as to whether plants stimulate or inhibit N_2O emissions. Recently, Hayashi et al. (2015) reviewed the effects of upland crops on N_2O emissions and found that planted soils emit approximately twice as much N_2O as unplanted soils with the same N rate, and the likely reason was stimulated heterotrophic denitrification as a result of root exudations. However, Jamali et al. (2016) found that N_2O emissions were 6.2 and 2.4 times higher from fallow clay loam and loam cores, respectively, compared to cores planted with wheat. This finding was attributed to the uptake of soil water and N by the plants. However, the effect of plants on soil N_2O emissions from dryland soils has rarely been investigated. In dryland regions, soil water and N are usually limited due to low precipitation and high evaporation (Delgado-Baquerizo et al. 2013). On the one hand, plants may increase N_2O emissions through root-derived labile C (Ni et al. 2012; Sey et al. 2010). On the other hand, plants consume water, compete for N with microbes, and decrease soil temperature through canopy shading, which create a soil condition less favorable for N_2O production (Ding et al. 2007; Jamali et al. 2016). In addition, plants could create anaerobic conditions in the rhizosphere due to root and rhizomicrobial respiration (Philippot et al. 2007) and stimulate N_2O reductase by releasing root exudation (Henry et al. 2008). These processes may have an

impact on the production and consumption of N_2O from denitrification. The stimulatory or suppressive effects of plants on N_2O fluxes will influence the overall N_2O emission budget in the plant-soil systems. Thus, further investigation of the role of plants in regulating N_2O fluxes from dryland farming regions is important for understanding the underlying mechanisms and the complex controls of greenhouse gas fluxes from agricultural ecosystems.

Spring maize is one of the most widely grown cereals (Liu et al. 2009) in the semiarid regions of northwestern China. High yield levels of maize ($> 14 \text{ Mg ha}^{-1}$) can be obtained with high inputs of N fertilizer (Bu et al. 2014), but the accompanied N_2O emissions should not be ignored. It is imperative to reduce greenhouse gas emissions while ensuring crop production. Maize plants, characterized by large and extensive root systems (Amos and Walters 2006), are integral components of agricultural systems that might affect the production and emission of N_2O by releasing root-derived organic C and altering soil environment (e.g., soil moisture, temperature). Therefore, understanding the effects of maize plants on N_2O emissions from dryland regions would improve our comprehensive understanding of the systematic process of N_2O emissions through plant-soil interaction. In order to accomplish this, we performed a 2-year field experiment to combine the measurements of N_2O emissions and subsoil N_2O concentrations at a depth of 0–50 cm from a dryland experimental farm in a region of northwestern China. The N_2O fluxes at the soil surface and in subsoil were measured and calculated by chamber and concentration gradient methods, respectively. The objectives of this experiment were (1) to determine the effect of maize plants on N_2O emissions and vertical dynamics within soil profiles, (2) to investigate the production and reduction of N_2O in subsoil by coupling the N_2O fluxes within soil profiles and at the surface, and (3) to identify the soil variables that affect subsoil N_2O concentrations and surface emissions. Our hypothesis was that the presence of maize plants would decrease N_2O emissions by decreasing soil water, temperature, and available N and therefore result in soil conditions less suitable for N_2O production in soil.

Materials and methods

Site description

The Changwu Agri-Ecological Station (35.28° N, 107.88° E, 1200 m altitude) is located on the Loess Plateau, China. The site is characterized by a semiarid continental climate. The annual mean air temperature is 9.1 °C. The average annual precipitation is 584 mm (73% rainfall during May–September), and the annual potential evaporation is 1560 mm. The major local cereal crops are winter wheat (*Triticum aestivum* L.) and spring maize (*Zea mays* L.), and

the dominant cropping system is one harvest a year. Agricultural production in this region depends on natural rainfall. The soil is a loam (Cumulic Haplustoll, USDA Soil Taxonomy System) developed from wind-deposited loess, with high permeability. The experimental field had been cultivated with winter wheat or spring maize for a long time prior to this experiment. Soil properties in the top 20 cm were bulk density 1.3 g cm^{-3} , pH 8.2, soil organic matter 14.2 g kg^{-1} , total N 1.0 g kg^{-1} , NO_3^- 19.6 mg kg^{-1} , exchangeable NH_4^+ 2.0 mg kg^{-1} , available phosphorous (Olsen) 21.5 mg kg^{-1} , and available potassium ($\text{NH}_4\text{OAc-K}$) 147.8 mg kg^{-1} . These analyses concern soil sampled in April 2014. A detailed description of the basic physical and chemical properties of soils in the 0–50 cm layer can be found in Yao et al. (2017).

Field experiments and crop management

The N_2O concentration was monitored in a field experiment organized in a completely randomized block design with three replicates, isolated by 1-m walkways. Each plot area was 56 m^2 ($7 \text{ m} \times 8 \text{ m}$). The field experiment started in 2014 and four treatments were setup: unplanted without N (C0), unplanted soil with 225 kg N ha^{-1} N (CN), maize-planted without N (P0), and maize-planted with 225 kg N ha^{-1} N (PN). Urea (N 46%) was applied in two splits in a ratio of 1:2: 75 kg N ha^{-1} as basal fertilizer and 150 kg N ha^{-1} as supplemental fertilizer. Calcium superphosphate (40 kg P ha^{-1}) and potassium sulfate ($80 \text{ kg K}_2\text{O ha}^{-1}$) were applied in all treatments. Before sowing, all mixed fertilizers were manually broadcast over the soil surface then tilled into the soil. The remaining 150 kg N ha^{-1} was applied using a hole-sowing machine during the 12th leaf stage (V12 stage, July 5, 2014 and July 3, 2015). Spring maize (var. *Pioneer 335*) was sown (April 30, 2014 and April 26, 2015) to a depth of 5 cm using a hand-powered hole-drilling machine and harvested on September 18, 2014 and September 13, 2015. The density was $65,000 \text{ plants ha}^{-1}$ and the distances between adjacent rows and hills were 50 and 30 cm, respectively. All plots were manually weeded periodically during the sampling period.

Sample collection and measurements

N₂O concentrations in the soil

Soil gas samples at depths of 10, 20, 30, 40, and 50 cm were collected with multiple sampling tubes (for more details, see Yao et al. (2017)). The sampling tubes were composed of five individual chambers which were isolated by PVC plates. Sixteen small holes were distributed in the lower section of each sampler and were covered by nylon mesh (0.038 mm). Each gas sampler was connected to the soil surface via an organic glass tubule (inner diameter 0.4 cm), and the end of the tubule was fitted with a three-way stopcock that allowed

collecting of subsoil gases at the soil surface. The three-way stopcocks were turned on only when sampling. The multiple sampling tubes were inserted into the holes made by a soil auger (inner diameter 5.0 cm), and the space between the tubes and the soil was back-filled in the original order. Soil gas sampling systems were installed at the center of each plot prior to planting and remained there during the measurement period. For the maize-planted treatments, gas sampling tubes were installed between the maize rows. Soil profile gas samples were collected between 8:30 a.m. and 11:30 a.m. Measurements were made weekly during the maize growing season and fortnightly or monthly during the fallow season. Each time, the gas samples in the sampling tubes and in the soil surface air (0 cm) were manually withdrawn using 20-mL syringes fitted with three-way stopcocks. In order to mix the air inside the samplers, syringes were extracted and purged slowly three times before collecting gas.

Chamber-based N₂O flux measurements

The surface N_2O emissions were measured manually using the closed static chamber method. Each stainless steel chamber consisted of a top chamber ($50 \times 30 \times 30 \text{ cm}$) and a base frame ($50 \times 30 \times 15 \text{ cm}$). The upper chamber had a $10 \times 10\text{-cm}$ hole at the center and was composed of two separate parts that were combined using hinges and airtight rubber seals. The bottoms of each part were also covered with airtight rubber seals. The upper chamber was covered with Styrofoam coating to minimize fluctuations of air temperature in the chamber during the sampling period. One fan was installed inside the chamber to promote the mixing of air. The frames were inserted into the soil to a depth of 15 cm, and one maize plant was placed in the center of the frame area. To collect gas samples, the top chambers were temporarily placed on the frames, which were combined closely by two clamps. The hole in the upper chamber was used to allow the maize plant to pass through the chamber top when the stalk was too high. Two pieces of soft polyethylene foam and preservative films ($1.2 \mu\text{m}$ thick) were used to seal the gap between the maize stalk and the hole in order to reduce the leakage of gas. Gas samples were taken using 50-mL syringes 0, 10, 20, and 30 min after enclosure. Surface flux samples were collected concurrently with the soil profile gas samples.

All the gas samples (within soil profile and of surface fluxes) were analyzed using a gas chromatograph (GC, Agilent 7890A, Shanghai, China) equipped with an electron capture detector (ECD). Gas samples analyses were done within 12 h on the sampling day. The ECD and column of the GC were set at 350 and $60 \text{ }^\circ\text{C}$, respectively. The carrier gas of GC was N_2 (99.999% purity). The N_2O fluxes were calculated from the linear increase in the concentration in the chamber.

Environmental and soil variables

The daily precipitation and daily mean air temperatures at 1.5 m above the soil surface were recorded by the nearby Changwu Meteorological Monitoring Station (within 50 m). Soil temperatures at depths of 10, 20, 30, 40, and 50 cm in each plot were measured using portable digital thermometers (JM624, Jinming Instrument Ltd., Tianjin, China) simultaneously as sampling gas. Soil samples in the layers of 0–10, 10–20, 20–30, 30–40, and 40–50 cm were collected to determine moisture weekly during the maize growing season and fortnightly or monthly during the fallow season. Soil samples in the top 20 cm were taken to determine the soil moisture and mineral N concentration. Soil moisture was not measured during the period of soil freezing (December to early March in the next year). The soil samples were oven-dried at 105 °C to a constant weight to determine soil gravimetric water content. Water-filled pore space (WFPS) was calculated by dividing the volumetric water content by total soil porosity. A 5.0-g sub-sample was used to determine mineral N (exchangeable NH_4^+ + NO_3^-) concentration extracted with 50 mL 1 M KCl solution, and the extracts were analyzed by colorimetric analysis on an automated flow injection analyzer (FLOWSYS, Italy). Soil available P was extracted with 0.5 mol L⁻¹ sodium bicarbonate at pH 8.5 and determined with a colourimetric method (Watanabe and Olsen 1965). Available K was extracted with 1.0 mol L⁻¹ ammonium acetate solution (pH 7.0) and measured with a flame photometer (Helmke and Sparks 1996). Soil organic C was analyzed by the dichromate oxidation method (Mebius 1960), and total N was analyzed by the Kjeldahl method (Bremner and Mulvaney 1982). Soil pH was measured with a standard combination electrode in a 1:2.5 soil-to-water ratio. Soil bulk density at the 0–50-cm soil depth, at an interval of 10 cm, was measured by the cutting-ring method (100 cm³) in the field before sowing and after harvesting in each year. In addition, measurements were taken from the 0–10- and 10–20-cm layers monthly after sowing for 2 months in both years.

Plant biomass sample

At the silking stage (July 18, 2014 and July 20, 2015), three adjacent plants in a row in each plot were cut to determine the leaf area index (LAI). The LAI was calculated as follows: $\text{LAI} = \text{leaf area (m}^2 \text{ plant}^{-1}) \times \text{population (plants ha}^{-1}) / 10,000 \text{ (m}^2 \text{ ha}^{-1})$. The leaf area was calculated using the following formula: $\text{Leaf area} = \text{leaf length} \times \text{maximum leaf width} \times 0.75$ (McKee 1964). Leaf length and maximal width of all green leaves were measured manually. At the physiological maturity stage, the grain yields were estimated for all of the plants selected from a 10-m² area in each plot. The leaf, stem, bract, ear axis, and grain were oven-dried at 105 °C for 30 min initially and then at 65 °C to constant weight. After

weighed, samples were ground for chemical analysis. Total N in plant was analyzed using the Kjeldahl digestion method (Bremner and Mulvaney 1982).

Calculations and statistical analyses

N₂O diffusive flux in the soil profile

The N₂O effluxes within soil profiles were calculated using the following equation based on Fick's law (Marshall 1959):

$$q = -D_p \frac{d_c}{d_z} \quad (1)$$

where q is the gas flux (g m⁻² s⁻¹), positive values are defined as gas moving upward, and negative values as moving towards deeper layers. D_p is the effective diffusion coefficient of N₂O in the soil (m² s⁻¹), and $\frac{d_c}{d_z}$ is the concentration gradient between two adjacent soil layers (g m⁻³ m⁻¹). D_p was calculated using the following model (Allaire et al. 2008):

$$D_p = D_0 \left(\frac{\varepsilon^{2.5}}{\Phi^2} \right) \quad (2)$$

where D_0 is the N₂O diffusivity in free air (m² s⁻¹) and ε and Φ are the soil air-filled porosity (m³ m⁻³) and total soil porosity (m³ m⁻³), respectively. The parameters ε and Φ were calculated using the Millington-Quirk model (Millington and Quirk 1961):

$$\Phi = 1 - \frac{\rho_b}{\rho_s} \quad (3)$$

$$\varepsilon = \Phi - \theta_v \quad (4)$$

$$\theta_v = \theta_m \times \rho_b \quad (5)$$

where ρ_b is the soil bulk density (g cm⁻³) and θ_v and θ_m are the soil volumetric water content (m³/m³) and gravimetric water content (g/g), respectively. D_0 is influenced by air temperature and pressure:

$$D_0 = D_s \left(\frac{T}{T_0} \right)^{1.75} \left(\frac{P_0}{P} \right) \quad (6)$$

where T is the temperature (K), P is the air pressure (Pa), and D_s is the D_0 at reference temperature T_0 (273.15 K) and reference barometric pressure P_0 (1 atm), given as 1.43 × 10⁻⁵ m² s⁻¹ (Pritchard and Currie 1982). For the N₂O effluxes in the subsoil, we used the bottom depth below each layer (10, 20, 30, 40, and 50 cm) representing the whole soil layers (0–10, 10–20, 20–30, 30–40, and 40–50 cm) in the following tables and figures for convenience.

Cumulative gas effluxes

The cumulative emissions were calculated using the following formula:

$$T = \sum_{i=1}^n (X_i + X_{i+1}) / 2 \times (t_{i+1} - t_i) \times 24 \quad (7)$$

where T (g N ha^{-1}) is the cumulative N_2O flux, X ($\text{g N ha}^{-1} \text{h}^{-1}$) is the average daily N_2O flux rate, i is the i th measurement, and $(t_{i+1} - t_i)$ is the number of days between two adjacent measurements.

All of the statistical analyses were carried out using SPSS 18.0. The distribution normality and variance uniformity of N_2O concentrations, N_2O fluxes, and soil variables were tested using the Kolmogorov-Smirnov and Levene tests ($P > 0.05$). The data were expressed as arithmetic means of the three replicate. The data were log-transformed when needed before analysis. The N_2O emission factor is the percentage of applied N lost as N_2O , calculated based on the difference in the total N_2O emissions from the fertilized and unfertilized soil. Two-way analysis of variance (ANOVA) by using maize planting and N fertilization as two fixed factors was applied to evaluate treatment effects on soil variable, gas fluxes within different growing season. One-way ANOVA was used to compare differences in grain yield and aboveground biomass and N uptake between treatments in the presence of maize plants. Spearman nonparametric rank correlation was used to estimate relationships between N_2O fluxes or concentrations and soil variables. Differences between treatments were considered significant compared to the least significant difference (LSD) calculations at $P < 0.05$.

Results

Grain yield, aboveground biomass, N uptake, and maximum LAI

Nitrogen fertilization significantly enhanced maize yield by 35.8% ($P < 0.01$) and 244.1% ($P < 0.01$) in 2014 and 2015, respectively (Table 1). Aboveground biomass and N uptake were significantly higher ($P < 0.01$) in the PN treatment than in the P0 treatment. The maize yield and aboveground N uptake in the P0 treatment were significantly higher ($P < 0.01$) in 2014 than in 2015. The application of N fertilizer significantly increased the maximum LAI in 2015 ($P < 0.001$), but no significant response was observed in 2014 ($P = 0.07$).

Soil temperature and WFPS

The air temperature ranged from -8.8 to 27.7 °C during the 2-year study period (Fig. 1). The mean soil temperatures are listed in Table 2. Over the 2-year period, the temperature of

the planted soil at depths of 10–50 cm ranged from 12.4 to 25.5 °C during the maize growing season. The average 2-year temperatures of the planted soil at depths of 10 to 50 cm were approximately 1 °C lower than those of the unplanted soil during the maize growing season.

The total precipitation during the maize growing season was 375 and 361 mm in 2014 and 2015, respectively (Fig. 1). Soil WFPS fluctuated with precipitation (Fig. 2, left panels). Unplanted soils were characterized by a smaller moisture range than the planted soils.

Soil mineral N

The mineral N concentrations in the upper 20 cm of the soil increased after N fertilization (Fig. 3). The exchangeable NH_4^+ concentrations decreased rapidly to a background level within 2 weeks (Fig. 3a). However, the NO_3^- concentrations remained high for more than 1 month. The mean mineral N concentrations measured in the CN treatment in 2014 and 2015 were 49.9 and 52.4 mg N kg^{-1} , respectively. These values were significantly ($P < 0.05$) decreased by the planting of maize in both years (40.7 and 38.0 mg N kg^{-1} in 2014 and 2015, respectively, in the PN treatment). Compared to the unfertilized treatments, N addition significantly ($P < 0.01$) increased the exchangeable NH_4^+ and NO_3^- concentrations.

Dynamics of N_2O concentration in soil profile

The N_2O concentrations within the soil profile did not show clear seasonal pattern (Fig. 2, right panels). Heavy rainfall dramatically increased the subsoil N_2O concentrations; this response was observed at all depths (e.g., August 10, 2014). Thereafter, the N_2O concentration decreased to the baseline level as time progressed. However, it took longer for the peaks in the deeper layers to disappear compared with those in the upper layers. The N_2O concentrations at the soil surface (0 cm) did not differ among the different treatments. During most of the experimental period, the N_2O concentration increased with soil depth. The mean N_2O concentrations at soil depths of 10 to 50 cm during the fallow season ranged from 338 to 455 ppb among the different treatments; these values were lower than those measured during the maize growing season. Compared with the unplanted treatments, the presence of maize significantly decreased ($P < 0.05$) the mean subsoil N_2O concentrations at depths of 10–40 cm during the maize growing season (Table 3). The application of N fertilizer significantly increased ($P < 0.01$) the N_2O concentrations throughout the soil profile.

Diffusive fluxes in the soil and surface emissions of N_2O

Similar to the pattern of the N_2O concentrations, the calculated N_2O diffusive effluxes at depths of 10 and 20 cm peaked after rainfall events (20 mm on June 20 and 93 mm on August 10,

Table 1 Grain yields, aboveground biomass, N uptake, and maximum LAI under N treatment

| Years | Treatment | Grain yield (Mg ha ⁻¹) | Aboveground biomass (Mg ha ⁻¹) | N uptake (kg N ha ⁻¹) | Maximum LAI (m ² m ⁻²) |
|-------|-----------|------------------------------------|--|-----------------------------------|---|
| 2014 | P0 | 8.3 ± 0.29 b | 13.7 ± 0.2 b | 117.7 ± 6.2 b | 4.4 ± 0.2 a |
| | PN | 11.3 ± 0.4 a | 20.2 ± 1.3 a | 215.6 ± 6.5 a | 5.2 ± 0.5 a |
| 2015 | P0 | 3.4 ± 0.5 b | 8.2 ± 1.3 b | 46.6 ± 2.4 b | 3.2 ± 0.2 b |
| | PN | 11.7 ± 0.1 a | 21.7 ± 0.4 a | 199.2 ± 13.2 a | 5.1 ± 0.2 a |

Mean values (mean ± stand deviation; $n = 3$) followed by different letters within a column are significantly different at $P < 0.05$

P0 maize-planted and N-unfertilized, PN maize-planted and N-fertilized, Maximum LAI (leaf area index) the LAI values at the silking stage

2014; 36 mm on June 2 and 6 mm on July 17, 2015) (Fig. 4). The N₂O efflux (May 5, 2014 and May 1, 2015) also peaks occurred after plowing and basal fertilization. The largest N₂O effluxes at a depth of 10 cm occurred on August 10, 2014 after a heavy rainfall event of 93 mm. The N₂O effluxes below the 20-cm depth were comparable and significantly ($P < 0.05$) lower than those in the 10-cm layer (Table 4). Planting maize significantly ($P < 0.05$) decreased the mean N₂O effluxes at a depth of 10 cm in the fertilized soil during the maize growing season. The average N₂O diffusive fluxes at depths of 10 and 20 cm were significantly increased ($P < 0.01$) by N inputs in both the planted and unplanted soil.

With respect to the unfertilized treatment, the presence of maize plants reduced the cumulative N₂O effluxes at a depth of 10 cm by 43% ($P < 0.05$) in 2014 and 10% ($P > 0.05$) in 2015 (Table 5). Under N fertilization, the presence of maize plants significantly decreased ($P < 0.05$) the cumulative N₂O effluxes by approximately 18% in both years. During the fallow season, no obvious difference was found among the different treatments.

The surface N₂O emissions generally exhibited a similar temporal rhythm as the soil diffusive fluxes at a depth of 10 cm (Fig. 4a, b). The overall estimated N₂O fluxes at depth of 10 and 20 cm were significantly correlated with surface N₂O emissions for all the treatments (Table 6). However, there was also a

disagreement on the N₂O fluxes between the two methods, especially when the fluxes were extremely high (Fig. 4a, b). For example, large surface N₂O emissions occurred from July 8 to 19, 2014 and on July 10, 2015 following topdressing N, whereas the estimated N₂O fluxes at a depth of 10 cm maintained at a background level during this period. The differences in the cumulative N₂O emissions derived by the chamber method among the different treatments were similar to those estimated using the gradient method at a depth of 10 cm (Table 5). The emission factor of the surface N₂O emissions during the maize growing season ranged from 0.11 to 0.14% (Table 5).

Relationships between soil variables and N₂O concentration and surface N₂O emissions

The N₂O concentrations at a depth of 10 cm exhibited a clear response pattern across the ranges of soil temperature and WFPS. The N₂O concentrations increased with soil temperature up to 17.5–22.5 °C (Fig. 5a). Above 17.5–22.5 °C, an increase in soil temperature decreased the N₂O concentrations. The N₂O concentrations at a depth of 10 cm were significantly correlated with soil temperatures during the fallow season, whereas no significant relationships were found during the maize growing season (Table S1). The N₂O concentration

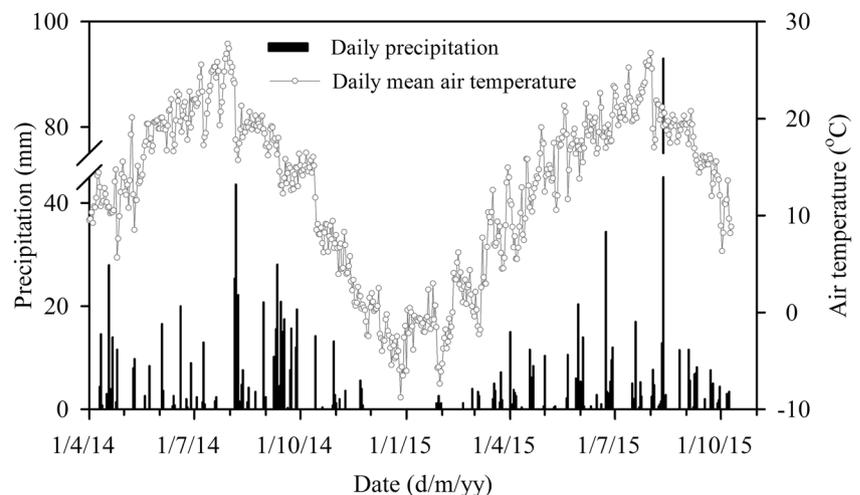
Fig. 1 Precipitation and mean daily air temperature during the study period

Table 2 The mean median and range of seasonal soil temperature (°C) at five soil depths in different treatments

| Treatment | Soil depths | Growing season in 2014 | | | Fallow season | | | Growing season in 2015 | | |
|------------------------|-------------|------------------------|--------|-----------|---------------|--------|-----------|------------------------|--------|-----------|
| | | Mean | Median | Range | Mean | Median | Range | Mean | Median | Range |
| Unplanted ^a | 10 cm | 19.9 | 19.7 | 12.6–27.1 | 4.9 | 3.3 | –1.3–16.5 | 19.4 | 20.5 | 12.8–24.8 |
| | 20 cm | 20.2 | 20.2 | 12.8–26.7 | 5.2 | 3.3 | –1.1–16.5 | 19.4 | 20.3 | 12.5–25.5 |
| | 30 cm | 20.4 | 20.8 | 13.6–27.5 | 5.7 | 3.9 | –0.5–17.1 | 19.8 | 20.7 | 12.6–25.9 |
| | 40 cm | 20.5 | 21.5 | 13.7–28.0 | 6.1 | 4.1 | 0.7–16.9 | 19.6 | 20.1 | 12.6–25.3 |
| | 50 cm | 20.2 | 21.0 | 13.3–27.0 | 6.5 | 4.9 | 0.8–16.9 | 19.6 | 20.3 | 12.5–25.0 |
| Planted | 10 cm | 18.8 | 18.8 | 12.4–23.8 | 4.4 | 3.0 | –1.2–16.5 | 18.5 | 19.3 | 13.1–22.7 |
| | 20 cm | 18.9 | 19.1 | 12.9–23.8 | 4.8 | 3.0 | –1.1–16.5 | 18.6 | 19.4 | 12.6–22.7 |
| | 30 cm | 19.3 | 19.6 | 13.7–24.4 | 5.3 | 3.4 | –0.5–17.0 | 18.9 | 19.7 | 12.5–22.8 |
| | 40 cm | 19.4 | 19.7 | 13.6–24.5 | 5.9 | 4.0 | 0.7–16.7 | 18.9 | 19.7 | 12.7–22.7 |
| | 50 cm | 19.1 | 19.8 | 13.2–23.8 | 6.3 | 4.6 | 0.8–16.5 | 18.8 | 19.5 | 12.4–22.2 |

^a Soil temperature for the unplanted (or planted) treatments represented the mean values for the unplanted (or planted) soil with and without N fertilizer

increased with soil WFPS and the highest values were measured at a WFPS between 50 and 60% (Fig. 5b). The N₂O concentrations decreased with the increasing soil WFPS above 60%. However, extremely high N₂O concentrations were observed approximately at a WFPS above 65% (Fig.

5b and S1). The N₂O concentrations were significantly correlated with soil WFPS during the maize growing season in 2014 (Table S1) but not in 2015 and the fallow season. The N₂O concentrations in the 20–50-cm layers presented a similar relationship with soil temperature and WFPS as observed

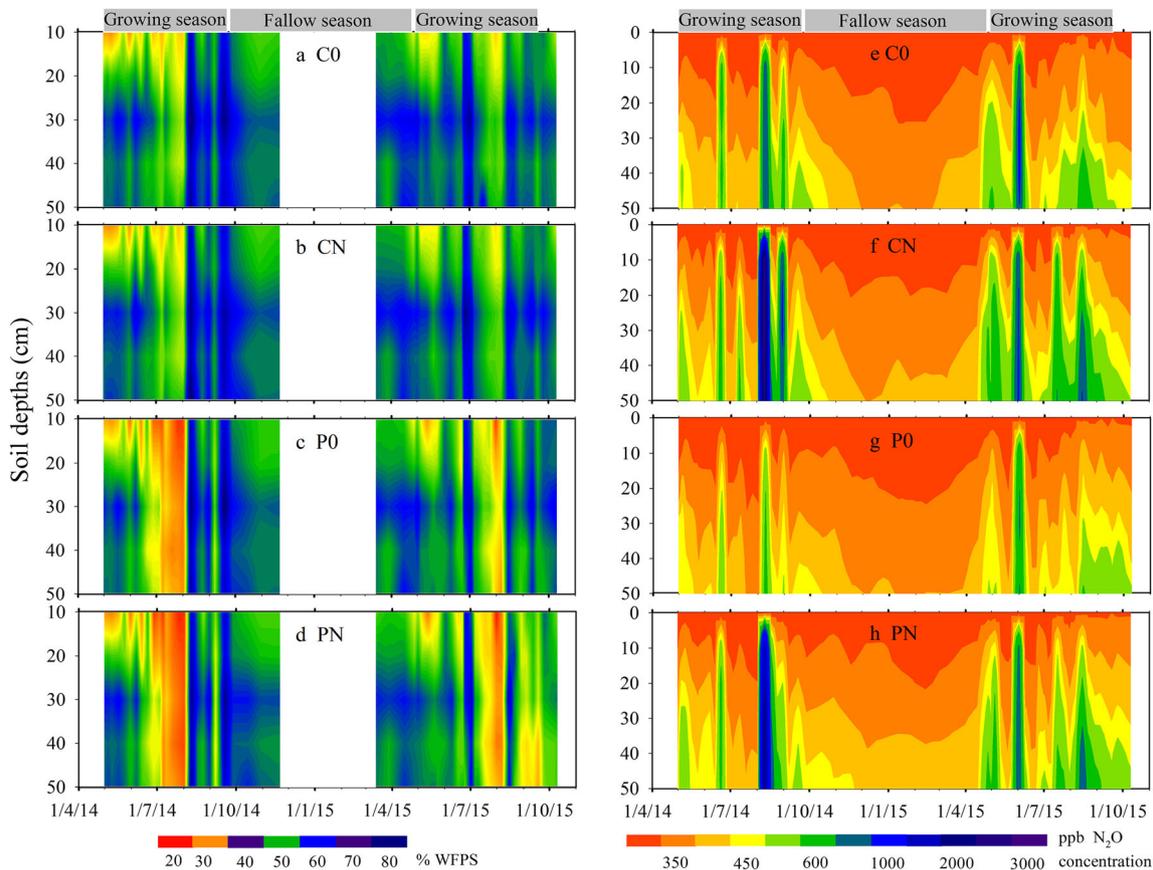


Fig. 2 Mean ($n = 3$) soil water-filled pore space (WFPS; **a–d**) and N₂O concentrations (**e–h**) interpolated between the six sampling depths in the top 50 cm of soil and the sampling dates for different treatments. C0,

unplanted and N-unfertilized; CN, unplanted and N-fertilized; P0, maize-planted and N-unfertilized; PN, maize-planted and N-fertilized

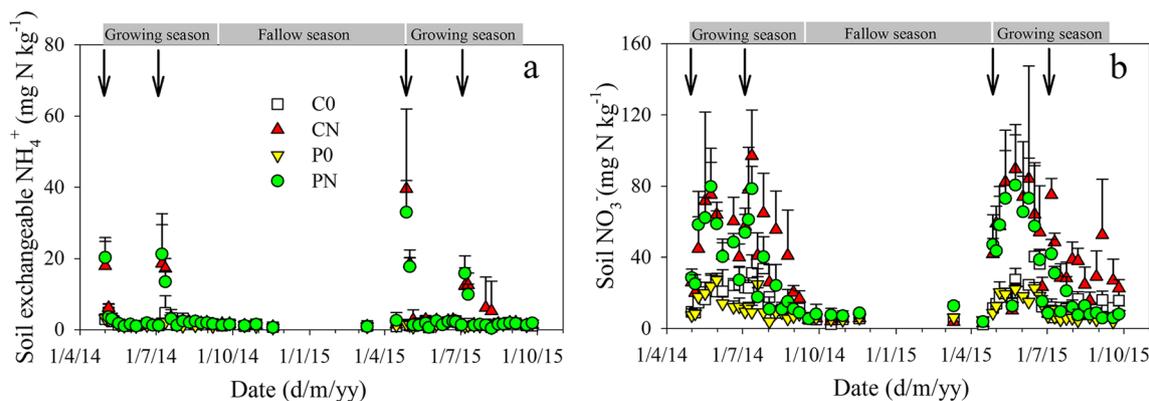


Fig. 3 Soil exchangeable NH_4^+ (a) and NO_3^- (b) concentrations in the top 20 cm for the different treatments over the course of the measurement period. The bars represent the standard deviations of the means ($n = 3$).

C0, unplanted and N-unfertilized; CN, unplanted and N-fertilized; P0, maize-planted and N-unfertilized; PN, maize-planted and N-fertilized. The solid arrows indicate fertilization

for N_2O concentrations in the 10-cm layer (Fig. S1). Similar to the results of the N_2O concentrations, the highest N_2O emissions were measured when the soil temperature was between 17.5 and 22.5 °C (Fig. 6a). Significant correlations between N_2O emissions and soil WFPS (except for the CN treatment) were observed during the maize growing season in 2014 (Table S3). The N_2O emissions were low when the WFPS was below 40% (Fig. 6b). The exchangeable NH_4^+ concentrations were less than 5 mg N kg^{-1} during most of the study period (Fig. 6c). The soil exchangeable NH_4^+ and NO_3^- concentrations in the top 20-cm layer were not significantly correlated with the N_2O emissions (Table S3).

Discussion

Subsoil N_2O concentration and efflux

The distribution of subsoil N_2O concentration varied with time and depth throughout the study period (Fig. 2, right panels). In most cases, the N_2O concentration increased with soil depth; a similar finding has been reported in many studies (Kellman and Kavanaugh 2008; Koehler et al. 2012; Nan et al. 2016; Wang et al. 2013). The N_2O concentrations at all soil depths remained low during most of the study period; however, they dramatically increased after rainfall events, especially

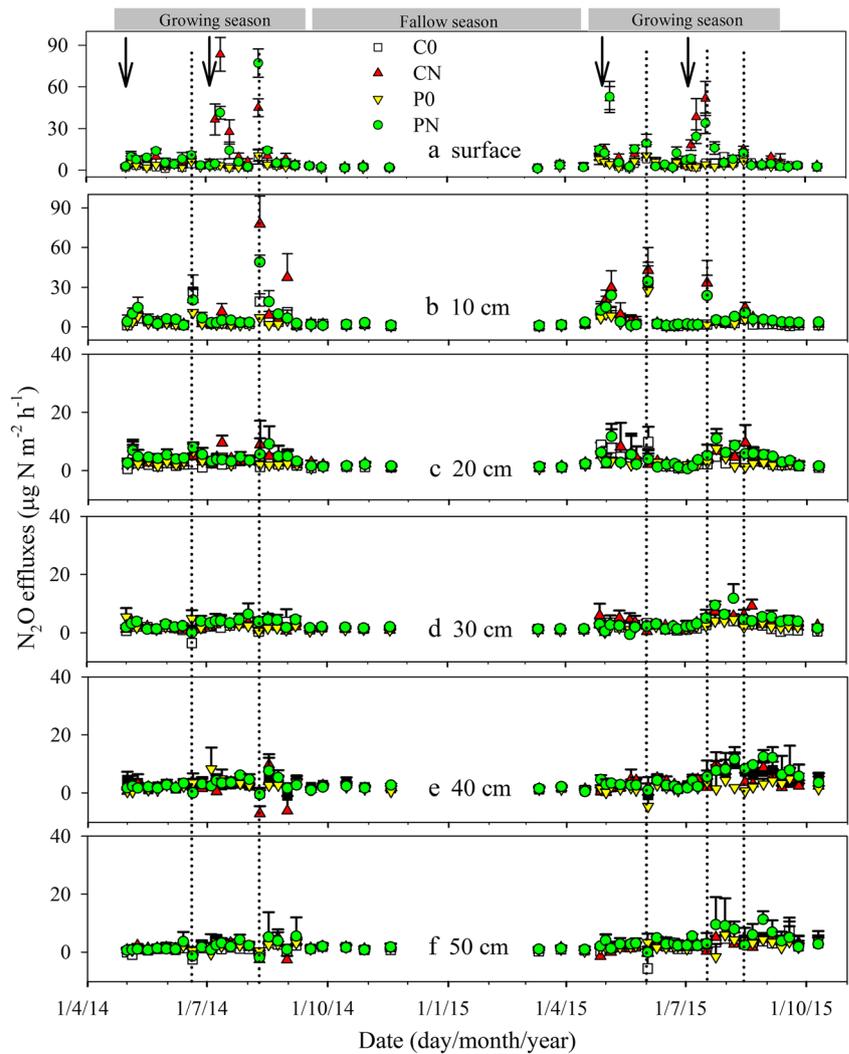
Table 3 Mean seasonal N_2O concentration at five soil depths in different treatments

| Season | Soil depth | N_2O concentration (ppb) | | | | Significance | | |
|------------------------|------------|--|------------|------------|-------------|--------------|--------------|-----------|
| | | C0 | CN | P0 | PN | Plant | N | Plant × N |
| Growing season in 2014 | 0 cm | 325 ± 2 a | 325 ± 1 a | 325 ± 1 a | 325 ± 1 a | 0.766 | 0.731 | 0.600 |
| | 10 cm | 394 ± 19 c | 503 ± 30 a | 356 ± 2 d | 444 ± 14 b | 0.001 | 0.000 | 0.352 |
| | 20 cm | 420 ± 20 c | 560 ± 28 a | 386 ± 10 c | 496 ± 21 b | 0.003 | 0.000 | 0.289 |
| | 30 cm | 454 ± 23 c | 592 ± 31 a | 418 ± 13 c | 542 ± 33 b | 0.013 | 0.000 | 0.570 |
| | 40 cm | 490 ± 26 b | 615 ± 31 a | 444 ± 19 b | 570 ± 41 a | 0.026 | 0.000 | 0.954 |
| Fallow season | 0 cm | 330 ± 1 a | 329 ± 1 a | 330 ± 2 a | 331 ± 0 a | 0.178 | 0.897 | 0.641 |
| | 10 cm | 341 ± 3 b | 342 ± 3 b | 343 ± 1 b | 348 ± 1 a | 0.012 | 0.059 | 0.074 |
| | 20 cm | 357 ± 4 b | 364 ± 3 a | 357 ± 1 b | 364 ± 4 a | 0.921 | 0.008 | 0.945 |
| | 30 cm | 380 ± 8 b | 385 ± 5 ab | 375 ± 3 b | 394 ± 8 a | 0.624 | 0.013 | 0.117 |
| | 40 cm | 405 ± 12 ab | 410 ± 7 ab | 398 ± 7 b | 424 ± 14 a | 0.622 | 0.033 | 0.110 |
| Growing season in 2015 | 0 cm | 334 ± 0 a | 333 ± 1 a | 333 ± 0 a | 334 ± 1 a | 0.333 | 0.503 | 0.089 |
| | 10 cm | 389 ± 13 c | 434 ± 5 a | 382 ± 6 c | 408 ± 13 b | 0.023 | 0.000 | 0.139 |
| | 20 cm | 434 ± 23 bc | 484 ± 9 a | 413 ± 8 c | 450 ± 12 b | 0.011 | 0.001 | 0.436 |
| | 30 cm | 477 ± 27 bc | 551 ± 21 a | 446 ± 8 c | 495 ± 19 b | 0.005 | 0.001 | 0.322 |
| | 40 cm | 534 ± 36 b | 607 ± 20 a | 468 ± 13 c | 548 ± 16 b | 0.002 | 0.000 | 0.796 |
| | 50 cm | 569 ± 48 b | 642 ± 10 a | 501 ± 34 c | 592 ± 20 ab | 0.012 | 0.002 | 0.635 |

Mean values ($n = 3$) followed by different letters within a row in the same seasons are significantly different at $P < 0.05$. Bold indicates significance ($P < 0.05$)

C0 unplanted and N-unfertilized, CN unplanted and N-fertilized, P0 maize-planted and N-unfertilized, PN maize-planted and N-fertilized

Fig. 4 Mean N₂O surface emissions (a) and diffusive effluxes within the soil profile (b–f) under different treatments over the course of the measurement period. C0, unplanted and N-unfertilized; CN, unplanted and N-fertilized; P0, maize-planted and N-unfertilized; PN, maize-planted and N-fertilized. The solid arrows indicate fertilization, and the dotted lines indicate rainfall



in the N fertilizer soils, implying that rainfall and N fertilization were the main factors controlling the N₂O concentrations.

The significant correlations between N₂O concentrations and the corresponding soil temperature at each soil depth during the fallow season indicated that low temperatures limited N₂O production. During the maize growing season in both years, the maximum N₂O concentrations were measured at temperatures ranging from 17.5 to 22.5 °C (Fig. 5a). Either lower or higher soil temperatures decreased the N₂O concentration. Lower temperature usually restricts microbial activities and subsequently the production of N₂O (Smith 2017). In our study, soil temperatures showed a similar seasonal pattern to air temperature and there was a significant decrease in soil WFPS with increasing soil temperature ($R = -0.25$, $P < 0.001$). Therefore, at higher temperatures, soil WFPS was the likely factor that limited the production and accumulation of N₂O within the soil profile. Cosentino et al. (2013) reported that soil WFPS decreased with topsoil (10 cm) temperatures and the effect of soil temperature on N₂O

emissions could be indirectly mediated by soil moisture in an agricultural field.

During the maize growing season in both years, the N₂O concentrations increased with soil WFPS in a range from 20 to 50%, and the highest N₂O concentrations were measured at a WFPS of 55–60% (except for the extremely high values at a WFPS > 65%). When the soil WFPS exceeded 60%, the N₂O concentrations decreased with soil WFPS. This was probably because N₂O produced in subsoil profiles were reduced to N₂ (Dalal et al. 2003). Higher WFPS increased the residence time for N₂O within soil profile and decreased soil O₂ content, which resulted more complete denitrification of N₂O to N₂ (Clough et al. 2005; van Groenigen et al. 2005). However, extremely high N₂O concentrations were observed at a WFPS > 65% (Fig. 5b and S1). These high N₂O concentrations were measured on August 10 following a heavy rainfall in 2014 (93 mm, from August 5 to 9). It has been reported that N₂O pulses usually occurred after heavy rainfall in soils affected by drought (Snider et al. 2015), and the magnitude of

Table 4 Mean seasonal N₂O surface emissions and diffusive effluxes within the soil profiles of different treatments

| Season | Soil depth | Mean N ₂ O effluxes ($\mu\text{g N m}^{-2} \text{h}^{-1}$) | | | | Significance | | |
|------------------------|------------|---|------------------|------------------|------------------|--------------|--------------|------------------|
| | | C0 | CN | P0 | PN | Plant | N | Plant \times N |
| Growing season in 2014 | Surface | 3.8 \pm 0.3 c | 13.3 \pm 0.3 a | 3.5 \pm 0.3 c | 11.6 \pm 0.2 b | 0.000 | 0.000 | 0.000 |
| | 10 cm | 5.2 \pm 0.9 c | 10.3 \pm 0.7 a | 3.1 \pm 0.1 d | 8.5 \pm 1.3 b | 0.006 | 0.000 | 0.766 |
| | 20 cm | 2.3 \pm 0.5 c | 4.1 \pm 0.5 ab | 3.1 \pm 0.8 bc | 4.7 \pm 0.5 a | 0.068 | 0.001 | 0.738 |
| Fallow season | Surface | 3.3 \pm 0.4 a | 3.3 \pm 0.8 a | 2.5 \pm 0.5 a | 3.5 \pm 1.1 a | 0.177 | 0.019 | 0.04 |
| | 10 cm | 2.5 \pm 0.5 a | 2.8 \pm 0.9 a | 2.0 \pm 0.2 a | 3.1 \pm 0.8 a | 0.750 | 0.101 | 0.350 |
| | 20 cm | 2.2 \pm 0.2 ab | 2.3 \pm 0.2 a | 1.8 \pm 0.3 b | 2.2 \pm 0.3 ab | 0.114 | 0.098 | 0.326 |
| Growing season in 2015 | Surface | 5.1 \pm 0.4 c | 13.6 \pm 0.2 a | 4.5 \pm 0.5 c | 11.8 \pm 0.4 b | 0.021 | 0.000 | 0.200 |
| | 10 cm | 4.9 \pm 1.0 c | 9.3 \pm 0.4 a | 4.3 \pm 0.5 c | 7.6 \pm 0.8 b | 0.032 | 0.000 | 0.228 |
| | 20 cm | 3.7 \pm 0.7 bc | 4.4 \pm 0.5 a | 2.8 \pm 0.3 c | 4.6 \pm 0.2 a | 0.246 | 0.002 | 0.087 |

Mean values ($n = 3$) followed by different letters within a row in the same seasons are significantly different at $P < 0.05$. Bold indicates significance ($P < 0.05$)

C0 unplanted and N-unfertilized, CN unplanted and N-fertilized, P0 maize-planted and N-unfertilized, PN maize-planted and N-fertilized

the N₂O pulse was related to soil moisture prior to wetting (Cai et al. 2016; Uchida et al. 2014). The lower the prewetting soil moisture, the larger the N₂O pulses (Cai et al. 2016; Uchida et al. 2014). This was mainly because the higher labile C mineralized from the dry soil following soil wetting (Harrison-Kirk et al. 2014) increased C sources and the activities of microbial organisms and therefore the N₂O production (Snider et al. 2015). In our study, the soil suffered a persistent drought lasted from late July to early August and therefore the subsequent heavy rainfall strongly stimulated the N₂O production. The higher soil moisture after rainfall blocked soil pores, which further restricted N₂O diffusion from the subsoil to the atmosphere, and therefore, large quantities of N₂O accumulated in the subsoil. The N₂O concentrations at each soil depth for all the treatments (except for the CN treatment at a depth 10 cm) showed a significant and positive relationship with soil WFPS (Table S1) during the maize growing season in 2014 but not in 2015. This was possibly because the soil

WFPS values during the maize growing seasons in 2014 were generally lower than those in 2015 (Fig. 2, left panels), and the response of N₂O concentrations to soil WFPS was more sensitive in 2014 than that in 2015. Moreover, the extremely high N₂O concentrations measured on August 10, 2014 strongly affected the general response of N₂O concentrations to WFPS.

Highest N₂O concentrations at a depth of 10 cm were observed when the exchangeable NH₄⁺ was less than 5 mg N kg⁻¹, indicating NH₄⁺ had no effect on the N₂O concentrations. High N₂O concentrations were measured when the NO₃⁻ concentrations exceeded 10 mg N kg⁻¹. Previous studies have indicated that N₂O production was not limited if soil NO₃⁻ concentration was above the threshold of 5 mg N kg⁻¹ in intensively managed agricultural field (Dobbie et al. 1999). Our results of no significant correlations between N₂O concentrations and NO₃⁻ indicated that the formation N₂O concentrations were restricted by other factors such as soil WFPS and labile C. Ju et al. (2011) found that N₂O emissions were

Table 5 Cumulative N₂O surface emissions and diffusive effluxes within the soil profiles of different treatments

| Season | Soil depth | Cumulative N ₂ O effluxes (g N ha ⁻¹) | | | | Significance | | |
|------------------------|------------|--|---------------------------------------|-----------------|--------------------------|--------------|--------------|------------------|
| | | C0 | CN | P0 | PN | Plant | N | Plant \times N |
| Growing season in 2014 | Surface | 127 \pm 11 c | 446 \pm 22 a (0.14) ^c | 117 \pm 9 c | 407 \pm 12 b (0.13) | 0.021 | 0.000 | 0.752 |
| | 10 cm | 184 \pm 33 c | 367 \pm 35 a | 106 \pm 4 d | 301 \pm 48 b | 0.006 | 0.000 | 0.643 |
| | 20 cm | 76 \pm 18 c | 135 \pm 16 ab | 101 \pm 27 bc | 160 \pm 14 a | 0.055 | 0.001 | 0.974 |
| Fallow season | Surface | 131 \pm 17 a | 133 \pm 12 a | 95 \pm 13 b | 125 \pm 13 a | 0.027 | 0.070 | 0.119 |
| | 10 cm | 78 \pm 17 a | 90 \pm 25 a | 73 \pm 3 a | 105 \pm 21 a | 0.478 | 0.120 | 0.291 |
| | 20 cm | 82 \pm 27 a | 81 \pm 8 a | 73 \pm 13 a | 93 \pm 21 a | 0.836 | 0.320 | 0.713 |
| Growing season in 2015 | Surface | 174 \pm 11 c | 462 \pm 29 a (0.13) | 152 \pm 13 c | 394 \pm 34 b (0.11) | 0.011 | 0.000 | 0.127 |
| | 10 cm | 174 \pm 39 c | 328 \pm 9 a | 157 \pm 15 c | 271 \pm 22 b | 0.029 | 0.000 | 0.188 |
| | 20 cm | 124 \pm 28 bc | 154 \pm 18 ab | 96 \pm 10 c | 161 \pm 8 a | 0.351 | 0.002 | 0.123 |

Mean values ($n = 3$) followed by different letters within a row in the same seasons are significantly different at $P < 0.05$. The values in the parentheses represent the emission factor. Bold indicates significance ($P < 0.05$)

C0 unplanted and N-unfertilized, CN unplanted and N-fertilized, P0 maize-planted and N-unfertilized, PN maize-planted and N-fertilized

Table 6 Spearman rank correlation coefficients (*R*) between surface N₂O emissions and modeled diffusive fluxes (μg N m⁻² h⁻¹) at different soil depths

| Soil depth | Overall | | C0 | | CN | | P0 | | PN | |
|------------|----------|--------------|----------|--------------|----------|--------------|----------|--------------|----------|--------------|
| | <i>R</i> | <i>P</i> |
| 10 cm | 0.586 | 0.000 | 0.477 | 0.000 | 0.593 | 0.000 | 0.581 | 0.000 | 0.554 | 0.000 |
| 20 cm | 0.493 | 0.000 | 0.394 | 0.003 | 0.549 | 0.000 | 0.255 | 0.048 | 0.499 | 0.000 |
| 30 cm | 0.311 | 0.005 | 0.356 | 0.008 | 0.330 | 0.014 | 0.137 | 0.317 | 0.195 | 0.154 |
| 40 cm | 0.070 | 0.304 | 0.029 | 0.831 | 0.056 | 0.685 | -0.089 | 0.519 | 0.056 | 0.683 |
| 50 cm | 0.065 | 0.339 | -0.026 | 0.848 | 0.011 | 0.937 | 0.085 | 0.539 | 0.020 | 0.882 |

Bold indicates significance (*P* < 0.05)

C0 unplanted and N-unfertilized, CN unplanted and N-fertilized, P0 maize-planted and N-unfertilized, PN maize-planted and N-fertilized

not correlated with soil NO₃⁻ because the low soil WFPS and readily oxidizable C limited N₂O production via denitrification in the calcareous soil in North China.

In the present study, positive N₂O fluxes in the 0–10-cm soil layers were generally higher than those in the deeper layers during the maize growing season (Fig. 4), and the fluxes in each layer of the 20–50-cm depth were comparable. As the N₂O turnover rate (production or consumption) in soil

is driven by the change in the diffusive flux from the center of one layer to the center of the next layer, the flux profiles can be interpreted as source and sink layers (Goldberg et al. 2008; Maier and Schack-Kirchner 2014). Our results suggest that most N₂O production occurred in the topsoil (0–10 cm), which is in agreement with other studies that suggested that most N₂O is produced in the 0–30-cm soil layers (Kusa et al. 2010; Pihlatie et al. 2007; Wang et al. 2013). Judging from the

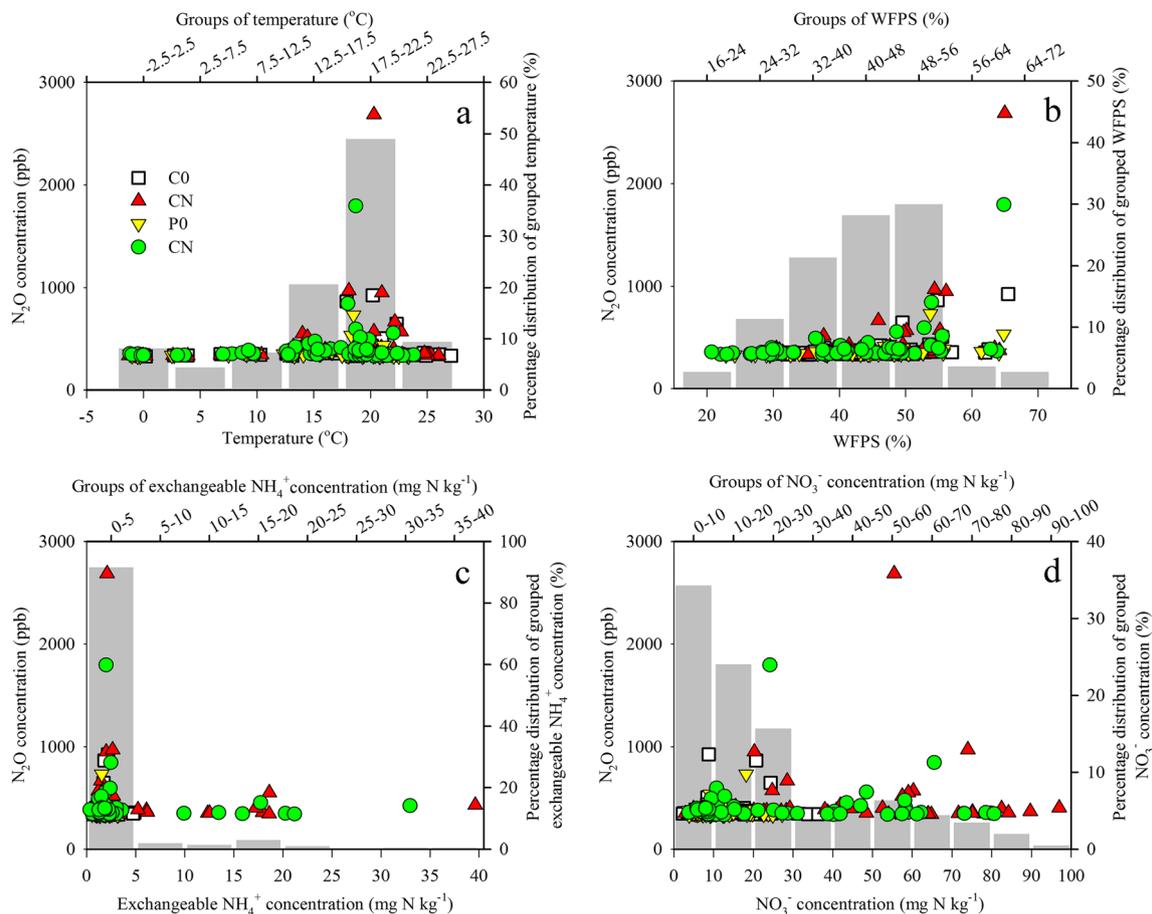


Fig. 5 The relationships between soil temperature (a), soil water-filled pore space (b), soil exchangeable NH₄⁺ concentration (c), soil NO₃⁻ concentration (d), and soil N₂O concentrations at a depth of 10 cm over

the course of the measurement period. C0, unplanted and N-unfertilized; CN, unplanted and N-fertilized; P0, maize-planted and N-unfertilized; PN, maize-planted and N-fertilized

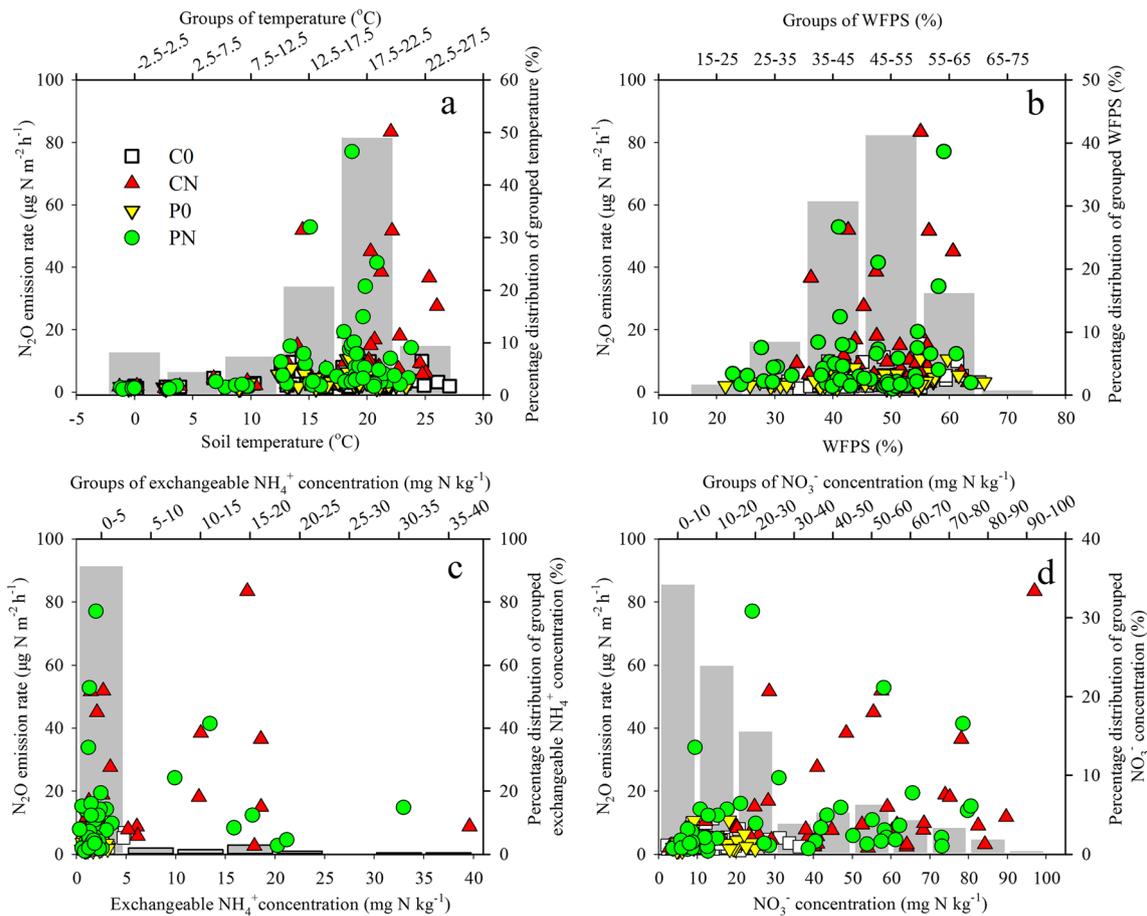


Fig. 6 The relationships between soil temperature (**a**), soil water-filled pore space (WFPS, **b**), soil exchangeable NH_4^+ concentration (**c**), soil NO_3^- concentration (**d**), and surface N_2O emissions. C0, unplanted and

N-unfertilized; CN, unplanted and N-fertilized; P0, maize-planted and N-unfertilized; PN, maize-planted and N-fertilized

positive and small N_2O diffusive fluxes, we inferred that the soil layers below 20 cm might be a minor source of N_2O because N_2O in the deep soil may contribute to surface N_2O emissions as it slowly diffused upwards or it may be reduced to N_2 before reaching atmosphere. Our results were consistent with the finding of Nan et al. (2016), who found that N_2O fluxes below 30 cm depth were approximately zero for a maize experiment near our study site.

N_2O surface fluxes and controlling factors

The surface N_2O emissions presented a similar relationship with soil WFPS as observed for the N_2O concentrations. Over the entire study period, the soil WFPS in the 0–20-cm layer seldom exceeded 60% (Fig. 6b); therefore, it is possible that the N_2O in the tested soil was mainly produced by nitrification during most of the study period (Abalos et al. 2017; Bateman and Baggs 2005; Ju et al. 2011). However, denitrification may also contribute to the large N_2O fluxes after intense rainfall (Snider et al. 2015; Uchida et al. 2014). For example, the N_2O pulses that occurred on August 10

following intense rainfall were most likely caused by denitrification because of the high soil WFPS and NO_3^- concentrations. The optimum WFPS for N_2O emission was between 55 and 60% (Fig. 6b), and the emissions decrease with the increase of WFPS above 60%. Similarly, Horváth et al. (2010) reported that N_2O emissions increased with soil moisture and peaked around at 50% WFPS, and then the emissions decreased with the increased of WFPS (> 50%) in sandy soils. This is because gas diffusion from the soil into the atmosphere is hindered to a large extent when the soil WFPS is above 60% (Nan et al. 2016; Zhou et al. 2016). Moreover, the decrease in N_2O emissions at soil WFPS > 60% is probably related to the reduction of N_2O to N_2 (Dalal et al. 2003). It has been reported that short periods of anoxic conditions after heavy rainfall event could increase N_2O reductase activity (Uchida et al. 2014).

Exchangeable NH_4^+ and NO_3^- were used as the substrates for the production of N_2O from nitrification and denitrification, respectively (Baggs 2011). As long as other factors such as soil moisture and labile C are not limiting, a rapid response of N_2O emissions to the N fertilization occurred (Smith 2017).

In our study, large N_2O pulses occurred after N fertilization (i.e., in July 2014 and in May and July 2015). During the pulses of N_2O emissions, the relatively low soil WFPS (40–50%) and the quick decrease of exchangeable NH_4^+ concentrations from the peak to a low level ($< 5 \text{ mg N kg}^{-1}$) within 1 week after N fertilization indicated that nitrification of the applied urea-N was likely the main process of N_2O production. It has been reported that ammonia oxidation and linked nitrifier denitrification (ND) are the major processes generating N_2O in the calcareous soil following applying urea-N (Huang et al. 2014). Our result of no significant relationship between N_2O emissions and exchangeable NH_4^+ was probably because the exchangeable NH_4^+ was quickly transformed to NO_3^- after N inputs. The N_2O emissions were not significantly correlated with NO_3^- -N, which was mainly because that N_2O production from the reduction of NO_3^- was usually limited by the low availability of labile organic C and the well-aerated conditions in the calcareous soil (Ju et al. 2011). It has been reported that large N_2O emissions were produced by denitrification following heavy rainfall (Abalos et al. 2017). When the soil conditions were favorable for denitrification, the rate of denitrification increased with soil NO_3^- concentrations (Dalal et al. 2003). In our study, the higher N_2O emissions measured on August 10 (following an intense rainfall) in the N-fertilized soil than that in the unfertilized soil was possibly attributed to the higher NO_3^- concentrations. Moreover, recent studies have suggested that N_2O emissions were more correlated with soil nitrite (NO_2^-) concentration than with exchangeable NH_4^+ and NO_3^- concentration (Ma et al. 2015). This was because the reduction of NO_2^- via nitrifier denitrification played an important role in the N_2O emissions (Ma et al. 2015; Huang et al. 2014). Therefore, to study the relationship between N_2O emissions and NO_2^- is needed in future study.

In our study, the emission factor (EF) values ranged from 0.11 to 0.14% (Table 5), which are lower than the default value of 1% reported by the IPCC (IPCC 2013) as well as most other studies (Chen et al. 2014; Liu et al. 2011; Zhang et al. 2016). However, the values are comparable to those (0.02–0.18%) reported by Abalos et al. (2017) for a rainfed system under a Mediterranean climate, as well as the results (0.18–0.23%) obtained by Wang et al. (2016) for a nearby cropped site. The low EF values in our study were primarily due to the low soil organic C content and low WFPS in our study site. It has been reported that N_2O emissions increased with soil organic C contents, because high soil organic C increased microbial activities, resulting in increased O_2 consumption and consequently favoring N_2O production via denitrification (Russow et al. 2008). Our value was lower than the 0.34% for a black soil (soil organic C content = 27.5 g kg^{-1}) in a rainfed maize field fertilized with 150 kg N ha^{-1} in Northeast China (Chen et al. 2014). The emission factor in our study is also lower than the 1.34% from an irrigated wheat-maize cropping system in northern

China (Ding et al. 2007). With the similar organic C content (7.28 g ka^{-1}) and N rate (250 kg ha^{-1}), the difference in emission factor was most likely attributed to the higher soil WFPS in their study (40 vs 53%).

It is noteworthy that although there is no substantial difference in the cumulative N_2O emissions and subsoil fluxes during the maize growing season between the 2 years, there were some differences in the mechanisms underlying for the N_2O production. The N_2O pulses that occurred within 2 weeks of N fertilization (July 12, 2014, May 5 and July 17, 2015) were mainly derived from the nitrification of the exchangeable NH_4^+ from urea hydrolysis. However, the N_2O pulse occurred on August 10 following a heavy rainfall was primarily driven by denitrification, because of the low exchangeable NH_4^+ concentrations and high soil WFPS.

Effect of maize plants on soil concentrations and surface emissions of N_2O

In our study, the presence of maize plants significantly ($P < 0.05$) decreased the N_2O concentration in the soil profile at a depth of 10–40 cm relative to the unplanted treatments during the maize growing season (Table 3). There are three possible explanations for this phenomenon: first, the mean WFPS (10–50 cm layers) and mineral N in the 0–20-cm layer during the maize growing season were significantly decreased by the presence of maize plants (Figs. 2 and 3). Second, the lower temperature of the planted soil might decrease microbial activities and subsequently the N_2O production. Third, higher gas diffusivity due to lower moisture and developed root systems in the planted soil also facilitated the diffusion of N_2O produced in the subsoil into the atmosphere, which resulted in the lower N_2O concentration in the planted soil. However, the second possibility was excluded: the average temperatures at soil depths of 10 to 50 cm during the maize growing season were approximately 19 and 20 °C for the planted and unplanted soil (Table 2), respectively, both of which fell into the range of optimum temperatures (17.5 to 22.5 °C) for the highest N_2O concentrations and surface emissions (Fig. 3). The results implied that during the maize growing season, the decrease in soil temperature resulting from canopy shading contributed little to the decrease in N_2O emissions.

The presence of maize plants significantly ($P < 0.05$) decreased the cumulative surface N_2O emissions in the maize growing season over the 2 years (Table 5). Similarly, Jamali et al. (2016) found that N_2O emissions from wheat-planted cores were significantly higher than those from fallow cores, which was attributed to plant uptake of soil water and available N. Ding et al. (2007) also reported that the presence of winter wheat plants significantly decreased N_2O emissions because of lower soil temperatures and available N caused by plant shading and uptake. However, our results are in contrast with most previous studies that showed plants promoted N_2O

emissions (Hayashi et al. 2015; Song and Zhang 2009; Verma et al. 2006). As already mentioned, maize plants significantly increased N_2O fluxes compared to bare soil in agricultural fields in China because plant-derived C stimulated N_2O production (Ding et al. 2007; Ni et al. 2012; Song and Zhang 2009). All of these field experiments have the following in common: there was no significant difference in the soil WFPS of the surface layers between the planted and unplanted treatments because of high rainfall during the maize growing season. Similarly, Uchida et al. (2011) also reported that the presence of pasture plants significantly increased N_2O fluxes when the soil WFPS (with and without plants) was controlled at 70%. However, the main factors controlling N_2O emissions, i.e., soil water and mineral N, are limited in semiarid areas (Delgado-Baquerizo et al. 2013). In our study, the exchangeable NH_4^+ and NO_3^- concentrations showed no significant effect on N_2O emissions, and therefore the primary driving factor caused the decrease in N_2O emissions in the planted soil was soil WFPS. Our results indicate that the plant-induced decrease in soil moisture was greater than the increase in root-derived C with respect to the effect on N_2O emissions in the current study.

The surface N_2O emissions after heavy rainfall events (e.g., 93 mm, from August 5 to August 9, 2014) were significantly higher ($P < 0.05$) for the PN treatment compared with the CN treatment (Fig. 4a). Probably, the soil WFPS was not the factor that caused the higher N_2O fluxes in the planted soil because there was no significant difference ($P > 0.05$) in the soil WFPS between the PN and CN treatments (c. 65%). Moreover, the soil WFPS values for all treatments were higher than 60% following heavy rainfall events (Fig. 2a); therefore, it is likely that denitrification played an important part in N_2O production during this period (Ju et al. 2011; Linn and Doran 1984). Second, the possible roles of soil temperature and mineral N were also ruled out because the soil temperatures measured in the PN (18.7 °C) and CN (20.3 °C) treatments fell within the optimum range of temperatures for N_2O production, and the higher NO_3^- concentrations in the unplanted soil were more beneficial to N_2O production. Third, the higher N_2O emissions from the planted soil compared with the unplanted soil can probably be attributed to the higher production of N_2O stimulated by root-derived labile C because available organic C from maize plants is an important driver that promotes N_2O production through denitrification (Henry et al. 2008; Qian et al. 1997; Uchida et al. 2011). However, this explanation was also excluded because of the higher subsoil N_2O concentration at a depth of 10 cm in the unplanted soil (1795 and 2688 ppb in the PN and CN treatments, respectively) (Fig. 2b). According to Fick's law, surface gas emissions depend not only on the gas concentration gradient but also on the effective diffusion coefficient. Therefore, in our study, the higher N_2O emissions from the planted soil after heavy rainfall events were likely due to the enhanced gas diffusion

associated with plant roots. The preferential flow ways through macro pores created by plant roots increased the gas and water movement within soil profile (Angers and Caron 1998; Bohn et al. 2011). Therefore, the water in the upper layers percolated more rapidly to deeper layer in the planted soil, which could facilitate the gas exchange between the soil and atmosphere (Jamali et al. 2016). Therefore, the higher diffusivity in the PN treatment resulted in more O_2 diffusion into soil, which did not favor the reduction of N_2O to N_2 . On the other hand, respiration of root and the root-associated microorganisms in the planted soil depleted more O_2 and created anaerobic conditions. The root-derived C and the low O_2 levels may stimulate more N_2O reduction to N_2 relative to the unplanted treatment (Henry et al. 2008; Klemmedtsson et al. 1987). Therefore, the actual effect of plants on denitrification was complex. In this study, the N_2O concentrations were higher in the CN treatment, whereas the surface N_2O emissions were lower compared to those of the PN treatment. We speculated that more N_2O was reduced before escaping to the atmosphere in the CN treatment. Further research was needed to study the effect of maize plants on N_2 emissions.

Comparison of the closed-chamber and concentration gradient method

The N_2O fluxes at a depth of 10 cm estimated using the gradient method and the surface N_2O emissions measured using the chamber method generally had a similar pattern (Fig. 4). A significant and positive relationship was also found between the measured and estimated fluxes (Table 6), indicating that the gradient method could be useful to measure N_2O emissions. Satisfactory agreement between the two methods has been reported by Yoh et al. (1997) for a volcanic ash soil, Maljanen et al. (2003) for a drained organic soil, and Zhou et al. (2016) for a rice-wheat rotation system.

However, it should be noted that the estimated N_2O fluxes did not completely correspond with the measured fluxes, particularly when the N_2O emissions were high (Fig. 4). Peak N_2O emissions were associated with increases in the WFPS following rainfall, which were sometimes under- or overestimated by the gradient method. As a result, there was a discrepancy between the two methods with respect to the cumulative seasonal N_2O fluxes. It has been suggested that gradient method does not perform well in the wet soil, especially after rainfall events (Clough et al. 2005; Kusa et al. 2008; Maljanen et al. 2003; Wolf et al. 2011), due to the vigorous N_2O production and consumption processes in the wet surface soil above the uppermost gas sampler (10 cm). The uppermost soil layers (above 10 cm) usually have high microbial activities and rates of N_2O production (Uchida et al. 2014) and transportation towards atmosphere (Wolf et al. 2011); however, it is difficult to accurately measure the gas concentration and diffusion coefficients in this zone (Kusa

et al. 2008; Maier and Schack-Kirchner 2014). In the present study, the shallowest gas sampler was installed at a depth of 10 cm. This sampler was not able to monitor the N₂O concentration above a depth of 10 cm. Therefore, the fluxes estimated using the gradient method may not represent the real N₂O fluxes if high amounts of N₂O are produced in the top few centimeters. Furthermore, the increase in soil moisture in the top few centimeters was underestimated by regular measurements that were integrated over a larger soil volume. Therefore, continuous monitoring of N₂O concentrations at shallower depths is necessary in future studies to capture the dynamics of N₂O in the surface layers. The estimated N₂O flux was calculated based on Fick's law, assuming that gas transportation within the soil profile and between the soil and atmosphere was entirely driven by diffusive transportation (Clough et al. 2005; Maier and Schack-Kirchner 2014). Therefore, neglecting N₂O diffusion in soil water and convective transportation in the flux calculation may also contribute to the disagreement between the two methods.

Conclusion

Our work showed that the presence of maize plants significantly decreased the N₂O surface emissions as well as the N₂O concentrations and diffusive fluxes in the top 10-cm layer. This pattern was probably due to the uptake of soil water by the maize plants, which resulted in less suitable soil conditions for N₂O production. The application of N fertilizer significantly increased N₂O surface emissions and the N₂O concentrations within the soil profile. The gradient method exhibited a similar pattern to the chamber measurements; however, there was a discrepancy between the two methods when the N₂O emissions were high. Our results are helpful in understanding the mechanism and drivers of N₂O fluxes in plant-soil systems under dryland farming.

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