

Nitrous oxide emission from highland winter wheat field after long-term fertilization

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Abstract. Nitrous oxide (N_2O) is an important greenhouse gas. N₂O emissions from soils vary with fertilization and cropping practices. The response of N₂O emission to fertilization of agricultural soils plays an important role in global N₂O emission. The objective of this study was to assess the seasonal pattern of N₂O fluxes and the annual N₂O emissions from a rain-fed winter wheat (Triticum aestivum L.) field in the Loess Plateau of China. A static flux chamber method was used to measure soil N₂O fluxes from 2006 to 2008. The study included 5 treatments with 3 replications in a randomized complete block design. Prior to initiating N2O measurements the treatments had received the same fertilization for 22 years. The fertilizer treatments were unfertilized control (CK), manure (M), nitrogen (N), nitrogen + phosphorus (NP), and nitrogen + phosphorus + manure (NPM). Soil N_2O fluxes in the highland winter wheat field were highly variable temporally and thus were fertilization dependent. The highest fluxes occurred in the warmer and wetter seasons. Relative to CK, m slightly increased N2O flux while N, NP and NPM treatments significantly increased N2O fluxes. The fertilizer induced increase in N2O flux occurred mainly in the first 30 days after fertilization. The increases were smaller in the relatively warm and dry year than in the cold and wet year. Combining phosphorous and/or manure with mineral N fertilizer partly offset the nitrogen fertilizer induced increase in N2O flux. N2O fluxes at the seedling stage were mainly controlled by nitrogen fertilization, while fluxes at other plant growth stages were influenced by plant



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and environmental conditions. The cumulative N2O emissions were always higher in the fertilized treatments than in the non-fertilized treatment (CK). Mineral and manure nitrogen fertilizer enhanced N2O emissions in wetter years compared to dryer years. Phosphorous fertilizer offset 0.50 and $1.26 \text{ kg N}_2\text{O-N} \text{ ha}^{-1}$ increases, while manure + phosphorous offset 0.43 and $1.04 \text{ kg } N_2 \text{O-N} \text{ ha}^{-1}$ increases by N fertilizer for the two observation years. Our results suggested that the contribution of single N fertilizer on N₂O emission was larger than that of NP and NPM and that manure and phosphorous had important roles in offsetting mineral N fertilizer induced N₂O emissions. Relative to agricultural production and N₂O emission, manure fertilization (M) should be recommended while single N fertilization (N) should be avoided for the highland winter wheat due to the higher biomass and grain yield and lower N2O flux and annual emission in m than in N.

1 Introduction

Nitrous oxide (N₂O), an important greenhouse gas in the atmosphere, has increased from pre-industrial concentrations of 270 ppb to 319 ppb in 2005 (Forster et al., 2007). Soil is acknowledged as the major source of N₂O, accounting for about 70% of total emissions. Agricultural soils account for a large proportion (70–81%) of the increase in N₂O emissions to the atmosphere, with the increase linked to increased N fertilizer use (Bouwman, 1990). A recent calculation showed that 3.3 Tg N₂O-N yr⁻¹ is emitted globally from fertilized cropland (Stehfest and Bouwman, 2006). However, the linkages between agricultural soil emissions and global emissions are still uncertain (Stehfest and Bouwman, 2006), and further understanding of N_2O emissions from cropped land are still necessary for accurate global N_2O emission prediction.

The response of soil N₂O emissions to N fertilizatioN has been widely studied in different ecosystems. Dittert et al. (2005) and Lampe et al. (2006) showed larger N₂O emissions from mineral N than from slurry treated grassland plots and demonstrated that the soil N pool was always clearly the major source of N2O. Zhang and Han (2008) reported a linear relationship between cumulative N₂O and N application rate in the semi-arid grassland of northern China, while Mc-Swiney & Robertson (2005) found a nonlinear response of N₂O flux to incremental fertilizer additions in a continuous maize (Zea mays L.) cropping system in southwest Michigan. Although the response pattern of soil N₂O emission to N fertilizers was not identical, the increased N₂O emission associated with mineral N fertilizer application is widely acknowledged (Bouwman et al., 2002). As for manure fertilizer, Davidson (2009) showed that manure has been important for N₂O emission since 1860, whereas mineral fertilizer became important only during the last half of the twentieth century, suggesting that the contribution of manure fertilizer to N₂O emission cannot be ignored. However, estimations by Flynn et al. (2005) indicated that manure makes a significantly smaller contribution than mineral N fertilizers to N₂O emissions from cropped soils. Many other studies also showed that N₂O emission due to manure were less than that due to mineral N fertilizers (Dambreville et al., 2008; Alluvione et al., 2010). Nevertheless, little research has been performed to study how N2O emissions respond to a combined application of manure and mineral N fertilizers. Combinations of manure and mineral N fertilizers are used extensively around the world.

Manure fertilizer has been extensively applied to farmland in China for more than 2000 years. In the past 50 years, with the use of chemical fertilizers, manure and chemical fertilizers were often used together to increase crop yields (Shen, 1998). For example, in the central Loess Plateau, the average manure application rate before 1960 was 22.9 ton/ha. Chemical fertilizers were not used before 1960. Yet after 1960 the manure application rate was 24.9 toN ha⁻¹ and the chemical fertilizer rate was 178 kg ha^{-1} (Hao et al., 1991). Therefore, even after chemical fertilizer applications became popular, manure still accounted for an important part of the total N applied to farmland soils in the area. Thus, the effects of manure on N₂O emissions should be investigated.

Wheat is an important contributor to the global food supply. The global wheat area in 2007 was 210 million ha, while the area in China was 23 million ha. N fertilization is fundamental for wheat production. In 1996, a total of 4.97 million tons of N fertilizer were applied for global cereal production, and wheat accounted for approximately 29% of the total (Raun and Johnson, 1999). However, the nitrogen use efficiency of wheat is relatively low and N losses attract much attention from scientists (Xing and Zhu, 2000; Silgram et al., 2001; Bouwman et al., 2005; Sudling et al., 2005; Vitousek et al., 2009). As global wheat production expands, N fertilizer applications for wheat production increase, and the N fertilizer induced N₂O emissions in wheat fields account for an increasing proportion of global N₂O emissions. Further studies of N fertilizer effects on wheat growth and N₂O emissions are needed in support of improving estimations of global N₂O emissions.

The response of N_2O emission to fertilization varied greatly with fertilization years (Hall and Matson, 1999), which could be ascribed to the unstable fertilization effects during the first several years of the experiment and the relatively stable effects during the middle or latter periods of the experiment. To avoid inconsistent treatment start-up responses of N_2O to fertilization, N_2O emissions under longterm fertilized conditions should be investigated in order to objectively determine the stable fertilization effects on N_2O emissions.

In this study, a two-year N₂O flux observation was conducted in a highland winter wheat field after 22 years fertilization in the Loess Plateau of China. The objective was to assess seasonal patterns of N₂O flux and annual emissions as affected by long-term fertilization practices in rain-fed winter wheat field plots.

2 Materials and methods

2.1 Study sites and experimental design

The long-term field experiment was initiated in September 1984 at the Agro-ecological Experiment Station of the Chinese Academy of Science, Changwu County, Shaanxi Province, China $(35^{\circ}12' \text{ N}, 107^{\circ}40' \text{ E})$. The average annual temperature of this site is 9.1 °C and annual precipitation is 585 mm. The soil is a Hei Lu soil according to the Chinese classification system, which corresponds to a Calcarid Regosol according to the FAO/UNESCO classification system (FAO/Unesco, 1988).

The cropping system was continuously cropped winter wheat (*Triticum aestivum* L.). The fertilizer treatments were unfertilized control (CK), manure (M), nitrogen (N), nitrogen + phosphorus (NP), and nitrogen + phosphorus + manure (NPM). Urea and superphosphate were used as the source of N and P. The manure came from cattle. In all of the fertilizer treatments, the N rate was 120 kg ha^{-1} , the P rate was 26.2 kg ha^{-1} , and the m rate was 75 ton ha^{-1} . The mean total N content of the manure was 1.97 g kg^{-1} and the available N was 91 mg kg^{-1} .

Fertilization treatments were replicated three times in a randomized complete block statistical design. Each plot was 10.3 m by 6.5 m. Routine crop management practices for this region were used. Prior to seeding, fertilizers were broadcast on the soil surface and then the land was plowed two times

Treatments	Organic C (Mg ha ⁻¹)	Total N (Mg ha ⁻¹)	Total P (Mg ha ⁻¹)	Available N (kg ha ⁻¹)	Available P $(kg ha^{-1})$	Available K (kg ha ⁻¹)	C/N
CK	16.5	2.3	1.3	110	13	368	7.3
М	24.1	2.6	1.5	158	65	1024	9.0
Ν	17.8	2.4	1.6	136	14	371	7.4
NP	20.1	2.3	2.0	148	36	624	8.8
NPM	24.9	2.9	2.2	182	102	849	8.9

 Table 1. Soil fertility after 22 years of fertilization management (0–20 cm).

with a cattle-drawn plow to a depth of about 10-cm. Wheat was sown in rows 20 cm apart. After seeding, the soil was raked to cover the seed. Weeds were removed by hand in all of the treatments. When the wheat reached maturity, it was harvested at the ground-level, the straw and grain were removed, and then the soil was plowed two times to a depth of about 15 cm.

2.2 Measurement of N₂O flux

The N_2O fluxes were measured from September 2006 to September 2008. The soil fertility conditions in each treatment before N_2O flux measurement are shown in Table 1.

A static closed-chamber technique was used to investigate N_2O flux. The chambers, with base area of $20 \text{ cm} \times 25 \text{ cm}$ and height of 20 cm, made of polyvinyl chloride, were inserted 10 cm into the ground in each plot. The chambers remained open (uncovered) between each measurement period. Chambers were closed (covered) to begin a measurement period. Four samples, at intervals of 15 min each, were collected from enclosed interspaces with a 25 mL glass gas-tight syringe. Tang et al. (2006) showed that greenhouse gases fluxes, including N₂O, measured at 9:00 a.m. were close to daily means, so we collected gas samples near 9:00 a.m. every 10 days in the winter and every 4–6 days in the other seasons.

A gas chromatograph (Shimadzu GC-14B), fitted with a 4 mm×3 m stainless steel column packed with Porapack Q (80–100 mesh) and an ⁶³Ni electron capture detector (ECD), was used to measure N₂O concentrations. Highpurity Ar₂/CH₄ (95% Ar₂ + 5% CH₄) was used as the carrier gas, and the flow rate was maintained at 40 mL min⁻¹. The column and the detector temperatures were set at 65 and 300 °C, respectively. The standard N₂O gas was supplied by the Japanese National Institute of Agro-Environmental Sciences.

2.3 Climate and soil environment measurements

The climate data were determined at a weather station at the Agro-ecological Experiment Station, part of the Chinese Ecosystem Research Network (CERN). Hourly values of rainfall and air temperature at a 1.2 m height were measured automatically at the weather station. Daily values, from 10:00 a.m. to 10:00 a.m. the following day, of rainfall and air temperature were determined from hourly values.

Soil temperature and moisture (volumetric water content) at a depth of 5 cm were monitored by a Delta-T Profile Probe (ML2X) and HH2 reader (Delta-T Devices Ltd, UK) in each plot at the time when gas samples were collected. Waterfilled pore space (WFPS) was calculated using the measured soil bulk density data and an assumed particle density value of 2.65 g cm^{-3} .

2.4 Statistical analysis

The frequency distributions of observed N_2O fluxes were tested. The N_2O flux data in this study showed log normal or highly skewed distributions. Therefore, the original data were log-transformed to meet the normality criteria before performing ANOVA analysis. The repeated measures ANOVA was used to test the effects of fertilization and growth stage on soil N_2O flux. A correlation analysis was conducted to assess the relationships between N_2O flux and soil temperature and WFPS for each fertilization treatment. All statistical analyses were performed using SAS software (SAS Institute, 1999).

3 Results

3.1 Overview of environmental conditions

In this study, an observation year included a winter wheat season (from September to June in the following year) and a fallow season (June to September). The mean daily air temperatures at the study site for the 2006–2007 and the 2007–2008 observation years were $10.8 \,^{\circ}$ C and $9.6 \,^{\circ}$ C, respectively. In these 2 observation years, the winter wheat seasons were characterized by relatively low air temperatures, which were $12.7 \,^{\circ}$ C and $13.9 \,^{\circ}$ C lower than those in the respective fallow seasons. Generally, the lowest daily air temperatures were observed in December and January (within the wheat season) while the highest daily air temperatures were observed in July (within the fallow season) (Fig. 1a). The rainfalls for the 2 observation years were 358



Fig. 1. Precipitation and air temperature (a), 5 cm depth soil temperature (b) and WFPS (c) for different fertilization treatments from September 2006 until September 2008.

and 509 mm, of which 55 and 53% fell during the periods of winter wheat growth (Fig. 1a). The average daily rainfalls were 0.72 and 0.99 mm for the wheat seasons, and 1.74 and 2.6 mm during the fallow seasons, respectively. Therefore, the study site was characterized by dry and cold wheat seasons and wet and warm fallow seasons.

Soil temperature (5 cm depth) varied seasonally in response to air temperature (Fig. 1b). The largest soil temperature (observed in CK) occurred in August, while the smallest soil temperature occurred (observed in NPM) in February. Soil temperatures in CK were 0-2.4 °C and 0-2.0 °C larger than in the fertilized treatment for 2006–2007 and 2007–2008 observation years depending on the sampling date. Nevertheless, the repeated measures ANOVA showed that fertilization did not significantly influence soil temperature.

Relatively large WFPS of surface soil was observed during the study period, and it varied temporally in response to rainfall and wheat growth (Fig. 1c). For the 2 observation years, WFPS was highest during the wet season and decreased during wheat growth. Soil WFPS was significantly influenced by fertilization. Generally, CK, N and NP had WFPS ranging from 37.3 to 77.1%, 34.5 to 81.5% and 35.1 to 82.8%, while m and NPM had WFPS ranging from 33.4 to 79.1%



Fig. 2. N₂O fluxes for different fertilization treatments from September 2006 until September 2008.

and 33.4 to 75.8%, respectively. The WFPS in CK, N and NP were 3 to 20% higher than in m and NPM.

3.2 Fertilization effects on N₂O fluxes

Soil N₂O fluxes were temporally variable and fertilization dependent (Fig. 2). For the CK treatment, N₂O fluxes ranged from 3 to $63 \,\mu g \, N_2 O \cdot N \, m^{-2} \, h^{-1}$, with average fluxes of 21 and $18 \,\mu g \, N_2 O \cdot N \, m^{-2} \, h^{-1}$ for 2006–2007 and 2007–2008 observation years. The largest N₂O fluxes occurred in warm and wet seasons. Application of manure slightly increased the N₂O flux. The average fluxes in the m treatment were 25 and 35% higher than those in CK for the 2 observation years, respectively. The application of N, NP and NPM significantly increased N₂O fluxes. For the observation period from 2006 to 2008, the fluxes in N, NP and NPM ranged from 4.9 to 624, 6.1 to 445, and 7.6 to 454 $\mu g \, N_2 O \cdot N \, m^{-2} \, h^{-1}$, with average fluxes of 64, 48 and 48 $\mu g \, N_2 O \cdot N \, m^{-2} \, h^{-1}$, which were 3.3, 2.5 and 2.5 times the average flux in CK.

Generally, the largest increase in N_2O flux due to fertilization was observed in the 2007–2008 observation year. The N_2O fluxes for the N and NP treatments in 2007–2008 were 1.1 times greater than those in 2006–2007.

Significant increases in N₂O flux due to fertilization occurred mainly in the first 30 days after fertilization (Table 2). For example, the average N₂O flux in the first 30 days after fertilization were 1057%, 681% and 614% larger in the N, NP and NPM treatments than in the CK treatment during the 2 observation years (2006-2008), while the average flux in the following periods (31–315 days) were 42%, 26% and 44% larger than in CK, respectively. Additionally, the effects of fertilization on N₂O fluxes in the first 30 days were

		Whole year			First 30 days after fertilization			Later period (31–365 days)		
		2006–2007	2007-2008	2006-2008	2006-2007	2007-2008	2006-2008	2006-2007	2007-2008	2006-2008
Precipitation (mm)		358	509	456	57	100	90	300	409	366
Soil temperature (°C)		10.8	9.6	10.1	13.9	10.6	12.1	10.8	9.5	9.9
	CK	21	18	20	22	24	23	21	17	19
	Μ	26	24	26	25	35	30	26	22	24
Flux (ug N ₂ O-N $m^{-2} h^{-1}$)	Ν	64	66	81	215	282	275	27	25	27
482	NP	48	48	59	141	197	183	26	20	24
	NPM	48	50	59	130	176	169	29	25	27
F		4.50	3.69	11.38	7.03	8.07	14.37	1.65	8.29	5.98
р		0.0015	0.0060	< 0.0001	< 0.0001	0.0002	< 0.0001	0.1621	< 0.0001	0.0001

Table 2. Mean N₂O fluxes during the first 30 days after fertilization and during the most of the year (the later period).

Table 3. N₂O fluxes in growing seasons and fallow season as affected by fertilization.

		Seedling	Tillering	Jointing	Booting	Heading to Maturity	Fallow season	F	р
		2006–200	7						
Precipitation (mm)		59	39	21	11	68	160		
Soil temperature (°C)		14.0	1.4	8.7	16.6	19.9	20.2		
	CK	22	15	17	15	25	26	2.48	0.0413
	М	27	20	24	22	29	32	1.68	0.1536
Flux (ug N ₂ O-N m ^{-2} h ^{-1})	Ν	232	15	15	23	29	39	12.88	< 0.0001
(PB2	NP	149	20	25	22	29	32	10.13	< 0.0001
	NPM	139	16	15	22	31	43	6.97	< 0.0001
F		7.03	5.47	11.40	2.54	0.15	1.68		
Р		0.0001	0.0006	0.0002	0.0603	0.9610	0.1626		
		2007-2008	8						
Precipitation (mm)		110	46	21	16	88	239		
Soil temperature (°C)		10.6	-0.7	10.0	15.0	19.1	19.7		
	CK	24	16	16	18	12	18	2.49	0.0432
	М	36	22	22	21	20	23	3.50	0.0087
Flux (ug N ₂ O-N m ^{-2} h ^{-1})	Ν	347	34	20	21	20	26	29.14	< 0.0001
	NP	243	22	19	18	13	25	26.71	< 0.0001
	NPM	220	19	25	27	30	31	12.11	< 0.0001
F		8.07	9.29	0.75	0.85	4.34	3.61		
Р		0.0002	< 0.0001	0.5671	0.5049	0.0069	0.0110		

dependent on temperature and rainfall. The increases were smaller in the relatively warm and dry 2006–2007 compared to the colder and wetter 2007-2008 for the M, N, NP and NPM treatments.

Relative to CK, the N treatment had the largest increases in N₂O flux with an increase range of 0–605 μ g N₂O-N m⁻² h⁻¹ during the 2 experimental years, while the m treatment had the lowest increase in flux, ranging from 0 to 38 μ g N₂O-N m⁻² h⁻¹. The increases by the NP and NPM treatments ranged from 0 to 426 μ g N₂O-N m⁻² h⁻¹ and 0 to 438 μ g N₂O-N m⁻² h⁻¹, respectively, indicating that phosphorous or manure could partly offset the single mineral N fertilizer induced increases in N_2O flux. In this study, the phosphorous and manure + phosphorous offset 36% and 35% of the increased fluxes associated with by mineral N fertilizer alone.

3.3 Seasonal patterns of N₂O fluxes as influenced by fertilization

The seasonal patterns of soil N_2O fluxes were mainly influenced by fertilization, wheat growth, and environmental conditions (Fig. 2 and Table 3). Both CK and m treatments had relatively low N_2O fluxes, and the fluxes in both treatments



Fig. 3. Changes in N_2O emissions with winter wheat growth for different fertilization treatments from September 2006 until September 2008.

could be viewed as background fluxes for the site. The background fluxes were higher in the relatively wet and warm seedling period and during the maturity stages and fallow season compared with the dryer and colder growth stages in 2006–2007. Larger background N₂O fluxes corresponded to larger soil WFPS and/or temperature. Fertilization significantly influenced treatments, N, NP, and NPM, and the fluxes were largest in the seedling stage with average values of $> 130 \,\mu g \, N_2 O \cdot N \, m^{-2} \, h^{-1}$ for both 2006–2007 and 2007–2008. However, in the other growing stages and the fallow season, the average fluxes were less than 45 $\mu g \, N_2 O \cdot N \, m^{-2} \, h^{-1}$, although they were relatively large during the dryer and warmer fallow season.

The seasonal patterns of N_2O flux varied with fertilizations. The fluxes in the fertilized treatments (M, N, NP, and NPM) were nearly always larger than in the CK treatment during the growing seasons and fallow seasons in 2006–2007 and 2007–2008. However, the fluxes in the fertilized treatments were not significantly different in the later growing stages and the fallow seasons. These results indicated that N₂O fluxes at the seedling stage were mainly controlled by nitrogen fertilization, while fluxes at the other growing stages were mainly influenced by plant and environmental conditions.

3.4 Overall emissions

The overall N₂O emissions were generally always larger in the fertilized treatments than in the CK treatment for the growing season and the fallow season (Fig. 3). Significant differences in emissions among the five treatments occurred in the seedling, tillering, jointing stages and the fallow season during 2006–2007, and in all of the growing stages and the fallow season for 2007–2008. The lowest increases of N₂O emissions during the growing season and fallow season oc-



Fig. 4. Above-ground biomass and grain yields of winter wheat for different fertilization treatments from September 2006 until September 2008.

curred in M, and the largest increases occurred in the N treatment. Although the NP and NPM treatments also resulted in increases in N_2O emissions, the increases were less than the single mineral N fertilizer (N treatment) increase, suggesting that phosphorous and manure fertilizers have the potential to inhibit the mineral N fertilizer increased N_2O emission in the studied area.

For the observation year 2007–2008, the emissions for all treatments were larger from seedling to booting and lower in heading to maturity and in the fallow season compared with 2006–2007. Totally, 191 mm and 130 mm of rain occurred from seedling to booting in 2007–2008 and 2006–2007, indicating that rainfall during this period may have had more influence on the N₂O emissions more than did the fertilizers. The effects of fertilization on emissions varied with rainfall. The annual emissions in CK and m were 10.7% and 5.1% less in the wetter 2007–2008 than in the dryer 2006–2007, while for N, NP and NPM, emissions were 41.3%, 22.1% and 26.6% larger in 2007–2008 than in 2006–2007, demonstrating that nitrogen fertilizer enhanced N₂O emissions more in the wetter year than in the dryer year, in the seedling to booting stages.



Fig. 5. Grain yield-based and grain N-yield-based N_2O -N emissions for different fertilization treatments from September 2006 until September 2008.

Because the major aim of fertilization is to increase crop yields, expressing N₂O emissions on a grain yield basis provides a useful option for evaluating fertilizer impacts. The biomass and grain yields in the NP, m and NPM treatments were significantly larger than those in the N and CK treatments for both 2006-2007 and 2007-2008 (Fig. 4). The crop grain yield-based and grain N-yield-based N₂O emissions were significantly larger in the N treatment than in the CK treatment, while those in the NP, M, and NPM treatments were lower than that in the CK treatment (except for the grain yield-based emissions in the NPM treatment in 2006–2007, which was similar to CK) (Fig. 5). Therefore, the combination of N fertilizer with P or manure improved the production and the environmental effects compared to single N fertilization alone.

4 Discussion

4.1 Fertilization effects on N₂O fluxes

In our experiment, N₂O flux peaks just after nitrogen fertilizer applications. The annual average fluxes due to fertilization were relatively large compared to reported fluxes from other ecosystems. Generally, the N₂O fluxes in forest soils and grassland are typically low, and increases associated with N fertilizer applications are also low compared with farmland (Davidson and Kingerlee, 1997, Hall and Matson, 1999; Venterea et al., 2003; Du et al., 2006; Zhang and Han, 2008). The limited effects of N fertilizer for forest and grassland soil might be due to the relatively low N application levels, while the relatively large input of N fertilizer to crop soil often results in a large N₂O flux (Wagner-Riddle et al., 2007; Barton et al., 2008; Scheer et al., 2008). Irrigation in cropped soil also enhances N₂O flux (Scheer et al., 2008). The variation of soil N₂O flux with ecosystem type and fertilization practices can be explained by changes in soil C/N ratio which is negatively related with N2O flux (Klemedtsson et al., 2005). Generally, top soils in forests and grasslands have larger C/N ratios than do farmland soils, and the soils with large C/N ratios have lower N₂O fluxes. The application of mineral N fertilizer to soil reduces the C/N ratio, and thus increases N₂O flux. Soil C/N ratios of the farmland in our site are larger than those observed by Scheer et al. (2008) and smaller than those by Wagner-Riddle et al. (2007) and Barton et al. (2008), and our fluxes in the CK and mineral N fertilized treatments are smaller than those reported by Scheer et al. (2008) and larger than those reported by Wagner-Riddle et al. (2007) and Barton et al. (2008). The application of manure fertilizer often increases the C/N ratio and consequently decreases the N₂O flux. The top soil C/N ratios followed the order of M>NPM>N while the fluxes followed the order of M<NPM<N.

The application of manure fertilizer often improves soil structure (Bronick and Lal, 2005), increases soil porosity, and decreases WFPS, which reduces the denitrification rate and thus decreases N_2O emissions. Liu et al. (2000) and Huo et al. (2008) reported that manure fertilizer has a large potential to increase soil porosity and aggregation in the Loess Plateau, which improves soil aeration. In our study, the WFPS in CK, N and NP were 3 to 20% higher than in m and NPM, which partly explains the manure effects on N_2O emissions.

Although several studies demonstrated that P promotes N_2O flux (Minami and Fukushi, 1983; Falkiner et al., 1993; Zhang and Han, 2008), we observed 24% and 27% lower fluxes in the NP treatment than in the N treatment in 2006–2007 and 2007-2008, indicating that P could alleviate single N fertilizer increases of N_2O flux. We assume that this effect is due to the fact that the NP treatment accelerated the uptake of soil mineral N which decreased soil nitrate and ammonium levels compared with the N treatment. The lower the soil N,

		СК	М	Ν	NP	NPM	All treatments	
		The whole observation period						
Soil temperature	r	0.2386	0.2058	0.0750	0.0839	0.1412	0.0833	
	p	0.0076	0.0217	0.4080	0.3544	0.1177	0.0381	
WFPS	r	0.1693	0.1243	0.3710	0.3163	0.3667	0.2760	
	p	0.0602	0.1691	< 0.0001	0.0003	< 0.0001	< 0.0001	
		The whole observation period except seedling stage						
Soil temperature	r	0.2515	0.2243	0.2170	0.2610	0.5171	0.2865	
	p	0.0097	0.0214	0.0260	0.0072	< 0.0001	< 0.0001	
WFPS	r	0.1489	0.0776	0.1562	0.1318	0.0224	0.0903	
	p	0.1294	0.4315	0.1117	0.1801	0.8208	0.0386	

Table 4. Co-relationships between N₂O fluxes and soil temperature and water-filled pore space (WFPS).

the lower the substrate for N_2O production. The uptakes of N by winter wheat in 2006–2007 and 2007–2008 were 109% and 67% higher in NP than in N, while the residual nitrate and ammonium in the top soil (0–10 cm) were 23% and 13%, and 35% and 6% less in the NP treatment than in the N treatment.

4.2 Seasonal patterns

The fertilizations significantly affected the seasonal patterns of soil N_2O fluxes, which were characterized by a peak flux within 30 days following fertilization, consistent with other findings (Hall and Matson, 1999). For the whole wheat season and fallow season, the flux was significantly related with soil temperature for each treatment. Except for the seedling period, the seasonal patterns of N_2O flux in all treatments were closely related with WFPS (Table 4), indicating that N_2O fluxes in the winter wheat fields of the study area were somewhat temperature and water dependent.

The dependence of N₂O production on soil temperature has been reported in many ecosystems (Dobbie and Smith, 2003a; Flynn et al., 2005; Wagner-Riddle et al., 2007; Zhang and Han, 2008). A threshold of 5 °C for N₂O productioN has been reported (Dobbie and Smith, 2003b). According to this threshold, there should be little N₂O flux in the littering stage. However, winter and spring thaw could magnify N₂O fluxes because freeze/thaw cycles have been related to enhanced microbial activity due to increased available carbon from freezing lysis (Christensen and Tiedje, 1990), and disintegrating aggregates (van Bochove et al., 2000), combined with sufficient soil water content during thawing (Lemke et al., 1998). Combined, these conditions can result in a relatively large flux in the littering stage. Wagner-Riddle et al. (2007) also observed increased N₂O in cold seasons, agreeing with our measurements. Additionally, large available N in soils during the stage following the seedling stage (when the fertilizer was applied) might be another reason for the higher fluxes in the nitrogen fertilized treatment.

In our study, 93% of the observed WFPS values were lower than 70%, which is the threshold for denitrification dominance (Davidson, 1992), suggesting that nitrification is likely the main source of N₂O in our study. The seasonal patterns of winter wheat growth and WFPS differed significantly with fertilization, indicating that the effects of fertilization on temporal N₂O fluxes in the growing season were mainly associated with wheat root activities and WFPS changes, which alter soil microorganism activities in N₂O production together with soil C and N conditions. The effects in the fallow season were dominated by the WFPS and soil C and N conditions.

5 Implications for N₂O emission and fertilization

At the global scale, annual N₂O emissions from cropped mineral soils ranged from 0.3 to $10.7 \text{ kg N}_2\text{O-N} \text{ ha}^{-1} \text{ yr}^{-1}$ (Stehfest and Bouwman, 2006). For wheat soils, the emission ranged from 0.5 to $3.7 \text{ kg N}_2\text{O-N} \text{ ha}^{-1} \text{ yr}^{-1}$ regardless of irrigation or not, while more than 75% of these observations were less than $1.6 \text{ kg N}_2\text{O-N} \text{ ha}^{-1} \text{ yr}^{-1}$ (Roelandt et al., 2005). The background emissions (CK treatment) in our rain-fed winter wheat field were 1.7 and $1.9 \text{ kg N}_2\text{O-N} \text{ ha}^{-1} \text{ yr}^{-1}$, while our fertilized emissions were 2.1-3.4 and $3.2-7.5 \text{ kg N}_2\text{O-N} \text{ ha}^{-1} \text{ yr}^{-1}$ for the 2006–2007 and 2007–2008 observation years. These were relatively large emission values.

The annual N₂O emissions from fertilized treatments were in the following order N>NPM and NP>>M, suggesting that the contribution of single N fertilizer alone was larger than that of NP and NPM. This finding agrees with other reports (Flynn et al., 2005; Dambreville et al., 2008; Alluvione et al., 2010). This result implies that the present assessment might be larger because manure and phosphorous fertilizers are widely used in agriculture together with mineral N fertilizer while its role in offsetting mineral N fertilizers induced N₂O emissions are usually neglected. Barton et al. (2008) also suggested that the observed emission factor in semiarid cropped soil is 60 times less than the values suggested by IPCC. Many estimations separate fertilizer N as mineral N and organic N (from manure fertilizer) (Flynn et al., 2005, Davidson, 2009) without considering the fact that organic N and mineral N and mineral N and P are often used together. These estimations often ignore the offsetting role of manure when it is applied together with mineral N fertilizers.

Our results further show that the m treatment had relatively large biomass and grain yield and relatively low N_2O fluxes and annual emissions. From the point of agricultural production and N_2O emission, M is recommended while single N fertilization alone is not recommended for highland winter wheat, when fertilizers are applied at the time of planting.

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