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Nitrous oxide emissions and maize yield as influenced by nitrogen fertilization and tillage operations in upland soil

Sung Un Kim^{1,2†}, Hyun Ho Lee^{1†}, Sung Min Moon¹, Hae Ri Han¹ and Chang Oh Hong^{1,2*}

Abstract

Previous studies simply focused on determining nitrous oxide (N₂O) emissions from the soil under different tillage operations and nitrogen (N) fertilizations without considering crop yield. Therefore, the objective of this study was to determine the effects of different tillage operations and N fertilizations on N₂O emissions and crop yield from upland soil. Two different tillage operations [conventional tillage (CT) and no-tillage (NT)] and N fertilizations [without urea (WOU) and with 186 kg N ha⁻¹ of urea (WU)] were established in a randomized block design with three replications on upland soil. Maize (*Zea mays*) was cultivated from 6th July to 4th October, 2018 (year 1), and from 15th April to 26th July, 2019 (year 2). The daily N₂O flux did not peak soon after tillage operation and N fertilization, but it was more related to the change in water-filled pore space (WFPS). The mean value of WFPS across N fertilizations and seasons (years) was higher in CT than in NT. The changes of nitrification and denitrification rates could be attributed to the differences in WFPS between CT and NT. Nitrification was the predominant process producing N₂O with CT, but denitrification was with NT. The application of urea increased cumulative N₂O emissions, while CT also increased it compared with NT. The order of the mean values of cumulative N₂O emissions across seasons from the highest to the lowest was as follows: CT + WU (7.12 kg N₂O ha⁻¹ year⁻¹) > NT + WU (5.69 kg N₂O ha⁻¹ year⁻¹) ≥ CT + WOU (5.02 kg N₂O ha⁻¹ year⁻¹) > NT + WOU (4.24 kg N₂O ha⁻¹ year⁻¹). Tillage operation did not affect the grain yield of maize or yield-scaled N₂O emissions (YSNE). However, the application of urea increased the grain yield of maize and decreased YSNE, implying it could reduce N₂O emission per unit of maize grain production. No-tillage management did not decrease YSNE value compared to CT operation, but N fertilization significantly decreased YSNE in the current study.

Keywords: Nitrous oxide, Tillage, Upland soil, Urea, Yield-scale N₂O emission

Introduction

Agricultural soils are the largest anthropogenic source of nitrous oxide (N₂O), which has 310 times greater global warming potential than carbon dioxide over a 100-year timeframe [1] (IPCC 2019). In addition, N₂O concentration in the atmosphere is increasing at 0.73 ppb per year due to anthropogenic activity [2, 3]. Annual N₂O

emission across the globe is 6.7 Tg N₂O year⁻¹, 60% of which is attributable to agricultural soil.

Agricultural soil is a complex environment, in which various microbial pathways are involved in the production and consumption of N₂O [4, 5]. Microbial processes producing N₂O, such as nitrification and denitrification, which depend on inorganic nitrogen (N) as their substrate, are responsible for most of the N₂O emitted from arable soil [6]. Therefore, the application of inorganic N fertilizer to arable soil may increase the rates of both microbial processes and N₂O emission.

Nitrous oxide emission is also affected by changes in the physical properties of soil. Bulk density associated

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with water holding capacity and aeration and different amounts of precipitation may change the percentage of water-filled pore space (WFPS) [7]. Change of WFPS is one of important factors affecting N₂O emissions from arable soil [8–11]. Nitrous oxide can be emitted by nitrification with a soil WFPS of 35–60%, which requires ammonium as an inorganic N substrate for aerobic respiration, whereas a soil WFPS above 60% (when O₂ is limited) induces a switch from aerobic to anaerobic respiration. Therefore, nitrate is an alternative electron acceptor used by microorganisms associated with denitrification that produces N₂O [10, 12–14]. Tillage operations directly cause physical changes in arable soil. No-tillage operation is an optimal form of management for improving the physical properties of arable soil, and is eco-friendly and economically favorable. Therefore, no-tillage and tillage operations may affect N₂O emission from arable soil differently.

It is crucial to reduce environmental pollution without compromising food security in the context of an increasing global population [15, 16]. Future sustainable agriculture should explore low N₂O emissions at high crop productivity for food security. Agricultural practices can be related to N₂O emission based on crop yield, referred to as yield-scaled N₂O emission (YSNE). There are many studies observed the effect of tillage operations and N fertilization on N₂O emission in the arable land [17–21]. Therefore, the objective of this study was to determine the effects of different tillage operations and N fertilizations on N₂O emission and crop yield from upland soil. To this end, changes of inorganic N, physical properties of soil, and YSNE were measured in a maize field for two consecutive years.

Materials and methods

Site description and experimental design

This study was conducted on upland soil located in Cheonghak-ri, Samrangjin-eup, Miryang City, Gyeongnam Province, South Korea (35°26′59″N; 128°48′30″E). The soil was well drained with 2–3% slope and its texture was loam (fine loamy, mixed, mesic family of Anthraquic Hapludalfs), containing 42.2% sand, 39.3% silt, and 18.5% clay. The specific chemical properties of the studied soil are shown in Table 1. Averaged annual precipitation and temperature have been 1215 mm and 12.9°C over the last decade, respectively. Seasonal precipitation data were collected from an automatic weather station in Miryang (Korea Meteorological Administration), which was 5 km away from the experimental site. The upland soil selected for this study was cultivated pepper for 7 years before October 2015. This field had not been cultivated since the end of October 2015, and the experimental field was established in November 2017 to compare different tillage operations [conventional tillage (CT) and

Table 1 Selected characteristics of the studied soil (n = 3)

Parameters	Value
pH (1:5 with H ₂ O)	6.89
Total organic carbon (g kg ⁻¹)	10.6
Total nitrogen (g kg ⁻¹)	1.55
C/N ratio	4.48
Inorganic nitrogen	
NH ₄ ⁺ (mg kg ⁻¹)	7.79
NO ₃ ⁻ (mg kg ⁻¹)	5.01
Available phosphorus (mg kg ⁻¹)	107
Bulk density (g cm ⁻³)	1.23
Exchangeable cation	
K (cmol _c kg ⁻¹)	0.62
Ca (cmol _c kg ⁻¹)	7.59
Mg (cmol _c kg ⁻¹)	1.91
Cation exchangeable capacity (cmol _c kg ⁻¹)	10.45
Soil separate	
Sand (%)	42.2
Silt (%)	39.3
Clay (%)	18.5
Soil texture	Loam

no-tillage (NT)] and N fertilizations [without urea (WOU) and with urea (WU)]. The present experiment included four treatments: CT + WOU, CT + WU, NT + WOU, and NT + WU. In the CT plots, tillage operations were conducted using a moldboard plough (20 cm deep shank with 60 cm spacing) before transplanting and after harvest. In the NT plots, no ploughing was performed and transplanting involved minimal soil disturbance. The experiments were arranged in a randomized block design with four replications and a plot size of 12 m² (3 × 4 m). Immediately after tillage operation, maize (*Zea mays* L.) seeds were sown in all plots on 2nd June, 2018. Unfortunately, the germination rate of maize was below 10% due to flooding. Because the total amount of precipitation was 273 mm from 10th June, 2018, to 5th July, 2018, we also irrigated a total of 80 mm between 2nd and 5th June after seeding. Therefore, surviving maize sprouts were removed from all plots and then maize seedlings grown in the greenhouse for 30 days were transplanted in all plots on 26th July, 2018, for year 1. In the same manner, maize was transplanted on 10th May, 2019, for year 2. In both years, maize was planted at a rate of 56,000 seeds ha⁻¹ with 30 × 60 cm between rows. A total of 93 kg N ha⁻¹ of urea as basal N fertilization was applied only to plots with urea (CT + WU and NT + WU) on 2nd June, 2018, while 186 kg N ha⁻¹ of urea as both basal and additional N fertilizations was applied to the same plots on 26th July, 2018, for year 1. For year 2, urea was once applied without split-addition at a rate of 186 kg N ha⁻¹ on 8 May, 2019. In addition, 35 kg P₂O₅ ha⁻¹ of fused phosphate and 74 kg K₂O

ha⁻¹ of potassium chloride were applied to the entire plot areas on 2nd June, 2018, and 8th May, 2019, for years 1 and 2, respectively. All maize in the entire plot was harvested on 4th October, 2018, and 26th July, 2019, for year 1 and year 2, respectively. The detailed information concerning the field management plan was shown in Table 2.

Nitrous oxide emission from soil

Daily N₂O flux and cumulative N₂O emission were measured by a closed chamber method [22] across the year. A static collar made of a PVC column (headspace; 24.8 cm diameter × 17 cm height) was installed at the center of each plot on 2nd November, 2017. The tops were closed to lids, and samples were collected after 0, 20, and 40 min using a 20 ml polypropylene syringe, and were transferred into 12 ml evacuated glass vials (Exetainer® 12 ml vial-evacuated 838 W; Labco, UK). The gas sampling was conducted twice each week during the growing season and once a week after the harvesting of maize. Based on previous studies [23], gas samples were collected from 10:00 a.m. to 12:00 a.m. throughout the year. The weeds and plant residues were eliminated in the chamber and each chamber was left open in the field throughout the experimental period. The temperature in the chamber during gas sampling was measured using a portable thermometer (WT-1; Elitech, UK). Nitrous oxide concentration was analyzed using a gas chromatograph mass spectrometer (GC-MS OP2020, Shimadzu, HP-PLOT Q Column). The N₂O flux for a day (N₂O g ha⁻¹ day⁻¹) and cumulative N₂O emission for a year (N₂O kg ha⁻¹ year⁻¹) were calculated using the following equations:

$$N_2O\ flux(N_2O\ g\ ha^{-1}\ day^{-1}) = (\Delta g/\Delta t) \times d \times (273/T) \times (V/A) \times k \times a,$$

$$Cumulative\ N_2O\ emissions(N_2O\ kg\ ha^{-1}\ year^{-1}) = \sum_i^n (R_i \times D_i),$$

where $\Delta g/\Delta t$ is the rate of change in gas concentration inside the chamber (g m⁻³ min⁻¹), d is the gas density (g m⁻³) at 273 K and 0.101 MPa pressure, T is the air temperature (K) inside the chamber, V is the volume of the chamber (m³), A is the surface area circumscribed by the chamber (m²), k is the time conversion factor (min day⁻¹), and a is the area convection coefficient (10,000 m² ha⁻¹). The air temperature measured in the chamber at the time of sampling was used to calculate fluxes. Cumulative N₂O emissions during the experimental period were calculated by multiplying the mean value of N₂O flux (N₂O g ha⁻¹ day⁻¹) (R_i) by the length of the period (D_i) and adding that amount to the previous cumulative total.

After estimating the dry mass of grain yield grown in each plot, YSNE was calculated by dividing cumulative N₂O emission by dried grain biomass of maize as follows.

$$YSNE\ (kg\ N_2O\ Mg^{-1}\ yield) = \frac{Cumulative\ N_2O\ emission}{Dried\ grain\ biomass}.$$

Soil sampling and analysis

Soil samples were collected once a month during the crop growing season and using a hand auger (0–15 cm depth) and core sampler (100 cm³) to analyze the soil properties. To determine the concentration of total N in soil and maize, it was quantified using an automated Kjeldahl analyzer (JP Selecta, PRO-NITRO-S, Spain). Measurements of the concentrations of inorganic N (NH₄⁺ and NO₃⁻) were performed using ion chromatography (Seal Analytical, AA500 Autoanalyzer, Germany).

Table 2 Treatment and field management practice of this study

Treatment	Tillage operation	Urea application	N fertilization rate (kg N ha ⁻¹)	
			Year 1	Year 2
CT+WOU	Conventional tillage (CT)	Without urea (WOU)	0	0
CT+WU	Conventional tillage (CT)	With urea (WU)	279	186
NT+WOU	No-tillage (NT)	Without urea (WOU)	0	0
NT+WU	No-tillage (NT)	With urea (WU)	279	186

Daily averaged volumetric water content from 0 to 10 cm depth was measured using a static soil moisture sensor (5TE Water Content, Temperature, and Electrical Conductivity; Decagon, USA) installed horizontally at a depth of 10 cm near the anchor. Soil bulk density was determined using a metal core can (100 cm³) to sample the undisturbed soil structure, and the sampled soil in the core was dried at 105 °C for 24 h [24]. Soil WFPS was calculated from the volumetric water content and the soil bulk density using the following equation:

$$WFPS = (\theta/soil\ porosity) \times 100,$$

where θ is the volumetric water content ($\text{cm}^3 \text{cm}^{-3}$). Soil porosity ($\text{cm}^3 \text{cm}^{-3}$) was calculated using a particle density value of 2.65g cm^{-3} .

Nitrification and denitrification rates were calculated from a substrate of the processes and its product using the following equation:

$$\text{Nitrification ratio} = \text{Nitrate}(\text{NO}_3^-) \text{ concentration} / \text{Ammonium}(\text{NH}_4^+) \text{ concentration},$$

$$\text{Denitrification ratio} = \text{Cumulative N}_2\text{O emission} / \text{Nitrate}(\text{NO}_3^-) \text{ concentration},$$

where the NH_4^+ and NO_3^- concentrations applied in the above equation were analyzed using soil after harvesting maize.

Statistical analysis

Mean values of cumulative N_2O emission, grain yield, and yield-scaled N_2O emission were analyzed by pairwise comparison. The least significant difference test (LSD) was used for multiple comparisons between the means, and performed only when the F-test result was significant in the range of $P < 0.05$. Statistical analysis was performed using Statistix software (version 9.0) (Statistix, 2008).

Results and discussion

Daily nitrous oxide flux

The change in N_2O flux was monitored for 2 years from November 2017 to October 2019 under different N fertilizations and tillage operations (Fig. 1a). The trend of N_2O flux over this timeframe was not similar to those of air and soil temperatures (Fig. 1a, b). Although the flux was relatively low during the cold and dry fallow season, it then peaked as the temperature reached its maximum in August 2018, but it did not peak in August 2019. Precipitation is one of the factors affecting N_2O emission from arable soil. Some studies have reported that N_2O flux peaked soon after a high-rainfall event, which induced soil to adopt an anaerobic state for N_2O production through denitrification [25]. However, in the current study, the peak of N_2O flux did not appear after high-rainfall events, despite the fact there were several such events over the 2 years. This result could be interpreted through the change in products of denitrification process depend on different WFPS [26, 27]. Denitrification is completely performed until N_2 instead of N_2O or NO in the above 75% of WFPS [28]. Thus the ratio of $\text{N}_2/\text{N}_2\text{O}$ increased in above 80% WFPS [27]. The N_2O flux did not peak soon after the application of urea, but increased dramatically 12 and 4 days after irrigation on 26th July, 2018, and 9th May, 2019, respectively, when the average

daily WFPS was near 60% (Fig. 1a, c). Water-filled pore spaces were 61% and 63% on 8th August, 2018, and 5th May, 2019, respectively (Fig. 1c). The daily N_2O flux may be related to the soil WFPS. Nitrification is the predominant process for N_2O production from soil at $< 60\%$ WFPS, whereas denitrification is the predominant pro-

cess at $> 60\%$ WFPS [26, 29, 30]. When the soil WFPS is $\sim 60\%$, N_2O is produced through both nitrification and denitrification [31–33]. The daily N_2O flux may increase dramatically because of both processes occurring simultaneously when the soil WFPS increases to near 60%, as found in this study. Based on the above results, N_2O flux was more related to the application of urea and WFPS rather than climatic events such as changes of temperature and precipitation in the current study.

Cumulative nitrous oxide emission

Tillage operation significantly affected the cumulative N_2O emission from soil (Table 3). The mean value of cumulative N_2O emission across N fertilizations and years with CT was higher than that with NT (Table 4). It was 5.90 and $5.14 \text{kg ha}^{-1} \text{year}^{-1}$ for CT and NT, respectively. Tillage operations directly affect the physical properties of soil, such as its bulk density. The changes of bulk density with different tillage operations directly affected WFPS, which was calculated with reference to soil bulk density. This indicated that the WFPS value could differ depending on soil bulk density, even with the same rainfall and irrigation. As shown in Table 5, the mean bulk density during the growing season of maize across N fertilizations and years with CT was significantly lower than that with NT. This implied that the soil porosity developed with CT and WFPS should have decreased. However, the mean value of WFPS across N fertilizations and years with CT was markedly higher than that with NT (Table 6). This is not surprising because tillage operation causes less downward movement of percolating water in the soil profile than no tillage does [34]. Continuous tillage operation causes the soil compaction below 20 cm of soil depth, which is the main reason for forming hardpan [35]. In a corn field study conducted for 3 years, Patni et al. observed that leachate drained 46% more under NT treatment than under CT treatment [36]. Therefore, WFPS could increase with CT in upland fields due to poor drainage. This higher WFPS with CT might provide soil water conditions that are more favorable to

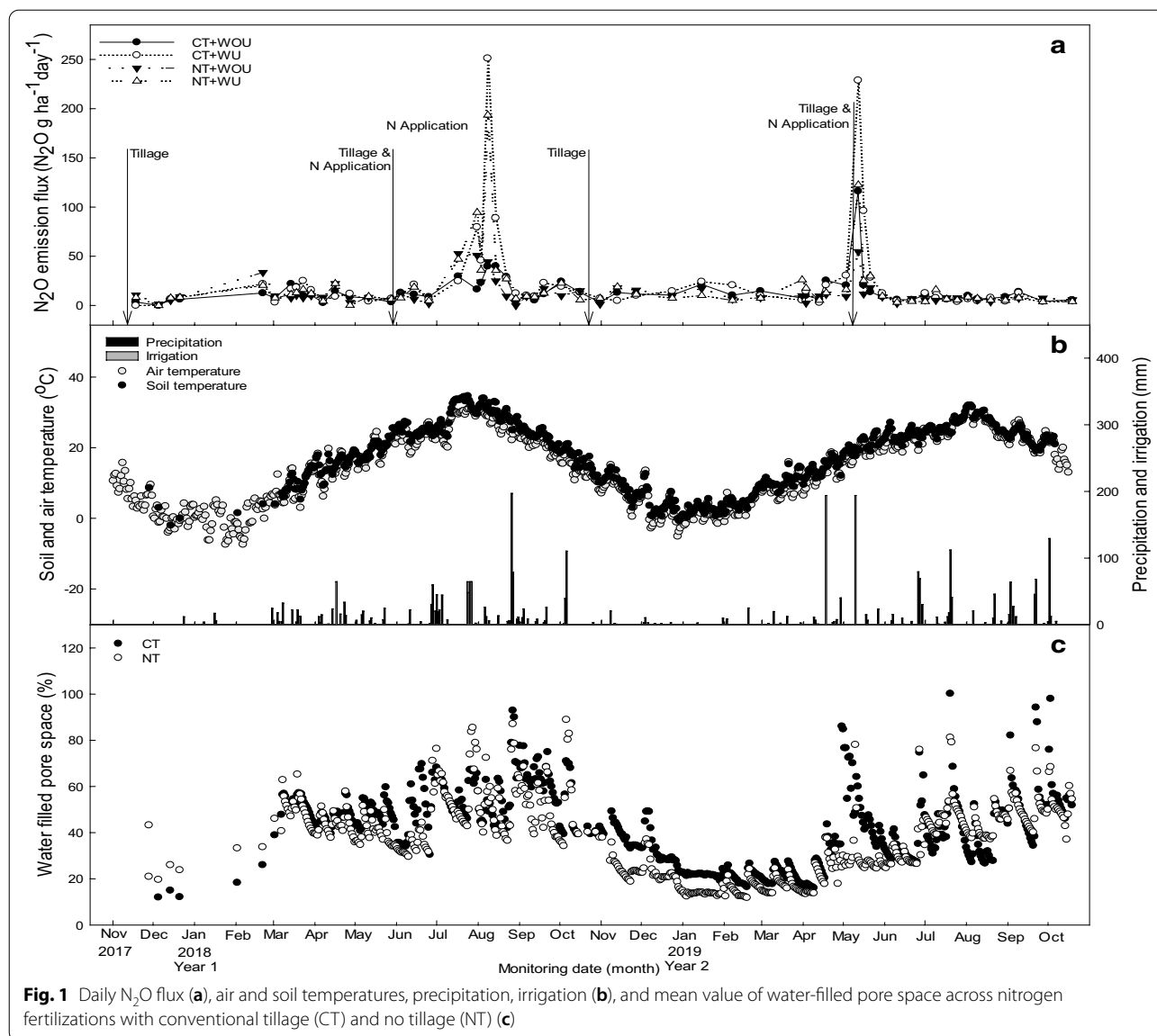


Fig. 1 Daily N₂O flux (a), air and soil temperatures, precipitation, irrigation (b), and mean value of water-filled pore space across nitrogen fertilizations with conventional tillage (CT) and no tillage (NT) (c)

Table 3 ANOVA and P-value of cumulative N₂O emission, grain yield, yield-scaled N₂O emission, bulk density, and water-filled pore space

Item	df	Cumulative N ₂ O emission	Grain yield	Yield-scaled N ₂ O emission	Bulk density	Water-filled pore space
Tillage operations (T)	1	0.002	NS	NS	< 0.001	< 0.001
Nitrogen fertilizations (N)	1	< 0.001	< 0.001	0.011	NS	NS
Year (Y)	1	< 0.001	0.005	NS	NS	< 0.001
T × N	1	0.003	NS	NS	NS	NS
T × Y	1	< 0.001	0.004	NS	NS	NS
N × Y	1	NS	< 0.004	< 0.001	NS	NS
T × N × Y	1	NS	NS	NS	NS	NS

NS not significant

Table 4 Cumulative N₂O emission from soil with different tillage operations (T), nitrogen fertilizations (N), and years (Y)

Year	Year 1			Year 2			Y mean ¹		
	Tillage operations	CT	NT	T mean	CT	NT	T mean	CT	NT
Nitrogen fertilizations									
Cumulative N₂O (kg ha⁻¹ year⁻¹)									
WOU	4.30 ^b (0.08)	5.73 ^a (0.26)	5.02 ^B (0.30)	4.99 ^a (0.22)	3.50 ^b (0.19)	4.24 ^B (0.31)	4.65 ^a (0.17)	4.61 ^a (0.45)	4.63 ^B (0.23)
WU	7.44 ^a (0.54)	6.79 ^a (0.31)	7.12 ^A (0.31)	6.85 ^a (0.27)	4.53 ^b (0.29)	5.69 ^A (0.47)	7.14 ^a (0.30)	5.66 ^b (0.47)	6.40 ^A (0.33)
N mean ³	5.87 ^a (0.64)	6.26 ^a (0.27)		5.92 ^a (0.39)	4.02 ^b (0.25)		5.90 ^a (0.36)	5.14 ^b (0.34)	
T × N mean ⁴	6.07 ^a (0.34)			4.97 ^b (0.33)					

Different lower- and upper-case letters denote significant differences at $p < 0.05$ in column and row comparisons, respectively

¹ Y mean: mean value across years

² T × Y: mean value across tillage operations and years

³ N mean: mean value across N fertilizations

⁴ T × N: mean value across tillage operations and N fertilizations

Standard error in brackets

Table 5 Soil bulk density during growing season of maize with different tillage operations (T), nitrogen fertilizations (N), and years (Y)

Year	Year 1			Year 2			Y mean ¹		
	Tillage operations	CT	NT	T mean	CT	NT	T mean	CT	NT
Nitrogen fertilizations									
Bulk density (g cm⁻¹)									
WOU	1.16 ^b (0.01)	1.38 ^a (0.07)	1.27 ^A (0.05)	1.14 ^b (0.07)	1.36 ^a (0.09)	1.25 ^A (0.06)	1.15 ^b (0.03)	1.39 ^a (0.06)	1.26 ^A (0.04)
WU	1.20 ^b (0.12)	1.40 ^a (0.09)	1.30 ^A (0.07)	1.18 ^b (0.08)	1.38 ^a (0.09)	1.28 ^A (0.06)	1.19 ^b (0.06)	1.37 ^a (0.05)	1.29 ^A (0.05)
N mean ³	1.18 ^b (0.05)	1.39 ^a (0.05)		1.16 ^b (0.05)	1.38 ^a (0.06)		1.17 ^b (0.03)	1.38 ^a (0.04)	
T × N mean ⁴	1.29 ^a (0.05)			1.27 ^a (0.05)					

Standard error in brackets. Different lower- and upper-case letters denote significant differences at $p < 0.05$ in column and row comparisons, respectively

¹ Y mean: mean value across years

² T × Y: mean value across tillage operations and years

³ N mean: mean value across N fertilizations

⁴ T × N: mean value across tillage operations and N fertilizations

Table 6 Water-filled pore space during growing season of maize with different tillage operations (T), nitrogen fertilizations (N), and years (Y)

Year	Year 1			Year 2			Y mean ¹		
	Tillage operations	CT	NT	T mean	CT	NT	T mean	CT	NT
Nitrogen fertilizations									
Water-filled pore space (%)									
WOU	56.4 ^a (3.22)	50.0 ^b (1.05)	53.2 ^A (1.80)	38.4 ^a (3.55)	29.9 ^b (3.21)	34.2 ^A (2.48)	47.4 ^a (4.55)	40.0 ^b (4.74)	43.7 ^A (3.33)
WU	57.0 ^a (2.68)	48.8 ^b (3.52)	52.9 ^A (2.33)	39.7 ^a (1.44)	29.8 ^b (1.79)	34.8 ^A (2.13)	48.4 ^a (4.09)	39.3 ^b (4.62)	43.8 ^A (3.24)
N mean ³	56.7 ^a (1.88)	49.4 ^b (1.67)		39.1 ^a (1.74)	29.8 ^b (1.64)		47.9 ^a (2.92)	39.6 ^b (3.16)	
T × N mean ⁴	53.1 ^a (1.62)			34.5 ^b (1.80)					

Standard error in brackets. Different lower- and upper-case letters denote significant differences at $p < 0.05$ in column and row comparisons, respectively

¹ Y mean: mean value across years

² T × Y: mean value across tillage operations and years

³ N mean: mean value across N fertilizations

⁴ T × N: mean value across tillage operations and N fertilizations

microorganisms associated with N₂O-producing processes such as nitrification and denitrification than with NT.

Nitrogen fertilization significantly affected cumulative N₂O emission (Table 3). The mean cumulative N₂O emission across tillage operations and years with WU was significantly higher than that with WOU (Table 4; 4.63 and 6.40 kg ha⁻¹ year⁻¹ for WOU and WU, respectively). The application of urea provides inorganic N (NH₄⁺ and NO₃⁻) as a substrate for nitrification and denitrification, which are responsible for most of the N₂O emitted from arable soil. The mean NH₄⁺ and NO₃⁻ concentrations in soil after transplanting across tillage operations and years with WU were significantly higher than those with WOU (Tables 7, 8).

Year significantly affected cumulative N₂O emission (Table 3). The mean cumulative N₂O emission across tillage operations and N fertilizations in year 1 was significantly higher than that in year 2 (Table 4). As mentioned above, different rates of urea were applied in the 2 years. In total, 279 and 186 kg N ha⁻¹ of urea were applied in year 1 and year 2, respectively. The greater supply of inorganic N to the soil increased the cumulative N₂O emission in year 1. In addition, different soil water contents might have affected the cumulative N₂O emission in the 2 years. The annual precipitation levels were 1300 and 1177 mm in year 1 and year 2, respectively. In addition, the mean WFPS across tillage operations and N fertilizations in year 1 was significantly higher than that in year 2 (Table 6). It was 53.1% and 34.5% in year 1 and year 2,

Table 7 Ammonium (NH₄⁺) concentrations in soil after transplanting with different tillages (T), nitrogen fertilizations (N), and years (Y)

Year	Year 1			Year 2			Y mean ¹		
	CT	NT	T mean	CT	NT	T mean	CT	NT	T × Y mean ²
Nitrogen fertilizations									
Ammonium (NH₄⁺) (mg kg⁻¹)									
WOU	7.12 ^a (0.15)	7.23 ^a (0.16)	7.17 ^A (0.10)	17.24 ^b (0.23)	20.51 ^a (0.11)	18.87 ^A (0.71)	12.18 ^b (1.92)	13.87 ^a (2.51)	13.02 ^B (1.54)
WU	8.27 ^a (1.04)	7.58 ^a (0.15)	7.92 ^A (0.50)	18.00 ^b (0.27)	21.19 ^a (0.75)	19.60 ^A (0.71)	13.13 ^a (1.90)	14.39 ^a (2.60)	13.76 ^A (1.56)
N mean ³	7.69 ^a (0.53)	7.41 ^a (0.12)		17.62 ^b (0.22)	20.85 ^a (0.37)		12.66 ^b (1.31)	14.13 ^a (1.56)	
T × N mean ⁴	7.55 ^b (0.27)			19.23 ^a (0.47)					

Standard error in brackets. Different lower- and upper-case letters denote significant differences at *p* < 0.05 in column and row comparisons, respectively

¹ Y mean: mean value across years

² T × Y: mean value across tillage operations and years

³ N mean: mean value across N fertilizations

⁴ T × N: mean value across tillage operations and N fertilizations

Table 8 Nitrate (NO₃⁻) concentrations in soil after transplanting with different tillage operations (T), nitrogen fertilizations (N), and years (Y)

Year	Year 1			Year 2			Y mean ¹		
	CT	NT	T mean	CT	NT	T mean	CT	NT	T × Y mean ²
Nitrate (NO₃⁻) (mg kg⁻¹)									
WOU	22.20 ^a (0.85)	4.92 ^b (1.45)	13.56 ^A (3.36)	2.67 ^a (0.27)	2.58 ^a (0.25)	2.63 ^B (0.17)	12.44 ^a (3.71)	3.75 ^b (0.81)	8.09 ^B (0.50)
WU	23.40 ^a (0.01)	4.20 ^b (0.07)	13.80 ^A (3.63)	5.30 ^a (1.17)	6.29 ^a (0.66)	5.79 ^A (0.65)	14.35 ^a (3.46)	5.25 ^b (1.41)	9.80 ^A (0.40)
N mean ³	22.80 ^a (0.45)	4.56 ^b (0.69)		3.99 ^a (0.75)	4.43 ^a (0.77)		13.39 ^A (2.21)	4.50 ^B (0.40)	
T × N mean ⁴	13.68 ^a (2.39)			4.21 ^b (0.52)					

Standard error in brackets. Different lower- and upper-case letters denote significant differences at *p* < 0.05 in column and row comparisons, respectively

¹ Y mean: mean value across years

² T × Y: mean value across tillage operations and years

³ N mean: mean value across N fertilizations

⁴ T × N: mean value across tillage operations and N fertilizations

respectively. The greater soil water content in year 1 produced more N₂O than in year 2. Ammonium and NO₃⁻ produced through the application of urea might have been consumed by different microbial processes in the 2 years due to the difference in WFPS between them. As shown in Fig. 2, the mean nitrification ratio across N fertilizations with CT was higher in year 2 than in year 1. Lower WFPS could provide more aerobic conditions to promote microbial activity associated with nitrification in year 2 than in year 1. Nitrous oxide can be emitted by nitrification with a soil WFPS of 35–60%, which requires NH₄⁺ for aerobic respiration [10, 12–14]. Most of the daily WFPS with CT during the growing season of maize in year 2 ranged from 35 to 60% (Fig. 1c). Soil WFPS above 60% induces a switch from aerobic respiration to anaerobic respiration. Therefore, NO₃⁻ is an alternative electron acceptor used by microorganisms associated with denitrification that produces N₂O. As shown in Fig. 1c, most of the daily WFPS with CT during the growing season of maize in year 1 was near 60%. Nitrification might have been the predominant process by which N₂O was produced in year 2, but denitrification was in year 1. This was evident by the relationships between daily N₂O flux, NH₄⁺ and NO₃⁻ concentration in soil (Fig. 3). Daily N₂O flux was more positively correlated with NO₃⁻ in year 1, but with NH₄⁺ in year 2.

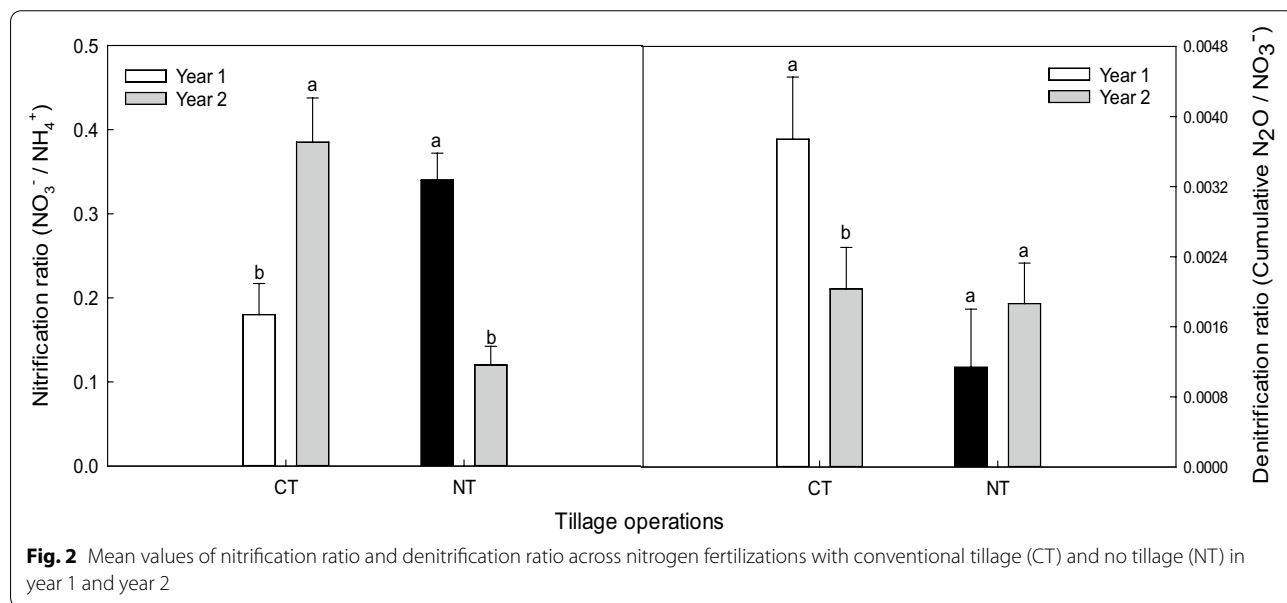
Interestingly, the mean nitrification ratio across N fertilizations with NT was significantly higher in year 1 than in year 2, but the mean denitrification ratio did not differ significantly between the 2 years, despite the fact that the WFPS of soil was higher in year 1 than in year 2 (Fig. 2). The mean values of WFPS across N fertilizations with

NT in year 1 and year 2 were 49.4% and 29.8%, respectively (Table 6). WFPS of 35–60% constitutes favorable soil water conditions for nitrification [26, 37]. However, both nitrification and denitrification become slow in water-limited conditions involving WFPS of < 35% [26, 37].

There was a significant tillage × N fertilization interaction for cumulative N₂O emission (Table 3). The mean values of cumulative N₂O emission across years between CT and NT under WOU did not differ significantly (Table 4). However, such emission was lower with NT under WU than with CT. This implies that NT operation had a pronounced effect on reducing N₂O emission with N fertilization. A higher level of inorganic N through urea application was likely to be transformed into N₂O with the elevation of WFPS by tillage. The mean value of WFPS across N fertilizations and years with CT was significantly higher than that with NT (Table 6).

Maize grain yield and yield-scaled N₂O emission

Nitrogen fertilization significantly affected the grain yield of maize and YSNE, but tillage operation did not (Table 3). The mean value of the grain yield of maize across tillage operations and years with WU was significantly higher than that with WOU (Table 9). However, the mean value of YSNE across tillage operations and years with WU was significantly lower than that with WOU, despite the fact that the mean value of cumulative N₂O emission across tillage operations and years with WU was higher than that with WOU (Tables 4, 10). The greater rate of increase of the grain yield of maize than the rate of cumulative N₂O emission with



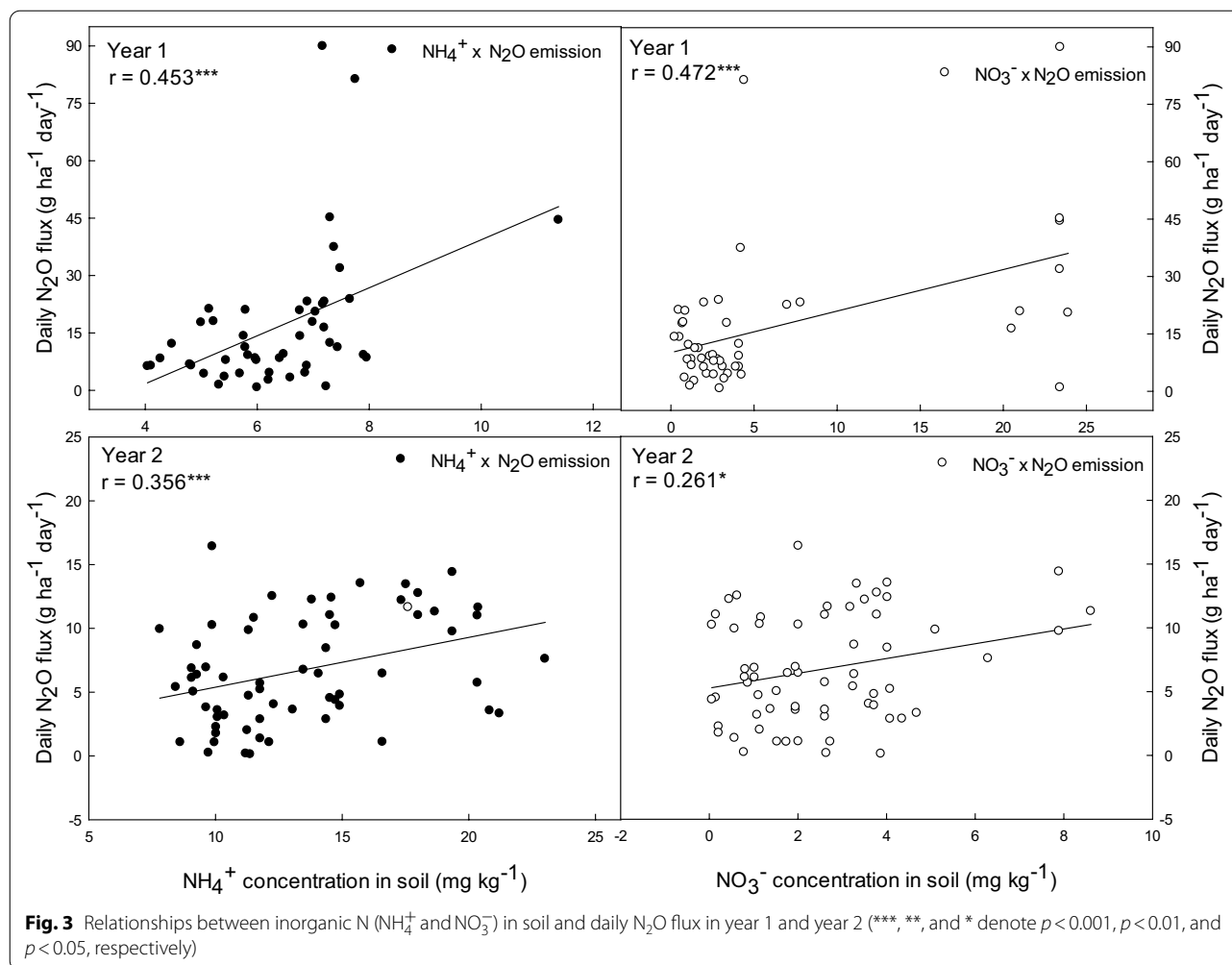


Table 9 Grain yield amended with different tillage operations (T), nitrogen fertilizations (N), and years (Y)

Year	Year 1			Year 2			Y mean ¹		
	CT	NT	N mean	CT	NT	N mean	CT	NT	T x Y mean ²
Grain yield (Mg ha⁻¹)									
WOU	3.93 ^b (0.86)	6.02 ^a (0.56)	4.97 ^A (0.62)	2.85 ^a (0.22)	1.50 ^b (0.04)	2.17 ^B (0.28)	3.39 ^a (0.46)	3.76 ^a (0.89)	3.57 ^B (0.42)
WU	5.41 ^a (1.01)	5.96 ^a (0.49)	5.68 ^A (0.56)	6.63 ^a (0.20)	4.87 ^b (0.67)	5.75 ^A (0.47)	6.02 ^a (0.56)	5.41 ^a (0.44)	5.72 ^A (0.57)
N mean ³	4.67 ^a (0.70)	5.99 ^a (0.35)		4.74 ^a (0.73)	3.18 ^b (0.71)		4.70 ^a (0.45)	4.59 ^a (0.57)	
T x N mean ⁴	5.33 ^a (0.41)			3.96 ^b (0.53)					

Standard error in brackets. Different lower- and upper-case letters denote significant differences at $p < 0.05$ in column and row comparisons, respectively

¹ Y mean: mean value across years

² T x Y: mean value across tillage operations and years

³ N mean: mean value across N fertilizations

⁴ T x N: mean value across tillage operations and N fertilizations

WU was primarily responsible for the lower YSNE. The value of YSNE reflects the kg cumulative N_2O emission per Mg of maize grain produced. The lower value of

YSNE with WU than with WOU indicates that the application of urea could reduce N_2O emission per unit of maize grain production. Zhao et al. [38] determined the

Table 10 Yield-scaled N₂O emission with different tillage operations (T), nitrogen fertilizations (N), and years (Y)

Year	Year 1			Year 2			Y mean ¹		
	Tillage operations	CT	NT	T mean	CT	NT	T mean	CT	NT
Nitrogen fertilizations				Yield-scaled N₂O (kg Mg⁻¹ year⁻¹)					
WOU	1.29 ^a (0.29)	1.00 ^b (0.16)	1.14 ^A (0.16)	1.80 ^a (0.21)	2.34 ^a (0.13)	2.07 ^A (0.15)	1.55 ^a (0.19)	1.67 ^a (0.27)	1.61 ^A (0.16)
WU	1.56 ^a (0.32)	1.16 ^a (0.09)	1.36 ^A (0.17)	1.04 ^a (0.05)	1.02 ^a (0.23)	1.03 ^B (0.11)	1.30 ^a (0.18)	1.09 ^a (0.12)	1.19 ^B (0.11)
N mean ³	1.42 ^a (0.21)	1.08 ^a (0.09)		1.42 ^a (0.13)	1.68 ^a (0.28)		1.42 ^a (0.13)	1.38 ^a (0.16)	
T × N mean ⁴	1.25 ^a (0.12)			1.55 ^a (0.16)					

Standard error in brackets. Different lower- and upper-case letters denote significant differences at $p < 0.05$ in column and row comparisons, respectively

¹ Y mean: mean value across years

² T × Y: mean value across tillage operations and years

³ N mean: mean value across N fertilizations

⁴ T × N: mean value across tillage operations and N fertilizations

change of YSNE in a maize field at N fertilization rates of 0–250 kg N ha⁻¹. They observed that the YSNE with N fertilization was lower than that with control up to 171 kg N ha⁻¹ and then increased at higher application rates.

Year significantly affected the grain yield of maize (Table 3). The mean value of grain yield of maize across tillage operations and N fertilizations was significantly higher in year 1 than that in year 2 (Table 9). As mentioned above, a higher rate of urea was applied in year 1 than in year 2. The greater grain yield of maize in year 1 was attributed to the greater supply of inorganic N, as this is a plant nutrient that is essential for growth and reproduction.

There was a significant tillage operation × year interaction for the grain yield of maize (Table 3). The mean grain yields of maize across N fertilizations with CT and with NT in year 1 did not significantly differ between them (Table 9). However, the mean grain yield of maize across N fertilizations with NT was significantly lower than that with CT in Year 2. Even though this study was initiated in November 2017, NT practice had been maintained for 4 years from October 2015 to October 2019. The effect of no tillage in reducing grain yield of maize seems to change over time. Pittelkow et al. investigated the effect of NT on crop yield through a global meta-analysis [39]. They reported that maize yield under NT management did not exceed that under CT in any experiment duration.

There was significant N fertilization × year interactions for grain yield of maize and YSNE (Table 3). The mean values of the grain yield of maize across tillage operations with WOU and WU in year 1 did not differ significantly between them (Table 9). However, the mean value of grain yield of maize across tillage operations with WU was significantly higher in year 2 than that with WOU. This grain yield response to N fertilization in both years affected

YSNE. The mean values of YSNE across tillage operations with WOU and WU in year 1 were not significantly different (Table 10). However, the mean value of YSNE across tillage operation with WU was significantly lower in year 2 than that with WOU. The similar rates of increase of grain yield of maize and cumulative N₂O emission with WU were responsible for the lack of a difference between YSNEs with WOU and WU in year 1 (Tables 4, 9, 10).

In conclusion, the results from a field study for 2 years clearly demonstrated the effect of N fertilization and tillage operation on N₂O emission from upland soil and maize grain yield. The application of urea increased the cumulative N₂O emission from maize-cultivated upland soil. Conventional tillage increased cumulative N₂O emission compared with NT. Different tillage operations had different effects on the nitrification ratio and denitrification ratio. The current study showed that nitrification was the predominant process producing N₂O with CT, but denitrification was with NT. Tillage operation did not affect the grain yield of maize and YSNE. However, the application of urea increased the grain yield of maize and decreased YSNE, implying that it could reduce N₂O emission per unit of maize grain production. Although the application of urea decreased YSNE, it increased cumulative N₂O emission. From the perspectives of the global environment and food security, future sustainable agriculture should explore systems with low N₂O emissions at high crop productivity. Therefore, as N fertilization is inevitable in agriculture, a combination of N fertilization and NT could be more environmentally and economically beneficial soil management. In addition, further research on different combinations of N fertilization, including the fertilizer type and application rate, and tillage operations, including conservation tillage and partial tillage, to reduce N₂O emission and maintain or increase crop yield should be conducted.

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Authors' contributions

SUK, HHL, SMM, and HRH carried out soil sampling, soil analyses, and data organization. COH participated in interpreting the obtained results and organizing the manuscript. All authors read and approved the final manuscript.

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