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Short-Term Responses of Nitrous Oxide Emissions and Concentration Profiles to Fertilization and Irrigation in Greenhouse Vegetable Cultivation*[1]

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ABSTRACT

In vegetable cultivation, the majority of N_2O emissions occur after fertilization; it is therefore important to understand any factors contributing to this process. An experiment was conducted to investigate short-term N_2O dynamics following topdressing in a greenhouse vegetable field in South China. During two topdressing processes, three different urea-N treatments with irrigation were conducted in May and June in a tomato (*Lycopersicum esculentum*) cultivation. The N_2O fluxes, soil concentration profiles and soil environments at the 0–60 cm depths at 10 cm intervals were measured both immediately prior to and 5 days after topdressing. The N_2O fluxes before topdressing ranged from 6.7±2.1 to 55.0±28.8 μg N m^{-2} h^{-1}; even higher numbers were recorded in highly fertilized plots. The NO_3^--N accumulation in the soil caused by vegetable cultivation during the 5 years prior to the start of the experiment, resulted in high background N_2O fluxes. One day after topdressing (1 DAT) in May and June, N_2O fluxes increased, which coincided with sharp increases in soil N_2O concentrations at depths of 2.5 and 15 cm and in NO_3^--N and NH_4^+-N contents at depths of 0–20 cm. From 1 to 5 DAT, fluctuations in the N_2O fluxes did not harmonize with the N_2O concentrations at a depth of 2.5 cm, which was attributed to different gas diffusion rates at depths of 0–10 cm. These results suggested that surface soil N and environmental conditions were crucial for determining the short-term N_2O ebullitions during topdressing in greenhouse vegetable cultivation.

Key Words: nitrogen application, N_2O, soil profile, topdressing, vegetable field

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INTRODUCTION

Nitrous oxide (N_2O) is a major greenhouse gas and a cause of great environmental concern, having a global warming potential of 310. N_2O also provides the third greatest contribution to the annual global greenhouse gas emission budget following CO_2 (IPCC, 2007a). In addition, N_2O is predicted to be main ozone depleting substance in the 21st century (Ravishankara *et al.*, 2009). Anthropogenic activities are enhancing the global emissions of N_2O and the agricultural use of fertilizer N has been recognized as one of the main sources of this trace gas (IPCC, 2007b).

China is one of the largest agricultural countries in the world. The total area of harvested crops in China accounts for approximately 13% of the world total (FAO, 2010). The consumption of chemical fertilizer N has also increased rapidly since the early 1970s, from 2.9 Tg in 1970 to 24.8 Tg in 1998, accounting for approximately 30% of the world total consumption (Zhu and Chen, 2002). Such a significant increase in N fertilizer usage could be the cause of high N_2O emissions from agricultural fields. Annual fertilizer N-induced direct N_2O emissions have been estimated at 115.7 Gg N $year^{-1}$ in the 1980s and 210.5 Gg N $year^{-1}$ in the 1990s (Zou *et al.*, 2010). Currently, vegetable cultivation has become more popular and is one of the most important sources of income for local farmers in the North and South of China (Chen *et al.*, 2004; Shi *et al.*, 2009). He *et al.* (2009) documented that greenhouse vegetable cultivation systems have received excessive amounts of N fertilizer. Chen *et al.* (2004) and Huang *et al.* (2006) reported that vegetable cultivation under greenhouse or open fields have received around

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1 000 kg N ha^{-1} $year^{-1}$ of fertilizer N in China. N recovery of harvested vegetable, however, was only 20%–50% of the applied N (Huang *et al.*, 2006). It could be expected that a significant amount of N_2O was produced as a consequence of vegetable production in China. Recently, the quantification of N_2O emissions from vegetable fields has been gradually gathering attentions in North and South China (Xiong *et al.*, 2006; He *et al.*, 2009; Wang *et al.*, 2011; Deng *et al.*, 2012; Jia *et al.*, 2012; Min *et al.*, 2012).

The majority of these studies have focused on N_2O fluxes from the soil to the atmosphere. It is, however, crucial to investigate N_2O dynamics in the soil to mitigate N_2O emission from vegetable cultivation, because N_2O emissions from fertilized soils are the result of production, consumption and transport in the soil. Unfortunately, much less information has been gathered regarding N_2O in the soil and its location of production in the soil profile during vegetable cultivation. N_2O production and consumption are controlled by a number of individual factors, including soil water content, temperature, mineral N content, available organic carbon and pH (Mosier *et al.*, 1998), which are all themselves influenced by agricultural management. The N_2O concentration could therefore be considerably different throughout the soil depth because these factors can vary within the soil profile (Van Groenigen *et al.*, 2005). Irrigation, topdressing and soil temperature have been reported as key factors in controlling N_2O emissions in greenhouse tomato fields (He *et al.*, 2009). Pulse-like N_2O emissions have been observed following topdressing (Xiong *et al.*, 2006; Min *et al.*, 2012), and N_2O flushes have accounted for more than 50% of the total emissions (He *et al.*, 2009; Min *et al.*, 2012). It is therefore important to understand the short-term N_2O dynamics in the soil after topdressing.

The purpose of this study was to elucidate N_2O dynamics in vegetable fields, by focusing on the short-term N_2O dynamics in greenhouse cultivation after topdressing because high N_2O emissions have often been reported following topdressing in previous studies. We attempted to measure N_2O emissions and concentration profiles under different fertilizer management systems, which had been continuously implemented for 5 consecutive years prior to the initiation of any sampling, and determine the mechanisms and factors affecting N_2O production and emission.

MATERIALS AND METHODS

Experimental site

The field experiment was carried out at a typical vegetable production farm (31° 14′ N and 119° 53′ E) in Yixing, Jiangsu Province, which is located at the center of the Taihu Lake region belonging to the Yangtze River Delta in China. Two typical commercial greenhouses were randomly selected for the field experiment. The two greenhouses were adjacent to each other, and have the same cultivation history. The soil at the study site was classified as an Anthrosol. The upper soil horizon (0–20 cm) contained 8.3% sand, 76.4% silt and 15.3% clay with a bulk density of 1.6 g cm^{-3} and was classified as a sandy clay loam (Huan *et al.*, 2007). The soil had a pH of 5.58 and electrical conductivity (EC) of 0.28 mS cm^{-1} with an extraction ratio of 1:5 (soil:water). At the beginning of the experiment, soil organic matter content, total N, NO_3^--N, Olsen-P and ammonium acetate extractable-K values were 24.9, 1.04, 0.04, 0.06 and 0.06 g kg^{-1}, respectively. Experimental treatments were randomly distributed throughout the two greenhouses. The average underground water table was 0.8 m during the summer at the experimental area (Min *et al.*, 2011). Each greenhouse was covered with polyethylene film (ground area 36 m × 6.0 m) and was without supplementary lighting or heating. From 2005 to 2009, three consecutive crops, including tomato (*Lycopersicum esculentum*), cucumber (*Cucumis sativus*) and celery (*Apium graveolens* cv. Dulce), were cultivated every year under three N managements with three replications (plot size of 7.0 m × 2.5 m), which were denoted as treatments C (receiving 234 kg N ha^{-1} $year^{-1}$ of organic manure), F1 (receiving 234 kg N ha^{-1} $year^{-1}$ of organic manure plus 522 kg N ha^{-1} $year^{-1}$ of urea, representing 3/5 of the conventional urea application rate) and F2 (receiving 234 kg N ha^{-1} $year^{-1}$ of organic manure plus 870 kg N ha^{-1} $year^{-1}$ of urea, representing a conventional urea application rate). Thus, C, F1 and F2 treatments were fertilized with 1 170, 3 780 and 5 520 kg N ha^{-1}, respectively, through the application of organic manure and urea prior to the beginning of cultivation in 2010.

Experimental design

The sampling was conducted following the 1st and 2nd topdressing periods during the cultivation of a tomato crop from April to July in 2010. The N fertilizer was applied at 0, 45 and 75 kg N ha^{-1} as topdressed urea in the treatments C, F1 and F2, respectively, in 2010. Each treatment was conducted with three replicates.

The tomato cultivation practice followed in 2010 was as follows: as basal fertilizer, urea (0, 90 and 150 kg N ha^{-1} for C, F1 and F2 treatments, respectively), organic manure (78 kg N ha^{-1} for all treatments), 150 kg ha^{-1} of K_2O and 150 kg ha^{-1} of P_2O_5 were evenly broadcasted onto the soil surface on April 8, 2010. Tomato plants with four leaves were transplanted into the seedbed and 7.2 mm of river water was applied as irrigation on April 11, 2010. The distances between the rows and hills were 50 and 25 cm, respectively. The first topdressing plus irrigation (6.5 mm), only irrigation (6.1 mm) and 2nd topdressing plus irrigation (7.2 mm) were applied, respectively, on May 7, June 5 and June 18, 2010. The amounts of topdressed urea-N were the same as those described above. The fruits were harvested between June 24 and July 20, 2010.

Sampling and field measurements

From the day of topdressing (0 day after topdressing, 0 DAT) on May 7 and June 18, continuous sampling was conducted for 6 days (Fig. 1). Each sampling was conducted in the morning.

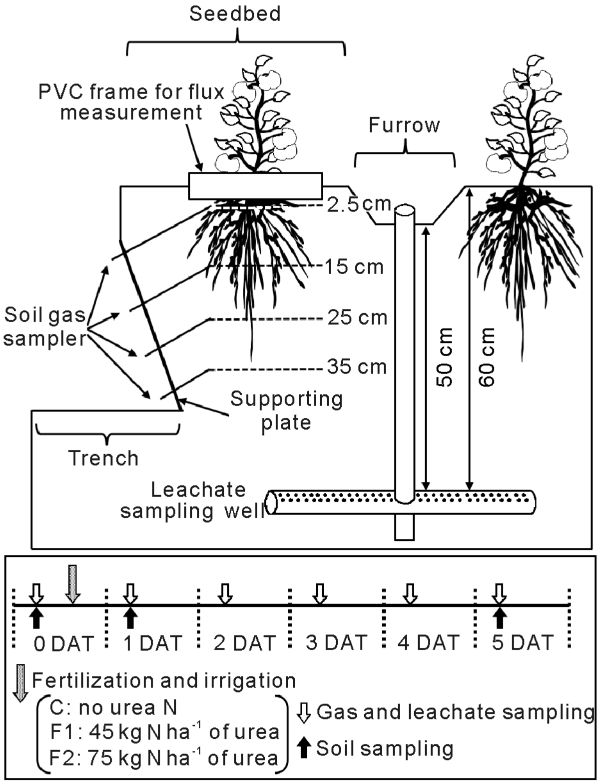

Fig. 1 Cross-sectional diagram for the installation of sampling equipment, and timing of fertilization, irrigation and sampling. DAT means day after topdressing.

The sampling equipment installed in each plot is also shown in Fig. 1. Gas samples for N_2O flux determination were collected using closed chambers (Terry *et al.*, 1981) made of plexiglas (60 cm × 70 cm × 60 cm or 60 cm × 70 cm × 120 cm in size, depending on the tomato height). The chambers were placed on a fixed polyvinyl chloride frame to enclose the soil and a tomato crop. Gas samples were collected in the morning (from 09:00 to 11:00) with a double-ended needle connected to an evacuated vial stopped with butyl rubber septum. For each N_2O flux measurement, four gas samples were withdrawn from each chamber at 15 min intervals. When the gas samples were collected, the air temperature and the temperature inside the chamber were also measured simultaneously.

The soil gases at depths of 2.5, 15, 25 and 35 cm below the seedbed were sampled through a 3-way stopcock attached to stainless steel soil gas samplers (outside diameter of 1.81 mm, inner diameter of 1.45 mm). The soil gas samplers were inserted into a plexiglas supporting plate in a trench dug into the side of each plot (Fig. 1). Five milliliters of the soil gas samples were transferred immediately into the 3 mL evacuated glass vials (SVG-3, Nichiden-Rika Glass Co., LTD., Hyougo, Japan) to ensure over pressure of sample in the vials. During every soil gas sampling procedure, soil temperature profiles at 2.5, 15, 25, 35 and 45 cm below the seedbed in each plot were measured using a thermometer (RTR-52, T&D Co., Nagano, Japan) modified for profile measurement.

Duplicate leachate samples from 60 cm below the seedbed were obtained from the two leachate sampling wells installed in a furrow in each plot (Fig. 1) for determination of mineral N (NH_4^+-N and NO_3^--N) in the leachate. The sampling wells were passive samplers, which had an outlet and two perforated tubes attached to one end of the sampler (Fig. 1). The sampler was installed so that the perforated tubes could collect leachate infiltrated from seedbed and furrow at a depth of 60 cm (Fig. 1). Prior to topdressing (0 DAT), the leachates in the wells were removed to avoid any mixing with the leachate resulting from the irrigation. Leachate sampling was initially conducted on 1 DAT by withdrawing leachate using a plastic syringe through the outlet, which was capped in between sampling to avoid contamination. During this study, no more leachate was observed in the sampler on 2–5 DAT in either May or June.

A soil core of 60 cm depth was sampled from the seedbed on 0, 1 and 5 DAT. The soil core was divided into 10 cm segments (*i.e.*, 0–10, 10–20, 20–30, 30–40, 40–50 and 50–60 cm) for each soil sample taken.

Sample analysis

The samples for N_2O flux measurement were taken to the laboratory and analyzed using a gas chromatograph (GC) (HP 5890 II, Hewlett-Packard, Palo Alto, CA, USA) with an electron capture detector (ECD) and N_2 as the carrier gas. The details for the GC-ECD method have been described by Xing *et al.* (2002). The N_2O concentrations in soil were also analyzed by GC (GC-14A, Shimadzu, Kyoto, Japan) with an ECD and N_2 mixed with CH_4 as the carrier gas.

The NH_4^+-N and NO_3^--N concentrations in the leachates were analyzed by ion chromatography system (ICS-90 and ICS-1000, Dionex, Sunnyvale, USA) after filtration through a 0.45 μm membrane filter.

To determine NH_4^+-N and NO_3^--N contents in each of the 10 cm soil segments sampled from the 60 cm soil cores, fresh soil was mixed thoroughly and sieved (5 mm mesh). Subsamples of 10 g were weighed and extracted by shaking with a 100 mL 1 mol L^{-1} KCl solution for 1 h. The filtrates were stored at −20 °C and analyzed with a continuous flow analyzer (Skalar Co., Breda, The Netherlands).

Determination of soil physical characteristics

Soil bulk density for each depth was determined from the segments of the 60 cm soil core collected at 0 DAT in May. The volumetric water content from each depth was determined gravimetrically after drying the subsample at 105 °C for 24 h. The water filled pore space (WFPS) was calculated using the bulk density (ρ, g cm^{-3}), volumetric water content (θ, %) and mean soil particle density (ρ_m, g cm^{-3}) (He *et al.*, 2009), as follows:

$$\text{WFPS} = \theta/[1 - (\rho/\rho_{\text{m}})] \times 100 \tag{1}$$

The diffusion coefficient of N_2O (D_{s}) was calculated from the soil tortuosity ($a^2/\varphi^{2/3}$) and diffusion coefficient in free air (D_{a}) (Jin and Jury, 1996; Yoh *et al.*, 1997), as follows:

$$D_{\text{s}} = (a^2/\varphi^{2/3})[D_{\text{a}} \times (T/273)^{1.7}] \tag{2}$$

where a (cm^3 cm^{-3}) is the volumetric air content, φ (cm^3 cm^{-3}) is the soil porosity, D_{a} is a constant of 0.143 cm^2 s^{-1} at standard temperature and pressure, and T (K) is the soil temperature. Volumetric air content and soil porosity were calculated from bulk density, particle density and volumetric water content (Jury and Horton, 2004).

Data analyses

Cumulative N_2O emissions from 0 to 5 DAT were estimated by trapezoidal integration of the mean N_2O flux over time.

All statistical analyses were conducted using SPSS version 16.0 (SPSS Inc., USA). Differences of NH_4^+-N and NO_3^--N contents and N_2O concentration in the soil between sampling dates or N treatments were analyzed by using independent samples *t*-test. The *t*-test was also applied for comparison of cumulative N_2O emissions between different topdressing periods. Correlations of N_2O concentration in the soil were performed with soil temperature, WFPS, and NH_4^+-N and NO_3^--N contents. Data from 0, 1 and 5 DAT was used for the analyses because mineral N data was determined on 0, 1 and 5 DAT only. To evaluate the effects of N fertilization management and the tomato growing season on NO_3^--N and NH_4^+-N concentrations in the leachate, a two-way analysis of variance (ANOVA) was performed.

RESULTS

Soil environment

As shown in Fig. 2, the soil temperature profiles revealed similar values and trends in all of the treatments for each topdressing period. Soil temperatures exhibited greater variability with sampling date at depths of 2.5 and 15 cm. Soil temperatures at depths of 2.5 and 15 cm in all treatments fluctuated from 16.7±0.3 to 28.3±2.1 °C in May and from 23.9±0.6 to 32.1±1.1 °C in June. At depths of 25–45 cm, the soil temperatures were relatively stable.

The WFPS was relatively higher in May than in June, especially at depths of 0–10 and 10–20 cm (Fig. 3). Prior to topdressing (0 DAT), the mean WFPS values in May at depths of 0–10 and 10–20 cm were 35.2%±2.9% and 40.4%±0.8%, respectively. In June, the corresponding values were 26.0%±0.8% and 33.1%±1.6%. After topdressing, these respective values increased to 42.1%±3.1% and 42.1%±0.9% in May and 31.9%±3.1% and 33.7%±2.7% in June. In the deeper layers (20–60 cm), WFPS values were 40%–50% in the majority of cases.

The NH_4^+-N content at the depth of 0–10 cm in the fertilized plots (treatments F1 and F2) was 8.7±5.3 and 33.1±18.3 mg N kg^{-1}, respectively (Fig. 4). Although these values were higher than that found in treatment C (6.3±2.1 mg N kg^{-1}), no significant differences were found between treatment C and the fertilized plots (treatments F1 and F2). With the exception of treatment F2 in June, the application of urea led to significant increases ($P < 0.05$) in the NH_4^+-N content at the depth of 0–10 cm in the fertilized plots at 1 DAT. The NH_4^+-N content at the depth of 0–10 cm in May increased from 8.7±5.3 to 105.4±28.2 mg N kg^{-1} and 33.1±18.3 to 104.0±17.0 mg N kg^{-1} in the F1 and F2 treatments, respectively. In June, they increased from 18.0±9.9 to 107.0±26.9 mg N kg^{-1} and 84.2±49.5 to 142.6±75.8 mg N kg^{-1} in the F1 and F2 treatments, respectively. In treatment C, slight de creases in the NH_4^+-N content were found at the depth of 0–10 cm

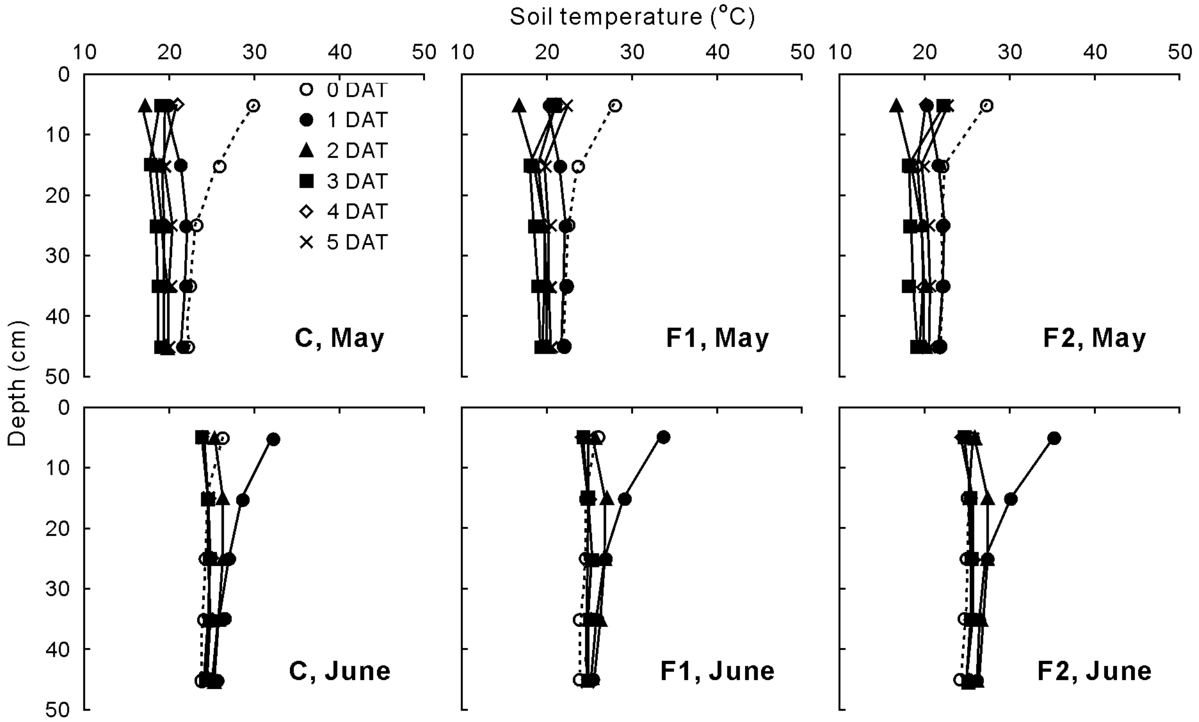

Fig. 2 Vertical profiles (2.5, 15, 25, 35 and 45 cm depths) of soil temperature in the treatments receiving 0, 45 and 75 kg N ha^{-1} of topdressed urea plus irrigation (treatments C, F1 and F2, respectively) in May and June, 2010. DAT means day after topdressing.

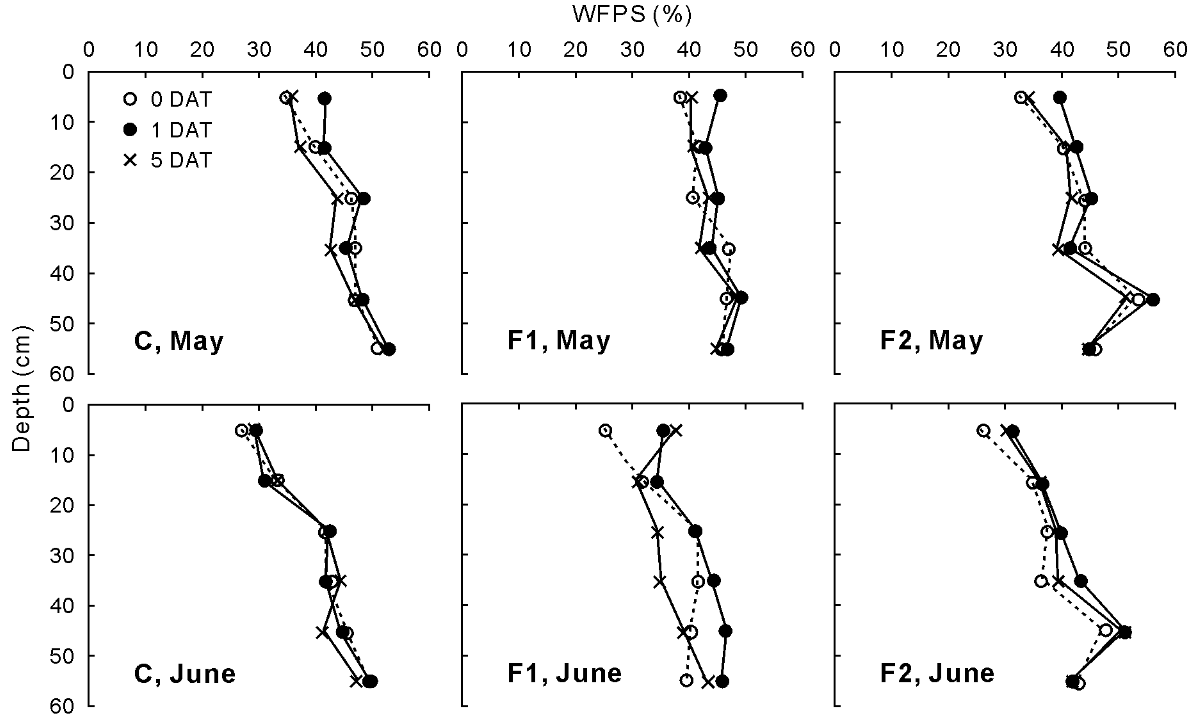

Fig. 3 Vertical profiles of water filled pore space (WFPS) in the treatments receiving 0, 45 and 75 kg N ha^{-1} of topdressed urea plus irrigation (treatments C, F1 and F2, respectively) in May and June, 2010. The data for each soil depth mean WFPS of 0–10, 10–20, 20–30, 30–40, 40–50 and 50–60 cm soil core segments, respectively. DAT means day after topdressing.

following irrigation (1 DAT) compared with those in 0 DAT in both topdressing periods. The NH_4^+-N content in the deeper soil layer (10–60 cm depth) showed smaller changes compared to those observed at the depth of 0–10 cm.

Fig. 5 shows the vertical NO_3^--N content profiles of the soil. Prior to fertilization, the NO_3^--N contents in the C treatment at the depth of 0–10 cm in May and

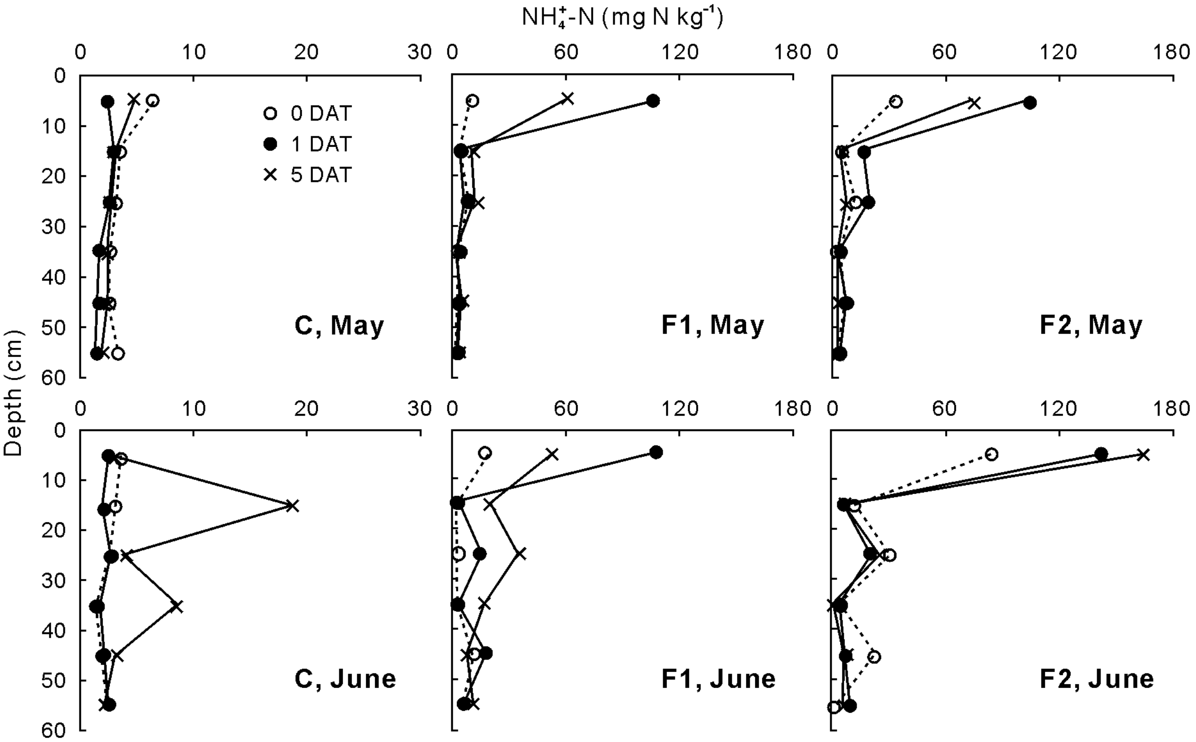

Fig. 4 Vertical profiles of NH_4^+-N contents in the treatments receiving 0, 45 and 75 kg N ha^{-1} of topdressed urea plus irrigation (treatments C, F1 and F2, respectively) in May and June, 2010. The data for each soil depth mean NH_4^+-N contents of 0–10, 10–20, 20–30, 30–40, 40–50 and 50–60 cm soil core segments, respectively. DAT means day after topdressing.

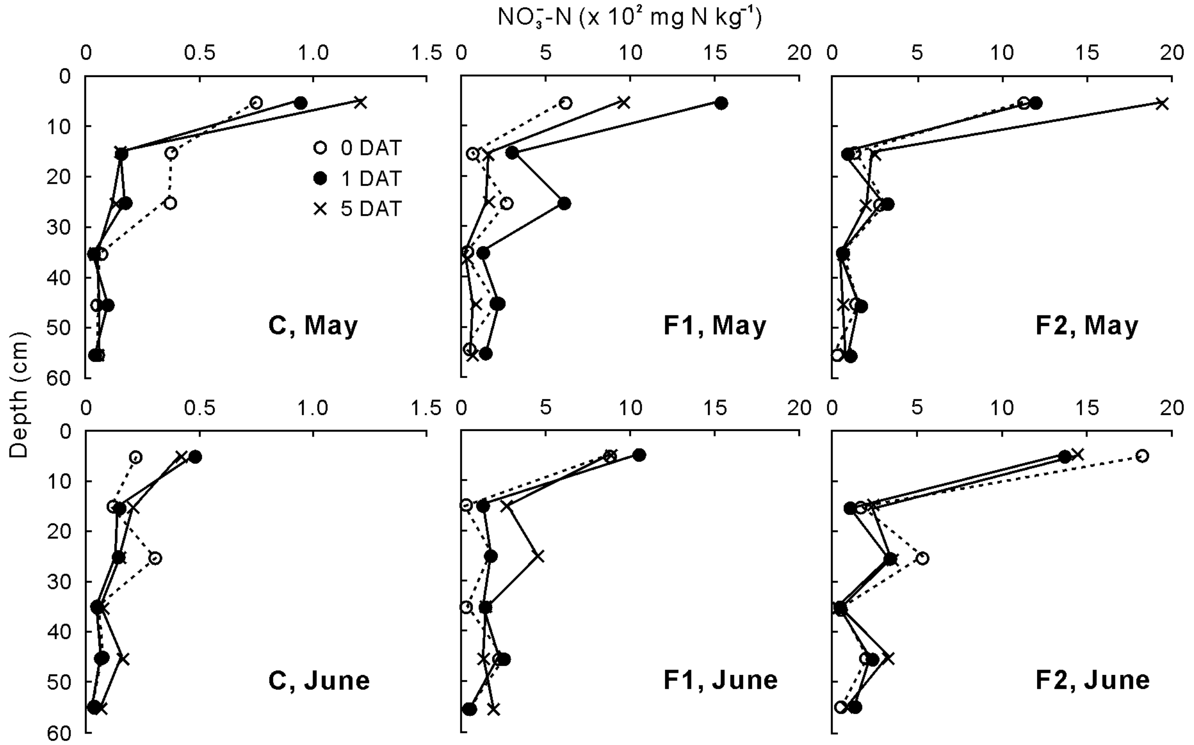

Fig. 5 Vertical profiles of NO_3^--N contents in the treatments receiving 0, 45 and 75 kg N h^{-1} of topdressed urea plus irrigation (treatments C, F1 and F2, respectively) in May and June, 2010. The data for each soil depth mean NO_3^--N contents of 0–10, 10–20, 20–30, 30–40, 40–50 and 50–60 cm soil core segments, respectively. DAT means day after topdressing.

June were 74.6±32.9 and 21.2±2.3 mg N kg^{-1}, respectively. In the fertilized plots, the corresponding values were 614±175 and 884±366 mg N kg^{-1} in the F1 treatment and 1 119±263 and 1 828±450 mg N kg^{-1} in the F2 treatment. These values were significantly higher ($P < 0.05$) in both months than those in the C treatment. The NO_3^--N contents were lower at the depth of 10–60 cm than those at 0–10 cm. Treatments F1 and F2, however, had higher NO_3^--N contents at the depth of 0–10 cm compared with the C treatment. After topdressing, the NO_3^--N contents at the depth of 0–10 cm in the majority of treatments increased slightly, although the increase was not significant.

N_2O flux from soil to the atmosphere

Prior to topdressing, the N_2O flux determined using the chamber method ranged from 6.7±2.1 to 55.0±28.8 μg N m^{-2} h^{-1} in May and 8.1±3.8 to 35.6±11.0 μg N m^{-2} h^{-1} in June (Fig. 6). Although fertilizer was not applied to the F1 and F2 treatments on 0 DAT, these treatments had higher N_2O fluxes than those from the C treatment. On 1 DAT, the N_2O fluxes of C, F1 and F2 had increased to 26.1±11.1, 63.1±32.1 and 96.6±35.0 μg N m^{-2} h^{-1} in May and 44.4±9.2, 105.7±87.1 and 233.8±127.7 μg N m^{-2} h^{-1} in June, respectively. Following the initial increases in N_2O flux, fluxes then fluctuated with different patterns between May and June (Fig. 6).

The cumulative N_2O emissions in treatments C, F1 and F2 from 0 to 5 DAT were 2.1±1.1, 6.7±1.4 and 9.4±0.8 mg N m^{-2} in May, and 1.8±0.3, 5.3±2.7 and 14.7±6.0 mg N m^{-2} in June, respectively. Within the same N treatments, no significant differences in the cumulative emission values were found between May and June.

Soil N_2O concentration

Fig. 7 shows N_2O concentrations in the soil for each treatment in May and June. As mineral N contents or N_2O flux, mean N_2O concentrations across the entire depth of treatments F1 and F2 were significantly higher ($P < 0.05$) than those found in treatment C in both topdressing periods on 0 DAT. The N_2O concentrations increased after topdressing (1 DAT), especially in the fertilized plots. The values then decreased over time to reach near background levels by 5 DAT. The N_2O concentrations were 0.9–1.8 times higher on 1 DAT than those recorded on 0 DAT in the C treatment. In the same manner, they were 1.0–17 and 0.5–9.6 times higher in the F1 and F2 treatments, respectively. The maximum N_2O concentrations were always observed in upper soil layers during the experimental period (depths of 2.5 and 15 cm).

Concentrations of mineral N in leachate

In this study, leaching of irrigation water was obse-

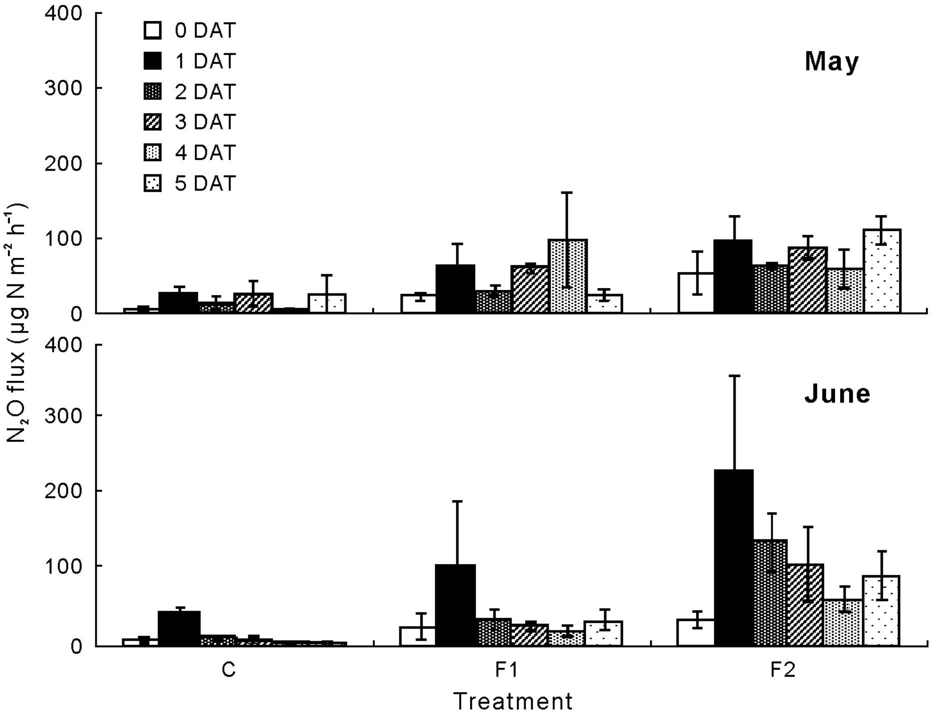

Fig. 6 Mean N_2O fluxes determined by chamber method in the treatments receiving 0, 45 and 75 kg N ha^{-1} of topdressed urea plus irrigation (treatments C, F1 and F2, respectively) in May and June, 2010. Vertical bars indicate standard deviations of the means ($n = 3$). DAT means day after topdressing.

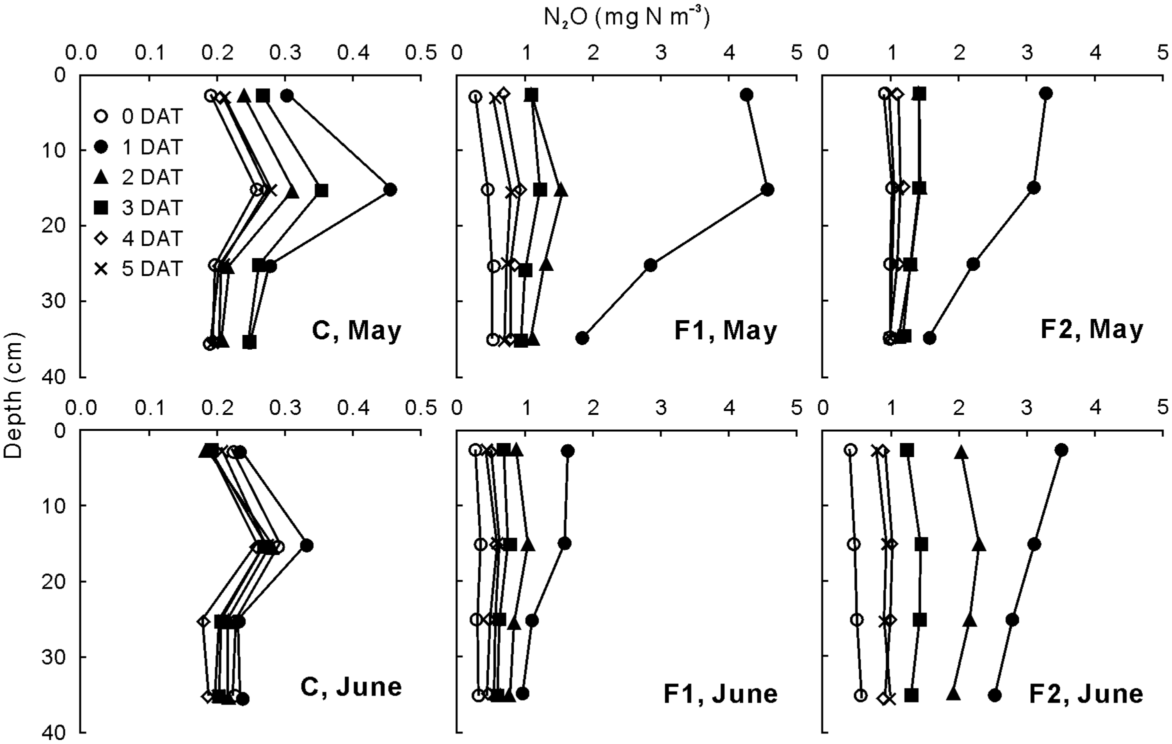

Fig. 7 N_2O concentrations in bulk soil of the treatments receiving 0, 45 and 75 kg N ha^{-1} of topdressed urea plus irrigation (treatments C, F1 and F2, respectively) in May and June, 2010. DAT means day after topdressing.

rved only on 1 DAT (data not shown). In some of the leachate sampling wells, no accumulation of the leachate was observed. Sample sizes in C, F1 and F2 were 3, 2 and 4 in May and 4, 5 and 5 in June, respectively. Table I shows the mean concentrations of NO_3^--N and NH_4^+-N in the leachates calculated from the leachate data collected. The NO_3^--N concentrations in the leachates were higher with higher N fertilization rates. Two-way ANOVA analysis showed that there was a significant fertilization effect on NO_3^--N concentration (Table I). The NH_4^+-N concentrations in the leachates were less than 1.0 mg N L^{-1} in both months.

TABLE I

Concentrations of NO_3^--N and NH_4^+-N in the leachates of the treatments receiving 0, 45 and 75 kg N ha^{-1} of topdressed urea plus irrigation (treatments C, F1 and F2, respectively) 1 day after topdressing in May and June, 2010

Treatment	NO_3^--N		NH_4^+-N	
	May	June	May	June
	mg N L^{-1}			
C	8.0±1.9[a)]	6.6±1.3	0.44±0.48	0.78±0.57
F1	13.0±4.1	8.6±3.3	0.22±0.32	ND[b)]
F2	22.7±12.3	20.5±14.3	0.25±0.51	0.82±0.57
Fertilizer	*		NS	
Season	NS[c)]		NS	
Fertilizer × season	NS		NS	

*Significant at $P < 0.05$.

[a)] Means±standard deviations. Sample sizes (n) in treatments C, F1 and F2 were 3, 2 and 4 in May and 4, 5 and 5 in June, respectively.

[b)] Not detectable.

[c)] Not significant.

DISCUSSION

In the greenhouse vegetable cultivation, N managements significantly affect the soil environment factors, such as soil pH, salinity and N content (Shi *et al.*, 2009). For the current research, the studied soils were subjected to different N management programs continuously for 5 years. Prior to the first topdressing (in May), significantly higher NO_3^--N contents were observed at a depth of 0–10 cm in the treatments F1 and F2 than those in the treatment C, which indicated an accumulation of applied N from consecutive cultivation or basal fertilizer. If basal fertilizer including organic N was accumulated at the depth of 0–10 cm, this would lead to an increase in N content at the same depth; however, they were only 83, 142 and 258 mg N kg^{-1} for treatments C, F1 and F2, respectively. This indicated that most of the NO_3^--N content at the depth of 0–10 cm at 0 DAT in May was derived from mineralized manure N and urea applied during earlier fertilization processes, as identified by Shi *et al.* (2009). Nitrogen accumulation in the surface soil in tomato cultivation has also been reported in previous studies (Jackson and Bloom, 1990; Min *et al.*, 2011) and is likely a result of furrow irrigation. In the majority of cases, the irrigation water was placed into a furrow between the seedbeds (Fig. 1). As a result, the solute in the soil below the furrow was leached, whereas the solute in the seedbed tended to accumulate (Jackson and Bloom, 1990). Thus, consecutive greenhouse cultivation considerably affected the soil NO_3^--N content.

The higher N_2O fluxes observed prior to topdressing (0 DAT) in the treatments F1 and F2, compared with treatment C, were clearly the results of N accumulated in the soil (Fig. 6). Relatively higher N_2O concentrations were observed at lower depths in the fertilized plots, which suggested that N_2O diffusion was occurring into the upper layer from the deeper soil. In contrast, the highest N_2O concentrations observed in this study were found at a depth of 15 cm in the treatment C (Fig. 7). The occurrence of a significant positive correlation between N_2O concentration at 35 cm and NO_3^--N concentration at 30–40 cm on 0 DAT (Table II) suggested that the differences of high N_2O concentration between treatment C and the fertilized treatments (F1 and F2) were caused by higher denitrification of NO_3^--N in the treatment plots at a depth of 30–45 cm, in which higher WFPS values were observed than those in the surface soil (Fig. 3 and 5). Thus, greenhouse fields which received a high amount of N had the potential to emit high background levels of N_2O because of the storage of significant amounts of NO_3^--N in the soil during cultivation and fallow periods. The upward diffusion of N_2O dissolved in the groundwater may have also been responsible for the N_2O emitted to the atmosphere in the treatments F1 and F2, as demonstrated by Minamikawa *et al.* (2011). In the studied area, the groundwater table during summer was 0.8 m, which was similar to the study conducted by Minamikawa *et al.* (2011). It has been reported that high NO_3^--N concentration in the groundwater under an arable field was related to high concentration of N_2O (Von der Heide *et al.*, 2008).

The observed rapid increases of N_2O flux following topdressing (1 DAT) are in agreement with the data reported in previous publications (Xiong *et al.*, 2006; He *et al.*, 2009; Min *et al.*, 2012). High concentrations of N_2O near the surface (depths of 2.5 and 15 cm) after topdressing in the fertilized treatments (1 DAT) suggested a shift in the hot spots of N_2O production from the deeper layers. Rapid changes in the N_2O concentration profile following fertilization and irrigation (or rainfall) have also been reported in several earlier studies (Yoh *et al.*, 1997; Nobre *et al.*, 2001). Yoh *et al.* (1997) suggested that nitrification was the major process for rapidly increasing of N_2O concentration in surface layer of the soil fertilized with $(NH_4)_2SO_4$, followed by a flush of N_2O into the atmosphere. In contrast, both the nitrification and denitrification processes might be responsible for the rapid increase of N_2O concentration in our case at depths of 2.5 and 15 cm, because of the presence of high concentrations of NO_3^--N and NH_4^+-N from urea at depths of 0–10 cm on 1 DAT (Figs. 4, 5 and 7). On 0 DAT, the fields were irrigated after broadcasting urea. The NO_3^--N accumulated in the surface soil would be converted to N_2O during this period because of the enhancement of deni-

TABLE II

Correlation coefficients between N_2O concentration and soil environmental factors within each depth and sampling time

Sampling time	Depth[a)]	Correlation coefficient			
		NH_4^+-N	NO_3^--N	Soil temperature	Water filled pore space
	cm				
0 day after topdressing	0–10	0.385	0.518	−0.142	0.030
	10–20	0.215	0.589	−0.859*	0.415
	20–30	0.403	0.590	−0.540	−0.005
	30–40	0.502	0.904*	−0.334	−0.098
1 day after topdressing	0–10	0.863*	0.952**	−0.166	0.377
	10–20	0.352	0.827*	−0.263	0.580
	20–30	0.622	0.917*	−0.122	−0.262
	30–40	0.584	0.364	0.094	−0.161
5 days after topdressing	0–10	0.791	0.992**	0.393	−0.069
	10–20	−0.386	0.844*	−0.091	0.508
	20–30	0.298	0.531	−0.120	−0.163
	30–40	−0.315	0.287	0.005	−0.476

*, **Significant at $P < 0.05$ and $P < 0.01$, respectively.

[a)] N_2O concentrations at 2.5, 15, 25 and 35 cm depths were used for the correlation analysis with soil environmental factors at 0–10, 10–20, 20–30 and 30–40 cm depths, respectively.

trification by irrigation (Sánchez-Martín *et al.*, 2008). Following on from that, the NH_4^+-N accumulated at a depth of 0–10 cm on 1 DAT would be nitrified on the wet soil of the seedbed after drying. The WFPS on 1 DAT at the depth of 0–20 cm was in agreement with the range of 35%–60%, in which nitrification existed as the dominant process for N_2O production (Bateman and Baggs, 2005). Denitrification was also possible in the anaerobic microsite under aerobic conditions (Smith *et al.*, 2003) with high NO_3^--N content in the surface soil (Fig. 5). These mechanisms for N_2O production are the reason why N_2O concentrations at the depth of 0–10 cm were positively correlated with both NH_4^+-N and NO_3^--N contents at the same depth (Table II). It can therefore be speculated that both denitrification and nitrification are responsible for N_2O production in the surface soil layer (0–10 cm) following topdressing in greenhouse cultivation. Shen *et al.* (2010), however, reported that greenhouse cultivation decreased nitrification activity through soil acidification. Zhu *et al.* (2011) also reported that the dominant N_2O production process of the greenhouse soil was denitrification under 50% of its water holding capacity. Further study is needed to clarify possible mechanisms for the high N_2O production observed during the short period of time after topdressing. From 2 DAT, N_2O concentrations across the entire range of depths decreased gradually (Fig. 7), implying that net N_2O production was occurring from only 0 to 1 DAT and not over the entire extended sampling period (5 DAT) in each topdressing period. These results suggested that the N_2O emitted to the atmosphere was mainly produced during the first day immediately after topdressing.

One might expect that N_2O production by denitrification of the NO_3^--N in the upper soil layer would be prevalent in the fertilized plots (F1 and F2) after topdressing. The N_2O concentrations at depths of both 25 and 35 cm, however, were lower than those in the upper soil layer from 1 to 5 DAT (Fig. 7). A higher NO_3^--N concentration was detected in the leachate sample in the plots receiving higher amounts of N (Table I), indicating the occurrence of NO_3^--N transport from the surface soil until sampling on 1 DAT. The observed low correlation coefficient between the N_2O concentration and NO_3^--N content at the depth of 30–40 cm on 1 DAT (Table II) implied low N_2O production by low NO_3^--N content. It was therefore suggested that the surface soil was a hot spot of N_2O production during the short period after topdressing. If denitrification of NO_3^--N in the surface soil layer was enhanced by irrigation, however, as mentioned above, leaching of N_2O produced in the surface soil layer would be expected. In addition, a higher NO_3^--N concentration in the leachate sample in the plots with higher N application rate (Table I) would lead to further production of N_2O by denitrification of NO_3^--N in the groundwater. It has been reported that N_2O emissions through surface runoff or leaching (indirect N_2O emission) (IPCC, 2006) provide great uncertainty to the global N_2O budget. Therefore, systematic study including air, soil and groundwater would be needed in the future to quantify indirect N_2O emissions in greenhouse vegetable cultivation.

Even though each treatment received the same amount of N in May and June (0, 45 and 75 kg N ha^{-1} for C, F1 and F2, respectively), the magnitude and patterns of the associated N_2O fluxes were different in the two months (Fig. 6). It was envisaged that the changes in N_2O fluxes would be closely related to N_2O concentrations at the depth of 2.5 cm because of the short distance from the soil to the surrounding atmosphere. However, the changes in N_2O concentrations at the depth of 2.5 cm did not coincide with the temporal change in N_2O fluxes (Figs. 6 and 7), suggesting that the mechanism of N_2O diffusion into the atmosphere was different between May and June. It is likely that the diffusive flux to the atmosphere was affected by the conditions near the surface soil because gas diffusion is affected by water content and temperature (Heincke and Kaupenjohann, 1999). Furthermore, diffusive transport also relates to O_2 availability in addition to the potential for N_2O to escape to the atmosphere (Petersen *et al.*, 2008). In this study, the mean diffusion coefficients calculated from the values of all treatments at the depth of 0–10 cm were 0.027 ± 0.002 and 0.042 ± 0.004 cm^2 s^{-1} in May and June, respectively (data not shown) and they were significantly different ($P < 0.05$). It could therefore be suggested that faster nitrification and release into the atmosphere resulted in a pulse-like N_2O emission in June, whereas slower nitrification and loss into the atmosphere in May resulted in a relatively constant N_2O flux in the wetter surface soil after topdressing (Figs. 3 and 6).

CONCLUSIONS

Consecutive vegetable cultivation over a 5-year period in greenhouse conditions affected the short-term N_2O dynamics before and after topdressing. Accumulation of NO_3^--N in the soil by consecutive vegetable cultivation resulted in higher levels of N_2O production and emission in the soil relative to the plots receiving lower amounts of fertilizer N, suggesting the presence of higher background N_2O emissions. Short-term responses in the N_2O concentration profiles after topdressing revealed that the source of N_2O emitted to the atmosphere was the surface layer (0–20 cm) of the soil. Denitrification of NO_3^--N and nitrification of fertilized NH_4^+-N accumulated at the depth of 0–20 cm were responsible for the N_2O production observed a day after topdressing. No increases in the N_2O concentrations across the soil depth (0–35 cm depth) from 2 DAT to the end of the sampling schedule (5 DAT) suggested that N_2O emitted to the atmosphere was mainly produced during the first day immediately after topdressing. Furthermore, water content and temperature in surface soil were related to N_2O production and its emission patterns (pulse or constant). It can therefore be concluded that N in the surface soil layer is crucial for determining short-term N_2O ebullitions after topdressing in greenhouse vegetable cultivation. More intensive and elaborate sampling is needed to clarify the mechanisms of N_2O production and emission to the atmosphere.

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