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Nitrous oxide fluxes from soil under different crops and fertilizer management

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ABSTRACT

The effect of mineral fertilizer (F) and mineral combined with organic fertilizer (MF) on N_2O flux in grassland and cornfield was investigated for one year in Southern Hokkaido, Japan. Annual N_2O flux was higher in grassland than in cornfield, and it was higher in MF plot (14.9 kg N/ha/period) than in F plot (11.1 kg N/ha/period) in grassland. However, in cornfield, the annual N_2O flux was equal between both plots (5.6 kg N/ha/period). These results clarified that high nitrogen application was not always responsible for the high soil N_2O flux. N_2O flux was significantly correlated with air, soil temperature and water-filled pore space. More than 80% of the annual N_2O flux occurred before freezing and less than 4% during melting period. Denitrification was the main process of N_2O flux during study, it was evidenced by the distribution of N_2O and NO ratio which is from 1 to 1000. The denitrification activity (DEA) potentially increased in grassland soil in the beginning and the end of winter season when NO_3-N was abundant; on the other hand the abundance of carbon potentially increased DEA in cornfield soil.

Keywords: emission; greenhouse-gasses; land use; manure; *Zea mays*; nutrient management

Nitrous oxide (N_2O) is a potent greenhouse gas with a global warming potential of 298 times of CO_2 in a 100-year time horizon (IPCC 2007) and is the largest contributor to the stratospheric ozone layer depletion (Ravishankara et al. 2009). Agricultural soil is an important source accounting for about 40% of the total N_2O emission (IPCC 2001).

The increasing demand of organic agriculture may increase the use of organic matter for crop production in future. Application of manure enriches organic carbon sequestration in the soil; however, it stimulates N_2O flux (Singurindy et al. 2009).

Soil N_2O is produced through nitrification and denitrification processes (Davidson et al. 2000) and it is affected by soil moisture, temperature, and pH (Bouwman 1990). In addition to environmental factors, nitrogen from fertilizer and manure stimulates soil N_2O production in grassland in central and northern Japan (Mori et al. 2008). On the other hand, Shimizu et al. (2013) reported that there was no significant difference in annual N_2O flux between plots with mineral fertilizer and plots with combination of organic and mineral fertilizer in four sites of grasslands in northern to southern Japan, although there was a significant difference among the sites and the years.

Another study from Dambreville et al. (2008) indicated that organic fertilizer also corresponded to higher peak of N_2O flux than mineral fertilizer in cornfield in Brittany, France even though annual N_2O flux was not significantly different between the fields. The surplus of nitrogen played a key role in soil N_2O flux that was higher in cornfield than grassland (Katayanagi et al. 2008).

A combination of mineral and organic fertilizer application in grassland and cornfield has become a part of field management in Japan. The amount of nitrogen that is high enough will affect soil N_2O production. The objective of this study is to clarify the effects of nutrient managements, land uses and soil environments on soil N_2O flux in Southern Hokkaido, Japan.

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MATERIAL AND METHODS

The study was conducted in grassland in the livestock experimental farm of Hokkaido University and a cornfield in Niikapu station, the National Livestock Breeding Centre, southern of Hokkaido, Japan, from the beginning of May 2013 to the end of April 2014. Before 2012, grassland site was a cornfield and it was re-established to grassland from 2012 until now. Otherwise cornfield site was managed as grassland before 2012 and it was converted to cornfield in 2012. The elevation of both sites is 50 m at a 1–2% incline and the climate is characterized as humid continental with cold winters and cool summers, without wet or dry seasons. The mean annual precipitation and annual temperature (1981–2010) are 1290 mm and 8.1°C, respectively. The soil at both sites is classified as Andisols (USDA) with topsoil layer depth is 19 cm and 26 cm at grassland and cornfield, respectively. Soil pH of the top soil layer is 5.3–5.5. Grassland site has poor drained soil while cornfield has well drained soil. Sand content is 0.77 g/g and dominates the soil texture in grassland, followed by silt 0.19 g/g and clay 0.037 g/g. In cornfield, soil fraction contents of 0.71 g/g sand, 0.21 g/g silt and 0.08 g/g clay were reported.

Plots of the size 10 × 10 m with four replications were established for mineral fertilizer (F) as well as for mineral and organic fertilizer (MF) in grassland and cornfield. On May 25, 2013, timothy (*Phleum pratense* L.) and white clover (*Trifolium repens* L.) were sown with 21 and 4 kg/ha in grassland, respectively. Due to high weed infestation, renovation was decided, grass was harvested on July 19 and herbicide was applied on August 13. The grass and soil were incorporated by rotary plough on September 20, 22, and 24. On September 27, 2013, 80 kg/ha timothy (*P. pratense* L.) was sown.

40 kg N/ha in the form of urea was applied by topdressing method to F and MF plots on May 25, 2013 concurrently with grass seed sowing. 37 t/ha (448 kg N/ha and 7.3 t C/ha) of mixed woody bark and cattle manure was applied by broadcasting machine to MF on October 17, 2012. The renovation comprised 17 t fresh manure/ha (194 kg N/ha and 3.5 t C/ha) which was applied again on September 12, 2013.

Cornfield was ploughed on April 26, and corn (*Zea mays* L.) was sown at 24.6 kg/ha on May 10, 2013 and harvested on September 20. Whole corn plants were removed from the field except 30 cm of stubble and root. The N rate of the mineral fertilizer was 103 kg N/ha that is taken from urea and applied in F and MF plots on June 2, 2013 by topdressing method. 29 t fresh manure/ha (191 kg N/ha and 4.3 t C/ha) was applied to MF plot on September 9–17, 2013 using the broadcasting machine.

The N application rates of mineral fertilizer in grassland and cornfield were at the recommended level on the basis of soil tests done by the livestock farm staff, whereas the manure application rates were the optimum rates used by farmers in the region that were based on adequacy of potassium application to the fields (Jin et al. 2010).

Climatic data were obtained from the Japan meteorological agency. The soil frost depth was measured by the ethylene blue method (Richard et al. 1976), soil temperature at 5 cm depth by thermocouple thermometer (CT220, Custom, Tokyo, Japan).

Soil samples (composite and core) were taken from 0–10 cm depth during study with four replications and extracted using deionized water (at 1:5 ratio of soil to water), and 2 mol/L KCl (at 1:10 ratio of soil to water) then filtered through a 0.2-μm membrane. Soil-water was extracted and then was used to measure water extractable organic carbon (WEOC) by TOC analyser (TOC-5000A, Shimadzu, Kyoto, Japan) and NO_3-N concentration employing ion chromatography (Dionex, Sunnyvale, USA). NH_4-N concentration was measured using the indophenol-blue method from soil-KCl extract by spectrophotometer (UV-Vis Mini spectrophotometer 1240, Shimadzu, Kyoto, Japan). Water-filled pore space (WFPS) was determined from soil core using the soil moisture content by frequency domain reflectometry (FDR) method (DIK-311A, Daiki, Saitama, Japan). WFPS was calculated from calibration curves of the FDR device reading (m^3/m^3) and percentage of the total porosity (Linn and Doran 1984).

The N_2O and NO fluxes measurements were conducted in 2–28 day intervals during the growing season and in 10–30 day intervals during the non-growing season, between 8:00 and 11:00 h each measuring day to minimize the effect of diurnal temperature variation. The stainless steel chamber (40 cm in diameter and 30 cm high) was used for the measurement (Toma and Hatano 2007). The samples of N_2O were collected for 0, 15 and 30 min and for 0 and 30 min for NO samples from the chambers using a 50 mL syringe. N_2O was analyzed using gas chromatography with ECD

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(GC-2014 Model, Shimadzu, Kyoto, Japan) and NO using NOx analyser (265 P Model, Kimoto Electric, Osaka, Japan). Gas flux is the gradient of gas concentration in chamber over time. The cumulative gas flux was calculated by:

$$\sum_{i=1}^{n} R_i \times 24 \times D_i$$

Where: R_i – mean gas flux (mg/m^2/h) of the two successive sampling dates; D_i – number of days in the sampling interval; n – number of sampling times.

To investigate denitrification activity (DEA) in the beginning and the end of the winter season, an acetylene block technique that inhibits the transformation of N_2O to N_2 (Tiedje 1994) was performed. A 10 g of fresh soil in 100 mL conical flask was incubated under anaerobic condition with different solutions. The solutions consisted of 1 g/L of chloramphenicol for four treatments, (1) distilled water (Ctrl); (2) glucose of 300 mg C/L (+C); (3) KNO_3 of 50 mg N/L (+N), and (4) glucose of 300 mg C/L + KNO_3 of 50 mg N/L (+C+N). The flasks were evacuated and flushed four times with N_2 to ensure anaerobic conditions, and acetylene (C_2H_2) gas was added to a final concentration of 10% (10 kPa) in the headspace. Gas was sampled after 24, 48 and 72 h using syringe and directly analyzed for N_2O concentration by gas chromatography. DEA were calculated from the linear increase of N_2O production against the time. Statistical analysis (*t*-paired test and Pearson correlation) was performed using SPSS 16.0 (SPSS Inc., Chicago, USA).

RESULTS AND DISCUSSION

N_2O flux increased after the application of mineral fertilizer, organic fertilizer and precipitation. Annual mean of N_2O flux was higher in the MF than in the F plot, in addition to the flux in grassland, which tended to be higher than in cornfield. Daily N_2O flux in the F plot was sometimes higher than in the MF plot. Both sites had similar patterns of N_2O flux until July, but they had different patterns after August. A significant high peak occurred on September 18 in grassland, but not in cornfield. N_2O flux during the freezing period was low compared to that before freezing; however, during the melting period, N_2O flux increased in all plots other than MF plot of grassland. Then, after melting period, N_2O flux increased in grassland, but not in cornfield (Figure 1). There was a significant positive correlation between N_2O

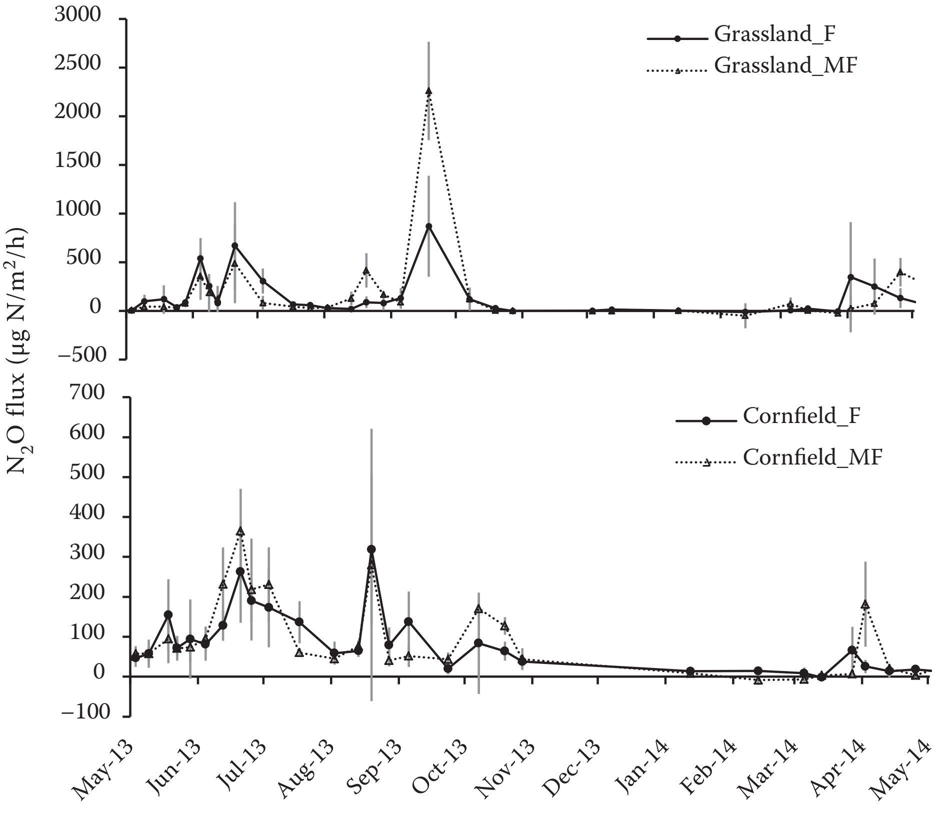

Figure 1. Soil N_2O flux in grassland and cornfield from 1 May 2013 to 30 April 2014. F – mineral fertilizer; MF – mineral combined with organic fertilizer

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Table 1. Correlation among N_2O flux and environmental variables

	N_2O flux	Air temperature	Soil temperature	NO_3-N	NH_4-N	WEOC
Air temperature	0.269**					
Soil temperature	0.276**	0.966**				
NO_3-N	0.050	0.143	0.242*			
NH_4-N	–0.016	0.044	0.118	0.441**		
WEOC	–0.002	0.075	0.065	–0.168	–0.201	
WFPS	0.212*	–0.225*	–0.265*	–0.191	–0.313**	0.027

*$P \geq 0.05$; **$P \geq 0.01$; WEOC – water extractable organic carbon; WFPS – water-filled pore space

flux and air, and soil temperature in both sites. A significant positive correlation also occurred between N_2O flux and WFPS. N_2O flux was not significantly correlated with NO_3-N, NH_4-N and WEOC (Table 1).

Annual N_2O flux in grassland was higher in MF than in F plot (14.9 and 11.1 kg N/ha/year, respectively). In cornfield, there was an equal annual N_2O flux between both plots (5.6 kg N/ha/year). Annual N_2O flux in both F and MF plots in grassland was remarkably higher than in cornfield (Table 2). More than 80% of the annual N_2O flux occurred before freezing. Cumulative N_2O flux during the melting period accounted for less than 4%. However, there was no contribution in MF plot of grassland during that period. After the melting period, cumulative N_2O flux was higher in grassland than in cornfield.

N_2O flux before soil freezing. The application of mineral fertilizer in all plots strongly influenced N_2O flux in June (Figure 1). Gagnon et al. (2011) obtained the similar result. Nitrification and denitrification are the process that control N_2O production in the soil. The ratio of N_2O and NO could be an indicator to find out the predominant process, nitrification or denitrification. Nitrification becomes the main process when the ratio is less than 1, on the other hand denitrification is the main process when the ratio is above 100. However if the ratio is 1–100, both nitrification and denitrification are the main processes in the soil N_2O production. In this study denitrification was the main process that increased N_2O flux in both grassland and cornfield (Figure 2)

Table 2. Cumulative N_2O flux (kg N/ha/period) in mineral (F) and mineral combined with organic fertilizer (MF) plots of grassland and cornfield

Period	Grassland				Cornfield			
	F		MF		F		MF	
	mean	SD	mean	SD	mean	SD	mean	SD
Before freezing	9.3	3.0	13.7	3.6	5.1	2.2	5.2	1.0
Freezing	0.0	0.1	0.0	1.1	0.2	0.1	0.0	0.1
Melting	0.4	0.7	0.0	0.1	0.2	0.1	0.2	0.1
After melting	1.4	1.6	1.3	0.4	0.1	0.1	0.3	0.1
Whole	11.1	2.9	14.9	4.5	5.6	2.2	5.6	1.0

Before freezing (1 May 2013–21 December 2013); freezing (22 December 2013–26 March 2014); melting (27 March 2014–2 April 2014); after melting (3 April 2014–30 April 2014); whole (1 May 2013–30 April 2014); SD – standard deviation

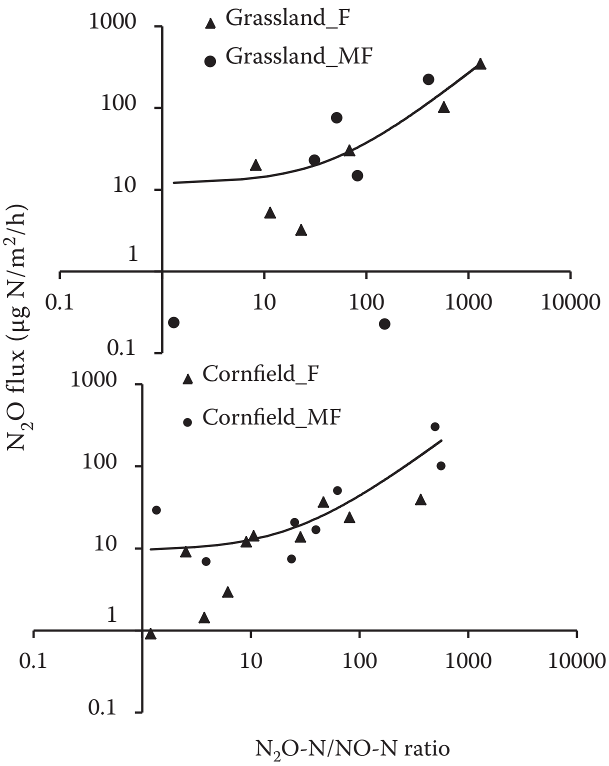

Figure 2. Relationship between N_2O flux and N_2O-N/NO-N ratio. F – mineral fertilizer; MF – mineral combined with organic fertilizer

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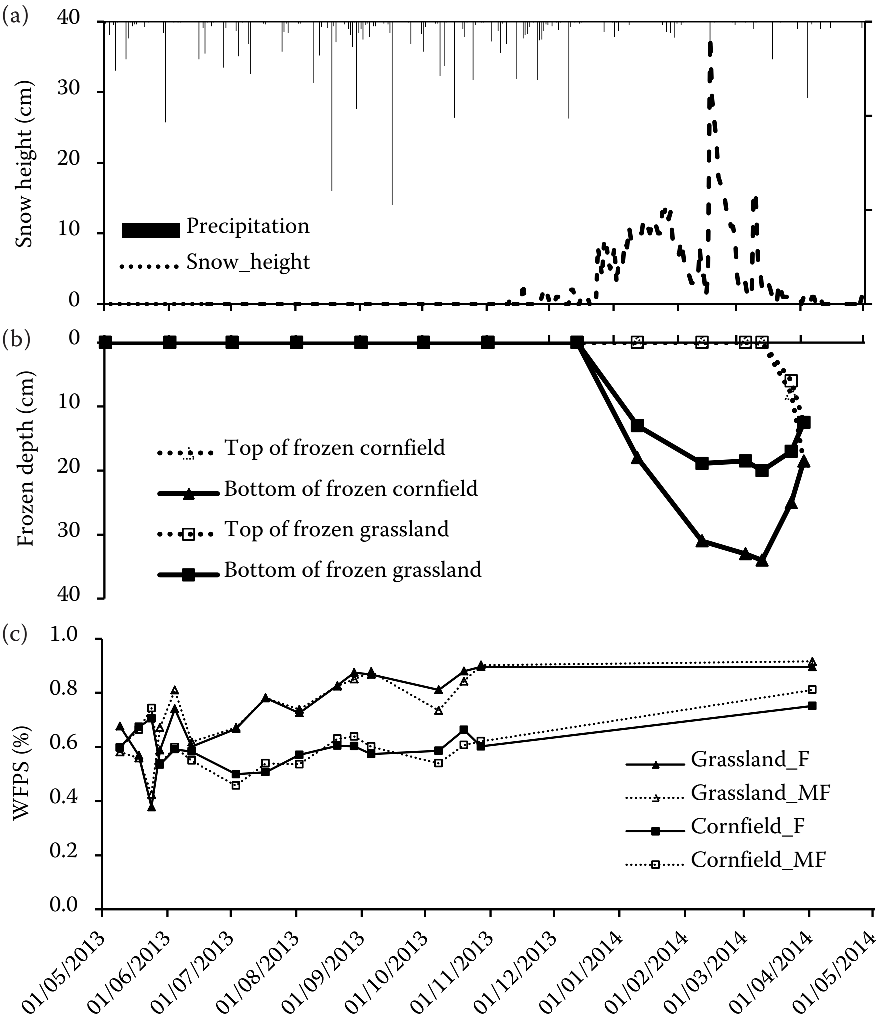

Figure 3. Seasonal pattern of (a) daily precipitation and snow depth; (b) soil frozen depth and, (c) water-filled pore space (WFPS) in grassland and cornfield from 1 May 2013 to 30 April 2014. F – mineral fertilizer; MF – mineral combined with organic fertilizer

which is also in accordance with Lipschultz et al. (1981). Higher WFPS in May and June (Figure 3c and Figure 1) supports the increase of N_2O flux along with denitrification. However, N_2O flux was similar in F and MF plots in both grassland and cornfield, regardless of the following tendencies; higher NO_3-N concentrations in F plot than in MF plot of both grassland and cornfield, while WEOC higher in MF plot than in F plot (Figure 4). This suggests that WEOC in F plot and NO_3-N in MF plot during May and June might be the limiting factors for denitrification next to soil moisture condition.

N_2O flux decreased concurrently with the decrease of NO_3-N concentration in July to middle of August in all plots (Figure 1). From the middle of August to the beginning of September, there was a peak of N_2O flux with precipitation (Figure 3a). The peak was relatively large in cornfield, but it was small in grassland. At that time, WEOC increased in all plots (Figure 4), and there was a peak of NO_3-N concentration in cornfield, yet a decline of NO_3-N concentration in grassland (Figure 4b). Therefore, the low NO_3-N concentration is thought to be the limiting factor for N_2O flux in grassland.

In grassland, there was a large peak of N_2O flux, i.e. 870 and 2260 μg N/m^2/h in F and MF plots, respectively, on September 18, but the peak did not appear in cornfield (Figure 1). This might be ascribed to the renovation in grassland. In the renovation, the decomposition of grass after the application of by herbicide and incorporation of top-dressed manure into the soil elevated the N_2O flux. On the other hand, in cornfield, there was a higher peak of N_2O flux in MF plot in October. Manure application on September had led to the increase of N_2O flux. After November, N_2O flux decreased.

Cumulative N_2O flux before freezing in grassland was higher than in cornfield. The N_2O flux is also higher in MF plot than in F plot in grassland (Table 2).

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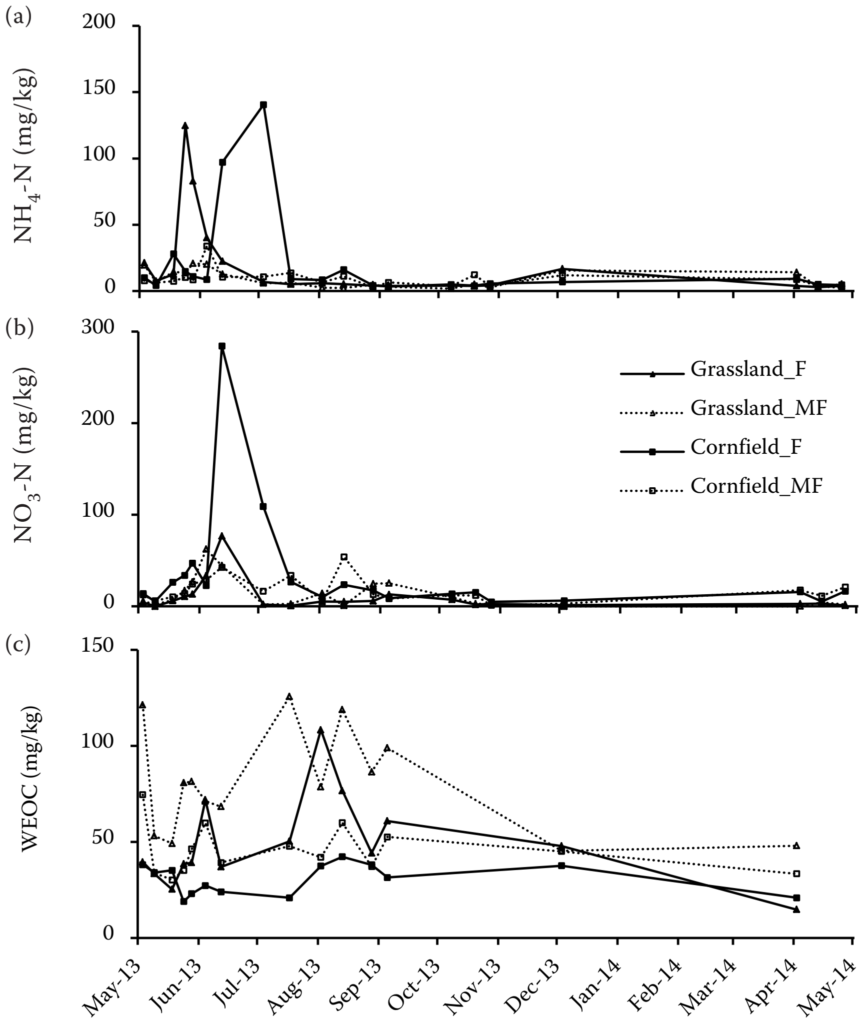

Figure 4. Concentration of (a) NH_4-N; (b) NO_3-N, and (c) water extractable organic carbon (WEOC) from soil at 0–10 cm depth in grassland and cornfield. F – mineral fertilizer; MF – mineral combined with organic fertilizer

Renovation resulted in higher cumulative N_2O flux in grassland. The grass biomass with carbon and nitrogen content 40.7% and 2.12%, respectively (C/N ratio 19), enriched soil organic matters after renovation and it was evidenced by the concentration of WEOC in both MF and F plots that were higher than in cornfield (Figure 1). High WEOC concentration was ascribed to the decomposition of incorporated organic matter. Organic matter decomposition might have led to ammonification and nitrification in grassland soils, which in turn increased denitrification and N_2O flux.

Unlike in grassland, the F and MF plots in cornfield showed a similar cumulative N_2O flux before the freezing period (Table 2). This situation did not reflect soil NO_3-N concentration that was lower in MF plot (18.3 mg/kg) than in F plot (38 mg/kg). Ammonia volatilization might have been a cause of the low soil NO_3-N concentration in MF plot. Ammonia volatilization from moist soil is 2–3 times higher than in air-dry soil (Meisinger and Jokela 2000). Soil was wet in MF plot in cornfield when the manure was applied on September 16, due to high precipitation of 97.5 mm on the same day.

Corn stubble with 30 cm height that remained in the cornfield after harvesting on September 2013 was one of the reasons for lower N_2O flux in cornfield than in grassland. Application of crop residues with carbon and nitrogen content 36.7% and 0.76% (C/N ratio 49.90) decrease organic matters decomposition (Toma and Hatano 2007) and usually leads to immobilization, which temporarily declined the availability of nitrogen and carbon in the soil that reduced N_2O flux in the cornfield.

N_2O flux after soil freezing. N_2O flux decreased significantly with the decrease of air and soil temperature (Table 1), Jin et al. (2010) reported the similar result. During the freezing period N_2O flux was very low below 30 μg/m^2/h, then it increased

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Table 3. Soil denitrification activity (DEA, μg N/kg/h) in the beginning and the end of winter season

Site		Treatment	The beginning of winter	The end of winter
Grassland	F	Ctrl	33.5 ± 16.1	19.2 ± 20.0
		+C	23.0 ± 23.9	39.9 ± 29.1
		+N	242.1 ± 184.7	102.2 ± 132.8
		+C+N	424.2 ± 227.4	262.8 ± 250.5
	MF	Ctrl	25.0 ± 11.3	88.5 ± 78.0
		+C	6.5 ± 3.9	241.3 ± 151.7
		+N	286.1 ± 13.3	251.1 ± 186.2
		+C+N	957.4 ± 37.5	355.9 ± 264.5
Cornfield	F	Ctrl	26.3 ± 10.2	77.1 ± 16.3
		+C	154.0 ± 46.7	101.0 ± 51.9
		+N	26.2 ± 9.3	84.7 ± 106.3
		+C+N	375.6 ± 61.0	206.7 ± 33.8
	MF	Ctrl	92.9 ± 70.5	37.9 ± 30.2
		+C	271.1 ± 64.9	86.6 ± 94.1
		+N	125.8 ± 94.9	92.7 ± 49.2
		+C+N	225.1 ± 221.8	127.0 ± 123.2

Data represent mean ± standard deviation. F – mineral fertilizer; MF – mineral combined with organic fertilizer; Ctrl – distilled water; +C – glucose of 300 mg C/L; +N – KNO_3 of 50 mg N/L; +C+N – glucose of 300 mg C/L + KNO_3 of 50 mg N/L

during the melting period in all plots except in MF plot of grassland (Figure 1). Low N_2O flux in MF plot was observed concurrently with very low NO_3-N concentration, which was 0.2 mg N/kg.

Although N_2O flux increased in melting period, the cumulative N_2O flux in melting period did not greatly contribute to annual flux, particularly the contribution in MF plot in grassland that was almost zero. In freezing and melting periods the contribution of N_2O in F and MF plots of grassland were 3.5% and 0%, respectively, whereas in cornfield N_2O flux contribution was 7.5% and 3.2% in F and MF plots, respectively. These results were considerably lower compared to other studies by Goossens et al. (2001) and Syvasalo et al. (2004).

In this study, the soil freezing disappeared on April 2 in 2014, but the previous investigation in 2005 showed that soil was totally melted on April 19 (Katayanagi and Hatano 2012). In that year, the starting date of the soil melting was rather earlier (March 24) than in 2014 (March 27). This circumstance decreased the contribution of N_2O flux during melting period. Melting of frozen soil proceeds from top and bottom, while ice layer remains at the depth of 15–20 cm (Figure 3b) and top soil is water-saturated in melting period. After melting period, the water drains from the soil and soil temperature increases, which was slightly shown by high N_2O flux in grassland (Figure 1).

In winter, denitrification was still a potential to be the main process of N_2O flux in both grassland and cornfield. It was potentially increasing when either carbon or nitrogen was available. In grassland soil, DEA increased in +N treatment ($P < 0.01$), but in cornfield soil DEA increased in +C treatment ($P = 0.05$) (Table 3). WEOC and NO_3-N concentrations had an important role in denitrification activity in grassland and cornfield. WEOC concentration in F and MF plots in grassland during freezing and melting was higher (15–48 mg/kg) than in cornfield; otherwise NO_3-N concentration was higher in cornfield than in grassland, i.e. 3–17 mg/kg (Figure 4). In cornfield, soil DEA in +N treatment did not increase differently than control (Table 3), indicating that NO_3-N in the soil was in sufficiency and therefore the addition gave a negative effect on denitrification activity. Luo et al. (1996) showed that the increase in NO_3-N concentration above 50 mg/kg soil was detrimental for denitrification activity. In cornfield, WEOC was always lower than in grassland. Carbon is very important as energy in denitrification. Low availability of carbon result in low denitrification and it is responsible for the abundance of NO_3-N concentration in the soil.

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REFERENCES

Bouwman A.F. (1990): Exchange of greenhouse gases between terrestrial ecosystems and the atmosphere. In: Bouwman A.F. (ed.): Soils and the Greenhouse Effect. Chichester, John Wiley and Sons, 61–127.

Davidson E.A., Keller M., Erickson H.E., Verchot L.V., Veldkamp E. (2000): Testing a conceptual model of soil emissions of nitrous and nitric oxides. BioScience, 50: 667–680.

Dambreville C., Morvan T., Germon J.C. (2008): N_2O emission in maize-crops fertilized with pig slurry, matured pig manure or ammonium nitrate in Brittany. Agriculture, Ecosystems and Environment, 123: 201–210.

Gagnon B., Ziadi N., Rochette P., Chantigny M.H., Angers D.A. (2011): Fertilizer source influenced nitrous oxide emissions from a clay soil under corn. Soil Science Society of America Journal, 75: 595–604.

Goossens A., De Visscher A., Boeckx P., Van Cleemput O. (2001): Two-years field study on the emission of N_2O from coarse and middle-textured Belgian soils with different land use. Nutrient Cycling in Agroecosystems, 60: 23–34.

IPCC (Intergovernmental Panel on Climate Change) (2001): Climate Change 2001: The Scientific Basis. New York, Cambridge University Press.

IPCC (Intergovernmental Panel on Climate Change) (2007): Climate Change 2007: Mitigation of Climate Change: Contribution of Working Group III to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change. New York, Cambridge University Press.

Jin T., Shimizu M., Marutani S., Desyatkin A.R., Iizuka N., Hata H., Hatano R. (2010): Effect of chemical fertilizer and manure application on N_2O emission from reed canary grassland in Hokkaido, Japan. Soil Science and Plant Nutrition, 56: 53–65.

Katayanagi N., Sawamoto T., Hayakawa A., Hatano R. (2008): Nitrous oxide and nitric oxide fluxes from cornfield, grassland, pasture and forest in a watershed in Southern Hokkaido, Japan. Soil Science and Plant Nutrition, 54: 662–680.

Katayanagi N., Hatano R. (2012): N_2O emissions during the freezing and thawing periods from six fields in a livestock farm, southern Hokkaido, Japan. Soil Science and Plant Nutrition, 58: 261–271.

Linn D.M., Doran J.W. (1984): Effect of water-filled pore space on carbon dioxide and nitrous oxide production in tilled and nontilled soils. Soil Science Society of America Journal, 48: 1267–1272.

Lipschultz F., Zafiriou O.C., Wofsy S.C., Mcelroy M.B., Valois F.W., Watson S.W. (1981): Production of NO and N_2O by soil nitrifying bacteria. Nature, 294: 641–643.

Luo J., White R.E., Ball P.R., Tillman R.W. (1996): Measuring denitrification activity in soils under pasture: Optimizing conditions for the short-term denitrification enzyme assay and effects of soil storage on denitrification activity. Soil Biology and Biochemistry, 28: 409–417.

Meisinger J., Jokela W.E. (2000): Managing Nutrients and Pathogens from Animal Agriculture (NRAES-130). New York, Ithaca, Natural Resources, Agricultural and Engineering Services.

Mori A., Hojito M., Shimizu M., Matsuura S., Miyaji T., Hatano R. (2008): N_2O and CH_4 fluxes from a volcanic grassland soil in Nasu, Japan: Comparison between manure plus fertilizer plot and fertilizer-only plot. Soil Science and Plant Nutrition, 54: 606–617.

Ravishankara A.R., Daniel J.S., Portmann R.W. (2009): Nitrous oxide (N_2O): The dominant ozone-depleting substance emitted in the 21st century. Science, 326: 123–125.

Richard J.A., Tobiasson W., Greatorex A. (1976): The Field Assembled Frost Gage. Hanover, US Army Corps of Engineers, Cold Regions Research and Engineering Laboratory.

Shimizu M., Hatano R., Arita T., Kouda Y., Mori A., Matsuura S., Niimi M., Jin T., Desyatkin A.R., Kawamura O., Hojito M., Miyata A. (2013): The effect of fertilizer and manure application on CH_4 and N_2O emissions from managed grasslands in Japan. Soil Science and Plant Nutrition, 59: 69–86.

Singurindy O., Molodovskaya M., Richards B.K., Steenhuis T.S. (2009): Nitrous oxide emission at low temperatures from manure-amended soils under corn (*Zea mays* L.). Agriculture, Ecosystems and Environment, 132: 74–81.

Syväsalo E., Regina K., Pihlatie M., Esala M. (2004): Emissions of nitrous oxide from boreal agricultural clay and loamy sand soils. Nutrient Cycling in Agroecosystems, 69: 155–165.

Tiedje J.M. (1994): Denitrifiers. In: Weaver R.W., Angle S., Bottomley P. (eds.): SSSA Book Series 5, Methods of Soil Analysis, Part 2: Microbiological and Biochemical Properties. Application Environmental Microbiology, SSSA, Madison, 245–257.

Toma Y., Hatano R. (2007): Effect of crop residue C:N ratio on N_2O emissions from Gray Lowland soil in Mikasa, Hokkaido, Japan. Soil Science and Plant Nutrition, 53: 198–205.

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