



Nitrous oxide emissions from grassland in spring and early summer at increasing nitrogen application rates

NIBIO REPORT | VOL. 6 | NO. 183 | 2020



Shahid Nadeem and Anne Kjersti Bakken Division for food production and Scociety

TITTEL/TITLE

Nitrous oxide emissions from grassland in spring and early summer at increasing nitrogen application rates

FORFATTER(E)/AUTHOR(S)

Shahid Nadeem, Anne Kjersti Bakken

	_				
DATO/DATE:	RAPPORT NR./ REPORT NO.:	TILGJENGELIGHET/AVAILABILITY:		PROSJEKTNR./PROJECT NO.:	SAKSNR./ARCHIVE
26.03.2021 6/183/2020 Op		en	10934	17/02938	
ISBN:		ISSN:	ANTALL SIDER/ NO. OF PAGES:	ANTALL VEDLEGG/ NO. OF APPENDICES:	
978-82-17-02739-3		2464-1162	26		

OPPDRAGSGIVER/EMPLOYER: Forskningsmidlene for jordbruk og matindustri/ Norwegian Research Funding for Agriculture and the Food Industry	KONTAKTPERSON/CONTACT PERSON:

STIKKORD/KEYWORDS :	FAGOMRÅDE/FIELD OF WORK:
Eng, lystgassutslipp, N ₂ O, nitrogengjødsling	Grovfôrdyrking
Leys, nitrogen fertilization, soil temperature, soil moisture	Forage production

SAMMENDRAG/SUMMARY:

Lystgassutslipp har blitt målt over to vekstsesonger i et feltforsøk i Trøndelag med gras gjødsla med stigende mengder nitrogen. Nedbør og temperatur varierte mye i de to åra. Lystgassutslippene var spesielt høye i perioder like etter vårgjødling og like etter gjødsling etter førsteslått dersom jordtemperaturen var over 10°C og jorda var godt fuktig. Avlingsmengden økte ikke i gjødslingsintervallet fra 24 til 32 kg N per daa og år, men lystgassutslippene økte betydelig i samme intervallet. I gjennomsnitt gikk 0,6% av tilført N tapt som lystgass i måleperioden, som ikke omfattet seinsommer, høst og vinter.

I rapporten diskuteres om balansert gjødsling, seinere gjødsling og deltgjødsling kan være gode strategier for å minske risikoen for denitrifikasjon og store lystgassutslipp fra eng om våren. Resultatene gir ikke grunnlag for å si at den offisielle utslippsfaktoren for lystgass på 1% av tilført N, bør heves, men samtidig er ikke vinterperioden med, og risikofaktorer som kløverinnslag og bruk av husdyrgjødsel ble ikke dekt av forsøket.

Summary

Nitrous oxide emissions from a perennial ley on a silty loam soil in central Norway have been recorded in spring and early summer over two years. In an experimental set up, five different N fertilization regimes were included, with the addition of 0-240-280-300-320 kg N per ha and year.



Emission rates were highest after fertilization in spring and early summer if the soil temperature was above 10°C and the soil moisture content was high. The grass yield did not vary significantly between the four treatments where N was added. The total nitrous oxide emissions per unit area was, however, twice as high at the highest level as at the "normal" level of 240 kg N ha⁻¹. On average, 0.6% of the added N was emitted as nitrous oxide. Different fertilization strategies that may reduce nitrous oxide emissions without reducing the yield or yield quality have been discussed. They include postponing fertilization until the crop is in a phase of rapid growth and split fertilization.

LAND/COUNTRY:	Norway	
FYLKE/COUNTY:	Trøndelag	
KOMMUNE/MUNICIPALITY:	Stjørdal	
STED/LOKALITET:	Kvithamar	
GODKJENT /APPROVED		PROSJEKTLEDER /PROJECT LEADER
Mats Höglind		Anne Kjersti Bakken
NAVN/NAME		NAVN/NAME



Preface

This report presents and discusses the results from a two-year study of N_2O emissions from a perennial grass crop according to nitrogen fertilization rate and weather conditions at a NIBIO research site in Stjørdal in Trøndelag.

The study was part of the project "Fertilization strategies for grasslands to reduce nitrous oxide emission in cold and wet springs" ("Gjødslingsstrategier for eng som minimerer lystgassutslipp i kjølige og våte vårer") financed by the Norwegian Research Funding for Agriculture and the Food Industry (Forskningsmidlene for jordbruk og matindustri) from 2018 to 2020.

Shahid Nadeem has designed the study, conducted all laboratory analyses and the field measurements in 2018. He has processed most of the data and been responsible for the report writing. Anne Kjersti Bakken has managed the field and conducted the field measurements in 2019 and contributed to the writing of the report.

The authors are thankful to the research technicians, advisors and researchers at NIBIO Kvithamar who took part in accomplishing the research activities, especially Anne Langerud, Randi Wikdahl and Dr. Lars Nesheim.

Moss, 26.03.21 Shahid Nadeem

Content

1	Introduction	6
	1.1 The nitrogen cycle and background for N ₂ O emission from agriculture	6
	1.2 N ₂ O emissions related to Norwegian agriculture	8
2	Material and Method	11
	2.1 Experimental site	. 11
	2.2 Treatment description and crop establishment	. 11
	2.3 Gas sampling	. 12
	2.4 Soil sampling	. 12
	2.5 Weather data	. 13
	2.6 Crop yield and nitrogen content	. 13
	2.7 Statistical analyses	. 14
3	Results	15
	3.1 Dry matter and N yields	. 15
	3.2 Temporal variation in N ₂ O emissions after fertilization	. 16
	3.3 Cumulative N ₂ O emissions	. 16
4	Discussion	20
	4.1 N ₂ O emissions according to soil moisture and temperature	. 20
	4.2 N_2O emissions according to fertilization rates and regimes	. 20
5	Conclusions	22
Re	ferences	23

1 Introduction

1.1 The nitrogen cycle and background for N₂O emission from agriculture

Nitrogen (N) is the most abundant element in the atmosphere (78 vol% N_2) where it is relatively stable apart from negligible N oxidation during electrical discharge (lightning). Anthropogenic activities seriously alter the N cycle (Sutton et al. 2011). Burning of fossil fuels, increased biological and massive industrial N fixation, which converts inactive N to reactive nitrogen (Nr), cause the main anthropogenic nitrogen loading of the biosphere (Galloway et al. 2004). The annual input of Nr to the biosphere today is more than double compared to preindustrial times, due to anthropogenic N fixation. This is mainly due to the escalating use of inorganic fertilizers needed to secure food production and to keep pace with a growing human population and its dietary preferences. Nitrogen application to the soil for increasing crop production could lead to high N losses to the atmosphere in the form of gaseous nitrous oxide (N₂O) emission, and nitrate (NO₃-) leaching beyond the crop root zone and runoff. All the N lost through leaching (NO₃-) or other ways will ultimately contribute to N₂O emissions directly or indirectly, these indirect emission are sometimes even larger than the direct emission down (Olesen et al. 2011). An efficient utilization of added N by plants will help to keep the N₂O

Nitrous oxide is one of the most important GHGs showing a steady increase in the atmosphere due to anthropogenic sources including crop management practices, particularly inorganic fertilizer application and fossil fuel combustion. Anthropogenic activities contribute more than 50% of total N₂O emissions on a global scale, to which agriculture activities have the biggest (81%) share (Isermann 1994). It is estimated that global annual N_2O emissions from agriculture are about 6 ×103 Gg N_2O -N. Direct N_2O emission from cultivation of arable soils contributes 42%, while indirect emissions from nitrate leaching, sewage, runoff and atmospheric N deposition account for 31% and animal production is responsible for the rest of 27% (Nevison 2000, IPCC 2007). The atmospheric N₂O concentration has increased about 20% over the past century and rises steadily at the rate of 0.25% annualy. Figure 1 shows the increase in N₂O concentration in the atmosphere from pre industrial times. Increasing atmospheric content of N₂O causes serious environmental concerns due to its high global warming potential of nearly 300 times more than the carbon dioxide in a 100 years perspective (IPCC 2007). Furthermore, high concentration of N₂O in the atmosphere could lead to ozon depletion by its breakdown to NO in the stratosphere (Crutzen & Lelieveld 2001, Ravishankra et al. 2009). In order to reduce the environmental footprint of food production we need to reduce the GHG emissions related to the crop production.



Figure 1. Increase in N₂O concentration in the atmosphere during the last century at 4 different measurement sites as shown in figure legend. Adapted from IPCC 2007 data in Eds. B. Metz, O. R. Davidson, P. R. Bosch, R. Dave and L. A. Meyer. Cambridge, UK; New York, NY: Cambridge University Press 2007.

Nitrous oxide in soils is produced mainly through two different microbial processes of nitrification and denitrification. Figure 2 summarizes the overall biological processes involved in the N fixation, and the return of fixed N to the atmosphere.



Figure 2. Microbiological N cycle, indicating the names of different processes that respire or assimilate nitrogen. Adapted from Thomson et al. (2012).

N₂O is formed as an intermediate product in the denitrification process. Additionally, N₂O can also be produced as a by-product in nitrification, nitrifier denitrification and dissimilatory nitrate reduction to ammonia (DNRA). N₂O emission from microbial processes of nitrification and denitrification is well explained by the hole in the pipe model illustrated in figure 3.



Figure 3. The 'hole in the pipe' model, describing the regulation of NO and N₂O flux: 1: The rates of nitrification and denitrification (the flow rate through the pipes). 2: The ratios of end products (size of the holes in the pipes). 3: Diffusion and consumption of gasses before they are lost to the atmosphere. Adopted from Firestone & Davidson (1989).

In agroecosystems, both nitrification and denitrification are regulated by a number of complex interlinked variables as shown in figure 4 and 5, which makes it difficult to predict N_2O emissions from agricultural soils in space and time due to these inter linked factors (Tiedje 1988, Beauchamp 1997, Stehfest & Bouwman 2006). Nitrous oxide emissions from cultivated soils are controlled by the availability of mineral N and degradable organic matter (Kaiser et al. 1998, Baggs et al. 2000, Aulakh et al. 2001), soil temperature (Stott et al. 1986, Smith et al. 1998), soil moisture (Marti 1984, Cheng et al. 2014) and pH (Simek & Cooper 2002, Cuhel et al. 2010, Liu et al. 2010). Fertilization regime (Eichner 1990, Bouwman 1996, MacKenzie et al. 1998, Smith et al. 1998, Tian et al. 2014), manure application and tillage operations (Choudhary et al. 2002, Venterea et al. 2005, Yonemura et al. 2014) affect N_2O emissions by directly or indirectly modulating these factors. Weather conditions in

the field change from day to day which make these emissions variable from day to day and even within a day, like diurnal variation related to the temperature.



Figure 4. Different variables regulating the nitrification process in agroecosystems. Adapted from Beauchamp (1997).



Figure 5. Different variables regulating the denitrification process in agro ecosystems. The importance of each of the three main factors (O₂, NO₃⁻ and C) is indicated by the thickness of the arrows. From Tiedje (1988).

1.2 N₂O emissions related to Norwegian agriculture

In Norway, GHG emissions from agriculture accounted for 9.2% of total annual emissions in 2015 (Snellingen Bye et al. 2016). CH_4 contributed 58%, while N_2O contributed about 38% of total GHG emissions. The National inventory report-2018-Norway documented that 74% of all N_2O emissions in Norway (Fig. 6) are related to the agriculture and agricultural soils being the most prominent contributors. Figure 7 shows the long term (1990-2018) contribution of different sectors to the overall N_2O emissions from Norway, where agriculture is the prominent and constant contributor throughout

this period. One reason for that constant share, could be that limited field scale data are available under Norwegian environmental conditions. Further, more than 90% of total NH_3 emissions originate from agriculture (Snellingen Bye et al. 2016), and about 1% of this NH_4 -N turns into N_2O -N (IPCC 2006), thus contributing to the total N_2O share of GHG emissions.

Ruminant production systems have a dominant position in Norwegian agriculture. About 60% of the cultivated area is under grassland production. Emissions from cultivated soil under grasslands therefore constitute a considerable part of the total agricultural GHG profile in the country. N_2O emissions from that type of soils are controlled by many factors like soil pH, temperature, moisture, cultivation and added N in the form of animal manure and mineral fertilizers. In most management regimes for silage production, at least half of the yearly dosage of N is added in spring, when plants are small and climatic conditions are cold and wet. The applied N may therefore be easily available to the microbes in the soil, and there is a high risk of N losses in form of N_2O .

Quantification of N₂O emission from Norwegian agricultural soils is limited. Currently the IPCC (2006) Tier 1 methodology is applied, which allows no differentiation according to regional and farmwise variation in soil characteristics and weather conditions. Previous studies conducted on grasslands by Hansen et al. (1993), Rivedal et al. (2013), Hansen et al. (2014) and Sturite et al. (2014) have shown great inter- and intra-annual variation in N₂O emissions, and that water saturation/oxygen tension as well as soil temperature can be important in regulating the N₂O emissions under ample amount of available N after fertilization. Still, it is no doubt that denitrifying microbes need easily available substrate N. In phases of initial or slow grass growth and N uptake, and simultaneously high N fertilization rates typically occurring in early spring in Norwegian grasslands, there might therefore be high risks for peaks in N losses.

In the present report, we have summarized and discussed the results from an experiment being part of the recent project "Fertlization strategies for grasslands to reduce nitrous oxide emission in cold and wet spring" financed by Norwegian Agriculture agency (Landbruksdirektoratet).

The over all aim of the project was to contribute knowledge of how N_2O emissions from Norwegian grasslands can be reduced.

The specific aims of the experiment was to quantify emission rates in early phases of spring and regrowth after cut when grass growth is slow and the soil N concentration is high after fertilization.



Figure 6. Distribution of $\,N_2O$ emissions by major sources during 2016 in Norway.

Source: Statistics Norway/Norwegian Environment Agency



 $\label{eq:sources} Figure \ 7. \ Distribution \ of \ N_2O \ emissions \ by \ sources \ during \ 1990-2016 \ in \ Norway.$ Source: Statistics Norway/Norwegian Environment Agency

2 Material and Method

2.1 Experimental site

A field trial was established in 2018 at a research farm (Kvithamar) of the Norwegian Institute of Bioeconomy Research (NIBIO) in Stjørdal, Trøndelag (63° 30 N, 10° 54 E; elevation 30 m a.s.l.). At Kvithamar the long-term normal value (1961-1990) for annual precipitation is 896 mm, of which 416 mm occurs during the growing season (May to September), and the average growing season temperature is 11.7 °C. The soil was a silty loam (21% clay and 63% silt) overlying silty clay loam, classified as typic Cryaquoll (Soil Survey Staff, 1998), Mollic Gleysol (WRB 1998) and an Orthic Humic Gleysol (CSSC, 1998). The plough layer contained 6-8% organic matter (by weight). The soil at the experimental site has a pH of 6.2. Weather condition at the site during the experimental period are shown in figure 8.



Study site at NIBIO research station Kvithamar (Stjørdal)

2.2 Treatment description and crop establishment

A grass mixture of timothy (*Phleum pratense* L.) variety Grindstad and meadow fescue (*Festuca pratensis* Huds.) variety Fure (0.8:0.2 by seed weight) was sown on 8 July in 2017 at a rate of 25 kg per ha. Just before sowing, 50 kg N ha⁻¹ was applied as NPK (22:3:10) compound fertilizer. All plots were harvested on 4 September in 2017, and thereafter, another 50 kg N ha⁻¹ was applied as compound fertilizer.

A field trial with five fertilization treatments was established in spring 2018. There were three replicates of each treatment arranged in three blocks. Each plot was 1.5 m \times 7.0 m.

The fertilization rates were 0, 240 (which is considered as normal fertilization rate in the area), 280, 300 and 320 kg N ha⁻¹ yr⁻¹ supplied from a 22:3:10 NPK fertilizer. Treatment details are given in table 1. In 2018, the zero N-treatment was supplied with 70 kg N ha⁻¹ after the second harvest to sustain grass growth and prevent weed and clover invation.

The dates for fertilizer application in 2018 and 2019, respectively, were 2 May and 26 April in spring, 26 and 7 June after 1st harvest and 29 and 22 July after 2nd harvest. All plots were harvested three times per year in June (26 and 7), July (29 and 22) and August/September (11 September and 30 August) in 2018 and 2019.

	ON	240N	280N	300N	320N
Spring	0	100	125	135	145
After 1 st harvest	0	70	85	95	105
After 2 nd harvest	0	70	70	70	70
SUM	0	240	280	300	320

Table 1. Nitrogen fertilization rate (kg N ha⁻¹) on field plots in 2018 and 2019.

2.3 Gas sampling

Nitrous oxide (N₂O) emissions were measured by manually operated static chambers (Nadeem et al. 2012), consisting of quadratic aluminium frames (51 cm × 51 cm, 20 cm depth) and chambers (51 cm × $51 \text{ cm} \times 19.5 \text{ cm}$ (height)). In spring 2018, the frames were inserted into the field by pushing 10-15 cm into the soil in each plot. The metal frames had a groove $(3 \text{ cm} \times 3 \text{ cm})$ on top, which was filled with water prior to deploying a chamber to ensure an air tight connection between frame and chamber (Maljanen et al. 2003). Aluminium chambers were equipped with a 3-way stop cock sampling port and a 3 mm diameter pressure equilibrium tube. N₂O flux measurements were carried out by deploying the chambers on the frames for 45 minutes, and taking 15 ml gas samples with 20 ml polypropylene syringes at regular intervals (0, 15, 30 and 45 min). The air inside the chamber was well mixed by pulling and pushing the plunger of the syringe 3-4 times to obtain a well-mixed sample from the chamber. The sample was transferred to a pre-evacuated 12 ml glass vial crimped with a butyl rubber septum. Gas sampling was done weekly or biweekly during the growing seasons of 2018 and 2019. Frames were removed just before harvesting and placed back shortly after. Samples were sent to Norwegian University of Life Sciences (NMBU) and were analysed by gas chromatography (Model 7890A, Agilant, Santa Clara, CA, US) using a 30 m wide-bore Poraplot Q (0.53 mm) column at 38 °C with back flushing and He as a carrier gas. The electron capture detector (ECD) conditions were 375 °C with 17 ml min⁻¹ ArCH₄ (10/90 vol%) as makeup gas. N_2O emission rates were calculated from the linear or quadratic change in chamber gas concentration using the equation

 $F_{N2O} = d_{N2O} / dt \times V_c / A \times M_n / V_m \times 60$

where F_{N2O} is the emission flux of N_2O in μ g N_2O -N m⁻² h⁻¹, d_{N2O} the rate of change in N_2O concentration (ppmv min⁻¹) in the chamber headspace, V_c the volume of chamber (L), A the area covered by the chamber (m²), M_n the molecular mass of N in N_2O (g mol⁻¹) and V_m the molecular volume of gas at chamber temperature (L mol⁻¹). Cumulative N_2O emissions (kg N_2O -N ha⁻¹ period⁻¹) were calculated assuming average flux between two measurement dates.

2.4 Soil sampling

Soil samples (0-20 cm) were taken at every date of gas sampling by a 30 mm diameter soil corer to determine soil moisture and in some instances mineral N (NH_4^+ and NO_3^-) content. In both years, 4 soil cores were sampled per plot and pooled to one composite sample. Soil moisture was determined

by drying 20 g fresh soil at 105 °C until constant weight. Water filled pore space (WFPS) was calculated as (gravimetric soil moisture × bulk density)/soil porosity, assuming a soil density of 2.65 g cm⁻³. Soil mineral N (NH₄⁺, NO₃⁻) was determined by extracting 45 g fresh soil with 50 ml 2M KCl solution. Soil extractants were centrifuged at 10,000 g for 15 minutes at 4 °C. NO₃⁻ and NO₂⁻ concentrations in the supernatant were determined by colorimetry using a microplate reader (Infinite F50, TECAN Austria GmbH) at 540 nm. Both NO₃⁻ and NO₂⁻ were measured by Griess reaction (Keeney & Nelson 1982), with NO₃⁻ being converted to NO₂⁻ by vanadium chloride (Doane & Horwath 2003).

2.5 Weather data

Weather data were collected from a nearby station located at the same research farm (Kvithamar, Stjørdal). Weather conditions in spring varied greatly between the two study years with 2018 being very dry and warm, while 2019 being cold and wet.



Figure 8. Weather data from Kvithamar weather station during the growing seasons 2018 and 2019. Dates for fertilization are indicated by arrows.

2.6 Crop yield and nitrogen content

All plots were harvested with a Haldrup experimental harvester at a stubble height of 7 cm. Raw weights from each plot were recorded and representative sub samples were taken to determine the dry matter content and plot dry weight. The sub samples were dried for approximately 48 hours at 60 °C. Sub samples from the dried plant material were chopped and grinded, and N content (crude protein content/6.25) was determined by NIRS analyses at NIBIO Løken (Fystro & Lunnan 2006).

2.7 Statistical analyses

Cumulative N_2O emissions within season, and yields of dry matter and nitrogen for each harvest and in sum for all harvests within season were analysed according to analyses of variance, with fertilization treatment as fixed effect and block as random. Means were separated by a post hoc Tukey test if the ANOVA model turned out significant (p<0.05).

3 Results

3.1 Dry matter and N yields

Dry matter yield (kg ha⁻¹) of all the three harvests in the two production years are presented in table 2. Dry matter yield was significantly higher in fertilized than in zero N treatments, but it did not vary significantly between the fertilized treatments in 2018. In the second harvest in 2019, however, the treatment with the highest N application rate returned higher yields than the rest of the treatments.

Table 2. Dry matter yield (kg ha⁻¹) of grass in three harvests in 2018 and 2019. Presented values are averages of three replicates. Means within rows marked with different letters were significantly different according to a Tukey test (p<0.05).

Treatment	ON	240N	280N	300N	320N
2018					
1 st harv.	3330ª	4750 ^b	4550 ^b	4580 ^b	4820 ^b
2 nd harv.	320ª	2440 ^b	2980 ^b	3120 ^b	3080 ^b
3 rd harv.	1920ª	2280 ^{ab}	2350 ^{ab}	2750 ^b	2560 ^{ab}
SUM	5580ª	9470 ^b	9880 ^b	10450 ^b	10460 ^b
2019					
1 st harv.	1930 ^a	5010 ^b	4850 ^b	4680 ^b	4420 ^b
2 nd harv.	1340ª	3690 ^b	3730 ^b	3880 ^b	4770 ^c
3 rd harv.	490ª	1460 ^b	1510 ^b	1530 ^b	1420 ^b
SUM	3770ª	10160 ^b	10090 ^b	10090 ^b	10610 ^b

Total N content in harvested plant biomass (N yields) are shown in table 3. Nitrogen uptake by plants increased with increasing fertilization rate, but total N uptake with increasing fertilizer was less than the applied extra N, thus leading to extra N available in soil (Figure 14). The N balance was negative (less N taken up in plants than applied as mineral N) in the warm and dry production year of 2018. In 2019, the proportion of applied N that was taken up by the grass crop seemed to be higher.

Table 3.	Nitrogen content in grass yield (kg N ha ⁻¹) in three harvests during 2018 and 2019. Presented values are
	averages of three replicates. Means within rows marked with different letters were significantly different
	according to a Tukey test (p<0.05).

Treatment	0 N	240N	280N	300N	320N
2018					
1 st harv.	50 ^a	89 ^b	93 ^b	90 ^b	96 ^b
2 nd harv.	4 ^a	36 ^b	53 ^c	52 ^{bc}	57 ^c
3 rd harv.	43ª	54 ^{ab}	59 ^{ab}	68 ^b	63 ^{ab}
SUM	97ª	179 ^b	205 ^{bc}	210 ^{bc}	215 ^c
2019			· · · ·		<u>. </u>
1 st harv.	32 ^a	110 ^b	116 ^b	122 ^b	116 ^b
2 nd harv.	22ª	82 ^b	82 ^b	81 ^b	109 ^b
3 rd harv.	11ª	39 ^b	38 ^b	39 ^b	41 ^b
SUM	65ª	231 ^b	236 ^b	242 ^b	266 ^b

3.2 Temporal variation in N₂O emissions after fertilization

Figure 9 shows the temporal variation in N_2O emission during spring and early summer of 2018. N_2O emissions increased rapidly after fertilization in the warm and wet spring, and remained high for two weeks after fertilization until it returned back to what seemed to be the normal background level. In the warm and dry summer of 2018 (Figure 8), we did not observe any emission peak after the 2^{nd} fertilization in June (Figure 9).

Figure 10 shows the N_2O emission pattern in 2019 over most of the growing season. Rather than high emission peaks after fertilization in spring we observed a slow and small increase over time in the cold and wet spring (Figure 8). However in late spring with high temperature and wet weather we observed high emission peaks right after the 2nd fertilization, which stayed high for the next 4 weeks (Figure 10). This year we prolonged our measurements into late summer to monitor residual effects of high fertilization rates on N₂O emission after the 2nd harvest, when all plots were supplied with equal amount of fertilizer. During this period, soils were very dry (Figure 13), and the emission were very low in all treatments (Figure 10).

3.3 Cumulative N₂O emissions

Cumulative N_2O emissions over the study period were significantly higher in fertlized grass as compared to the zero N treatment (Figure 11). An increase in the fertilization rate from 240 to 320 kg N ha⁻¹ doubled the total N_2O emission. In average, 0.6% of applied N was lost as N_2O during the 3 months of study in both years. Total dry matter yield did not increase with increasing fertlization rate above 240 kg N ha⁻¹ (table 2), while total cumulative N_2O emissions increased significantly from 240 to 320 kg N ha⁻¹ (Figures 11 and 12).



Figure 9. Temporal variation in N₂O emission from a grass field under different fertilization regimes during spring and early summer in 2018. Presented values are averages of 3 replicates.



Figure 10. Temporal variation in N₂O emission from a grass field under different fertilization regimes during spring and early summer in 2019. Presented values are averages of 3 replicates.



Figure 11. Cumulative N₂O emissions from grass swards according to fertilization regime over the measurement periods of 83 and 96 days during 2018 and 2019, respectively. Columns within years marked with different letters were significantly different according to a Tukey test (p<0.05).



Figure 12. Relationship between the N₂O emission, total DM yield and fertilization rate during the two years of study. Presented values are the average of 3 replicates in each treatment.



Figure 13. Soil temperature and volumetric water content (VWC) at 5 cm depth during the two years of study. Data were collected from loggers installed at the experimental site.



Figure 14. Mineral N (NH₄⁺and NO₃⁻) content in soil during the study period of 2018 and 2019. Presented values are the average of three replicates of each treatment.

4 Discussion

4.1 N₂O emissions according to soil moisture and temperature

Weather plays a significant role in controlling the magnitude of N_2O emissions from agricultural soils. It has previously been shown that N_2O emission rates are highly dependent on soil moisture content (Marti 1984, Cheng et al. 2014) and temperature (Stott et al. 1986, Smith et al. 1998). Smith et al. (1998) confirmed that the increase in N_2O emissions rates with increasing temperature also is related to the increasing respiratory demand for O_2 , resulting in more and larger anaerobic zones.

The weather conditions were very different beween the two years of study on the present investigation site. Emissions rates were highest in spring 2018 and after first cut in 2019 when soil moisture and temperature were high, whereas low emission rates were observed when soils were cold and/or dry after 1st harvest in 2018 and in early spring in 2019, thus shows the importance of weather conditions in controlling the N_2O emissions between the seasons and years.

Optimum soil moisture conditions favouring denitrification and its higher $N_2O:N_2$ product ratio are in the range of 60-80% water filled pore space (WFPS) depending on the soil type (Davidson et al. 2000, Skiba & Smith 2000). At higher soil moisture content (over 80% WFPS) the $N_2O:N_2$ product ratio of denitrification decreases and the major end product will be N_2 (Weier et al. 1993, Butterbach-Bahl et al. 2013). At very low soil moisture content (more aerated conditions) nitrification is the main dominating process, which has lower $N_2O:N$ product ratio than denitrification. In our study, peak N_2O emission were observed when the volumetric water content were in the range of 0.30 – 0.35 (60-70% WFPS), which is in agreement with numerous previous studies. Regarding the soil temperature in our study, high emissions were only observed at temperatures above 10°C, which shows that microbial activity is retareded at low temperature.

If this knowledge was to be applied for the development of fertilization regimes that prevent N₂O emissions, one might in first hand suggest that N should be applied in early spring when soil temperatures are low. However, under such conditions, plants will deplete the pool of mineral N in the soil very slowly which leaves surplus substrate for denitrifying microbes for a longer period of time. Thus, the best option will likely be to apply the N in periods with rapid plant growth although they may coincide with periods with high risks of denitrification and N₂O emissions. This dilemma will probably also occur if springs will be warmer and even wetter in future (Hanssen-Bauer et al. 2015).

The effects of soil water content on N_2O emission rates may have been indirect via N availability in the soil as well. If it is too dry, fertilizer granules will be slowly dissolved and the diffusion and bulk flow of nitrogen solved in water will also be retarded. This might have been the situation at the application of fertilizer after the first cut in 2018. Although there were a few rain showers and a sudden and short increase in soil water content around the fertilization date, a large water deficit had accumulated after a long periode of high temperatures and no precipitation. Both the mineral N content in the soil and the N_2O emissions were shown to be low, giving some support to the hypothesis of low substrate availability due to diffusion constraints. However, the concentration of both NO_3^- and NH_4^+ was as high as under the high emission episodes in spring the same year (Figure 14).

4.2 N_2O emissions according to fertilization rates and regimes

The yield response to N application above 240 kg ha⁻¹ was low or absent in both years in the present experiment, whereas N_2O emission rates increased considerably in all intervals between 240 and 320 kg ha⁻¹. This illustrates that the option of balancing N application rates to crop growth potential stands as a valid practice for reducing N_2O emissions, both in absolute terms and relative to crop yield. This holds even if the "balancing dosage" applied in spring (100 kg N) or after regrowth (70 kg N) is far

higher than what is taken up by the grass crop during the following 2-3 weeks, when surplus N is easily available for the microbes to nitrify and denitrify and cause gaseous N losses as N_2O and N_2 . Although all fertilization rates from 240 to 320 kg N ha⁻¹ caused high N surplus relative to plant uptake in spring and after cut, it seemed to be very important for total N_2O emissions how large surpluses were. Total emissions were twice as high at 320 kg N ha⁻¹ as at 240 kg N ha⁻¹.

When evaluating the environmental burden of food and feed production, it might also be relevant to express N_2O emission intensity per crop yield and not only per area farmed land. In the present study, this ratio was lower for the 240N treatment than for the oN treatment. It should, however be noted that this holds only for emissions during the growing season. We have only accounted for emissions in non-continuous periods from late April until August, and not for those occurring during autumn and winter. Residual N accumulated in soil and remaining plant parts were likely higher in the 240N regime than in the zero N regime, and Sturite et al. (2014) have demonstrated that these pools can be sources for high N_2O emission from grassland during winter time and may account a substaintial part of total annual N_2O emissions.

Total N_2O emissions during the current measurement period (three months) amounted to nearly 0.6 % of the N applied as fertilizer in both years. These results are in line with previous findings in cereal production in south east Norway (Nadeem et al. 2015), where N_2O emissions ranges from 0.5-0.6% during the growing season (May-October). This proportion (0.6%) is lower than the IPCC emission factor of 1%, but again we have to take into regard that the present study has covered only one fourth of the year, and that the N_2O measurements were not continuous. Annually, total N_2O emissions are mainly controlled by event driven peaks, like after fertilization, drying-rewetting, freezing-thawing, and soil cultivation (crop residue incorporation) etc, most of them were not captured in this study.

In order to lower the N dosage that is applied in early phases of spring growth and regrowth, which implies risks for high N₂O emission rates, split fertilization within growing periods may be an alternative, at least for the part of N supplied from mineral fertilizers. Late spreading of animal manure implies risks for contamination of silages with unwanted microorganisms, and will probably involve damage and lodging of the crop, irrespective of spreading method. Broadcasted granulated fertilizers supplied according to plant demand by use of precise and high capacity centrifugal spreaders under optimal weather conditions may, however, be an option with acceptable trafficking in the standing crop.

5 Conclusions

- Critical conditions for N_2O emissions from soil were warm and wet periods after fertilization, before small plants efficiently absorbed added N.
- Moderate N fertilization could reduce N₂O emissions to some extent.
- Heavy fertilization in spring did not lead to increased grass yield but increased total N₂O emissions.
- Under the predicted warm and wet spring conditions in Norway in future, heavy fertilization would imply high risks for N losses and N₂O emissions.
- Late or split fertilization according to predicted grass plant N uptake capacity could be a preventive measure, but will be difficult to implement.
- Avoiding heavy fertilization during periods of high soil temperature and moisture content could be another preventive measure to reduce N₂O emissions. Such a practice will, however, likely imply risks of suboptimal nitrogen supply when growing conditions are good.
- This study, covering only parts of the year, gave no basis for suggesting adjustments of the official N_2O emission factor of 1% of added nitrogen.
- Animal manure application and clover inclusion in the perennial crop would probably have added significantly to total N_2O emissions.

References

- Aulakh, M.S., Khera, T.S., Doran, J.W. & Bronson, K.F. 2001. Denitrification, N₂O and CO₂ fluxes in rice-wheat cropping system as affected by crop residues, fertilizer N and legume green manure. Biology and Fertility of Soils 34 (6): 375-389.
- Baggs, E.M., Rees, R.M., Smith, K.A. & Vinten, A.J.A. 2000. Nitrous oxide emission from soils after incorporating crop residues. Soil Use and Management 16 (2): 82-87.
- Beauchamp, E. 1997. Nitrous oxide emission from agricultural soils. Canadian Journal of Soil Science 77 (2): 113-123.
- Bouwman, A.F. 1996. Direct emission of nitrous oxide from agricultural soils. Nutrient Cycling in Agroecosystems 46 (1): 53-70.
- Butterbach-Bahl, K., Baggs, E.M., Dannenmann, M., Kiese, R. & Zechmeister-Boltenstern, S. 2013. Nitrous oxide emissions from soils: how well do we understand the processes and their controls? Phil Trans R Soc B 368: 20130122: <u>http://dx.doi.org/10.1098/rstb.2013.0122</u>
- Chadwick, D., Sommer, S., Thorman, R., Fangueiro, D., Cardenas, L., Amon, B. & Misselbrook, T. 2011. Manure management: Implications for greenhouse gas emissions. Animal Feed Science and Technology 166–167: 514–531.
- Cheng, Y., Wang, J., Wang, S.Q., Zhang, J.B. & Cai, Z.C. 2014. Effects of soil moisture on gross N transformations and N₂O emission in acid subtropical forest soils. Biol Fertil Soils. 50: 1099–1108.
- Choudhary, M., Akramkhanov, A. & Saggar, S. 2002. Nitrous oxide emissions from a New Zealand cropped soil: tillage effects, spatial and seasonal variability. Agriculture, Ecosystems & Environment 93 (1-3): 33-43.
- Crutzen, P.J. & Lelieveld, J. 2001. Human impacts on atmospheric chemistry. Annu. Rev. Earth Planet. Sci. 29: 17–45.
- CSSC., 1998. The Canadian system of soil classification. Soil classification working group (3rd ed., pp. 187). Ottawa, New Zealand: CSSC.
- Cuhel, J., Simek, M., Laughlin, R. J., Bru, D., Cheneby, D., Watson, C. J. & Philippot, L. 2010. Insights into the Effect of Soil pH on N₂O and N-2 Emissions and Denitrifier Community Size and Activity. Applied and Environmental Microbiology 76 (6): 1870-1878.
- Davidson, E.A., Keller, M., Erickson, H.E., Verchot, L.V. & Veldkamp, E. 2000 Testing a conceptual model of soil emissions of nitrous oxides. Bioscience, Washington, DC. 50 (8): 667-680.
- Doane, T.A. & Horwath, W.R. 2003. Spectrophotometric determination of nitrate with a single reagent. Analytical Letters 36: 2713-2722.
- Eichner, M. J. 1990. Nitrous oxide emissions from fertilized soils: summary of available data. Journal of Environmental Quality 19 (2): 272-280.
- Firestone, M.K. & Davidson, E.A. 1989. "Microbial basis of NO and N₂O production and consumption in soil." In M.O.Andreae and D.S.Schimel, editors, Exchange of trace gases between terrestrial ecosystems and the atmosphere. John Wiley and Sons Ltd. Chichester. 7-21 pp.
- Fystro, G. & Lunnan, T. 2006. Analyser av grovfôrkvalitet på NIRS. Bioforsk FOKUS 1 (3): 180-181.
- Galloway, J.N., Dentener, F.J., Capone, D.G., Boyer, E.W., Howarth, R.W., Seitzinger, S.P., Asner, G.P., Cleveland, C.C., Green, P.A., Holland, E.A., et al., 2004. Nitrogen Cycles: Past, Present, and Future. Biogeochemistry 70 (2): 153-226.

- Hansen S., Bernard M-E., Rochette P., Whalen, J.K. & Dörsch, P. 2014. Nitrous oxide emissions from a fertile grassland in Western Norway following the application of inorganic and organic fertilizers. Nutr. Cycl. Agroecosyst 98: 71-85.
- Hansen, S., Maehlum, J.E. & Bakken, L.R., 1993. N₂O and CH₄ fluxes in soil influenced by fertilization and tractor traffic. Soil Biol. Biochem. 25: 621-630.
- Hanssen-Bauer, I., Førland, E.J., Haddeland, I., Hisdal, H., Mayer, S., Nesje, A., Nilsen, J.E.Ø.,
 Sandven, S., Sandø, A.B., Sorteberg, A. & Ådlandsvik, B. 2015. KLIMA I NORGE 2100:
 Kunnskapsgrunnlag for klimatilpasning oppdatert i 2015. NCCS report no. 2/2015. 204 sider (in Norwegian).
- IPCC. 2006. In: Eggleston, H.A., Buendia, L., Miwa, K., Ngara, T. & Tanabe, K. (Eds), IPCC guidelines for national greenhouse gas inventories, prepared by the National greenhouse inventories programme, IGES, Japan.
- IPCC. 2007. Climate change 2007: the physical science basis: contribution of Working Group I to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change. Cambridge Univ. Press.
- Isermann, K. 1994. Agriculture's share in the emission of trace gases affecting the climate and some cause-oriented proposals for sufficiently reducing this share. Environ. Pollut. 83: 95-111.
- Kaiser, E.A., Kohrs, K., Kücke, M., Schnug, E., Munch, J. & Heinemeyer, O. 1998. Nitrous oxide release from arable soil: importance of perennial forage crops. Biology and Fertility of Soils 28 (1): 36-43.
- Keeney, D.R. & Nelson, D.W. 1982. Nitrogen inorganic forms. In: Methods of soil analysis, Part 2-Chemical and microbiological properties, Eds. A.L. Page, R.H. Miller and D. R. Keeney. pp. 643-698. American Society of Agronomy Inc. and Soil Science Society of America, Inc., Madison, Wisconsin, USA.
- Liu, B. B., Morkved, P. T., Frostegard, A. & Bakken, L. R. 2010. Denitrification gene pools, transcription and kinetics of NO, N2O and N-2 production as affected by soil pH. Fems Microbiology Ecology 72 (3): 407-417.
- MacKenzie, A. F., Fan, M. X. & Cadrin, F. 1998. Nitrous oxide emission in three years as affected by tillage, corn-soybean-alfalfa rotations, and nitrogen fertilization. Journal of Environmental Quality 27 (3): 698-703.
- Marti M. 1984. Kontinuierlicher Getreidebau ohne Plug im Süd¬osten Norwegens Wirkung auf Ertrag, Physikalsche und Chemische Bodenparameter. (Continuous cereal production with ploughless cultivation in South-Eastern Norway - Effects on yields and soil physical and chemical parametres). (in German with English summary). Dr. scient thesis, Agricultural University of Norway, 155 pp.
- Nadeem, S., Hansen, S., Azzaroli Bleken, M. & Dörsch, P. 2012. N₂O emission from organic barley cultivation as affected by green manure management. Biogeosciences 9: 2747-2759.
- Nadeem, S., Børresen, T. & Dörsch, P. 2015. Effect of fertilization rate and ploughing time on nitrous oxide emissions in a long-term cereal trail in south east Norway. Biology and Fertility of Soils 51: 353–365. <u>https://doi.org/10.1007/s00374-014-0979-7</u>
- Nevison, C. 2000. Review of the IPCC methodology for estimating nitrous oxide emissions associated with agricultural leaching and runoff. Chemosphere-Global Change Science 2: 493-500.
- Olesen, J. E., Schelde, K., Weiske, A., Weisbjerg, M. R., Asman, W.A.H. & Djurhuus, J. 2006. Modelling greenhouse gas emissions from European conventional and organic dairy farms. Agric. Ecosyst. Eviron 112(2–3): 207–220.

- Ravishankara A.R., Daniel J.S. & Portmann R.W. 2009. Nitrous oxide (N₂O): the dominant ozonedepleting substance emitted in the 21st century. Science 326:123.
- Rivedal, S., Hansen, S. & Dörsch, P. 2013. Lystgassutslepp frå eng. Verknad av jordpakking etter gjødsling. Bondevennen 116 (22): 14-15.
- Simek, M. & Cooper, J. E. 2002. The influence of soil pH on denitrification: progress towards the understanding of this interaction over the last 50 years. European Journal of Soil Science 53 (3): 345-354.
- Skiba, U. & Smith, K.A. 2000. The control of nitrous oxide emissions from agricultural and natural soils. Chemosphere-global change science 2: 379-386.
- Smith, K., Thomson, P., Clayton, H., McTaggart, I. & Conen, F. 1998. Effects of temperature, water content and nitrogen fertilisation on emissions of nitrous oxide by soils. Atmospheric Environment, 32 (19): 3301-3309.
- Snellingen Bye, A., Aarstad, P.A., Løvberget, A.I. & Høie,H. 2016. Jordbruk og miljø. Tilstand og utvikling, Statistisk sentralbyra, Statistics Norway, Oslo, 160 pp.
- Soil Survey Staff, 1998. Keys to soil taxonomy (8th ed., p. 326). Washington, DC: USDA.
- Stehfest, E. & Bouwman L. 2006. N₂O and NO emission from agricultural fields and soils under natural vegetation: summarizing available measurement data and modelling of annual emissions. Nutr Cycl in Agroecosyst. 74: 207-228.
- Stott, D. E., Elliott, L. F., Papendick, R. I. & Campbell, G. S. 1986. Low- temperature or low water potential effects on the micrbial decomposition of wheat residue. Soil Biology & Biochemistry 18 (6): 577-582.
- Sturite I., Rivedal, S. & Dörsch, P. 2014. Effects of fertilization and soil compaction on nitrous oxide (N₂O) emissions in grassland. Grassland Science in Europe, Vol. 19: EGF at 50: the future of European Grasslands.
- Sutton, M. A., Howard, C. M., Erisman, J. W., Billen, G., Bleeker, A., Grennfelt, P., van Grinsven, H. & Grizzetti, B. 2011. Assessing our nitrogen inheritance. In Sutton, M. A., Howard, C. M., Erisman, J. W., Billen, G., Bleeker, A., Grennfelt, P., Van Grinsven, H. & Grizzetti, B. (eds) The European nitrogen assessment: sources, effects and policy perspectives. Cambridge: Cambridge University Press. 1-6.
- Thomson, A. J., Giannopoulos, G., Pretty, J., Baggs, E. M. & Richardson, D. J., 2012. Biological sources and sinks of nitrous oxide and strategies to mitigate emissions. Philosophical Transactions of the Royal Society B-Biological Sciences 367 (1593): 1157-1168.
- Tian X.F., Hu H.W., Ding Q., Song M.H., Xu X.L., Zheng Y. & Guo L.D. 2014. Influence of nitrogen fertilization on soil ammonia oxidizer and denitrifier abundance, microbial biomass, and enzyme activities in an alpine meadow. Biol Fertil Soils 50:703–713.
- Tiedje, J. M. 1988. Ecology of denitrification and dissimilatory nitrate reduction to ammonium. In: Zehnder, A. J. B. (ed), Biology of anaerobic microorganisms. John Wiley & sons, New York. pp. 179-244.
- Venterea, R. T., Burger, M. & Spokas, K. A. 2005. Nitrogen oxide and methane emissions under varying tillage and fertilizer management. J. Environ. Qual. 34: 1467–1477.
- Weier, K.L., J.W. Doran, Power, J.F. & Walters, D.T. 1993. Denitrification and the Dinitrogen/Nitrous Oxide Ratio as Affected by Soil Water, Available Carbon, and Nitrate. Soil Sci. Soc. Am. J. 57: 66-72.

- WRB 1998. World reference base for soil resources. World Soil Resources Report 84 (pp. 88). Rome, Italy: ISRIC and FAO.
- Yonemura S., Nouchi I., Nishimura S., Sakurai G., Togami K. & Yagi K. 2014. Soil respiration, N₂O, and CH_4 emissions from an Andisol under conventional-tillage and no-tillage cultivation for 4 years. Biol Fertil Soils 50:63–74.



NIBIO - Norwegian Institute of Bioeconomy Research was established July 1 2015 as a merger between the Norwegian Institute for Agricultural and Environmental Research, the Norwegian Agricultural Economics Research Institute and Norwegian Forest and Landscape Institute.

The basis of bioeconomics is the utilisation and management of fresh photosynthesis, rather than a fossile economy based on preserved photosynthesis (oil). NIBIO is to become the leading national centre for development of knowledge in bioeconomics. The goal of the Institute is to contribute to food security, sustainable resource management, innovation and value creation through research and knowledge production within food, forestry and other biobased industries. The Institute will deliver research, managerial support and knowledge for use in national preparedness, as well as for businesses and the society at large. NIBIO is owned by the Ministry of Agriculture and Food as an administrative agency with special authorization and its own board. The main office is located at Ås. The Institute has several regional divisions and a branch office in Oslo.

Cover photo: Anne Kjersti Bakken