Nitrous oxide emissions from drained organic and mineral soil: a study on hemi boreal Spruce forests

Mohammad Aurangojeb

UNIVERSITY OF GOTHENBURG
FACULTY OF SCIENCE

Licentiate Thesis
University of Gothenburg
Department of Earth Sciences
Gothenburg, Sweden 2017

Nitrous oxide emissions from drained Organic and mineral soil: a study on hemi boreal Spruce forests

Mohammad Aurangojeb

Nitrous oxide emissions from drained Organic and mineral soil: a study on hemi boreal Spruce forests

Internet ID: http://hdl.handle.net/2077/51523
Printed by Kompendiet AB, Gothenburg, 2017

Copyright © Mohammad Aurangojeb 2017
Distribution: Department of Earth Sciences, University of Gothenburg, Sweden
To my family
Mohammad Aurangojeb

**Abstract**

Terrestrial soils are the major source of N$_2$O, related to the soil N cycle and influenced by many factors. Forest soils have in general lower emission than soils in agricultural use. However higher emission than commonly found in agriculture can be found for some forest soils, as for the drained peat soil at the Skogaryd research site.

To understand the N$_2$O flux size and its controls two soil types existing in the Skogaryd area were investigated, drained peat and mineral soil, having high fertility and the same type of spruce forest. Thus we were able to keep weather conditions similar and only the soil types were different. Field measurements were conducted during three years, where soil emissions were sampled by manual closed chambers. To investigate soil gross N turnover processes at the organic site a $^{15}$N tracer field study was conducted for control plots and plots without either only roots or both roots and ectomycorrhizae.

Over the years the average emission from the drained organic soil was six times higher than from the mineral soil (4.2 ± 0.1 and 0.7 ± 0.1 kg N$_2$O ha$^{-1}$ yr$^{-1}$, respectively), despite slightly more fertile soil at the mineral site. The emissions varied over the year for both the organic and mineral soils where the large emissions were found during summer especially following precipitation after dry periods. Precipitation and temperature are thus influencing factors. The higher emission for the organic site was initially suggested connected to a larger soil organic matter (SOM) content of this soil, in percentage of top 0.30 m soil, however the SOM amount (kg/m$^2$) was similar for both sites, thus other suggestions were sought for. For the mineral site, both above ground biomass and mycorrhizae growth were much higher than those for the organic site resulting in a higher nitrogen demand and less N available for nitrification and denitrification. The importance of mycorrhizae was also shown in the trenching experiment at the organic site. Here the presence of roots and mycorrhizae stimulated microbial NH$_4^+$ immobilization more than NH$_4^+$ oxidation to NO$_3^-$, explaining a lower N$_2$O emission.

In summary, the findings of this thesis showed that N$_2$O emission differed between drained organic and mineral soil with higher emissions from the former in same climate conditions. It also suggest that tree roots and mycorrhizae affect soil N cycle through uptake of mineralized N and by stimulating microbial N immobilization thereby keep the N$_2$O emission down.

**Keywords:** N$_2$O emissions, organic soil, mineral soil, spruce forest, soil organic matter, $^{15}$N tracer, roots and mycorrhizae, mineralization, immobilization, nitrification, denitrification.
Populärvetenskaplig sammanfattning

Den största källan för tredje viktigaste växthusgasen lustgas (N$_2$O) är från marken, vilket är naturligt men påverkas av markanvändning. Lustgasen bildas främst i nitrifikation och denitrifikation och påverkas av många faktorer. Skogsmark har vanligtvis lägre emission än jordbruksmark, men även skogsmark kan ha höga emissioner. Till exempel så har dränerad torvmark vid Skogaryd forskningsstation visats ha hög avgång av lustgas.

För att förstå varför så mycket N$_2$O avgår och vad som påverkar flödet undersöks två närliggande områden i Skogaryd vilka har olika typ av mark, dränerad torv och en mineraljord, med liknande bördighet och samma typ av granskog. Mätningar i fält gjordes under tre år, där gaser som avges från marken samlades upp manuellt med hjälp av kammare. Dessutom undersöks med spårämnesanalys ($^{15}$N) de processer som omsätter kväve (N) i den organogena jordens kontrollyta, och ytor där antingen bara rötter exkluderats eller både rötter och mykorrhizasvampar.

Emission av lustgas var sex gånger högre från den dränerade torvjorden jämfört med mineraljorden, i medeltal 4.2 ± 0.1 respektive 0.7 ± 0.1 kg N$_2$O ha$^{-1}$ år$^{-1}$, trots en något högre bördighet för mineraljorden. Emissionerna varierade också med årstiden på båda ytorna, där de största emissionerna ägde rum under sommaren och särskilt då i samband med regn efter en torr period. Nederbörd och temperatur visade sig vara viktiga faktorer som påverkar emissionen. Till en början förklarades den högre emissionen på den organogenan ytan med att marken har en högre halt organiskt material i ytjorden, men den totala mängden av organiskt material i ytjorden var lika så en annan förklaring söktes efter. Något som skiljde sig åt mellan ytorna var skogens tillväxt ovan jord samt tillväxt av mykorrhizasvampar i jorden, vilka båda var mycket högre på mineraljorden. Eftersom dessa efterfrågar mer kväve blir mindre kväve tillgängligt för nitrifikation och denitrifikation. Betydelsen av mykorrhiza visades också genom ett experiment där rötter eller både rötter och mykorrhizasvampar hållits undan från experimentjorden. Närvaro av både rötter och mykorrhizasvampar ökar på mikroorganismernas upptag av kväve och mindre blir då över för nitrifikation, vilket kan förklara en lägre N$_2$O emission.

Sammanfattningsvis, resultat i denna avhandling visar att under samma väderförhållanden skiljer sig N$_2$O-emission mellan dränerad torvmark och mineralmark, där torvmarken hade högre emission. Resultaten pekar också på att trädens rötter och dess mykorrhiza påverkar kväve-cyklen genom att själva ta upp kväve och stimulera markmikroorganisms kväveupptag, och därigenom hålla nere N$_2$O emissionen.
List of papers

This thesis includes following two papers:


M. Aurangojeb conducted field work, data collection, data analyses and writing of the paper together with supervisor


M. Aurangojeb was responsible for N$_2$O data collection, data analyses, took part in situ $^{15}$N labelling experiment and contributed to part of writing
Table of content

Introduction ..................................................................................................................................................... 9

Materials and Method .................................................................................................................................... 13

  Site descriptions ........................................................................................................................................ 13

  N\textsubscript{2}O Flux Measurements ........................................................................................................ 14

  Measurement of abiotic variable and Soil properties ................................................................................. 14

  \textsuperscript{15}N labelling, soil sampling and Analysis of \textsuperscript{15}N ..................................................... 15

Result and discussion .................................................................................................................................... 16

  N\textsubscript{2}O emission from forested drained organic and mineral soils ..................................................... 16

  Factors influencing soil N\textsubscript{2}O emissions ....................................................................................... 17

Conclusions .................................................................................................................................................... 22

Acknowledgement ......................................................................................................................................... 23

References ..................................................................................................................................................... 24
Part I

Summary
Introduction

The earth’s surface temperature has increased by 0.85 [0.65 to 1.06] °C, over the period 1880–2012, due to increased emissions of greenhouse gases (GHGs) to the atmosphere (Hartmann et al., 2013). Carbon dioxide (CO\textsubscript{2}), methane (CH\textsubscript{4}) and nitrous oxide (N\textsubscript{2}O) are the most important anthropogenic GHGs and the atmospheric concentration of these gases has increased significantly since the preindustrial time. Among these anthropogenic GHGs, the emission of N\textsubscript{2}O is of particular concern because of its high global warming potential which is 265 times that of CO\textsubscript{2} in a 100 year perspective (Myhre et al., 2013). In addition to the global warming potential, the N\textsubscript{2}O gas is projected as the largest stratospheric ozone-depleting substance for the remainder of this century (Ravishankara et al., 2009).

Nitrous oxide emissions from terrestrial soils are assumed to be the major source of atmospheric N\textsubscript{2}O (IPCC, 2007). And the emissions from terrestrial soils are found to be largely influenced by anthropogenic activities including land use and land use change (Leppelt et al., 2014), e.g. agricultural soils are the largest source of N\textsubscript{2}O emission but forest soils have in general low emission. Also, natural undrained peatlands are known as a minor source for N\textsubscript{2}O emission (Martikainen et al., 1993; Von Arnold et al., 2005). However when peatlands are drained for agriculture and forestry, the decomposition rate of the organic matter of peat increases releasing both carbon (C) and nitrogen (N) and thus leading to enhanced N\textsubscript{2}O emission (Kasimir-Klemedtsson et al., 1997; Martikainen et al., 1993). Globally around 60 Mha of peatland have already been drained for agriculture or forestry which is 0.3 percent of world’s land cover (FAO, 2012). In Sweden, 1.5 Mha of forested drained peatland exists which is 6 % of total of the total 23 Mha productive forestry area and N\textsubscript{2}O emission from these forests on drained peat were found to be of the size 15% of the total anthropogenic N\textsubscript{2}O emission from Sweden (Ernfors et al., 2008).

High emissions of N\textsubscript{2}O have been recorded from forested drained peat soils in temperate and boreal region (Klemedtsson et al., 2005; Maljanen et al., 2012). A review study compiling data on emission measurement of N\textsubscript{2}O in forested soils showed emission from mineral soils to be low compared to drained organic soils (Maljanen et al., 2010). However, our knowledge on flux difference between soil types is primarily based on individual studies which focused on either the drained peat or mineral soils. Diverse environmental conditions make it difficult to compare N\textsubscript{2}O emissions from different studies since in diverse conditions other factors than soil type could be important for emission. Therefore, to compare N\textsubscript{2}O emissions between different types of soil, it is important to try to keep other conditions such as weather and vegetation as similar as possible.

In this thesis the N\textsubscript{2}O emissions from two different types of forested soils: a drained organic soil (Histosol) and a drained mineral soil (Umbrisol) were investigated (Paper I). The sites were closely located (within 1 km) and both of them were afforested with Norway spruce after abandonment of agricultural activities, thus exposed to the same land use history, climate conditions and vegetation composition. The work hypothesis was N\textsubscript{2}O emission from forested drained organic soil are higher compared to mineral soil in same climatic conditions.
since drained organic soil have higher soil organic matter (SOM) content (in percentage) compared to the mineral soil.

The emission of N\(_2\)O from terrestrial ecosystems is directly related to the soil N cycle, which is complex in nature as it includes several simultaneously occurring processes (Hart et al., 1994; Myrold and Tiedje, 1986; Nason and Myrold, 1991). Briefly, the soil N cycle includes mineralization, immobilization, nitrification and denitrification processes. The major terrestrial reservoir of N is SOM. In the mineralization process the organic N compound of SOM is transformed into ammonium (NH\(_4^+\)) which is then either taken up by plants or immobilized by microbes (Booth et al., 2005), or used by nitrifiers for nitrification. In nitrification, oxidation of NH\(_4^+\) (autotrophic nitrification) or organic N compound (heterotrophic nitrification) produce nitrate (NO\(_3^-\)) via nitrite (NO\(_2^-\)) and N\(_2\)O is produced as a byproduct of the reactions (Wrage et al., 2001). Denitrification is the reduction of NO\(_3^-\) to molecular N\(_2\) via N\(_2\)O and is a heterotrophic process which takes place under anaerobic conditions as heterotrophic denitrifiers use NO\(_3^-\) as a terminal electron acceptor only when O\(_2\) is unavailable. The microbial nitrification and denitrification are the major N transformation processes involved in N\(_2\)O production in soil (Firestone and Davidson, 1989). In addition, nitrifier denitrification and chemodenitrification are known N\(_2\)O producing processes in soils (Wrage et al., 2001). These processes of the N cycle are influenced by the environment, such as ecosystem type, soil type, land management, weather and climate, and living communities of plants and heterotrophs (Canary et al., 2000; Chapman et al., 2006; Gödde and Conrad, 2000; Mary et al., 1996).

Plants affect soil N cycling through several mechanisms; uptake of N, retain in tissues which are then slowly released via roots turnover in rhizosphere. Roots turn over and associated mineralization are known to be a major component of soil available N in rhizosphere (Frank and Groffman, 2009). Also, plants host a variety of microbial communities in their rhizosphere and enhance the growth and activity of microorganisms through exudation of labile carbon (C) via roots (Frank and Groffman, 2009; Hütsch et al., 2002). Additionally, the exudation of labile C via plant roots stimulate production of microbial enzymes for degradation of complex soil organic N compound which in turn facilitated nutrient availability for plant uptake (Frank and Groffman, 2009). Plant and microbes interaction, thereby, influence soil N dynamics and higher gross N mineralization in the rhizosphere compared to bulk soil, has already been observed in previous studies conducted in laboratory condition (Herman et al., 2006; Landi et al., 2006).

Most of the plant roots in temperate and boreal forests host ectomycorrhizae fungi (ECM) (Taylor et al., 2000) which play a key role in uptake of nutrients through enhancing the availability N to plant (Powell and Klironomos, 2007). A few studies have investigated the effect of roots/ECM on gross N transformation rates in situ (Holub et al., 2005; Ross et al., 2001), however the effect of ECM on gross N transformation is not clear. In a previous study by Ernfors et al. (2011), using trenching experiment on organic soils at Skogaryd, noticed two times higher N\(_2\)O emissions after exclusion of roots and mycorrhizal mycelia. This higher emission was explained by increased N availability for N\(_2\)O producing
microorganisms due to reduced plant uptake of N from soil through the mycorrhizal fungi. In the paper II of this study, the gross rates of N transformation on the same site was investigated using in situ $^{15}$N tracer study to elucidate how internal N transformation rates changed as a consequence of exclusion of roots and roots plus ECM which resulted in enhanced N$_2$O emissions. The understanding of the influence of soil-plant’s roots and ECM interaction on soil N turnover could improve our understanding of plants control on N$_2$O emissions. This understanding is included into some models. One example is the process based CoupModel which recently been calibrated on the Skogaryd organic site and used to simulate N$_2$O emission (He et al., 2016a; He et al., 2016b). Here the ground water levels together with nutrient uptake by roots were the most influential factors. However, here the ECM interaction was not yet included which may have improved the result. Modelling N$_2$O emission is always difficult due the many influential factors, processes and thresholds needed to pass for high emissions to occur.

In contrast to this study gross soil N dynamics in soil are traditionally determined by $^{15}$N pool dilution experiments in the laboratory, where soils are often mixed and/or sieved which may alter factors that influence soil N transformations, such as N pool sizes and mobility and, root biomass and the microbial community structure, especially ectomycorrhizal hyphae (Frank and Groffman, 2009). For instance, Booth et al. (2006) noticed that soil mixing promotes gross mineralization and NH$_4^+$ consumption. Therefore, the virtual core approach proposed by Rütting et al. (2011) was used in the study of paper II which allowed us investigating gross N dynamics under field conditions in minimum disturbed soils.

In this study process-related gross N transformation rates were quantified by a numerical data analysis based on a $^{15}$N tracing model where parameters are optimized using the Markov chain Monte Carlo (MCMC) parameter optimization technique (Müller et al., 2007). The advantage of the $^{15}$N tracing model with numerical data analysis is that numerical $^{15}$N tracing model provides the advantage to estimate gross nitrogen transformation rates from several simultaneously occurring gross nitrogen transformation processes, while analytical equations quantify only total gross production and consumption rates of the labelled N pools (Barraclough and Puri, 1995; Schimel, 1996). Moreover, this approach allows longer study periods (1–2 weeks) than commonly used isotope dilution experiments with analytical data analysis (usually 1–2 days)(Rütting et al., 2011). Combining this calculating method and the virtual core approach allowed us to reveal the interaction between soil, plant roots and their associated microbial communities including mycorrhizae and N transformation rates in field condition. As root exudates stimulate microbial activity, here the hypothesis was that trenching reduces both gross mineralization and NH$_4^+$ immobilization rates; and due to decreased NH$_4^+$ immobilization the relative importance of nitrification for NH$_4^+$ consumption increases which results in higher soil N$_2$O emissions after exclusion of roots and ECM.
Aims

The aims of this thesis were to:
• Quantify and compare N\textsubscript{2}O fluxes from afforested drained organic and mineral soils (Paper I)
• Elucidate how plants and their mycorrhizal symbionts control soil N cycling and affect N\textsubscript{2}O emissions from forest soils (Paper II)
Materials and Method

Site descriptions

**Fig. 1:** Organic and mineral site at Skogaryd Research Catchment

For the work described in paper I, field measurement were conducted at two closely located sub sites at the Skogaryd catchment, a part of the SITES station network (www.fieldsites.se), located in southwest Sweden (58°23′N, 12°09′E) (Fig. 1). The subsites were on two different types of soil; Histosol and Umbrisol (FAO 2015) referred to as organic site and mineral site, respectively in this thesis. Both are similarly drained (described in paper I). Experiments for paper II were performed on the organic subsite only. The sub sites were drained in the 1870s and used for agriculture until afforested with Norway spruce in the 1950s. At both sites Norway spruce (*Picea abies*) dominates the forest, with some Birch trees (*Betula verrucosa*) and a sparse field and bottom layer. Important characteristics of the sites are given in table (Table 1). The long term (1961–1990) mean annual temperature was 6.4°C and mean annual precipitation 709 mm, recorded at a nearby weather station in Vänersborg, situated 12 km from the study area (Alexandersson and Eggertsson Karlström, 2001).

At each sub site the \( \text{N}_2\text{O} \) fluxes were measured from three measurement stations: O1, O2, and O6 at the organic site, established in a previous trenching experiment (Ernfors et al., 2011), and M1, M2, and M3 at the mineral site, established during this study. The distances between stations were 11-28 m at the organic site and 8-29 m at the mineral site. Each station was comprised of two flux measurement plots and there were three collars installed with a maximum distance of 1.5-5 m apart in each plot. Thus, a total of 18 collars were present at each site (Fig. 1, paper I) for \( \text{N}_2\text{O} \) measurement. In the trenching experiment by Ernfors et al. (2011), the three collars of each plot at the organic site were randomly assigned to one of
the three treatments: (a) control (ctrl), (b) roots excluded (exclR) and (c) roots and ECM excluded (exclRM). Detailed description of the trenching can be found in Ernfors et al. (2011). For comparability with the mineral site, N\textsubscript{2}O emissions data only from the control chambers of organic site were used in paper I.

Table 1. Some important characteristics of organic and mineral site

<table>
<thead>
<tr>
<th></th>
<th>Organic Site</th>
<th>Mineral Site</th>
</tr>
</thead>
<tbody>
<tr>
<td>SOM content (%) †</td>
<td>74(± 8)</td>
<td>17(± 3)</td>
</tr>
<tr>
<td>SOM amount (kg/m\textsuperscript{2}) †</td>
<td>50 ± (6)</td>
<td>55± (12)</td>
</tr>
<tr>
<td>Bulk Density (g/cm\textsuperscript{3}) †</td>
<td>0.2(± 0.0)</td>
<td>1.1(± 0.1)</td>
</tr>
<tr>
<td>Tree age in 2010</td>
<td>60 years</td>
<td>60 years</td>
</tr>
<tr>
<td>Above-ground biomass (ton dry weight ha\textsuperscript{-1})</td>
<td>180±0.9</td>
<td>300±1</td>
</tr>
</tbody>
</table>

Note: †, mean over 0.05-0.30 m depth

\textit{N_{2}O Flux Measurements}

The flux measurements were conducted using dark stainless steel chamber as described in Ernfors et al. (2011). During August 2010 to July 2013, I conducted N\textsubscript{2}O flux measurement from all plots at both organic and mineral site and flux data from control chambers were used in the work described in paper I. For the work of the paper II, flux data from all control and trenched chambers measured during 2010-2013 at the organic site and the flux data measured by Ernfors et al. (2011) for the period of July 2006 to Dec 2009 were used. Fluxes of N\textsubscript{2}O at the soil surface were generally measured biweekly during the morning or early afternoon. A detailed description of the chamber and the procedure of gas sampling is given in paper II and Ernfors et al. (2011). The collected gas samples were analyzed by gas chromatography (Agilent 7890A, Agilent Technologies, Santa Clara, CA, USA) equipped with an autosampler (7697A). The N\textsubscript{2}O fluxes were calculated from the slope of the linear regression of gas concentrations plotted against time.

\textit{Measurement of abiotic variable and Soil properties}

Air temperature data were collected with Campbell 107 Temperature Probes (Campbell Scientific Inc) at a level of 2 m above the ground at the organic site. Soil temperatures were manually measured at two depths (0.1 and 0.2 m) at both sites, concurrently with the gas sampling (paper I, II). Groundwater level (GWL) was only measured at the organic site since the mineral site had a compact hard soil layers at a depth of around 0.4 m which made its difficult to install the tubes. The measurements were performed manually using a plumb
line lowered into perforated tubes inserted to a soil depth of 1.5 m next to each chamber. Soil samples were collected close to the chambers for determination of soil SOM, pH (KCl), total carbon (C), total nitrogen (N) content and C/N ratio. The SOM content was determined by loss-on-ignition where the soil samples were dried at 65°C for 48 hours and then burned at 550°C for 6 hours. The SOM content was calculated from the weight loss of the soil samples. To determine the total N and C, the oven dried soil samples were milled in order to homogenize and the samples were then analyzed with an elemental analyzer couple to isotope ratio mass spectrometer (IRMS) (20-20, Sercon Ltd, Crewe, Cheshire, UK).

15N labelling, soil sampling and Analysis of 15N

In paper II, for investigating the gross soil N dynamic under field conditions the soil at organic site was labelled with 15N using the virtual soil core approach (Rütting et al., 2011; Staelens et al., 2012) in May 2013. The 15N labelling was conducted by injecting either 15NH4NO3 or NH415NO3 (99%) into the soils and the detail description of 15 N labelling and soil sampling is available in paper II. Gross N mineralization and nitrification was calculated using the 15N tracing model Ntrace. Detailed description of this process is given in the paper II.
Nitrous oxide emissions from drained Organic and mineral soil: a study on hemi boreal Spruce forests

**Result and discussion**

*N₂O emission from forested drained organic and mineral soils*

Both, drained organic and mineral soils were overall net sources for N₂O emission with the median value of flux rates 38 and 6 µg m⁻² h⁻¹ at the organic and mineral site respectively, although at a few occasions both soils also act as a sink. A pronounced seasonal pattern in N₂O emissions was observed at both sites with occasions of high fluxes to occur in summer (Fig. 4 in *paper I*). The annual emissions for all years were higher at the organic site (Fig. 2) and over the three years period this site showed in average six times higher N₂O flux than the mineral site (Table 2 in *paper I*), which agrees with the result reported by Maljanen et al. (2010) showing higher N₂O emission from drained organic soils compared to the mineral soils in a review study on N₂O emissions.

<table>
<thead>
<tr>
<th>Year</th>
<th>Organic site (kg N₂O ha⁻¹ yr⁻¹)</th>
<th>Mineral site (kg N₂O ha⁻¹ yr⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td>'10/11</td>
<td>2</td>
<td>0</td>
</tr>
<tr>
<td>'11/12</td>
<td>4</td>
<td>2</td>
</tr>
<tr>
<td>'12/13</td>
<td>6</td>
<td>4</td>
</tr>
</tbody>
</table>

**Fig 2:** Annual nitrous oxide (N₂O) emission from organic and mineral soil in Skogaryd catchment in south-western Sweden and error bars show standard errors of the mean

The annual N₂O fluxes from the organic site ranged between 4.0 and 4.4 kg N₂O ha⁻¹ during the period 2010 to 2013, which is within the range of the reported values for drained organic soils (0.4–8.1 kg N₂O ha⁻¹ yr⁻¹: (Alm et al., 2007; Maljanen et al., 2003; Von Arnold et al., 2005; Yamulki et al., 2013). However, the emissions were smaller and with a lower variability between years (year to year) compared to the earlier period measured by Ernfors et al. (2011), where the annual emissions were between 5.4 and 11.2 kg N₂O ha⁻¹ in the period 2006 to 2009 for exactly the same plots. Between years, the variation in emissions can be
explained by summer months which in the earlier studies were found to be both drier and warmer compared to the years of the present study. This is also supported by the relatively higher N$_2$O fluxes in the latest study year (2013) which experienced warmer and drier summer and compared to the preceding two years. Drawdown of ground water table as a consequence of drought increases the thickness of aerobic surface peat layer (Alm et al., 1999) which can undergo faster decomposition and subsequent mineralization leading to increase availability of mineral N. This could be the cause of higher emission at the organic site during the warmer years as we can see in the paper II that increase availability of mineral N resulted in higher N$_2$O emission. However, the high variability in N$_2$O emissions between years underpins the need to compare sites when conditions are similar, since even for the same site the annual emissions vary.

The annual N$_2$O emissions from the mineral site ranged between 0.6 to 0.7 kg N$_2$O ha$^{-1}$ yr$^{-1}$. To my knowledge, no data on N$_2$O emissions from Umbrisol is available. Nevertheless, the emissions were in the range (0- 1.0 kg N$_2$O ha$^{-1}$ yr$^{-1}$) to what Gundersen et al. (2012) found when summarizing on N$_2$O emissions from temperate forest located on a range of mineral soils including Podzol, Cambisol, Gleysol and Arenosol.

**Factors influencing soil N$_2$O emissions**

The pattern of N$_2$O emission was similar at both sites i.e. increases/decreases of N$_2$O emission occurred simultaneously most of the time (Fig. 4 in paper I). Most of the high flux occasions of N$_2$O were observed in summer between late May and early September at both sites and these high fluxes often occurred following rainfall after a dry period. High N$_2$O fluxes were also observed in March and April at the organic site and these high fluxes were related to thawing of frozen soils when the air temperature raised to above 0°C (Fig. 4 in paper I). In contrast to the organic site the N$_2$O emission from mineral site was not found to increase during thawing.

Within site the SOM was the most important factor influencing the overall size of N$_2$O emissions from forested drained organic and mineral soil. Table 2 gives an overview of the station specific SOM and emissions data. The greatest N$_2$O emission was found from the station having highest SOM content (%) at both sites. This finding was also confirmed by the observed correlation between the N$_2$O emissions and SOM content (%) within both organic ($\rho$=0.99, p<0.01) and mineral site ($\rho$=0.55, p<0.05) (Table 3 in paper I).

Since weather conditions as well as forest age and vegetation were similar, the difference in emission between two sites; organic and mineral site, could be related to their soil properties. The 4-fold higher SOM content (%) of the organic site compared to the mineral site could be taken as an explanation for the higher emissions. However, because of higher bulk density of mineral soils the total SOM amount (kg per volume of soil) in the upper 0.30 m soil was found to be similar (Table 2) to that of the organic sites even though the soil had lower SOM content (%). Thus it could be asked why the mineral site had lower emissions than the organic site. One possible explanations could be that the mineral soil SOM is protected against microbial decomposition by association with minerals establishing physical barriers.
Nitrous oxide emissions from drained Organic and mineral soil: a study on hemi boreal Spruce forests

between SOM and microbes or enzymes (Six et al., 2004), or intermolecular interactions between organic and inorganic substances which decrease the availability of the organic substrate for decomposition (Guggenberger and Kaiser, 2003). On the other hand, in peat little inorganic substances exists (Päivänen, 1995) why this inhibition is less and the SOM in peat is highly decomposable when water-level is lowered (Clymo, 1984) following drainage for forestry. The forests are not fertilized, thus, N-deposition and the mineralization of SOM are the sources of inorganic N delivering to the trees as well as for microbial nitrification and denitrification. Therefore, it can be argued that the lower emissions for our mineral soil might result from lower mineralization of mineral soil SOM. However, the higher tree growth as indicated by its higher above ground biomass at this site (Table 1) does not support low mineralization of SOM at this site.

Table 2: N\textsubscript{2}O fluxes and abiotic factors at the stations of organic (O1, O2, O6) and mineral site (M1, M2, M3)

<table>
<thead>
<tr>
<th>Site</th>
<th>Station</th>
<th>O1</th>
<th>O2</th>
<th>O6</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Organic site</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>N\textsubscript{2}O (kg ha\textsuperscript{-1} yr\textsuperscript{-1})</td>
<td>3.7 (± 0.5)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>6.20 (± 0.7)&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.3 (± 0.9)&lt;sup&gt;b&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>SOM content (%)</td>
<td>66 (± 1)</td>
<td>89 (± 5)</td>
<td>65 (± 6.0)</td>
<td></td>
</tr>
<tr>
<td>SOM amount (Kg/m\textsuperscript{2})</td>
<td>40 (± 3.0)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>60 (± 3.0)</td>
<td>45 (± 5.0)&lt;sup&gt;b&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>C/N ratio</td>
<td>28 (± 4.0)</td>
<td>24 (± 1.0)</td>
<td>25 (± 0.3)</td>
<td></td>
</tr>
<tr>
<td>pH</td>
<td>3.9 (± 0.1)&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.2 (± 0.0)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>3.7 (± 0.1)&lt;sup&gt;a&lt;/sup&gt;</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th><strong>Mineral site</strong></th>
<th></th>
<th>M1</th>
<th>M2</th>
<th>M3</th>
</tr>
</thead>
<tbody>
<tr>
<td>N\textsubscript{2}O (kg ha\textsuperscript{-1} yr\textsuperscript{-1})</td>
<td>1.6 (± 0.0)&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.3 (± 0.0)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.3 (± 0.0)&lt;sup&gt;a&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>SOM content (%)</td>
<td>22 (± 3)&lt;sup&gt;a&lt;/sup&gt;</td>
<td>18 (± 2)&lt;sup&gt;a&lt;/sup&gt;</td>
<td>10 (± 6.0)&lt;sup&gt;a&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>SOM amount (Kg/m\textsuperscript{2})</td>
<td>75 (± 9.0)&lt;sup&gt;a&lt;/sup&gt;</td>
<td>60 (± 6.0)&lt;sup&gt;a&lt;/sup&gt;</td>
<td>35 (± 2.0)&lt;sup&gt;a&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>C/N ratio</td>
<td>16 (± 0.5)</td>
<td>19 (± 0.4)</td>
<td>20 (± 0.3)</td>
<td></td>
</tr>
<tr>
<td>pH</td>
<td>3.8 (± 0.1)</td>
<td>3.8 (± 0.0)</td>
<td>3.8 (± 0.0)</td>
<td></td>
</tr>
</tbody>
</table>

**Note:** Parameters that were significantly different between stations within each site are followed by different letters within a row of the site.

Numbers in parentheses are the standard errors of the mean

The availability of mineral N in forest ecosystems were found to be controlled by the competition between plants and soil microorganisms and it is recognized that plants can successfully compete with microorganisms for N (Schimel and Bennett, 2004). Plants are growing more vigorously at the mineral site, as seen in its higher above ground biomass.
(300±1 ton dry weight ha\(^{-1}\)) compared to the organic sites (180±0.9 ton dry weight ha\(^{-1}\)), despite the age of the forests in two sub sites are similar. Thus, it is likely that the forest on the mineral site had a larger N uptake compared to the organic site. As for the above ground biomass, root associated ectomycorrhizal mycelia growth (43.3 and 7.5 g/m\(^2\) at the mineral and organic site, respectively, measured in 2009 to 2010) was also found to be higher at the mineral site. The nutrient absorbing surface area for a plant can be increased (Chapman et al., 2006) by increasing the production of its roots associated mycorrhizal hyphal surface area (Simard et al., 2002) which even can lead to a decreased N availability in soil (Högberg et al., 2006). Thus, the higher root associated ECM production at the mineral site probably resulted in lower N availability in soil and thereby lower \(\text{N}_2\text{O}\) emissions. This suggestion is supported by the findings of the trenching experiment of paper II where \(\text{N}_2\text{O}\) emissions were found to be higher in the plots where roots and ECM were excluded. Also, He et al. (2016a) showed that plants uptake of N has significant influence on soil N availability and thereby on \(\text{N}_2\text{O}\) emission in a recent study investigating the factors affecting \(\text{N}_2\text{O}\) emission at our organic site using a process-based model, CoupModel.

Over the entire 6 years period in paper II, the mean \(\text{N}_2\text{O}\) emission rate, was higher by 62\% in the exclIR treatment (mean: 9.5 kg \(\text{N}_2\text{O}\) ha\(^{-1}\) yr\(^{-1}\)) compared to the control (mean: 5.9 kg \(\text{N}_2\text{O}\) ha\(^{-1}\) yr\(^{-1}\)) and the exclRM treatment showed almost tripled (mean: 17.1 kg \(\text{N}_2\text{O}\) ha\(^{-1}\) yr\(^{-1}\)) \(\text{N}_2\text{O}\) emissions compared to control. However, the rate of the gross mineralization was higher in control and the value was almost triple of that found in the exclRM treatment (4.49 and 1.34 µg N g\(^{-1}\) d\(^{-1}\) for the control and exclRM plot, respectively (Table 3). The gross mineralization rate did not differ between control and exclIR treatment. Thus this finding indicates that the mineralization itself is not controlling the \(\text{N}_2\text{O}\) emissions at our organic site. The gross mineralization rate found in control chambers was similar to the gross mineralization rates, 3.5 to 5.9 µg N g\(^{-1}\) d\(^{-1}\) observed for organic soils forested with birch and poplar (Münchmeyer, 2001; Willison et al., 1998).

As stated above, the SOM mineralization is the main source for mineralized N at the organic site. Consequently, one obvious mechanism by which \(\text{N}_2\text{O}\) emission was decreased despite higher gross mineralization in control plots could be plant uptake of mineralized N leading to decreased N availability for the microbes to produce \(\text{N}_2\text{O}\). Also, the microbial \(\text{NH}_4^+\) immobilization (\(I_{\text{NH}4}\)) rate was highest at the control and after trenching the rate was decreased by 20\% and 64\% in the exclIR and exclRM treatment, respectively (Table 3). The decreased microbial \(I_{\text{NH}4}\) after trenching probably resulted from the declined microbial activity because of reduced input of labile C via roots (Kaiser et al., 2011). Thus, the combined N uptake by plants and microbes reduced concentration of mineral N as the substrate for \(\text{N}_2\text{O}\) emission in the control plots.

Besides immobilization of \(\text{NH}_4^+\) (\(I_{\text{NH}4}\)). Oxidation of \(\text{NH}_4^+\) to \(\text{NO}_3^-\) by nitrifier (autotrophic nitrification notes as \(O_{\text{NH}4}\)) is another the process that competes for ammonium produced from mineralization. The relative dominance of the processes competing for \(\text{NH}_4^+\), immobilization (\(I_{\text{NH}4}\)) and autotrophic nitrification (\(O_{\text{NH}4}\)), can be expressed in the ratio N/I. Using all treatments, including control and trenched, strong correlation between \(\text{N}_2\text{O}\)
Nitrous oxide emissions from drained Organic and mineral soil: a study on hemi boreal Spruce forests

emissions (averages over 6 years) and the N/I ratio was found (Fig. 5 in paper II). We cannot
tell in which process the N$_2$O was produced, where nitrification is one suggestion. However,
NO$_3^-$ produced by nitrification is needed for denitrification which could be the main pathway
of N$_2$O production in our investigated organic site (Björk et al., 2010; He et al., 2016a), the
N/I ratio could then be a proxy for predicting N$_2$O emissions.

Table 3: Gross N transformation rates (mean ± standard deviation) for an organic forest soil in south-
western Sweden, for control soil and soil with exclusion of roots (ExclR) or exclusion of roots and
ectomycorrhiza (ExclRM).

<table>
<thead>
<tr>
<th>N transformation</th>
<th>Kinetic</th>
<th>Transformation rate (µg N g$^{-1}$ d$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Control</td>
</tr>
<tr>
<td>$M_{SON}$ Mineralization of SON</td>
<td>0</td>
<td>4.49 (0.55)$^a$</td>
</tr>
<tr>
<td>$I_{NH4}$ Immobilization of NH$_4^+$</td>
<td>1</td>
<td>2.21 (0.44)$^a$</td>
</tr>
<tr>
<td>$O_{SON}$ Oxidation of SON to NO$_3^-$</td>
<td>0</td>
<td>5.67 (0.54)$^a$</td>
</tr>
<tr>
<td>$I_{NO3}$ Immobilization of NO$_3^-$</td>
<td>1</td>
<td>0.95 (0.29) n.d.</td>
</tr>
<tr>
<td>$O_{NH4}$ Oxidation of NH$_4^+$ to NO$_3^-$</td>
<td>1</td>
<td>1.68 (0.36)$^a$</td>
</tr>
<tr>
<td>$D_{NO3}$ Dissimilatory NO$_3^-$ reduction to NH$_4^+$</td>
<td>1</td>
<td>1.42 (0.18)$^b$</td>
</tr>
<tr>
<td>$L_{NH4}$ Losses of NH$_4^+$</td>
<td>1</td>
<td>1.34 (0.30)$^b$</td>
</tr>
<tr>
<td>$L_{NO3}$ Losses of NO$_3^-$</td>
<td>1</td>
<td>4.48 (0.39)$^a$</td>
</tr>
</tbody>
</table>

Note: Gross rates that were significant different between treatments are followed by different
letters within a row. Kinetics were either zero order or first order; n.d. = not detected.

Similar to gross mineralization rate, the total gross nitrification was highest in the control plot
which was followed by the exclIR and exclRM treatment. These findings are consistent with
the observed positive relation between these two processes by Booth et al. (2005). However,
this result is different from the findings of Ross et al. (2001) and Kaiser et al. (2011), who
found increased gross nitrification despite decreased gross mineralization after trenching
(Ross et al., 2001) or tree girdling (Kaiser et al., 2011). In controls we observed a total gross
nitrification, production of NO$_3^-$ from soil organic N ($O_{SON}$) and NH$_4^+$ pool ($O_{NH4}$), rate of
6.3–7.4 µg N g$^{-1}$ d$^{-1}$ at Skogaryd (paper II) that were similar or within the range of values, 0–
10 µg N g$^{-1}$ d$^{-1}$, reported in the literature for temperate forests (Booth et al., 2005).
Heterotrophic nitrification ($O_{SON}$) had the largest contribution, 77% of the total nitrification.
Such dominance of $O_{SON}$ for NO$_3^-$ production in forest soils has also been demonstrated by
others (Hart et al., 1997; Rütting et al., 2008; Staelens et al., 2012; Zhu et al., 2011). Even
though the total nitrification rate was lower in exclR treatment, the rate of autotrophic nitrification was found to be higher by 41% compared to the control (Table 3). Thus, the relative importance of autotrophic nitrification to the total nitrification was increased in this in exclR treatment. The increase in autotrophic nitrification rate in the exclR treatment was probably dependent to some extent on the decrease production of root derived (or released) nitrification inhibitors (Paavolainen et al., 1998; White, 1986). It could also be that in absence roots competition, more NH$_4^+$ is available for microbes for autotrophic nitrification. However, the rate of O$_{NH4}$ did not differed significantly between the exclRM and control treatment.

Nitrate immobilization (I$_{NO3}$) was only detectable in the control plots but not in the trenched treatments. Nitrate immobilization generally increases with increasing C availability (Booth et al., 2005; Hart et al., 1994) due to increased microbial activity and, hence, increased microbial N demand. The absence of I$_{NO3}$ in the trenched plots (Table 3) thus, indicates reduced C availability.

Overall the findings of the study in paper II indicate roots and ECM have large influence on soil N turnover rates and that gross rates of N transformation, gross mineralization and nitrification in forested drained organic soil is enhanced in presence of plant roots and ECM. These enhanced rates of gross mineralization are however balanced by the high rates of N immobilization in presence of roots and ECM resulting in relatively low N availability for nitrification and denitrification. Hence soil N$_2$O emissions decreased in presence of plants roots and ECM.
Conclusions

From the studies presented in this thesis I conclude that,

- Nitrous oxide emission differs between forested drained Histosol and Umbrisol soil. Drained Histosol is a stronger source for N$_2$O emissions than Umbrisol.
- Plant roots and their mycorrhizal symbionts influence N transformation rates in forested soil. Gross mineralization–ammonium (NH$_4^+$) immobilization turnover increase in presence of roots and mycorrhizae probably due to high inputs of labile C stimulating microbial activity.
- The N$_2$O emission is positively related to the ratio of NH$_4^+$ oxidation (autotrophic nitrification) to NH$_4^+$ immobilization in forest soils.
- Plants uptake of mineral N as well as the stimulated microbial NH$_4^+$ immobilization in presence of roots and ECM reduce the importance of nitrification for NH$_4^+$ consumption and thereby decrease N$_2$O emissions from forested soils.
Acknowledgement

First of all, I am grateful to the Almighty Allah (su) who gives His blessings not only to accomplish this research work successfully but for all my life. He is the one who made me capable to complete this study.

I would like to thank to my supervisor Åsa Kasimir for her guidance and all advice concerning academic thinking, writing and research throughout this study. I want to thank Prof. Leif Klemedtsson for his valuable advice and suggestions during my study. Thanks also to my examiner Prof. Roland Barthel for his valuable suggestions in completing this thesis. I am grateful to Tobias Rütting for all the positive criticisms and insightful comments and for all helps to complete the studies. I am also grateful to Christine Achberger for her encouragement in the beginning of my research work at GEO.

I would like express a large thanks to Mark Johnson, the Head of the department, for his valuable discussions and for his help when needed.

I want to thank Per Weslien for sharing your great experiences and knowledge in solving difficulties in wide range of problems, and Robert Björk for nice and very helpful discussion about mycorrhizae. I am grateful to David Allbrand who has been always with me out in the field, and Aila Schachinger who help me in lab works.

It has been pleasure for me to meet all the people at GEO and to have a chat with them now and then. I want to thank Prof. Rodney Stevens for your generosity and for your wonderfully supportive attitude. Thanks to Fredrik Lindberg and Alexander Walther. It has been pleasure for me to work with you in GIS teaching. Special thanks to Anna-Karin, Tage Vowles, Yumei Hu and Tinghai Ou for sharing family life. I would like to thank Tariqul Islam and Hongxing He for their company during coffee break. Thanks also to Ewa Steen Stackelius for patiently organizing many administrative things in order for me during this study.

BECC (Biodiversity and Ecosystem services in a Changing Climate) is gratefully acknowledged for financing support.

I would like to thank to my parents who live far away from me but they always motivated and fervently encouraged me to complete my work. My warmest and deepest thank to my son Ayaan Nazef. After a long and hard day’s work or sometimes when everything feels difficult and suffocating, there is nothing better than come to home and see your smiling face. Finally, thanks to my wife, Dr. Shamima Nazmin Akhi, for all her never-ending love and encouragement and being my friend all the way through.
References


Nitrous oxide emissions from drained Organic and mineral soil: a study on hemi boreal Spruce forests


27


