

# The effect of geothermal soil warming on the production of carbon dioxide (CO<sub>2</sub>), methane (CH<sub>4</sub>), nitrous oxide (N<sub>2</sub>O), nitric oxide (NO) and nitrous acid (HONO) from forest soil in southern Iceland

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## ABSTRACT

Geothermal areas can be local sources of greenhouse gases, both directly from the geothermal system or because of soil warming effects on biological sources. In this study we repeated field measurements methane (CH<sub>4</sub>) and nitrous oxide (N<sub>2</sub>O) fluxes along the soil temperature (T<sub>s</sub>) gradient in a Sitka spruce (*Picea sitchensis*) stand at the ForHot study site in southern Iceland, where geothermal soil warming had started eight years earlier. We complemented these results with *in situ* measurements of carbon dioxide (CO<sub>2</sub>) and topsoil sampled in the same plots to study the production rates of those gases at 20 °C in the laboratory, as well as nitric oxide (NO) and nitrous acid (HONO). We showed that the eight year long exposure to elevated T<sub>s</sub> had changed the topsoil, including its microbial properties and the production potentials of these gases. However, the production rates of CO<sub>2</sub>, CH<sub>4</sub> and N<sub>2</sub>O measured in laboratory conditions did not clearly follow the *in situ* fluxes. We discuss both adaptation of microbes and origin of greenhouse gases (depth patterns and microbial vs. geothermal sources) as possible reasons for these discrepancies.

Keywords: geothermal soil warming; *Picea sitchensis*; temperature sensitivity; volcanic soil

## YFIRLIT

Áhrif hækkaðs jarðvegshita á myndun koldíoxíðs (CO<sub>2</sub>), metans (CH<sub>4</sub>), hláturgass (N<sub>2</sub>O), nituroxíðs (NO) og nitraðrar síru (HONO) í skógarjarðvegi á Suðurlandi.

Jarðhitasvæði geta verið uppsprettur ýmissa gróðurhúsalofttegunda, annað hvort beint upp úr jarðhitakerfinu eða vegna áhrifa aukins jarðvegshita á ýmsa lífræna ferla. Í þessari rannsókn endurtókum við mælingar á flæði metans (CH<sub>4</sub>) og hláturgass (N<sub>2</sub>O) með auknum jarðvegshita (T<sub>s</sub>) í foldu í sitkagreniskógi (*Picea sitchensis*) á ForHot rannsóknasvæðinu á Suðurlandi, þar sem jarðhitasvæði hafði fæst undir átta árum áður. Við bættum einnig við mælingum á losun koldíoxíðs (CO<sub>2</sub>) í foldu og bárum -niðurstöðurnar saman við losun þessara sömu gastegunda og nituroxíðs (NO) og nitraðrar síru (HONO) úr jarðvegskjörnum úr sömu reitum sem mældir voru við 20 °C á rannsóknastofu. Niðurstöðurnar sýndu að átta ára jarðvegshlýnun hafði bæði breytt efnasamsetningu og örveruflóru reitanna og þar með getu til að framleiða áður nefndar lofttegundir. Hinsvegar breyttist framleiðslugeta CO<sub>2</sub>, CH<sub>4</sub> og N<sub>2</sub>O við 20 °C ekki reglulega með auknum T<sub>s</sub> í foldu. Við ræðum bæði aðlögun örvera að auknum hita og hvernig uppruni gróðurhúsalofttegunda (úr mismunandi dýpi í jarðvegi og hvort hann er lífræðilegur eða jarðfræðilegur) getur mögulega útskýrt þær niðurstöður sem við fengum.

## INTRODUCTION

Carbon dioxide (CO<sub>2</sub>), methane (CH<sub>4</sub>) and nitrous oxide (N<sub>2</sub>O) are important greenhouse gases. The concentration of these gases in the atmosphere has increased since industrialization began due to enhanced anthropogenic sources; however, there are also important natural sources for these gases, such as soils. Carbon dioxide is produced in soils during respiration by plants, soil animals and soil microorganisms. Soil temperature and moisture are the most important regulators of soil respiration (Chapin et al. 2002). Methane is formed in soils by anaerobic methanogenesis (Le Mer & Roger 2001) and the production of CH<sub>4</sub> is primarily controlled by oxygen content, but is additionally controlled by soil temperature, pH, vegetation, moisture and salinity (Le Mer & Roger 2001). Nitrous oxide is mainly produced by microbial aerobic nitrification and anaerobic denitrification in soils (Priemé & Christensen 2001, Smith 2017) and these processes are regulated by several factors such as temperature, moisture, pH and N-availability (Smith 2017).

Soils can be sources also for some other reactive gases like nitric oxide (NO) and nitrous acid (HONO), and the emissions of these gases can also be affected by soil temperature (Pilegaard 2013). Nitric oxide is not a greenhouse gas, but it reacts to form smog and acid rain and it is also important to the formation of tropospheric ozone (Heil et al. 2016). Nitrous acid is also a reactive gas in the atmosphere and has been linked to the production of N<sub>2</sub>O and NO (Su et al. 2011, Maljanen et al. 2013). Nitrous acid is not a greenhouse gas; instead, it contributes to the formation of hydroxyl radicals ( $\cdot$ OH), which are strong oxidizing molecules and can oxidize atmospheric CH<sub>4</sub> (Riedel & Lassey 2008). The soil-related sources and formation pathways of HONO are not yet well known. Photosensitized reduction of nitrogen dioxide (NO<sub>2</sub>) on aerosol surfaces containing humic acid is considered an important source of HONO (Stemmler et al. 2006) but it is also produced in soils (Su et al. 2011, Oswald et al. 2013, Maljanen et al. 2013, Scharko et al. 2015, Bhattarai et al. 2018).

Temperature affects the production and

consumption of the greenhouse gases (CH<sub>4</sub>, N<sub>2</sub>O and CO<sub>2</sub>) and also NO. Thus, greenhouse gas fluxes from ecosystems are expected to change with the warmer temperatures resulting from climate change. The effects of increasing temperature can be studied using manipulation experiments with artificial warming or by using natural temperature gradients. We measured earlier (in 2012-2014) the emissions of N<sub>2</sub>O and CH<sub>4</sub> from natural geothermal temperature gradients in southern Iceland and found significant differences in the flux rates with changing soil temperature from normal ambient temperatures (from 2 to 14°C) up to an elevation of +40 °C (Maljanen et al. 2017). Here, we repeated the field measurements in 2016 and complemented them with laboratory studies on the production rates of the three greenhouse gases (CO<sub>2</sub>, CH<sub>4</sub> and N<sub>2</sub>O) and also with analysis of HONO and NO production rates in 2016. We hypothesize that volcanic soils can also produce HONO and NO and the exposure to high soil temperatures might have an effect on their emission rates. We also hypothesize that, similar to the trace gas emissions for CH<sub>4</sub> and N<sub>2</sub>O as published by Maljanen et al. (2017), CO<sub>2</sub> emissions are temperature sensitive, but that soil microbes have adapted to the exposure to elevated temperatures (Poeplau et al. 2017). Those adaptations will be reflected in the CO<sub>2</sub>, N<sub>2</sub>O, and CH<sub>4</sub> production rates in laboratory incubations at room temperature. In addition to biological formation of the greenhouse gases, the role of geothermal outgassing of CO<sub>2</sub>, CH<sub>4</sub> and N<sub>2</sub>O through the soil, especially in the warmest plots, is discussed.

## METHODS

### *Soil sampling and analysis*

Soil samples were collected in July 2016 from the ForHot experimental site at a natural geothermal temperature gradient within a 50-year-old Sitka spruce stand [*Picea sitchensis* (Bong.) Carr.] at Reykir in southern Iceland. The soil type is Silandic Andosol (also termed Brown Andosol; Arnalds 2015), soil texture silty loam with a relatively high pH (5.5 - 7.0) and large soil water retention capacity (O’Gorman

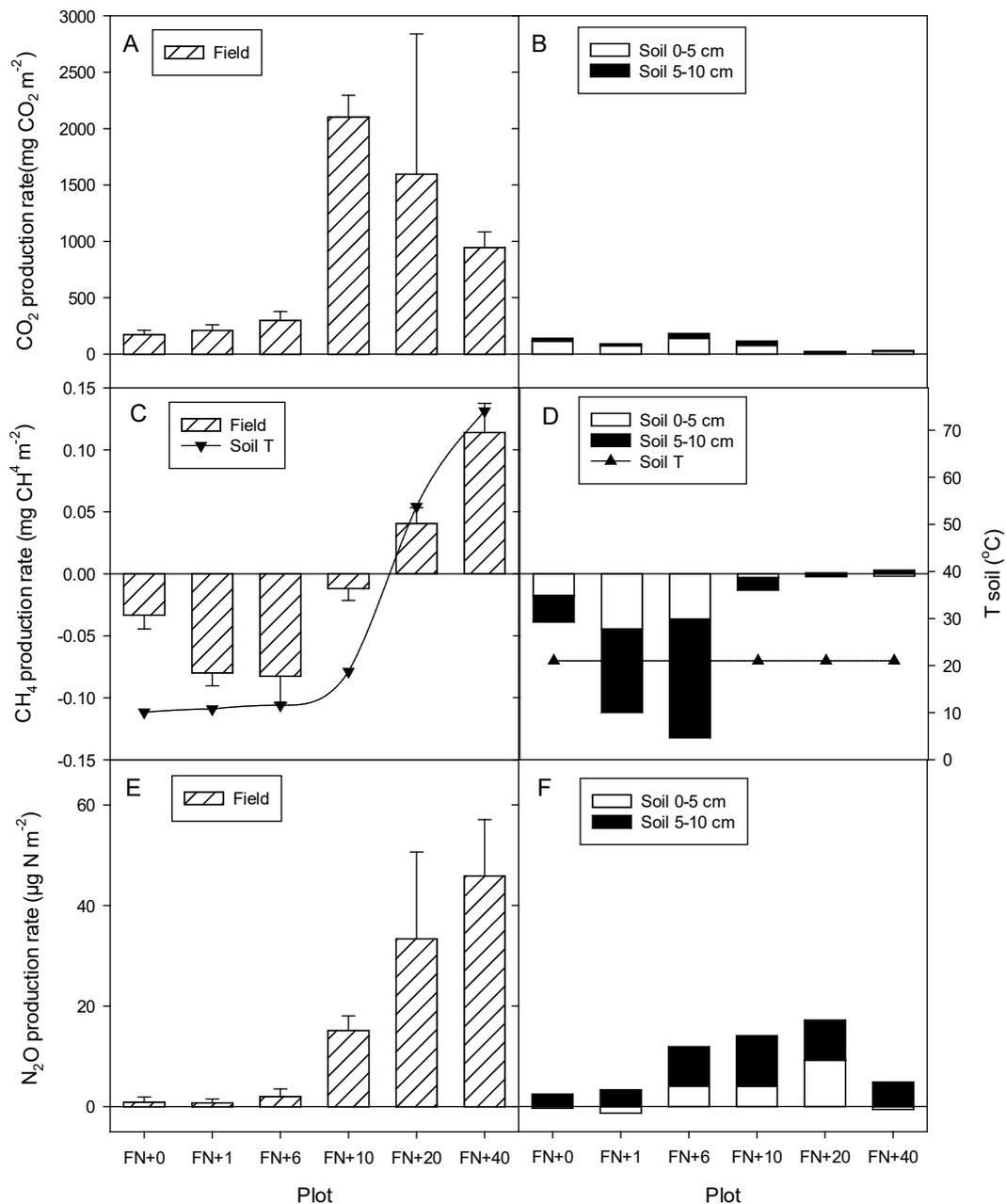
et al. 2014). More details about the ForHot experiment can be found in Sigurdsson et al. (2016), Maljanen et al. (2017), Gargallo-Garriga et al. (2017) and Leblans et al. (2017). This temperature gradient was formed in May 2008 after a major earthquake started geothermal bedrock warming under an originally unheated site (Sigurdsson et al. 2016), and hence the soils within the affected area had only been warm for eight years when the study took place. The sampling plots were located both outside the affected area (on unwarmed soil temperature;  $T_s$ ) and within it at a different elevated  $T_s'$ , with actual temperatures reaching up to 75°C. The plots were named according to the warming levels measured in 2012 with site code FN and temperature elevation as +X °C, similarly as in Maljanen et al. (2017).

The actual measured temperatures during sampling in July 2016 are shown in Figs. 1, 2 and Table 1. The thin litter layer was removed before sampling. The soil surface in the dense Sitka spruce plantation was mainly bare without any green vegetation except on plot FN+10,

where grasses were covering 20% of the surface after some tree mortality had opened up the stand, and on FN+20 (100% moss cover), where all Sitka spruce trees had been killed due to high soil temperatures (O’Gorman et al. 2014, Sigurdsson et al. 2016). Samples were collected from two depths, 0-5 cm and 5-10 cm, by cutting a square hole (10 x 10 cm) with a knife. Four subsamples were pooled and sieved (4 mm) and within two days samples were transported to Finland for analysis. Soil pH and electric conductivity (EC) were measured from a sample:water slurry (30:50 v/v). Total carbon (C) and nitrogen (N) were analyzed with an elemental analyzer (Thermo Finnigan, Germany, Flash EA 1112). For analysis of nitrate ( $\text{NO}_3^-$ ) and nitrite ( $\text{NO}_2^-$ ) 30 ml soil and 100 ml milliQ- $\text{H}_2\text{O}$  were shaken at 175 rpm for one hour and then filtered and analyzed with an ion chromatograph (DX 120, Dionex Corporation, USA). Gravimetric moisture ( $\text{g}_{\text{H}_2\text{O}} \text{g}_{\text{dry soil}}^{-1}$ ) was determined by drying the samples for 24 h at 105 °C.

**Table 1.** Soil properties in top 0-5 and 5-10 cm of soil at the study plots in July 2016; average values from two replicate analyses are shown except C and N from four replicate analyses. Soil temperatures were measured at the depths of 5 and 10 cm.

Plot	T (°C)	pH (H <sub>2</sub> O)	EC (μS cm <sup>-1</sup> )	GM (%)	C (%)	N (%)	C:N
0-5 cm							
FN+0	10.1	5.9	31	41.9	7.8	0.48	16.3
FN+1	10.7	6.4	20	48.3	14.9	0.62	23.0
FN+6	11.3	6.1	19	52.1	17.3	0.80	21.8
FN+10	18.1	6.0	36	64.4	11.7	0.79	14.7
FN+20	49.5	5.4	155	49.7	6.0	0.49	12.1
FN+40	62.6	5.6	259	37.9	3.8	0.34	11.1
5-10 cm							
FN+0	10.2	6.1	32	42.2	9.0	0.41	21.9
FN+1	10.9	6.1	36	40.9	6.0	0.36	16.8
FN+6	11.7	5.8	34	53.7	7.6	0.52	14.6
FN+10	18.9	5.9	26	64.1	7.3	0.44	16.4
FN+20	54.5	5.8	69	39.5	2.8	0.36	7.7
FN+40	75.0	6.7	66	38.9	1.1	0.15	7.5



**Figure 1.** Measured average field (hatched bars) and laboratory (black and white bars) production rates of CO<sub>2</sub> (A, B), CH<sub>4</sub> (C, D) and N<sub>2</sub>O (E, F). Standard deviation (n=3) shown for field measurements, laboratory incubations were made without replicates due to small amount of samples available. The laboratory production rates of samples from 0-5 cm (white bar) and 5-10 cm (black bar) were measured at +20 °C. The line with triangles down (C) shows the actual soil (10 cm) temperatures in the field and the line with triangles up (D) shows the temperature in the laboratory during gas production measurements.

### *Field gas flux measurements*

At the same time that the soil sampling took place (in July 2016), field measurements of CO<sub>2</sub>, N<sub>2</sub>O and CH<sub>4</sub> emissions were made along the soil temperature gradient using the static chamber method (Maljanen et al. 2017), with three replicate chambers at each gas sampling plots which were covered unwarmed soil and soils warmed up to 75 °C at a depth of 10 cm (Fig. 1, Table 1). The metal flux chambers (ø = 26 cm, h = 30 cm) had a hole in the top for a sampling line and for a capillary line to avoid pressure effect. Prior to sampling, the sharp edge at the bottom of the chamber was twisted 3-5 cm into soil and the top was sealed with a rubber septum. A total of six gas samples (25 ml) were collected with a 60 ml syringe (Terumo) 5, 10, 20, 30, 40 and 60 min after installing the chamber. Within 8 h, the samples were injected into 12 ml Labco pre-evacuated vials (Labco Excetainer®) for gas analysis with a gas chromatograph (Agilent 7890B, Agilent Technologies, USA) at University of Eastern Finland (UEF) (Maljanen et al. 2017). Soil temperatures were recorded manually at each sampling location next to the chambers at depths from 5, 10, and 20 cm. Field measurements of NO and HONO were not applicable and these gases were only studied in the laboratory.

### *Soil gas concentrations*

Soil CO<sub>2</sub>, CH<sub>4</sub> and N<sub>2</sub>O concentrations were measured at the sampling points simultaneously with the gas flux measurements. Therefore, gas samples of 20 ml were taken into a 60-ml syringe with a stainless steel sampling probe (ø = 3 mm, l = 50 cm) at four soil depths: 10, 20, 30 and 40 cm. Samples were treated and analyzed at UEF as described above.

### *Laboratory gas flux measurements*

The gas flux measurements in the laboratory at UEF were made at room temperature (20°C). Soil samples from each sampling point and depth (no replicates due to the small amount of sample which we were allowed to take from this unique experimental site) were placed in a PVC cylinder (ø 10 cm) to simulate field conditions

and the bottom of the cylinder was sealed with aluminum foil. Nitrous oxide, CH<sub>4</sub> and CO<sub>2</sub> fluxes were measured with a static chamber system and samples were taken with syringes from the headspace of the chamber at intervals of 5, 10, 15 and 20 min after closing the PVC chamber (vol. 1.0 l) fitted on the PVC cylinder. Gas samples were treated and were analyzed at UEF with a gas chromatograph as described above. For the HONO and NO emission measurements the cylinders were placed in a Teflon chamber (volume 3.2 l, details in Bhattarai et al. 2018) and the gas fluxes were measured by the dynamic flow chamber method. A commercial HONO analyzer (LOPAP, QUMA Elektronik & Analytik GmbH, Germany; detailed description of the principle of the device in Heland et al. 2001) was used for HONO and a Thermo 42i NOx analyzer (Thermo Fisher Scientific, USA) was used for NO. The flow rate in the HONO measurements was 4 l min<sup>-1</sup> and for NO 0.5 l min<sup>-1</sup>.

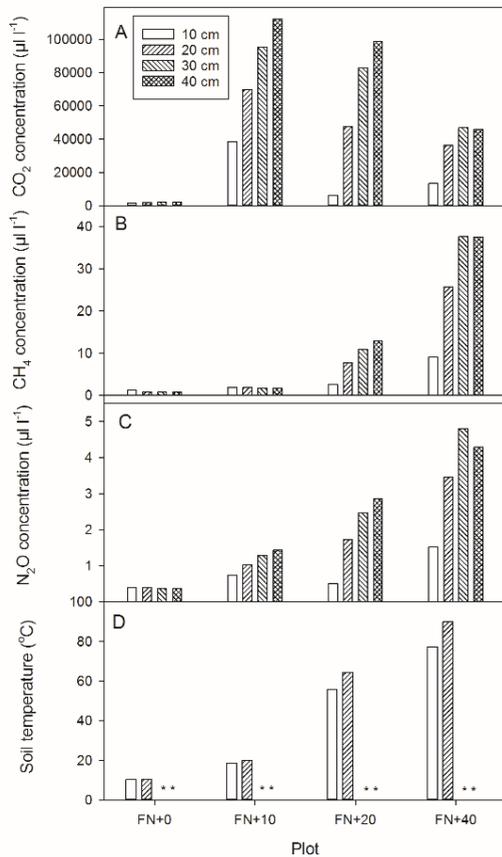
### *Statistics*

The correlations between field gas fluxes, gas production rates and soil parameters were tested using a non-parametric Spearman correlation since Levene's test showed that flux data was not normally distributed (SPSS 21 statistical program, IBM Inc., NY, USA).

## RESULTS

### *Soil properties*

The temperature range in the soils sampled in 2016 was from 10 (unwarmed ambient temperature) to 75 °C at 10 cm depth (Table 1). Soil pH varied between 5.4. and 6.7 and there was no clear correlation with temperature; however, the highest pH was measured from the warmest FN+40 plot at a depth of 5-10 cm. Soil electrical conductivity increased with increasing temperature. Soil gravimetric moisture varied between 38 and 64% and was highest in plot FN+10 and lowest in FN+40. Carbon and N concentrations were not well correlated with temperature; however, the lowest values were measured from the warmest plot FN+40 (Table 1).



**Figure 2.** Concentrations of CO<sub>2</sub> (A), CH<sub>4</sub> (B), and N<sub>2</sub>O (C) in soil at depths of 10, 20 30 and 40 cm. Soil temperature at depths of 10 and 20 cm shown in the bottom (D). \* ND, Soil temperature was not measured in the deeper layers.

#### Carbon dioxide fluxes and soil concentrations

CO<sub>2</sub> emissions from the three coolest plots (FN+0, FN+1 and FN+6) were quite similar when measured in the field and in the laboratory at 20 °C (Fig. 1). In the field, the highest CO<sub>2</sub> emissions were found at FN+10, and were much higher in the field than was found in the laboratory at similar temperatures. The CO<sub>2</sub> emissions from FN+20 and FN+40 were also much higher than those measured at constant temperature in the laboratory (Fig. 1). The lowest CO<sub>2</sub> production rates (at 20 °C) in the laboratory were measured in soils from the warmest plots.

The soil CO<sub>2</sub> concentrations were the lowest in the coolest plot and increased with depth (from 1,400 to 2,200 μl l<sup>-1</sup>), whereas the highest CO<sub>2</sub> concentrations were also measured from plot FN+10, up to 112,000 μl l<sup>-1</sup> at depth of 40 cm (Fig. 2).

#### Methane fluxes and soil concentrations

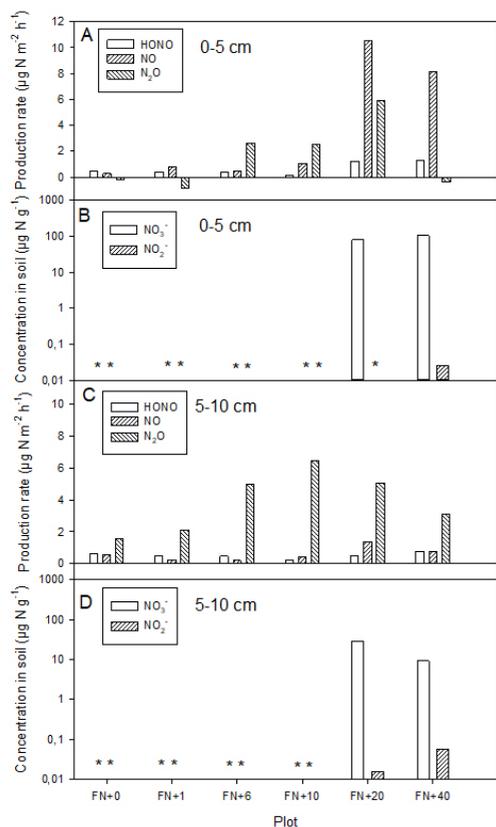
There was clear CH<sub>4</sub> uptake by the coolest soils, both in the field and in the laboratory measurements, and the uptake rates were rather similar between field and laboratory measurements, especially for plot FN+10 where field temperature was close to laboratory temperature (Fig. 1). However, the warmest soils had small CH<sub>4</sub> emissions in the field and they did not produce (or consume) any CH<sub>4</sub> in the laboratory experiment. Soil concentrations of CH<sub>4</sub> ran between 0.7 to 1.3 μl l<sup>-1</sup> in the coolest FN+0 plot, decreasing with depth, but in the warmer plots concentrations increased with depth up to 38 μl l<sup>-1</sup> (Fig. 2)

#### Nitrous oxide fluxes and soil concentrations

Nitrous oxide emissions measured from the field increased with increasing soil temperature ( $p < 0.001$ ,  $r = 0.985$ ) (Fig. 1). In the laboratory experiment there was no linear correlation between N<sub>2</sub>O production rates and the original field soil temperature. The highest N<sub>2</sub>O production rate was measured from plot FN+20 (depth 0-5 cm) and from plot FN+10 (depth 5-10 cm). Nitrous oxide concentrations (from 0.38 to 0.39 μl l<sup>-1</sup>) in the coolest plot (FN+0) were slightly higher than the ambient (0.33 μl l<sup>-1</sup>) concentration and did not differ between depths (10-40 cm). However, in the warmest plots, N<sub>2</sub>O concentrations were higher than ambient and increased with depth up to 4.3 μl l<sup>-1</sup> (Fig. 2). The concentrations of NO<sub>3</sub><sup>-</sup> and NO<sub>2</sub><sup>-</sup> in soil were below the detection limit (0.01 μg N g<sup>-1</sup>) in all other plots except in the warmest plots FN+20 and FN+40 (Fig. 3).

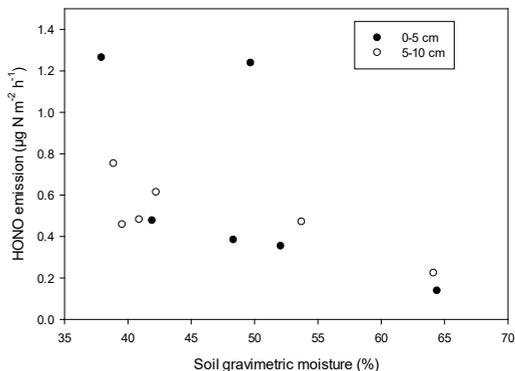
#### HONO and NO production rates in the laboratory

In addition to N<sub>2</sub>O, the soils all produced NO and HONO, which were measured only in the



**Figure 3.** Production rates of HONO, NO and  $N_2O$  measured at 20 °C in the laboratory from soil samples collected from depths 0-5 cm (A) and 5-10 cm (C) shown together with soil  $NO_3^-$  and  $NO_2^-$  concentrations (B, D). \* = concentration below detection limit ( $0.01 \mu\text{g N g}^{-1}$ ). Production rate measurement were made from a single sample without replicates due to small amount of sample. Note the logarithmic y-axis for  $NO_3^-$  concentration.

laboratory (Fig. 3). The production rates of NO and HONO were higher in the 0-5 cm samples, especially in the warmest plots, whereas the  $N_2O$  production rate was higher in the 5-10 cm samples. The NO and HONO emissions were associated with the availability of  $NO_3^-/NO_2^-$  in the soil, which were barely above the detection limit ( $0.01 \mu\text{g N g}^{-1}$ ) in the samples from the two warmest plots (Fig. 3). The highest NO emissions rates were measured from the soils from the warmest plots, but HONO emissions



**Figure 4.** HONO emissions from soil samples from depths 0-5 (black circles) and 5-10 cm (open circles) plotted with gravimetric soil moisture.

were less variable. HONO emissions correlated negatively with soil moisture (depth 0-5 cm  $p = 0.042$ ,  $r = -0.829$ ; depth 5-10 cm  $p = 0.005$ ,  $r = -0.943$ ). The HONO and NO production rates did not correlate with any other measured parameters (Fig. 4).

## DISCUSSION

### Field $CO_2$ , $CH_4$ and $N_2O$ fluxes

The  $CO_2$  emissions in the field were lowest at the low temperatures and very high from the three warmest plots. Field soil  $CO_2$  emissions seemed to peak on plot FN+10 where soil temperature at a depth of 10 cm was about 20 °C. However, the field  $CO_2$  emissions decreased when the temperature reached 40 to 70 °C. This can be explained by heat stress at higher temperatures, where soil microbes can die or operate at suboptimal conditions for mesophiles. Such temperature optimum for soil respiration, however, is not commonly found in nature or in warming experiments (Chapin et al. 2002, Lu et al. 2012) with less warming. Additionally, the respiration by the ground vegetation may have enhanced  $CO_2$  fluxes in plot FN+10, since this was the only plot with a significant amount of living grasses on the surface. There could also have been a change in microbial community structure from mesophiles to thermophiles. Crowther & Bradford (2013) showed that warm-acclimated microbes had lower growth and respiration rates at intermediate

temperatures than cold-acclimated isolates. The third potential reason for reduced  $\text{CO}_2$  flux rates at the highest temperatures would be a resource limitation for the soil microbes due to loss of soil organic matter after eight years of soil warming, similar to what has been found in a nearby grassland that was also warmed from 2008 (Poeplau et al. 2017). This last hypothesis was partly confirmed by our finding that soil carbon concentrations decreased at the highest soil temperatures (Table 1) and with the laboratory measurements with soils sampled from plots with topsoil temperature above 20 °C, since there also the lowest soil respiration rates were found at higher initial temperatures (Fig. 3).

Nitrous oxide emissions increased with increasing soil temperature, whereas  $\text{CH}_4$  was showing negative flux (uptake) which increased with moderate warming (up to +5 °C), but thereafter net  $\text{CH}_4$  emissions were observed. High emissions of  $\text{N}_2\text{O}$  and  $\text{CH}_4$  from the warmest plots, and thus a significant warming response of the trace gas fluxes over the entire temperature range, were also recorded in an earlier study at the same site (Maljanen et al. 2017). The flux rates at the coolest plots (up to +5 °C) were also similar to those reported for boreal forest sites on mineral soil (Maljanen et al. 2010).

The soils from the coolest plots showed  $\text{CH}_4$  uptake, both in the field and laboratory (see below; Fig 1). The methane uptake rate first increased with a slight increase of *in situ* temperature, but soils from the warmest plots were emitting  $\text{CH}_4$ . This may indicate that the warmer soil conditions could be unfavourable for methane oxidizers because upland forest soils generally have some  $\text{CH}_4$  uptake (LeMer & Roger 2001). The  $\text{CH}_4$  emissions from the field are difficult to explain based on the biological control mechanisms. The soils were drier and most probably more aerobic at higher temperatures (Table 1), and C concentration was also low, and therefore these factors did not support the biological  $\text{CH}_4$  production from organic matter in anaerobic conditions. Thus,  $\text{CH}_4$  produced from abiotic geothermal sources

could potentially contribute to the  $\text{CH}_4$  efflux at the ForHot sites. Such processes are known from deep geological sources where abiotic  $\text{CH}_4$  can be emitted (Etiope & Sherwood Lollar 2013, Klusman et al. 2000).

Nitrous oxide emissions increased linearly with temperature under field conditions with the highest rates observed at 75 °C in soil (depth 10 cm). Since the microbial production of  $\text{N}_2\text{O}$  is favoured if there are both aerobic and anaerobic microsites (nitrification in aerobic conditions and denitrification in anaerobic conditions), this is theoretically possible, although thermophilic denitrifiers/nitrifiers would need to be operating at such high temperatures. There is some evidence of such thermophilic nitrifiers can still be active above 70 °C (de la Torre et al. 2008). We detected also high nitrate concentrations in the warmest FN+40 plot, which could support the high nitrification rate found there. However, the geothermal sources of  $\text{N}_2\text{O}$  may also play a role, as suggested earlier (Maljanen et al. 2017)

#### *Comparison to earlier measurements*

High emissions of  $\text{N}_2\text{O}$  and  $\text{CH}_4$  were measured from the warmest plots. Nitrous oxide emissions measured from the field during the growing seasons were higher in 2012-2014 (up to 150  $\mu\text{g N}_2\text{O m}^{-2} \text{h}^{-1}$ ) than in our present measurements from 2016 (up to 30  $\mu\text{g N}_2\text{O m}^{-2} \text{h}^{-1}$ ). Methane emissions in 2016 were similarly lower from the warmest plots (up to 0.1  $\text{mg CH}_4 \text{m}^{-2} \text{h}^{-1}$ ) than in 2012-2014 (up to 1.25  $\text{mg CH}_4 \text{m}^{-2} \text{h}^{-1}$ ) (Maljanen et al. 2017). This may be due to changes in the geothermal system between these years because the soil temperature had also significantly increased in the warmest plots from 2012-2014; the maximum temperature recorded at a depth of 10 cm in 2016 had changed from 51 (Maljanen et al. 2017) to 75 °C. Such large fluctuations in  $T_s$  seem to affect mostly the warmest parts of the forest temperature gradients, an area where the warming gradient is very steep (Sigurdsson et al. 2016). Similar irregular shifts in the spatial boundaries within this hottest area (>+20 °C) were also recorded with continuous measurements of  $T_s$  in 2011-2012 (Sigurdsson et al. unpublished data). Comparable unexplained

shifts or pulses have also been repeatedly observed in drill-holes for hot water in a nearby geothermal field (Helgadottir, pers. comm.).

#### *Field versus laboratory measurements of CO<sub>2</sub>, CH<sub>4</sub> and N<sub>2</sub>O fluxes*

As expected, field fluxes did not correlate well with laboratory fluxes measured at a standard temperature of 20 °C. However, the CO<sub>2</sub> emissions also differed when comparing similar field and laboratory temperatures; i.e. the field emissions of CO<sub>2</sub> were several times higher in the plot FN+10, where field soil temperature (19 °C) was close to the temperature in the laboratory (20 °C), where a 10 cm deep soil sample was incubated. This could have been due to disturbance effects caused by sieving and re-packing the soils in the laboratory. However, this could also indicate that CO<sub>2</sub> (as well as CH<sub>4</sub> and N<sub>2</sub>O at the warmest temperatures, see below), were produced at lower soil depths in the field (soil depth was on average ca. 70 cm; Sigurdsson et al. 2016), possibly even from deeper sources where some geothermal CO<sub>2</sub> can outgas from the nearest volcano's (Mt. Hengill) magma chamber. Geothermal CO<sub>2</sub> could be transported through the bedrock with geothermal water and mainly outgassed through the soil where the geothermal soil warming was highest (and where geothermal channels were closest to the surface). Such phenomena are known to occur very close to geothermal fumaroles in the same area as the ForHot experiment is located (Fridriksson et al. 2006) and are also known from other studies on volcanic soils (Rey 2015).

The laboratory incubations also pointed toward some potential non-biological (geothermal) sources of CH<sub>4</sub>, since laboratory production rates did not mirror the emissions measured in the field. The laboratory experiment also showed that the warmest soils did not have the potential to oxidize atmospheric CH<sub>4</sub> at room temperature, most probably due to changes in methanotroph communities.

In the field, N<sub>2</sub>O emissions increased with increasing temperature with the highest rates observed at 75 °C in the soil. However, in the laboratory experiment, where N<sub>2</sub>O emissions

were only measured at +20 °C, the highest production rates were found in soils from 20-40 °C soil temperature (FN+10 and FN+20), which is likely the optimum temperature for N<sub>2</sub>O production (Barnard et al. 2005). Therefore, we cannot totally exclude the possibility of a deeper geothermal N<sub>2</sub>O source at the highest soil temperatures, a phenomenon that has been little studied for nitrogen gases (Klusman et al. 2000).

#### *The soil CO<sub>2</sub>, CH<sub>4</sub> and N<sub>2</sub>O concentrations*

Soil gas concentrations were in line with the flux results from the field experiments, with highest concentrations of N<sub>2</sub>O and CH<sub>4</sub> found at the warmest temperatures, while CO<sub>2</sub> concentrations were highest at FN+10. Methane concentrations increased in the warmer soils with depth, hinting at other than a biological source in this well aerated upland soil (LeMer & Roger 2001). As expected, CH<sub>4</sub> concentrations in the coolest plot decreased, however, with depth, indicating oxidation of atmospheric CH<sub>4</sub> and no significant CH<sub>4</sub> source from the deeper soil layers. This finding also supported the suspected geothermal source of CH<sub>4</sub> at high temperatures, as discussed earlier.

#### *The potential geothermal outgassing*

At present, we can only speculate about a geothermal source of greenhouse gases from the warmest plots of the natural geothermal temperature gradients in southern Iceland. The potential geothermal outgassing of CO<sub>2</sub>, CH<sub>4</sub> and N<sub>2</sub>O in the warmest soils needs to be studied further at the ForHot field sites, e.g. using stable isotope ratios of N and C ( $\delta^{15}\text{N}$  of N<sub>2</sub>O and  $\delta^{13}\text{C}$  of CH<sub>4</sub> and CO<sub>2</sub>). Also, it is of high importance to determine if such geothermal outgassing might be limited only to the warmest part of the existing geothermal temperature gradients. The soil surface gas exchange measurements with analysis of isotopic ratios in the gases could be applied at the gradient for enhancing our understanding of how much geothermal outgassing is affecting the flux rates and how biological processes are affected by the soil temperature alone.

### *HONO and NO emissions*

Little is known about the origins of HONO emissions (Maljanen et al. 2013). The volcanic soils of the Sitka spruce stand had the potential to emit HONO and NO, as was found in the incubation experiment. These NO and HONO emissions were, however, lower than those measured with similar methods from, for example, agricultural soils (Bhattarai et al. 2018, Oswald et al. 2013), but higher than those measured from coniferous forest soils in Finland (Maljanen et al. 2013). The highest emissions of HONO and NO were measured from the top soil layer of 0–5 cm. These emissions were linked to availability of  $\text{NO}_3^-/\text{NO}_2^-$  as reported by Su et al. (2011) and also soil moisture (Fig. 4). HONO and NO emissions increased with decreasing soil moisture, similar to results reported by Oswald et al. (2013). However, NO and HONO emissions were not tightly associated with  $\text{N}_2\text{O}$  production.

### *Conclusions*

The warmest plots in the natural temperature gradient in volcanic forest soil also emitted, in addition to previously reported  $\text{CH}_4$  and  $\text{N}_2\text{O}$ , more  $\text{CO}_2$  than the plots under ambient *in situ* soil temperature. These volcanic soils were also sources of NO and HONO in the laboratory measurements. When soils sampled from these plots with variable soil temperatures were incubated at a standard temperature (20°C), the production rates of  $\text{CH}_4$ ,  $\text{N}_2\text{O}$  and  $\text{CO}_2$  were still different between the plots, but temperature effects varied considerably between laboratory and field. The very high *in situ* emissions from the warmest plots and low emissions from laboratory incubations may possibly indicate adaptation mechanisms, but hint also at some geothermal rather than biological source for these gases. More studies are needed to confirm that, but if this assumption holds true, source partitioning with stable isotopes needs to be carried out on  $\text{CO}_2$ ,  $\text{CH}_4$  and  $\text{N}_2\text{O}$  emitted from these soils before interpreting temperature effects on greenhouse gas emissions from natural geothermal temperature gradients like ForHot sites.

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### REFERENCES

- Arnalds O 2015.** *The Soils of Iceland*. Springer, Netherlands. 183 p.
- Barnard R, Leadley PW & Hungate BA 2005.** Global change, nitrification, and denitrification: A review. *Global Biogeochemical Cycles* 19, GB1007.  
<http://dx.doi.org/10.1029/2004GB002282>.
- Bhattarai HR, Virkajärvi P, Yli-Pirilä P & Maljanen M 2017.** Emissions of atmospherically important nitrous acid (HONO) gas from northern grassland soil increases in the presence of nitrite ( $\text{NO}_2^-$ ). *Agriculture Ecosystems and Environment* 256, 194–199.  
<https://doi.org/10.1016/j.agee.2018.01.017>
- Chapin III FS, Matson PA & Mooney HA 2002.** *Principles of Terrestrial Ecosystem Ecology*. Springer, New York, Berlin, London. 392 p.
- Crowther TW & Bradford MA 2013.** Thermal acclimation in widespread heterotrophic soil microbes. *Ecology Letters* 6, 469–477.  
<http://dx.doi.org/10.1111/ele.12069>
- Etioppe G & Sherwood Lollar B 2013.** Abiotic methane on earth. *Reviews of Geophysics* 51, 276–299.  
<http://dx.doi.org/10.1002/rog.20011>.
- Fridriksson T, Kristjánsson BR, Ármannsson H, Margrétardóttir E, Ólafsdóttir S & Chiodini G 2006.**  $\text{CO}_2$  emissions and heat flow through soil, fumaroles, and steam heated mud pools at the Reykjanes geothermal area, SW Iceland. *Applied Geochemistry* 21, 1551–1569.  
<http://dx.doi.org/10.1016/j.apgeochem.2006.04.006>.

- Gargallo-Garriga A, Ayala-Roque M, Sardans J, Bartrons M, Granda V, Sigurdsson BD, Leblans N, Oravec M, Urban O, Janssens I & Peñuelas J 2017.** Impact of Soil Warming on the Plant Metabolome of Icelandic Grasslands. *Metabolites* 7, 44.  
<http://dx.doi.org/10.3390/metabo7030044>
- Heil J, Vereecken H & Brueggemann N 2016.** A review of chemical reactions of nitrification intermediates and their role in nitrogen cycling and nitrogen trace gas formation in soil. *European Journal of Soil Science* 67, 23-39.  
<http://dx.doi.org/10.1111/ejss.12306>
- Heland J, Kleffmann J, Kurtenbach R & Wiesen P 2001.** A new instrument to measure gaseous nitrous acid (HONO) in the atmosphere. *Environmental Science & Technology* 35, 3207-3212.
- Klusman RW, Moore JN & LeRoy MP 2000.** Potential for surface gas flux measurements in exploration and surface evaluation of geothermal resources. *Geothermics* 29, 637-670.  
[http://dx.doi.org/10.1016/S0375-6505\(00\)00036-5](http://dx.doi.org/10.1016/S0375-6505(00)00036-5)
- Leblans, NIW, Sigurdsson BD, Aerts R, Vicca S, Magnússon B & Janssens IA 2017.** Icelandic grasslands as long-term C sinks under elevated N inputs. *Biogeochemistry* 134, 279-299.  
<http://dx.doi.org/10.1007/s10533-017-0362-5>
- Le Mer J & Roger P 2001.** Production, oxidation, emission and consumption of methane by soils: A review. *European Journal of Soil Biology* 37, 25-50.  
[http://dx.doi.org/10.1016/S1164-5563\(01\)01067-6](http://dx.doi.org/10.1016/S1164-5563(01)01067-6)
- Lu M, Zhou X, Yang Q, Li H, Luo Y, Fang C, Chen J, Yang X & Li B 2012.** Responses of ecosystem carbon cycle to experimental warming: A meta-analysis. *Ecology* 94, 726-738.  
<http://dx.doi.org/10.1890/12-0279.1>
- Maljanen M, Yli-Moijala H, Biasi C, Leblans NIW, De Boeck HJ, Bjarnadóttir B & Sigurdsson BD 2017.** The emissions of nitrous oxide and methane from natural soil temperature gradients in a volcanic area in southwest Iceland. *Soil Biology and Biochemistry* 109, 70-80.  
<http://dx.doi.org/10.1016/j.soilbio.2017.01.021>
- Maljanen M, Yli-Pirilä P, Hytönen J, Joutsensaari J & Martikainen PJ 2013.** Acidic northern soils as sources of atmospheric nitrous acid (HONO). *Soil Biology and Biochemistry* 67, 94-97.  
<http://dx.doi.org/10.1016/j.soilbio.2013.08.013>
- Maljanen M, Sigurdsson BD, Gudmundsson J, Óskarsson H, Huttunen JT & Martikainen PJ 2010.** Land-use and greenhouse gas balances of peatlands in the Nordic countries - Present knowledge and gaps. *Biogeosciences* 7, 2711-2738.  
<http://dx.doi.org/10.5194/bg-7-2711-2010>
- O’Gorman E, Benstead JP, Cross WF, Friberg N, Hood JM, Johnson PW, Sigurdsson BD & Woodward G 2014.** Climate change and geothermal ecosystems: Natural laboratories, sentinel systems, and future refugia. *Global Change Biology* 20, 3291-3299.  
<http://dx.doi.org/10.1111/gcb.12602>
- Oswald R, Behrendt T, Ermel M, Wu D, Su H, Cheng Y, Breuninger C, Moravek A, Mougín E, Delon C, Loubet B, Pommerening-Roser A, Sörgel M, Pöschl U, Hoffmann T, Andreae MO, Meixner FX & Trebs I 2013.** HONO emissions from soil bacteria as a major source of atmospheric reactive nitrogen. *Science* 341, 1233-1235.  
<http://dx.doi.org/10.1126/science.1242266>
- Pilegaard K 2013.** Processes regulating nitric oxide emissions from soils. *Philosophical Transactions of the Royal Society B-Biological Sciences* 368, 20130126.  
<http://dx.doi.org/10.1098/rstb.2013.0126>
- Poeplau C, Kätterer T, Leblans NIW & Sigurdsson BD 2017.** Sensitivity of soil carbon fractions and their specific stabilisation mechanisms to extreme soil warming in a subarctic grassland. *Global Change Biology* 23, 1316-1327.  
<http://dx.doi.org/10.1111/gcb.13491>
- Priemé A & Christensen S 2001.** Natural perturbations, drying-wetting and freezing-thawing cycles, and the emission of nitrous oxide, carbon dioxide and methane from farmed organic soils. *Soil Biology and Biochemistry* 33, 2083-2091.  
[http://dx.doi.org/10.1016/S0038-0717\(01\)00140-7](http://dx.doi.org/10.1016/S0038-0717(01)00140-7)
- Rey A 2015.** Mind the gap: non-biological processes contributing to soil CO<sub>2</sub> efflux. *Global Change Biology* 21, 1752-1761.  
<http://dx.doi.org/10.1111/gcb.12821>
- Riedel K & Lassey K 2008.** Detergent of the atmosphere. *Water & Atmosphere* 16, 22-23.
- Scharko NK, Schütte UME, Berke AE, Banina L, Peel HR, Donaldson MA, Hemmerich C, White JR & Raff JD 2015.** Combined flux chamber and

genomics approach links nitrous acid emissions to ammonia oxidizing bacteria and archaea in urban and agricultural soil. *Environmental Science & Technology* 49, 13825–13834.

<http://dx.doi.org/10.1021/acs.est.5b00838>

**Sigurðsson BD, Leblans NIW, Dauwe S, Guðmundsdóttir E, Gundersen P, Gunnarsdóttir GE, Holmstrup M, Ilieva-Makulec K, Kätterer T, Marteinsdóttir B, Maljanen M, Oddsdóttir ES, Ostonen I, Peñuelas J, Poeplau C, Richter A, Sigurðsson P, van Bodegom P, Wallander H, Weedon J & Janssens I 2016.** Geothermal ecosystems as natural climate change experiments: The ForHot research site in Iceland as a case study. *Icelandic Agricultural Sciences* 29, 53-71.

<http://dx.doi.org/10.16886/IAS.2016.05>

**Smith KA 2017.** Changing views of nitrous oxide emissions from agricultural soil: Key controlling processes and assessment at different spatial scales. *European Journal of Soil Science* 68, 137-155.

<http://dx.doi.org/10.1111/ejss.12409>

**Stemmler K, Ammann M, Donders C, Kleffmann J, George C 2006.** Photosensitized reduction of nitrogen dioxide on humic acid as a source of nitrous acid. *Nature* 440, 195–198.

<http://dx.doi.org/10.1038/nature04603>

**Su H, Cheng Y, Oswald R, Behrendt T, Trebs I, Meixner FX, Andreae MO, Cheng P, Zhang Y & Pöschl U 2011.** Soil nitrite as a source of atmospheric HONO and OH radicals. *Science* 333, 1616–1618.

<http://dx.doi.org/10.1126/science.1207687>

**de la Torre JR, Walker CB, Ingalls AE, Könneke M & Stahl DA 2008.** Cultivation of a thermophilic ammonia oxidizing archaea on synthesizing crenarchaeol. *Environmental Microbiology* 10, 810–818.

<http://dx.doi.org/10.1111/j.1462-2920.2007.01506.x>

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