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The impact of ploughing intensively managed temperate grasslands on N$_2$O, CH$_4$ and CO$_2$ fluxes

J. Drewera, M. Andersona, P.E. Levyb, B. Scholtesb, C. Helftera, J. Parkerb, R.M. Reesb, U.M. Skiba

$^a$CEH, Bush Estate, Penicuik, EH26 0QB, Scotland, UK
$^b$SRUC, West Mains Road, Edinburgh, EH9 3JG Scotland, UK

Correspondence:

Julia Drewer
NERC Centre for Ecology and Hydrology
Bush Estate
Penicuik
EH26 0QB
Tel. +44 131 4454343
Fax. +44 131 4453943
juew@ceh.ac.uk

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Abstract

Background and aims
Temperate grasslands are a globally important component of agricultural production systems and a major contributor to the exchange of greenhouse gases (GHG) between the biosphere and atmosphere. Many intensively managed grazed grasslands in NW Europe are ploughed and reseeded occasionally in order to improve their productivity. Here, we examined the impact of ploughing on the emission of GHGs a grassland.

Methods
To study these interactions we measured soil GHG fluxes using the static chamber method in addition to the net ecosystem exchange (NEE) of CO₂ by eddy covariance from two adjacent fields. Until ploughing one field in 2012 and the other in 2014, management of these intensively grazed grasslands was almost the same and typical for the study region.

Results
The effect on N₂O is small, but distinguishable from the effects of N fertilisation, soil temperature and soil moisture. Tillage-induced N₂O fluxes were close to expectations based on the IPCC default methodology. By far the dominant effect on the GHG balance was the temporary reduction in GPP.

Conclusions
Ploughing and reseeding can substantially influence short-term GHG emissions. Therefore tillage-induced fluxes ought to be considered when estimating greenhouse gas fluxes or budgets from grasslands that are periodically ploughed.
Introduction

Grasslands rank among the world’s most extensive ecosystems and are used for forage production and animal grazing (Campbell and Stafford Smith 2000). They cover 22% of the EU-25 land area, accounting for 80 million ha (EEA 2005). Managed grasslands are major source of emissions of N\textsubscript{2}O, CO\textsubscript{2} and CH\textsubscript{4}, if grazed by ruminants. Emission rates depend on soil management, soil type, climate and interannual climate variability (Skiba et al, 2012, Jones et al. 2005).

In order to maintain high harvest yields and optimal grass growth for grazing, renovation activities, such as ploughing and harrowing, are periodically carried out on intensively managed grasslands. To maximise productivity, these grasslands are heavily fertilised and therefore known large sources of N\textsubscript{2}O (Davies et al. 2001, Soussana et al, 2007).

Tillage is defined as the mechanical manipulation of soil conditions to support crop production, including ploughing and harrowing operations (Brady and Weil 2002). Depending on local soil properties and weather patterns, grassland tillage can increase grass yield and improve soil structure and aeration through drainage, which is often necessary in order to maintain productivity. On the other hand, this mechanical agitation is known to change soil properties and thereby can affect the net GHG exchange of grasslands (Ball et al. 2014).

Pagliai et al. (2004) showed that soil porosity can decrease under conventionally tilled loam soils, and by reducing the size and the continuity of pores, water conductivity decreases. Conventional tillage (particularly in wet soils) can increase subsoil compaction, promoting conditions that are associated with increased rates of denitrification (Uchida et al. 2008). On
the other hand, conventional tillage can be beneficial for certain soil types, such as poorly
drained and compactable soils (Ball et al. 1999). Other studies reported that long-term
ploughing practices resulted in soil organic matter (SOM) losses (Eriksen and Jensen 2001),
microbial biomass and water-stable aggregation decrease as well as lower potentially
mineralisable N (Karlen et al. 2013). Generally, the impact on ploughing on soil properties
depends on the soil type and weather conditions, thus resulting in many contrasting reports in
the literature (Soane et al. 2012). Ball et al. (1999) reported that high rates of N$_2$O emissions
were mainly associated with rainfall patterns and compact arable soils, and no strong
correlation between soil tillage and N$_2$O emissions was found. In contrast, Kessavalou et al.
(1998) found a 100% increase in N$_2$O emissions from a loam soil after a tillage event during
fallow, which agrees with other studies (Estavillo et al. 2002). For poorly drained grasslands,
conventional tillage can be used as a mitigation method to increase soil porosity and water
infiltration. As a consequence, denitrification rates can decrease and N$_2$O emissions are
reduced (MacDonald et al. 2011).

We report detailed data which allowed comparison of the effect of ploughing on GHG
exchange at the long-term field study site, Easter Bush, South East Scotland. Two adjacent,
predominately sheep grazed grasslands under the same management, were ploughed two
years apart and thereby provided the opportunity to evaluate the magnitude of ploughing-
induced GHG fluxes. This was not a designed experiment, but reflects common farming
practice in this region, and can therefore provide useful information directly relevant to this
kind of land management.

Our questions were:

(1) Do ploughing and associated management operations increase N$_2$O, CH$_4$ and CO$_2$
flaxes?
How variable are ploughing-induced emissions?

Materials and Methods

Site description

The study site is located at Easter Bush, 10 km south of Edinburgh, Scotland, in a mesothermal maritime climate (latitude 55°52’N, longitude 3°2’W). The two adjacent fields (North Field (NF) and South Field (SF)) are managed grasslands (>90% Lolium perenne). The soil is an imperfectly drained sandy clay loam (FAO classification: eutric cambisol) with a clay content varying from 20 - 26% and a pH varying from 5 to 6 (in H2O), depending when the soil was last limed. Soils were limed prior to the ploughing in 2012 and the soil pH was 6.1 (in H2O) during the 2012 - 2014 study period. During extended periods of rain, these fields tend to have localised waterlogging due to an insufficient drainage system. A meteorological station positioned between these two fields provides continuous measurements, with data averaged over 30 min periods. Rainfall amount is measured using a tipping bucket and air temperature at a height of 1.5 m above ground. The 10 year mean (1 Jan 2004 – 31 Dec 2014) air temperature was 8.8 °C and rainfall 958 mm with a variation of less than 100 mm from the 10-year mean.

Agronomic management of both fields was very similar. In the 10 years prior to the ploughing experiment, the fields were predominately grazed by sheep and occasionally, for short periods, by cattle in 2004-2006, and on the NF in August and September 2012. Livestock was sporadically removed from the fields for periods of several days up to several weeks. The 10 year average livestock density was 0.84 LSU ha⁻¹ y⁻¹, cattle contributed only with 0.05 LSU ha⁻¹ y⁻¹. In order to maintain high grass yields, the fields receive mineral N fertiliser, mainly as ammonium nitrate (NH₄NO₃), but occasionally as NPK compound
fertiliser or urea. The 10 year average nitrogen (N) fertiliser application rate was 194 kg N ha$^{-1}$ y$^{-1}$, usually split across three applications during spring and early summer months (Skiba et al. 2013).

Foregoing ploughing and reseeding the grass was killed using Glyphosate (Table 1). The South field (SF) and the North field (NF) were ploughed on 1st May 2012 and on 20th May 2014, respectively, with a mouldboard plough to a depth of 30 cm. The fields were harrowed, reseeded and rolled a few days after both ploughing events. All management operations and fertiliser applications during the study periods in both years are summarised in Table 1 and the management operations were essentially identical for the two years. It is common practice not to apply N fertiliser until the grass is well established. Therefore only the NF received N fertiliser on the 28th of May 2012, and only the SF was fertilised on the 9th May 2014. In 2012 GHG flux measurements were made for 39 days before ploughing and 142 days after ploughing and in 2014 a shorter study provided the same measurements for 67 days before ploughing and 34 days after ploughing.

Measurements of soil $N_2O$, $CH_4$ and $CO_2$ fluxes

The static chamber method (Clayton et al. 1994) was used for $N_2O$ and $CH_4$ flux measurements. Round static chambers (diameter = 40 cm) consisting of opaque polypropylene bases, were installed on each field; 20 (10 in each field) in 2012 and 10 (5 in each field) in 2014, respectively. The bases of 10 cm height were inserted into the ground to a depth of approximately 5 cm for the entire study period to allow free grazing. Lids of 20 cm height, were fastened onto the bases using four strong clips, only during the 60 minute measurement periods. A strip of commercially available draft excluder glued onto flange of the lid provided a gas tight seal between chamber and lid. The lids were fitted with a pressure...
compensation plug to maintain ambient pressure in the chambers during and after sample
removal. Gas samples were taken at regular intervals over one hour (0, 30, 60 min in 2012
and 0, 20, 40, 60 min in 2014) for each chamber. A three way tap was used for gas sample
removal using a 100 ml syringe. 20 ml glass vials were filled with a double needle system to
flush the vials with five times their volume. The samples and three sets of four certified
standard concentrations (N₂O, CH₄, CO₂ in N₂ with 20% O₂) were analysed at CEH on an
HP5890 Series II gas chromatograph (Hewlett Packard (Agilent Technologies) UK Ltd.,
Stockport, UK) with electron capture detector (ECD) for N₂O analysis and flame ionization
detector (FID) for CH₄ analysis. These detectors were setup in parallel allowing the analysis
of the two GHGs at the same time. Limit of detection was 7 ppb for N₂O and 0.07 ppm for
CH₄. Peak integration was carried out with Clarity chromatography software (DataApex,
Prague, Czech Republic). The flux F (μg m⁻² s⁻¹) for each sequence of gas samples from the
different chambers was calculated according to Equation 1:

\[ F = \frac{dc}{dt} \times \frac{\rho V}{A} \]  (Equation 1)

Where \( \frac{dc}{dt} \) is the concentration (C, μmol mol⁻¹) change over time (t, in s), which was
calculated by linear regression.

\( \frac{\rho V}{A} \) is the number of molecules in the enclosure volume to ground surface ratio, where ρ is the
density of air (mol m⁻³),

V (m³) is the air volume in the chamber and

A (m²) is the surface area in the chamber (Levy et al. 2012).

In addition, ecosystem CO₂ respiration rates, which is the sum of soil and vegetation CO₂
respiration, were measured close to each chamber location using an opaque closed dynamic
chamber (volume: 0.001171 m³) covering 0.0078 m² of soil for 120 s with an EGM-4 infrared gas analyser (IRGA: InfraRed Gas Analyser) (PP Systems; Hitchin, Hertfordshire, England). Taking into account the soil temperature, fluxes were calculated based on the linear increase of CO₂ concentrations. In 2012, the short-term physical release of CO₂ immediately after ploughing the SF was investigated from 4 random locations. First soil respiration measurements were made within 10 – 19 minutes after the plough turned the soil over and were repeated at intervals up to almost 3 hours. Thereafter CO₂ respiration rates (bulk soil and vegetation), were always measured at approximately the same time and adjacent to the chambers used for N₂O and CH₄ flux measurements, both in 2012 and 2014.

Auxiliary physical and chemical soil measurements

Other environmental parameters were measured during time of chamber enclosure as possible explanatory variables for correlation with recorded GHG fluxes. Soil temperature was measured with a handheld Omega HH370 temperature probe (Omega Engineering UK Ltd., Manchester, UK) for each chamber location at a depth of 10 cm. Volumetric soil moisture content (VSM) was measured at a depth of 7 cm with a handheld Theta probe HH 2 moisture meter (Delta T-Devices, Cambridge, UK) horizontally inserted at four points around each chamber. Gravimetric moisture content (GWC) was occasionally measured to calibrate the Theta probes. In order to determine bulk density, total C/N, ammonium (NH₄⁺) and nitrate (NO₃⁻) concentrations, soil cores were taken around each of the chamber locations. Soil samples for determination of bulk density were collected using a galvanised iron ring (98.17 cm³) with a sharp edge that was inserted in the upper soil layer with a hammer to 5 cm depth without compaction. Samples were oven-dried at 105 °C until constant weight (usually 48 hours) and bulk density (g cm⁻³) was calculated based on the dry weight occupying the volume of the ring.
For $\text{NH}_4^+$ and $\text{NO}_3^-$ analysis 15 g of fresh soil was mixed in plastic flasks with 50 ml of 1 M KCl solution made up with deionised water. The flasks were put on a Stuart Orbital Shaker SSL1 (Barloworld Scientific Ltd., Stone, UK) set to 100 rpm for 1 hour. The extract was filtered with Whatman 42 filter papers and poured into vials that were stored frozen thereafter. Defrosted samples were then analysed with a SAN++ Automated Wet Chemistry Analyzer (Skalar Analytical B.V., Breda, Netherlands). To determine total soil C and N, samples were oven-dried at 105°C and ground with a mixer mill MM200 (Retsch GmbH & Co. KG, Haan, Germany) at CEH. Between 10 and 20 mg of each soil sample was transferred to tin capsules and analysed together with four standards of aspartic acid with a Flash 2000 Elemental Analyzer (Thermo Fisher Scientific, Cambridge, UK).

*Net ecosystem exchange of CO₂*

In addition to the above described ecosystem respiration rates, we measured the net ecosystem exchange of CO₂. In order to measure from the ploughed and unploughed fields simultaneously we installed a mobile eddy covariance (EC) system in addition to our permanent, long-term system, in both years.

*Long-term eddy-covariance system*

Fluxes of CO₂ have been measured continuously by eddy-covariance (EC) at Easter Bush since 2002. The EC mast is located along the fence line which separates the NF from the SF (Figure 1). The EC system consists of a Gill WindmasterPro ultrasonic anemometer for the measurement of 3D wind vector components and sonic temperature (20 Hz data), and of a LICOR 7000 closed-path infrared gas analyser (IRGA) operating at 10 Hz for the simultaneous measurement of CO₂ and H₂O mole fractions. Air is sampled at 10 l min⁻¹, 20
cm below the mid-point between the anemometer’s transducers (effective measurement height of 2.5 m) through a 10 m long Dekabon© line (OD ¼”). Data is captured and processed offline into half-hourly fluxes using in-house software written in LabView™ (National Instruments). Data capture was high in the period 9th May - 20th Aug 2012 (85%), with a 52% to 48% split between measured fluxes originating from the SF and the NF respectively. The extent of the flux footprint of the long-term EC system during the 2012 measurement period relevant to the ploughing experiment is shown in Figure 1. The footprint statistics used for this figure were obtained with the analytical Kormann-Meixner footprint model for non-neutral stratification (Kormann and Meixner 2001). In 2014, total data capture after filtering was 84% for the long-term EC system with a 71% to 29% split between measured fluxes originating from the SF and the NF respectively.

**Mobile eddy covariance system in 2012**

The prevailing wind direction pre- and post-ploughing was from the N/NW and not the usual S/SE. This means that the long-term EC system mainly measured CO₂ fluxes from the unploughed grassland in the NF. Therefore a temporary mast was erected in the SF in April 2012 (Figure 1) to achieve the direct temporal comparison of F_{CO₂} from the ploughed and unploughed field for wind directions in the range ~ N-NW to N-NE. The SF system was a Campbell Scientific EC150 open-path infrared gas analyser for CO₂ and H₂O combined with a Campbell CSAT3 ultrasonic anemometer, with effective measurement height of 1.90 m. Data were logged at 20 Hz to a Campbell Scientific CR3000 data logger and processed offline. The SF system provided 3245 half-hourly average flux in total in the period 9th May - 20th Aug 2012 (66% of possible half-hourly data points during this measuring period), of which 926 (28%) corresponded to wind directions in the range ~ N-NW to N-NE. Low turbulence (u* < 0.1 m s⁻¹) and periods of rain accounted for over 95% of missing data.
Mobile eddy covariance system in 2014

The prevailing wind direction was SE and the above mentioned long-term eddy covariance system provided the measurements for the SF (which in 2014 was the newly established grass sward, after ploughing in 2012). A mobile system, different to the mobile system used in 2012, was erected in the NF in May 2014 prior to the ploughing of the field on 20th May 2014 (Figure 1) and was removed on 4th Aug 2014. The EC system consisted of a Metek USA-1 ultrasonic anemometer operating at 20 Hz and a Licor 7000 closed-path infrared gas analysed measuring CO₂ and H₂O mole fractions at 10 Hz. Air was sampled 20 cm below the mid-point between the anemometer's transducers (effective measurement height of 2.3 m) at 8 l min⁻¹ through a 1.5 m long piece of Dekabon® tubing (OD ¼”). Data was logged by a laptop running an in-house data acquisition software written in LabView™ and were processed offline. Data capture was 58% with 47% of available data points attributable to the North field. After standard filtering and quality control (Helfter et al. 2015), there remained 25% of high quality data (19% daytime and 6% night time data). The IRGA was run with a scrubbing column (1:1 mixture of soda lime and drierite) in front of the reference cell rather than a supply of N₂; exhaustion of the chemicals was the greatest cause of data loss (> 80%).

Data analysis

For comparing soil properties before and after the ploughing event, paired t-tests were carried out and results with p<0.05 regarded as significant.

In an attempt to separate the effects of fertilisation and ploughing on N₂O flux, we used a simple model which describes the expected response to fertilisation. The N₂O flux was expected to increase to a peak value some time after the date of fertilisation, and show an exponential decline thereafter. We used the lognormal density function to represent this
pattern in time. Using data from all fertilisation events, we fitted two parameters, \( \mu \) and \( \sigma \). Conventionally, these represent the mean and standard deviation of the log-transformed data. However, in this context, \( \mu \) represents the time delay between fertilisation and the peak flux occurring, and \( \sigma \) represents a decay rate parameter. By expressing the flux data appropriately, these parameters can be found as the mean and standard deviation of a transformed data set, so numerical optimisation is not required. A scaling coefficient was derived by linear regression of these predictions on the observations. In this way, we found the best fit to the observations, given a lognormal-shape pattern following fertilisation. This procedure was applied only to \( \text{N}_2\text{O} \) fluxes, as there was no similar a priori expectation of a response of \( \text{CH}_4 \) or \( \text{CO}_2 \) fluxes to fertilisation.

We statistically analysed whether \( \text{N}_2\text{O} \) fluxes were related to ploughing using a mixed-effects model (Pinheiro and Bates, 2004). This expressed the \( \text{N}_2\text{O} \) flux in terms of four fixed effects: soil temperature, soil moisture, the predicted response following fertilisation, and whether ploughing had recently taken place or not. We also included two nested random effects, accounting for repeated measurements on individual chambers, which were nested within the two fields. For \( \text{CH}_4 \) and \( \text{CO}_2 \), we could fit a simpler model with the same random effects, but only the three fixed effects of soil temperature, soil moisture, and ploughing. All analyses were performed on log-transformed fluxes, so that the data met normality assumptions. To allow for negative values, an offset of 50 was added to \( \text{CH}_4 \) fluxes.

**Results**

*Rainfall, Temperature and soil moisture*

The rainfall patterns in 2012 and 2014 were similar. Cumulative rainfall over the two months prior to ploughing in 2012 was 118 mm, compared with 136 mm in 2014 (Figure 2c,d). Both
ploughing events were followed by a similarly wet period: 100 mm for the month of May 2012, and 116 mm during the post-ploughing month in 2014, around twice the long-term mean for May.

In 2012, the average air and soil temperatures in the two weeks before ploughing and one week after ploughing stayed below 10 °C (Figure 2a, 3a). The air temperature only increased to double figures (15 °C) on the 21 May, and stayed between 12 and 18 °C until the end of the measurement period. There was no significant rainfall the week before and the week after ploughing, but from the 31 May (i.e. almost one month after ploughing) rainfall frequency and amount increased (Figure 2c). Because of these cold, dry conditions, germination was very slow.

In 2014, the soil temperature was around 5 °C warmer at the time of ploughing, compared with 2012 (Figure 3a). Soil temperature rose fairly steadily from 12 °C to 20 °C over the study period following ploughing. In both years, soil temperature increased after ploughing, and the increase was greater in the ploughed field than in the unploughed field (Figure 3a). Unlike in 2012, there was no rainfall in the week before ploughing and reseeding in 2014 (Figure 2d), but frequent rain showers within two weeks of the ploughing event together with the warmer temperatures facilitated fast germination and almost complete canopy closure by the end of this much shorter study period.

Volumetric soil moisture (VSM) content in 2012 was larger in the NF than the SF irrespective of the ploughing (Figure 3b). In 2014 the VSM in the NF decreased from 70-90% to <30%. The downward trend was stronger after ploughing. The unploughed SF did not show this trend and even showed a slight increase in VSM in June to a maximum of around 60% from averages around 40% previously (Figure 3b).
Soil properties

Bulk density, total C and N, and KCl extractable NH$_4^+$ and NO$_3^-$ for the top 10 cm were measured one week before and one and five weeks after ploughing from both ploughed and non-ploughed fields in both years (Table 2). Both ploughing events significantly increased the soil bulk density of the top 5 cm by 37%, from 0.75 g cm$^{-3}$ to 1.19 g cm$^{-3}$. The small differences in bulk densities between 2012 and 2014 shown in Table 2 are not significant. Total C/N ratio was lower in 2012 than 2014 for both fields, none of the differences between years and fields were significant. In 2012 and 2014 differences in NH$_4^+$ and NO$_3^-$ concentrations were not significant for the two fields before ploughing. After ploughing the NH$_4^+$ and NO$_3^-$ concentrations were larger from the ploughed fields compared to the unploughed field, both 1 and 5 weeks after ploughing. These differences were significant for NH$_4^+$ on both post-ploughing dates in 2012 (p<0.001), and for NO$_3^-$ 1 week after ploughing in both years (p<0.05). In 2012 SF and NF NH$_4^+$ and NO$_3^-$ increased with time between pre-ploughing and 1 week later, and also between the 1 week and 5 week measurements. Differences were significant at p<0.05 and above for all, except for NO$_3^-$ concentrations from the SF 1 and 5 weeks after ploughing and the NF pre and 1 week after ploughing. In 2014 there was no significant change in NH$_4^+$ and NO$_3^-$ concentrations from the unploughed SF.

N$_2$O fluxes

Background mean fluxes in early spring in both years were <5 µg m$^{-2}$ h$^{-1}$ N$_2$O-N (Figure 4a). After fertilisation events, N$_2$O fluxes generally showed a peak followed by a decline, and the lognormal density function approximates this pattern in the data reasonably well (fitted lines in Figure 4a). After both ploughing events, N$_2$O fluxes showed a strong deviation from the pattern expected from fertilisation alone, and increased approximately linearly over the
following month, up to around 200 µg N₂O-N m⁻² h⁻¹ in 2012, and to 1300 µg N₂O-N m⁻² h⁻¹ in 2014 (red points in Figure 4a). However, soil temperatures also increased over both periods, so we cannot interpret this simply as a response to ploughing. To separate the effects of fertilisation, temperature and soil moisture from that of ploughing, we used the mixed-effects model analysis. This shows a strong indication that N₂O fluxes were higher after ploughing, after accounting for the effect of fertilisation, temperature and soil moisture (Table 3). Because the mixed-effects model is fitted to the log-transformed flux, the interpretation of the coefficients is not as straightforward as in the normal case. The exponentiated coefficients are interpreted as the proportional change in flux for a unit change in the independent variable. To translate these into more meaningful units, we calculate the absolute effect size as the difference in the fitted mixed model predictions with and without ploughing, at the mean level of all other inputs (Table 4). This predicts that fluxes were higher after ploughing by 14.1 µg N₂O-N m⁻² h⁻¹ in 2012, and 49.9 µg N₂O-N m⁻² h⁻¹ in 2014. By comparison with the average magnitude of fluxes after fertilisation events, we would expect fluxes to be on average 96 µg m⁻² h⁻¹ higher, if 1% of 70 kg N ha⁻¹ were released as N₂O in the 30 days following fertiliser application (although we would expect this to follow the lognormal pattern in time described previously). We thus estimate that ploughing has an effect which is ~14 - 52 % of that of typical N fertilisation.

**CH₄ fluxes**

Both positive and negative CH₄ fluxes were measured in both years. In early spring in both years on both fields, background fluxes ranged from uptake of a few tens of µg CH₄-C m⁻² h⁻¹ to positive emission fluxes of a few tens of µg CH₄-C m⁻² h⁻¹. After ploughing in May 2012, fluxes from the SF increased to a few hundreds of µg CH₄-C m⁻² h⁻¹ (Figure 4b, red points) whilst fluxes from the unploughed NF remained in the order of a few tens of µg CH₄-C m⁻² h⁻¹.
After ploughing of the NF in 2014, CH$_4$ fluxes increased to >5000 µg CH$_4$-C m$^{-2}$ h$^{-1}$. Fluxes also increased from the SF but only to about 500 µg CH$_4$-C m$^{-2}$ h$^{-1}$. Again, we used the mixed-effects model to separate the effect of ploughing from the effects of temperature and soil moisture (Table 3). This showed a strong effect of temperature, a weak effect of soil moisture, and a variable response to ploughing. Ploughing decreased CH$_4$, fluxes by 11 µg m$^{-2}$ h$^{-1}$ in 2012, and increased them by 36.5 µg m$^{-2}$ h$^{-1}$ in 2014 (Table 4). In the absence of ploughing, fertilisation in May in 2012 appeared to increase CH$_4$ fluxes, and to a lesser extent in August 2012, but there was no effect apparent in 2014 (Figure 4b).

Ecosystem respiration rates

Although variable, all 4 random locations on the ploughing day on 2012 demonstrated the immediate increase in CO$_2$ respiration within the first 30 min after the plough passed that particular area (Figure 5). This physical release of CO$_2$ remained for at least 3 hours, and fluxes then returned to near-background levels after around 80-90 min.

Ecosystem respiration rates in 2012 were on average around 250 mg CO$_2$-C m$^{-2}$ h$^{-1}$ in early spring for both fields (Figure 4c). Results from the mixed-model analysis show that ploughing decreased ecosystem respiration quite consistently, as well as showing a strong positive response to temperature (Table 3). The net effect of ploughing was to decrease ecosystem respiration by 71-85 mg CO$_2$-C m$^{-2}$ h$^{-1}$ (Table 4). An effect of fertilisation separate from that of temperature was not easily discernible.

Net ecosystem exchange of CO$_2$ measured by eddy covariance
There was a greater than usual occurrence of wind from the N-NW in the summer of 2012 which resulted in the near 50:50 split of data collected from NF and SF (Figure 1). The 70:30 split in favour of winds blowing from the SW observed in 2014 is more typical for the site.

The two ploughing events in 2012 and 2014 exhibited multiple similarities in terms of NEE (Figure 6). Daytime uptake of CO$_2$ by the ploughed field ceased after ploughing and fluxes remained positive for approximately 40 days after the event (Figure 6a and c). This is most obvious at ploughing of the NF in 2014 with highest coverage of eddy covariance data (Figure 6c). After ca. 40 days, CO$_2$ uptake in the ploughed and re-sown field was comparable to the non-ploughed field in each year; however, the variability in daytime NEE in the two fields was large (2-3 times larger in 2014 than in 2012; Figure 6a and c). Night time fluxes of CO$_2$ were not statistically different between fields in 2012 (Figure 6b) and the temporal variability was consistent with variations in soil temperature (weak positive correlation of fluxes with soil temperature which peaked in both fields ca. 27 days after ploughing; Figure 3a). Night time fluxes in the ploughed NF also followed the upward trend in soil temperature observed in 2014 (Figure 6d and Figure 3a). In contrast, night time respiration in the SF was larger than in the ploughed NF, it was more scattered and did not exhibit a clear correlation with soil temperature. Ploughing had a transient effect on CO$_2$ fluxes at Easter Bush, with a full recovery of the sink strength observed within 1.5 to 2 months after ploughing and re-sowing.

Daytime and night time CO$_2$ fluxes measured by EC increased sharply from the day of ploughing in 2014 and peaked 3 days later (Figure 6c and d) which we attribute to the combined effects of the physical removal of the CO$_2$ sink and the release of CO$_2$ from upturned soil layers.
Ploughing caused a net release of carbon of the order of 120 g CO₂-C m⁻² (95% confidence interval range 87 to 153 g CO₂-C m⁻²) during the month following the 2014 ploughing event. Data coverage for the ploughed SF during the month following the 2012 ploughing event was too sparse for the calculation of reliable cumulative fluxes. However, in light of Figure 6 it seems reasonable to assume that the net carbon loss in 2012 would be of similar magnitude as that observed in 2014 under similar meteorological conditions.

Discussion

Our results show that ploughing increased N₂O emissions, decreased ecosystem respiration, and had a mixed effect on CH₄ fluxes. We can estimate the total impact of ploughing by adding the increase in N₂O emissions, accounting for their relative global warming potential, to the net release of carbon following ploughing. We assume the net effect on CH₄ is small enough to be negligible. If N₂O emissions are increased by 14-50 µg N₂O-N m⁻² h⁻¹ over the month following ploughing, converting this to total mass of N₂O and CO₂ equivalent units using a global warming potential of 298 (IPCC 2014), we obtain values of 4-17 g CO₂-eq m⁻². This is small compared to the 440 g CO₂ m⁻² released as CO₂ in the month following ploughing, and gives a total of 444 - 457 g CO₂-eq m⁻². To put this into context, this represents 55% of the average harvest yield at this site when managed for hay or silage rather than grazing (Jones et al., submitted). Alternatively, the ploughing loss represents 7% of average GPP at the site. The purpose of ploughing is to increase sward productivity, so GPP would be expected to be larger in subsequent months and years. Whether the ploughing operation is GHG-neutral depends on the magnitude and duration of this longer-term effect on GPP, as this determines when/whether the increased carbon uptake offsets the short-term net source induced by the ploughing operation. This is difficult to discern without a longer term study.
The ploughing-induced increases in N\textsubscript{2}O emissions were rather different in 2012 and 2014, at 14 and 50 μg N\textsubscript{2}O-N m\textsuperscript{-2} h\textsuperscript{-1}, respectively. Because we have accounted for the effects of temperature and soil moisture in the analysis, it is not likely that this is due to differences in weather conditions. The difference between years may be related to different N contents in the vegetation at the time of ploughing. The increase in N\textsubscript{2}O emissions following ploughing is most likely due to increases in nitrogen inputs from mineralisation of the organic N in plant litter. In 2014, the ploughing took place six weeks after a fertilisation event, so the N stock in the vegetation was presumably higher than in 2012, when the field had not been fertilised that year. However, the difference in N\textsubscript{2}O emission between ploughing events is not clearly reflected in the measured ammonium and nitrate concentrations (Table 2). Similar short lived N\textsubscript{2}O emissions after tillage events on managed grassland were measured by other authors (Davies et al. 2001; Velthof et al. 2010; Merbold et al. 2014) and (Ball et al. 1997; Estavillo et al. 2002) linked these to increases in soil NO\textsubscript{3} concentrations, following mineralisation of the organic N in plant litter. An analysis of 39 studies in Europe concluded that incorporation of crop residue into the soil by ploughing resulted in a 6 fold increase in soil respiration rates and 12 fold increase in N\textsubscript{2}O emissions (Lethinen et al., 2014). The IPCC default inventory methodology for incorporation of crop residue (De Klein 2013) would predict an N\textsubscript{2}O emission of around 50 μg N\textsubscript{2}O-N m\textsuperscript{-2} h\textsuperscript{-1} for our site, based on estimates of biomass and plant N content from Jones et al (submitted), a shoot:root ratio of 1.5, using the 1% default emission factor, and assuming this were emitted over a month. This is very close to our higher value, obtained in 2014. The average fertiliser-induced N\textsubscript{2}O emission over the 3 weeks after fertilisation for the whole study period ranged from 0.29% to 2.94%.
Mineral agricultural soils tend to be only small sources and sinks for CH₄, unless irrigated. This is also the case for the Easter Bush fields, for which the average annual CH₄ fluxes were 3.4 μg CH₄-C m⁻² h⁻¹ for the period 2007 – 2010 (Skiba et al, 2013). On the ploughed field an additional CH₄ source was the decomposition of the ploughed under grass turf, which provided the labile carbon compounds and anaerobicity required for methanogenesis, and possibly was responsible for the slightly larger CH₄ emissions (Figure 4b). In 2014, CH₄ emissions were much larger from the ploughed NF, than the unploughed SF (Figure 4b). It is likely that under these warmer conditions, the main CH₄ source was the decomposition of the grass turf (Yamulki and Yarvis 2002).

A number of studies reported no conclusive evidence of tillage impacting soil microbial respiration rates in the long term (Yamulki and Jarvis, 2002, Jones et al. 2005, Ball et al, 1999). Our observations show a small but consistent decrease in ecosystem respiration rate following ploughing. However, it is important to make the distinction between soil respiration rate and ecosystem respiration rate (ie. including the above-ground plants), as the system definitions are different. When comparing ecosystem respiration rate before and after ploughing, the total biomass is initially the same, except the plants are over-turned, mostly dead and no longer respiring. The ecosystem respiration rate will therefore generally decrease. When comparing soil respiration rate before and after ploughing, the total biomass is generally increased after ploughing, as the above-ground plant material is now incorporated in to the soil. The soil respiration rate will therefore generally decrease. The physical release of CO₂ trapped in soil air for several days immediately after the ploughing of grassland soils (Kessavalou et al. 1998) and arable soils (Reicosky 1997; Vinten et al. 2002, Omonde et al. 2007) only makes a small contribution to the overall CO₂ emissions.
Our estimated net release of $4.0 \text{ g CO}_2\text{-C m}^{-2} \text{ d}^{-1}$ (95% confidence interval range 2.9 to 5.1 g CO$_2$-C m$^{-2}$ d$^{-1}$) following the 2014 ploughing event is consistent with other European studies (e.g. Merbold et al. (2014): 2.8 g CO$_2$-C m$^{-2}$ d$^{-1}$ for a restored grassland in Switzerland; Willems et al. (2011): 3.1 ± 1.2 g CO$_2$-C m$^{-2}$ d$^{-1}$ for a grassland in Ireland). In contrast, the unploughed SF had a net flux of -0.9 g CO$_2$-C m$^{-2}$ d$^{-1}$ (95% confidence interval range -2.7 to 0.9 g CO$_2$-C m$^{-2}$ d$^{-1}$) for the same 2014 time period.

The tillage management considerably changed the soil physical and chemical properties, broadly in the same manner on both fields in both years. Both tillage events, increased the bulk density in the top 5 cm soil depth from 0.77 g cm$^{-3}$ to 1.22 g cm$^{-3}$ (Table 2). The ploughing induced increase in bulk density is caused by the mechanically disruption of stable soil aggregates and mixing lighter more organic top soil with heavier mineral soil from the deeper layers. After the soil is rolled, the newly arranged soil aggregates are compacted and porosity and conductivity between pores decrease in the upper top soil layer (Ball, 2013). The reduction of soil aggregation increases evaporation (Six et al., 1998) and explains our observed reduction in soil moisture content after ploughing from both fields (Figure 3b). Average volumetric soil moisture content from the ploughed fields in 2012 (SF) and 2014 (NF) were 44% and 21% lower than from the unploughed fields.

Mineralisation rates are also favoured by the physical turnover of soil and break up of aggregates during ploughing by exposing new surfaces to the more oxygen rich atmosphere and by ploughing in the grass turf. Depending on the C/N ratio of the plant material, incorporation can either lead to immobilisation or mineralisation (Davis et al., 2001). At Easter Bush, the C/N ratios in the top 10 cm of the soil did not change significantly over the 6 week period, 1 week before to 5 weeks after tillage (Table 2). We observed a 10 and 5 fold increase in top soil (0 -10 cm) NH$_4^+$ and NO$_3^-$ concentrations in the first 5 weeks after ploughing in 2012 from the ploughed SF, but also a 7 and 5 fold increase in NH$_4^+$ and NO$_3^-$
concentrations from the unploughed NF. This implies that the raised concentrations are a result of several factors: weather and ploughing on the SF and climate and excreta and urine from the sheep grazed NF. The reason for the much larger NH$_4^+$ concentrations before ploughing in 2014 compared to 2012 are not obvious. In 2014 ploughing resulted in a significant decrease of NH$_4^+$ and increase of NO$_3^-$ concentrations, presumably caused by nitrification (Table 2). With hindsight, total C and N and NH$_4^+$ and NO$_3^-$ concentrations should have been measured for the entire plough depth (30 cm). The mixing of the soil layers and incorporation of the turf to the deeper layers will have created hotspots of mineralisation/immobilisation, which we could not account for by the 0-10 cm soil analysis.

Conclusions

Ploughing significantly increased fluxes of N$_2$O, reduced ecosystem respiration rate, and had a variable effect on CH$_4$ fluxes. The effect on N$_2$O is small, but distinguishable from the effects of N fertilisation, soil temperature and soil moisture. Tillage-induced N$_2$O fluxes were close to expectations based on the IPCC default methodology. By far the dominant effect on the GHG balance was the temporary reduction in GPP.

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Figure Captions

Figure 1: Satellite image (Google Earth; imagery date July 2012) showing the outline of the South field (SF) and the North field (NF), and the locations of the three eddy-covariance systems used during the two ploughing events of 2012 and 2014. The long-term, fixed eddy-covariance system (“EC fenceline”) is located along the fence which separates the two fields. A temporary eddy-covariance system was deployed in the SF (“EC (May – August 2012)”) during the spring and summer of 2012 to monitor pre- and post-ploughing fluxes within the ploughed field. A different system (see materials and methods section for details) was deployed in the NF (“EC (May-August 2014”) during the spring and summer of 2014. Overlain onto the satellite image are median values of $x_{\text{max}}$ (red line), $x_{50}$ (green line) and $x_{70}$ (purple line) (distance in meters from the EC mast where peak, 50% and 70% of the measured fluxes originated, respectively) for spring and summer 2012 as in this instance fluxes from the same tower could come from either field and plotted per 10 deg wind direction bins. These footprint statistics were obtained with the analytical Kormann-Meixner footprint model for non-neutral stratification (Kormann and Meixner, 2001).

Figure 2: Average daily air temperature (°C) (a, b) and daily rainfall (mm) (c, d) in 2012 (left, a & c) and 2014 (right, b & d). Ploughing was on the 1st May in 2012 and the 20th May in 2014 indicated by the dashed vertical red line.

Figure 3: a) Soil Temperature (°C) and b) Volumetric Soil Moisture (%) in 2012 (left panel) and 2014 (right panel) for North Field (NF) and South Field (SF), respectively. Measurements after ploughing in red and unploughed in blue. Fertilisation events indicated by blue horizontal line and ploughing by red horizontal line. To aid visualisation a smooth line was fitted through the data points.
Figure 4: Log fluxes of N$_2$O (a), CH$_4$ (b) [µg m$^{-2}$ h$^{-1}$] and CO$_2$ (c) [mg m$^{-2}$ h$^{-1}$] in 2012 (left panel) and 2014 (right panel) for North Field (NF) and South Field (SF), respectively. Measurements after ploughing in red and unploughed in blue. Fertilisation events indicated by blue horizontal line and ploughing by red horizontal line. A simple exponential decay after fertilisation fitted as blue line through log N$_2$O fluxes to indicate fertilisation induced predicted flux.

Figure 5: Soil CO$_2$ respiration rates on the day of ploughing. The bars represent average values from 4 measurement positions, the error bars are standard deviation. Time is the period in minutes after the plough passed the 4 plots on 5 repeated occasions.

Figure 6: Day time and night time fluxes of carbon dioxide (CO$_2$) measured by an eddy-covariance system installed along the fence line separating the north field (NF) and the south field (SF); (a)-(b) 2012 fluxes and (c)-(d) 2014 fluxes.