



1 **The influence of tillage on N<sub>2</sub>O fluxes from an intensively managed grazed**  
2 **grassland in Scotland**

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11

12 **Abstract**

13 Intensively managed grass production in high rainfall temperate climate zones is a globally important source of  
14 N<sub>2</sub>O. Many of these grasslands are occasionally tilled and can lead to increased N<sub>2</sub>O emissions. This was  
15 investigated by comparing N<sub>2</sub>O fluxes from two adjacent intensively managed grazed grasslands in Scotland, one  
16 of which was tilled. A combination of eddy covariance, high resolution dynamic chamber and static chamber  
17 methods greatly improved the temporal and spatial coverage of N<sub>2</sub>O fluxes before and after the tillage event and  
18 is recommended to be followed in future studies.

19 Total cumulative fluxes calculated for the tilled and un-tilled fields over the 175 day measurement period were  
20  $2.45 \pm 0.27$  and  $2.08 \pm 0.23$  kg N<sub>2</sub>O-N ha<sup>-1</sup>, respectively. N<sub>2</sub>O emissions from the tilled field increased significantly  
21 for several days immediately after ploughing and remained elevated for approximately two months after the tillage  
22 event contributing to an estimated increase in N<sub>2</sub>O fluxes of  $1.08 \pm 0.14$  kg N<sub>2</sub>O-N ha<sup>-1</sup>. Cumulative fluxes  
23 calculated over a 28 day period in August after the application of 70 kg-N ha<sup>-1</sup> as ammonium nitrate to both fields  
24 were estimated at  $0.42 \pm 0.15$  and  $0.75 \pm 0.14$  kg N<sub>2</sub>O N ha<sup>-1</sup> for the tilled and un-tilled fields, respectively. The  
25 tillage event appears to have substantially increased N<sub>2</sub>O fluxes from the tilled grassland field over a two month  
26 period; however, this increase may have been fractionally offset by a decrease in emissions after the August  
27 fertilisation event.

28



29 **1 Introduction**

30 Modern agriculture and intensive land management practices are believed to contribute over 39 % of total global  
31 anthropogenic emissions of the greenhouse gas (GHG) nitrous oxide (N<sub>2</sub>O) (IPCC, 2014). N<sub>2</sub>O is a naturally  
32 occurring GHG released into the atmosphere by the microbial processes of nitrification and denitrification which  
33 occur in soils and aquatic systems (Davidson et al., 2000; Seitzinger et al., 2000). Human activities which alter  
34 environmental conditions can have a significant impact on natural microbial processes which in turn can increase  
35 N<sub>2</sub>O emissions. Agricultural activities such as the use of nitrogen fertilisers, livestock production and land use  
36 changes are all important sources of anthropogenic N<sub>2</sub>O from agricultural soils (Fowler et al., 2013).

37 There is still large uncertainty associated with the quantification of N<sub>2</sub>O emissions released from  
38 agricultural soils on a national and global scale due to the large spatial and temporal variability of N<sub>2</sub>O fluxes  
39 (Cowan et al., 2015; Jahangir et al., 2011; Mathieu et al., 2006). Many past experiments have measured the release  
40 of N<sub>2</sub>O from soils after the application of nitrogen fertilisers - which are believed to be the most significant  
41 contributor to the rise of N<sub>2</sub>O emissions since pre-industrial times (e.g. Bouwman et al., 2002; Dobbie et al.,  
42 1999). Other causes of N<sub>2</sub>O emissions from agricultural soils, such as tillage and compaction, are less well  
43 documented, thus preventing effective assessment of their contribution to the overall annual N<sub>2</sub>O flux from the  
44 agricultural sector.

45 The use of nitrogen fertilisers (Abdalla et al., 2010; Yamulki and Jarvis, 2002), the presence of crop  
46 residues (Baggs et al., 2003; Mutegi et al., 2010), soil compaction (Ball et al., 2008; Yamulki and Jarvis, 2002)  
47 and the regularity and method of tillage (Sheehy et al., 2013) have all been reported to affect tillage induced N<sub>2</sub>O  
48 emissions. Changes in N<sub>2</sub>O emissions after tillage events are believed to be partly due to altering the bulk density,  
49 water filled pore space (WFPS) and oxygen availability in soils which can lead to an increase or decrease in  
50 nitrification and denitrification rates depending on environmental conditions (Elmi et al., 2003; Palma et al.,  
51 1997). The addition of nitrogen to tilled soils in the form of decaying plant matter (crop residues) is a recognised  
52 potential source of N<sub>2</sub>O; however the emissions associated with specific crop residues are not well quantified.  
53 Currently the IPCC emission inventories estimate that 1 % of all organic nitrogen applied to soils as crop residues  
54 will be emitted in the form of N<sub>2</sub>O (IPCC, 2006).

55 The large number of variables which may alter microbiological processes in tilled soils can lead to a wide  
56 variety of results between experiments carried out at different field sites under different meteorological conditions.



57 As a result, tillage events in agricultural fields have been reported to have very different effects on N<sub>2</sub>O production.  
58 Some experiments have reported large increases in N<sub>2</sub>O emissions varying from 0.89 to 3.37 Kg N ha<sup>-1</sup> (i.e.  
59 Chatskikh and Olesen, 2007; Merbold et al., 2014; Omonode et al., 2011; Pinto et al., 2004; Yamulki and Jarvis,  
60 2002) dependant of fertiliser application post-tillage, whereas others have shown a zero (i.e. Boeckx et al., 2011;  
61 Choudhary et al., 2002) or potentially negative effect of tillage (-0.88 Kg N ha<sup>-1</sup>, Tan et al., 2009). There is no  
62 consensus among these studies when quantifying the effect of different drivers of N<sub>2</sub>O production; however, it is  
63 common that factors influencing the aerobicity of the soil (such as WFPS and bulk density) are cited as influential  
64 in most tillage studies.

65 Improving our understanding of N<sub>2</sub>O fluxes from tillage events is important, especially in countries such  
66 as the UK, where agriculture accounts for approximately 70 % of the total land coverage (DEFRA, 2012) and  
67 tillage is widely practiced. Improved grasslands alone account for 25 % of the total land coverage of the UK  
68 (Morton et al., 2011). Although permanent grasslands are tilled less often than arable land, the conversion between  
69 arable and grazed pasture is a fairly common occurrence which requires a tillage event. Due to the large number  
70 of tillage events which occur annually across the country it is possible that even small perturbations in fluxes  
71 caused by these events could contribute significantly to the total national inventory of anthropogenic N<sub>2</sub>O  
72 emissions. Unlike arable fields, many of which are tilled annually, intensively managed grasslands are only tilled  
73 occasionally (generally every 5 to 20 years depending on soil conditions and grazing intensity), either for  
74 conversion to arable use or to improve grass sward productivity. The regularity of sward renewal depends  
75 primarily on the condition of the grass available for grazing and desired stocking density and is entirely dependent  
76 on the opinion and experience of farm managers in different climates. Few experiments have been carried out on  
77 GHG emissions resulting from the tillage of grassland fields due to the infrequency of these events. The aim of  
78 this work was therefore (i) to use multiple N<sub>2</sub>O flux measurement methodologies to add to the understanding of  
79 the magnitude of N<sub>2</sub>O fluxes from grasslands tilled for sward renewal, (ii) develop an improved statistical  
80 methodology which allows for uncertainties in cumulative flux emissions to be calculated for the event, and (iii)  
81 compare our cumulative flux emissions with those predicted using the current IPCC methodology for crop residue  
82 emissions.

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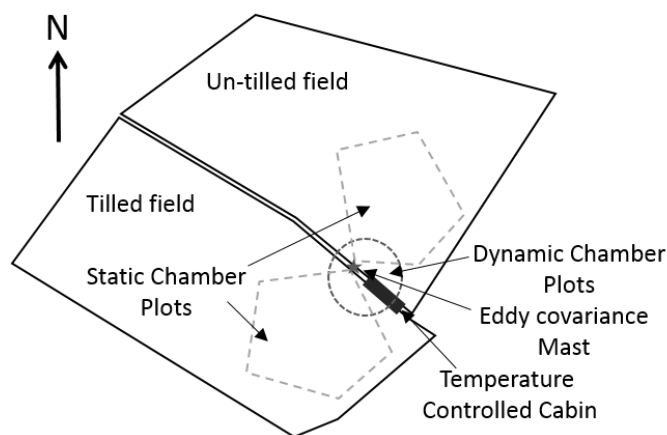


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## 86 2 Materials and method

### 87 2.1 Field site

88 Fluxes of  $N_2O$  were measured from an area of intensively managed, grazed grassland (Easter Bush, Scotland,  $55^\circ$   
89  $51' 55.30''N$ ,  $3^\circ 12' 22.17''W$ ) before and after a tillage event on the 1<sup>st</sup> of May 2012, and were compared with  
90 fluxes measured from an adjacent grassland which remained un-tilled (as described in Jones et al., 2011) (Figure  
91 1). The climate is temperate maritime, with an average annual rainfall of 921 mm and average annual air  
92 temperature of  $9^\circ C$  (in the period 2001–2011). The two fields (each approximately 5.4 ha) have been managed  
93 for intensive livestock production for at least twenty years, and since 2002 were predominately grazed by sheep.  
94 The average stocking densities were  $0.7 \text{ LSU ha}^{-1}$  (livestock units) and average N fertiliser application rates have  
95 been approximately  $200 \text{ kg N ha}^{-1} \text{ y}^{-1}$ . Mainly  $NH_4NO_3$  or NPK compound fertilisers were applied in three split  
96 applications usually between March and July (Skiba et al., 2013).



97

98 **Figure 1**  $N_2O$  fluxes were measured from two adjacent grassland fields at the Easter Bush Farm (Penicuik,  
99 Scotland). The north field remained un-tilled, while the south field was ploughed on the 1<sup>st</sup> of May 2012. An eddy  
100 covariance mast was setup next to a permanent cabin positioned between the fields. Dynamic chamber  
101 measurements were made within a 30 m radius of the cabin. Static chambers were spread out within the fetch of  
102 the eddy covariance mast.



103           The soils are clay loams with a sand/silt/clay texture of 28/20/52 and 24/19/57 for the top 30 cm in the  
 104 un-tilled and tilled fields, respectively with a pH of 5.1 (in H<sub>2</sub>O). They are classed as an imperfectly drained  
 105 Macmerry soil of the Rowanhill association (eutric cambisol, FAO classification). A drainage system had been  
 106 installed about 50 years ago, but is no longer functioning well, resulting in frequent occurrence of surface water  
 107 during rainy periods. The fields had not been tilled for at least twenty years, and the farmer had reported reduced  
 108 fertility and productivity. This together with the poor drainage led to the decision to till both fields.

109           In the first stage, only one field (also called the South Field in Jones et al., 2011) was tilled (Table 1). In  
 110 preparation, glyphosphate (1.5 l ha<sup>-1</sup>) was applied to kill the grass three days prior ploughing on the 27<sup>th</sup> of  
 111 April. The addition of glyphosphate to the soil is considered standard practice when grassland fields are  
 112 ploughed in the area and the effect that it may have had on microbiological processes or the decay of the grass  
 113 materials ploughed into the soil are considered to be part of the tillage event as a whole in this study. The field  
 114 was ploughed to a depth of 30 cm on the 1<sup>st</sup> of May 2012. Two days after ploughing the field was harrowed, then  
 115 rolled and sown with ryegrass (*Lolium perenne* L.) three days after ploughing. The un-tilled field (also called the  
 116 North field in (Jones et al., 2011)) was managed as usual and grazed by sheep (approximately 30 sheep ha<sup>-1</sup>).  
 117 Fertilisation events continued as normal on the un-tilled field which received two ammonium nitrate (Nitram)  
 118 fertiliser applications of 70 kg-N ha<sup>-1</sup>, one on the 28<sup>th</sup> of May and the second on the 9<sup>th</sup> of August. The tilled field  
 119 only received a 70 kg-N ha<sup>-1</sup> Nitram application on the 9<sup>th</sup> of August approximately four months after the tillage  
 120 event.

121           **Table 1** Field management events for both the tilled and un-tilled fields in 2012.

Date	Tilled Field (South)	Un-Tilled Field (North)
27 <sup>th</sup> April 2012	Glyphosphate application (1.5 l ha <sup>-1</sup> )	
1 <sup>st</sup> May 2012	Ploughing at 30 cm depth	
3 <sup>rd</sup> May 2012	Harrowing, seeding & rolling	
28 <sup>th</sup> May 2012		70 kg-N ha <sup>-1</sup> Nitram application
9 <sup>th</sup> August	70 kg-N ha <sup>-1</sup> Nitram application	70 kg-N ha <sup>-1</sup> Nitram application

122

123           Biomass samples were collected from the fields before tillage. Twenty soil cores (12 cm deep and 5.8  
 124 cm diameter) were extracted from the fields. All above ground biomass was removed from the soil and dried in



125 an oven at 80°C until constant weight. Once dry the above ground biomass was weighed. The remaining soil cores  
126 were broken up by hand and dried at 100°C until constant weight. After drying, the root materials were separated  
127 from the soils by hand and weighed. Sub samples of the dried plant materials were prepared for combustion  
128 elemental analysis of total carbon and nitrogen contents (vario EL cube, Elemaentar, Hanau, Germany).

## 129 2.2 Flux Measurements

130 N<sub>2</sub>O fluxes were measured from both tilled and un-tilled fields over a seven month period using three measurement  
131 methodologies; eddy covariance, static chamber and dynamic chamber techniques. Eddy covariance was the  
132 primary measurement methodology used; however, due to unpredictable changes in wind direction at the site it  
133 was necessary to deploy chamber methodology to ensure that both fields were measured periodically during the  
134 experiment. The dynamic chamber measurements was used as a cost effective way to provide many (>30) high  
135 resolution N<sub>2</sub>O fluxes on the days immediately after tillage without the need for time consuming GC lab analysis  
136 required by static chambers.

137 An eddy covariance system was installed on the 27<sup>th</sup> of March on the field boundary (See Figure 1). An  
138 ultra-sonic anemometer (WindMaster Pro 3-axis, Gill, Lymington, UK) mounted at 2.2 m was used to measure  
139 fluctuations in 3-D wind components at a frequency of 10 Hz. Mixing ratios of N<sub>2</sub>O, H<sub>2</sub>O and CO<sub>2</sub> were measured  
140 at 10 Hz by a quantum cascade laser (QCL) gas analyser (CW-QC-TILDAS-76-CS, Aerodyne Research Inc.,  
141 Billerica, MA, USA), housed in a temperature controlled cabin. The inlet line to the QCL was a 13.5 m length of  
142 Dekabon tubing (0.25 inch outer diameter), with a flow rate of approximately 13 l min<sup>-1</sup>. Fluxes were calculated  
143 at 30 min intervals using the EddyPro software (Version 5.2.1) (Li-Cor, Lincoln, NE, U.S.A.), based on the  
144 covariance between the N<sub>2</sub>O concentration ( $\chi$ ) and vertical wind speed ( $w$ ):

$$145 \quad F_{\chi} = \overline{\chi'w'} \quad (\text{Eq. 1})$$

146 In the processing, we applied double coordinate rotation (vertical and crosswind), spike removal, block  
147 averaging, and time lag removal by covariance maximisation. Correction for the frequency response of the system,  
148 both high and low-frequency losses, were made using the method of Moncrieff et al., (1997). Corrections for  
149 density fluctuations were applied on a half-hourly basis using the method of (Burba et al., 2012). The quality  
150 control scheme of Mauder & Foken, (2004) was used to remove poor quality flux measurements (their category  
151 2). Data were also rejected on the basis of high instrumental noise, and friction velocity ( $u^*$ ) values less than 0.05



152 m s<sup>-1</sup>. Fluxes measured with a mean wind direction between 150 and 300 degrees were classed as from the tilled  
153 field; those measured at greater than 320 and less than 130 degrees were classed as from the un-tilled field. The  
154 remaining data were disregarded due to obstruction of the wind by the cabin and fence line. Standard  
155 meteorological variables (rainfall, air temperature and soil temperature) were recorded by a nearby weather  
156 station.

157 N<sub>2</sub>O was also measured from both fields using static chamber and dynamic chamber techniques. The  
158 static chambers consisted of a cylindrical polyvinyl chloride (PVC) plastic pipe of 38 cm inner diameter (ID) and  
159 22 cm height. These chambers were inserted 5 cm into the soil, giving a headspace of approximately 20.4 l.  
160 Chambers were closed for 40 mins, during which time three 100-ml gas samples were collected via a syringe and  
161 a three-way tap fitted to the lid, at t = 0, 20 and 40 mins. After each measurement, chamber height was measured  
162 at five points to estimate the chamber volume. Gas samples were stored in 20 ml glass vials which were flushed  
163 with 100 ml of air in the syringe using a double needle. Samples were analysed using a Hewlett Packard 5890  
164 series II gas chromatograph (Agilent Technologies, Stockport, fitted with an electron capture detector) (Skiba et  
165 al., 2013).

166 Ten static chambers were positioned in each of the fields, within the estimated flux footprint of the eddy  
167 covariance system (10 to 200 m from the mast). Static chambers in the un-tilled field were kept in the same  
168 position for the duration of the experiment. Chambers in the tilled field were occasionally moved to allow access  
169 to farm vehicles during the different stages of the tillage operation. Chamber measurements were carried out  
170 between 9:00 and 15:00 on the measurement dates. Fluxes were calculated as:

$$171 \quad F = \frac{dC}{dt} \cdot \frac{\rho V}{A} \quad (\text{Eq. 2})$$

172 where  $F$  is the gas flux from the soil (nmol m<sup>-2</sup> s<sup>-1</sup>),  $dC/dt$  is the rate of change in concentration with time in nmol  
173 mol<sup>-1</sup> s<sup>-1</sup> estimated by linear regression,  $\rho$  is the density of air in mol m<sup>-3</sup>,  $V$  is the volume of the chamber in m<sup>3</sup>  
174 and  $A$  is the ground area enclosed by the chamber in m<sup>2</sup>.

175 Fluxes were also measured using the QCL in a closed, dynamic chamber system (Cowan et al., 2014a).  
176 A chamber (39 cm ID, 22 cm high) was placed onto a stainless steel collar inserted several cm into the soil (on  
177 average 5 cm) prior to measurement. Two 30 m lengths of 3/8 inch ID Tygon<sup>®</sup> tubing connected the chamber to  
178 the inlet of the QCL and the outlet of a vacuum pump (SH-110, Varian Inc, CA, USA) to form a closed system.



179 This allowed a 30-m possible radius from the instrument cabin in which the chamber could be placed (Figure 1).  
180 A flow rate of approximately 6 to 7 L min<sup>-1</sup> was used, with a lag time of approximately 22 seconds between the  
181 chamber and analyser. Fluxes of N<sub>2</sub>O were calculated with 1-Hz data over three minutes, using both linear and  
182 non-linear asymptotic regression methods (Levy et al., 2011; Pedersen et al., 2010). Using a mixture of goodness-  
183 of-fit statistics and visual inspection, the regression method that provided the best fit for the time series of mixing  
184 ratios of N<sub>2</sub>O was chosen for each individual measurement. The detection limit of individual fluxes calculated by  
185 this method was approximately 0.04 nmol m<sup>-2</sup> s<sup>-1</sup> compared to 0.4 nmol m<sup>-2</sup> s<sup>-1</sup> when using the static chambers  
186 (Cowan et al., 2014a, 2014b).

187 In the first few days after the tillage event, the wind direction was north-easterly, meaning that the eddy  
188 covariance system could not record fluxes from the tilled field (to the south-west). The dynamic chamber  
189 measurements were primarily used to fill this gap in the eddy covariance time series with high precision chamber  
190 measurements.

191

### 192 2.3 Gap filling

193 Because the eddy covariance system was placed on the field boundary, observations could only be made on a  
194 single field at any given time. Furthermore, some data were missing because of instrument failure and some had  
195 to be rejected according to the quality control criteria used. In order to estimate cumulative fluxes from both  
196 fields, temporal interpolation of the missing data points was required. However, in the absence of a well-validated  
197 process-based model for N<sub>2</sub>O fluxes on which to base predictions, it is not obvious how this is best achieved. The  
198 most common approach is to linearly interpolate in time between flux measurements. In this study, a general  
199 additive model (GAM) was used as an alternative approach, which accounted for temporal patterns at a range of  
200 time scales and nonlinear responses to environmental variables, implemented using the *mgcv* package in the R  
201 software (Wood, 2006).

202 We fitted the GAM with the same model terms to the separate data sets from the tilled and un-tilled  
203 fields. The terms included were air temperature, soil temperature, precipitation, and time. Additional terms for  
204 temperature and precipitation aggregated over longer intervals (1, 6, 12, 24 and 48 hours preceding the flux  
205 measurement) were examined and included where they improved the fit. The GAM allows for non-linearity by





206 fitting a smooth response with cubic splines. The degree of smoothing is optimised by the algorithm, but was also  
207 adjusted subjectively, such that the model was not over-fitting to noise in the data. Observations from eddy  
208 covariance and the two chamber methods were given equal weighting.

209 Predictions from the GAM were used to fill gaps when observations were not available. Uncertainty in  
210 predictions was estimated by simulating 2000 replicate time series from the GAM, given the uncertainty in the  
211 fitted parameters, to estimate the posterior distribution. The quantiles of this posterior distribution provided the  
212 95 % credibility interval at each predicted time step. To calculate cumulative fluxes, observed fluxes were used  
213 with their associated uncertainties when available; otherwise the GAM predictions were used.

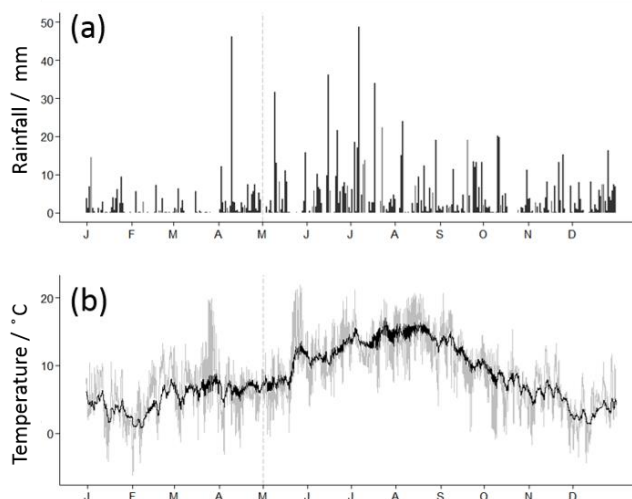
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## 215 **3 Results**

### 216 **3.1 Meteorological data**

217 A total of 1191 mm of rain was recorded in 2012, higher than the average annual rainfall of 921 mm (2001 to  
218 2011) for the Easter Bush area (Figure 2a). Historically, the wind direction at the field site is predominantly south-  
219 westerly (85 %). However, during the measurement campaign, the wind direction was split more evenly between  
220 the tilled and un-tilled fields (Figure 3). This allowed a better basis for comparison of N<sub>2</sub>O fluxes from the two  
221 fields, although data coverage for each field was low, 27 % and 25 % for tilled and un-tilled respectively.

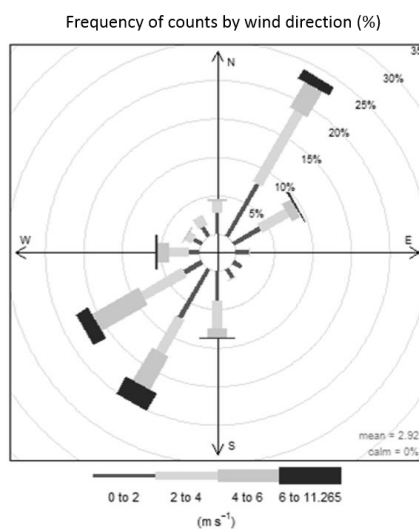
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224 **Figure 2** (a) Accumulated daily rainfall at the Easter Bush Field site during the year 2012. (b) Air temperature at  
225 height 3 m (grey) and soil temperature (black) recorded at the Easter Bush field site during the year 2012. Tillage  
226 occurred on the 1<sup>st</sup> of May 2012 (grey dashed vertical line).

227



228

229 **Figure 3** Wind rose plot for the Easter Bush field site during eddy covariance measurements (March – October  
230 2012).



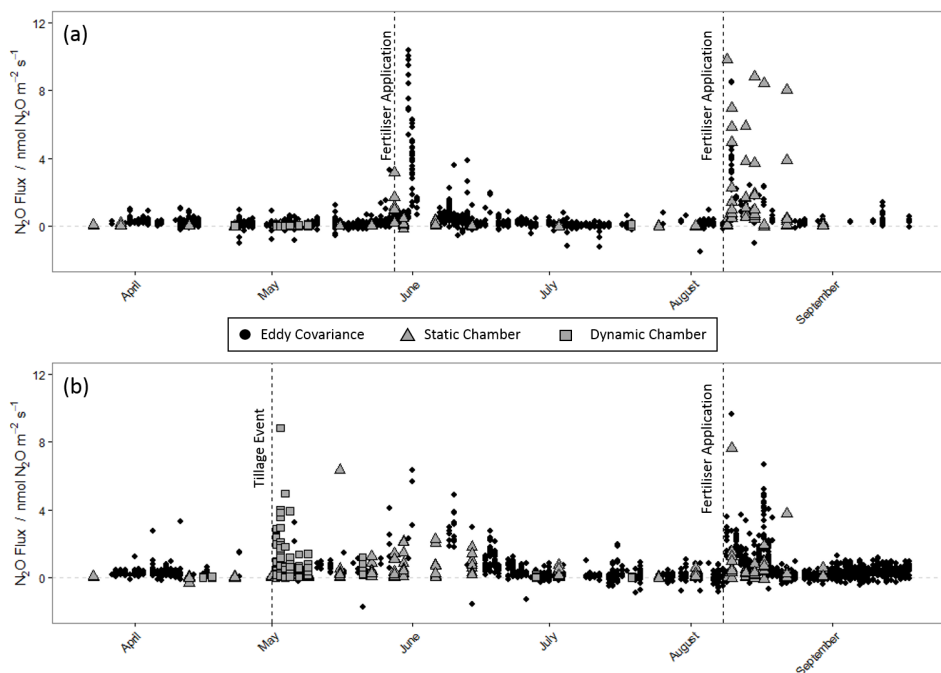
### 231 3.2 Comparison of N<sub>2</sub>O fluxes measured from the un-tilled and tilled fields

232 Before the tillage event, N<sub>2</sub>O fluxes were similar in the tilled and untilled fields. In both cases, around 90 % of  
233 fluxes were below 0.5 nmol m<sup>-2</sup> s<sup>-1</sup> (Figure 4). All three fertilisation events (the two fertiliser events in the untilled  
234 filed and single fertiliser event in the tilled field) were characterised by an emission peak of 5-10 nmol m<sup>-2</sup> s<sup>-1</sup>  
235 lasting a few days, which declined over the following days and weeks, often with considerable variability and  
236 some apparent secondary peaks (Figure 5). Fluxes had returned to background levels (<0.5 nmol m<sup>-2</sup> s<sup>-1</sup>) within  
237 28 days of each of the fertilisation events. Fluxes measured by all methods agreed reasonably well in magnitude,  
238 and there is no strong evidence for a systematic bias, given the differences in the spatial and temporal sampling  
239 (for a more specific insight see e.g. Cowan et al., 2014a).

240 The tillage event also produced an increase in emissions, and although the peak was less clearly defined,  
241 the effect was more prolonged. Fluxes generally ranged from ~0 to 1.0 nmol m<sup>-2</sup> s<sup>-1</sup> in the days before tillage and  
242 ~0 to 8.8 nmol m<sup>-2</sup> s<sup>-1</sup> in the week immediately after tillage (Figure 4b). Fluxes from the tilled field from mid to  
243 late May were approximately 1 nmol m<sup>-2</sup> s<sup>-1</sup> higher than from the untilled field (before the latter was fertilised).  
244 There followed an apparent increase in N<sub>2</sub>O fluxes lasting approximately four weeks from the tilled field from  
245 late May to late June, peaking in the first week in June (Figure 5c). Unfortunately, data coverage was rather low  
246 during this period due to changes in wind direction and a one week period in which the QCL was unavailable.  
247 Because the tilled field had not been fertilised since the previous year, we infer that the increased fluxes were a  
248 result of the tillage event. Fluxes in the tilled field returned to pre-tillage magnitude during July. By July, a new  
249 sward of grass had grown in the tilled field, but sheep were not re-introduced into the field until late August.

250 The GAM method was used to gap fill flux data to calculate cumulative fluxes (Figure 6). Cumulative  
251 N<sub>2</sub>O fluxes calculated for the tilled and un-tilled fields for the 175 days from 27th of March to the 18th of October  
252 were 2.45 ± 0.27 and 2.08 ± 0.23 kg N<sub>2</sub>O-N ha<sup>-1</sup>, respectively (Figure 7).

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254

255 **Figure 4** Fluxes of  $N_2O$  from the (a) un-tilled and (b) tilled fields measured at the Easter Bush field site in 2012.

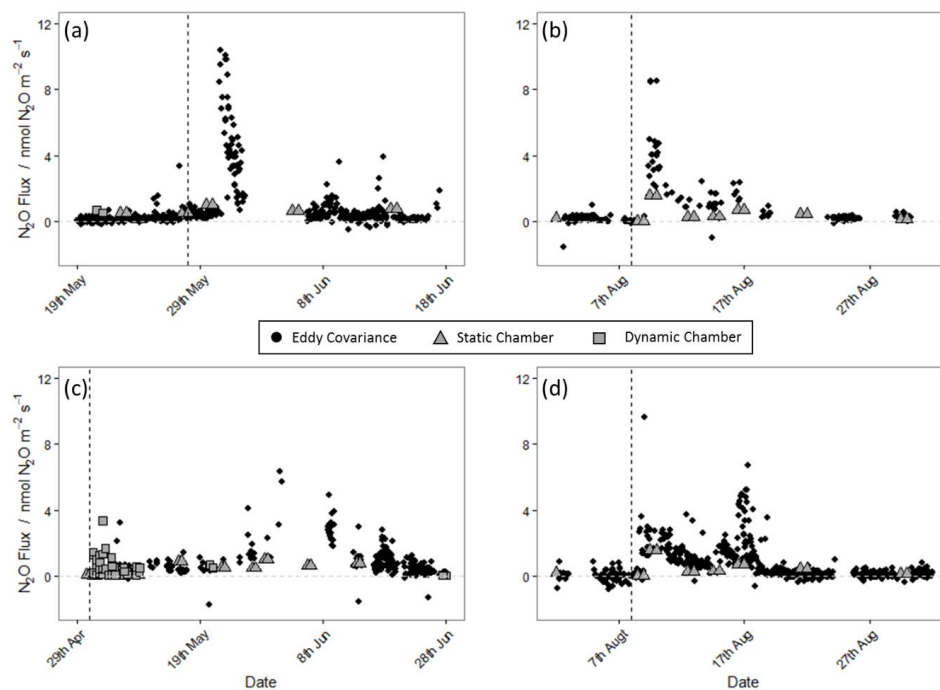
256 Fertiliser was applied to the un-tilled field on the 28<sup>th</sup> of May and to both fields on 9<sup>th</sup> August. Tillage began on

257 1<sup>st</sup> May. The Y-axis is limited to  $12 \text{ nmol m}^{-2} \text{ s}^{-1}$  for better comparison between the fields. Only four static chamber

258 measurements in the north field recorded fluxes above  $12 \text{ nmol m}^{-2} \text{ s}^{-1}$  in the first few days after the August

259 fertilisation.

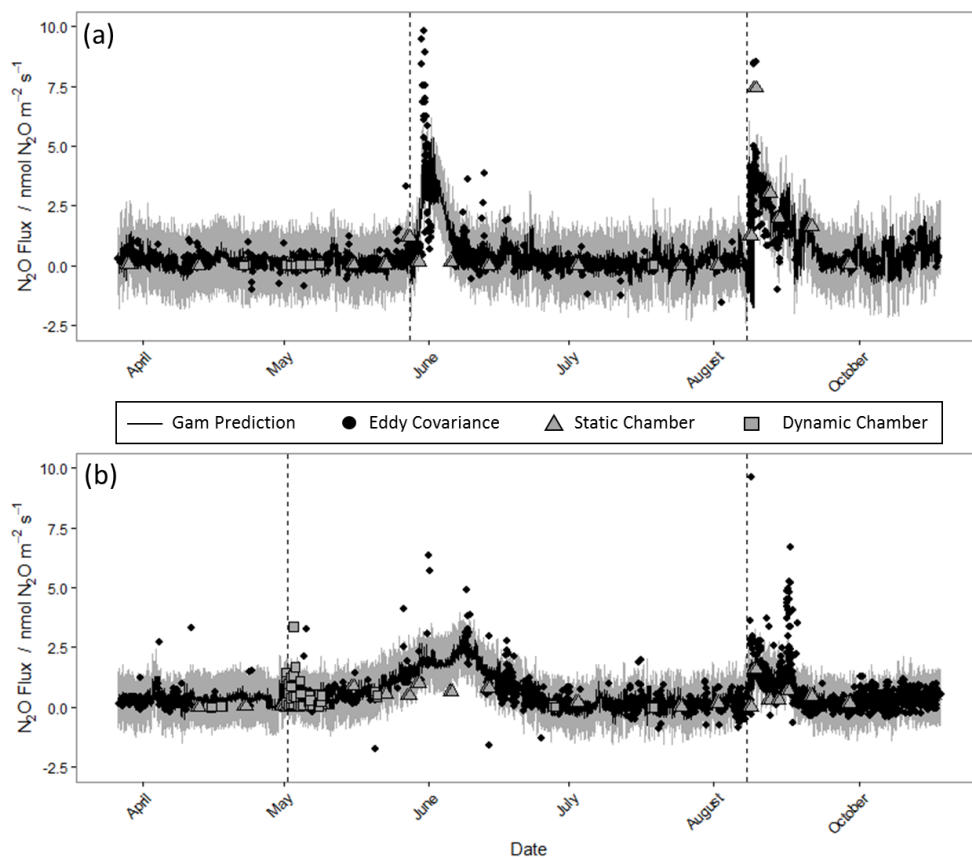
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262 **Figure 5** Elevated fluxes of N<sub>2</sub>O were measured from the un-tilled field after (a) a 70 kg-N ha<sup>-1</sup> application of  
263 Nitram on the 28<sup>th</sup> of May and (b) a second 70 kg-N ha<sup>-1</sup> application of Nitram on the 9<sup>th</sup> of August. Elevated  
264 fluxes of N<sub>2</sub>O were measured from the tilled field (c) immediately after the tillage event on the 1<sup>st</sup> of May which  
265 remained above pre-tillage magnitude until the end of June. Elevated fluxes of N<sub>2</sub>O were also observed from the  
266 tilled field (d) after a 70 kg-N ha<sup>-1</sup> application of Nitram on the 9<sup>th</sup> of August.

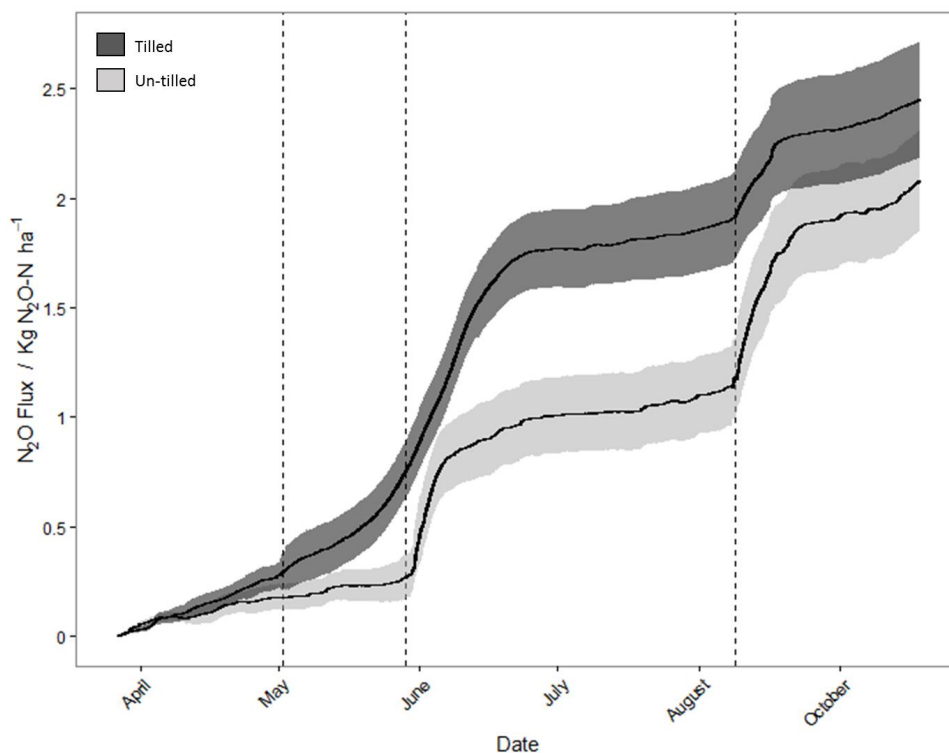
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269 **Figure 6** The GAM method provides an estimated N<sub>2</sub>O flux which can be used to gap fill measurements from  
270 both the (a) un-tilled and (b) tilled fields at 30 min intervals. The 95 % confidence interval in the estimated flux  
271 reported by the GAM is included (grey). Tillage and fertiliser dates are indicated (vertical lines)

272



273

274 **Figure 7** Cumulative flux is calculated for the tilled (dark) and un-tilled fields (light) using the gap filled flux  
275 data. The propagated 95 % confidence intervals estimated using the sum of least squares method (grey areas).  
276 Fertiliser was applied to the un-tilled field on the 28<sup>th</sup> of May and to both fields on the 9<sup>th</sup> of August and tillage  
277 occurred on the 1<sup>st</sup> of May (black dashed vertical lines).

### 278 3.3 Biomass measurements

279 Biomass measurements made from the tilled field before tillage estimated that there was an average of  $369 \pm 310$   
280  $\text{g m}^{-2}$  of grass materials (dried weight) growing across the field with a root to shoot ratio of  $\sim 1.5$ . The elemental  
281 analysis of the dry grass materials measured an average total carbon content of 45.7 % and nitrogen content of 2.5  
282 %. Based on these measurements it is estimated that the tillage event added a total of  $93.6 \text{ kg ha}^{-1}$  of nitrogen to  
283 the field in the form of crop residues.

284



285    **4       Discussion**

286    **4.1      The influence of tillage on N<sub>2</sub>O fluxes**

287    The comparison of pre-tillage and post-tillage fluxes from the tilled field suggests that the tillage event was  
288    directly responsible for an immediate increase in N<sub>2</sub>O fluxes (Figure 4b & 5c). N<sub>2</sub>O fluxes significantly larger  
289    than those measured pre-tillage were observed from the tilled field over two separate periods during which no  
290    changes in N<sub>2</sub>O fluxes were observed in the adjacent un-tilled field. The initial increase in N<sub>2</sub>O flux from the tilled  
291    field occurs directly after the disturbance of the soil caused by ploughing and harrowing. In the two month period  
292    in which fluxes from the tilled field were elevated, a total of  $1.47 \pm 0.16$  kg N<sub>2</sub>O-N ha<sup>-1</sup> was estimated to have  
293    been released. Assuming fluxes in the tilled field had remained at approximately pre-tillage magnitude ( $\sim 0.27$   
294    nmol m<sup>-2</sup> s<sup>-1</sup>) had the tillage event not taken place, it can be concluded that the tillage event contributed to an  
295    additional  $1.08 \pm 0.14$  kg N<sub>2</sub>O-N ha<sup>-1</sup> emitted from the field over a two month period.

296           Increases in N<sub>2</sub>O flux lasting up to two months after grassland tillage events have been observed before  
297    in other studies using both static chamber and eddy covariance measurements (Chatskikh and Olesen, 2007;  
298    Merbold et al., 2014). Reported fluxes can be relatively high over a sustained period of time (several days or  
299    weeks) and similar in magnitude to those recorded after fertilisation events. The mechanisms driving these large  
300    sustained fluxes are believed to be partly due to the mineralisation of organic materials in the soils (decaying grass  
301    materials from the previous sward in tilled grasslands) (Baggs et al., 2003; Hellebrand, 1998; Pimentel et al.,  
302    2015). The large quantities of decaying organic matter ploughed into the soils would have provided a gradual  
303    release of carbon and nitrogen into the soils, which provide substrate for the microbial processes of nitrification  
304    and denitrification (Pimentel et al., 2015; Seastedt et al., 1992). According to IPCC estimates, 1 % of N added to  
305    soils in the form of crop residues can be expected to be released as N<sub>2</sub>O (IPCC, 2006). Based on our pre-tillage  
306    biomass measurements (93.6 kg N ha<sup>-1</sup>) if these estimates were true we would expect to see N<sub>2</sub>O fluxes of  
307    approximately 0.94 kg N<sub>2</sub>O-N ha<sup>-1</sup> from the field. This estimated value is within the range of uncertainty of our  
308    calculated cumulative fluxes in this study ( $1.08 \pm 0.14$  kg N<sub>2</sub>O-N ha<sup>-1</sup>). High emissions from crop residues tilled  
309    into arable crops have been recorded in similar wet soils with high clay content (Ball, 1999) which may indicate  
310    a similar process is occurring under these conditions at other field sites in the area.

311           Large N<sub>2</sub>O fluxes (> 0.5 nmol m<sup>-2</sup> s<sup>-1</sup>) are observed from both fields after fertilisation events. Elevated  
312    fluxes recorded from the fields after fertilisation typically last three to four weeks with an occasional large spike





313 lasting 24 to 48 hours before returning to pre-fertilisation levels. This month long period in which the majority of  
314 large fluxes occur after fertilisation is also generally observed by other similar studies from the local area (Skiba  
315 et al., 2013; Smith et al., 2012). Assuming the majority of N<sub>2</sub>O emitted after a fertilisation event occurs within a  
316 28 day period after the fertiliser application, the 28 day cumulative flux emissions associated with the fertilisation  
317 events on the 28<sup>th</sup> of May and 9<sup>th</sup> of August on the un-tilled field were  $0.72 \pm 0.14$  and  $0.75 \pm 0.14$  kg N<sub>2</sub>O-N ha<sup>-1</sup>  
318 <sup>1</sup>, respectively. The 28 day cumulative flux emissions associated with the fertilisation event on the tilled field was  
319  $0.42 \pm 0.15$  kg N<sub>2</sub>O-N ha<sup>-1</sup>. As each fertilisation event consisted of the same application rate of 70 kg N ha<sup>-1</sup> of  
320 Nitram pellets, these cumulative fluxes account for a 1.03, 1.07 and 0.60 % of the total applied nitrogen in each  
321 case. Assuming the 28 day periods account well for the emission factors of the fertiliser events, these results agree  
322 well with the generic 1 % value reported by the IPCC for N fertiliser events (IPCC, 2014).

323         The reason for the large difference in fluxes associated with the fertiliser events between the two fields  
324 in this study is unknown. As the weather was the same for both fields it is assumed that temperature and rainfall  
325 was not a factor. After sward renewal on the tilled field, the grass grew back very thick and appeared much  
326 healthier than the more established grass in the un-tilled field. One theory is that the healthier, more productive  
327 grass in the tilled field consumed more of the available fertiliser than in the un-tilled field, leaving less N available  
328 for N<sub>2</sub>O producing microbial processes. Another possible explanation is that physical changes to the soil since  
329 tillage (such as bulk density, WFPS and soil organic carbon content) altered the microbial processes in a way that  
330 reduced N<sub>2</sub>O emissions from the fertiliser event (Ball et al., 2008; Choudhary et al., 2002; Davidson et al., 2000;  
331 Turner et al., 2008). From the limited soil data available, it is not possible to say for certain why fluxes from the  
332 tilled field in August were lower than those recorded in the adjacent field. As the driving forces of N<sub>2</sub>O emissions  
333 from agricultural soils are not understood well in general, it complicates our ability to comprehend what is  
334 happening at the microbial level.

#### 335     **4.2     Gap filling of N<sub>2</sub>O fluxes**

336 Gap-filling N<sub>2</sub>O flux measurements is difficult due to the lack of reliable process-based models on which to base  
337 predictions. N<sub>2</sub>O fluxes are believed to be driven primarily by the availability of nitrogen compounds in the soils  
338 (ammonium and nitrate) (Davidson et al., 2000) as well as physical properties of the soil such as WFPS, aerobic  
339 extent, soil type, temperature and compaction (Ball et al., 2008; Choudhary et al., 2002; Davidson et al., 2000;  
340 Turner et al., 2008). The collection of these data on a temporal/spatial scale which would allow these models to  
341 be applied is not often logistically possible or affordable. The GAM method used in this study incorporates



342 readily-available meteorological data with the temporal pattern in the data, to provide an empirical but practical  
343 means of temporal interpolation, which makes use of more information than simple linear interpolation. Although  
344 the GAM method has proved useful, we would also emphasise the dangers of extrapolating to conditions beyond  
345 those to which the model was fitted. For example, as we have not measured fluxes during the cold months in  
346 winter, the GAM is unable to reliably predict fluxes in temperatures lower than those measured during the study.  
347 A two week gap in measurements in early June when the QCL instrument was out of commission has left a large  
348 gap in data, which makes a considerable contribution to the total uncertainty in the cumulative flux following the  
349 tillage event.

350 In this study spatial variability was not explicitly accounted for in the cumulative flux uncertainty and  
351 this remains a potentially large error. Eddy covariance is able to integrate over a large area of the field (several  
352 100 m<sup>2</sup>) but these measurements are still subject to an element of spatial variability which is unaccounted for. Any  
353 study which plans to report cumulative flux estimates should consider how to minimise the uncertainties which  
354 arise when interpolating and/or extrapolating measurements to larger temporal and spatial scales (e.g. from  
355 occasional chamber measurements to annual field-scale emissions). Further studies may require more complex  
356 statistical analysis of uncertainties using methods such as Bayesian statistics to improve uncertainty estimates in  
357 methodology.

358

## 359 5 Conclusion

360 Total cumulative fluxes calculated for the tilled and un-tilled fields over a 175 day period were  $2.45 \pm 0.27$  and  
361  $2.08 \pm 0.23$  kg N<sub>2</sub>O-N ha<sup>-1</sup>, respectively. N<sub>2</sub>O emissions after tillage were relatively large and sustained, similar  
362 in magnitude to a nitrogen fertilisation event. The tillage event is estimated to be responsible for emissions of  $1.08$   
363  $\pm 0.14$  kg N<sub>2</sub>O-N ha<sup>-1</sup> over a two month period after tillage. Further differences in N<sub>2</sub>O fluxes were observed  
364 between the tilled and un-tilled fields after a subsequent nitrogen fertilisation in August 2012. Cumulative fluxes  
365 of N<sub>2</sub>O estimated for a four week period after fertilisation for the tilled and un-tilled fields were  $0.42 \pm 0.15$  and  
366  $0.75 \pm 0.14$  kg N<sub>2</sub>O-N ha<sup>-1</sup>, respectively. It is uncertain whether the tillage event or other factors were the reason  
367 for lower emissions from the tilled field after the fertilisation event. The results reported in this study agrees with  
368 several other studies that nitrogen added to soils in the form of crop residues may contribute to high emissions of  
369 N<sub>2</sub>O similar to those expected from fertiliser events. This observation highlights a potentially large un-quantified  
370 and poorly understood source of anthropogenic N<sub>2</sub>O emissions at a global scale. The study also highlights the



371 need for more detailed investigation of the microbiological processes in soils to describe processes driving  
372 emissions from tilled soils and crop residues which also remain poorly understood.

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