

1 **Nitrous oxide emissions after incorporation of winter oilseed rape (*Brassica napus* L.)**  
2 **residues under two different tillage treatments**

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21 **Abstract**

22 The aim of this study was to investigate the effect of crop residues from winter oilseed rape on  
23 N<sub>2</sub>O emissions from a loamy soil, and to determine the effect of different tillage practices on  
24 N<sub>2</sub>O fluxes. We therefore conducted a field experiment where crop residues of winter oil seed  
25 rape (*Brassica napus* L., OSR) were replaced with <sup>15</sup>N labelled OSR residues. Nitrous oxide  
26 (N<sub>2</sub>O) emissions and <sup>15</sup>N abundance in the N<sub>2</sub>O were determined for a period of 11 months after  
27 harvest of OSR and in the succeeding crop winter wheat (*Triticum aestivum* L.) cultivated on a  
28 Haplic Luvisol in South Germany. Measurements were carried out with the closed chamber  
29 method in a treatment with conventional tillage (CT) and in a treatment with reduced soil tillage  
30 (RT). In both tillage treatments we also determined N<sub>2</sub>O fluxes in control plots where we  
31 completely removed the crop residues.

32 High N<sub>2</sub>O fluxes occurred in a short period just after OSR residue replacement in fall and after  
33 N-fertilization to winter wheat in the following spring. Although N<sub>2</sub>O emissions differed for  
34 distinct treatments and sub-periods, cumulative N<sub>2</sub>O emissions over the whole investigation  
35 period (299 days) ranged between 1.7 kg and 2.4 kg N<sub>2</sub>O-N ha<sup>-1</sup> with no significant treatment  
36 effects. More than half of the cumulative emissions occurred during the first eight weeks after  
37 OSR replacement highlighting the importance of this post-harvest period for annual N<sub>2</sub>O  
38 budgets of OSR.

39 The contribution of residue N to the N<sub>2</sub>O emission was low and explained by the high C/N-  
40 ratio fostering immobilization of mineral N. In total only 0.03 % of the N<sub>2</sub>O-N emitted in the  
41 conventional tillage treatment and 0.06 % in the reduced tillage treatment stemmed directly  
42 from the crop residues. The <sup>15</sup>N recovery in the treatments with crop residues was 62.8 % (CT)  
43 and 75.1 % (RT) with more than 97 % of the recovered <sup>15</sup>N in the top soil.

44 Despite our measurements did not cover an entire year, the low contribution of the OSR residues  
45 to the direct N<sub>2</sub>O emissions shows, that the current IPCC tier 1 approach, which assumes an EF

46 of 1.00 %, strongly overestimated direct emissions from OSR crop residues. Furthermore, we  
47 could not observe any relationship between tillage and crop residues on N<sub>2</sub>O emission, only  
48 during the winter period were N<sub>2</sub>O emissions from reduced tillage significantly higher  
49 compared to conventional tillage. Annual N<sub>2</sub>O emission from RT and CT did not differ.

50

## 51 1 Introduction

52 In 2014 the acreage of winter oilseed rape (OSR) in the European Union (EU) was  $6.7 \times 10^6$  ha  
53 with an average yield of  $3.6 \text{ Mg ha}^{-1}$  (FAOSTAT, 2017). The production of OSR in the EU  
54 increased between 1993 and 2014 by 43.1 % (FAOSTAT, 2017). This development was mainly  
55 a result of the higher demand for biodiesel due to the Renewable Energy Directive (RED, 2009),  
56 implying that a share of 10 % of transport energy need is made by biofuels. Rapeseed oil is the  
57 most common feedstock for biodiesel, which is the major biofuel in Europe (Hamelinck et al.,  
58 2012). Cultivation of oilseed rape (including production of fertilizers and agro-chemicals,  
59 transportation of the feedstock to a biofuels production plant and the use of fertilizers) as  
60 feedstock for biodiesel production in Europe contributes between 75 and 86 % of total GHG  
61 emissions ( $66.7\text{-}119.5 \text{ g CO}_2 \text{ MJ}_{\text{fuel}}^{-1}$ ) (Hoefnagels et al., 2010).

62 N<sub>2</sub>O is a climate-relevant trace gas and it is also involved in stratospheric ozone depletion  
63 (Granli & Bøckman, 1994; Crutzen, 1981; Sagggar et al., 2004). N<sub>2</sub>O has a 298 times higher  
64 specific heat adsorption potential when compared to the same mass of carbon dioxide (CO<sub>2</sub>)  
65 (Mhyre et al., 2013). In 2015, the atmospheric N<sub>2</sub>O concentration was 328 ppb and about 21 %  
66 higher than in the pre-industrial era (WMO, 2016). More than 50 % of the anthropogenic N<sub>2</sub>O  
67 is emitted from agricultural soils (Clais et al., 2013). The main source for N<sub>2</sub>O from agricultural  
68 soils is microbial denitrification in soil compartments with low oxygen availability.  
69 Nitrification, the microbial oxidation of ammonia to nitrate is a further N<sub>2</sub>O source with a

70 probably lower contribution to N<sub>2</sub>O emissions from agricultural soils when compared to  
71 denitrification (*Flessa et al.*, 1996). Apart from these two processes, the share of further N  
72 transformation processes in soils is currently under discussion (i.e. *Shaw et al.*, 2006;  
73 *Butterbach-Bahl et al.*, 2013). Since all these processes rely on mineral N as substrate, N-input  
74 (N fertilization or N in crop residues) generally increases N<sub>2</sub>O emission from soils (*Stehfest &*  
75 *Bouwman*, 2006). Several studies indicated a strong correlation between the mineral N contents  
76 of the top soil or the N surpluses with the N<sub>2</sub>O emissions from arable fields (*Kaiser & Ruser*,  
77 2000; *Van Groenigen et al.*, 2010).

78 Oilseed rape is known for its high N demand during early growth stages and a low N removal  
79 with the seeds resulting in high N surplus which are susceptible to gaseous or leaching losses  
80 into the environment (*Rathke et al.*, 2006). N uptake by OSR plants ends early (*Malagoli et al.*,  
81 2005) whereas mineralization of organic soil N proceeds. As a result, mineral N under OSR is  
82 generally high in the harvest period (*Christen & Fried*, 2011).

83 Leaving crop residues in the field has many positive environmental effects such as nutrient  
84 transfer over winter, carbon sequestration, and reduction of soil erosion (*Chen et al.*, 2013). On  
85 the other hand, adverse effects as i.e. increased N<sub>2</sub>O emissions after incorporation of crop  
86 residues were reported (*Baggs et al.*, 2000; *Chen et al.*, 2013). *Moiser et al.* (1998) estimated a  
87 global production of 0.4 million tons of N<sub>2</sub>O-N yr<sup>-1</sup> from crop residues.

88 Winter N<sub>2</sub>O emissions can account for 50 % of the annual N<sub>2</sub>O emissions if distinct frost/thaw  
89 cycles occur during this period (*Kaiser & Ruser*, 2000; *Jungkunst et al.*, 2006). *Kaiser et al.*  
90 (1998) showed that N<sub>2</sub>O emissions during the winter season decreased with increasing C/N-  
91 ratio of crop residues.

92 On the one hand, high C/N-ratios (well above 30) can lead to a short-term immobilization of  
93 mineral N and thus reduce the substrate availability for N<sub>2</sub>O production. On the other hand,

94 crop residues release easily available C. The turn-over of this C and the associated oxygen (O<sub>2</sub>)  
95 consumption can lead to anaerobic conditions favoring denitrification and thus stimulating N<sub>2</sub>O  
96 release (*Flessa & Beese*, 1995). As shown by *Ruser et al.* (2017) NO<sub>3</sub> concentrations in the top  
97 soil after OSR harvest slightly decreased but they were obviously still sufficient to enhance  
98 N<sub>2</sub>O emissions in the post-harvest season at five study sites in Germany. Due to the incomplete  
99 immobilization of mineral N it can therefore not generally be assumed that OSR crop residues  
100 with a high C/N-ratio reduce N<sub>2</sub>O emission. In their meta-analysis, *Chen et al.* (2013) reported  
101 slightly positive effects for C/N- ratios between 45 and 100 on N<sub>2</sub>O emission from agricultural  
102 soils. Even the application of *Miscanthus x giganteus* residues with C/N-ratio of 297 induced  
103 N<sub>2</sub>O emissions which were significantly higher when compared to a control without crop  
104 residues. In all these studies, increased N<sub>2</sub>O emissions induced by crop residues with a high  
105 C/N ratio were explained with the formation of anaerobic microsites as a result of the short-  
106 term availability of easily decomposable C during the decomposition of crop residues thus  
107 favoring denitrification and N<sub>2</sub>O release (*Li et al.*, 2013).

108 The IPCC's (2006) methodology assumes that 1 % of the residue-N is emitted into the  
109 atmosphere as direct N<sub>2</sub>O emission within the first year after application. Several investigations  
110 reported emission factors (EFs) for crop residues between 0.62 and 2.8 % (*Kaiser et al.*, 1998;  
111 *Harrison et al.*, 2002; *Millar et al.*, 2004; *Vinther et al.*, 2004; *Novoa & Tejada*, 2006). The high  
112 variability of EFs reported for crop residues was the reason why *Delgado et al.* (2010) suggested  
113 adjusting the EF according to the C/N ratio.

114 Reduced tillage (RT) is defined as abstaining from ploughing, i.e. tillage practise without soil  
115 inversion (*Townsend et al.*, 2016). Reduced soil disturbances in combination with crop residue  
116 retention in the field was shown to be efficient in increasing organic C and N stocks in the  
117 uppermost soil layer (*Al-Sheikh et al.*, 2005; *Ghimire et al.*, 2012), soil erosion control and water  
118 conservation (*Krauss et al.*, 2017). Results of studies on the effect of tillage on N<sub>2</sub>O emissions

119 are contradictory. When compared to conventional tillage, RT reduced N<sub>2</sub>O emissions (i.e.  
120 *Koga, 2013; Wang & Dalal, 2015*), resulted in similar N<sub>2</sub>O emissions (i.e. *Abdalla et al., 2013;*  
121 *Negassa et al., 2015*) or, which was the majority of experiments, increased emission (i.e. *Baggs*  
122 *et al., 2000; Venterea et al., 2005*). *Lognoul et al. (2017)* reported 10 times higher emissions  
123 under a seven-year-old reduced tillage system than under conventional tillage. This result was  
124 attributed to higher total N and SOC contents, and a larger microbial biomass in the uppermost  
125 soil layer, caused by limited digging and mixing of crop residues under RT. Conditions such as  
126 increased soil moisture and availability of organic C compounds as energy supplier favour  
127 denitrification and they are therefore crucial for N<sub>2</sub>O production and release from soils under  
128 RT. Similarly, the reason for less aeration inside RT soil could be the presence of higher water-  
129 filled porosity thus lowering O<sub>2</sub> diffusion into the soil (*Linn & Doran, 1984*).

130 The most common tillage method for OSR in Germany is ploughing followed by harrowing but  
131 direct drilling is also applied (*Rathke et al., 2006*). Particularly in dry summers, the latter  
132 practice has the advantage of conserving soil moisture and therefore it is becoming more  
133 popular (*Rathke et al., 2006*). OSR yield seems not to be affected by RT (*Bonari et al., 1995;*  
134 *Christen et al., 2003*).

135 The effect of OSR crops residues on N<sub>2</sub>O emissions during the post-harvest period have hardly  
136 been investigated, therefore the main hypotheses of this study were: (1) as a result of incomplete  
137 immobilization of mineral N after harvest, N<sub>2</sub>O emissions are stimulated through OSR crop  
138 residues which provide easily available C, which thus favor anaerobic conditions, (2)  
139 nevertheless, due to the high C/N-ratio of OSR residues, the emission factor derived from <sup>15</sup>N  
140 labelling technique of crop residues is lower than the IPCC default of 1 %, and (3) N<sub>2</sub>O fluxes  
141 from reduced tillage system are higher than from the conventional system after OSR crop  
142 residue application.

## 143 2 Material & Methods

### 144 2.1 Study site

145 The field site was located on the research station Ihinger Hof (University of Hohenheim), South  
146 Germany (48°44'40.7"N, 8°55'26.4"E). The station is located 478 m above sea level, the mean  
147 annual temperature is 8.3 °C and the long-term annual precipitation is 738 mm. The soil of the  
148 study site was classified as Luvisol (Table 1) with a high silt content. The experiment was  
149 conducted from the end of July 2014 to June 2015. Before the trial, the site was used as arable  
150 land with OSR (var. Visby) as preceding crop, fertilized with 180 kg N ha<sup>-1</sup>. In October 2014,  
151 winter wheat (*Triticum aestivum* L., var. Julius) was sown as a succeeding crop and fertilized  
152 with calcium ammonium nitrate in 3 doses (30, 80, and 55 kg N ha<sup>-1</sup> at BBCH stages 24, 32,  
153 and 61 resp. inflorescence emergence, *Meier*, 2001) in spring to summer 2015.

154 ((Table 1))

### 155 2.2 Preparation of <sup>15</sup>N labeled crop residues

156 To label OSR residues with <sup>15</sup>N, young plants (BBCH 3) were transplanted from field to pots  
157 (volume: 80 l, surface area 0.4 m<sup>2</sup>) filled with a mixture of clayey loam and sand (6:4 w:w) and  
158 equipped with a closed water circulation system to avoid leaching losses. We chose the same  
159 planting density as under field conditions (35 plants m<sup>-2</sup>, corresponding to 14 plants per pot).  
160 <sup>15</sup>N enriched potassium nitrate (KNO<sub>3</sub>) with 60 atom % <sup>15</sup>N was used for fertilization. The N  
161 amount used for labeling (180 kg N ha<sup>-1</sup>, corresponding to 6.8 g N per pot based on the pot  
162 surface) also followed field conditions. In order to avoid sulphur (S) deficiency, 90 kg S ha<sup>-1</sup>  
163 (corresponding to 3.4 g S per pot based on the pot surface) was applied as Kieserite (MgSO<sub>4</sub> ·  
164 H<sub>2</sub>O). Plants were harvested 15 weeks after transplanting (BBCH 85). The plants were bulked  
165 and separated into pod, litter, stem, grain and (washed) roots before oven drying at 60°C. After  
166 drying, the different plant parts were roughly cut into 2-6 cm pieces. Separately, aliquots of

167 each part were ground finely to determine the C- and N-concentration, and the  $^{15}\text{N}$  abundance  
168 with an isotope ratio mass spectrometer (IRMS, Delta C; Finnigan MAT, Germany) coupled  
169 with an elemental analyzer (EA 1108; Fisons Instrument, Italy). The final average  $^{15}\text{N}$   
170 abundance of the crop residues was 14.4 atom% with a C/N-ratio of 51.7. The share of each  
171 plant part and the  $^{15}\text{N}$  abundance in each part as well as in the mixture applied to the field is  
172 shown in Table 2.

173 ((Table 2))

### 174 2.3 Field preparation

175 The experiment was conducted by inserting into an existing tillage trial (split-plot design, four  
176 blocks). Two tillage treatments were applied: ploughed (CT) and reduced (RT). Ploughing was  
177 done using a mouldboard plough to a depth of 30 cm. Reduced tillage was done using a chisel  
178 plough to a depth of 15 cm. The tillage experiment was initiated in 2012. After harvest of OSR  
179 in 2014, two mini plots were placed in each plot of the tillage experiment: one plot without (-  
180 CR) and one plot with (+CR)  $^{15}\text{N}$ -labeled OSR residues. The size of each mini plot was 0.6 m  
181 x 0.5 m. The mini plot area was cleared and free of stubble, and roots of the OSR were removed  
182 and replaced against  $^{15}\text{N}$  labelled roots. Proper isolation of the residue treated mini plots was  
183 maintained by the use of metallic plates. Base-rings of closed chambers were inserted approx.  
184 10 cm deep in the center of each mini plot. Finally, the residual  $^{15}\text{N}$ -labelled crop residues (stem,  
185 pod, litter, seeds) were mulched and evenly distributed on the surface of the mini plots (total  
186 crop residues  $455 \text{ g plot}^{-1}$  corresponding to  $135 \text{ kg N ha}^{-1}$ ). Simultaneously with the remaining  
187 field, tillage measures were carried out at the end of September. To avoid mechanical carriage  
188 of the  $^{15}\text{N}$  labeled material by tillage machinery, tillage in the mini plots was simulated  
189 manually.

190



191 2.4 Determination of trace gas fluxes

192 The N<sub>2</sub>O flux measurements were conducted using the closed chamber method (*Hutchinson &*  
193 *Mosier*, 1981). Fluxes were determined weekly in the morning with additional event driven  
194 samplings after N-fertilization, strong rainfall and during thawing of frozen soil. As shown by  
195 *Flessa et al.* (2002), this sampling strategy significantly reduces the error of a weekly sampling  
196 scheme with an error of approximately 10 % when compared to high resolution measurements.  
197 The circular, dark vented chambers had an inner diameter of 30 cm and were described in detail  
198 by *Flessa et al.* (1995). During the closure period of 45 minutes, we periodically took four gas  
199 samples out of the chambers' atmosphere using a syringe and transferred the gas sample into  
200 evacuated glass vials (22.5 ml). In order to determine the <sup>15</sup>N-N<sub>2</sub>O abundance, we took two  
201 further samples, after 0 (t<sub>n</sub>) and 45 minutes (t<sub>n+1</sub>).

202 N<sub>2</sub>O and CO<sub>2</sub> concentrations in the gas samples were measured with a greenhouse gas analyzer  
203 equipped with a <sup>63</sup>Ni electron capture detector (ECD) (Scion 450-GC, Bruker) connected to an  
204 autosampler (GX-281, Gilson). The software package Compass CDS (Bruker, 2012) was used  
205 to calculate trace gas concentrations. For a consortium of gas analyzing laboratories including  
206 our lab, *Ruser et al.* (2017) calculated a flux detection limit lower than 22 µg N<sub>2</sub>O-N m<sup>-2</sup> h<sup>-1</sup> for  
207 90 % of the flux measurements.

208 <sup>15</sup>N-N<sub>2</sub>O was determined with an Isotope Ratio Mass Spectrometer (IRMS) delta plus (Finnigan  
209 MAT, Bremen, Germany) coupled with a fully automated PreCon-Interface for preparing the  
210 N<sub>2</sub>O from the air sample (*Brand*, 1995).

211 As described by *Leiber-Sauheitl et al.* (2013) in more detail, N<sub>2</sub>O fluxes were calculated with  
212 the HMR package (*Pedersen*, 2012) adjusted by a script, created by *R. Fuß*. The script selects  
213 automatically the most suitable model for each flux being either a robust linear or a non-linear  
214 (HMR) model.

215 Cumulative emissions of N<sub>2</sub>O were integrated per chamber with:

216 Equation 1:

217 
$$\text{Cumulative N}_2\text{O-N} = \sum_i^n \frac{(t_{i+1}-t_i) \times (f_i + f_{i+1})}{2}$$

218 with cumulative N<sub>2</sub>O-N in kg ha<sup>-1</sup>, t = sampling time [h] and f = gas flux [kg ha<sup>-1</sup> h<sup>-1</sup>].

219 The δ values were calculated with following equation recommended by *Tilsner et al. (2003)*:

220 Equation 2:

221 
$$\delta x_{\text{emitted}} = \frac{\delta x_{t_{n+1}} \times c(\text{N}_2\text{O})_{t_{n+1}} - \delta x_{t_n} \times c(\text{N}_2\text{O})_{t_n}}{c(\text{N}_2\text{O})_{t_{n+1}} - c(\text{N}_2\text{O})_{t_n}}$$

222 with δx = is the value of the heavy isotope x (‰) and c = concentration of N<sub>2</sub>O (ppm).

223 N input related emission factor (NEF) were calculated as

224 Equation 3:

225 
$$\text{NEF} = \frac{\text{N}_2\text{O}_{\text{treatment}}}{\text{N}_{\text{input}}} \times 100$$

226 with NEF in % and cumulative N<sub>2</sub>O-N emissions and N-inputs (crop residue and fertilizer N)

227 in kg ha<sup>-1</sup>.

228

229 Crop residue related emission factors (EF<sub>CR</sub>) were calculated as

230 Equation 4:

231 
$$\text{EF}_{\text{CR}} = \frac{\text{N}_2\text{O}_{\text{CR}}}{\text{N}_{\text{input}}} \times 100$$

232 with  $EF_{CR}$  in % and cumulative  $N_2O$ -N emissions emitted by crop residues and N-inputs (crop  
233 residue and fertilizer N) in  $kg\ ha^{-1}$ .

## 234 2.5 Soil sampling, laboratory analyses and weather conditions

235 Soil samples were taken four times during the experimental period to determine the mineral  
236 nitrogen content ( $N_{min}$ ). Due to the small area of the mini plots, a more frequent sampling design  
237 was not possible. In the CT plots samples were taken from 0-30 cm depth, in the RT plots we  
238 sampled soil from 0-15 cm and from 15-30 cm. For  $N_{min}$  analysis, 20 g of fresh soil was  
239 extracted with 80 ml of 0.0125 M  $CaCl_2$  solution. The concentrations of  $NH_4^+$ -N and  $NO_3^-$ -N  
240 were quantified using a fully automated flow injection analyzer (FIAstar 5000, FOSS,  
241 Denmark).

242

243 Soil moisture was determined gravimetrically by drying an aliquot of fresh soil at  $105^\circ C$  for 24  
244 h. The calculation of the water-filled pore space (WFPS) was explained in detail by *Ruser et al.*  
245 (1998). In addition, the volumetric water content of the soil was measured in the field  
246 simultaneously to the gas measurement in every plot using a mobile soil water monitoring probe  
247 (EasyTest FOM/mts, Lublin, Poland). These measurements were carried out as long as the soil  
248 was not too dry or frozen thus impeding the use of the probe.

249  $^{15}N$  abundance in the  $NH_4^+$  and  $NO_3^-$  pool was determined using a diffusion procedure  
250 according to *Brooks et al.* (1989). For the diffusion method, the  $CaCl_2$  extract from  $N_{min}$  analysis  
251 was used. Isotope Ratio Mass Spectrometer (delta plus, Finnigan MAT, Bremen) coupled with  
252 a CN elemental analyzer (Euro EA, Eurovector, Milano, Italy) was used to determine  $^{15}N$   
253 abundance in  $N_{min}$  fractions.

254 Air temperature and precipitation were provided by the climate station of the research station.

## 255 2.6 Plant sampling

256 At the end of the experiment, aboveground wheat biomass on the mini plots was harvested on  
257 May 26<sup>th</sup> 2015. Plants were dried at 60°C for 48 h. An aliquot was ground and C- and N-  
258 contents were determined with a CN elemental analyzer (VarioMax, Elementar  
259 Analysensysteme GmbH). A further aliquot was used to measure the <sup>15</sup>N enrichment of the  
260 wheat.

261 The percentage of recovery of <sup>15</sup>N in soil or plant was calculated according to *Hauck and*  
262 *Bremner* (1976).

263 Equation 5:

$$264 \quad {}^{15}\text{N}_{\text{RCE}} = 100 \times \frac{p \times (c-b)}{f \times (a-b)}$$

265 Where p is the total N in soil or plant (kg N ha<sup>-1</sup>), f the total amount of <sup>15</sup>N applied with the  
266 crop residues, a the <sup>15</sup>N abundance in the crop residues [atom%], b the <sup>15</sup>N abundance in the  
267 treatments without crop residues (unlabeled) and, c the <sup>15</sup>N abundance in soil or plant samples.

268

## 269 2.7 Statistical analyses and further calculations

270 Cumulative N<sub>2</sub>O emissions were divided in three periods: post-harvest period from harvest to  
271 tillage (31.07.14 – 01.10.14), winter period (02.10.14 - 02.03.15), and vegetation period  
272 (03.03.15 – 29.05.15). For comparison of the cumulative N<sub>2</sub>O emissions, an ANOVA was  
273 performed using the PROC MIXED procedure by SAS 9.4 (SAS Institute, 2016). For N<sub>2</sub>O  
274 emissions over time, repeated measures of ANOVA were performed using the PROC MIXED  
275 procedure, with an autoregressive AR(1) covariance structure to acknowledge for proximate  
276 correlation.

277 The models for N<sub>2</sub>O were as follows:

278 (1)  $\mu_{ij} = \mu + \alpha_i + \beta_{j1} + (\alpha \times \beta)_{ij} + (\gamma_{ij1} \times \beta_j) + \delta_{ij}$

279 (2)  $\mu_{ij} = \mu + \alpha_i + \beta_{j2} + (\alpha \times \beta)_{ij} + (\gamma_{ij2} \times \beta_j) + \delta_{ij}$

280 with  $\mu$ = general effect,  $\alpha$ = CR (crop residues),  $\beta$ = tillage,  $\gamma$ = WFPS, and  $\delta$ = CO<sub>2</sub>

281 WFPS and CO<sub>2</sub> were entered as covariants. CO<sub>2</sub> x CR and CO<sub>2</sub> x tillage were not significant

282 and removed from the model.

283 The model for CO<sub>2</sub> was as follows:

284  $\mu_{ij} = \mu + \alpha_i + \beta_j + \delta_{ij}$

285 with  $\mu$ = general effect,  $\alpha$ = CR (crop residues),  $\beta$ = tillage,  $\delta$ = CO<sub>2</sub>

286 WFPS was entered as covariant. WFPS x tillage were not significant and removed from the

287 model.

288 Tests for normality and homogeneous variance were performed graphically. Natural log-

289 transformation (*Parkin & Robinson, 1993*) of the N<sub>2</sub>O and CO<sub>2</sub> emissions data was carried out

290 prior to the analysis of variance. LSMEANS were calculated and compared with an LSD-test

291 at  $\alpha = 5\%$ . Ln Daily flux standard deviations were back transformed with the delta method.

## 292 3 Results and discussion

### 293 3.1 Weather conditions

294 Several heavy rainfalls (>10 mm d<sup>-1</sup>) occurred during the first four weeks of our measurements

295 (Figure 1a). The first heavy rainfall within this period occurred with approximately 12 mm d<sup>-1</sup>

296 two days after surface application of the residues. Highest daily amount of rainfall within the

297 whole investigation period was measured on August 26<sup>th</sup> (40 mm d<sup>-1</sup>). With the beginning of

298 the vegetation period in 2015, distinct phases without precipitation (two weeks or longer) were

299 followed by intense rainfall events in March, April and May.

300 Although mean air temperature dropped on two occasions below 0°C for a longer period in  
301 winter, no permanent frost was recorded in 5 cm soil depth (Figure 1a). The first frost period  
302 with mean daily air temperature below zero occurred at the end of December 2014 and lasted  
303 five days. A second pronounced frost was measured for a period of 10 days at the beginning of  
304 February.

305 The precipitation in the whole experimental period (July 2014 to May 2015) was 776 mm  
306 (Figure 1a). It was higher than the long-term mean in this period (620 mm).

307 ((Figure 1))

## 308 3.2 Trace gas fluxes

### 309 3.2.1 Temporal nitrous oxide flux dynamics

310 Figure 1b shows mean N<sub>2</sub>O fluxes during the study period. Increased N<sub>2</sub>O fluxes were measured  
311 immediately after crop residue application in conjunction with precipitation (Figures 1a, 1b).  
312 Except for two sampling dates at the end of August, the N<sub>2</sub>O fluxes increased for more than  
313 two months and showed a similar course to the CO<sub>2</sub> fluxes (Figure 1c). In the period between  
314 the beginning of the experiment and the end of September 2014 we found a positive correlation  
315 between N<sub>2</sub>O and CO<sub>2</sub> flux rates ( $r = 0.54$ ,  $p < 0.001$ ,  $n = 4$ , Pearson correlation).

316 Such an increase in N<sub>2</sub>O release after crop residue application has frequently been observed  
317 (*Flessa & Beese, 1995; Baggs et al., 2003; Millar & Baggs, 2004*). Amendment of crop residues  
318 provides easily available labile carbon and nitrogen as substrates which in turn can increase the  
319 microbial activity in the soil. Resulting rapid oxygen consumption by microbes during  
320 respiration decreases the redox potential and thus favours conditions for denitrification (*Flessa*  
321 *& Beese, 1995; Azam et al., 2002; Miller et al. 2008*).

322 In the first two weeks after residue application, CO<sub>2</sub>- and N<sub>2</sub>O-fluxes from the treatments with  
323 crop residues were significantly higher compared to the treatments without crop residues  
324 (Figure 1b). By the beginning of September, the higher fluxes were measured in the CT – CR  
325 treatment.

326 This result confirmed observations from a laboratory experiment on the effect of OSR residues  
327 with different C/N-ratios on N<sub>2</sub>O fluxes from the topsoil of our study site (*Herr, 2015*). After  
328 an initial phase where OSR residues stimulated N<sub>2</sub>O release, the N<sub>2</sub>O emissions decreased  
329 below emissions from a treatment without crop residues indicating a microbial immobilization  
330 of mineral N as a result of the wide C/N-ratio. Several further studies indicated a net NO<sub>3</sub><sup>-</sup>  
331 immobilization after application of crop residues with high C/N-ratios (*Chaves et al., 2007*;  
332 *Kaewpradit et al., 2008*; *Chen et al., 2013*). Immobilization therefore reduces the availability  
333 of mineral N which serves as substrate for microbial N<sub>2</sub>O production in soils and thus decreases  
334 N<sub>2</sub>O emissions (*Huang et al., 2004*).

335 In the RT + CR treatments we could not observe any mineral N immobilization effect of crop  
336 residues on N<sub>2</sub>O fluxes (Figure 1b) in September; the fluxes were as low as in the treatment  
337 CT+CR and the reason for this phenomenon remained unclear.

338 After tillage, N<sub>2</sub>O fluxes were low until the beginning of December. Low soil moisture contents  
339 below 60 % WFPS did not allow for anaerobic conditions necessary for denitrification (*Dobbie*  
340 *et al., 1999*; *Skiba & Ball, 2002*; *Batemann & Baggs, 2005*).

341 Frost-thaw events occurred between December and March resulting in moderate but steadily  
342 elevated N<sub>2</sub>O fluxes up to 50 µg N<sub>2</sub>O-N m<sup>-2</sup> h<sup>-1</sup> which was considerably higher than the  
343 background emission in late autumn. These increased background fluxes during winter are in  
344 accordance with other studies, whereas we did not measure pronounced flux peaks during  
345 thawing of frozen soil (*Flessa et al., 1995*; *Röver et al., 1998*; *Kammann et a., 1998*; *Kaiser &*

346 *Ruser*, 2000). The absence of pronounced N<sub>2</sub>O pulses during thawing may be explained by the  
347 short durations of the frost periods and by the mild temperature conditions. It was shown that  
348 N<sub>2</sub>O pulses during thawing of frozen soil increase with increasing duration of frost periods and  
349 with severity of the soil freezing (*Teepe et al.*, 2004; *Risk et al.*, 2013; *Xu et al.*, 2016).

350 At the end of winter, in the middle of March, N<sub>2</sub>O emissions declined to the background level.  
351 N<sub>2</sub>O emissions after fertilization were influenced by rainfall events. The highest mean flux in  
352 this period was measured with 175 µg N<sub>2</sub>O-N m<sup>-2</sup> h<sup>-1</sup> in the RT + CR treatment following two  
353 precipitation events with 20 and 10 mm d<sup>-1</sup> at the beginning of May.

354 High fluxes after N-fertilization and rainfall are usually explained by enhanced denitrification  
355 due to the increased availability of nitrate as substrate for N<sub>2</sub>O production and anaerobic  
356 conditions as a result of increased soil water contents (*Flessa et al.*, 1995; *MacKenzie et al.*,  
357 1997).

358 We did not find significant effects of the treatments on median daily flux rate either of the crop  
359 residues ( $p = 0.78$ ) or of the tillage treatment ( $p = 0.57$ ) and WFPS ( $p = 0.41$ ).

360 There was a strong correlation ( $p < 0.0001$ ) between CO<sub>2</sub> fluxes and N<sub>2</sub>O fluxes. This indicated  
361 heterotrophic microbial denitrification as a main N<sub>2</sub>O source during the investigated period.  
362 The C availability is an essential factor controlling denitrification (*Knowles* 1982; *Beauchamp*  
363 *et al.*, 1989), directly by increasing the energy and electron donator for denitrifiers, and  
364 indirectly through enhanced microbial growth thereby stimulating high O<sub>2</sub> consumption  
365 (*Beauchamp et al.*, 1989; *Garcia-Montiel et al.*, 2003, *Gillam et al.*, 2008).

366



### 367 3.2.2 Cumulative N<sub>2</sub>O emission

368 Total cumulative N<sub>2</sub>O emissions over the whole measuring period varied from 1.7 to 2.4 kg  
369 N<sub>2</sub>O-N ha<sup>-1</sup> with no significant differences between the treatments (Figure 2). Although our  
370 measurement period covered only 299 days, the order of magnitude of cumulative emissions  
371 was similar to annual emissions reported from OSR fields (*Ruser et al., 2017*) as well as from  
372 other arable crops (*Jungkunst et al., 2006*).

373 Between 50-68 % of total N<sub>2</sub>O emission was released during the post-harvest period covering  
374 only two months, highlighting the importance of that period for N<sub>2</sub>O budgets in OSR  
375 production. Soil tillage did not affect the emissions in this period.

376 A high share of post-harvest N<sub>2</sub>O emissions to the total annual N<sub>2</sub>O loss was also reported from  
377 *Ruser et al. (2017)* who measured trace gas fluxes at five study sites representative for German  
378 winter oilseed rape production. They explained the high post-harvest emissions with increased  
379 nitrate contents combined with O<sub>2</sub> consumption during the turn-over of the OSR residues thus  
380 favoring N<sub>2</sub>O release from denitrification.

381 Due to the mild winter conditions this period accounted for only 18-28 % of the total emission.  
382 In this period, N<sub>2</sub>O emission from the reduced tillage system was significantly higher than from  
383 the conventional treatment.

384 Higher N<sub>2</sub>O fluxes under reduced tillage were often observed (e.g. *Johnson et al., 2005*;  
385 *Venterea et al., 2005*), particularly in the first years after transition from conventional to reduced  
386 tillage (*D'Haene et al., 2008*). The reason for the higher emission was the often reported higher  
387 soil moisture in RT systems favouring denitrification (*Aulakh et al., 1984*; *Staley et al., 1990*;  
388 *Palma et al., 1997*; *MacKenzie et al., 1997*). Our results confirmed these earlier observations  
389 since soil moisture in the RT treatments were predominantly higher when compared to the CT  
390 treatments (Figure 1d).

391

((Figure 2))

392 The share of the N<sub>2</sub>O emission during the vegetation period (of the succeeding winter wheat)  
393 to the total emission varied between 14-32 % (Figure 2). Crop residues stimulated N<sub>2</sub>O emission  
394 during the subsequent cropping season. This effect was significant in the RT treatment ( $p =$   
395 0.03) and appeared in tendency in the CT treatment. CO<sub>2</sub> fluxes in this period showed the same  
396 trend but since we used dark chambers and therefore could not differentiate between soil  
397 respiration and dark respiration from photosynthesis of the plants within our chambers, we do  
398 not present this data, although aboveground wheat biomass was apparently similar in all  
399 investigated plots. We assume that this crop residue effect on N<sub>2</sub>O emissions during the  
400 succeeding cropping season of wheat in the RT+CR treatment was induced by mineralisation  
401 of the OSR residues which was indicated by an increase <sup>15</sup>N abundance (%) in the nitrate pool  
402 between May and June (Table 3). In contrast there was no change of the <sup>15</sup>N abundance in the  
403 nitrate pool of the CT+CR treatment in the same period. Consequently, this effect did not occur  
404 in the CT+CR treatment. The reason for different crop residue response in this period remained  
405 unclear; the slightly drier conditions in the CT+CR treatment might have contributed here.

406

((Table 3))

407

### 408 3.3 Contribution of crop residue N to N<sub>2</sub>O emission and <sup>15</sup>N recovery

409 Only 4.2 % in CT+CR and 5.2 % in RT+CT (no significant difference) of the N released as  
410 N<sub>2</sub>O during the investigated period stemmed directly from the applied OSR crop residues. N<sub>2</sub>O  
411 production from crop residue N occurred mainly in the first two months before tillage (Figure  
412 3). This corresponds with *Baggs et al. (2000)* who found 65 % of the measured N<sub>2</sub>O emissions  
413 during two weeks after crop residues incorporation. We interpret this as rapid stimulation of  
414 microbial decomposition of the residues (*Shen et al., 1989*) related with anaerobic conditions

415 resulting from microbial respiration which was in connection with the increased C supply as  
416 substrate for denitrification favoring higher N<sub>2</sub>O emissions.

417 Table 3 shows the mean soil N<sub>min</sub> (NH<sub>4</sub><sup>+</sup> and NO<sub>3</sub><sup>-</sup>) contents of the treatments, as well as the  
418 share of <sup>15</sup>N N<sub>min</sub>, from the + CR treatments. N<sub>min</sub> values were similar in the + CR treatments.  
419 In May the content was highest with approx. 88 kg N ha<sup>-1</sup> after N-fertilization. The highest share  
420 of the crop residue N calculated from <sup>15</sup>NH<sub>4</sub><sup>+</sup> and <sup>15</sup>NO<sub>3</sub><sup>-</sup> was measured in December with 4.8 %  
421 in the CT treatment and 6.3 % in the RT treatment. In spring and early summer, the share of  
422 crop residues dropped down to approx. 1.0 % in CT+CR and 1.7 % in RT+CR. This low  
423 contribution of <sup>15</sup>N to the total mineral N explained the low share of <sup>15</sup>N-N<sub>2</sub>O in the N<sub>2</sub>O fluxes.

424 ((Figure 3))

425 The low contribution from crop residues on total N<sub>2</sub>O emissions presumably resulted from the  
426 wide C/N-ratio of the OSR crop residues. *Pfab* (2011) reported that 38 % of the total N<sub>2</sub>O  
427 emission stemmed directly from cauliflower residues as a contrasting material (C/N-ratio:  
428 10.4).

429 The total <sup>15</sup>N-recovery rate after the experimental period (Table 4) was 65.0 % (CT) and 75.1 %  
430 (RT) respectively. Since N losses from crop residue as N<sub>2</sub>O were low (0.06 and 0.09 % of N  
431 added, Table 4) main pathways for N losses were presumably over the leaching pathway (NO<sub>3</sub><sup>-</sup>  
432 or as dissolved organic N) or gaseous as NH<sub>3</sub>, NO or elemental N<sub>2</sub>. Other field studies found  
433 similar recoveries covering ranges between 60 % and 85 % (*Jensen et al.*, 1997; *Garza et al.*,  
434 2009; *Zhang et al.*, 2010). Most of the applied <sup>15</sup>N was found in soil (97 %). We assume that  
435 the main reason for the high portion of soil-N to the total recovery of the <sup>15</sup>N applied was the  
436 slow turnover of the high C/N-ratio crop residues as a result of immobilization. In an incubation  
437 experiment, *Trinsoutrot et al.* (2000) indicated N immobilization after incorporation of low N  
438 OSR crop residue material even 186 days after application of the residues.

439

((Table 4))

440 The main proportion of the  $^{15}\text{N}$  recovery after termination of the measurements in June was  
441 found in the soil fraction (Table 4). This suggests that most of the crop residues were not  
442 completely mineralized at that time and that there was still potential for  $\text{N}_2\text{O}$  emission from that  
443 source in consecutive years. *Jensen et al. (1997)* found approx. 90 % of the total soil N, resulted  
444 from the incorporated  $^{15}\text{N}$  rapeseed straw (C/N-ratio 80), after seven months, indicating that  
445 there was no loss of straw N during winter, supporting our results.

446 The total N-input in +CR treatments was 245.3 kg and in –CR treatments 110 kg N ha<sup>-1</sup>. The N  
447 input related emissions factors (NEF) are shown in Table 5. The NEF of 1.84 and 1.57 %  
448 respectively from the treatments without crop residues were distinctly higher compared to the  
449 NEF of 0.77 and 0.98 % respectively from treatments with crop residues. These results  
450 supported the assumption of immobilization effect due to OSR crop residues. The calculated  
451 NEF are within the range of 0.3 to 3 % suggested by the IPCC (2006) for N-input related direct  
452  $\text{N}_2\text{O}$  emissions.

453 Due to the  $^{15}\text{N}$  labeling it was possible to calculate a separate emission factor for the crop  
454 residues. The  $\text{EF}_{\text{CR}}$  were very low with 0.03 (CT) and 0.05 % (RT) (Table 5). Here the IPCC  
455 (2006) is overestimating the share of OSR crop residues.

456

((Table 5))

457

#### 458 4 Conclusions

459 Despite a stimulating effect of crop residues in the RT+CR treatment over the vegetation period  
460 after crop residue incorporation,  $\text{N}_2\text{O}$  emission was not affected over the total experimental  
461 period. Therefore, our first hypothesis that crop residues increase  $\text{N}_2\text{O}$  emissions must be

462 declined. In contrast, our second hypothesis of a low emission factor (EF) for OSR crop residues  
463 due to a high C/N-ratio was confirmed. The low EF from the crop residues was  
464 overcompensated by the obviously high EF from mineral N fertilization of the succeeding  
465 wheat crop, thus resulting in EF within the range of the IPCC Tier 1 approach when related to  
466 the total N-input.

467 From the low direct contribution of crop residues to N<sub>2</sub>O emission during the post-harvest  
468 period we conclude that the increased N<sub>2</sub>O fluxes after OSR harvest reported in other studies  
469 were predominantly a result of increased N<sub>min</sub> contents already apparent during the harvest  
470 period (1) due to the early ending of N uptake by OSR and (2) due to proceeding soil N  
471 mineralization increasing the N<sub>min</sub> pool.

472 Furthermore, we found higher N<sub>2</sub>O emissions in the RT treatment in the winter period but due  
473 to the low contribution of the winter fluxes to the total cumulative N<sub>2</sub>O emission this effect was  
474 not significant for the entire investigation period. Consequently, we had also to decline our third  
475 hypothesis of higher N<sub>2</sub>O emissions in the RT system.

476

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483

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486

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744 **Table 1:**

745

746 Soil chemical and physical characteristics of the experimental site.

Soil depth	Organic C	Organic N	pH <sup>s</sup>	Sand	Silt	Clay
cm	mg C kg <sup>-1</sup>	mg N kg <sup>-1</sup>			%	
0-30	16.8	2.0	6.8	3.2	78.2	18.6
30-60	9.2	1.2	7.0	3.2	77.3	19.5
60-90	5.3	0.7	7.1	7.0	64.3	28.7

747 <sup>s</sup>pH 0.01 M CaCl<sub>2</sub>

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750

751 **Table 2:**

752 Proportions of the plant parts and their <sup>15</sup>N abundance in the mixture of the OSR crop residues

753 applied to field.

Plant part	Amount [g plot <sup>-1</sup> ]	Proportions of the mixture [%]	<sup>15</sup> N abundance [atom% <sup>15</sup> N]
Root	50	11.0	9.7
Pod	100	22.0	22.6
Litter	165	36.3	12.3
Stem	130	28.6	12.1
Seed	10	2.1	22.5
Total	455	100	14.4

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757 **Table 3:**

758 Mean  $N_{\min}$  content ( $\pm$  SE) and share of  $^{15}\text{N}$ - $N_{\min}$  in  $^{15}\text{N}$  crop residue amended treatments,  
 759 conventional tillage (CT) in 0-30 cm and reduced tillage (RT) subdivided into 0-15 and 15-30  
 760 cm with (+CR) and without (-CR) crop residue return.

Treatment	Date	depth cm	$N_{\min}$		$^{15}\text{N}$ - $N_{\min}$		
			kg N ha <sup>-1</sup>	SE	kg N ha <sup>-1</sup>	SE	%
CT + CR	09.09.2014	0-30	32.0	1.6	1.1	0.4	3.5
	02.12.2014	0-30	31.3	3.3	1.5	0.1	4.8
	12.05.2015	0-30	88.7	25.3	0.8	0.2	0.9
	02.06.2015	0-30	48.3	10.3	0.5	0.1	1.0
RT + CR	09.09.2014	0-30	26.0	0.8	1.4	0.2	5.5
	02.12.2014	0-15	12.7	1.7	1.0	0.1	7.7
		15-30	14.3	1.5	0.7	0.2	4.9
		$\Sigma$	27.0		1.7		6.3
	12.05.2015	0-15	52.8	5.4	0.6	0.1	1.2
		15-30	34.7	10.7	0.2	0.0	0.6
		$\Sigma$	87.5		0.8		0.9
	02.06.2015	0-15	23.3	1.7	0.3	0.1	1.1
15-30		16.1	2.6	0.2	0.1	1.3	
$\Sigma$		29.4		0.5		1.7	
CT - CR	09.09.2014	0-30	28.6	2.3			
	02.06.2015	0-30	105.7	9.4			
RT - CR	09.09.2014	0-30	11.8	0.7			
	02.06.2015	0-30	36.0	9.5			

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762

763 **Table 4:**

764  $^{15}\text{N}$  recovery ( $^{15}\text{N}_{\text{RCE}}$ ) of  $^{15}\text{N}$ - crop residues under conventional tillage (CT+CR) and reduced  
765 tillage (RT+CR) in  $\text{N}_2\text{O-N}$ , biomass (winter wheat) and soil.

Treatment	$\text{N}_2\text{O-N}$	Biomass*	Soil <sup>§</sup>	$\sum^{15}\text{N}_{\text{RCE}}$
		%		
CT + CR	0.06	2.2	62.8	65.0
RT + CR	0.09	2.1	72.9	75.1

766 \*aboveground biomass + roots; <sup>§</sup>plough layer 0-30 cm.

767

768 **Table 5:**

769 N<sub>2</sub>O emission factor for total N-input (NEF) and separately for crop residues (EF<sub>CR</sub>).

Treatment	NEF	EF <sub>CR</sub>
	[%]	
CT + CR	0.77	0.03
CT - CR	1.84	-
RT + CR	0.98	0.05
RT - CR	1.57	-

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772 **Figure captions**

773 **Fig. 1:** Daily precipitation, mean daily air and soil temperature (a), mean N<sub>2</sub>O flux rates (n=4;  
774 ± SE) (b), mean CO<sub>2</sub> flux rates (c), and mean water-filled pore space (WFPS %) (d) as affected  
775 by soil tillage and crop residue amendment during the investigation period.

776

777 **Fig. 2:** Mean cumulative N<sub>2</sub>O emission (n=4) divided into post-harvest, winter and vegetation  
778 periods as affected by soil tillage and crop residue amendment. Different letters indicate  
779 statistical significant differences between treatments within one measuring period (Tukey-Test;  
780 p < 0.05).

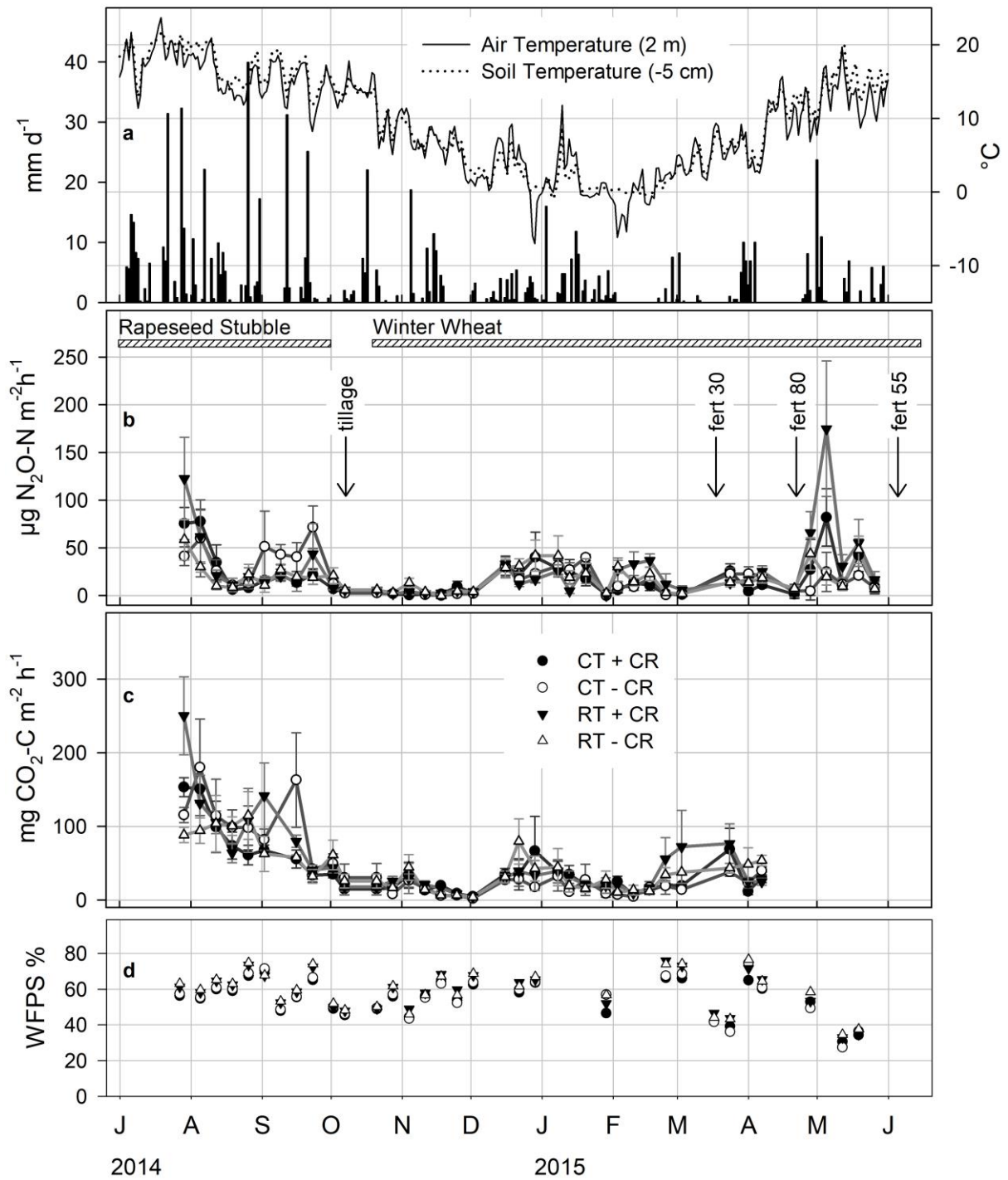
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782 **Fig. 3:** Mean cumulative N<sub>2</sub>O emission as affected by tillage (CT and RT) and crop residue (+  
783 CR and – CR) (left Y-axis) and <sup>15</sup>N-N<sub>2</sub>O emissions from labelled crop residues (n=4, mean ±  
784 standard error) (right Y-axis).

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787 **Figure 1**



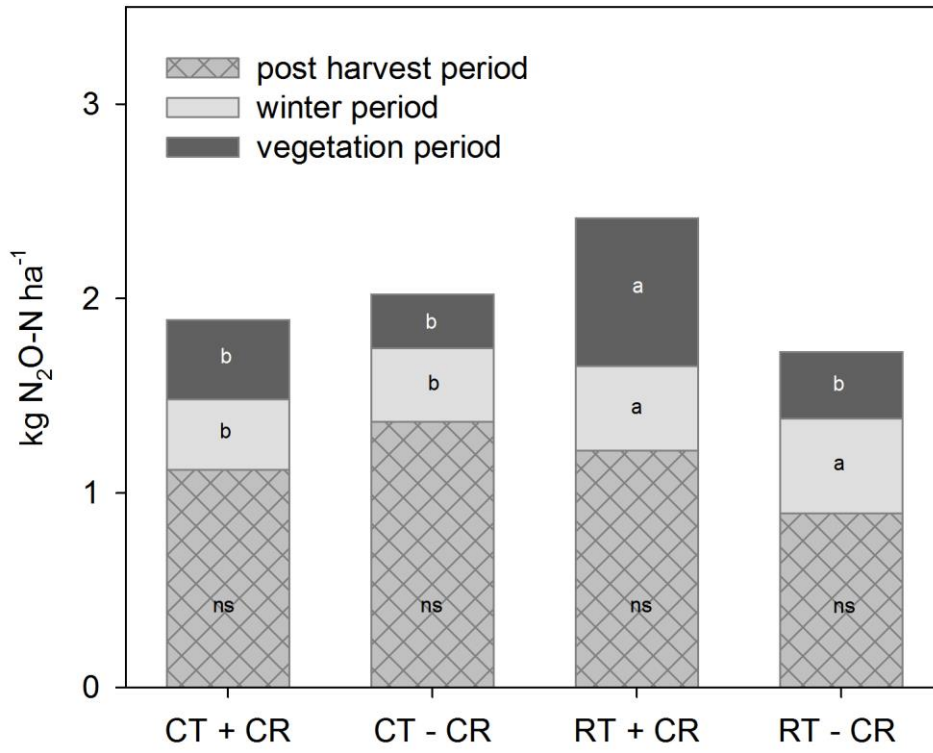
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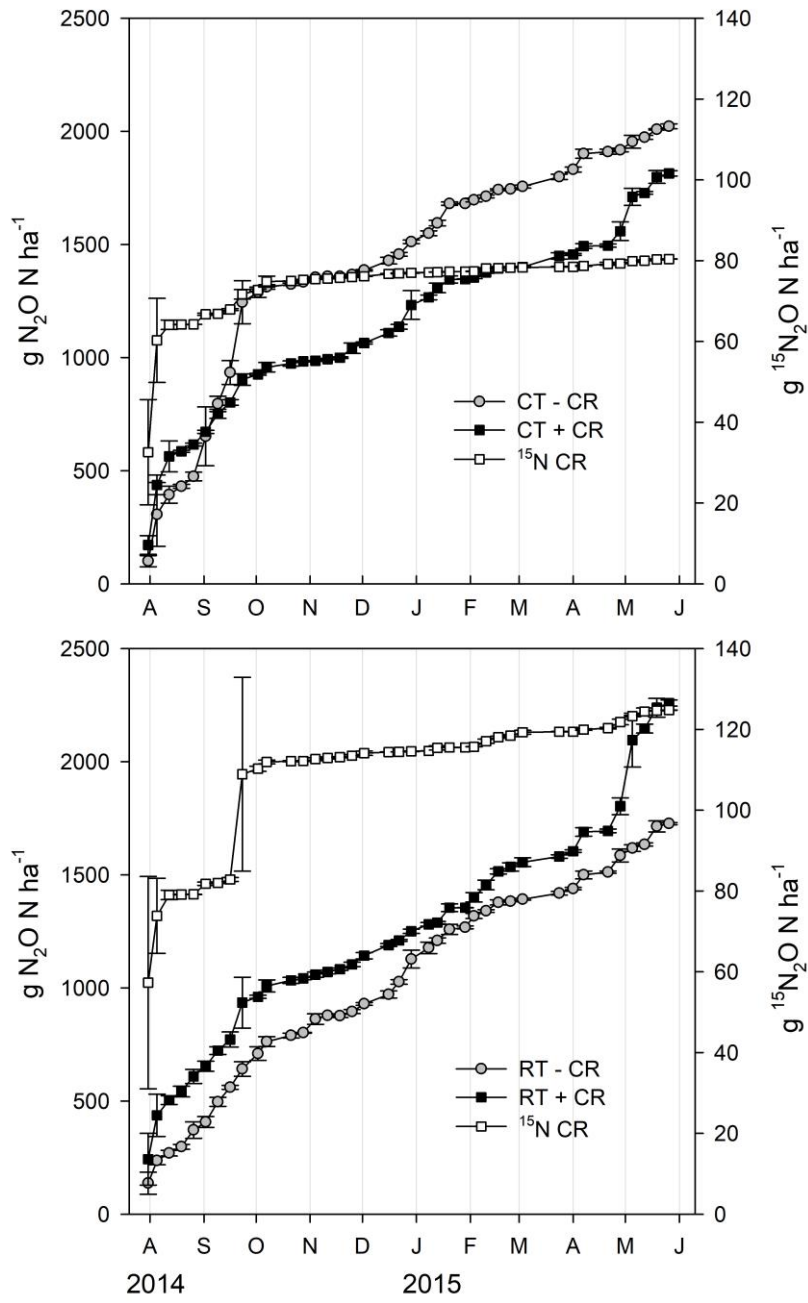


791 **Figure 2**



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