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TITLE: Mustard catch crop enhances denitrification in shallow groundwater
beneath a spring barley field

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19 **Mustard catch crop enhances denitrification in shallow**
20 **groundwater beneath a spring barley field**

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44 **Abstract**

45 Over-winter green cover crops have been reported to increase dissolved organic
46 carbon (DOC) concentrations in groundwater, which can be used as an energy source
47 for denitrifiers. This study investigates the impact of a mustard catch crop on *in situ*
48 denitrification and nitrous oxide (N₂O) emissions from an aquifer overlain by arable
49 land. Denitrification rates and N₂O-N/(N₂O-N+N₂-N) mole fractions were measured
50 *in situ* with a push-pull method in shallow groundwater under a spring barley system
51 in experimental plots with and without a mustard cover crop. The results suggest that a
52 mustard cover crop could substantially enhance reduction of groundwater nitrate
53 (NO₃⁻-N) via denitrification without significantly increasing N₂O emissions. Mean
54 total denitrification (TDN) rates below mustard cover crop and no cover crop were
55 7.61 and 0.002 μg kg⁻¹ d⁻¹, respectively. Estimated N₂O-N/(N₂O-N+N₂-N) ratios,
56 being 0.001 and 1.0 below mustard cover crop and no cover crop respectively,
57 indicate that denitrification below mustard cover crop reduces N₂O to N₂, unlike the
58 plot with no cover crop. The observed enhanced denitrification under the mustard
59 cover crop may result from the higher groundwater DOC under mustard cover crop
60 (1.53 mg L⁻¹) than no cover crop (0.90 mg L⁻¹) being added by the root exudates and
61 root masses of mustard. This study gives insights into the missing piece in agricultural
62 nitrogen (N) balance and groundwater derived N₂O emissions under arable land and
63 thus helps minimise the uncertainty in agricultural N and N₂O-N balances.

64

65 *Key Words:* cover crop, denitrification, shallow groundwater, DOC, N₂O-N, N₂-N

66

67 **1 Introduction**

68 Groundwater contamination by NO_3^- -N is a cause of concern for the environment
69 (Galloway et al., 2008). Aquifer discharge of NO_3^- -N into streams, lakes, rivers and
70 coastal transitional waters can increase the risk of eutrophication in surface waters
71 (Stark and Richards, 2008). Excessive NO_3^- -N leaching to groundwater below arable
72 land in a spring barley system, where land is left fallow over winter, has been reported
73 before by Hooker et al. (2008). In tillage farming, cover crops reduce NO_3^- -N
74 leaching to groundwater through the uptake of N during the fallow period between
75 crop harvest and subsequent planting of the next crop (Shepherd et al., 1993). Over a
76 three years period mustard sown has been found to reduce mean groundwater NO_3^- -N
77 concentration by c. 25% (Premrov et al., 2012). The mean DOC concentrations were
78 found to be significantly higher by c. 32% under the mustard cover crop than under no
79 cover crop, suggesting that mustard may help reduce groundwater NO_3^- -N occurrence
80 by (i) taking up soil N and/or supplying DOC in groundwater to enhance
81 denitrification.

82

83 Nitrate reduction into un-reactive N via denitrification can be accompanied by the
84 emissions of N_2O , a potent greenhouse gas with global warming potential of 298
85 (IPCC, 2007). The contribution of the leached NO_3^- -N with associated groundwater to
86 indirect N_2O -N emissions is well recognised (IPCC, 2007) but the dynamics of N_2O
87 production and reduction *in situ* in groundwater is not yet well understood (Clough et
88 al., 2007). Moreover, in measuring denitrification in groundwater, it is often unclear if
89 denitrification products are produced in situ or if they have been leached from surface
90 soils (Groffman et al., 1998). An estimation of N_2O -N/(N_2O -N+ N_2 -N) ratios is
91 necessary to know the potential of pollution swapping for NO_3^- -N to N_2O -N.

92 Moreover, quantification of the end product of denitrification, N_2-N , is also important
93 to minimise uncertainty in the agricultural N balance (Galloway et al., 2004).

94

95 While previous research recognised the importance of mustard as an over winter
96 cover crop in reducing NO_3^-N leaching to groundwater during the winter recharge
97 (Hooker et al., 2008; Premrov et al., 2012), there are no reports on the effect of cover
98 crops on groundwater denitrification and the N_2O or N_2 transformation rates. This
99 information is crucial to better understand N cycling below an arable system and to
100 improve land management. The objective of this experiment was to investigate the
101 effect of a mustard cover crop on *in-situ* denitrification rates and $N_2O-N/(N_2O-N+N_2-$
102 $N)$ ratios in shallow groundwater under a spring barley cropping system.

103

104 **2 Methodology**

105 **2.1 Site and experimental design**

106 The experiment was carried out during February - March, 2011 at Oak park Research
107 Centre, Co. Carlow, Ireland ($52^{\circ}51'43''$ N, $6^{\circ}54'53''$ W) in a shallow sand/gravel
108 aquifer (water table <2.5 m below ground level, bgl). The top soil is a well drained
109 sandy loam overlying inter-bedded layers of sand, gravel and silt/clay. The shallow
110 fluvioglacial sand and gravel aquifer is underlain by a deeper Carboniferous limestone
111 aquifer. Two over winter treatments within a spring barley system have been
112 cultivated since 2006: (1) mustard cover crop and (2) no cover crop, as part of a larger
113 experiment on the effect of over winter green cover on NO_3^-N leaching. Three
114 independent piezometers (PVC pipe; 0.03 m i. d. and 1.0 m screen section) were
115 installed in each treatment to a depth of 4 m bgl. The treatment plots were oriented to

116 the dominant groundwater flow to ensure hydrogeological homogeneity and to
117 minimise lateral flow. Leaching is the dominant hydrological pathway as overland
118 flow on this site was considered negligible due to the free draining nature of soils and
119 subsoils. Inorganic N fertiliser “Super Nett” (27% N and 3.7% S) and KCl were used
120 at a rate ranged from 115 - 135 kg N and 35 - 91 kg Cl ha⁻¹. Spring barley was grown
121 during March to August. Mustard cover crop was grown during November to April
122 and ploughed prior to the next barley cropping.

123

124 **2.2 *In-situ* push-pull method**

125 Denitrification rates in groundwater were measured *in situ* using a Push-Pull method
126 described by Addy et al. (2002). In brief, the push-pull method consists of collecting
127 groundwater from a well, amending it with ¹⁵N-enriched NO₃⁻-N and a conservative
128 tracer (bromide), injecting the solution in the aquifer (“push”), incubating for 4-h and
129 pumping back (“pull”). Ten L of groundwater (fill 43.4 kg of aquifer materials; bulk
130 density: 1.65 g cm⁻³; porosity: 38%) was collected from each well (depth 4 m bgl) in a
131 plastic container (carboy) using a peristaltic pump (Model 410, Solinst Canada Ltd.)
132 and immediately enriched with 50 mg L⁻¹ Br⁻ (as KBr) and 50 mg L⁻¹ isotopically
133 enriched (50 atom % ¹⁵N) NO₃⁻-N (as KNO₃⁻-N). The dosing solutions (i.e., the 10 L
134 of amended groundwater) were pushed into the wells (4 m bgl) at a low rate (15 L h⁻¹)
135 with a peristaltic pump to minimise changes in the hydraulic potential surrounding the
136 well. To ensure that the DO content of the enriched solution is same to the collected
137 groundwater, DO was monitored using a probe (Multi 340i/SET, WTW, Germany). A
138 small quantity of the dosing solution (targeted 500 ml) was left at the bottom of the
139 carboy to measure dissolved gases and hydrochemistry.

140

141 Incubation time was set at 4 h because previous study on this site indicated that longer
142 or shorter than 4 h incubations can respectively, reduce recovery of injected solution
143 or detection of denitrification products (Jahangir et al., 2012a). After incubation for 4
144 h, 20 L of groundwater was pulled from each well at the same rate (15 L h^{-1}) as during
145 the push phase to avoid generating gas bubbles within the gas-impermeable Teflon
146 tubing. Groundwater samples were collected at 2 L intervals into 160 ml glass serum
147 bottles for dissolved $\text{N}_2\text{O-N}$ and $\text{N}_2\text{-N}$ analysis and into 50 ml plastic tubes for the
148 measurement of hydrochemical parameters. The Teflon outlet was placed at the
149 bottom of the glass bottle, gradually filled with groundwater and immediately sealed
150 with butyl rubber septa and aluminium crimp caps (Wheaton, USA). No visible air
151 bubbles were observed in the sample. In a preliminary test, such samples taken
152 directly in the glass bottles; or indirectly using a syringe attached to an air-tight
153 sampling tube (Teflon) connected to the outlet of the pump did not show any
154 significant differences in dissolved N_2O , CO_2 and CH_4 concentrations. All samples
155 were submerged under water in a polystyrene box and stored at 4°C until analysed
156 for dissolved gases, ions and hydrochemistry within one week of collection.

157

158 **2.3 Dissolved gases and hydrochemical analyses**

159 Denitrification products in groundwater ($\text{N}_2\text{O-N}$ and $\text{N}_2\text{-N}$) were extracted using the
160 phase equilibration headspace extraction technique (Davidson and Firestone, 1988)
161 with helium (He ; BOC, Linde Group, Germany) filling the headspace (He : water 3:1;
162 v/v). In brief, samples in the serum bottles were shaken for 13 min on a Gyrotory
163 shaker (Model G-10, New Brunswick Scientific Co., USA) and left for a standing
164 period of 63 min (Jahangir et al., 2012c). Headspace samples were then taken in 12 ml
165 exetainers (Labco Inc. Wycomb, UK) using a syringe after injecting additional 15 ml

166 of high purity *He* for the analysis of N₂O and N₂ concentrations and the ¹⁵N
167 enrichment of N₂O and N₂. Concentrations and isotopic composition of N₂O-N and
168 N₂-N were determined on a dual-inlet isotope ratio mass spectrometer (Stable Isotope
169 Facility, UC Davis, CA) as described by Mosier and Schimel (1993).

170

171 Dissolved N₂O-N and N₂-N concentrations were calculated using the three highest
172 recovery values (plume core; being estimated from the recovery of tracer in the pulled
173 water) within sample replicates (Harrison et al., 2011) to minimise the effects of
174 uncertainty of estimation due to physical attenuation. For each piezometer,
175 conservative tracer (Br⁻) recovery was estimated as C/C₀, where C was the tracer's
176 concentrations in the pulled groundwater following incubation and C₀ was tracer's
177 concentrations in the original pushed groundwater (Freeze and Cherry, 1979). The
178 masses of dissolved N₂O-N and N₂-N gases (μg) were calculated from the headspace
179 extraction samples using equations and constants provided by Mosier and
180 Klemetsson (1994). The total mass of N₂O-N or N₂-N was then transformed to the
181 mass of ¹⁵N₂O-N or ¹⁵N₂-N multiplying it by the respective ¹⁵N sample enrichment
182 proportion (ratio of pulled atom% of the dissolved N₂O-N and N₂-N to pushed NO₃⁻-
183 N atom%, both corrected for ambient atom%). Gas production rates for ¹⁵N₂O-N and
184 ¹⁵N₂-N were expressed as μg N kg⁻¹ soil d⁻¹ following Eq. 1 below:

185

$$186 \text{ Rates } \mu\text{g N kg}^{-1}\text{d}^{-1} = \frac{\text{Total mass of } ^{15}\text{N}_2\text{O} - \text{N and } ^{15}\text{N}_2 - \text{N per volume of water pulled}}{\text{Dry mass of soil per volume of water * incubation period}} \quad \text{Eq. 1}$$

187

188 Mass of aquifer materials was calculated for individual depths at each well. The TDN
189 rates were the sum of ¹⁵N₂O-N and ¹⁵N₂ generation rates. Groundwater table depth
190 was measured using an electrical dip meter. Groundwater pH, electrical conductivity

191 (EC) and redox-potential (Eh) were measured on site using a multi-probe (Troll
192 19500, In Situ Inc. USA). Groundwater was analysed for NO_3^- -N and Br^- on DX-120
193 ion chromatography (Metrohm UK Ltd.). The DOC was analysed using Total Organic
194 Carbon Analyser (TOC-V cph/cpn; Shimadzu Corporation, Japan). Groundwater total
195 oxidised N, nitrite (NO_2^-), ammonium (NH_4^+) were analyzed by Aquakem 600
196 Discrete Analyser (Aquakem 600A, 01621 Vantaa, Finland).

197

198 **2.4 Statistical Analysis**

199 The measured denitrification rates and $\text{N}_2\text{O-N}/(\text{N}_2\text{O-N}+\text{N}_2\text{-N})$ ratios were not
200 normally distributed. The non-parametric test ‘Mann–Whitney U test’ (Ott, 1993) was
201 performed to determine significant differences in denitrification rates observed
202 between the mustard cover crop and no cover crop treatments. Differences between
203 the two treatments for hydrochemical properties were tested by a *t*-test. All statistical
204 analyses were performed on GenStat version 13 (VSN Intl Ltd., UK).

205

206 **3 Results**

207 **3.1 Ambient hydrochemical properties**

208 Nitrate concentrations under the mustard cover crop were significantly lower ($p <$
209 0.05) than without any cover crop (Table 1). Despite being low, DOC concentrations
210 were 44% higher ($p < 0.05$) in groundwater beneath mustard cover crop cultivated
211 plots than the no cover crop (Table 1). Other hydrochemical parameters (DO, pH, EC
212 and SO_4^{2-}) were statistically similar ($p > 0.05$) in groundwater below treatments,
213 except for Eh which was significantly lower below mustard cover crop (Table 1).

214

215 3.2 In situ push-pull tracer recovery

216 The tracer recovery data showed that injected plume was dispersed steadily from the
217 screen section into the aquifer (Fig. 1). After the 4 h incubation the highest mean
218 recovery of 59-66% of the injected concentrations of Br^- , was sufficient to calculate
219 the non-reactive losses of the injected NO_3^- -N. After pulling back 20 L of water (twice
220 the injected volume), the tracer reached the background concentration. Another
221 implication of tracer recovery is that injected plume was dispersed uniformly across
222 the aquifer materials even though it occupied only small amount of sediments (20 L =
223 ~ 87 kg).

224

225 3.3 Denitrification rates

226 The N_2O -N production rates in shallow groundwater were similar ($p > 0.05$) below
227 the mustard cover crop and the no cover crop treatments, with mean values of 0.003
228 and 0.002 $\mu\text{g N kg}^{-1} \text{d}^{-1}$, respectively (Fig. 2a). There was no N_2 -N production
229 detected in shallow groundwater beneath spring barley with no cover crop, whereas
230 the N_2 -N production rate was 7.61 $\mu\text{g N kg}^{-1} \text{d}^{-1}$ (Fig. 2b) beneath the mustard cover
231 crop treatment. The TDN rates beneath the mustard cover crop and no cover crop
232 treatments were 7.61 and 0.002 $\mu\text{g kg}^{-1} \text{N d}^{-1}$, respectively (Fig. 3a). Consequently,
233 the $\text{N}_2\text{O-N}/(\text{N}_2\text{O-N}+\text{N}_2\text{-N})$ ratio was about 1.0 beneath the no cover crop treatment,
234 whereas the ratio was much lower (0.001) beneath mustard cover crop (Fig. 3b). No
235 NH_4^+ accumulation in groundwater was detected.

236

237 4 Discussions

238 4.1 Impacts of mustard cover crop on groundwater geochemistry

239 Dissolved nutrients such as NO_3^- -N and DOC are transported rapidly to groundwater
240 on free draining sites with high rainfall. Our previous research on this experimental
241 site showed that the mustard cover crop significantly reduced NO_3^- -N and increased
242 DOC in groundwater compared to no cover crop (Premrov et al., 2012). Groundwater
243 NO_3^- -N was also found to be significantly negatively correlated with groundwater
244 DOC and temperature and positively correlated with EC, suggesting that
245 denitrification might be playing a role. High DO concentrations ($\sim 10 \text{ mg L}^{-1}$) in such
246 aquifers are due to the free draining and aerobic nature of the sediments. Moreover,
247 despite a significant increase in DOC below mustard cover crop, it does not seem to
248 be sufficient to significantly reduce the DO (10.3 mg L^{-1} oxygen uses up 3.8 mg C L^{-1} ,
249 Buss et al., 2005). A numerically lower value of DO below mustard cover crop (10.2
250 mg L^{-1}) compared to no cover crop (10.7 mg L^{-1}) indicates that some of the DO might
251 have been consumed by DOC. From a monthly monitoring during 2009 to 2011 in 5,
252 12 and 22 m bgl at this site (out side these plots), Jahangir et al. (2013) reported mean
253 groundwater DO concentrations of 7.1, 6.2 and 4.8 mg L^{-1} , respectively. They also
254 reported mean DOC concentrations of 1.1, 0.08 and 0.06 mg L^{-1} in 5, 12 and 22 m
255 bgl, respectively. Decrease in groundwater DO and DOC concentrations along
256 vertical gradients implies that consumption of DO corresponds to the consumption of
257 DOC while passing through the sediments. Nonetheless, the added DOC may create
258 some localised anoxic microsites within the aquifer sediments in some local pockets
259 of low permeability (Premrov et al., 2012) where inter-bedded clay band or silt/clay
260 lenses are in subsoils. The observed Eh (178 to 195 mV) does not correspond to the
261 high DO may be because it is unlikely that groundwater will be at equilibrium with

262 respect to redox in spatially complex geochemical conditions (Christensen et al.,
263 2000). In practice, systems seldom exhibit strict redox zone boundaries as a number of
264 redox reactions may occur simultaneously in any single aquifer block (McGuire et al.,
265 2002). In addition, redox conditions in an aquifer can best be defined by
266 simultaneously measuring several redox couples e.g., DO concentrations, ferrous iron
267 and manganese, or other reduces species (Postma et al., 1991).

268

269 **4.2 Role of mustard cover crop on denitrification rates**

270 Adoption of a mustard cover crop within a spring barley rotation significantly
271 increased groundwater denitrification rates. The denitrification process below mustard
272 cover crop has been found to reduce approximately $0.033 \text{ mg N L}^{-1} \text{ d}^{-1}$ (equivalent to
273 the TDN rate of $7.61 \text{ } \mu\text{g kg}^{-1} \text{ d}^{-1}$) accounting for 0.07% of the injected NO_3^- -N within
274 this short incubation time (c. 4 h), whereas it was negligible below no cover crop
275 system. The 4 h incubation time was sufficient for measuring denitrification products
276 in groundwater as evident by the study on which the methodology was based (Kellog
277 et al., 2005). The mean ambient NO_3^- -N concentrations of 13.6 and 20.2 mg N L^{-1}
278 respectively, in groundwater below the mustard cover crop and no cover crop systems
279 were in agreement with the denitrification results i.e., lower ambient NO_3^- -N
280 corresponded to higher TDN rates. Even though, the observed denitrification rates
281 within this short incubation do not seem to be sufficient to completely reduce the high
282 groundwater NO_3^- -N concentrations, while considering the aquifer residence time
283 (saturated hydraulic conductivity $3.3 \times 10^{-2} \text{ m d}^{-1}$ estimated by Jahangir et al., 2013)
284 can further help reduce the net NO_3^- -N. [The higher denitrification rates can be](#)
285 [attributed mainly to the increased DOC concentrations, coupled with physico-](#)
286 [chemical changes in the groundwater originating from root exudates from beneath the](#)

287 mustard cover crop. Mustard cover crop residues, being ploughed and incorporated
288 into the soil, may increase DOC availability in groundwater. The Eh was in a
289 favourable range for denitrification to occur because Eh values <250 mV have been
290 reported to be favourable for denitrification (Korom, 1992). Despite the high DO
291 concentrations, the increase in denitrification rates can possibly be due to the presence
292 of some microsites which have comparatively more anaerobic environment than the
293 surrounding areas (e.g., presence of inter-bedded clay band or silt/clay lenses in
294 subsoils). In this shallow aquifer, existence of silt/clay lenses in the subsoils and
295 glacial tills were already reported by previous authors (Premrov et al., 2012).
296 Denitrifying microbes are essentially facultative anaerobes, even though aerobic
297 denitrification also likely to occur (Cannavo et al., 2004). The occurrence of aerobic
298 denitrification is possible because aerobic denitrification can occur at even 80%
299 oxygen saturation conditions in groundwater (Carter et al., 1995). Even though
300 aerobic denitrification (~80% air saturation) can take place in groundwater,
301 denitrification actually seems more likely under locally anaerobic conditions within
302 microsites in particulate organic matter (Hammersley and Howes, 2002),
303 heterogeneous organic-rich patches of sediments (Jacinthe et al., 1998) or biofilms
304 (Seiler and Vomberg, 2005). In a monthly monitoring during 2009-2011, Jahangir et
305 al. (2013) measured mean N₂O and denitrified N₂ concentrations of 0.042 and 0.69
306 mg N L⁻¹, respectively in groundwater in 5 m bgl at this site (outside these plots)
307 wherein the DO concentrations were 7.1 mg L⁻¹. Therefore, these findings indicate
308 that, though low, denitrification occurs in these relatively aerobic environments.
309 Therefore, it can be concluded that in such aquifers denitrification is C limited. Lack
310 of organic carbon to provide energy to heterotrophic micro-organisms is usually

311 identified as the major factor limiting denitrification rates in aquifers (Devito et al.,
312 2000; Rivett et al., 2008).

313

314 The intermediate product of groundwater denitrification is important as N₂O is a
315 potent greenhouse gas and indirect emissions of N₂O from leached N contribute up to
316 25% of field scale total N₂O emissions (Reay et al., 2009). Denitrification under the
317 mustard cover crop resulted in nearly 100% N₂-N production, an un-reactive and
318 environmentally benign form of N, whereas the product of denitrification in the no
319 cover crop treatment was 100% N₂O. Even though, the *in situ* N₂O generation rates
320 were similar below mustard cover crop to no cover crop (Fig. 2a), mustard cover crop
321 showed the potential of reducing N₂O further to N₂ while passing through and from
322 the subsoils and aquifer sediments. This result is comparable with the subsoil
323 denitrification study by Jahangir et al. (2012b) who concluded that increased C supply
324 to subsoils (1.3 m bgl) in poorly drained grazed grassland increased denitrification
325 rates with N₂-N as the main end product (94% of TDN). Therefore, at the low DOC
326 level below the no cover crop, N₂O produced by denitrification was not reduced
327 further to N₂. The experiment was conducted during one season (February –March,
328 2011) because dissolved N₂O and N₂ in groundwater at this site were previously
329 observed to be similar throughout the year (Jahangir et al., 2013). No NH₄⁺
330 production was detected in this study, suggesting that shallow groundwater within this
331 4 m bgl did not contribute to dissimilatory NO₃⁻-N reduction to ammonium (DNRA).
332 In addition, NH₄⁺ when produced can be fixed in the clay lattice.

333

334 **5 Conclusions**

335 Introduction of a cover crop (mustard) into the agricultural management activities has
336 appeared to add DOC to groundwater. The added DOC either can serve either as an
337 energy source for denitrifiers or consume DO in the percolating water and create
338 anaerobic sites capable to denitrification. Results from the present study suggest that
339 the introduction of a mustard cover crop in the spring barley tillage areas can enhance
340 denitrification rates. Although the TDN rates are low, when combined with aquifer
341 residence times, denitrification could substantially reduce groundwater NO_3^- -N
342 concentrations. In addition, $\text{N}_2\text{O-N}/(\text{N}_2\text{O-N}+\text{N}_2\text{-N})$ ratios in this study indicate that
343 the end product of denitrification below the spring barley-mustard cover crop system
344 was $\text{N}_2\text{-N}$ (ratio 0.001), suggesting that inclusion of a mustard cover crop with spring
345 barley reduces indirect N_2O emissions to the atmosphere. On the contrary, under no
346 cover crop system the $\text{N}_2\text{O-N}/(\text{N}_2\text{O-N}+\text{N}_2\text{-N})$ ratios were 1.0, indicating shallow
347 groundwater as a potent source of indirect N_2O emissions. This study gives an insight
348 into the indirect pathway of N losses (as $\text{N}_2\text{O-N}$ or $\text{N}_2\text{-N}$) below different
349 management systems in arable land and thus should be accounted for agricultural N
350 balance to minimise the existing uncertainties.

351

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356

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460 *Table 1* Hydrochemical properties in two differently managed arable plots (mean \pm

461 SE; n=3)

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484 *Figure 1* Relative concentration profiles of conservative tracer (Br^-) beneath spring
485 barley with cover crop and without cover crop rotations from the 4-h *in-situ* NO_3^-
486 push-pull test; the term C represents the concentration of the sample pulled from the
487 well. The term C_0 represents the concentration of the solution originally pushed into
488 the well.

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490 *Figure 2* $\text{N}_2\text{O-N}$, (a) and $\text{N}_2\text{-N}$, (b) production rates in two different cropping systems:
491 spring barley with mustard cover crop and spring barley with no cover crop.

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493 *Figure 3* TDN (Total denitrification = $\text{N}_2\text{O-N} + \text{N}_2\text{-N}$) (a), and $\text{N}_2\text{O-N}$ mole fraction
494 ($\text{N}_2\text{O-N} / \text{N}_2\text{O-N} + \text{N}_2\text{-N}$), (b) in two different cropping systems: spring barley with
495 mustard cover crop and with no cover crop.

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