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Interactive comment on "N_2O and N_2 losses from simulated injection of biogas digestate depend mainly on soil texture, moisture and temperature" *by* Sebastian Rainer Fiedler et al.

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Dear Sir or Madam

We acknowledge your reasonable doubts regarding our manuscript and accept that there are, from an in situ point of view, methodically conditioned pitfalls, which prevent conclusions about the dynamics of short-term emissions of N2O, N2 and CO2 after soil amendments with 'injected' biogas digestate (BD). However, in the following, we would like to respond to your major points of criticism and argue why our approach should be accepted as an opportunity to get insights into relevant human induced biogeochemical processes, which are very difficult to examine with any other approach.

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Indeed, the ambition of our study was not to draw general conclusions about the dynamics over the whole period of increased emissions after the amendment. Rather, we aimed to detect the importance of different factors (soil texture, water-filled pore space and high nutrient concentrations resulting from spacing between injection slits) on the short-term emissions potentials, especially of N2, after amendments with BD, which has never been done before.

The determination of N2 emissions from soils is a delicate matter since the atmosphere consists to a large part of N2; hence, it is difficult to detect concentration changes in amounts relevant for soil processes. Generally, there still exist only two appropriate methods to tackle this challenge: 15N labelling and the use of an N2 free artificial headspace (Lewicka-Szczebak et al., 2014). Up to now, both methods, used separately or in combination, are only applicable for laboratory incubations. Recent efforts to determine N2 fluxes on field sites confirmed the need to continue the development of an in situ method (Lewicka-Szczebak et al., 2017). Since BD is a rather heterogeneous substrate, 15N labelling of ammonium would end up in a relative uneven distribution of the latter. Nevertheless, to establish a device for the helium oxygen method is also a protracted issue due to the efforts needed to achieve complete tightness against contamination from air born N2. In other words, we were able to perform an incubation to determine N2 fluxes from soils and such data have a high value. Moreover, such measurements on BD are generally rare and we are the first to provide data on N2 emissions from such high concentrations as they appear after injective application. Further, by applying the N2/(N2 + N2O) ratio to field measurements, an estimation of N2 emissions in situ might be possible.

Generally, the initial phase, i.e., the first week after fertilizer application, is crucial for N2O emissions (Dobbie et al., 1999; Kaiser and Heinemeyer, 1996) and most probably also for N2 because the same processes are involved. Regarding BD, first N2O peaks were observed within the first and third day in incubation experiments, which indicate a rather immediate reaction also for N2 at least in vitro (Köster et al., 2011; Köster et

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al., 2015; Senbayram et al., 2009). Therefore, we deduce our consecutive two days of measurements as appropriate. Nevertheless, these former studies recorded a second plateau of N2O emission consistently after around two weeks, which would imply an incubation duration of at least three weeks. Transferred to our incubation, this would mean a duration of at least 27 weeks, which involve a huge logistical effort for systems maintaining a nearly N2 free headspace. However, these authors operated at a higher WFPS than we in our study. At a lower WFPS of 65%, Senbayram et al. (2009) measured only one peak within two days without a repeated increase later, regardless the amount of applied BD. Thus, we assume a single peak shortly after application holds also true for our incubation. On the bottom line, the efforts for a study of the dynamics of N2 emissions harmonised along the lines of your criticism were out of our scope and are probably more appropriate for projects lasting for years that focus this single issue. However, we observed ourselves only a few days of increased N2O emissions in field trials under optimal conditions (Hagemann et al., 2016). Hence, we are convinced that the duration of measurements we have chosen are appropriate to get a first feeling for the impact of the different conditions of soil texture, water-filled pore space and applied BD concentration.

Our method is up to date for the investigation of soil related N2 emissions and has been applied successfully by several working groups, e.g., Köster et al. (2011) and Wang et al. (2011). For this method it is mandatory to keep the vessels at low temperature at the beginning: since we wanted to record the de novo production of N2O and N2, these two days without measurements are needed to rid the soil of this gases by diffusion into the virtually nitrogen free headspace. We specified the methods accordingly: "[. . .] to remove residues of N2 from soil cores by diffusion, including a restricted N2 production by decreased microbial activity" (lines 131 – 132). Thus, such a low temperature at the beginning is also obligate to reach a static state with no restricted N gas production. As a result, we had to perform a shift in temperature anyway and we just extended this period of low temperature for an additional check for emissions activity during these conditions, which also gave us the opportunity to have a comparison of

the effects these two different temperature regimes. Wang et al. (2011) and Wang et al. (2013), respectively, also presented a similar course of temperature shift. However, the temperature dependence of microbial processes like denitrification is well known (Phillips et al., 2015) and should no longer be emphasised in the revised manuscript.

Finally, we chose to switch to the headspace from an oxic state to anaerobicity to "to determine the current potential for N2O and N2 generation in a completely anaerobic soil matrix" (122 – 123), which is important to get a clue about actual potential for these gaseous N losses after BD application. We included the latter to the manuscript: "[. . .] to determine the generation of N2O and N2 in a completely anaerobic soil matrix. The latter step is important to get a clue about the actual potential for these gaseous N losses after highly concentrated BD application" (lines 138 – 140). Further, the microbes in soil associated with the production of N2O and N2 are able to react fast to changing environmental conditions by utilising existing enzymes within minutes or by de novo synthesis within 4 – 8 hours (Rudaz et al., 1991). Wang et al. (2011, 2013) showed in similar studies to ours that the emission of N2 and N2O peaked within less than 24 hours after switching their headspace from oxic to anaerobic conditions, which emphasise our study design as appropriate. Thus, the immediate and strong increase of N2 emissions in the sandy soil and the obvious changes in the silty soil on the last day of our study represent very likely the emission potential.

However, your concerns about the duration of our study might be a critical issue; but again, our aim was not to study the dynamics after BD application. Rather it was to estimate the effect of the differing conditions on gaseous N2 losses. Moreover, the fluxes from the anaerobic headspace indicate that we captured the actual potentials. On the one hand, we observed no changes of N2O in the clayey silt, which suggest no further increase would have awaited if we had extended the incubation period. The increased N2 emissions showed 'only' the potential, which would have aroused if the soil cores had been complete completely anaerobic. The latter has, however, no implications for mineral soils since such conditions are unlikely to occur 'in reality'. On

the other hand, the extremely increased N2 emissions from the loamy sand verify that this soil permitted abundantly oxygen diffusion, which let us assume a prevention of a possible emission increase in the former oxic headspace. Hence, we are convinced, again, that the duration of our measurements were appropriate to distinguish the effect of the examined conditions. Nevertheless, we will discuss the latter assumptions with respect to the incubation duration in a new inserted section on limitations (423 – 447)

Further, we did not reproduce injection of BD per se, but rather the conditions, which would establish in soils after this application method with regard to the high nutrient concentrations: “repacked to reach nutrient concentrations comparable to that in injection bands” (114). We also emphasised this statement in the abstract, now: “[. . .] the effect of high nutrient concentrations on N2 losses as they may appear after injection of BD [. . .]” (13 – 14), “Hence, we performed an incubation experiment with soil cores in a helium-oxygen atmosphere to examine the influence of soil substrate (loamy sand, clayey silt), water-filled pore space (WFPS; 35, 55, 75%) and application rate (0, 17.6 and 35.2 mL BD per soil core [250 cm^3]) on the emissions of N2O, N2 and CO2 after the application of high loads of BD” (14 – 17) and, finally, “Our results suggest a larger potential for N2O formation in the fine-textured clayey silt compared to the coarse loamy sand after applying high concentrations of BD as appearing after injection [. . .]” (22 – 24). Nevertheless, a reproduction of injection as done by Markfoged et al. (2011) is quiet impossible for our method due to the huge airtight apparatus which would be needed. This would be, again, rather an issue for a long-term study. However, to treat the present study like an immediate incorporation of a slurry band as recommended, would assume indeed unrealistic high application rates. Neverteless, we practiced injection of BD also in a field study (Fiedler and Jurasinski, 2015) and did not observe “very heterogeneous distribution of BD” as you stated in your review (which is another issue than the heterogeneity within BD). Additionally, we varied these nutrient concentrations with respect to a differencing spacing between injections rows, which result in a doubling of the application rate with a doubling of the row spacing to get the same applied amount per area, e.g. 160 kg N ha-1. Thus, yes, we had mixed twice as much

for the 320 kg N ha-1 treatment. However, we renamed the treatments (low BD and high BD, respectively) and gave the actual BD application per soil core (see above).

Likewise we tuned our conclusions down to the immediate observed effects and omit now the speculations about the overall emissions in field situations and recommend further investigations on this issue: “Nevertheless, our results show the need for further investigations on the dynamics and the duration of the observed effects and their significance for field conditions.” (25 – 26) Regarding the emission units, we would prefer to keep the reference to area. First, we would lose some comparability with other studies (e.g., Eickenscheidt et al., 2014) and any, though rough, estimation of area-related N2 emissions would become impossible. Second, if we were going to use a reference to mass, then a reference to dry matter of BD or the amount of applied N might be more useful, because the amount of soil does not change within each of both soil treatments. However, as our results show already now, there is no effect of the amount of BD on the emission rates. A new calculation may result in decreased rates per unit BD, but this finding could be discussed as is. Third, every approach that relies on a hypothetical mass of soil (or on area), which would be affected directly by BD application, has to be kept speculative as long as there is no comparable research on the effect of different injection techniques in situ.

Overall, we acknowledge some methodical pitfall we need to discuss, but we do not think that they justify such a rigorous rejection of our manuscript. First, the duration of our study was appropriate to catch the first emission peak: we did not examine the dynamics (which could last weeks), rather than the effect of different environmental conditions on the actual potential of N2 and N2O emissions. Second, we simulated the BD concentration in soil after injection rather than the injection band per se, which would not be realisable for an available Helium-Oxygen incubation system. However, we can comprehend your doubts about the limited relationships to real conditions in the field and would weaken our conclusions we draw, but it is difficult to perform such studies in situ, because current methods does not permit a direct measurement of

N2 fluxes hereunder. Therefore, the use of an artificial atmosphere is prerequisite to reproduce basic numbers, e.g., for a extrapolation to field by N2O/(N2 + N2O) product ratios.

Yours faithfully

Sebastian Fiedler and co-authors

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