Response to reviewer 2

Note: Reviewer titles (overview, specific comments) are shown in **bold**. Reviewer comments (RC) are enumerated, with corresponding *author comments (AC) in italic*.

Overview

It is true that a lack of measurement precision so far prevents a sound in-situ assessment on the role of nutrient-poor ecosystems in N_2O cycling and their potential to consume N_2O. E.g. in Huth et al. (2022) Restoration Ecology, 30, e13490, we found a tendency of N_2O uptake in Sphagnum-moss dominated plots, but due to the low precision of the GC sampling and measurements, we could not determine if that was actually significantly different from 0 or not. Therefore I much appreciate the efforts made by Triches et al. to test high-precision N_2O measurements in a low-flux environment as this will substantially help elucidating the role of northern nutrient-poor ecosystems in global N_2O cycling. The manuscript is generally well-written and fits nicely into the scope of the journal Atmospheric Measurement Techniques. I only had some minor comments and suggestions to make (especially in the discussion), except for the point that at very low N_2O fluxes, CO_2 uptake or efflux due to transparent or non-transparent chamber measurements could actually become a factor in either enriching or diluting N_2O during closure time. Since I'm wondering if this may explain the differing results between the transparent and non-transparent chamber measurements I would encourage the authors to use the Aeris CO_2 data and check, if a correction similar to water vapor would change the results. If CO_2 data is not available, I believe this should at least be thoroughly discussed.

We thank Dr. Huth for the kind words and thorough review, as well as for providing some relevant references that we had missed. We agree that possible interference of N_2O *and* CO_2 *fluxes during the chamber closure time may cause issues. We know that tests on the Aeris Mira Ultra* N_2O CO_2 *analyser have been conducted in 2023, showing a significant* N_2O CO_2 *crosstalk with a decrease of -0.008 ppb* N_2O *per ppm* CO_2*. Considering that the change in* CO_2 *concentrations we measured in the field are generally around 60-80 ppm with a simultaneous decrease of 0.6-0.7 ppb of* N_2O *(20 x 0.008 = 0.16), this suggests that the difference between dark and light cannot only be explained due to the sensors' interference. We have also analysed our* N_2O *and* CO_2 *data thoroughly and have observed some trends. However, we hope Dr. Huth will understand that*

this is a discussion for another manuscript, which is indeed in preparation. For the follow-up manuscript, we will, as suggested by Dr. Huth, add a small laboratory analysis to check how realistic CO_2 concentration changes affect the N_2O concentrations in our instrument.

Specific comments

Abstract:

RC 1. L.15-19: Please shortly mention your chamber height, because closure times are dependent on that.

AC 1. Thanks, we will make sure to mention it. Please see our abstract in the revised manuscript.

Introduction:

RC 2. L.33: Why not give credit to the early studies, e.g. Martikainen et al. (1993) Nature, 366, 51–53 or Nykänen et al. (1995) Journal of Biogeography, 22, 351–357.

AC 2. We agree, thank you. We will add the first suggested study as reference.

RC 3. L.34: If N availability is low, N_2O uptake might be expected (Buchen et al. 2019, Soil Biology and Biochemistry, 130, 63-72) but up to now it was extremely hard to detect, e.g. via Helium incubation studies (ibid). The value of this study to me is that the role of (northern) nutrient-poor ecosystems in N_2O cycling and potential uptake could now be elucidated.

AC 3. We will adjust the text accordingly in the introduction and conclusion: "Until about 15 years ago, only few studies investigated N_2O fluxes in the (sub) Arctic, where soils often have a very low availability of reactive N (Virkkala et al., 2024) and thus are not expected to emit amounts of N_2O relevant for the global climate (Voigt et al., 2020; Christensen et al., 1999; Grogan et al., 2004; Martikainen et al., 1993). In these low N ecosystems, N_2O uptake could be expected, but has, so far, not been confirmed in field studies (Buchen et al., 2019; Schlesinger, 2013)."

RC 4. L.61: Please add: "under a fixed chamber height", because closure times are directly depending on it (see Fiedler et al. 2022).

AC 4. Thanks for the reviewer comment, we will revise the manuscript as suggested.

RC 5. L.75: Was the chamber really dark? In general, the terms "light" and "dark" measurements can easily be misleading (e.g. our non-transparent chambers/our shading tarps are usually white to increase reflection and reduce chamber heating and I guess you did not really measure light, did you?), I would suggest you just use "transparent" and "non-transparent" (or "opaque") measurements/chambers etc. throughout the manuscript.

AC 5. Very good point. We only used transparent chambers; for the "dark" measurements, we indeed just added a reflecting tarp on top of the chamber, which also covered the PAR sensor. We agree that transparent and opaque is more easily understandable and inclusive, and will change the manuscript accordingly.

2 Methods

RC 6. L.166: Does the Aeris analyser do not give dry mole fractions of the target gas? If so, shortly mention here or where you introduce the analyser.

AC 6. It does. We will add this to the introduction of the analyser: "To measure N_2O *concentrations, we used the Aeris MIRA Ultra* N_2O*/*CO_2 *(from now onward: Aeris-*N_2O*) analyser (Aeris Technologies; sensitivity: 0.2 ppb/s for* CO_2 *and* N_2O*, frequency: 1 Hz). As most PGA, the Aeris-*N_2O *provides dry mole fractions of the target gas. We performed several laboratory tests to assess the signal stability (i.e., drifts and stabilisation time), uncertainties, noise level, and water interference of the Aeris-*N_2O*. The analyser was left to sample ambient air for approximately 15 hrs to evaluate the signal stability (see section 3.1)."*

3 Results and Discussion

RC 7. L. 226: Does this warming period occur every time once you turn on the analyser or was this first-use? Does this have implications for field application? I did not quite get if it is a problem, because at different RH, N_2O concentration seems to be stable. Please just quickly inform the reader, if the analyser warm up may pose a problem or not.

AC 7. This warming period occurs every time the analyser is turned on. The implications for the field applications are that we never turned off the instrument during the whole field campaign, which was recommended by the company and is an important information for the reader. We will add this information as follows at the end of chapter 3.1: "Overall,

our conducted laboratory tests indicated that the Aeris-N_2O was a suitable instrument for measuring low N_2O fluxes, showing low noise and water interference, along with negligible signal drift after the laser warms up. Nevertheless, the long warm-up period (approximately 5 h) of the analyser needs to be taken into account, as this can be a limiting factor for certain applications. To mitigate this, the Aeris-N_2O remained powered on throughout the whole field campaign."

RC 8. 240: It's called atmospheric sign convention and this could be stated already in the methods under your flux calculation procedure.

AC 8. Thanks. We will add this after the flux calculation equation: "To report our flux rates, we used the atmospheric sign convention, i. e., negative signs for an uptake of N_2O into the soil, and positive signs for emissions."

RC 9. 243-244: Complicated sentence and you already said in the methods, what a measurement period is. Please shorten and rephrase.

AC 9. We agree and will rephrase the whole paragraph to: "At our site, we commonly observed net N_2O consumption, suggesting an atmospheric sink, with a mean flux of -0.469 μg N_2O-N m^{-2} h^{-1}, with a 95% confidence interval (CI) of (-0.60,-0.3) during a chamber closure time of 10 min. Our calculated mean flux during transparent measurements was 0.361 μg N_2O-N m^{-2} h^{-1}, with a 95% CI of (0.24,0.48) during a chamber closure time of 10 min (Table **??***). For opaque measurements, our calculated flux was -1.29 μg N_2O-N m^{-2} h^{-1}, with a 95% CI of (-1.45,-1.13), indicating that our opaque measurements represent a real biochemical process, rather than an experimental artefact, in the (sub-) Arctic ecosystem. Nevertheless, the impact of environmental drivers on N_2O fluxes, including the transparent and opaque measurements, is beyond the scope of this study. Overall, we collected 338 samples, with 60-90 % of N_2O fluxes above the detectable limit. We therefore also acknowledge the possibility of unknown chamber artefacts that may remain undiscovered and could affect the interpretation of our data."*

RC 10. L.245ff: This is my major point that needs some attention and discussion: At low N_2O concentration changes,CO_2 concentration changes due to respiration (non-transparent measurments: N_2O gets diluted, indicates uptake) or net uptake (transparent measurements: N_2O gets enriched, indicates efflux) might become a factor. Either recalculate fluxes with a

CO_2 correction (like for water vapour), or add a short paragraph on this topic. If it is easily doable for you, you might also check in the lab, how realistic CO_2 concentration changes affect N_2O concentration and add it to the manuscript.

AC 10. We thank Dr. Huth for the detailed description of his major point. For its discussion, we would like to refer to our response to his overview comment (see above, lines 17ff).

RC 11. L314ff: Yes, but it also could just indicate CO_2 saturation during closure time, hence a decrease in N_2O dilution.

AC 11. Please see our response to the overview comment (lines 17ff).

RC 12. L317: It's not really due to fewer sampling points but rather due to the lower absolute change in concentration, that is below the one needed for flux detection see also Fiedler et al. (2022)

AC 11. Correct. We will rephrase to: "In contrast to this, as mentioned above, more than 40% of the fluxes were below the MDF at 3-minute closure time. This confirms that very short closure times can lead to higher uncertainties of flux estimates because the concentration changes are too small to be accurately detected (Fiedler et al., 2022)."

RC 13. L320: It is not the chamber size, it's (effective) chamber height or V:A-ratio, that is determining concentration change and measurement length.

AC 13. Correct, thank you. We will rephrase to: "The optimal closure time depends on factors such as chamber height, micro habitat, and the duration of the field campaign."

RC 14. L321: Jungkunst et al. (2018) Journal of Plant Nutrition and Soil Science, 181, 7-11 actually assessed the trade off between reducing temporal accuracy of flux measurements to gain more spatial replicates. Might want to cite this here.

AC 14. Thank you, we will add it.

RC 15. L336: What do you mean by LM and non-LM models exclude each other? The common approach is that if the difference between the two is non-significant, the simpler model should be used.

AC 15. Thank you for pointing out this formulation. We will adjust the whole paragraph as follows: "Linear and non-linear models for concentration data during chamber closure may typically be seen as alternatives, not complementary

approaches. However, the non-linear fitting includes the linear fitting as a special case. When using a generic exponential function ae^{-bt} to fit data, where a and b are positive constants to be fitted, it can be approximated to a linear function if the data points are distributed linearly. This is because the exponential function can be expanded as a series, and when the rate constant b is small, the linear function dominates. Namely, the first three terms of the serial expansion are $a(1-bt+(bt)^2/2$, but when b is very small, i.e., $b << 1$, it is reduced to $a(1-bt)$, which is the linear form. The slope of the linear term is $-ab$; if we take the time derivative of the original exponential function to calculate the slope, it gives$-abe^{-bt}$. When we expand it as a series and only take the first order term as $b << 1$, we again obtain the simplified $-ab$ as slope. This means that if the data points are linear, the exponential fitting will automatically reduce to a linear fitting with the same slope. With our results, we show that for N_2O fluxes, indeed, flux estimates were reduced to the linear model and yielded identical results as the non-linear model."

RC 16. L343: Again, I don't understand this statement. If the data does not show non-linearity, linear models are sufficient. In any case, this statement should be rephrased, because I don't get how you assess that the relation between exponential and linear models do not appear to be recognised within the chamber community.

AC 16. We agree and refer to the AC 15 above.

RC 17. L350ff: That is true, but in theory, calculating fluxes from closed-chamber measurements actually assume non-disturbance conditions and non-linear models are a tool to calculate fluxes from disturbed measurements. Therefore it is also justifiable to reduce chamber closure time to the most linear part, because this signifies non-disturbance in your measurement. In other words, if concentration change is significantly different from linearity, chamber closure time was too long (or it wasn't properly sealed etc.).

AC 17. Thank you for this comment. We agree that these are the assumptions for closed-chamber measurements and we do not question the practice of reducing chamber closure times to the most linear part; indeed, we encourage short chamber closure times. However, several studies assessing these assumptions have concluded that chamber measurements are sensitive to errors (e.g., (Kutzbach et al., 2007; Pavelka et al., 2018)). On top of that, there are only few data sets addressing N_2O fluxes from nutrient-poor ecosystems and the high spatial and temporal variability of these fluxes

makes it challenging to assess them (Butterbach-Bahl et al., 2013). In our data, there is no evidence for oversaturation in the head space, i.e., too long chamber closure times, but non-linearity could still be observed. We further argue that non-linearity can be a result of a concentration change of the flux only, which can be corrected with non-linear fitting. This is why we encourage future research to include non-linear flux calculation to their linear calculation, and, ideally, justify their model choice with some metric.

RC 18. L365ff: Does accuracy increase due to the fact that more GC samples better represent non-linearity of the data? Please discuss!

AC 18. Thanks for the comment. We will address this in this statement: "The underestimation of GC fluxes may occur as a result of a smoothed out curve: when only few data points are available, variations in curves are naturally reduced. Furthermore, the precision of our GC was 1.9 ppb compared to 0.2 ppb of the Aeris-$\mathrm{N_2O}$*, resulting in less accurate measurements of the* $\mathrm{N_2O}$ *concentrations. This may lead to a loss of detail in the curve, particularly in the peak values of the* $\mathrm{N_2O}$ *concentrations, which can result in underestimation of the flux."*

RC 19. L.394: which = with?

AC 19. Thanks, we will revise the manuscript as follows: "Second, low $\mathrm{N_2O}$ *fluxes tend to be very scattered, with large noise in comparison to the actual trend, i.e., the change in concentration during chamber closure."*

4 Conclusion

RC 20. Much of this is a repetition from previous paragraphs. Consider shortening and focusing on the main outcomes and recommendations.

AC 20. Thank you. We will revise the conclusion accordingly, so that it includes the main outcomes, such as

- *the laboratory work, showing that with the Aeris-*$\mathrm{N_2O}$ *and our manual chamber system, we are able to report very low* $\mathrm{N_2O}$*fluxes,*
- *our recommended chamber closure times, which can differ for transparent and opaque measurements,*
- *reasons for using* ***all data points*** *when calculating flux estimates using the HM model.*

We further aim to give recommendations on why PGAs should be considered for future N_2O flux studies, and try to encourage future research on N_2O fluxes in nutrient-poor ecosystems. Please see our conclusions in the revised manuscript.

Appendix

RC 21. Figure A3: That's a really nice figure that I believe would be well-placed within the main text.

AC 21. Thank you. We will place figure A3 within the main text.

References

Buchen, C., Roobroeck, D., Augustin, J., Behrendt, U., Boeckx, P., and Ulrich, A.: High N2O Consumption Potential of Weakly Disturbed Fen Mires with Dissimilar Denitrifier Community Structure, Soil Biology and Biochemistry, 130, 63–72, https://doi.org/10.1016/j.soilbio.2018.12.001, 2019.

Butterbach-Bahl, K., Baggs, E. M., Dannenmann, M., Kiese, R., and Zechmeister-Boltenstern, S.: Nitrous oxide emissions from soils: how well do we understand the processes and their controls?, Philosophical Transactions of the Royal Society B: Biological Sciences, 368, 20130 122, https://doi.org/10.1098/rstb.2013.0122, number: 1621, 2013.

Christensen, T. R., Michelsen, A., and Jonasson, S.: Exchange of CH4 and N2O in a subarctic heath soil: effects of inorganic N and P and amino acid addition, Soil Biology and Biochemistry, 31, 637–641, 1999.

Fiedler, J., Fuß, R., Glatzel, S., Hagemann, U., Huth, V., Jordan, S., Jurasinski, G., Kutzbach, L., Maier, M., Schäfer, K., Weber, T., and Weymann, D.: BEST PRACTICE GUIDELINE: Measurement of carbon dioxide, methane and nitrous oxide fluxes between soil-vegetation-systems and the atmosphere using non-steady state chambers, 2022.

Grogan, P., Michelsen, A., Ambus, P., and Jonasson, S.: Freeze–thaw regime effects on carbon and nitrogen dynamics in sub-arctic heath tundra mesocosms, Soil Biology and Biochemistry, 36, 641–654, https://doi.org/10.1016/j.soilbio.2003.12.007, 2004.

Kutzbach, L., Schneider, J., Sachs, T., Giebels, M., Nykänen, H., Shurpali, N. J., Martikainen, P. J., Alm, J., and Wilmking, M.: CO₂ flux determination by closed-chamber methods can be seriously biased by inappropriate application of linear regression, Biogeosciences, 4, 1005–1025, https://doi.org/10.5194/bg-4-1005-2007, 2007.

Martikainen, P. J., Nykänen, H., Crill, P., and Silvola, J.: Effect of a Lowered Water Table on Nitrous Oxide Fluxes from Northern Peatlands, Nature, 366, 51–53, https://doi.org/10.1038/366051a0, 1993.

Pavelka, M., Acosta, M., Kiese, R., Altimir, N., Brümmer, C., Crill, P., Darenova, E., Fuß, R., Gielen, B., Graf, A., Klemedtsson, L., Lohila, A., Longdoz, B., Lindroth, A., Nilsson, M., Jiménez, S. M., Merbold, L., Montagnani, L., Peichl, M., Pihlatie, M., Pumpanen, J., Ortiz, P. S., Silvennoinen, H., Skiba, U., Vestin, P., Weslien, P., Janous, D., and Kutsch, W.: Standardisation of chamber technique for CO2, N2O and CH4 fluxes measurements from terrestrial ecosystems, International Agrophysics, 32, 569–587, https://doi.org/10.1515/intag-2017-0045, number: 4, 2018.

Schlesinger, W. H.: An Estimate of the Global Sink for Nitrous Oxide in Soils, Global Change Biology, 19, 2929–2931, https://doi.org/10.1111/gcb.12239, 2013.

Virkkala, A.-M., Niittynen, P., Kemppinen, J., Marushchak, M. E., Voigt, C., Hensgens, G., Kerttula, J., Happonen, K., Tyystjärvi, V., Biasi, C., Hultman, J., Rinne, J., and Luoto, M.: High-resolution spatial patterns and drivers of terrestrial ecosystem carbon dioxide, methane, and nitrous oxide fluxes in the tundra, Biogeosciences, 21, 335–355, https://doi.org/10.5194/bg-21-335-2024, 2024.

Voigt, C., Marushchak, M. E., Abbott, B. W., Biasi, C., Elberling, B., Siciliano, S. D., Sonnentag, O., Stewart, K. J., Yang, Y., and Martikainen, P. J.: Nitrous oxide emissions from permafrost-affected soils, Nature Reviews Earth & Environment, 1, 420–434, https://doi.org/10.1038/s43017-020-0063-9, 2020.