

Biogeosciences Discuss., referee comment RC1  
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## Comment on bg-2022-76

Anonymous Referee #1

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Referee comment on "Spatial and temporal variation of delta13C values of methane emitted from a temperate mire: Methanogenesis, methanotrophy, and hysteresis" by Janne Rinne et al., Biogeosciences Discuss., <https://doi.org/10.5194/bg-2022-76-RC1>, 2022

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### General Comments

The authors present an interesting and valuable dataset showing temporal and spatial patterns of mire methane flux and its  $^{13}\text{C}$  signature. They aimed to disentangle the relative importance of methanotrophy vs methanogenesis as well as the availability of substrates for methanogenesis for explaining temporal and spatial variability in their data. Secondary goals were to describe the methane associated prokaryotic community and compare the mire-level  $^{13}\text{C}$  signature from upscaled measurements (using their chambers and a land cover map for the mire derived from a previous study) to nocturnal boundary layer measurements. While the data itself are useful, and the upscaled  $^{13}\text{C}$  method successful, there are substantial issues. Primarily that the data presented are insufficient to fully test their hypotheses.

### Spatial (HS1 and HS2):

HS1 proposes that variation (in methane flux and  $^{13}\text{C}$  signature) is due to spatial changes in methane consumption, while HS2 proposes variation is due to spatial shifts from hydrogenotrophic to acetoclastic methane production.

These are not mutually exclusive, which is not inherently an issue, although they are treated as such in the study. Without information on the spatial distribution of methanotrophs or the respective groups of methanogens (or their substrates) or well-constrained values for the expected  $^{13}\text{C}$  signatures from individual processes, any conclusions on their relative contribution to the data is conjecture.

## Temporal (HT1 – HT3)

HT1 states that temporal variation (in methane flux and  $^{13}\text{C}$  signature) is driven by temperature. HT2 proposes temporal shifts from hydrogenotrophic to acetoclastic methane production. HT3 wisely combines them and proposes there will be a time lag between temperature and production of substrate (presumably of acetate) in the ecosystem that produces a hysteretic out and back arc in the data.

The presence of HT3 resolves the issue of exclusivity there, however the issue of being able to ascribe  $^{13}\text{C}$  signature changes to changes in microbial processes without any constraining values or direct measurement of those processes remains. Additionally, although the evidence from their data is evenly split between a temperature-driven response (see point clouds in Figure 9) and a response indicative of hysteresis, they conclude that HT3 is supported.

While the dataset is strong, its strength is mainly in describing spatial and temporal variation in methane flux and signature. If data are available from the site on the  $^{13}\text{C}$  signature of soil C, this might be enough to draw conclusions on microbial processes based on assumptions regarding fractionation rates. Otherwise, the framing of the study should shift to focus on its strengths; I can imagine an analysis of hotspots and hot moments, and/or the relationship between fluxes, vegetative cover, and water depth.

### Specific comments:

Additionally, there are a few issues that are reducing the clarity of the authors' message. One is the use of the phrase "trophic status", which is used in the manuscript to indicate both seasonal build-up of plant-derived carbon as well as the metabolic pathway of methanogenic archaea. Neither of these is likely the default interpretation that readers will be using when they first encounter the phrase. Distinct phrases should be used (and explained on first use) for these two phenomena and the link between them should be made explicit. Another issue is the description of the Keeling plot method, which currently leaves the reader to put together that the mixing ratio is based on the up-scaled land cover values, unless my interpretation is widely off-base (see L168 & 219). Please clarify this. Finally, the formatting for different taxonomic levels is non-standard and inconsistent throughout the manuscript.

Technical corrections:

66 Replace "mires" with "wetlands", as this statement applies to all wetlands ecosystems, rather than mires specifically

71 This implies the phase-change fractionation leads to biological (or kinetic) fraction, which is not true. It would be more accurate to describe biotic and abiotic fraction processes as just that, two separate chemical phenomena

74 Introduce the reader to what makes a mire ecosystem distinct from other wetland types here

93 Alternative to what?

175 Is this plot within the chamber itself or around the chamber?

189 Define RMSE

207-214 unit formatting, "spectrometer"

239 Totally fine to use gDNA as shorthand for "genomic DNA", but it should be defined on first use.

268 Spatial and temporal fluxes/signatures as well? This is unclear because of the lack of a separate statistical analysis section

Figure 8 takes up a lot of space it is, without providing a lot of information. The 10 day periods could be further collapsed into 4 blocks throughout the growing season, perhaps with a trend line. Or a subset of representative panels could be shown and the remainder moved to a supplement.

374 Is this mire nearby, how different is the climate/vegetation? Some quantitative context for the comparison would be helpful. Also, is an overlap of one methanogenic genus meaningful? It seems likely be mere chance.

