## **Response to Darcy Rush.**

On behalf of all co-authors, I would like to thank Darcy Rush for her helpful and positive comments. Below are listed our answers to her comments, including changes in the text.

This paper by Coffinet et al., entitled "Structural elucidation and environmental distributions of butanetriol and pentanetriol dialkyl glycerol tetraethers (BDGTs and PDGTs)" employs 2-dimensional nuclear magnetic resonance (2D-NMR) to confirm the structure of a previously unconfirmed lipids: BDGT. Furthermore, by investigating the 13C isotopic composition of biphytanes originating from these lipids in marine sediments, and comparing them with potential carbon sources (CH4, TOC, DIC), the authors go on to suggest potential source organisms, or rather carbon metabolisms, of these lipids. The authors do well in not over-interpreting their environmental results. They speculate that BDGTs and PDGTs likely are produced by a diverse archaeal community, and not just one confirmed synthesizer, the methanogen Methanomassiliicoccus luminyensis. I appreciate that they go on to suggest why these lipids may be found in such a range of archaea: BDGTs and PDGTs might boost membrane stability under low energy conditions found in deep sediments. Overall, I find this to be a useful paper for the community in general, and worthy of publication in Biogeosciences.

There are a couple of points that I would like the authors to address, however. I find the structural elucidation of BDGT-0 to be the most important aspect of this study. Far too few NMR experiments are performed in organic geochemistry, as these involve a lot of effort and lab hours and often we do not have enough starting material. I appreciate that the authors took the time to undertake the work here. However, I feel that they are doing themselves a disservice by not publishing the 2D correlation results of these experiments. They report having performed COSY, TOCSY, HMBC, and HSQC experiments. I suggest including at least a couple of these in Table 1. For example, they refer to the HMBC and HSQC results (line 171) confirming a butanetriol end group, and for clarity/transparency, I feel it would be useful to have these included in Table 1.

We agree with Darcy Rush and will add those results, which allowed us to confirm the butanetriol backbone, in Table 1 as an additional column.

Furthermore, for some of the most important hydrogen shifts at the very least, I suggest that they also include the coupling constants (J) and multiplicities, especially for those of the "C" carbons, which are the unusual components of this molecule.

There is severe overlap in the  $1D^{-1}H$  dimension that precluded reliable extraction of coupling constants (the de-symmetrisation of BDGT-0 compared to GDGT-0 leads to extensive overlap). This is the reason why we did not report J-coupling constants. Nevertheless, we will now include a  $1D^{-1}H$  spectrum as a supplementary figure, which clarifies this point. We will also include several other J-coupling constant values with cautionary notes to the table. This includes the C4' methyl group that is an overlapped doublet.

Finally, the number of significant figures on the hydrogens associated with A4, A4' etc is more (i.e. 1.022 - 1.105) than for that of all the other reported hydrogen shifts. Why is this? I also found a couple of lost 0s in the table (likely an artifact of program used to generate the table).

We thank Darcy Rush for spotting this and have corrected significant figures and formatting issues. Table 1 will be modified to enhance clarity.

## Minor comments:

Line 9: Czech Republic?

The Czech government adopted "Czechia" as the official name of the country in English in 2016 and this is consistent with the designation by the UN's database for official country names (cf. United Nations Group of Experts on Geographical Names, UNGEGN List of Country Names (New York: United Nations, 2017) available from unstats.un.org/unsd/geoinfo/UNGEGN/docs/11th-uncsgn-docs/E\_Conf.105\_13\_CRP.13\_15\_UNGEGN%20WG%20Country%20Names%20Document.pdf).

Line 29: Consider replacing amphiphilicity with "amphiphilic nature" Line 31: sn should be in italics Line 52: lipids Line 52: currently the only

Line 55: capacity change to capability

Line 134: as follows

Line 261: , as well as the absence of PDGTs

Line 264: a higher relative

Sentence starting line 317: There are two "therebys" in this sentence. Consider removing one.

Line 327: Thus, it seems likely

Caption for Figure 5: The last sentence should mention that the calculation of \_13CCO2 using the fractionation -10.7‰ comes from the measured values of DIC. This is clear in the text, but not here. *These comments will all be taken into account in the revised manuscript.*