

## ***Interactive comment on “Microbial activity, methane production, and carbon storage in Early Holocene North Sea peats” by Tanya J. R. Lippmann et al.***

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General comments: The authors have done a lot of work creating datasets to characterise the geochemistry, biogeochemistry, botany, microbial community, and carbon of submerged peats from the southern North Sea. Marine sediments are an understudied aspect of C cycle and climate and specifically the area of the North sea investigated here with its sandwiched layer of peat. The carbon pool of the region is estimated and potential emissions calculated. The potential for biological conversion of this carbon pool to CH<sub>4</sub> is analysed via microbial taxonomic survey and incubations. This study shows a decoupling between CH<sub>4</sub> production and C storage and posits that there is

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threat of re-coupling.

In general, the ideas are clearly defined though a little more work connecting ideas within paragraphs in the introduction and connecting the theory presented in the introduction to what was done in the study will assist readers in following the work. Further, adding explanations and justification for method choices will also assist readers. It is currently unclear why certain data was collected and analyses done. Some of the datasets are underutilized. Analyses connecting the different datasets could increase the value of the manuscript (MS) to a variety of audiences. There are a few instances in the discussion that reference data or results not detailed in the results section.

Specific comments: Figure 1A states that the southern North sea was inundated due to anthropogenic caused sea level rise. Is this a typo? Figure 1C would be great if it was even higher resolution covering just the section sampled ie the rectangle in 1B, with sample locations marked. Maybe even the location of the 'special' samples coded in a different colour or symbol. Just to help the readers visualize what was done.

The methods section, especially the computational description needs more detail (or citations) and should include versions of software used and parameters chosen. -Why was sequencing and culture carried out on different samples/cores? It would make more sense to survey the community that was the base for incubations. Please provide justification for this choice. -I also ask for justification for choice of 60°C annealing temperature for the initial amplification. Please also check the citation for the bacterial reverse primer it is the same paper as for the forward. -I could not find the deposited sequence data as there was no listing in Genbank found for the BioProject identifier. -Why was a qPCR carried out? -2.6.3 why is cloning mentioned in the subtitle? Was cloning done? Please provide method, results, and justification for using this method. And integrate into results, discussion and conclusion. -The link to the core data also is not yet working.

I believe that the physical, chemical, botanical, and radiocarbon dating (others?) were

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all done in order to establish how and when the peats formed and maybe what quality of carbon they hold. A large portion of the MS describes sampling, testing, physical qualities of the cores so it would be worth stating why these attributes were analysed as I did not notice this explicitly stated anywhere. Providing justification and motivation for choices will help the reader (who is unlikely to have the same level of expertise as the authorship team) to understand the work.

Published literature documents both co-occurrence and (spatial and temporal) separation of methanogenesis and methanotrophy. There is also substantial literature on the ANME archaea which I did not notice specifically and clearly mentioned in this MS. Here are some randomly chosen non-exhaustive examples for your consideration: <https://sfamjournals.onlinelibrary.wiley.com/doi/10.1111/1462-2920.13096> <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5104750/> <https://pubmed.ncbi.nlm.nih.gov/30664670/> <https://aem.asm.org/content/74/13/3985>

I would love to see greater use made of the core chemical data. Perhaps a multivariate approach comparing microbial community membership to chemistry would be very interesting and tie together major components of the data presented here. Similarly, there are a few places in the MS where the C, CH<sub>4</sub>, or organic content of the peats is referred to in comparison to microbes but I did not notice a statistical analysis to back up any comparison. This could add value to the MS.

I have concerns about the NMDSs presented. My reading of the manuscript is that there were 12 samples sequenced. On an NMDS where the samples are mapped onto species space there should therefore only be 12 dots. Please provide details of computational methods used so that what has been plotted on the NMDS is understood. For an example of the level of method detail required and correct plotting of an NMDS see e.g. <https://www.pnas.org/content/115/47/11994>

The MS states early on that it looks at C storage and CH<sub>4</sub> seepage/accumulation etc. Please check the MS for typos 'CH<sub>4</sub> storage' or do you have evidence that CH<sub>4</sub> is

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trapped in the peat? Is that why the term CH<sub>4</sub> storage is used? I would have guessed that the CH<sub>4</sub> currently in the peat deposits can bubble up through the overlying clay and sands into the water column where (depending on factors that might be worth listing) it is consumed by methanotrophs in the water column or emitted to the atmosphere. This could make an interesting discussion point for this MS.

There is a statement in the abstract and conclusion that the C in the peats could be converted to CH<sub>4</sub> under other circumstances. What other circumstances? Your MS shows and states that the remaining C is not accessible to methanogens so what would make it available? This would be an interesting discussion point.

Technical corrections: -Community structure was not studied. Community membership was, please change this throughout MS. -There is a mix of 'methane' and 'CH<sub>4</sub>' throughout the MS please pick one. -Ln 779 methanogenic bacteria – is this a typo? -'activity assay' refers to e.g testing catalase activity in a lab. This study documents incubations not activity assays. Please be careful about using the word 'activity' (including in the title) throughout the MS.

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