

Interactive comment on “Vertical profiles of sediment methanogenic potential and communities in two plateau freshwater lakes” by Yuyin Yang et al.

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1) General comments

This manuscript describes and compares methanogenesis in sediments from an eutrophic and a mesotrophic lake, with a focus on methanogens potential activity, abundance and diversity along vertical profiles. The results presented here are of interest. However, this manuscript could be significantly improved. Indeed, the current version

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suffers from (i) a lack of general conclusion (i.e. the discussion and conclusion are superficial), (ii) a writing style that is sometimes confusing for the reader, (iii) poor-quality figures, and (iv) a lack of geochemical background. Concerning this last point, the geochemical background is available in the SI but totally absent from the main text and the discussion. This information could significantly help the discussion and the interpretation of the results.

2) Specific comments

a. Abstract

- l. 25-26: “changes of methanogens”. Do you mean change in abundance or taxonomic change? Please be more specific.
- l. 35-36: For each lake, specify if it is mesotrophic or eutrophic.
- l. 33-43: This second part of the abstract looks like a list of results. It would be nice to have here some concluding remarks, i.e. put these results in the context of what we know about methanogenesis in these environments.
- l. 46-47: I am not sure that it makes sense to choose keywords that already appear in the title and/or in the abstract.

b. Introduction

- l. 77-78: This short description of methanogenesis pathways is over-simplified. Please expand or at least specify that those are the main pathways (and not the only ones).
- l. 81: What do you mean here by “theoretical ratio”. Do you mean that we consider that, in most of natural environments, 2 molecules of CH₄ are produced by acetoclastic methanogen per molecule of CH₄ produced by hydrogenotrophic methanogens? Please be more specific.
- l.85-86: Does the 16S or mcrA sequencing give some insights about the methano-

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genesis pathway?

- l. 93: What is a humic lake? Could you please define it?
- l. 96: What do you mean by substrate here? Are you talking about the source of electron (hydrogen, acetate)?

c. Materials and methods

- l. 108-109: Here we are missing some background information about these 2 lakes. I propose the following: Indicate where these 2 lakes are located on a map, and describe them by listing some key features (that make these lakes eutrophic or mesotrophic, and/or that are relevant for this study). This information could be included in the figure containing the map.
- l. 113-114: Is this information relevant to the study?
- l. 120: The physicochemical analyses are simply absent from the manuscript. They should be included in the main text and more importantly in the discussion.
- l. 125-127: At this point, we don't want to know where to find the results, but how did you proceed to obtain these results.
- l. 137: What is the final CH₃F concentration in the incubations?
- l. 130-141: How did you calculate the rates from these incubations?
- l. 144-161: What is the qPCR efficiency and how was inhibition tested?
- l. 166: What kind of triplicates are you talking about? Biological, technical?
- l. 170-171: "After subsampling to the lowest number of sequences". What do you mean here?
- l. 163-176: Did you check your sequences for chimeras?
- l. 174-176: Which method did you use for taxonomic annotation (Silva is only a

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database)

- l. 180: Which method did you use for detecting chimeras?
- l. 186-187: The PCoA and environment clusters analysis are missing from the manuscript.

d. Results

- l. 195: I guess DW stands for dry weight. You should mention in the methods that the rates were calculated relative to the dry weight of sediments (and how you measured the dry weight of the sediments).
- l. 201: I think you mean HMP, not MPP.
- l. 204-207: It would be nice to see these results. A plot including MPP, AMP and HMP would help the reader to understand the results.
- l. 226: What is this normalization?
- l. 228: What is exactly the library coverage? I guess it's the proportion of 16S from the community that was sequenced. How was it calculated?
- l. 251: If you use this annotation, you should define it somewhere (in the Methods).
- l. 268-271: Which taxonomic order these genera belong to?
- l. 303-304: Which clustering method was used here?

e. Discussion

- l. 320-467: Here the structure chosen by the author for comparing their results with the ones from other previous studies looks a bit strange to me. First, they describe what was observed in other studies, and then, in a second sentence, they summarize what they found. This is confusing for the reader. I would do it the other way around, i.e. "we observed this, which is consistent (or not) with previous observations". - l. 320: In which kind of environment are you referring to?

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- l. 334-335: What do you mean by geological constitute, geographic regions and water type. Please be more specific and/or develop a little bit.
- l. 339-341: What is the correlation between TOC (and other environmental parameters) and MPP, HMP and AMP?
- l. 343-346: Did you find similar OTUs between these 2 lakes sediments?
- l. 357: Please develop.
- l. 359-361: I don't understand what the authors mean here.
- l. 365-366: This is not clear. Should we conclude that shallow and eutrophic lakes are not stratified?
- l. 370-371: Please develop here a bit more.
- l. 373-374: How is it possible to assess the abundance of methanotrophs with qPCR based on archaeal 16S primers? This makes no sense to me.
- l. 377: Consistent with which results?
- l. 379-380: What is the range of archaeal abundance described in the literature?
- l. 383: Different archaeal primers combinations were used for 16S quantification and sequencing. That could be an explanation for the different ratios observed.
- l. 383-384: Do you mean that organisms can have multiple copies of *mcrA*? Or of 16S rRNA gene? Not really clear. Please develop and back-up with genomic data from literature.
- l. 394-395: "but the change pattern. . . was not clear". I don't understand it. Can you clarify?
- l. 397-398: Can you point out the results you are discussing here?
- l. 418: Could you briefly introduce Taihu Lake?

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- l. 420: "remarkably" is too strong here. I don't find this result very surprising. . .
- l. 420: Please point out a figure to illustrate these findings.
- l. 412: Where do you see that depth is a key factor for archaeal community structure?
- l. 425-426: It seems that you are describing quantitative changes. But are they qualitative changes as well (at the genus level)?
- l. 437-440: I would like to see this data.
- l. 442: What do you mean by "the seventh"? The 7th to be discovered or the 7th most important? Not clear.
- l. 454-461: The congruency of phylogenetic trees does not depend on the relative proportion of the different OTUs. In fact, this has not impact. Do the 2 different trees have the same shape? Are the clusters the same between these 2 trees?

f. Figures

- Fig. 1 and 2: Flip over the axis to have the depth as vertical and descending axis. Precise which lake is eutrophic and which one is mesotrophic.
- Fig. 1: Merge the 2 plots so it's easier to compare the results. Indicates MPP, AMP and HMP, and the AMP/HMP ratio.
- Fig. 2: Use log scale for gene abundance.
- Fig. 3: You should make this figure more informative. I suggest the following: you could start from the genus level. You indicate only genera that represent at least few % of the archaeal community (define a threshold) and merge the ones below this threshold in a category called "other <family name>". Then you do the same with the next taxonomic level (family), merging the small ones into the higher level (i.e. class), and so one. It's a little bit more of work to produce this figure, but at the end you have a plot that indicates the dominating groups, independently of the taxonomic level. An

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alternative is the Krona charts (<https://sourceforge.net/p/krona/home/krona/>).

- Fig. 4: Highlight the E and D samples with a color code.
- Fig. 5: The horizontal distances are not defined. Please use a color-code as well.

3) Technical corrections

a. Abstract

- l. 25: "layer depth" is confusing. Please change it for "depth" throughout the whole document.

b. Results

- l. 191-193: These 2 first sentences are not clear. You should re-write them as follow: "In this study, MPP varied remarkably with both lake and sediment depth except for the uppermost sediments layers which are remarkably similar between the two lakes (Figure 1)."
- l. 213: Remove "However".
- l. 214: Rephrase "and showed an increase with depth. . ."
- l. 220: Which depth these samples (D6 and E5) correspond to?

c. Discussion

- l. 379: Please rephrase: ". . .the abundance of archaea. . ."
- l. 381: Specify here that this first ratio was calculated with qPCR results.

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