

## ***Interactive comment on “Lack of <sup>13</sup>C-label incorporation suggests low turnover rates of thaumarchaeal intact polar tetraether lipids in sediments from the Iceland Shelf” by S. K. Lengger et al.***

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# General comments:

Thank you for giving an opportunity to read the Research Article entitled, “Lack of <sup>13</sup>C-label incorporation suggests low turnover rates of thaumarchaeal intact polar tetraether lipids in sediments from the Iceland Shelf”, by S.K. Lengger et al. The authors tackled the important issue with relevance to the role of carbon cycle mediated by putative

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Thaumarchaeota in deep-sea sediment. The stable isotope probing (SIP) by <sup>13</sup>C-labelled bicarbonate, pyruvate, amino acids and glucose are unique experimental set up to understand enigmatic benthic archaeal behaviors.

Potentially, the entire story is of wide interest in biogeochemistry and microbial ecology for the research of deep-sea carbon cycles. The paper is mostly well written (especially for method section, including Figures and Table data), however, I have some concerns as follows. I would like to recommend publication in Biogeosciences, however, the author needs minor revision on a point-by-point basis.

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# Minor concerns and specific issues to address:

[1] What is the fraction (%) of benthic Thaumarchaeota relatives to total sedimentary archaeal community ?

I totally agree with the context of lines 1-9 on page 12809. However, the fraction of benthic Thaumarchaeota in the present sample is still unclear through the manuscript. How much is the contribution of Thaumarchaeota in the sedimentary archaeal community ? I guess there are other major benthic groups including Miscellaneous Crenarchaeotic Group (MCG) and marine benthic group (MBG) in the sediment, while I understand the lipid-based rough estimation (probably, mixing with planktonic lipids) for crenarchaeol as noted in lines 21-26 on page 12821.

For example, please see: Lloyd et al., (2013) Nature, 496, 215-218. Kubo et al., (2012) ISME Journal, 6, 1949-1965. Gagen et al., (2013) Applied Environ Microbiol., doi: 10.1128/AEM.02153-13.

I think the author should state this critical description in the Material and Method, for example, based on quantitative polymerase chain reaction (qPCR) and 16S ribosomal RNA. Alternatively, I would suggest that the title could be changed without the term of Thaumarchaeal, i.e., “Lack of <sup>13</sup>C-label incorporation suggests low turnover rates

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of benthic archaeal intact polar tetraether lipids in sediments from the Iceland Shelf”, which is more simple and comprehensive.

[2] Can the  $^{13}\text{C}$ -SIP experiment up to 6 days simulate actual in-situ physiological condition for sub-surface archaeal community ?

I think the author need to state clearly that laboratory  $^{13}\text{C}$ -incubation experiments are not lumped in the same category as in-situ experiments because most of benthic archaea (including sedimentary Thaumarchaeota, MCG and MBG) are mostly unculturable by conventional laboratory-based studies. Laboratory-based  $^{13}\text{C}$ -SIP incubation studies are, therefore, different from in-situ  $^{13}\text{C}$ -SIP studies due to changing microbial physiological status and the unnatural physico-chemical factors.

Another minor concern is the incubation time up to 4-6 days. I understood the author's notion about the time scale in line 20 on page 12824 and the logic for estimation of the turn over rate. However, for example, considering about Lin et al. Environ Microbiol (2013) for 468 days incubation, I suppose that 4-6 days may not be a significant incubation span and too narrow slice of time to demonstrate  $^{13}\text{C}$ -uptaking behaviors for deep-sea uncultured benthic microbial community. The readers also may ask and wonder if, “why only several days ? ”. I think the author needs to address this point.

[3] Is there rapid response of benthic Thaumarchaeota ?

Please note that Takano et al. Nature Geosci. (2010) reported the rapid response of benthic Thaumarchaeota (described as Marine Group I, determined by qPCR and 16S rRNA) by the in-situ  $^{13}\text{C}$ -glucose incubation experiments during initial 9 days. Could you mention this rapid blooming event driven by benthic Thaumarchaeota ? If there were similar responses in the present approach by S. Lengger and co-workers, please describe whether it occurred in the deep-sea sediment. I expect the most active layer for benthic Thaumarchaeota is core-top section (i.e., sediment-water interface).

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# Other comments:

P12809, L19, L27:

Need to check the typo, Thaumarchaeote should be Thaumarchaeota.

P12810, L2:

There is no reference list for Ouverney et al. (2000). Is that Ouverney and Fuhrman (2000) ? The author's confirmation is needed.

P12811, L4-8:

This is somewhat misleading introduction. Please note that Takano et al. (2010) observed  $^{13}\text{C}$ -incorporation processes in benthic archaeal biphytanyl chains up to -9.3‰ (vs. PDB) during 9 days in-situ incubation experiment (also, same timing for blooming of benthic Thaumarchaeota), while biphytanyl chains were then rebounded to -22.0‰ (vs. PDB) after 405 days. Please see the Figure 3 and Supplementary Table 2 in the reference, and reorganize the context.

P12814, L13:

Could you provide the name of 16 amino acids or state the serial number ?

P12814, L25:

$^{13}\text{DIC}$  should be  $^{13}\text{C-DIC}$ .

P12817, L17:

I could not find out Schouten et al. (2007) in the reference list. The author's confirmation is needed.

P12823, L19-P12824, L9:

With respect to in-situ  $^{13}\text{C}$ -culture experiment for tracing sedimentary archaeal GDGTs, Nomaki et al. also reported significant  $^{13}\text{C}$ -incorporation (+213.8‰ in cal-

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darchaeol and +61.4‰ in crenarchaeol in 9 days, relatives to initial in-situ incubation status of 0 day) by using <sup>13</sup>C-labeled *Chlorella* sp., as organic <sup>13</sup>C-substrate to investigate the in-situ benthic response by a sinking primary production. Please see, Nomaki et al., (2011) *Marine Ecol Prog Ser*, 431, 11-24, and also slightly re-organize the discussion into those lines on page 12823.

P12826, L26-P12827, L4:

The author needs to modify this misleading discussion, likewise line 4-8 on page 12811. Please note that they reported the biphytanyl chains were slightly <sup>13</sup>C-labelled up to -9.3 ‰ (vs. PDB) during 9 days in-situ incubation. Additionally, the author also needs to consider the consequence of <sup>13</sup>C-incorporation to archaeal GDGTs reported by Nomaki et al., *MEPS* (2011).

P12834, L9-11:

Please check the latest reference information or note the DOI.

P12844, Fig. 6, (a) Station 1:

The data plot at the depth of 0-1 cm by using bicarbonate is missing. Presumably, this section is most active sediment-water interface. If the author can reflect on this, please update on it.

That's all.

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Interactive comment on *Biogeosciences Discuss.*, 10, 12807, 2013.