Biogeosciences Discuss., 7, C902–C906, 2010
www.biogeosciences-discuss.net/7/C902/2010/

Biogeosciences Discussions

Interactive comment on "Microbial conversion of inorganic carbon to dimethyl sulfide in anoxic lake sediment (Plußsee, Germany)" *by* Y.-S. Lin et al.

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Received and published: 8 May 2010

Review of: Microbial conversion of inorganic carbon to dimethyl sulfide in anoxic lake sediment

Y.S. Lin et al

The study by Lin et al. provides experimental evidence and a proposed mechanism linked to methanogenesis for the biological production of dimethylsulfide (DMS) in anoxic lake sediment. The studies are well designed, logical, clearly described and, consequently, the results are compelling. Several key observations from the numerous incubation experiments support the general conclusion that DMS is produced primarily from bicarbonate and methanethiol (MT), in part from methyl addition to sulfides, but

not from methyl group donors (by O-demethylation). The experiments described in this manuscript did not allow the investigators to decipher the origins of the MT with the exception that the primary source is neither bicarbonate nor methoxylated compounds. Several intriguing possibilities (in particular methionine as a precursor) are suggested, but, to the authors' credit none are defended. The results from this study represent a major advance in linking carbon and sulfur cycles in higher temperature (but not necessarily thermophilic) systems. In fact, Fig. 1 is text book material for linking methylated sulfides to carbon pools in anoxic sediments. Nevertheless, given the results were all obtained from incubations conducted at 55oC in freshwater sediments, it is not clear to this reviewer if the authors suggestion that this DMS-forming pathway is applicable to sulfate-methane transition zones that generally occur at relatively low temperatures (e.g. <5oC) in marine sediment. The most likely setting for observing this process in the nature is within "hydrothermally influenced" systems.

Without reservation, I recommend this manuscript for publication. Below, I offer some comments and suggested edits that may improve this fine manuscript.

Specific Comments: This reviewer does not have direct experience performing the radiolabelling experiments described in the methods. It would be advisable for someone with such experience to comment. However, I have conducted experiments with radiolabelled sulfur, and, therefore, have a general understanding of how these experiments should be conducted. Based on the description, justifications, caveats and qualifications provided by the authors, I feel the experiments were designed and executed with remarkable sophistication and care. Fundamentally, all of the parameters used to calculate the rates using Eq. 1 were clearly described and qualified. Nothing was left to the imagination, which is good (and frankly infrequent).

Regarding the substrate addition experiment, the authors suggest there is no stoichiometric relationship between the production of DMS and MT. I mildly disagree with this point as the rate of the DMS production moving across the columns in Fig 2 increases continuously while that of MT production decreases continuously. To me, this inverse

relationship implies increasing rates of DMS production are linked to a diminishing accumulation of MT, as would be expected from Eq. 2. I agree there is not a 1:1 stoichiometric ratio for the relationship, which is certainly related to the uncertainty in the source of the MT. This relationship is brought forward in the first paragraph of the discussion, which indicates an important experimental result. However, the point of discussion is based on the inhibition study. . .I believe this experiment also supports that important conclusion. Consequently, I feel this point should be emphasized now to prepare the reader for that discussion point.

For the same section, I was also initially confounded by the suppression of methane production when H2 was added. Later, this trend was explained to be the result of enhanced production of fermentative products (e.g., propionate), which makes sense, but I had to wait a long time for that resolution. IF there is any data from these experiments that suggests this was the case here, it might be reported in closer proximity to the statement about H2 suppressing methanogenesis. What troubles me is that acetate did not accumulate, which makes me wonder why propionate would be expected to do so. If actetate concentrations were measured, propionate would also have been.

From the 13C-HCO3 labelling experiment, the authors conclude the results support the hypothesis that 13C-enrichement of DMS suggests direct incorporation of bicarbonate into DMS. I do not argue that conclusion as the d13C of the DMS was clearly affected by this process. However, from the reported results (maximum d13C of +119%o for DMS) I cannot get a feel for how important this carbon source was for the accumulated DMS. This is simply a matter of how much the bicarbonate was enriched, but this is not included in the results. If it is a pure 13C source, +119 per mil enrichment is not the primary carbon source. . .or is it? A mass balance calculation would be interested and is advised. The natural abundance mass balance discussion on page 2583, I believe, assumes 50% of the carbon in the DMS comes from bicarbonate (as required by Eq. 2). . .doing the mass balance with the labeled substrates should actually provide a better estimate of the bicarbonate contribution than the natural abundance calculation.

Question. In the substrate addition experiments, it was clearly demonstrated that the addition of O-methyl compounds did not stimulate DMS or MT production. Given that, on page 2582, Lines 4-6, why would one expect MT pool in the 35S experiment to be derived from O-methyl pools? Is the reader expected to ignore the results from the preceding experiments?

The discussion about the methanogens being stressed (not thermophilic) is interesting. But given that, how important would one expect this process to be in hydrothermally affected systems? Wouldn't one expect the thermophiles to competitively exclude the stressed methanogens in a hydrothermal system? I can see why this would not occur in the experiments conducted from lake sediments as thermophiles are likely absent. But in a hydrothermal environment this might not be the case. There are certainly other ways of looking at this (all speculative), but not addressed here. But in any case I would be careful about extrapolating the results from this study to hydrothermal field. . .at least use caution.

Best of luck identifying the source of the MT! (My bets on the amino acids).

As a final comment on this wonderful paper, to test the idea of transmethylation from lignin to sulfide, consider conducting incubations with marine and terrestrial organic matter sources (i.e., lignin bearing and non-lignin bearing sources.)

Technical and Editorial Suggestions:

In the abstract, stating the 35S labeling studies demonstrated a "slow" process is vague. By some measures, all of the rates from this study were "slow". Perhaps, this could be rectified by simply saying the rates were slow relative to the others, or simply eliminate that part of the sentence as it must be slow if it accounted for <10% of the DMS.

On pages 2571 and 2572, replace "Firstly, Secondly, Thirdly and Lastly" with "First, Second, Third and Last." That way it is now written sounds a bit odd.

Page 2575, First paragraph, Section 2.5: The statement “x was not added. . .in order not to decrease. . .the specific activity” is very difficult to digest. Unless there is a reason for using that text that has escaped me, why not simplify this by writing “x was not added. . .to maintain a low specific activity”. After all, to “not decrease” is “to maintain.”

Line 11, same paragraph: . . .was added with the redox indicator resazurin. . . (to let the reader know why resazurin was added.)

Line 13, same paragraph: Consider amending end of sentence, . . .became colorless, which indicated . . ..

Page 2577, line 27: To state that “A (add this article) small amount of liquid was added” is vague. I imagine that the “small amount” is roughly equivalent to the lowest amount reported in the study. To be clear, simply provide the amount that was injected into whatever volume.

Page 2580, line 9. What does “these compounds were in the range that generated microcosm response in previous studies” mean? Please clarify this point.

Page 2582, Line 1: delete “a”

Page 2587, Line 13. To say methane starts to build up at the SMTZ is incorrect. Sulfate and methane from spatially separated sources are consumed there. It is a region of consumption, not initiation of production. That occurs immediately BELOW the SMTZ.

Thank you for giving me the opportunity to review this manuscript. –john pohlman

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Interactive comment on Biogeosciences Discuss., 7, 2569, 2010.