

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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This manuscript describes an interesting study of dimethylsulfide (DMS) formation in anoxic lake sediments incubated at elevated temperatures (55 C). The authors have provided evidence that reduction of CO₂ to the stage of methyl coenzyme M by methanogens, and then subsequent methylation of methanethiol (MT) by methyl CoM leads to DMS formation. This might be a new mechanism for the formation of dimethylsulfide (DMS) in anoxic freshwater sediments, although a previous study (Stets et al) also implicated methanogens in methanethiol methylation leading to DMS formation in acidic freshwater peats. In any case, the present study provides strong evidence for such a mechanism in Lake Plußsee and I think this is an important contribution that deserves to be published.

The paper is generally well written, though it could use improvement in a few places (see comments below). The data are of high quality and clearly presented.

The abstract could use some work to make it clearer and more logical. It is understandable – after one has read the entire manuscript, but on its own, I don't think it has a logical flow. For example, on line 14 the sentence "Labeling with $\text{NaH}^{13}\text{CO}_3$ showed that incorporation of bicarbonate into DMS occurred through methylation of MT (methanethiol)" does not make it clear that the bicarbonate was fixed via a reduction pathway of methanogenesis (thereby explaining the light ^{13}C of the methyl of DMS) and it was methyl Co-M that was the methyl donor for methanethiol methylation. Likewise, the conclusions don't logically follow from preceding sentences. It would be better to say something like: BES inhibited DMS formation suggesting that methanogens were involved in production of DMS. Something like that.

It might be worth pointing out somewhere in the manuscript that DMS consuming methanogens apparently use the reverse of the final methylation to generate methyl Co-M from Co-M, releasing MT in the process.

What were the sulfide concentrations in the lake sediment slurries? In the presence of sulfide, the inhibitor BES is converted slowly to Co-M (the compound BES mimics) – see Kiene, 1991. I doubt that this would have affected any of their results, but at the elevated temperatures used in this study perhaps it might have been important. In any case, it is something the authors should be aware of.

Was the pH of the sediments measured? Flushing with N_2 could remove CO_2 and could change the pH and of course the addition of HCO_3^- could also affect (or buffer) the pH. How might this have affected the results?

P2571, L29. Add hyphen: DMSP-limited. . .

P2573, L10. Reword. Our study tested whether this reaction is . . .

P 2575, L2-4. This is somewhat unclear. Was the 10 mmol/L NaHCO_3 $^{13}\text{-C}$ labeled? I

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don't think so, but this could be worded more clearly. What do you mean by background level? L 5-7. Did the storage at -20C kill the samples?? And what about potential abiotic reactions for DMS and MT when samples were frozen and then heated at 60 C for 20 min? Can they be ruled out?

P2575, L 23. It seems a pity that the first time point after H₂S addition was made 2 h after the 35S-H₂S was added. I understand that they did this to allow equilibration, but it would have been interesting to observe the equilibration kinetics.

What is the rationale for assuming that only the dissolved phase compounds could be methylated to form DMS? Since most of the added 35S-H₂S partitioned into solid phases, if this assumption is wrong, then it could lead to a large error

P2579, L17. Indicate whether there was any time trend for DMS in the autoclaved control. L18. ...AUTOCLAVED control. ...

P2579, L24. ...bicarbonate WHEN ADDED SEPARATELY...

L26. Add comma after In combination, ...

P2580. L6. ...formation WAS... (use past tense in these situations)

L12. Change improved to increased. ...none of the OTHER four substrates significantly INCREASED...

L24. ...was 60% inhibited by X mM BES

P2581. Top. The lack of effect of BES on acetate formation argues that there was no competition for H₂ among methanogens and acetogens. Correct? But the Methanogens were CO₂ limited? Is this common for Lake Plußsee – that Methanogens were CO₂ limited?

L24. Indicated (past tense).

P2582. L 12-13. It's not clear to me what you mean by conclusion #2 – that H₂S vapor

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was incompletely fixed and trapped.

P2583. L 3. Make it clear that H₂ alone did not stimulate DMS formation while bicarbonate alone did, and both TOGETHER stimulated DMS even more. As written this sentence is a bit misleading.

P2585, L4. Are you implying here that Methanogens were growing (or at least conserving energy) with the process of DMS formation? If you don't know that to be the case, then perhaps you should point out that growth by this mechanism remains to be determined, even if it is thermodynamically feasible.

L23. Re-order words. However, from these experiments we cannot yet identify. . .

P2586, L19. From what type of system was the $\delta^{13}\text{C}$ of S-adenosylmethionine methyl groups determined?

I doubt that demethylation of isotopically light DMS could explain the light methanethiol if methanethiol is the main precursor of DMS. That would imply a tight cycle, yet substantial MT is lost to other reactions (abiotic and biotic) so there must be some other major source.

Citations

Kiene, R.P., 1991. Evidence for the biological turnover of thiols in anoxic marine sediments. *Biogeochem.*, 13: 117-135.

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