

Interactive comment on “Microbial methanogenesis in the sulfate-reducing zone in sediments from Eckernförde Bay, SW Baltic Sea” by Johanna Maltby et al.

Anonymous Referee #1

Received and published: 16 May 2017

Shallow littoral sediments are a poorly constrained source of methane to marine and brackish water columns. Normally, methane fluxes from marine sediments into the water column are restricted by the large fluxes of sulfate available to microbial sulfate reduction taking place in the sediments. This “microbial lid” on methane effluxes derives in part from the competitive advantage of organoclastic sulfate reducing bacteria versus methanogens for buried reactive organic carbon substrates, and also to the direct oxidation of upward diffusing methane by methanotrophic sulfate reducing prokaryotes. However, methane ebullition from deeper layers into the surface sediments, or the production of methane from non-competitive substrates (e.g. methyl amines, or methanol) may contribute significantly to the methane flux into bottom waters. It is the latter pro-

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cess that the authors of this study seek to address and quantify in Eckernförde Bay sediments. Their approach can be divided into two parts: 1) a seasonal study of sediment methane biogeochemistry, including rate measurements, and 2) an experimental enrichment to examine the effect of methanol as a potential non-competitive substrate in Bognis Eck sediments.

Maltby and co-authors present a detailed seasonal data set showing geochemical and experimental data collected over two years from the shallow, organic-rich sediments of Eckernförde Bay in the Baltic Sea. Although it has been known now for decades that minor amounts of methane forms in sulfate-reducing sediments from methanogenesis of non-competitive substrates, the role that this process plays in Eckernförde deep waters was not clear prior to this study. The data and outcome of the present study are consistent with previous studies of methanogenesis using non-competitive substrates and suggests that methane derived from non-competitive substrates may be a source of methane for the Eckernförde deep water. This study adds to the data and knowledge concerning sediment biogeochemical processes for Bognis Eck, which has been the site of a successful string of studies investigating the biogeochemistry of deep anoxic waters and the underlying sediments in Eckernförde Bay. The geochemical data is of high quality. The down core experimental tracer data is also of good quality, although I have reservations about interpretation of some of the experiments (see Major Issues below).

Nevertheless, there are a number of points in the manuscript that the authors need to address.

Major issues:

1. Section 3.4.1 (and Methods – lines 235-244) Net methanogenesis: These rates do not necessarily represent methanogenesis in the presence of sulfate. Were the sulfate concentrations monitored during the incubations? There are no time course data of sulfate (nor methane) shown for these experiments. As the incubations were

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performed over four weeks, the chances that sulfate became depleted within several days at many of the depths is very likely given SR rates of up to $10000 \text{ nmol cm}^{-3} \text{ day}^{-1}$. Therefore, the direct comparison of the ^{14}C labeled hydrogenotrophic rates with Net Methanogenesis rates are not at all valid. The Net MG rates are very likely a severe overestimation of actual in situ rates of methanogenesis.

Likewise, the Manipulated Methanogenesis experiments are not described in enough detail to evaluate them properly. Were these experiments performed like the Net Methanogenesis experiments? Or were they performed over shorter period of time using radiolabeled bicarbonate?

2. I am not sure how insightful the ^{13}C -labeled methanol enrichments are for understanding the role of non-competitive substrates at this site. First of all, no in situ methanol concentrations are provided. Secondly, and more importantly, the authors added methanol up to 10 mM. These are enrichment concentrations that are not likely to reflect environmental conditions. Enrichment, or growth on methanol, is what they see in the experiments, as shown in Figures 6 and 7. The conclusion that these enriched organisms represent the in situ organisms and metabolisms is not tenable. This experiment does not even shed light on whether or not there was non-competitive methanogenesis occurring in the experiments slurries themselves. What happened to sulfate during this experiment? Was there still sulfate present after 10 days?

3. The Discussion needs to be made more concise. The authors should directly address the stated main point of the manuscript: Is there methanogenesis in the sulfate reducing zone, does it proceed via non-competitive substrates, and is it at all important for methane fluxes to the deep water? The discussion as written now is, to a large extent, a reiteration of the results with some commentary. It also tends to drift off into unwarranted speculation. Some parts that could be excised without detriment:

a. Lines 564 and following : “possible” additional sources of carbon and the production of hydrogen

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b. Lines 626 “Reaction of sulfide with methyl groups and organic matter. . .discussion is beside the point.

c. Lines 646 Discussion of dissolution of CO₂ in water was already discussed earlier in Results.

d. Section 670 The discussion on temperature is speculative and I am not sure where it is leading.

e. Lines 783 and following: The discussion of deep methanogenesis (below the SMTZ) appears to be beyond the scope of the manuscript (i.e. methanogenesis in surface sediments)

One means of shortening the discussion might be to delete or severely scale-back to the discussion revolving around the PCA analysis. I do not see how the analysis and resulting discussion adds anything new to our understanding of the controls on methanogenesis in marine sediments. In considering such a discussion, it might be worth for the authors to revisit the seminal articles on this topic by Crill and Martens (L&O 1983 and GCA 1986).

Specific Comments:

Line 282. This sentence is confusing. “Fast oxygen consumption” does not correlate with “slowed microbial activity”.

Figures 1 and 2. The postage stamp size plots (at least in the BG Discussions version) are difficult to read. Perhaps taking the water column data out and combining it into a separate figure would help?

Lines 424-434. I would not put so much emphasis on the single bottom points of the gravity core.

Line 469: The hydrogenotrophic methanogenic activity at 45 cm depth at the sulfate-methane transition zone may be in part due to tracer back flux associated with AOM

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(see Holler et al., PNAS 2011).

Figure 7: What is the difference between the methane concentration in this figure and in Figure 6? Why not combine Figures 6 & 7?

Lines 525 and following: What are the criteria for calling something a “strong” or “weak” correlation. Line 554 It might be good to briefly describe how BES works as an inhibitor, and why it has no effect here.

Line 566 How deep is bioturbation in Bognis Eck? And was the shell at 20 cm living or just debris? Figure 8: Based on what criteria was 0-5 cm depth for integrated methanogenesis chosen, whereas, similar data, but from 0-25 cm is shown in Figure 4?

Line 614: Again, this looks like a growth curve.

Line 637: These organisms became dominant due to the highly enriched methanol concentrations employed. This does not say anything about their importance under in situ conditions.

Line 690 and following: Changing sulfate concentration-depth profiles as a response to changing salinity conditions indicates that this is a non-steady-state situation. Ergo, it is not possible to use this as an indication of microbial sulfate reduction.

Line 841: How does the fueling of AOM above the SMTZ cause methanogenesis to play an “underestimated” role? I would expect that AOM would minimize the impact of methanogenesis on the water column methane budget.

Technical comments:

Line 138 “that” instead of “which”

Line 437 “Content” not “concentration” for POC wt%

Line 612: Sentence is confusing: “of” rather than “if”? Also, the population changes

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to the new conditions; you do not have any evidence for adaption (and evolutionary concept).

Interactive comment on Biogeosciences Discuss., doi:10.5194/bg-2017-36, 2017.

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