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Interactive comment on “Miniaturized biosignature analysis reveals implications for the formation of cold seep carbonates at Hydrate Ridge (off Oregon, USA)” by T. Leefmann et al.

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General Comments:

The work submitted by Leefmann and co-authors is introducing a novel approach to extract lipid biomarkers of micro-drilled carbonate phases from a cold seep carbonate by using a miniaturized extraction protocol. The method enables to measure lipid biomarkers in mg-sized samples. This is the first time that a miniaturization approach was applied to sub-recent seep carbonates to further elucidate the biomarker inventory of specific carbonate phases. In their study they are able to show that specific carbonate precipitates (here: whitish aragonite) were formed by biofilms of a former

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active Anaerobic Oxidation of Methane (AOM) consortium, whereas the formation of other carbonate phases seems not to be or only poorly related to AOM. Overall the manuscript is well written and consistent, and should be published after minor revision.

Specific Comments:

The inventory of lipid biomarkers as revealed by the miniaturized approach are consistent with findings from earlier studies from Hydrate Ridge (e. g. Elvert et al., 2005). The signatures found in the whitish aragonite clearly report AOM as responsible process forming this carbonate phase. For the other phases, especially the gray micrite, AOM cannot be excluded or was at least partially responsible for the formation of this phase. It would be very interesting to see differences in stable carbon isotopes of specific compounds, but I can imagine that it is not possible to measure miniaturized samples on the GC-IRMS. Therefore I suggest to include the bulk stable carbon isotope values of the drilled samples in Table 1. These results will at least provide additional data whether the carbonates are showing AOM signatures or not.

The second comment is dealing with the presence of lipid biomarkers from sulfate-reducing bacteria (SRB) in the carbonates. Since the SRB thriving at seeps in the consortia have never been found to synthesize DAGEs it is still not as clear-cut if DAGEs are sourced by SRBs living in the AOM consortium or if they were sourced from other, unknown SRBs, or other unknown bacteria. I am aware that most authors use DAGEs as SRB markers, but I would prefer to see distribution patterns of typical SRB fatty acids as well. It appears to be from the results presented here, that no specific SRB fatty acids (for example terminally branched fatty acids) were preserved. Maybe this is because the samples were not saponified prior extraction? Fatty acids which have been part of the AOM consortium may still be ester-bond or stored within the carbonate lattice and cannot be released by extraction only. The fatty acids presented in this study are most likely sourced only by autochthonous organisms. Therefore I reckon that it may be beneficial to saponify at least samples 7r1 and 9 (those with the highest concentrations) to test whether SRB fatty acids can be released or not after saponifi-

cation. Since SRB-specific fatty acids could have been traced back to the Oligocene (see Peckmann and Thiel, 2004; Lincoln Creek site), I would expect at least terminally-branched fatty acids to have the potential to be excellently preserved in the studied Hydrate Ridge samples.

The last comment is dealing with a) the presence and stability of hydroxyarchaeol and b) the using of the hydroxyarchaeol/archaeol ratio to discriminate ANME-1 from ANME-2:

a) Starting from page 4449, line 27 the authors pointed out that hydroxyarchaeol is rarely present in the fossil record and explained this fact by preferential degradation of this compound. It is true that Peckmann and Thiel (2004) only were able to report hydroxyarchaeol in trace amounts from the Oligocene Lincoln Creek, but not in the other Neogene and Mesozoic sites. However, in a recently published article in Organic Geochemistry hydroxyarchaeol was detected in three different ancient locations with strong ^{13}C -depletions (see Birgel et al., in press). Although no hydroxyarchaeol/archaeol ratios have been shown in their article, at least hydroxyarchaeol was still present in Neogene samples. I suggest that the statement should be modified.

b) Though the concentrations of hydroxyarchaeol are as high or even higher than those of archaeol and the ratio in the whitish aragonite are in 5 out of 8 samples >1 and might indicate that ANME-2 were the major consortium precipitating whitish aragonite, ANME-1 cannot be excluded as further candidates participating in carbonate formation, since no data of GDGTs are available. In Blumenberg et al. (2004) mixed ANME-1 and ANME-2 consortia provided hydroxyarchaeol/archaeol ratios varying from 0.6-1.4. Three out of the eight analysed whitish aragonite samples showed similar ratios. The authors discussed that preferential degradation lead to these ratios, but still the presence of ANME-1 cannot be excluded entirely. I am aware that it is unlikely or even impossible to measure GDGTs in miniaturized samples, but I would suggest to weaken the argument that the whitish aragonite is almost only precipitated by ANME-2.

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Technical corrections:

p. 4444, line 21: ...are exiting the seafloor.

p. 4444, line 24: ...a consortium of...

I hope that my comments are helpful for the authors

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