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Interactive comment

## Interactive comment on "Biogeochemical and microbiological evidence for methane-related archaeal communities at active submarine mud volcanoes on the Canadian Beaufort Sea slope" by Dong-Hun Lee et al.

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This paper reports the occurrence of AOM in the active mud volcanos on the continental slope of the Canadian Beaufort Sea, based on the analysis of lipid biomarker and 16S rRNA gene in three sediment cores. The authors state that archaea of the ANME-2c and ANME-3 clades participated in the process of methane consumption, by the relative high ratio of sn-2-hydroxyarchaeol to archaeol and the phylogenetic identities. They then speculate that the difference in distribution of ANME-2c and ANME-3 in these mud volcanos is under the control of methane flux changes. The results of this

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study can further help us to understand the biogeochemical cycling of methane in the seafloor mud volcano environments.

Generally, it is a very interesting study, especially the detection of extreme depleted  $\delta$ 13C value of sn-2- and sn-3-hydroxyarchaeol. The approach is scientifically valid and the conclusions are reasonably sound. However the manuscript has some questions and inadequate discussion that require resolution.

- 1. The content of the abstract section needs some more substance. For example, the mechanism of ANME distribution, as discussed in section 4.3, should be presented in this section.
- 2.Some expression is inaccurate, such as "bulk elements" at line 101 and "Bulk geochemical analysis" at line 105. The "Bulk geochemical analysis" only contains the bulk total organic carbon and its carbon stable isotope composition in this manuscript. However, "bulk elements" indicates that the data also contain other chemical parameters such as the major and trace elements of the sediments.
- 3.In the section 4.1 (Line 292-306), the authors suggest that AOM was mainly coupled with sulfate reduction in this area. However, sulfate became depleted at depth of 0.20 (MV270), 0.20 (MV420) and 0.45m bsf (MV740) indicated by data of Paull et al., 2015. The reason why biomarker of AOM was detected in the sulfate depleted zone in this study need to be clarified.
- 4.In section 4.3, the authors speculate that the distribution of ANME-2c and ANME-3 is under control of methane flux. Specifically, ANME-2c has a preferential niche in habitats with lower methane fluxes, while ANME-3 is apt to present in setting with relative higher thermal gradients (corresponding to higher methane fluxes). However, ANME-2c and ANME-3 both present in the upper zone (0-0.2m bsf) of sediment core MV282 with similar abundance. Please provide an explanation for this phenomenon.
- 5. Line 384 to 388, the authors suggest that archaeol are originated from methanogen-

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esis rather than AOM, due to their relative enriched  $\delta 13C$  values (-79.8% to -38.5% Table S1) than the ascending methane in the MVs (about -64%. This explanation seems a little superficial. If archaeol was relate to methanogenesis in this area, the  $\delta 13C$  value of archaeol should be much heavier than those of methane and dissolved inorganic carbon in the methanogenesis zone. However, the difference in  $\delta 13C$  value between archaeol and methane, especially in MV282, seems not large enough to draw this conclusion. Moreover, if this conclusion is correct, the phenomenon that the occurrence of methanogens in the AOM zone should be explained in the manuscript, and references should be added to support this conclusion.

6.Line 390-392, "previous studies...showed that GDGTs were mostly absent in ANME-2-dominanted settings". Please clarify why ANME-2 and GDGTs were both detected in your data.

7. There are some spelling mistakes throughout the manuscript (e.g., Line 284 "Several of the lines of evidence" should be "Several lines of evidence". Line 416 "OUT c1698" should be "OTU c1698").

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