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Interactive comment on "Different methanotrophic potentials in stratified polar fjord waters (Storfjorden, Spitsbergen) identified by using a combination of methane oxidation techniques" *by* S. Mau et al.

S. Mau et al.

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Referee: This manuscript describes a field study of methane oxidation rates in a stratified fjord system in a polar environment. The authors used two different methane tracers, labeled with either 3H or 14C, with the 3H tracer being almost non-perturbing of the natural methane concentration and the 14C tracer causing >10-fold increases in methane concentrations. The 3H tracer therefore provided rates that would be close to the in situ rates, while the 14C tracer would give the potential rates at near-saturating substrate concentrations. In addition to depth profiles of the rates, the authors also

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present some del 13C values for the dissolved methane pools as a function of depth in the fjord. All of the data is interpreted in the context of three different water masses, representing different depth layers. It is a well written manuscript and presents some interesting and useful data. I have just a few comments and suggestions offered to help improve the manuscript.

The distinction between the 3H-CH4 and 14CH4 methods is not always clear throughout the paper. It was not just a different isotope that was used, but a very different concentration of CH4 tracer that was applied in the two cases. While they describe this in the method, the distinction is lost in the Results and I think they should try to make this clearer throughout the text.

Author: We will clarify this in the revised version of the manuscript.

Referee: Note –not to the authors but to the journal. It is extremely inconvenient to review the "print version" of the manuscript, with page sections numbered 6463 etc, with seemingly random breaks. A continuous line numbering would help.

Referee: Abstract. L14. Add comma after surface L18.at 60 m, AND PEAK RATES WERE found in ArW/BSW L19. I believe it should be 13C not 14C that were increasing in residual methane pool. L25 attesting TO the ubiquitous L 27. I think you need something after "unusually long" – what does it mean? Also, spelling error in Methylosphera Introduction. L10. PREdominantly Also, replace carbonate with CO2 as that is the actual substrate. Sec 6464. L5. Remove "of" L7 has proven TO BE

Author: All these suggested corrections will be done.

Referee: L8. I think it is incorrect to state that the two tracers are converted at the same rate as the natural pool of methane since the 14C increased the concentration 10-fold and actually reduced the rate constant for most incubations.

Author: In the incubations, radio-labeled and non-labeled CH4 are converted at almost the same rate (not accounting for kinetic isotope effects) (see Ward et al. 1987, Na-

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ture, V: 327, p: 226-229). However, the reviewer is right that methane turnover in the incubation vials, particularly in those spiked with 14C-CH4 tracer, is different than the in situ methane oxidation. In the discussion part, we invoke that this is related to the different concentrations of CH4 in the incubations and in situ.

Referee: L11. Exist not exists. Sec6465. L2. Water not waters

Author: All these suggested corrections will be done.

Referee: Sec 6466. L22. Remove "gas mixture comprised" It is not needed and it wasn't a mixture, just one isotope in each sample, right? Or did you add both isotopes to the same sample?

Author: It is correct, that either 3H-CH4 or 14C-CH4 were added to a sample (not both at once). However, 3H-CH4, tracer was diluted with N2. We will clarify this in the revised version of the manuscript.

Referee: Sec 6467. L4. It is not clear what you mean by "ambient". It could mean in situ concentration, or the ambient concentration in the particular sample (which in the case of the 14C would me much higher than in situ).

Author: Ambient stands here for in situ CH4 concentrations. We will exchange ambient with in situ in the sentenced pointed out by the reviewer.

Referee:L14. WERE carried out.

Author: will be changed

Referee: L20. What percentage of the remaining methane was in the headspace?

Author: Almost all of the dissolved methane accumulates in the headspace as solubility of CH4 in NaOH solutions is very low.

Referee: L21. Was trapped L27. Phenylethylamine Sec 6469 L13. Add "at 60 m" after respecticvely. L16.decreased WITH DEPTH Sec 6470. L6.showed DISTINCT DGGE

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L15. Add comma after waters Sec 6471. L5 attesting TO

Author: All these suggested corrections will be done.

Referee: L8.What does it mean that none of the amplicons matched known pmoA genes? Are they sure they did this right? This section seems weak.

Author: A specific gene sequence was amplified using A189f/A682r primers. These primers are designed as the starting point for replication of a pmoA-sequence. The A189f/A682r is the first pmoA primer set targeting the pmoA gene (Holmes, 1995) that is still extensively used in environmental studies (McDonald et al., 2008). The primer pair was designed predominantly for sequences of terrestrial organisms and may, therefore, not be optimal for marine samples, which could contain MOx communities with a modified pmoA. Furthermore, in some studies, this primer set resulted in a limited retrieval of the diversity of methanotrophs and delivered nonspecific PCR products (Bourne, D.G., McDonald, I.R. and Murell, J.C, (2001), Comparison of pmoA PCR primer sets as tools for investigating methanotroph diversity in three Danisch soils, AEM 67: 3802-3809). We designed clone libraries of PCR-products with the A189f/A682r primer set and compared the gene sequences with known and published gene sequences. Unfortunately, we could not always find a match to known pmoA sequences. This may either indicate novel pmoA types or unspecific PCR-products (McDonald et al., 2008). We will clarify this in the revised version of the manuscript.

Referee: L22. Use salt instead of ion L23.Delete “with” Sec 6472. L3. Replace “with” with “at”

Author: All these suggested corrections will be done.

Referee: L10. There are probably other possibilities besides DMSP for the source of the methane in the water column.

Author: Other links have not been discovered in this area. Therefore, we wrote that it is a ‘probable’ source. However, in other ocean environments, methylphosphonate

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was found as a potential methanogenic substrate (Karl et al., 2008; Nat. Geosci., 1, 473-478). However, it was invoked that the phosphonate functions as a P-source in phosphate-limited environments, which is not necessarily the case in our study site. We will add this information in the revised version of the manuscript.

Referee: Section 6474. Just a discussion point. The deep population of methanotrophs might be poised to respond to methane release events, which might have been missed in the snap-shot sampling

Author: In Damm et al., 2007, it is suggested that these methane release events occur generally during the winter as a result of polynyas forming brines, which descend and induce turbulence at the sediment - water interface (see 2.1). Release events during spring and summer have not been reported. However, we have discussed that the deep-water MOx community is probably adapted to high CH4 concentrations as they occur during winter time.

Referee: Sec 6475. L1attesting TO THE ubiquitous L17. Add “IN CONTRAST,” the comparably short L25. Give the range of enrichment factors rather than just 1order of magnitude. Check all references for typos. Bender and Conrad has a typo in the title.

Author: All these suggested corrections will be done.

Referee: I printed the figures in color and in Fig 2 it was difficult to see the stations and the text labeling the coastal current arrow. It was easier to see on the computer screen.

Author: We will change the color of station dots and the text ‘coastal current’ to white to increase contrast and readability.

Interactive comment on Biogeosciences Discuss., 10, 6461, 2013.