We thank the reviewer for their helpful comments and constructive suggestions which have resulted in a much-improved manuscript overall. Below you can find our full response to each point raised.

Response to Anonymous Referee #3

Wen et al. address microbial controls of high methane emission after re-wetting in two temperate peatlands with contrasting geochemistry. There is very little information available on microbiology of re-wetted peatlands, so as the first study of re-wetted non-acidic fens, this study is very welcome. The manuscript is clearly written and easy to follow. The molecular analyses for microbes have been carried out with care (testing for sample inhibition in qPCR, pooling three different PCR products to reduce amplification bias, checking the taxonomic affiliations of OTUs in ARB). This is not a study of rewetting effects, because no samples from before re-wetting or from a non- rewetted control site are available. However, in addition to providing much needed information on re-wetted peatlands, the results contain some interesting details such as the strikingly patchy distribution of ANME-2d.

My biggest concern is that the main result is based on comparison of two different qPCR assays (mcrA vs. pmoA). Such a direct comparison of values assumes nearly absolute quantification, which is not realistic for environmental samples (different limitations in coverage for each primer pair etc). Comparisons of values of one assay between samples, on the other hand, do not rely on this assumption in the same way. The previous examples of pristine wetlands used as support (l. 413-421, 450-452) similarly rely on comparisons of two different qPCR assays. If/when these studies have used different methods and primers as this study, the comparisons become even more problematic, even when made at the broad level of orders of magnitude. I do not disagree with the overall conclusion that high numbers of methanogens the most likely reason for the high methane fluxes, **b**ut I would strongly recommend addressing this limitation in the discussion and modifying the text on l. 404-421 and elsewhere, including the title of the manuscript. Maybe strengthening the interpretation of microbial community results in relation to geochemistry could provide an alternative main message.

Please find our reply to these concerns below.

In addition, I am wondering about the role of methanotrophs in completely inundated peat and in the water layer. It is very much expected that methanotrophic activity would be low considering that in both sites the sampled peat was inundated. The optimal peat layer for methanotrophs where both methane and oxygen are readily available is largely missing (which the authors do address in the end of the manuscript). However, such conditions could be present in the water layer. I realise the water layer is out of the scope of this study, but are there reasons to exclude it from discussion or assume it plays no role in methane oxidation?

It is indeed possible that oxidation may be occurring in the water column. It is true, however, that the water column was beyond the scope of this manuscript. Recent, preliminary data for Zarnekow show methanotrophs in high abundance associated with ceratophyllum in the water column (unpublished data, still in progress). Nevertheless, even if oxidation is occurring in the water column in these two sites it is clearly not significant enough to keep methane concentrations and emissions low as demonstrated by

the flux data (added) in the revised manuscript. This is now mentioned in the discussion in lines 464-466.

Minor comments:
1. l. 190-193 Did the primers contain sequencing adapters and barcodes or were they added later?

Yes, the primers contained barcodes. A phrase was added to the manuscript in line 212 to denote that the primers contained barcodes in the 5'-end.

2. l. 234-235 Please remove the word 'all' from 'suitable for detecting all aerobic methanotrophic Proteobacteria' or change to 'all known' or similar (we cannot assume to be able to detect the full diversity).

As suggested we have changed the text in line 259 to instead say "all *known* aerobic methanotrophic Proteobacteria".

3. l. 318-319, l. 360-361 The Hütelmoor samples show higher within-site variation, but the samples were also taken much further apart from each other. Could this not explain the larger variation? On l. 360-361, the sentence could be understood to suggest the difference is due to brackish vs. freshwater.

Though in the study the Zarnekow samples were taken closer together, we know from previous work at the site that there is indeed less variation across the Zarnekow peatland (e.g. Zak and Gelbrecht 2007). Thus, taking the cores further apart in Zarnekow in this study would not have resulted in greater variation in our measured variable.

4. l. 360 Please change 'significant' to another word because no statistical testing was carried out for differences of community composition.

The phrase 'significant variation' was changed to 'large variation' in line 386 in the revised version of the manuscript.

5. l. 415 I do not think it is possible to compare PCR-based relative abundances between different studies, unless the studies used completely identical methods and equipment. Was this the case with Liebner et al. 2015?

It is an inevitable limitation that the methods and equipment of different studies are not completely identical. This is not only a limitation of our work but a general issue for meta-studies. All comparisons regarding bacterial, methanotrophic and methanogenic abundance are based on universal primer combinations of the respective groups. The primers we used for the bacterial 16S rRNA and mcrA genes of this study are identical with the primers used in Liebner et al. 2015. With regards to pmoA, both studies used universal primer combinations including identical forward primers, but as a result of initial testing different reverse primers. Further, the same qPCR technology was used. In addition, we compared the ratio of methanogenic to methanotrophic abundances and the fraction of methanotrophs in relation to the total bacterial community based on two independent methods, namely qPCR and sequencing, instead of the direct methanogenic or

methanotrophic abundances. This kind of 'normalization' mitigates the bias of different experiments and makes the results more reasonable and reliable. As suggested, we now discuss this potential limitation in our revised manuscript in lines 448-453.
Further, we have revised the title of our manuscript. Our revised manuscript title is as follows, "Predominance of methanogens over methanotrophs in rewetted fens - a possible explanation for the observed high methane emissions?".