

Interactive comment on “Lake mixing regime selects methane-oxidation kinetics of the methanotroph assemblage” by Magdalena J. Mayr et al.

Anonymous Referee #3

Received and published: 9 March 2020

Dear Authors,

This manuscript describes the study of methane oxidation (MOX) during lake overturn in Lake Rotsee in Switzerland. You combine measurements of MOX kinetics with meta-transcriptomic analyses of methane monooxygenase genes and report differences between epi- and hypolimnion during stratification and a convergence of MOX kinetics and gene expression during lake water mixing. You conclude that methane oxidizers with well-adapted kinetics occupy distinct niches in stratified lakes.

While I think the report of kinetic parameter of methane oxidation is of great relevance, however, I found that the manuscript suffers from a lack of clarity and over-

C1

simplifications. Most importantly, it's unclear how the central conclusion, that well-adapted methanotrophs inhabit niches depending on methane availability (in hypo- and epilimnion), is reached. Wouldn't a match between in situ CH₄ concentration and K_m (not normalized per cell) be a stronger indication of such an adaptation?

I also believe a better use of the metatranscriptomic data could help to strengthen this point. A finer taxonomic resolution based on the pmoCAB genes and a more quantitative characterization of the community turnover should be possible – and could help to make the point that indeed there are distinct populations of MOB that are adapted to in situ CH₄ concentration. Accordingly, I think that Figure 3 a1-c1 is not the ideal way to convey this important point. Maybe a combination of SI Fig. 2 (which I think shows quite nicely the convergence towards similar gene expression patterns in January, with a Figure showing the taxonomic composition of MOB during lake overturn would be a better choice.

Moreover, I was somewhat irritated by the rather vague description of the environmental conditions during lake overturn. The traditional definition of lake stratification and hence the difference between epi- and hypolimnion based on temperature rather than oxygen. And while the manuscript addresses MOX during lake overturn, you refer to the oxycline for sampling. I understand that the temperature profiles shown in SI Fig 1 may not be as clear as the oxygen profiles shown in Fig. 1 – but I would advise to show all profiles (also conductivity which should explain the inverse stratification pattern in December) and to be very clear with the definition of overturn, thermos- and oxycline.

Finally, given the relatively low number of samples and the fact that the pattern was (only) observed in Lake Rotsee, I think the manuscript should be thoroughly rewritten to make clear that this may reflect a specific situation in the (relatively eutrophic) Lake Rotsee. Also, there are several cases of speculation or exaggerated extrapolation, which should be avoided.

Please also consider specific comments below:

C2

L 11 In freshwater lakes... so, this excludes saline lakes? Consider removing “fresh-water”

L 14 we tested the hypothesis that methanotroph assemblages in a seasonally stratified lake...

L 18 consider a brief explanation of the meaning “half-saturation constant” here

L19 ...Km differed by two orders of magnitude – but in the results it seems that they differed between 15 and 0.7 μM (a factor of ~ 20)

L 25 ...90% of what?

L28 can you talk about a climate IMPACT of lacustrine systems?

L 31 anoxic habitats... In the oxygen-depleted hypolimnion... repetitive

L 47 kinetic traits ... Use kinetic parameter instead (see L 48)

L 58 ... Lake Rotsee...

L 63 ex situ consider replacing with “laboratory incubations”

L 73 four or five campaigns?

L 77 and onward. Please provide more detail on this method including how the killed controls were treated.

L 91 how were Schott bottles sealed air-tight?

L 110 we determined the in-situ MOX rate ... in duplicate ex-situ incubations... Confusing, please rewrite.

L 161 an 167 reads shorter than 400 or 300 bp were removed?

L 183 aerobic methane oxidation likely contributed to this oxygen depletion in the epilimnion. This seems very speculative for me. Could a back of the envelope calculation, e.g. knowing the volume and CH_4 concentration in the hypolimnion and the stoichiom-

C3

etry of MOX be used to support this speculation?

L 228 critical phase – critical for what?

L 233 specific affinity towards methane... unclear what is meant here.

L 235 was the convergence only driven by changes in kinetic parameter in the epilimnion (or also in the hypolimnion as seems apparent from Fig. 2 a)

L 289 remove “as in many other stratified lakes” – too speculative (or include references, but I would not advise so in the conclusion part)

L 295 adaptation to oligotrophic conditions – Lake Rotsee can not be considered oligotrophic

L 298 transport of methane into the epilimnion provided and advantage for fast-growing MOB over slower competitors. This is not shown (at least in this manuscript) and should be removed.

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