Responses to comments on Manuscript Preprint egusphere-2023-1745 by Clayer et al.

Title: Technical Note: Preventing CO2 overestimation from mercuric or copper (II) chloride preservation of dissolved greenhouse gases in freshwater samples

Reviewers' comments are underlined for clarity. Line numbers refer to line in the revised (clean version) version of the manuscript, if not specified otherwise.

REVIEWER REPORT(S):

Reviewer 1:

We are grateful to the reviewer, Thank you so much for this constructive and detailed review. Below we provide some preliminary response to the each reviewer's comments.

Specific comments:

- I am happy to see that these authors spent the time to do the analyses and writing of a technical note paper, as I believe it can prove very valuable for a wide audience and can improve the methods and results of many future studies. The use of certain chemicals to preserve samples for gas analysis is very common, and it is therefore really nice that there is research on the effects of the conservation by several methods. I therefore want to thank the authors of this manuscript for working on this topic, which I think is highly suitable for the Biogeosciences journal.

Thank you, it is nice to see that our work was appreciated.

- However, I am currently unable to assess the results of this paper, because I lack key information on the reliability of the results. I think this paper has a good potential, but the aspects that are missing are key for the interpretation of all results, so they need to be clarified before I could properly assess the quality of the whole paper.

We are sorry to read this and apologize for taking your time. We have now reorganized the method section and have split the results and discussion section to streamline and clarify the manuscript. See L. 116-215 for method description.

- I find it difficult that I cannot see if the actions of adding the substances to the samples itself changed something in the gas concentrations. There are differences in the gas concentrations at t0. This can be attributed to rapid effects of the interaction between the added chemicals and the sample, but it can also be due to sample contamination or degassing during handling. T0 is taken within 24 hours, but it remains unclear if this is after 1 hour for certain samples, and after 23 hours for others. There is also no miliQ or demineralized water control. To have these issues addressed, in a reply to this comment but also in the manuscript, would give me more certainty about the results.

Unfortunately, the experimental design was imperfect, and would have been nice to have some benchmark steps such as pH measurements of the samples right after preservative addition. Now to

overcome these shortcomings, we perform an additional 24h incubation to document the impact of preservatives on pH. See L. 174-187 and results section L. 437-442 and new Fig.2.

Note also that we start the discussion by discussing the validity of using the unfixed samples as representative for "real concentrations". See L. 494-519.

- I would think it benefits the paper if results and discussion are separated into different sections, but I leave it up to the editor to decide on this, as I know there is also personal preference involved on that topic.

This is exactly what we have done to streamline the paper.

- Another key issues that I would like to hear more on is why the t0 concentrations of CO2 of the different treatments is that different. You write some about it, but as I see this as a major factor for many of your interpretations, I would like to have it addressed more in depth and more clearly. Why are there no error bars on the Cu and Hg boxes of CO2?

We now start the discussion by discussing the validity of using the unfixed samples as representative for "real concentrations" and describing and explaining the difference seen at T = 24h. See L. 494-519.

There are no error bars on the Cu and Hg boxes because the points plot all together and that the 25th and 75th are also covered the 100% points, i.e., the 6 replicates. We have now added a description of the box plot in Fig. 1.

- Another important overarching issue is that it seems O2 significantly decreased in the samples with CuCl2. Is this indeed the case? This is of major importance, as it would suggest incomplete inhibition of microbial processes, and would affect all results for the CuCl2 treated samples.

Yes, there is a significant decrease in O2 over time which is likely due to incomplete inhibition of microbial processes. We didn't focus because the acidification of the samples is already a sufficient reason for disapproving its use as preservative. We make a clear point that CuCl2 and HgCl2 should be avoided.

- Specific comments on certain parts of the manuscript are included below.

Introduction

- The first paragraph is a bit odd. I think it is most important that the reader knows why it is needed to preserve the samples, not what they are used for in the end. I would put more emphasis on the processes in the bottles that will change the concentration.

Agreed, we have rephrased it. See L. 41-51

- I think it is important to also mention that ZnCl2 and CuCl2 are often used as toxic inhibitors. And give some details on why it seems certain researchers pick which inhibitor (or put that in the discussion). Problems with the disposal of HgCl2 would be good to mention as well, as well as the costs associated with it.

Agreed. However, we didn't put more focus on HgCl2 and gave more space to AgNO3 and CuCl2 in the introduction to re-equilibrate. See L. 75-79 as well as L. 88-89.

- 48 + 49-50. Based on what did you decide to pick these papers as references?

We performed a wide literature search for studies specifically mentioning HgCl2 as preservative for dissolved gas samples in their methods.

- 75 – 81. The tone of this paragraph is also a bit odd. You state quite suddenly that CO2 concentrations will be overestimated by HgCl2, while that is not really clear from the previous paragraphs. And the last line is way too firm for an introduction. The introduction should state the current state of the art, not opinions.

This paragraph has been removed.

- 82 – 92. I would structure this paragraph differently. First, in neutral words (so no effective, overestimation, etc) explain what you investigated. Then state in a few sentence the key findings of your research.

Agreed, we have rephrased the paragraph and hopefully clarified it. See L. 97-111.

Methods

- 96. Header 'study area' does not cover the content of the paragraph.

Agreed, we renamed the header to represent the content of the paragraph, see L. 115

- 98. What is carefully collected? Explain in more detail.

The sample procedure is now described in much more detail. See L. 116-131

- 100. Avoid = limit. And not gas loss, but gas exchange with the atmosphere.

Agreed. Corrected.

- 107. That cannot be uniform for all Norwegian lakes, right? Do you mean that is the case for this specific lake, or do you mean this lake was like the lakes tested in that de Wit paper?

Yes, this is now corrected, see L. 130-131

- 112. T=0 could be at any moment during the first 24 hours? Or was it the same for all samples? Were they randomized for the moment of analysis, or were some treatments done first and others later?

This is now clearly described. First time point is T = 24h. See L. 142-147

- 112. Were the same bottles measured at t0, t1 and t2, or were the bottles sacrificed?

This is now clearly described. See L. 139-148

- 113. 'as the water samples were collected'. Which water samples? Is it not the same as the 5L bottles? Unclear.

This sentence has been completely rephrased

- 123. Unclear what 'it' means.

"it" refers to "estimated toxicity". This sentence has been completely rephrased.

- 129. Why were they stored cold? In general, poisoned sampled are not kept cool, as far as I know.

This is how we have worked so far to be on the safe side (see e.g., Clayer et al. 2021), and this also prevents the samples to be subjected to changes in room temperature. Keeping water samples cold and dark is the best way to optimize sample preservation. There is no guarantee that poisoned samples will be 100% inert, keep them cold helps to limit any microbial activity.

- 133. Remove 'unfortunately'.

Done.

- 127. Was the same amount of liquid added to all samples? Was the miliQ flushed to remove the target gasses? What was the volume of the sample bottles?

Yes, the same volume of 240µL was added to all bottles. And no miliQ water was not flushed, but given its small volume, its concentration is unsignificant for the water samples. This is now clarified.

- 141. Sealed with what?

The samples were sealed with gas tight butyl rubber stoppers as for the first experiment. We used the sample bottles and caps for both experiments. This is now clarified L. 153-154.

Results and discussion

- Fig. 1. Please split the graphs of O2 and CO2 and CH4 and N2O, it is now not clear that boxes 5-8 are a different gas. 100% saturation at which temperature? That of the lake water, at the 4 degrees of the storage, or the temperature during measurement?

Everything is reported back to *in situ* temperature. This is now clarified in the caption. And the graph has been split. See new Fig. 1

- Table 3. This table is a bit hard to read, while it does contain very interesting information. Can you add some lines or italic or bold text or something, to make it easier for the reader to focus? What is the ice free season number made up of? Please explain in caption. Also write in the caption what diff is (I know it seems obvious but still good to write down).

The style of the journal is to avoid additional line, but we removed the "%" symbol on row 3 and 6, remove the decimal on the row 1 and 2, and arrange the alignments (probably increase the spacing a little) to improve visibility. The ice-free season is the average over the whole experiment duration from April to November. This is now clarified in the caption.

Note also that the "Preservatives" labels were inversed, lower values (1st and 4th rows) are from "unfixed" samples analyzed for DIC while the highest values were those from samples fixed with HgCl2 (2nd and 5th rows).

- Fig. 3. Is the lower panel a useful addition? Or can I already get the same info from the upper panel?

We decided to keep it as it is informative and helps to ensure we use the correct equations and formulations for pH and [H+].

- Fig. 4. Isn't this the exact same info as in table 3? No need to show it twice. I think the graph is much nicer, it brings across your point very clearly.

Table 3 and Fig. 4 both display the monthly mean, yes. In addition, Fig. 4 displays the observations and interpolated daily data. Table 4 also shows the relative difference between fixed and unfixed samples in % as well as the mean for the whole duration of the experiment. Since this is a technical paper, we believe it is useful to keep both for clarity and depending on the reader's affinity for table or figure. However, we agree that we should specifically state that these two objects display the same data. This is now clarified in the captions.

- 357-362. Please address why the t0 concentration in the inhibited samples was so different from the control sample, and why the range in concentrations was much larger. Is it because of sample contamination with air during the addition of the inhibitors? If this is the case, that is not necessarily a bad thing, if you then also explain that that is one of the risks of inhibitor additons to samples.

This is now addressed in the start of the discussion L. 494-519

- 382. Do you suggest that the microbial processes are not inhibited, or that there are abiotic proceses at play? If it's the microbial processes, then how is it possible that these microbes are not inhibited, but the ones using O2 are?

Yes, we suggest that microbial processes are not completely inhibited. Note that certain metabolic pathways can be selectively inhibited and N2O is likely much more sensitive (being at 10's nM levels) to subtle microbial activity than O2 (being at 100's µM levels). Given the none essential character of this information, we decided, after all, to not highlight it. The acidification caused by CuCl2 and HgCl2 is already a sufficient reason to reject its use.

- 386. Please mention whether there were statistically significant changes (between timepoints or between treatments) for N2.

There were no significant changes between timepoints or between treatments for N2. This has been added (L. 435).

- 408. In the CuCl2 treated samples, you have both O2 consumption and CO2 production. Why do you think these are not linked?

Yes, good point. CO2 production in the CuCl2 treated samples is likely partially link to O2 consumption through microbial respiration. However, the CO2 production being much larger than O2 consumption, an additional source of CO2 is needed. This is described L. 525-532

- I have not provided detailed comments on the later sections, as I think it important to know more about the CO2 results first, like I stated in my starting comments.

We hope, we have provided some clarifications. Thank you for your assessment.

Reviewer 2:

- General comments
- The technical note describes outcomes from an experiment examining the suitability of three preservatives for the quantification of dissolved gas concentrations, and another

experiment to determine the feasibility of HgCl2 preservation to derive CO2 fluxes from freshwater systems. Despite being toxic, HgCl2 is a commonly used chemical that prevents biological degradation of gas dissolved in water, even though alternatives exist. The study shows that these alternatives are effective and suggests substituting HgCl2 for less toxic preservatives. The results of this study are technically relevant, help reduce and avoid errors in flux estimation and support the implementation of user-friendlier substitutes for the preservation of freshwater samples. I think this study could be very valuable for many researchers in the field and I would like to thank the authors for their nice work. However, the manuscript would benefit from clarifications, and more details especially regarding the Methods are needed before publication.

We are grateful to the reviewer, Thank you so much for this thorough, constructive, and in-depth review. Thank you for pointing out concrete needs for clarifications and improvements. Below we provide response to the main comments, to all numbered comments and technical corrections.

Specific comments:

- Are the studied lakes representative for other lakes or waterbodies? Please clearly outline limitations of this study in terms of impact and application in a broader sense.

Thank you for raising this. Svartkulp is particularly representative of Northern Hemisphere lakes, typically found in granitic bedrock regions in North-East America and Scandinavia. It is a typical low-productivity, heterotrophic, slightly acidic to neutral, moderately humic lake. Similar lakes are found in Southern Norway (de Wit et al., 2023), large parts of Sweden (Valina et al. 2014), and Finland, Atlantic Canada (Houle et al., 2022), Ontario, Québec and North-East USA (Skjelkvåle and de Wit 2011; Weyhenmeyer et al., 2019). Note that even if Svartkulp is among the best buffered lakes in Norway, our findings are also relevant to more acidic, lower ionic strength lakes found in Norway and large parts of Northern Canada.

Lundebyvannet, is also representative of a large group of these Northern lakes, however, it is quite a productive lake with high photosynthetic activity, which is more of a end-member case (e.g., worst-case scenario for Norway, related to CO2 flux overestimation with HgCl2 fixation).

This is now highlighted in the Site description sections L. 131-136 and 214-216, as well as in the discussion (L. 642-646) throughout the manuscript.

- Is it feasible to assume unfixed samples to represent “real” concentrations/fluxes, as control? Could you discuss this further and eventually consider renaming “control” to “unfixed” for the first experiment?

We have reorganized the results and discussion and now start with a discussion on how representatives these unfixed samples are to real conditions. See L. 496-519. For clarity we decided to have a more focused Results section and separate Discussion.

- The Methods section lacks necessary detail. I would suggest to restructure the section to make the experimental setup and respective study lakes clearer. E.g. in the study area section Lake Lundebyvannet should also be introduced, and ideally both lakes should be presented with the same level of detail relevant to the respective experiments. More importantly, I'm missing information on sampling procedures and their feasibility. Since this

is a technical note, I believe the Methods should be sound. I added several comments in this regard below.

Excellent point. We have now described the sampling methods in much more details and present both lakes with a similar level of details. Please see L. 116-158 and 192-213

- I think the results of this study could be put into clear recommendations for future studies, and this could be part of the abstract and expressed more clearly in the discussion.

Thank you for another nice suggestion. We have now added some recommendations to the abstract (L. 32-34) as well as within a designated section in the discussion (L. 648-650 and 664-670)

- The Introduction could benefit from adding some information about the other preservatives studied. The application of $HgCl_2$ is broadly introduced (could be shortened), but the description of the substitutes dealt with in this study falls short. Are there any other studies where $CuCl_2$ or $AgNO_3$ were used to determine dissolved gas concentrations? Are there differences expected between the application of those two?

Agreed, we re-organize and streamlined the introduction. To our knowledge there is no study where $CuCl_2$ or $AgNO_3$ were used to determine dissolved gas concentrations, however, there is one study showing that $CuCl_2$ amendments to soil lowered the pH, we now refer to it. See L. 75-78, as well as L. 89-90.

Please note, the numbers at the beginning of each comment denote the line numbers.

1. 26-27: can you be more specific about time periods (3w, 3m)?

Yes, see now L. 27.

2. 29: are low ionic strength / high DOC lakes representative?

Yes, see L. 30-31

3. 30: are these estimations valid for other lakes?

Yes, see L. 30-31.

4. 31: I think explicitly adding recommendations here would be useful.

Yes, see L. 32-34

5. 59: better in regards to what?

This has been rephrased, see L. 63-65.

6. 67: what is the impact of higher H^+ concentrations?

See l. 86-89

7. 70: could you elaborate why this leads to an overestimation of CO_2 concentration?

Yes, we will elaborate here. See also l. 86-89

8. 82-92: it would help the reader if it was made clearer here that two different experiments were conducted and two different lakes were sampled for that, for example by 1)... 2)...

Agreed, we rephrased here see L. 97-111

9. 87: This assumes that unfixed samples are the control, or "real results". Is that feasible?

Agreed, we removed the term control and now refer to unfixed or unamended samples, e.g., L. 106-107.

10. 99: Did you collect the water from the surface? Did you use anything other than the bottles to avoid bubbling or degassing? Were samples temperature controlled (or otherwise controlled) between sampling and analysis?

This is now better described, see L. 116-130

11. 100: slowly poured - a bit vague? How could you guarantee no degassing?

This is now better described, see L. 139-158.

12. 103: Are the results you got from the water samples representative for lakes in the region in terms of magnitude? Do the numbers represent means of the sub-samples or was each sub-sample used for determination of one of the parameters?

Yes, see also our response to your 1st comment and see L. 131-136.

13. 105: As far as I understand, the concentration of platinum does not necessarily describe the color characteristics of water?

This has been removed since it is not essential.

14. 106: how did you measure the temperature? is this an important information if the water was transported to the lab? Or did you preserve this temperature during transport?

This is now clarified (L. 128-129), 18.5C was in the lake, we also monitored the temperature in the lab.

15. 111: technically 3 treatments and one control. Which of the scenarios would presumably result in the most "real" concentration?

See our response to 2nd comment above and start of the discussion.

16. 112: why did you choose these time steps? could you elaborate if these times are representative?

These times are now justified, see L. 145-147.

17. 116: Is the preparation of the solutions part of the experiment? do the yielded concentrations have an uncertainty? or would a derivation not have an impact on the outcome?

This is now described L. 159-163.

18. 133: Did the fact that pH was not measured affect your study? Or was that one reason to use the PHREEQC model?

Admittedly, the experimental design was imperfect and pH measurements following fixation should have been included. Now to overcome this, we added a small 24h incubation where pH was measured following the same treatment, see L. 174-188 and results L. 438-443 as well as new Fig.2

19. 137: Is the sampling strategy outlined different than the one for the first experiment? How did you achieve sampling water from different depths? It would also be nice if both lakes were described with the same level of detail.

The description of the sampling methods has been revised and should now be clear. See L. 192-213

20. 145: Could you clarify the purpose of DIC analysis in this study?

DIC analyses were performed to obtain an independent estimation of CO2 and DIC concentration, compared to the GC analysis. This is now added at l. 107-110.

21. 147: Add name of TOC analyzer and/or merge with other sections below to avoid repetition. You state that samples were not fixed – why not?

This sentence has been removed.

22. 151: Did you compare the pH data with that measured with the pH-meter as mentioned above, or why measure twice?

We apologize, this is an error. pH was not measured in the laboratory, we only used the pH data from the *in situ* HOBO sensor. This sentence will be corrected.

23. 160: The temperature was recorded during shaking – do you mean the water temperature? What was the purpose?

Yes, the water temperature (sorry for the lack of clarity) was recorded during shaking. This is now clarified with Eq. 1 L. 246-252.

24. 171: Do you mean ambient air was used for calibration? Did you know the concentrations of the ambient air?

Yes, ambient air was used for O2 and N2 calibration. Ambient air is measured regularly and is stable through time.

25. 175 section: I think it would help to directly add formulas in a section in the appendix for better understanding and reproducibility.

Good suggestion, thank you. Eq. 1 was added

26. 187: Could you explain the purpose of DIC analysis here or in earlier sections. Are the CO2 concentrations calculated in addition to the concentrations measured by GC for comparison? I think it may not always be clear where you used measured or calculated CO2 concentrations.

This is introduced earlier l. 107-110.

27. 244: Is it important to mention what files were input and output files? For someone who doesn't know the program, this info seems meaningless.

This is now clarified. See l. 312-313

28. 255: Is this analysis done in retrospect to make up for not measuring sample pH directly after storage (among other things)?

Yes partly. Now we have added a 24h incubation experiment to complement as well. See see L. 174-188 and results L. 438-443 as well as new Fig.2.

29. 320: What temperature did you use to determine the Schmidt number?

This is now clarified. See l. 388

30. 328 f: It is nice to have different temporal resolutions, but what is the purpose of that for this study? Would your measurements not reflect instantaneous fluxes (could maybe be considered as daily fluxes) rather than weekly?

The main idea to show fluxes with these three temporal aggregations is to highlight the magnitude of the mis-estimation of the fluxes when HgCl2 is used as preservatives for the water samples. The error magnitude can be much larger over shorter timescales (see. E.g., Fig. 4).

31. 340: A rather general comment to this study: what is the assumption about the development of gas concentration in between times 0, 3w, 3m? E.g., what would the concentration after 2w or 2m supposedly look like? Did you examine that?

We haven't looked at other time points, this is difficult to predict. We try to only present the data we have and not over interpret. We don't believe there should be any revision related to this comment.

32. 362: Would preservation with AgNO3 then be preferable rather than with HgCl2 due to its toxicity? Can you draw conclusions regarding CH4 from your results?

Yes definitely. These recommendations are now given in the abstract and discussion. L. 496-521

33. 364, Fig. 1: Concentrations of all gases (except CO2) show largest ranges for AgNO3 addition (largest bars) after 3w. Is there an explanation for that? Add to caption: what do the boxplots show, presumably 25th and 75th percentiles and the median?

Unfortunately, no we have no explanation for the relatively larger range for the AgNO3 fixed samples after 3w. We added the description of the boxplots in the caption. See l. 429-430

34. 375 f: Are you arguing that this process is slowed down in freshwater?

Not necessarily slowed down because of freshwaters, there are many parameters playing a potential role here, e.g., temperature, substrate concentration, etc. Rees et al. (2021) performed their incubations at ambient temperatures which is the most likely explanation for the difference seen with our observations. Our samples were stored at 4C for 3 months. This is now clarified. See L. 563-567.

35. 380-381: Is this assumption reflected in your results by the decrease of N2 concentration? Is there also an explanation for the N2O consumption following production?

The interpretation of small changes in the N2 data should be avoided since none of the groups are significantly different from each other. N2O consumption following production has been observed by others, but no specific explanation was proposed.

36. 385: Did you perform a statistical test here too? Is it worthwhile mentioning that the concentrations seem to have the opposite response over time than N2O?

Yes, we did a statistical test for N2, this is now added L. 436. The changes for N2 and N2O do not have the same magnitude, µM for N2, nM for N2O. The expected changes in N2 from the process mentioned in comment #35 is not detectable.

37. 422: The opposite of what you state in the text is shown in Tab. 3. Is there a mistake in the labels?

Yes, there is a mistake in the labels in Table 3, this has now been corrected.

38. 426 f: Wouldn't we expect to see a shift in pH then in Fig. 2 (top panel)? It appears as if the fixed and unfixed samples have the same pH?

We apologize for the confusion. The pH plotted here in the *in situ* pH which is the same for both sample sets. pH was only determined with sensors in situ, this is now clarified in the caption.

39. 435, Tab. 3: What was the reason to show fluxes calculated following Cole and Caraco and not the other wind-based models here?

This was to avoid overloading the table, the values are different but the relative differences between fixed and unfixed samples is bound to the original concentration data. We believe there is no point in showing three models showing the same differences.

40. 466: Why was this cut-off of 20 µM chosen?

This 20 µM cut-off was chosen as the maximum likely error from e.g., pH error of 0.05. This is clarified L. 673-674.

41. 503: Which of those shown in Tab. 3 and Fig. 4 were obtained from DIC analyses?

This is now clarified in the caption of now Fig. 5. See L. 692-693.

42. 507 f: This estimate is only valid for the tested lakes. What would be the implication for other lakes? Is this also valid for sea water samples? Do you have recommendations or a protocol that should be followed? And what about greenhouse gases other than CO2?

Excellent questions, thank you. This is now addressed in section 4.4 from L. 635 as well as section 4.1.

Technical corrections

1. 18: what regulations are there? Or do you mean something like “complex handling” instead of regulation?

We refer to the Minemata convention as described in the introduction and conclusion. We clarified L. 18

2. 49: check brackets

Thank you!

3. 66: use abbreviation DOC

Done, thank you.

4. 75: the paragraph could be moved to discussion

Done, thank you.

5. 87: I don't think it's necessary to mention the storage temperature here.

Removed, thank you.

6. 95: determination or rather quantification?

Corrected, thank you.

7. 100: replace “gas loss” with “degassing” throughout.

Corrected, thank you.

8. 106: I think following the journal's guideline you would want to state what NIVA stands for.

Corrected, thank you.

9. 125: silver, not Silver

Corrected, thank you.

10. 132: Add name of gas chromatograph. Maybe it would make sense to merge this section with the Gas chromatography section further below, and move some information to the supplement.

We decided to keep the gas chromatography description below since it also applies to Lundebyvannet samples.

11. 193: I think this description is great, but could be partially merged with sections above and equations moved to a separate section in the supplement.

We preferred to keep everything for clarify, This is a method paper, we decided a thorough description of the methods.

12. 198: pK or K?

These are in fact pK.

13. 205: for completeness state value/equation of K?

The equation can be found in Stumm & Morgan.

14. 206: rather than “given” use “approximated”

Corrected thank you.

15. 208: add altitude “above sea level”

Corrected thank you.

16. 212-214: stick to either air-water or water-atmosphere interface

Corrected thank you.

17. 230: add: in percent

Corrected thank you.

18. 239: without knowing (the power of) this program, I would suggest to move this section or part of it to the supplement, and maybe worth explaining briefly what the program does starting with what PHREEQC stands for. How well does the program perform in predicting variables?

We believe that this section is given with the appropriate level of details and doesn't belong to the SI. It helps the interested reader. In addition, the justification comes early in the description L. 310-312. It is not the place here to describe PHREEQC and what it does.

19. 258: thermodynamically

Corrected thank you.

20. 318: double-check equation numbering

Corrected thank you.

21. 357: The concentration of CH4 [across experiments] ranged…

Corrected thank you.

22. 377: Maybe reword to something like: the prevalence of N2O production in [...] was attributed to favoring more acid conditions.

This sentence has been displaced and rephrased.

23. 411: samples not sampled

This sentence has been displaced and rephrased.

24. 418: indices, not indexes (also change in table)

Corrected, thank you.

25. 432: maybe not necessary to mention Lake Lundebyvannet twice in the caption

Corrected, thank you.

26. 435, Tab. 3: Correct column labels (preservative-addition and None reversed). Remove one "Lake" in caption. Mention what Diff (%) means. No need to include % in each column. Following the journal's guidelines, all figures and tables should be denoted with abbreviated Fig. and Tab. Double-check throughout the manuscript.

Corrected, thank you.

27. 445: as instead of than

Corrected, thank you.

28. 461, Fig. 3: What does i stand for? pH for each sample? I don't think it is needed in the x-axis label then.

Yes, yes it stands for each sample. this has been corrected, thank you.

29. 499: has instead of would have

Corrected, thank you.

30. 503: Fig. S3 shows daily fluxes, not monthly as stated in caption, right?

Yes, this is now corrected, thank you.

31. 505: "in reality" is based on samples without fixation? is that feasible? Did you mean to cite Fig. 2 instead of Fig. 3?

See our response to your second main comment. Yes, we referred to Fig. 2, which is now Fig. 3.

References:

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