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Reply on RC2

Mélissa Laurent et al.

Author comment on "Relationships between greenhouse gas production and landscape position during short-term permafrost thaw under anaerobic conditions in the Lena Delta" by Mélissa Laurent et al., Biogeosciences Discuss., https://doi.org/10.5194/bg-2022-122-AC2, 2022

Laurent Biogeosciences Reviews & Response to reviewers:

We thank the three reviewers for their helpful comments and suggestions, which help to improve our manuscript. In response to the thoughtful and constructive comments from the reviewers, we made major revisions to this manuscript throughout all sections and most figures. A summary of these changes is given here, as well as the detailed response to reviewers below. Attached is a PDF file containing the responses to all reviewers. The major changes include:

- *Most of the reviewers pointed out that the results would be more relevant and interesting if the incubation time was longer in order to overcome the lag time before methane production that we observed. Unexpectedly, our results showed that after two months of incubation at 20degC, under anaerobic conditions, only one sample layer produced CH4. We originally focused on a short-term incubation because we believe that it is essential to quantify C production under realistic timescale of a growing season (~60 days) in Kurungnakh Island because the aim of this study was to quantify the C production during the growing season under wet conditions communities and identify factors (microbial abundance, substrate availability) that would limit CH4 production in this case study site. However, since only the active layer of the floodplain started producing CH4, we decided to keep the incubation running to see whether the other cores would produce CH4.*
- *We've included this additional incubation data from days 68 to 363 of the extended experiment. We incorporated additional incubation data into the revised manuscript throughout and have produced a new figure with the cumulative production over a 363-day period. The revised manuscript now shows anaerobic CO2 and CH4 production over a 363-day period.*

To summarize the results from this longer incubation period, the floodplain core produced CH4 within the first 60 days due to the already established methanogen communities, as we showed in the initial manuscript. After 6 months of incubation, the permafrost layers from the Yedoma cores started producing CH4. This important result was not included in the earlier manuscript version with the shorter incubation time, as noted by all the Reviewers. Old Figure 4 shows that the permafrost layer in P15 and P16 has a lower methanogen concentration than P17-A, so we attribute the difference in lag time primarily due to the time required for the Yedoma samples to activate the methanogen communities *and to produce CH4. We hypothesize that the lack of methanogens in the P16-A and P15-A could be due to the dry condition induced by the landscape position. This indicates that methanogenesis is unlikely established after permafrost thaw in these sediments unless colonized by methanogens and the lack of response of CH4 to the glucose addition and continued anaerobic CO2 production also reduces the likelihood that substrate availability limits CH4 production despite the lower C abundance compared with the floodplain soil (Table 1).*

Additionally, the results section has been clarified to distinguish missing versus zero data within the microbial dataset.

- *The introduction has been revised to address the concerns of the reviewers, to be more precise and specific about permafrost carbon, the permafrost carbon feedback, and earlier incubation studies. We both narrow the focus to findings from earlier incubation experiments and elaborate on the specifics of the findings regarding the landscape position. In detail, we include a definition, discuss what differs across landscape positions, and expand on the links between microbial abundance and CO2 and CH4 production.*
- *We substantially revised the methods section to include more details addressing the criticisms of the reviewers and clarified terminology throughout the manuscript (e.g. "production").*
- *We substantially revised the discussion in order to address the criticism from the reviewers about the overly broad implications of this study despite the limited number of permafrost cores. It is now substantially shorter. We remove Figure 5 (the conceptual diagram) in the revisions. We clarify throughout the manuscript that this is a case study based on permafrost cores from Kurungnakh Island, Siberia, Russia. We shortened and narrowed the discussion to a case study, which aims to understand and quantify the potential C production in this limited region by integrating information about the influence of landscape position, microbial data, and, soil parameters to understand the factors controlling C production in this site within the Yedoma dominated region. We would like to note, however, the importance of these particular permafrost cores collected at a remote field site in Arctic Siberia, and this data given that it is no longer possible to re-sample at these sites in the foreseeable future.*

Reviewer 2:

This paper aims to address some major knowledge gaps on a consequential subject--namely, the controls on potential carbon feedbacks from warming in permafrost regions. These controls are poorly understood due to the many interacting factors (e.g. temperatures, redox conditions, organic matter quality, composition of legacy microbial communities, etc.) that affect CH4 and CO2 release, and this paper does contribute somewhat to the knowledge base. However, I found this paper to be lacking in terms of the strength of the findings.

First of all, only three cores were analyzed, despite the heterogeneity of the landscape. This is partially compensated by comparisons with other studies, including an extensive discussion of the results in comparison to other cores from the same region analyzed by Herbst (2022).

As explained in the general answer, this study is a case study in Kurungnakh Island. The geologic history of this site is well known, however few studies have worked on the potential CH4 and CO2 production after permafrost thaw at two temperatures. Here, we contribute to quantify and understand the potential C loss after permafrost thaw along a slope profile from the active and permafrost layers, and compare them with a floodplain in Kurungakh Island. Our findings show the landscape position plays a key role in the establishment of methanogen communities during the growing season. It is likely that *under field condition, only the floodplain produce CH4 during the growing season, this season might not be long enough for the upland and slope to establish methanogen communities. However, with longer thaw time they produce CH4 in the permafrost layer.*

As mentioned above, our study is a case study of Kurungnakh Island. We worked with three cores, but we analysed those cores at two depths and incubated them at two different temperatures. We would like to point out that there is still a lack of data regarding incubation studies in Siberia, and only few studies have been working with the active layer and permafrost layer, and two incubation temperatures. Finally, to establish pan-arctic dataset and compare data throughout the Arctic, smaller studies are essential.

However, the bibliographic entry for Herbst (2022) did not include a link to that manuscript, and I was unable to find it through a web search. Is that manuscript planned to be published in the near future?

The Master's thesis from Herbst (2022) is now available via a permalink on the AWI preprint server EPIC, we added the link in the bibliography section.

Second, the conclusions about microbial abundances do not seem supported by the sparse amount of data shown in Fig. 4. This issue might be partially addressed by better delineating zero abundances vs. truly missing values in the figure, but that depends on how much of the data is actually missing. (see specific comments below)

Thank you for this comment. Based this comment and the one below, we edited Figure 4 and clarified which data was missing and which is zero. We revised the text to clarify that no methanogen was detected for the samples without values and that these should be interpreted as zero values. Methanogen were found only in permafrost layers of P15 and P16, and the active layers of P15-P17. After one year of incubation, the results show CH4 production only for the samples where methanogens were detected (except P15-A).

Finally, the Discussion and Conclusions include numerous statements about how the results can improve predictions of greenhouse gas release under permafrost thaw, but the most significant result (higher methanogenesis in the floodplain active layer) doesn't seem to directly address the effects of permafrost thaw, as the P17-A sample is from the active layer of a floodplain--unless that floodplain location is part of a thermokarst feature, or that sample was formerly part of the permafrost before active layer deepening; but this is unclear from the site description.

Given this comment and the feedback from the other reviewers about the discussion and the shortcomings of the incubation length (major revisions summary #2, #5), we have changed the scope of the discussion to focus more on the relevant results. Additionally, we are now able to quantify the potential effects of permafrost thaw by demonstrating that the lag time before methane production in these thawed upland soils exceeds the length of the current growing season, so CH4 produciton will likely not be an immediate effect of this yedoma thaw under an active layer deepening scenario unless there is some abrupt thaw and landscape change.

Specific comments:

li 83-85: Remove extraneous reference text outside the parentheses (3 occurrences).

Thanks for the comment, we have corrected the text.

li 132-133: After looking at Fuchs et al. (2018), I could not find any information about the "relationship between absolute water content and bulk density."

Thank you for pointing this out. First, we apologize for the wrong reference, which may have caused this confusion. We corrected the reference by "Fuchs 2019". In his thesis, Fuchs determined the bulk density and the water content of one thousand samples from the Lena Delta. To calculate the bulk density, he divided the dry weight of a sample by its initial volume. With the data from the bulk density and the water content, they established a transfer function to determine the bulk density of a sample when the volume is unknown. As we explained in the manuscript, we did not calculate the bulk density, but estimated it according to this transfer function. In addition, the samples used for this study come from the same area as the samples used to establish this correlation. We changed the manuscript and explained the bulk density estimation in more details.

li 142-143: How much sterilized tap water was added to the low-moisture samples?

We calculated the amount of water to add to reach 30% of water according to the water content, and the weight (dry and wet) of the samples. Therefore, the amount of added water differs for each sample.

li 164-165: Does the "cumulative emissions" used for calculating the Glucose Factor also include the time before the day 60 glucose additions? If so, this factor might be overly sensitive to random variations in production rates before the additions. (Also, the wording of this sentence is unclear. Suggested rephrasing: "The impact of glucose on CH4 and CO2 production was quantified as a glucose factor, calculated using the cumulative C emissions at 67 days:")

This is a very good point. In the revisions, we will calculate the glucose factor only after glucose addition and see if we have different values. If so, we will modify the results and the discussion based on the results.

li 167-168: Related to the above, the phrase "total CH4 production rate at i days" implies an instantaneous rate measured at several (i) timepoints, as opposed to cumulative only at 67 days (from line 164). Which method was used? (If only cumulative, then the phrase "at i days" seems extraneous.)

Thank you for this comment. We used only the cumulative C emissions. Therefore, we deleted the phrase "at i days" and replace the words "production rate" by "production".

li 173: I assume "after glucose addition" means at Day 67? Please clarify.

We clarified the text by adding "67 days".

li 202: Typo ("Kuskal-Wallis" should be "Kruskal-Wallis").

Thanks for the comment, we corrected the text.

.**li 216:** "the lowest [C:N] were in P17": Did you mean P16 (Table 1)?

Yes, that is correct, we meant P16, thanks for your comment.

Table 2: Typo in second-to-last row of first column (first occurrence of "P17-F" seems like it should be "P17-A").

Thanks for the comment, we corrected the text.

li 247: It would be clearer to cite Table 3 (from which the 42.53 ± 15.79 value is directly derived) in addition to Figure 3.

Revised as suggested.

li 251-252, "P15 and P16 behaved similarly, with higher CH_4 production for the active layer at 4 °C than at 20 °C": This doesn't appear to be true for P16, based on its active layer Q10 being >1 (see Table 2).

Thank you for pointing out this mistake. P16-A has higher CH_4 production at 20°C. We corrected the manuscript.

li 253, "...and no difference for the permafrost layer": This also seems surprising, given that for P16 (Fig. 2b), the blue line (permafrost at 4 degC) is noticably higher than the red (permafrost at 20 degC).

Even though the blue line is higher than the red one, the values of the production rates from the samples at 4°C are still very low and cannot be considered as a real methane production from the samples (samples are still in lag time). In addition, the error bars for the permafrost layer at 4°C and 20°C overlap, meaning that the values cannot be statistically considered as significantly different (supported also with the statistic test). We discussed this in the section 4.1.

Figure 2: Several comments:

- Dashes are missing from the lines in Fig. 2b.

Thank you for this remark. We modified the figure.

- In Fig. 2c, due to the very high Active Layer 20 degC values, it's impossible to see what's happening with the other samples. Would it be possible to create another version of this panel (perhaps for the Supplement) with the very high CH_4 values removed, so that the differences in the other lines can be seen?

Thank you for this good remark. We added a zoom in version in the supplementary figures to see the behaviour of the other samples.

- Some of the plots, particularly Fig. 2f, show negative CO_2 production rates. How would you explain these?

These negative production rates appear mainly the days where we flushed the samples, therefore it is likely due to the flushing. In the revisions, we replace this figure with one showing the cumulative CH_4 and CO_2 production where the trends are clearer.

Figure 4, li 320-321, "Absence of values for some samples is due to either low DNA concentration or failure in qPCR run.": Can you indicate on the figure (maybe using a symbol) which empty values were due to which cause (low concentration vs. failed qPCR run)? This delineation of zero vs. missing values would help a lot with the interpretation of this figure, as a zero (or below detection limit) concentration still represents the information that concentrations were low, as opposed to not measured at all.

Thank you for this helpful comment. We edited Figure 4 based on your comment, by adding the expression "below detection limit", or "not detected".

li 330: Wrong table references for CO_2 production (and move "Table 1" reference to line 329 or 331 about C and N contents)?

Revised as suggested, and Figure 2 added to support "low CO_2 production throughout the incubation".

li 390-391, "methanogen concentration before incubation showed the highest numbers in the floodplain (Figure 4c)": I can't tell whether this statement is supported by Figure 4, as the zero values aren't distinguished from an absence of measurement (see Figure 4 comment above). If all the empty values are actually missing (i.e. due to failed qPCR), then no direct comparisons of the pre-incubation samples would be possible between P17 and the other sites.

Half of the values from the "pre-incubation" were not measured. In the revised version, we carefully compared the values and specified that the microbial results were in line with the CH4 production, but missing values (likely due to too low concentration) make it difficult to establish a comparison with certainty.

li 396, "little change in methanogen quantity after 60 days of incubation": This doesn't appear true for the P16 permafrost layer incubated at 20 degC, which had much higher mcrA (Fig. 4b).

Yes, that is correct. We changed the text according to the comment and specify that this does not apply for P16 at 20degC.

li 404, "after permafrost thaw": Are the portions of the floodplains sampled by Herbst (2022) part of thermokarst features?

The samples incubated by Herbst (2022) were not part of thermokarst features. They incubated samples from the floodplains, and samples from the permafrost layer belonging to the same cores.

li 451: Typo; "three time" should be "three time**s**".

Revised as suggested.

li 461-463: Invalid sentence structure; did you mean for the end to read "**CH4 production** will likely increase" ?

Yes, you are right, thanks for pointing out this mistake.

li 472, "methanotrophic": Did you mean "methanogenic" ? or both?

We meant "methanogenic". We changed the manuscript.

Supplementary Figure 1: Which incubation temperature is shown here (or is it an average of both)?

The incubation temperature shown in this figure is 20degC. We added the temperature to the figure title and the figure.

Please also note the supplement to this comment:
https://bg.copernicus.org/preprints/bg-2022-122/bg-2022-122-AC2-supplement.pdf