Comment on bg-2022-37

Anonymous Referee #1

Referee comment on "Temperature sensitivity of soil organic carbon respiration along the Rwenzori montane forests elevational transect in Uganda" by Joseph Okello et al., Biogeosciences Discuss., https://doi.org/10.5194/bg-2022-37-RC1, 2022

The manuscript by Okello et al. presents a potentially interesting dataset examining the sensitivity of soil organic carbon stocks to projected temperature changes. The work fits well in the scope of BG, and I much appreciate the important work done – but the presentation of the data and the interpretation does require a substantial amount of clarification and improvement before being reconsidered. Detailed comments below.

Dear reviewer, thank you very much for appreciating our work and equally for the meticulous review and insightful suggestions to further improve the manuscript. In the following specific sections, we pay attention to address the concerns raised.

Main comments & suggestions -Terminology:

*throughout the manuscript, the terminology related to stable isotopes is really not OK. For example, the authors refer to "the ¹³C depletion factor" or "isotopic depletion factor" (L167) – that is not an accepted term in the literature, what you are referring to is termed fractionation (epsilon).

Thank you for this remark. Indeed, as you correctly identified, our study focused on isotopic fractionation (epsilon), i.e. the change in the stable isotope composition of C during the transformation of SOC to emitted CO₂, as a result of discrimination against ¹³C during SOC transformation that involved physical and chemical processes.

We indeed also realised that, strictly speaking, it was incorrect to refer to epsilon as a factor, because in contrast to "alpha" (which we no longer mention in the new version of the manuscript), epsilon is a difference.

Further, we agree to rearrange Equation 2 in the new version of the manuscript as follows:

"

$$\varepsilon = \left(\left(\frac{\text{R-CO}_2}{\text{R-SOC}} \right) - 1 \right) * 1000$$
 (2)

Where:

R-CO₂ is the ratio of 13 C to 12 C in the emitted CO₂

R-SOC is the ratio of ¹³C to ¹²C in soil organic carbon (substrate)."

*other examples: L 321 "13C and content of soil organic carbon relatively increased"

We agree to revise this in the new version of the manuscript as follows: "Further, in warmed soils, the $\delta^{13}C$ values and soil organic carbon content increased and decreased, respectively."

*Keeling mass balance approach (line 155-162): this is just a mass balance approach, a Keeling plot is something quite different; equation 1 does not appear in the Keeling (1958) paper you refer to.

Thank you for this observation. Indeed, we agree that this is simply a mass balance approach and we shall add citations of this approach. While indeed Keeling (1958) used this approach in the Keeling plot method over several points of measurements, we used the mass balance approach for only two CO₂ measurement points. As such we just calculated the mass balance using equation 1 (line 165). We agree to rephrase this statement in the new version of the manuscript as follows:

"To determine the $\delta^{13}C$ values of the respired CO₂, we used a mass balance approach (Phillips and Gregg, 2001).

$$\delta^{13}\text{C-CO}_2 \approx \frac{[\text{CO}_2]_{final} * \delta^{13}\text{C-CO}_{2final} - [\text{CO}_2]_{initial} * \delta^{13}\text{C-CO}_{2initial}}{[\text{CO}_2]_{final} - [\text{CO}_2]_{initial}}$$
(1)

*L 146, 190, 289: " δ^{13} C isotopic composition": again, this is not appropriately phrased. Use either " δ^{13} C values" or "the C stable isotope composition" but not combinations of the two. We agree to rephrase throughout the manuscripts and use the term " $\delta^{13}C$ values". For instance for those specific lines:

L146,. "Immediately, one gas sample was taken using a 45 mL syringe. The ambient CO_2 concentration and its $\delta^{13}C$ value at "open condition" was analysed using Cavity Ring-Down Spectrometer, (G2113-I, CRDS CO_2 analyser, Picarro, United States)." See line 155-157. (The "open condition" is the initial sampling time (T_0))

L190. "The top 10 cm of the soil cores was collected (i.e. the soil layer with the highest C content and most active in C cycling), homogenised, air-dried, and sieved (2 mm mesh size) for additional laboratory incubation experiments, in order to assess the effect of two years of in situ warming on: (i) CO₂ respiration rates; (ii) the AE and Q₁₀ coefficient; and (iii) SOC content and δ^{13} C values." See line 196-199.

L289. "Further, to check for changes in CO₂ emission rates, AE, Q_{10} , SOC content and $\delta^{13}C$ values between control and in situ warmed soil at each elevation cluster, we used a Wilcoxon test." See line 299-300.

*enrichment in ¹³C isotope (L478): enrichment in ¹³C

We agree to rephrase as suggested.

*L373: $\delta^{13}CO_2 \rightarrow \delta^{13}C - CO_2$ or $\delta^{13}C$ of CO_2

Thank you for the observation, we agree to rephrase this to " $\delta^{13}C$ -CO₂" in the new version of the manuscript.

-PLFA data: there is a short section in the Methods outlining the extraction and derivatization of PLFA, and then basically nothing. No info on how PLFA were identified and quantified, no information on how the resulting data were treated – assigned to microbial groups etc. The data are presented later on as concentrations of PLFA representing gram-positive, gram-negative bacteria, fungi, etc. but no information or references are given; no mention of this in the Methods, and very little real discussion of

these data. Either add all this info, or remove them if the data don't contribute much to the story.

Thank you for this insight. We agree to add some procedures and citations of the method used from line 132. i.e. "Eventually, the phospholipid fatty acids were converted to methyl esters, which were subsequently analysed using gas chromatography (GC, Trace GC, Thermo Scientific, Bremen, Germany)" following the methods described by Denef et al. (2007) and Huygens et al. (2011).

Additionally we agree to add the following: "We determined the ratios of the peak area of each individual PLFA to that of C16:0, a universal PLFA occurring in the membranes of all organisms. PLFA ratios less than 0.02 were excluded from the data set (Drijber et al., 2000). PLFA was assigned to microbial fungal group following (Zelles, 1997) and (Chung et al., 2007). While PLFA assignment to bacterial group and to gram-positive and gram negative bacteria followed the procedure described by Kroppenstedt (1985) and Frostegård and Bååth (1996)" see line 135-140.

-precipitation: L101:7000 mm should be 700mm probably or something in that order of magnitude at least. The study sites cover a wide range of precipitation, and there is also a clear difference between Kibale and Rwenzori (as in: precipitation is higher in Kibale than at the lowest elevation along the Rwenzori transect)- however, the effect of precipitation on the data from the translocation experiments is not discussed at all; this should be worth some discussion.

We agree that there must have been an error in precipitation data at elevation of 1760 m. The available rainfall data from the Uganda Wildlife Authority (2012-2016) indicated unusually high rainfall at two of the six elevations where data were collected. When picking the ranges of rainfall, we selected one of these high values. We have now used the ranges to only include rainfall data for the realistic four data points (all with rainfall amounts between 1500-1800 mm per annum). As such, rainfall ranged from 1570 mm at 1760 m to 1806 mm at 4230. Therefore we propose to use these rainfall amounts in the new version of the manuscript (see line 94-95; 101-102).

Further, while we used this data to give an indication of rainfall in the Rwenzori Mountains, the data are not specific to our study plots (only one weather station is within the studied site

elevation). Therefore we cannot conclusively tell the rainfall trend in the elevational gradient. Due to this uncertainty, in our study plots we focused on directly monitoring the soil moisture content which indeed directly affects microbial activities. We discussed the effect of soil moisture elaborately. For this reason, we did monitor CO_2 efflux under in situ conditions in two key periods (start of rain and mid rainy season). Indeed, the results indicated an increase in CO_2 efflux following increase in soil moisture in the mid rainy season. Because of this we noted that soil moisture has an effect on CO_2 efflux (see section 4.3 line 528-541). Additionally, under laboratory condition when moisture content was standardized, we noted that CO_2 emission did not differ along the elevational gradient, but decreased linearly when standardised per amount of SOC (i.e., effect of temperature is isolated, line 371-372; 374-375; 459-460).

-L111-116: No mention is made on whether samples were acidified to remove potential carbonates (or carbonates precipitation from the soil solution during soil drying). C/N ratios are reported but you need to specify whether these are weight/weight or molar ratios. For a proper interpretation of data, specify the reproducibility of your measurements (e.g. for δ^{13} C) and mention which standards were used. In Table 1, specify then if these concentrations refer to organic or total C. If total C, then you may want to add a note of caution in the interpretation of differences between δ^{13} C of soil C and CO₂ produced.

Thank you for these remarks. Practically at the prevailing soil pH-KCl (5.4-3.3, see table 1), the soil is quite acidic, and the presence of carbonates is negligible. Secondly, in the wet tropics (where there is precipitation surplus), carbonates are commonly dissolved and leached down to deeper soil layers, yet we only sample the top 30 cm of soil. Given these reasons, the total carbon measured was taken as the soil organic carbon. Therefore, agree to explicitly mention this in the revised version of the manuscript in the methodology section from line 117-118.

-section 2.3: provide a description of how temperature was controlled during these incubations.

We agree with this comment, in the new version of the manuscript, we agree to mentioned it in line 150-151.:

-section 2.3: mention how you coped with removing a 45 mL gas sample from your incubation jars: was this volume replaced with air while taking these samples, if not how was the pressure difference accounted for in your measurements?

We agree that this is needed. We wish to clarify that in the in situ CO₂ measurements, the gas chamber was equipped with a vent tube to minimize pressure differences (mentioned in line 220-221). Additionally, for in situ CO₂ measurements, only 15 mL gas sample was taken at a time, (yet the total volume of the chamber was about 4 L). While for laboratory incubation, headspace gas sampling for the determination of soil CO₂ fluxes was done twice, directly at the beginning before fully closing the jar and at the end of the incubation period. For gas sampling a gas tight syringe (45 mL) was used, which was immediately gas-tightly closed after taking the gas sample. Gas samples were analyzed immediately using Cavity Ring-Down Spectrometer, (G2113-I, CRDS CO₂ analyser, Picarro, United States). Thus, during incubation the pressure in the jar was not affected. Moreover, immediately after the second sampling, the jars were opened and only covered with parafilm to equilibrate until the following day.

-L208: why refer to the "slope of the CO₂ concentration in function of time"? If I understand well, you simply have measurement at the start and end of the incubation ?

We appreciate this suggestion. Indeed, this we agree to rephrase as follows: "Eventually, the CO_2 emission rate was determined as change in headspace CO_2 concentrations (t_1-t_0) divided by the incubation time (24 hours) for laboratory experiments. Still for the in situ measurements we calculated a slope representing increase of CO_2 concentrations (N = 5) over chamber closure time (90 minutes)".

-L199 and further: any reason to go for 50 mL jars here instead of 1 L jars (as in section 2.3) ? For the δ^{13} C measurements, it's important to convince the reader that the data you collected from these experiments are valid: you are in a closed system, where you sample gaseous CO2 for δ^{13} C analysis, but you also have an aqueous phase. CO₂ will equilibrate between the two, and there is a (small) degree of isotope fractionation involved. The smaller the headspace volume compared to the volume of soil (and thus water), the higher the possible bias in resulting δ^{13} C -CO₂ data if not accounted for. It might be negligible in your setup, but you need to provide arguments to show this.

Thank you for this comment. We are aware of the isotopic fractionation between headspace and liquid phase, though this section you refer to is only about CO_2 headspace concentration measurements, but not isotopes. Further, as clarified in this experiment, firstly, we ensured that the headspace volume and moisture content was constant for all the samples. Secondly, in preliminary experiments we ensured that headspace CO_2 concentrations increased linearly over a time period of 24 hours.

-Section 2.5: while I am aware that much of the literature refers to soil CO₂ flux measurements using closed chambers as "soil respiration", one should avoid keeping using this terminology in use; what is measured is not total in situ soil respiration but the diffusive flux of CO₂ from the soil. This diffusive flux is governed by the gradient of CO₂ concentrations / partial pressures and is thus influenced by e.g. porosity, water content etc. Part of the CO₂ produced by soil respiration is lost via percolation and groundwater losses.

We concur with this statement. In the new version of the manuscript, we agree to explicitly mention in line 70-71 that we measured the in situ diffusive flux of CO_2 from the soil and used it as a close proxy for CO_2 respiration (hereafter referred to as " CO_2 respiration").

-Chamber deployment time (L224): 90 minutes seems excessively long for chamber closures, especially given the low chamber surface area. Pavelka et al. (2018, doi:10.1515/intag-2017 0045) and other recommend much lower chamber closure times, in the order of 5 minutes. Were the chambers equipped with a fan to ensure proper mixing within the chambers ? If the CO₂ increase was not linear, this has implications for your fluxes as well as δ^{13} C data interpretation.

Thank you for this comment. We are aware of the effect of increasing gas concentrations in the headspace on CO_2 diffusive fluxes out of the soil. While shorter chamber closure times are indeed preferable, this could not be realised under field conditions. At the time of the field campaigns, we did not have the possibility to do continuous measurements in situ (as the instrument broke down), so this was the best alternative. Further, we purposely took headspace air samples every 30 minutes interval, so that it was possible to check linearity with time and indeed the CO_2 concentration was always linear over the 90 minutes intervals (see line 204-206). Secondly, the gas chambers were fitted with a vent tube to minimise any changes in

pressure. As such, the conditions for a non-steady state closed chamber flux measurements were still respected. We agree to explicitly mention this statement in the new version of the manuscript from line 232.

Not clear, by the way, if all δ^{13} C -CO₂ measurements were made using a Picarro G2113, this is only mentioned for the t0 samples in section 2.3. If other CO₂ samples were measured using other methods, add the necessary info (equipment, standards, reproducibility) to your Methods section.

Thank you for this comment. For clarification, in the laboratory incubation in section 2.3, we used Picarro G2213 for both T0 and T24 as mentioned (line 155-157). While for section 2.4 (about the soil mesocosm translocation experiment), we used a gas chromatograph (Finnigan Trace GC Ultra, Thermo Electron Corporation, Milan, Italy) fitted with a thermal conductivity detector (mentioned in line 214-215). The gas chromatography measurements were only for CO_2 concentration and not for ${}^{13}C$.

-L237: linear regression: if you have 2 data points only, then avoid referring to this as 'fitting a linear regression to the concentrations over time'.

Thank you for this suggestion. We concur and adapted as above in the comment about L208.

-L402-403: 'the SOC contents of warmed soil were relatively lower than those of control (soils) along the elevational transect': While Figure 4e may indeed suggest this, this does not appear to be the case for the lower elevation sites + even for the higher elevations the large error bars do not suggest that this difference is significant. Quantifying small changes in SOC stocks is challenging – if the difference is not statistically significant, then avoid phrasing in the way it is currently done. If you do feel confident that these are robust differences, you need to provide statistical justification + provide an estimated analytical error on your bulk density and %C (or OC, see elsewhere) data. The same comes back on L 484 where you claim that SOC was relatively lower in warmed as compared to control [samples] – if these differences are not significant then such statements should be rephrased; this is what others will pick out as conclusions in subsequent work.

Thank you for this insight. Indeed we agree with this comment. For this reason, we first mentioned the following: "Results revealed that at each elevation cluster, there was no significant difference in the studied parameter between control and warmed soil (SI Table 3)" The high spatial and temporal variability in measurements of soil diffusive CO₂ fluxes often preclude powerful statistical tests of small treatment effects (Davidson and Janssens, 2006), as is usually the case in warming experiments. This effect is even more critical in montane ecosystems with different slope positions and aspects. Therefore, a parallel linear regression model fit along the entire elevation can help to reveal some "trend" between warmed and control treatments for soils taken at different elevations. To clarify further, these parallel linear model only reveals some trend but doesn't test for significant differences. The regression slopes indeed revealed a trend between control and warmed samples when fitted side by side.

Finally, we have added the statistics for SOC and $\delta^{13}C$ and also the slopes and intercepts of the linear mixed effect model for SOC and $\delta^{13}C$ for both control and warmed treatments in SI, Table 3.

-L403-404: the data presented in Figure 4f show a surprisingly large difference in δ^{13} C values, albeit with relatively high standard deviations. It would be good to provide statistics for this: for which elevations are these differences significant or not ? Again, I assume that for the high elevation sites, they are not statistically different – which should not be a surprise, as you have very organic soils here for which you would need to have a very high turnover rate to see any differences in δ^{13} C of the SOC pool after 2 years. You could likely do some back-of-the envelope calculations here.

Thank you for this insight. We agree with this comment. As mentioned earlier, L402-403 and L484, there were no significant differences between control and warmed samples for a given elevation, while using linear mixed effect model regression analysis across entire elevational gradient allowed to show a trend between warmed and control treatments. As such we agree to clarify in the new version that "the remaining SOC tended to be more enriched in ¹³C in warmed than in control, and the SOC tended to be lower in warmed than in control treatment". We agree to also add the statistics for SOC and δ^{13} C in SI, Table 3 along with the slopes and intercepts of the linear mixed effect model regression fit in the new version of the manuscript.

-Discussion, section 4.1. The discussion on differences in δ^{13} C between CO₂ and soil (organic) carbon needs to be reconsidered. The current discussion assumes that there should be a relationship between rates of mineralization and isotope fractionation during respiration. On line 480, you refer to Amundson et al. (2003) to back up this idea – but this is a paper that only discusses nitrogen stable isotope ratios in soils. As far as I'm aware, there is no sound evidence in the literature that the degree of isotope fractionation (if any) during respiration would be related to either respiration rates, or temperature as you hint at in the first paragraph of section 4.1. You also refer to Andrews et al. (2000) and Natelhoffer & Fry (1988) here, but these do not really back up such statements: (i) Natelhoffer & Fry merely demonstrate that the SOC pool is typically enriched in 13C as mineralization progresses, without unambiguously demonstrating via which mechanisms (selective mineralization or degradation, Suess effect etc – for an updated discussion see e.g. Ehleringer et al. 2000 Ecological Applications 10: 412-422 and subsequent literature); and (ii) Andrews et al. (2000) should be interpreted carefully here. Granted, they observed similar patterns for soils from FACE experiments and control soils, but note that they do not invoke kinetics in offering an explanation to their data: "The increase in respiration rate across the entire temperature range and the enrichment in ¹³C only at 4°C rule out a strictly kinetic explanation for the observed carbon isotope fractionation. In addition, there is no theory that suggests a very different ratio of reaction rates of ¹³C compared to ¹²C in slow versus fast reactions (Agren et al., 1996). We suggest that the shift in carbon isotopic ratios in respired CO₂ is the result of a shift in the use of carbon substrates in the soil". Hence, likely better to refer to a shift in δ^{13} C or to "apparent fractionation" then to fractionation. Note also that they observed a strong change between the first days of incubations and subsequent days, and that there are some methodological aspects to consider when interpreting their data: high volume of soil (and water) compared to headspace, and complete flushing of the headspace with CO₂-free air (which implies that CO₂ dissolved in soil water remained and will re-equilibrate, this dissolved CO₂ has a different δ^{13} C value that the headspace CO₂, etc).

In short, this entire section at the moment lacks a solid empirical or theoretical basis to interpret differences in δ^{13} C data observed to the influence of temperature or higher respiration rates. The same holds true for how some of the conclusions are expressed,

We appreciate this insight. We agree to revise the text in this section accordingly. First, we fully agree to refer to apparent fractionation rather than fractionation sensu stricto.

Secondly, we propose to add $\delta^{15}N$ data in Table 1 to also show the trend in $\delta^{15}N$ signatures along elevation. Indeed, similar to $\delta^{13}C$ signatures, the $\delta^{15}N$ signatures decreased linearly with elevation. This indicates a more closed N cycling with increasing elevation. The results of $\delta^{13}C$ $\delta^{15}N$ signatures and CO₂ emission rates along elevation suggest a lower mineralisation at higher elevations. Similarly, the apparent $\delta^{13}C$ fractionation decreased linearly along the temperature gradient. This could indeed be due to a decrease in apparent fractionation and a shift in the use of carbon substrate. Finally, we agree to update some citations in this section. Therefore, we agree to revise the text in 4.1 (from line 459-473) as follows:

"The specific heterotrophic CO₂ emission decreased with increasing elevation, partly in response to effects of lower temperatures on microbial activity (Zimmermann et al., 2009). In support of the temperature effect on CO_2 respiration (Figure 3(b)), the apparent fractionation during SOC transformation to emitted CO_2 was also temperature-dependent. The emitted CO_2 at the warmer, lower elevations showed a higher apparent fractionation (and subsequently became relatively more depleted in ${}^{13}C$) than at the colder, higher elevations (Figure 3(c)). This observation may result from a shift in the use of carbon substrates along the temperature gradient or potentially ¹³C discrimination during decomposition by soil micro-organisms (Andrews et al., 2000; Ehleringer et al., 2000; Natelhoffer and Fry, 1988). Indeed, the nitrogen stable isotope composition also indicated a decrease in $\delta^{15}N$ value with elevation (SI, Table 3). As such, based on the link between the nitrogen stable isotope composition of ecosystems and nitrogen cycling (Amundson et al., 2003; Boeckx et al., 2005), higher degrees of isotopic discrimination may indeed indicate a gradient in the rate of soil organic matter transformation processes likely including specific heterotrophic CO_2 emission. Our results imply that at higher elevations, even though SOC contents were high, microbial SOC decomposition was limited by lower temperatures (Zimmermann et al., 2009). In addition, low CO₂ emission at high elevations may as well partly be the result of the low soil pH values as those negatively affect microbial activity (Walse et al., 1998) and, thus, respiration (Figure 3(c), SI, Table 1). Further, a low pH also facilitates the stabilisation of organic matter through complexation reactions with iron and aluminum ions, which become soluble at a low pH (Lützow et al., 2006)."

e.g. L 536-539: the statements made are not convincingly supported at the moment

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Thank you for this comment. Following the revisions in section 4.1 above, we propose revise this statement to indicate the apparent fractionation.

L511-512: 'low soil moisture content limited microbial CO₂ respiration at high elevations: I do not see such lower moisture content anywhere in the data. Given the strong gradient in precipitation, I would expect to see rather the opposite ?

Thank you for this comment. The soil moisture content data indicated that the soil moisture content increased in the mid rainy season as compared to the start of the rainy season. Subsequently, CO_2 respiration increased in the same trend. Additionally, in the laboratory incubation, when soil moisture content was set uniform at 60% WFPS, we observed no difference in CO_2 respiration except when standardised per unit soil organic carbon, where it decreased linearly with elevation (suggesting an isolated temperature effect). These results indicate a boost of microbial activity probably due to the increase in soil moisture content in the wet season. Secondly, along the elevation gradient, soil moisture content tended to decrease with elevation in the mid rainy season (slope =-0.8, $R^2m = 0.25$, SI, Figure 3). While this trend is not significant, the average WFPS was highest in the lowest elevation (57.2 % compared to the rest of the elevation from 1750-3000 (44.8 to 44.5 %, SI, Table 3), and the same trend was observed for CO_2 emission. For instance, CO_2 emission correlated strongly with water-filled pore space in the mid rainy season (P = 0.01, SI, Table 1). On the other hand, soil pH decreased linearly with elevation in the same trend as specific CO_2 emission. Subsequently, a significant correlation was observed between soil pH and CO_2 emission (SI Table 1).

Therefore, we agree to revise in the new manuscript to point at the correlations observed rather than the causal relationships.

Further, as already clarified about precipitation under comment L101 above, we do not have sufficient data within our sites elevations to determine any trend in precipitation

-L528-530: "we showed that...": rephrase this, limit to what you really unambiguously demonstrate, relationships are not necessarily causal. For example, I do not see strong direct evidence that soil moisture or pH had a direct effect on soil respiration along your gradient.

While we appreciate that several variables change along elevation which makes it difficult to assess the effect of individual variables, we could point some relationships. Therefore, we propose to revise the language according to identify the correlative variables with CO₂ emission along the elevational gradient as explained under comment L511-512 above.

Minor / textual comments

-L20: insight into temperature sensitivity: insight into the temperature sensitivity:

Thank you, we agree to revise the sentence as suggested.

-L25 and further: temperature sensitivity: make it explicit that you are referring to Q₁₀ values here.

Thank you for this suggestion. We agree to revise as suggested

-L38: make it explicit that you refer to terrestrial primary production, not global (terrestrial + marine).

Thank you for this suggestion. We agree to rephrase the statement to clarify this.

-L61: delete "of the CO2 respiration from soil"

Thank you for this suggestion. We agree to revise as suggested.

-L89: in the eastern slope: on the eastern slope.

Thank you for this correction.

-Equation 1: bit of an odd choice of symbols – (F, f, I, i)

Thank you for this comment. We agree to revise the symbol as in the above comment under line 155-162, i.e.

$$\delta^{13}\text{C-CO}_2 \approx \frac{[\text{CO}_2]_{final} * \delta^{13}\text{C-CO}_{2final} - [\text{CO}_2]_{initial} * \delta^{13}\text{C-CO}_{2initial}}{[\text{CO}_2]_{final} - [\text{CO}_2]_{initial}}$$
(1)"

'-L172: why "increment" ? I think this can be deleted.

Thank you for the suggestion, we agree to delete the word.

Comment on bg-2022-37

Anonymous Referee #2

Referee comment on "Temperature sensitivity of soil organic carbon respiration along the Rwenzori montane forests elevational transect in Uganda" by Joseph Okello et al., Biogeosciences Discuss., https://doi.org/10.5194/bg-2022-37-RC2, 2022

It is my pleasure to read and review this manuscript written by Joseph Okello et al. I congratulate the authors on a very substantial piece of work, nicely written up, general nicely documented and discussed by the authors with novelty design and solid data. Indeed, it is interesting work. Indeed, the authors offer a manuscript that illustrates interesting findings supporting some hypotheses raised during the last years: first, that soil organic carbon respiration positively responses to soil temperature; second, that mineralization and depletion of readily available carbon in soil is also a regulator of soil organic carbon variation with the changing of soil physicochemical properties and microbial community-induced by climate warming over time. Overall I support publication of this work, yet I have some comments to be considered (moderate revisions).

Thank you very much for appreciating our work and equally for the careful review and insightful suggestions to further improve the manuscript. We are greatly humbled by your support!

Small comments are on Abstract /Conclusion to present the findings of the selected soil microbial community to be involved in the SOC respiration processes of Q10 models. And it is better to give a feedback to the findings. Also, SOC should be given an abbreviation in the beginning of the abstract.

We appreciate these suggestions. We agree to revise the manuscript to give feedback on microbial community that the microbial community structure was not affected along the climate gradient. Additionally, as suggested we shall abbreviate soil organic carbon as SOC in the abstract. It is a pity that we couldn't discuss more on microbial community. We noted that microbial community structure did not show significant effects with altitude nor CO₂ emission. We feel these results of microbial community structure along the climate gradient are important to include in the abstract. The result is consistent with several studies that found no effect of temperature on microbial community structure e.g. (Karhu et al., 2014; Nazaries et al., 2015; Wei et al., 2014).

Introduction: authors should give that the effect of soil microbial community on SOC during climate warming is not yet well established. Maybe this can be added to the introduction to better develop the current study. Not?

Thank you for this suggestion. Indeed, we agree to add a statement in the introduction about the controversies on the effect of soil microbial community on SOC in response to climate warming. i.e. "Several studies reported reduced microbial biomass in response to warming being linked to either depletion of labile carbon (Bradford et al., 2008; Knorr et al., 2005) or a decrease in carbon use efficiency (Allison et al., 2010; Tucker et al., 2013). However, other studies found no effect of climate warming on microbial community (Karhu et al., 2014; Nazaries et al., 2015; Wei et al., 2014). This means that the changes in soil CO₂ emissions upon warming result from alteration in the activity of native microbial community without altering microbial community structure."

Our study on microbial community along the microclimate gradient in montane forests is consistent with the latter findings.

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