

# Interactive comment on “Seasonality, drivers, and isotopic composition of soil CO<sub>2</sub> fluxes from tropical forests of the Congo Basin” by Simon Baumgartner et al.

Anonymous Referee #2

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General comments:

The paper by Baumgartner et al on soil CO<sub>2</sub> emissions from tropical rainforests in the Congo Basin is relevant and mostly well written. It addresses the knowledge gap on GHG fluxes from the African continent, which is still critically under-researched and represents one of the main causes of uncertainty in global GHG budgets. The paper is generally well structured, and the results mostly support the drawn conclusions. There are a few areas, however, that could do with a little revision and rewriting, and some of the conclusions based on the isotopic signature of different ecosystem C compartments might be a little speculative and could do with some rephrasing. Furthermore, some details on the experimental setup are missing and should be added to the materials and methods section. The statistics are sound but could be presented in a more attractive format. But if the authors address these concerns in an adequate manner, I am convinced that this paper can be a valuable contribution to Biogeosciences.

We thank the Referee for the positive review and constructive comments. We take their point that some conclusions based on the isotopic signatures are somewhat speculative and we will tone them down in the revised manuscript. We will also edit the materials and methods section so that the experimental set up is clearer to the readers. Furthermore, we will remake Figure 3 and include information from the statistical model. To avoid confusion, we will split up Table 1 into two parallel analyses (one for montane and one for lowland).

Specific comments:

Introduction p.1 L17: fungi are also considered to be microorganisms. Therefore it is enough to say microbial respiration, or alternatively, fungal and bacterial respiration.

Thank you for the specificity, this will be changed in the manuscript.

L19: the reference for global C flux via soil photosynthesis is a bit old, I suggest using the numbers from the latest IPCC report.

In this sentence, we present the global soil respiration rate from Bond-Lamperty and Thomson, 2010. To our knowledge, in the latest IPCC report, soil respiration rates were only presented together with the C loss via fires.

p. 2 L15-34: The authors highlight why it is crucial to understand soil respiration especially in ecosystems that are less well researched (i.e. tropical African rainforests). This paragraph is a bit lengthy because I think the reader of Biogeosciences is aware of that fact. Please shorten this paragraph, and instead add some information on <sup>13</sup>C partitioning throughout the C cascade of tropical rainforests, and what different δ<sup>13</sup>C values can mean, as this will guide the reader towards the research questions.

We agree that we can condense this paragraph and introduce the <sup>13</sup>C partitioning.

p. 3 L4-9: what were your hypotheses?

Since our objectives were to quantify annual soil CO<sub>2</sub> fluxes from forests of the Congo Basin and to assess differences between forest types, this was primarily a descriptive study. As such, we did not formulate specific hypotheses to test. Furthermore, it would not be correct scientific practice to formulate hypotheses after the fact. Hence, we refrained from formulating hypotheses now.

Material and methods: p. 3 L15, L19-20, and throughout the manuscript: please don't confuse the terms "average" and "mean". The (geometric) mean is a form of the average, in addition to the median and the modus. It should, therefore, be "mean annual rainfall" and "mean annual temperature". This should also be addressed throughout the results and discussion section (e.g. mean flux, etc).

This will be changed in the manuscript.

p. 4: the section on soil CO<sub>2</sub> flux measurements lacks some important details: how big were the study areas and plots? How many plots were installed per site? What was the vegetation composition (dominant tree species, presence or absence of dense understorey, basal area of trees, etc)? Did you use 3 flux chambers per site or per plot (i.e. more per site)? How were the chambers arranged in plots (e.g. distance from large trees, understorey vegetation, depressions/mounds, etc)?

We thank the reviewer for requesting more details regarding the flux measurements. We installed one plot in a mixed forest site in the montane forest. There we used seven chambers for the short-term campaigns and three chambers in the first year of long-term measurements. In the second year of the long-term measurements, we increased the number of chambers to five. In the lowland forests of Yoko and Yangambi, we installed two plots in each site, one in a mixed forest and one in a mono-dominant forest, where more than 60% of the basal area consists of the species *Gilbertiodendron dewevrei*. For the short-term campaigns, we used four chambers in the mixed forest and three chambers in the mono-dominant forest. We started the long-term campaign with four chambers in the mixed forest and two chambers in the mono-dominant forest and after a year, we proceeded with five chambers in the mixed forest and stopped sampling in the mono-dominant forest. The chambers were randomly placed between trees and we avoided hills and depressions. We will make the respective section of the manuscript clearer and add more information on the study areas (dominant tree species, understory presence, basal area of trees, etc.) and chamber arrangements.

One more note on the number of replicates for CO<sub>2</sub> flux measurements: This is not 100% clear from the authors' description, but if I understand correctly, only 3 flux chambers were installed per study site. This is critical because spatial heterogeneity of soil respiration has been described in numerous previous studies, and this could lead to under- or overestimation of soil flux estimates. However, there are a couple of points that the authors could use to address this shortcoming: first, they have measured soil CO<sub>2</sub> flux not only in one but in 3 lowland rainforests, and they could look at the difference between sites to describe spatial heterogeneity in the region. Second, if the flux chambers were always installed following a similar scheme, e.g. always at a fixed distance from trees, they would still be comparable even if not 100% representing absolute fluxes. Third, data on GHG fluxes from Africa are very scarce, and one of the reasons is the difficulty in getting research material into or out of the respective countries. I know from personal experience that it can be very difficult to buy or import even simple building material to construct flux chambers, and shipping of environmental samples can be complicated and often requires a lot of paperwork. I can imagine that the situation in DRC might have been similar. Therefore, for future studies on GHG fluxes in regions that are not easily accessible, I recommend the use of the gas-pooling technique by which gas samples from multiple (usually 3-6) chambers are put into the same GC vial, which can help to cover spatial

heterogeneity while at the same time reducing the total number of samples. Nevertheless, even if the number of replicates is low and this probably introduces some uncertainty, this information on the magnitude of fluxes and their dynamics is still highly valuable, and I therefore still recommend the study for publication in BG.

Thank you for this comment. As stated above, we used a minimum of three chambers per site. However, for the short-term campaigns, we used up to seven chambers per site. The results from these short-term campaigns showed relatively low variability between chambers at the same site (C.V. of 22% in the lowland forest and 16% in the montane forest) and thus we decided to reduce the number of chambers due to the limited number of evacuated vials for gas sampling (four were used per chamber per sampling for this study). Additionally, the reviewer correctly acknowledged that we measured CO<sub>2</sub> fluxes at three different lowland forest sites, which are separated by more than 100 km. The average fluxes of these three sites were also similar (inter-site C.V. of 25%) which gave us further confidence that even those periods where only three chambers were used are representative. We will add this information to the manuscript. As the reviewer points out already, it was not easy to get materials into the DRC. Therefore, we were only able to increase replication with additional visits to the sites, taking more material to the DRC. Ideally, this would have been done from the start but was not possible due to logistical constraints. Moreover, we appreciate the suggestion to use gas-pooling and will consider adopting this technique in the future.

p. 5 d13C measurements L24-25: wouldn't drawing 3 analytical samples of 20 ml each from the headspace of a 110 ml vial create an underpressure? How did the authors address this?

When withdrawing the samples from the 110 mL vials, a luer-stopcock between syringe and needle was used to avoid underpressure problems when removing the needle from the vial headspace during subsampling. That is, after withdrawal of 25 mL of sample, the luer-stopcock valve between needle and syringe was closed and the syringe was removed from the headspace. After, the plunger was pushed to 20 mL before opening the valve and injecting the subsample to 20 mL Labco vials. This procedure was repeated 3 times. The precision of three analytical replicates was excellent, with a maximum standard deviation of 0.25‰. We will add a more detailed description of the sampling to the manuscript.

L31: how many litter traps were installed per site, and how were they arranged? L33: how many soil samples were collected per site? What was their arrangement (e.g. distance to chambers, distance to trees, etc)?

Eight litter traps were installed per site and arranged in two rows of four. There was a distance of eight meters between traps. Soil samples were collected at three random positions at each site. We thank the reviewer for their attention to detail and will add this information to the manuscript.

Statistics L6: you assumed little year-to-year variability of your data, but did you actually check if the climatic conditions (rainfall, temperature, moisture) varied between years?

We compiled the flux, temperature and WFPS data in weekly bins for easier presentation of the seasonality of the fluxes. However, for statistical analysis (influence of soil temperature and soil moisture on soil CO<sub>2</sub> fluxes) we used the individual fluxes with the actual soil temperature and soil moisture conditions during each flux measurement. As a result, the weekly bins did not affect the results of the statistical analysis.

p. 6 Figure 2: there are two dips in WFPS in March and October in the lowland forest, where WFPS dropped rapidly from c. 30 to 20%, and then recovered within a week or so – do you have an explanation for this?

The two observed dips are located right within the peak rainy season, therefore, speedy recovery of soil moisture can be expected.

p. 7 Table 1: I know that the R output of lmer looks like this, but it's not very convenient for the reader to understand the results of the statistical analysis. For example, for d13C there is a significant effect of "Montane forest – stream CO<sub>2</sub>". This is ambiguous: does it mean that the d13C of stream-CO<sub>2</sub> is different in montane and lowland forests? Or that the d13C of stream-CO<sub>2</sub> is different from the d13C of the other compartments (soil CO<sub>2</sub>, litterfall, SOC) only in the montane but not the lowland forest? Please use a different way to present these results as they are critical. For example, as a start you could add letters/starts to Figure 2, presenting sign. differences between compartments via different letters, and differences between forests via stars (or something like that).

We agree with the reviewer that we can improve this presentation, to ensure unambiguous interpretation. The p-values (as well as R<sup>2</sup>s) in the case of linear mixed effect models are only estimations of p-values, and should be interpreted with caution either way, hence the interpretation of the table was mainly meant for the effect sizes. We suggest that we remake Figure 3 and include the model information in a new Figure 3. For the ease of interpretation, we will split both the figure and modeling up in two parallel analyses (one for lowland, one for montane). This will avoid confusion with the interpretation of too many interaction effects, but the effect sizes of both models will still allow the reader to interpret both inter and intra-forest type effects on δ<sup>13</sup>C. A table of this 'split-up' analysis (much like the current Table 1), could then go to supplementary materials. We thank the reviewer for making this clear, it is in our best interest that the readership of the paper can easily interpret the data we show.

L4-6: You state that stream-CO<sub>2</sub> was significantly depleted in the wet season in lowland forests but not montane forests. However, in Table 1 you state "montane forest – wet season – stream CO<sub>2</sub>" to be significant. Isn't this contradictory (or just another example of how Table 1 could be misinterpreted)?

In Table 1 "Montane forest – Wet season – Stream CO<sub>2</sub>" has a P-value of 0.42. However, we fully agree that the current table, including three factors and two interaction effects, creates confusion, and will change this for the next version of this manuscript.

Discussion p. 8 L5-7: Move this to the results section.

Thank you for the suggestion, this will be moved to the suggested section in the revised draft.

p. 10 L9-12: Careful, while it is true that with increasing dry season length soil CO<sub>2</sub> fluxes might decrease, but it is not clear how future more erratic rainfall patterns and the corresponding more extreme drying-rewetting events will affect respiration, and whether potential CO<sub>2</sub> pulses after rewetting compensate or outweigh reduced soil respiration.

We thank the reviewer for providing this qualification and will rewrite this statement to reflect a larger degree of uncertainty, for example, that there could also be CO<sub>2</sub> pulses that compensate for lower respiration as a result of these extreme drying-rewetting events.

L25-30: Good call! I agree that the correlations between soil CO<sub>2</sub> flux and temperature in tropical systems that show very little annual variation should be handled with care. In your case, they might

be significant simply because your sample size is large enough, but I would not over-interpret them. As you correctly state, moisture and C availability are likely the bigger players here.

We are glad that the reviewer agrees with our conservative interpretation.

p.11 L1-4: soil moisture not only controls O<sub>2</sub> diffusion but also the diffusion of C substrates to soil microorganisms. Therefore, the response of respiration to moisture is more often an effect of C limitation (at low moisture) than an effect of O<sub>2</sub> limitation (which really only becomes critical at very high moisture contents). Please add this to the discussion, and I recommend these papers on the mechanisms underlying this: Manzoni S, Moyano F, Kätterer T, Schimel J (2016) Modeling coupled enzymatic and solute transport controls on ecomposition in drying soils. *Soil Biol Biochem* 95:275–287. doi: 10.1016/j.soilbio.2016.01.006 Moyano FE, Manzoni S, Chenu C (2013) Responses of soil heterotrophic respiration to moisture availability: An exploration of processes and models. *Soil Biol Biochem* 59:72–85. doi: 10.1016/j.soilbio.2013.01.002 Moyano FE, Vasilyeva N, Bouckaert L, et al (2012) The moisture response of soil heterotrophic respiration: Interaction with soil properties. *Biogeosciences* 9:1173–1182. doi: 10.5194/bg-9-1173-2012

We thank the reviewer for this nuanced perspective and will integrate the point along with the mentioned references into the discussion.

L20: you mention photosynthesis, yet this was not measured and is therefore a bit speculative.

We agree with the reviewer that using the term “photosynthesis” here is a bit misleading in the sense that it indeed does sound this parameter had been measured. We suggest to re-write this section avoiding the term photosynthesis: “In this study, the link between C assimilation and soil CO<sub>2</sub> is evident through [...]”

p. 12 L4: which canopy processes other than photosynthesis could those be? Furthermore, how do you think that vegetation composition might affect δ<sup>13</sup>C, and could this explain differences between lowland and montane forests? Can different trees have different leaf δ<sup>13</sup>C signatures, which could be reflected throughout the C cascade?

These are two very good points raised by the reviewer. We were mainly thinking of stomatal conductance as the other important canopy process determining <sup>13</sup>C discrimination. Since there is an interplay between photosynthesis and stomatal conductance on <sup>13</sup>C discrimination, we lumped these two processes together (as canopy processes). We will specify in the text how we define canopy processes.

The effect of altitude on δ<sup>13</sup>C of canopy leaves is well known (Körner et al., 1988, Hultine & Marshall, 2000; Chen et al., 2015) and can be explained by a combination of factors and the two consistent patterns associated with increasing elevation are a decrease in atmospheric pressure and in temperature. The decrease in O<sub>2</sub> partial pressure and temperature supposedly promotes a decline in ci/ca and the direct implication of this decline is that δ<sup>13</sup>C values become less negative (Wang et al., 2017). We will further elaborate on this issue in the new version of the manuscript.

L6: what are the mechanisms underlying the enrichment of <sup>13</sup>C at lower temperatures?

In the subsequent sentence, we explain that temperature changes can result in shifts in microbial communities, which can impact fractionation during heterotrophic soil respiration (Andrews et al., 2000).

Conclusions This is mostly a repetition of the results. Please instead give the “message of the story” – what are the implications of the results you found? What are questions that remain open? And what have we learned?

We will rephrase the conclusion and add implications of our results and address remaining research questions.

L24: how were the sites different in vegetation composition? Please describe in the M&M section and also address in discussion

We will add this to the site description and discuss it accordingly.

L27: what does this indicate, that that there was no temperature dependency of soil respiration between sites?

As our results suggests that respiration in the lowland forests is substrate limited, we reason that the higher temperatures, compared to the montane forest, will not results in an increased soil CO<sub>2</sub> flux. We amend tis sentence and clarify this in the manuscript.

p. 13 L4: you conclude the paper with the statement that these forests might become C sources under a warming climate, yet you did not find a strong effect of temperature! Instead, you could state that changes of C balance might happen in response to more erratic rainfalls and weather extremes.

We will rephrase the final statement in our conclusion in a manner that it better fits our observations.

Appendix A: Method supplement L6-25: please use the past tense throughout this section.

The Method supplement will be modified to the past tense.

p. 16 Figure 3A: change x-axis labels of panel d to the format HH:MM (e.g. 10:00, 15:00,...) to make it clear that those are hours.

The figure will be adjusted accordingly.

Technical corrections: p. 5 L30: please correct "...during the wet season from October to May"

This will be changed.

p. 6 L9: please correct "values were found" (use past tense throughout the results section)

The results section will be modified to the past tense.

L8 and elsewhere: You very often use the term "respectively"; however, I'm not a big fan of it, for two reasons: first, sentences become very complex and sometimes hard to understand when using this term, and second, it forces the reader to jump back and forth between the end and the start of the sentence, which disrupts the flow of reading. Very often, you'll find that your sentence won't actually become any longer if instead of using "respectively", you describe the results one after the other, in this case, this could be "The mean [instead of "average", see my earlier comment] annual values we measured in this study in the Congo basin, which are 3.83  $\mu\text{mol m}^{-2} \text{s}^{-1}$  for the montane forest and 3.69  $\mu\text{mol m}^{-2} \text{s}^{-1}$  for the lowland forest, are within the range of reported values from

other tropical forests. " I propose that you revise the MS and try to reduce the use of "respectively". This will make the paper easier to follow.

We very much agree with this comment and will rephrase those sentences and try to make it easier to read.

L14: please rearrange "...and they were rather low compared to our flux rates"

The sentence will be rearranged.

p.9 L2: "...showed marked seasonality [comma] with a 34 % decrease during the dry season [comma] whereas.."

We will add the commas.

L4: please rephrase "however, the decrease they found was not as pronounced as..."

p. 10 L22: "statistically significant correlation"

L18: please rephrase "play a crucial role in controlling soil respiration"

p. 11: L4: please add a comma here, otherwise the phrase is misleading: "stress soil microbial communities, and autotrophic respiration"

L10: please rephrase as this is otherwise misleading "While soil respiration in lowland forests is most likely C-limited, respiration in montane forests seems to be more sensitive to environmental conditions and could represent a potentially large C source with climate change."

p. 12 L17: enrichment does not occur in the "location" but in the movement from one compartment to another. Please rephrase "the highest enrichment occurs in the last step from soil to stream-dissolved CO<sub>2</sub>".

L25: please rephrase: "However, in contrast to the lowland forest, the montane forest site exhibited strong seasonality of soil respiration, primarily driven by WFPS during the dry season."

We will rephrase these sentences and will add commas where needed.

## References

Andrews, J. A., Matamala, R., Westover, K. M., and Schlesinger, W. H.: Temperature effects on the diversity of soil heterotrophs and the  $\delta^{13}\text{C}$  of soil-respired CO<sub>2</sub>, *Soil Biology and Biochemistry*, 32, 699 – 706, 2000.

Chen, L., Flynn, D. F. B., Zhang, X., Gao, X., Lin, L., Luo, J., Zhao, C.: Divergent patterns of foliar  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  in *Quercus aquifolioides* with an altitudinal transect on the Tibetan Plateau: an integrated study based on multiple key leaf functional traits, *Journal of Plant Ecology*, 8, 303 – 312, 2015.

Hultine, K. R., Marshall, J. D.: Altitude trends in conifer leaf morphology and stable carbon isotope composition. *Oecologia*, 123, 32 – 40, 2000.

Körner, C., Farquhar, G. D., Roksandic, Z.: A global survey of carbon isotope discrimination in plants from high altitude. *Oecologia*, 74, 623 – 632, 1988.

Wang, M., Liu, G., Jin, T., Li, Z., Gong, L., Wang, H., Ye, X.: Age- related changes of leaf traits and stoichiometry in alpine shrub (*Rhododendron agglutinatum*) along altitudinal gradient. *Journal of Mountain Science*, 14, 106-118, 2017.