Biogeosciences Discussions

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Comment on bg-2022-199

Hannah Holland-Moritz (Referee)

Referee comment on "Recently fixed carbon fuels microbial activity several meters below the soil surface" by Andrea Scheibe et al., Biogeosciences Discuss., https://doi.org/10.5194/bg-2022-199-RC2, 2022

In this preprint the authors use carbon (C) isotopes (^{14}C and ^{13}C) to determine the source and age of respired soil carbon in three sites along a climatic gradient in the Coastal Cordillera of Chile. They find that respired CO2-C was of more recent origin than soil organic C and that C in deeper soils was older than surface C. They then use these results along with total DNA extracted from the soils to conclude that microbial decomposition is primarily of new carbon, rather than old in these soils. I found this manuscript to be an important contribution to our understanding of the origin and age of microbially-processed soil carbon and generally an interesting well-written piece. My chief concerns are to do with the authors' use of total DNA as equivalent to microbial DNA, and therefor a proxy for microbial activity (Line 137), in some of the authors statistical methods relating respired total ^{14}C to respired ^{14}C (Figure 2c), and the interpretations regarding the influence of primary productivity (Line 228, conclusions, and final line in abstract), which I have detailed below.

Specific comments below:

Line 49-50: It would be nice to include an explanation for why the arid site was dug less deeply than the other two, perhaps in the methods section 2.2.

Line 73: Do the authors have any estimation of the differences in isotopes that might be due to the differences in sampling year and how that compares to the ranges seen in their results? This is particularly important for the ^{13}C-isotope results. I could see those as being particularly sensitive to differences in temperature and moisture conditions between the two sampling years.

Line 114: For methodological transparency, the authors should include a brief explanation about how they determined the pre-incubation period for each environment.

Line 137: Rather than referring to this measurement as microbial DNA, the authors should refer to it as total DNA. It likely not only includes DNA from microbial sources (e.g. microbial eukaryotes, fungi, and prokaryotes), but also DNA from non-microbial sources such as plant roots or soil arthropods. Although proportionally the non-microbial DNA is likely to be low in comparison, it is likely to vary among soil types (based on the amount of vegetation and moisture of the soil) and depth. Therefore it likely that the soils not only contain to contain different proportions of microbial:non-microbial DNA in different soils but also at different depths. The authors therefore should be cautious in their interpretation of total DNA as a proxy for microbial activity and should adjust their discussion accordingly. One relatively easy experimental way around this, would be to use qPCR to quantify the abundance of 16S and ITS gene copies in each soil sample's DNA. These numbers would be a more accurate quantification of microbial abundance at least, even though not all organisms possessing those genes are likely to have been active during incubation.

Section 2.7 – not having much disciplinary expertise in this area, I defer to others who do in evaluation of the methods. However, I appreciate the explanation in the last paragraph about accurate estimation of ages, which is helpful for non-experts in ^{14}C dating such as myself. It is also helpful that the authors imply that the carbon they observe likely is derived from the last 1000 years, and would be further helpful for their non-expert audience to know where the estimate of 1000 years comes from. Furthermore in their discussion, the authors describe a similar study in permafrost that was able to estimate ages using ^{14}C, if there is any way to get at least a range of ages from this data to compare to that study, I would find it very useful in interpreting the results.

Figure 2c – The R^2 value of the arid linear model is quite low compared to the mediterranean and humid sites. I wonder if a simple linear model is even appropriate for this relationship as it seems that depth, along with other co-correlates are at play. In fact, for the humid site as well, the mean and variance appear to be related with higher mean values having less variance at more shallow sites which violates the assumption of equal variance of residuals which means that their estimated slopes may be incorrect. The authors should address this low fit in the text and potentially may find some helpful solutions in this guide: https://academic.macewan.ca/burok/Stat378/notes/remedies.pdf

Line 228, conclusions, and final line in abstract: I'm not sure the authors have made a very strong argument that processes of the soil are highly dependent on primary productivity aboveground. Although the results do indicate that newer carbon is being respired, an alternative explanation could be that the newer carbon is simply being re-cycled among the microbial community as the community turns over. The incubations are not directly measuring the influence of primary productivity since they are plant-free incubations and there aren't measurements for primary productivity at each site. I recommend a more conservative interpretation.

Section 4.2: The authors may also want to consider the process of priming in their interpretation of this observation (see for example Bernard et al. 2022 for a nice review on the subject - https://besjournals.onlinelibrary.wiley.com/doi/full/10.1111/1365-2435.14038).

Technical corrections:

Line 98 – Typo, I believe “weight” should be “weighed”

Line 118 – typo in the specification of volume of gas: there are extra spaces between the number and the units. Additionally the units of liters should be abbreviated with a capital “L” rather than lowercase “l”.