

## Answer to the comments of reviewer 1

In this paper, the authors present results from a unique study comparing soil carbon stock and  $^{14}\text{C}$  values to soil respiration fluxes and  $^{14}\text{C}$  values during soil incubation of soils from 3 sites with distinct climate to up to 6 meters depth. What is truly unique about this study is the depth to which the authors sampled and incubated soils. I also find the depth increments to be impressively fine, adding to the value of these data (especially for modeling). The authors found that even at several m depth, microbial respiration during incubations is fueled by relatively younger C. While this finding is not unique, the comparison of sites across a climate gradient and the depth to which this study sampled in the soil profile is quite unique.

The paper is reasonably well written, but could be improved with some editing and revisions to the figures. These are relatively minor issues. More concerning, however, is the lack of statistical analyses or descriptions to support the authors' interpretations and conclusions. This manuscript needs some description of the statistical analyses used to provide the reported errors (are they standard deviation or standard error? something else?) and at minimum some simple statistical tests to look at differences between the sites and the  $^{14}\text{C}$  of TOC vs respired  $\text{CO}_2$ -C. This could be simple: A t-test to test the statistical significance of the TOC vs  $\text{CO}_2$  could be done easily by calculated the difference and testing if it is different from 0 - this could be a paired test so the same site/depth are compared to one another.

We thank the reviewer very much for the positive and constructive review of our manuscript. We added the results of the statistical analyses to all figures, and we now explain the statistical analyses that we conducted in a newly added section (section 2.8) in Material and Methods. In addition, we improved the manuscript according to the detailed comments (see below).

More detailed comments are below:

The title is great - I love that the main finding is right there! Thanks!

L 19 "higher than of the soil" is missing a "that" or something similar We added "that".

L 23 "strong microbial decomposition" - what is meant by "strong"? Perhaps a different word is better. We replaced "strong" by "most".

L 24 "which is likely due to stabilization" - I do not follow this logic. Do we know that decomposition stops at depth because of stabilization? I do not know that that we know that for sure. Perhaps "possibly" instead of "likely" or "partly" or be more specific about what you mean exactly. We added "that leads to enrichment of  $^{13}\text{C}$ " in this sentence and replaced "likely" by "possibly", as suggested.

L 30 "topsoils" are not generally thought of as the top meter (often this is the surface or A horizon, maybe the top 10 cm or 20 cm or even 30 cm, but certainly not the top meter!) Consider just saying in the top meter here and elsewhere, "top meter" is not much longer than "topsoil". We replaced "topsoil" by "uppermost meter of soils" here, and checked the correct use of the term topsoil throughout the manuscript.

L 35 the following paragraph is confusing to me - do you mean in the field? Incubations? Can you be more specific? We added "in incubations" in line 36 and "measured in the field" in line 42.

L 43 you need references to back up the statement that the total CO<sub>2</sub> is largely composed of CO<sub>2</sub> respired by roots. And what do you mean by largely? Half? Can you be more specific? What did these studies find? We replaced “is” by “can be” and we added a reference.

L 44 This paragraph would be a good place to clarify that you did laboratory incubations. It is not clear that you used incubations until the methods on L 92. We added “in incubations” in line 52.

L 137 This looks like a total DNA extraction with no clean up to remove plant or animal DNA - is this true? If so you should not call it microbial DNA. If you did remove plant and animal DNA please explain this more clearly. I think you can still use total DNA as a proxy for microbial DNA and biomass but you should be clear that this is what you have. Yes, in order to clarify this we removed “microbial”.

L 149 use “flushed” or “scrubbed” instead of “rinsed” done as suggested.

L 152-153 I don't follow - samples were collected at multiple time points over 11 or 8 months and checked for CO<sub>2</sub> concentration? It sounded like it was collected for 14C as it was written but the results seem to have only one time point for each site. I don't think you need to provide so much detail here - you can just say CO<sub>2</sub> concentrations were monitored and you collected the samples for 14C when there was enough or when you cutoff the incubation (at 11 months?) Please clarify. There is also detailed description of the respiration rate sampling earlier, but those data are not presented - why is that? We removed the details and simply say that the CO<sub>2</sub> concentration was monitored. The respiration rate is presented in a figure in the Supplement.

L 167 what is the reported analytical uncertainty? were soils pretreated for carbonates? were there carbonates (especially at the arid site?) The soils did not contain carbonates (as clearly stated in line 86. We now added the following sentence to the description of the AMS measurement “The analytical uncertainty is 2 ‰”.

L 172 I know what you mean, but the phrasing is awkward. You could say “...model to the data, but there was not enough data to constrain such a model” that said you could make assumptions or do a 2 compartment model with the respiration data, if you wanted to. (I don't think you do though!) We changed the sentence as suggested.

L175 - there is no description of statistics for the results! How were trends and differences assessed? Are these differences statistically significant? What are the reported uncertainties? Standard deviation or error or something else? Where is the respiration rate data? The respiration data was and is in the Supplement. All figure present(ed) standard errors and this is/was clearly explained in the figure captions. We now added results of statistical tests to all figures (see our answer to the first point raised by the reviewer).

L 204 Why do you say that most C respired by microorganisms is likely directly derived by roots? Can you be more specific? You might consider referring at some point to Phillips et al 2013: <https://bg.copernicus.org/articles/10/7999/2013/> We added “This is further supported by Philips et al (2013) who measured <sup>14</sup>C-CO<sub>2</sub> in a temperate hardwood forest, and found that rhizodeposition was an important driver of microbial respiration”.

L 223 add citations (you could use Phillips et al 2013 but there are likely others) We added the suggested citation.

L 227 What "interaction" do you refer to? Can you be more specific? This sentence is very awkwardly worded, please revise. [We revised the sentence.](#)

L 233 This paragraph is difficult to follow - is it about decomposition or about  $^{13}\text{C}$ ? It seems to go back and forth but not clearly explain how the two are connected. [We improved several sentences in this paragraph.](#)

L 237 What do you mean by "this"? Stabilization would not cause enrichment in  $^{13}\text{C}$ . [We replaced "this" by "organic matter decomposition is likely restricted to the topsoil".](#)

L 240 I do not follow how this is connected to  $^{13}\text{C}$  or the rest of the paragraph, it feels disconnected from the rest. [We shortened and changed this sentence.](#)

L 244-6 it would only be similar if C was cycling the same way, it seems like C cycles more slowly at the arid site and you would expect to see a smaller Suess effect there, which you do see. I would just cut this last sentence and reword the first part of the previous sentence to something like, "it is possible this is partly a result of the dilution of atmospheric  $^{13}\text{C}$ ....." [Done as suggested.](#)

L 253 Earlier in the paper suggests that  $^{13}\text{C}$  of roots were used for this interpretation too, but it looks like no? Are those data helpful here? Were there carbonates anywhere (it looks like maybe not, but it would be helpful if you could clarify this). [No, there were no carbonates \(we had stated this clearly in the description of the soils\).](#)

L 250 There is no discussion about the differences in vegetation causing some of the differences between sites. You should at least consider these differences since there is definitely a difference in vegetation cover across sites. [We added the following sentence "In addition, the higher decomposition of organic matter at greater depth at the humid site compared to the other sites might also be related to the trees at this site that seem to cause a larger root density in deeper soil horizons than the vegetation at the other sites, and thus likely stimulate microbial activity in the subsoil."](#)

L 254 can you add some more citations? Maybe <https://doi.org/10.1007/s10533-020-00725-z> [Done as suggested](#)

L 260 by "a different carbon pool" do you mean "a portion of the TOC pool"? [Yes. We revised this sentence.](#)

L 263 don't you think this is mostly because of water? these sites are so dry! [Yes, that is what we say \("water limitation"\).](#)

L 270 this comparison to permafrost seems very out of place here. You could develop this comparison by adding more context to the Intro and to the Discussion (with citations). If you think this important it could make the paper more interesting. [We refer to permafrost soils in the second paragraph of the Introduction. We now also added a comparison to permafrost soils in the Discussion.](#)

L 273 what do you mean by "tightly connected"? Can you be more specific? It seems like the deeper soil is connected to the shallow soil, but are the shallow soils really connected to the deeper soils? [We specify this in the same sentence \("that processes in the deep soil depend on carbon that recently entered the ecosystem through  \$\text{CO}\_2\$  fixation"\).](#)

L 277 where will the data be? the paper has been accepted as a discussion paper, where can the data be found? [We are currently in the process of publishing all data in the International Soil Radiocarbon Database, and the doi will be available very soon.](#)

Figure 2 for figure c, you could add a 1:1 line to show where the points would fall if respired C = TOC (since the axis are not the same) [Figure 2c already included a 1:1 line. For better recognition, we now added the 1:1 line in the legend.](#)

Figure 3 It doesn't seem like the axis need to go to -16 delta 13C. Can you plot these across a smaller range to make it easier to see the differences? Maybe -22 to -30 permil? [Done as suggested](#)

Figure 4 It is difficult to compare the panels arranged this way - maybe you could plott all 3 sites together with an inset or break in the axis to help show the differences? Something more like figure 5? [Done as suggested](#)