

Interactive comment on “Validation of a coupled $\delta^2\text{H}_{n\text{-alkane}} - \delta^{18}\text{O}_{\text{sugar}}$ paleohygrometer approach based on a climate chamber experiment” by Johannes Hepp et al.

Anonymous Referee #1

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Review of BG-2020-434 Validation of a coupled $\delta^2\text{H}_{n\text{-alkane}} - \delta^{18}\text{O}_{\text{sugar}}$ paleohygrometer approach based on a climate chamber experiment Johannes Hepp, Christoph Mayr, Kazimierz Rozanski, Imke Kathrin Schäfer, Mario Tuthorn, Bruno Glaser, Dieter Juchelka, Willibald Stichler, Roland Zech and Michael Zech.

Dear associate editor, Johannes Hepp and co-authors,

To start with my conclusion, I think this should be published, it is a beautiful dataset and I have the feeling that this dual isotope method for paleo reconstructions will become more and more important. I do have some issues with the manuscript as is. I had difficulties keeping track of the story a little, it is complex material, but I have the idea

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the authors have added to, at least, my confusion. I will try to explain what I mean. The measured data clearly shows the isotopic link (similarity) between source water, soil water and xylem water and leaf water moving away from the source water. This leaf water is assumed to be the source for biosynthesis, to some extend at least and on top of that there is biological isotope fractionation. The idea of using both isotopes is that hydrogen isotopes are highly affected by biological fractionation and oxygen isotopes not or less. One reason for that, I read would be O exchange between hemicellulose and leaf water. I had hoped oxygen would also be less dependent on leaf water evaporation, which it could be in paleorecords were bulk soils will be used and not only leaves. Which made me wonder how leaf derived sugars compare to stem or whole plant derived sugars? Especially for the paleo applications the authors mention as a reason for simplifying some of the formulas.

I have noticed that there is measured leaf water, but also sometimes leaf water isotope values reconstructed based on measured n-alkanes, especially in the comparisons with literature data. On top of that there is evaporative site water isotopic composition especially for H isotopes. At least that was my impression. There is also biosynthetic water that might be different, or water at the site of biosynthesis, again especially for hydrogen. Cytosol versus chloroplast. The role of NADPH is also mentioned. This helps explain the variability, but a lot falls under biosynthetic fractionation, so it explains variability in fractionation. It makes sense all these different versions of water and their impact, but the way the manuscript is organized no I find it extremely confusing. I would suggest a discussion along the main lines ending with some possible explanations for the “scatter”. One of the reasons I ask this is because apparently this is also going on for oxygen, leaf water isn't leaf water for oxygens isotopes either. There are isotopic and sucrose synthesis gradients that have to be taken into account. Again, it makes sense, but it is very confusing. And what does this mean for paleo applications? Where are the sugars coming from in paleosols? How is that related to this study? In general, what would be the effect of the oxygen exchange mentioned in the manuscript on paleo samples?

The authors mention that incubation 4 and 8 are the same, or the climate rooms are, the results are not? Any ideas what might be the reason? Some of the biosynthetic or synthesis water issues mentioned above? Or something different? Already the measured leaf water isotopes are different especially for Eucalyptus.

The authors calculate deuterium excess, I have been told that in highly evaporative systems also the slope of the meteoric waterline could be lower than 8. Could that be applicable to these kinds of systems as well?

Specific comments:

It would be great if the symbols in figure 1 could be a bit larger, the difference between squares and triangles is almost invisible.

Would it be possible to indicate the T and RH of the different chambers at least in the figure legend, but if possible, in the figure itself?

In figure 2 it would be nice if d was defined in the legend.

The per mil in the introduction is fine, the other two I would replace with %.

Line 226: n-alkane and sugar biomarkers Line 246: from the latter Line 247: ones or values, not once. Line 278: all three plant spp. Line 415: This could point.... I have a hard time connecting "this" to the previous sentences. I got lost here a little. Line 472: despite without? Line 472/473: bulk leaf is less enriched than the leaf water at the evaporative sites. Confusing. Bulk leaf water? The measured leaf water is bulk leaf water I assume? Why not use "leaf water" for this and call the other water, "water at the evaporative sites within the leaf" or something similar. Line 506: This is a very weird way to refer to a figure. You made the figure, it is not measured or observed and therefore a piece of evidence in your reasoning. The sentence starting at therefore till LEL's) can be deleted, I think. Line 550: Or the fact that bulk leaf water as measured does not capture the variability of water within the leaf and potentially important for biosynthesis. At least that has been mentioned several times already.

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I do think fractionation and variability therein is important, but the authors discussed these different leaf water to extensive to not mention here. Line 553: introduces

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