

Hydrol. Earth Syst. Sci. Discuss., referee comment RC2
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Comment on hess-2022-37

Anonymous Referee #2

Referee comment on "Technical note: Conservative storage of water vapour – practical in situ sampling of stable isotopes in tree stems" by Ruth-Kristina Magh et al., Hydrol. Earth Syst. Sci. Discuss., <https://doi.org/10.5194/hess-2022-37-RC2>, 2022

The work presented in HESSD by Magh and colleagues describes a new approach to sample xylem water for isotopic analysis in trees. In this technical note, the authors conducted a lab trial and a field trial to show the suitability of the proposed method. The general motivation of the study is to provide a cost-efficient approach to overcome the limitations of tree water sampling in field conditions and the potential bias of cryogenic extraction. This is important as this is a current point of debate in the literature (i.e. cryogenic limitation) and the proposed method is interesting.

While the study has a great design and is innovative, and the method provides reliable results in the lab, my main concern is that the proposed approach does not seem to provide reliable data from trees in the field (Fig. 7). This is a holdback, as this should be reliable if the goal is to obtain stem water and overcome limitations. It would be important to present more data in the field, i.e., more than one day, and possibly, in more than one tree (since the approach is "easy to use and cost-effective"), and different environmental conditions, i.e. wet vs dry.

The authors show a statistically significant difference between the *in situ*, defined in the paper as the "gold standard"/ "true value" and the VSVS data, even after the correction for storage is applied. This difference is evident even for the zero-day storage (Fig. 7) for d18O. While for d2H, there is no difference with some days (e.g. 1 and 7), it did not show a clear pattern. Thus, the reliability of the data is uncertain because the effect seemed random (e.g., why is 0-day results different from the *in situ* and not 1-day?). This is difficult to understand with a single trial in the field.

Suppose one can sample vapour with higher or lower water content (ppmV), as the authors even experienced (L166-168), and as we know the water content of the wood changes largely depending on the water status (e.g. high water content in the wood because of well-water conditions or low water content due to water stress), how would that interfere with VSVS and storage time, or even the proposed correction? Lab trials

usually show fractionation in vapour samples with low ppm. Since the authors are offering/reporting a new approach, it would be interesting to understand this before we, as a community, start to apply the technique broadly. Would it be beneficial to use larger vials during drier periods? Have the author's tested different volumes of vials for larger vapour storage?

Regarding the borehole, have the authors monitored the wounding effect? Similar to sap flow systems, where the wounding effect can influence sap flow rate measurements (e.g. Wiedemann et al., 2016; Peters et al., 2018), one would expect that we observe a similar effect when sampling for precise natural abundance where the boreholes are much larger than the small sap flow needle. Can the authors comment on this and discuss this limitation? For example, if multiple samples need to be collected from a forest in high-temporal resolution, for how long can one rely on the same borehole? Additionally, conifer species tend to produce resin near the wound, this could additionally result in spectral contamination. How could one define if spectral contamination is an issue in this system?

I think this study is missing a direct comparison with the cryogenic system. In the introduction, the authors refer to the potential bias of the method while this is also the "state-of-the-art extraction process" in this field, so how do the results compare with cryo? Or even the direct-equilibrium bag method?

Specific comments:

Title: The title mentions soil, but no tests or trials are done for soil in this work.

In line 36, the author refers to "matrix-bound" (assuming soil, as the previously mentioned soil matrix in the above paragraph). Still, in the last lines (L40-42) of this paragraph (L36-42), the authors use references that discuss cryogenic bias that relates to plants (Chen et al., 2020 and Allen and Kirchner, 2021). It would be helpful to be clearer and refer specifically to plant cryogenic bias in the text. Or, if the authors want to refer to cryogenic bias in both soil and plants, it would be helpful to mention it more clearly with appropriate references to both cases.

Lines 45- 46. What do the authors mean by altering their physiology?

Line 62: Add Kuhnhammer et al., 2021 along with Beyer et al., 2020 here as well.

Line 81: What was the volume of the standard water (in the larger vial)? Did the authors try different volumes of crimp neck sample vials? Why 50 ml was the selected volume?

Line 139: Give the scientific name to the two species.

Line 141: Give an estimate of the "several days" (e.g. ~ 5 days)

Line 143: It would be helpful to the reader if you already refer to the schematic figure here (Fig S1), and perhaps bring it to the main body of the paper (nice figure!).

L144: Maybe state clearly that one of the scots pine previously connected with the *in situ* system was monitored with the new system VSVS.

L149: Was the borehole flushed again or any treatment used after the five weeks before the change in the system? Did the authors detect any wounding effect in the borehole? Since this is a technical note, these details should be clearer so others can replicate the method.

L161: What was the air temperature in the field during the vapour sampling? How does change in air temperature affect the sampling (e.g. from wood to air)? Or if sampling in days with different air temperatures? It would be important to include the first answer here since this is a methods paper and the later ones in the discussion.

L175: It is not too clear why the in-situ $n = 2$. How was it determined? Please clarify this part or re-arrange the text to be more explicit.

L252-253: It would be helpful to show the atmospheric data in the supplementary information along with the samples.

L282: It would be helpful to see the data similarly to Figure 6. The raw along with the corrected for the field measurement.

L280-282: Didn't VSVS also fail to return the in-situ when compared with the other days (i.e. 1, 3, 7 and 14) and not only the "0-day"? Perhaps state it more directly.

L290-295. "3.3 Time and Cost Efforts" – This is not a result per se but part of the discussion. For a more comprehensive comparison, one should also state what type of cryogenic extraction the authors refer to, as the reference is not enough as this is important to the reader. The time efficiency one should also discuss field-set uptime (e.g.

how long does it take to set up the VSVS and in situ in the field?). This would be helpful to understand if short-term studies would still benefit from this approach.

L308-309: This is a bit of an overstatement for the field conditions. The VSVS proposed method results were not statistically similar to the defined "gold standard"/"true value" (*in situ*) for d18O and were somewhat similar for the d2H.

Fig. 6 Very minor comment: It would be nice to align the x-axis between the two plots.

Fig. 7 Very minor comment: Makes the asterisks larger; it is difficult to see them.

References:

Wiedemann, A., Marañón-jiménez, S., Rebmann, C., Herbst, M., & Cuntz, M. (2016). An empirical study of the wound effect on sap flux density measured with thermal dissipation probes. *Tree Physiology*, 36, 1471–1484. <https://doi.org/10.1093/treephys/tpw071>

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