Comment on hess-2022-178  
Anonymous Referee #2


General comments

In the manuscript ‘On uncertainties in plant water isotopic composition following extraction by cryogenic vacuum distillation’ by Haoyu Diao et al. the authors investigated the biases caused by CVD extraction of plant samples. In different experiments they tested the influence of H-exchange effects, absolute water amount, and evaporation and sublimation enrichments on the isotopic composition (δ18-O, δ2-H) of the extracted water. The manuscript is well structured and nicely written. The topic fits well to the scope of the journal and appears to be of interest for isotope hydrologists. However, the discussion could be more comprehensive to make things clearer. Most of my comments and remarks match those of Referee 1, who raised many important points already.

Specific comments

L. 128 ff: I suspect that drying at 60°C for 24h might eventually be too low and/ or too short. Was this procedure tested in advance? Did you check e.g. by weighing the samples after another 24h whether there was still some weight loss or not? Additionally, the ‘appropriate’ procedure could also be different for the twigs and the small segments. If your stem segments were not completely dry, the residual water will of course be strongly enriched, which will affect your results.

I’m not surprised that the differences in the isotope data are dependent on AWA rather than on RWA (e.g. L. 224, L.252). Possible reasons could be
very small water droplets are exposed to ambient air during collection (L. 180-181)
for small samples the sample volume might be too small relative to the size of the
extraction line i.e. the volume which is filled with water vapour. Can you estimate the
size (volume) of your system?

and should be considered in the discussion.

L. 322: You assume that the effect of small amounts is different, depending on the isotope
ratio? At least for isotope values in the range of your study it should not be in a
measurable range.

Figure 4: How can the huge differences in SD between D18-O and D2-H (Fig. 4a and c) be
explained?

**Technical corrections**

Throughout the manuscript: isotope fractionation should almost everywhere be singular
not plural, please check.

L. 71: 'liquid-vapour' instead of 'liquid-vapor'

L. 99: and L. 263: 'induced' instead of 'introduced'?

L. 310/ 311: ‘...allowed for investigating...’ instead of ‘...allowed us to investigate...’

L. 340: ...mean values ± 1 SD like in Fig. 3?

Figure S1: Typo in legend of Experiment 2 'Abosolute water amount'