

Interactive comment on “Effects of grass leaf anatomy, development and light/dark alternation on the triple oxygen isotope signature of leaf water and phytoliths: insights for a new proxy of continental atmospheric humidity” by Anne Alexandre et al.

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General comment: This paper examines the triple oxygen isotope systematics (expressed as $^{17}\text{O}_{\text{excess}}$) of leaf water and the respective phytoliths. The overall goal is to use phytolith oxygen isotopic composition as a humidity proxy. A general correlation between phytolith $^{17}\text{O}_{\text{excess}}$ and relative humidity had already been established in a previous publication. The growth chamber experiments presented is this new pa-

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per aim to better understand the full spectrum of processes that may affect phytolith $17\text{O}_{\text{excess}}$ (e.g. dark/light cycles, changing T and RH with dark/light cycles). These experiments and the respective data are important to develop an accurate relative humidity proxy. I think the modelling part can be improved, which would also make this paper more significant for a broader scientific community.

Specific comments: In their experiment 1, the authors investigate how leaf water composition changes between different growth stages and along the leaf. In the revised version the authors modeled the expected evaporation trend by extending the model of Farquhar and Gan (2003) for $17\text{O}_{\text{excess}}$. The model curves are concave in triple oxygen isotope space and clearly differ from the convex model curves recently published for a series of evaporitic ponds (see Surma et al. 2018). Both models are based on the Craig and Gordon model, so this discrepancy comes unexpected. I discussed this puzzling observation with my PhD student Claudia Voigt and she discovered, that calculating the parameter h' both from 17α 's and 18α 's (not just 18α 's) changes the curvature of the model to a convex form (i.e. identical to Surma et al. 2018). At first sight it appears as if the data fit better to such a revised model but possibly more processes need to be considered (e.g. mixing). If the authors manage to improve the plant water model, their paper would become significant for many other fields. Leaf water controls the triple oxygen isotopic composition of O_2 (produced from leaf water) and CO_2 (equilibrates with leaf water). It's well worth the effort to improve the model.

I found it especially interesting, that the measured phytolith data cannot be modeled from measured leaf water using published equilibrium fractionation factors. If the published equilibrium fractionation factors are correct, kinetic effects must be responsible for the observed offset. Or the measured leaf water is not representative of local leaf water from which the phytoliths form. To me, the changing λ values along the leaf seem to imply that the kinetic effects are not identical over the length of the blade. Is it possible to explain the data via contrasting fractionation factors during active (via enzymes) and/or passive (via evaporation) phytolith formation?

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Line 120: The isotopic composition of the vapor in air is identical to that of irrigation water. If these two reservoirs have any chance to exchange, vapor in air would be driven to lower values (i.e. the two reservoirs equilibrate). The agar agar prevents such an exchange to some degree. I assume that water vapor in the air is constantly exchanged to ensure constant RH and vapor isotopic composition. Is this correct? The vapor isotopic composition has a strong effect on the evaporation trajectories in triple oxygen isotope space, so if partial equilibration occurs that would be important to know.

Line 231: The main reason why the sheath comprises a lower oxygen isotopic composition than the blade is not the lower transpiration rate. As a thought experiment, assume that transpiration rates in the sheath and the blade are identical. The 'source water' of the sheath would be irrigation water with low d18O. But the source water to the blade would be evaporated water from the sheath with somewhat enriched d18O. In this simple model the blade could have a far lower transpiration rate than the sheath and still comprise higher d18O.

Model for the prediction of phytoliths in Figure 3: The empirical $\lambda_{\text{Phyto-LW}}$ as calculated from this data is used to predict the triple oxygen isotopic composition of the phytoliths, which is circular. If the published $\theta_{\text{silica-water}} = 0.524$ is used, the ^{17}O -excess values would be far off (as shown in Figure S1). Present the model using $\lambda = 0.524$ in Figure 1 (not only in Figure S1).

Section 7 (Conclusions): The first paragraph is confusing to me. Grass height and leaf height are mentioned here for the first time. Of course experiment 1 shows that leaf water composition changes along the leaf as predicted by the model, but this fractionation is not related to absolute height but to l/m . So a large (or high) leaf would carry the same bulk isotopic information as a short leaf (as stated at the end of paragraph 2). Also, I would not mix up the kinetic effects story with the RH story in the same paragraph.

Technical corrections: Line 57: Do not use the term distillation processes. In one of

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the references you cite (Steig et al. 2014) a distillation experiment is conducted where 17O excess changes over 90 per meg. Distillation processes can be governed both by equilibrium fractionation or kinetic fractionation depending on the set up. Line 124: provide 1 significant digit for the $\text{d}18\text{O}$ isotopic composition. Line 190: Please specify how the working O_2 gas was calibrated relative to SMOW or point to Alexandre et al. 2018. Provide the SMOW calibrated values for the internal quartz laboratory standard (Boulangé) and explain how that calibration was done. Ideally, provide a comparison of this laboratory internal standard to international standards with published $\text{D}17\text{O}$ on SMOW scale. This is crucial for recalculating the data in case of any revised calibration. Line 215: Do you mean Figure 2 (not 1)? Line 230: Table 1? Line 235: The good fit of the linear correlation seems impressive at first sight but the irrigation water is not included in that regression. If the linear regression (presented in the first manuscript version) is extrapolated, the irrigation water clearly falls below the line. I advise against using linear regressions because evaporation trends are best represented by curves. Line 241: These $\lambda_{\text{Phyto-LW}}$ are significantly lower than the expected equilibrium fractionation between silicates and water ($\theta_{\text{silica-water}} = 0.524$ for the $5\text{--}35^\circ\text{C}$ temperature range). The average reader won't remember that value so you may want to note that discrepancy here. Line 252: Remind the reader that RH and T changed with the light/dark alternations in this experiment. Line 287: The second ii) should be iii). Line 304: source not tsource. Table S3: If the leaf temperature is reduced from 20.4 to 18.4, the RH at the site of evaporation changes, so RH with respect to the leaf temperature (not air temperature) should be used as also recommended by Farquhar and Gan (2003). The Reference list is missing in the revised version. Caption of Fig. 3: $17\alpha = 18\alpha\lambda$ not $17\alpha = 17\alpha\lambda$ Clean up the legend of Fig. 3. (e.g. use $\lambda=0.52x$)

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