Importance:

The stable isotope of oxygen (d18O), is routinely used to characterize ecosystem processes from the cell to the globe (Werner et al., 2011, biogeosciences). The enzymatic activity of carbonic anhydrase (CA) enhances the exchange of the source water d18O signal with CO2 emitted from points of respiration. We can better constrain estimates of ecosystem metabolism by increasing our knowledge of CA activity under different environmental conditions. Sauze et al. set out to fill these gaps related to soil in our understanding through a series of innovative experiments and modeling. They quantified the isotopic exchange rate (k_{iso}) between H2O and CO2 in soils with a range of pH.

The authors devised small incubation units to which they could add water of known isotopic composition, known CO2 gas, and varying amounts of α-CA (28 and 80 mg L^{-1}). By controlling the environment, mainly through maintaining a near constant relative humidity, the authors were able to isolate the impact of soil pH on CA as determined by k_{iso}.

They found that the experimental results from three of the soils were congruent with their modeling. For the other three soils that did not conform to the modeling prediction, the authors infer that heterogeneity from the soil column, non steady-state conditions, or phosphate ions may have led to the aberrations.

The experiment was carried out well and the analyses were quite thorough, although a few statistical analyses with regards to differences in the isotopic composition of the source water and extracted water are needed. The manuscript needs a better description of the experiments performed and the hypotheses/questions asked of each steady-state iteration. I suggest a simple table or figure will suffice. This will let the reader keep better track of 1) some of the terminology used to describe different parameters (e.g., native, non-native), 2) sample size, and the results in general. In the introduction the real-world differences between α and β CA should be discussed. Furthermore, the use of α-CA can be defended more strongly upfront rather than toward the conclusion of the manuscript. I also am surprised by the differences in d18O between the soil extracted and equilibrated water values. This is important evidence in the ongoing debate of the two water world hypotheses (Kirchner, James W. "A double paradox in catchment hydrology and geochemistry." *Hydrological Processes* 17, no. 4 (2003): 871-874.; McDonnell, J. J. (2014), The two water worlds hypothesis: ecohydrological separation of water between streams and trees?. WIREs Water, 1: 323–329.; Sprenger, M., H. Leistert, K. Gimbel, and M. Weiler (2016), Illuminating hydrological processes at the soil-vegetation-atmosphere interface with water stable isotopes, Rev. Geophys., 54, 674–704.Vargas, A. I., Schaffer, B., Yuhong, L. and Sternberg, L. d. S. L. (2017), Testing plant use of mobile vs immobile soil water sources using stable isotope experiments. New Phytologist, 215: 582–594.).

Detailed comments:

Page 1
Line 12: delete "nonetheless"

Page 2
Line 3: Biosphere "absorbs" - unconventional way of describing flux and pools
Line 17: delete "advanced over recent years" given the literature list dating back to 1997 (and earlier?), the method is established.
Line 37- perhaps simple instead of crude.
Line 40- since you introduce these values can you report the accepted global photosynthesis rates?

Page3

Line 4: You introduce microbial communities as a possible explanation, but not how the microbes might alter CA activity. I assume it might have something to do with total biomass and functional characteristics of the communities.
Line 18: please provide a citation for this value.
Line 23- I think you mean soil solution here.

Perhaps specify in your hypotheses that you are primarily interested in the direct effects of pH and not the indirect effects anticipated from shifts in microbial diversity and function.

Page 4
Line 30: I suggest to introduce the "native" or control term here and please explain the situation to which you arrived at estimates of "un-catalyzed rates" (page 9 line 28).
Line 32: Why were these concentrations chosen?
Lines32-33: Please edit this sentence, especially "the measurement soils without".
Line 38: Introduce the water bath here and how its temperature was maintained.

Page 5
Line 3: The description of the two gases here is lacking. Is there not also CO2 in the compressed air-tank? It is not clear at this point why you have the two different gas sources. Please explain how you achieved different d18O compositions in the two gas sources.
Line 24: each is singular in this case: "Each line was measured", "only the last 40s of measurement was averaged"
Line 29- do you mean over the measurement time? i.e., the measurement period?

Page 6

Line 1: what do you mean precisely by "were propagated"?
Line 17. The equation in parentheses is difficult to decipher, perhaps separate it from the text.
Line 25: Can you better define piston velocity here? It looks like eqn. 5 is a formulation of Fick's diffusion implementing Henry's law. In this case, is the piston velocity expressing a minimal exchange of gas at the soil-water boundary during equilibrium? Tans (1998) also discusses piston velocity within this context.
Line 29: delete "so-called"

Page 7
Line 11: D_{iso} is not defined here
Line 28: add carbonyl sulfide along with OCS

Page 8

Line 10: refrigerator instead of "fridge"
Line 25: please edit" "to help vaporise the water under vacuum immediately upon injection", this does not read well.
Line 28: I assume this is a data filter and not a physical filter used in the analysis.
Line 29: "of the measurements"; please edit this whole sentence.

Page 9
Line 8: what is "near-common"?
Line8: perhaps, inform the reader before the calculation is described that it is desirable to verify the k_{iso} and d_{sw-eq} independently.
Line9: what is "sequence" referring to here?
Line 20: the difference is roughly 1 per mil, can you check that the difference is significant?
Line 28: why native? Is this the same as the control?
Line 29: do they mention "un-catalyzed" before?

Equation 12: let the reader know that $(k_{cat}/K_m)max$ is explained previously in equation 10.

Line 33: I suggest to separate Fig. 6a and 6b. Is the apparent peak and subsequent decline of K_{iso} explained in the results or the discussion?

Page 10
Line 19: This is a little confusing, it can be read as th ough your are using exogenous CA as a tool to predict the enhancement in soil CA activity, or it can be read as a general question asking if we can predict the enhancement in soil CA activity when additional CA is introduced.

Line 20: Since this is the first sentence of your discussion, perhaps you can set the reader up for what the topic of discussion is for this paragraph. The term native doesn't appear until page 9 line 28 and I don't think the term non-native is ever defined in the prior text. This exemplifies why a table or figure explaining the experiment will help the reader.

How is the fact that three of your soils did not conform to your model reconciled within fig 6?

Line11: this sentence is a little convoluted, it reads at first as if the soil pH is going to have a response when in fact this metric is intrinsic to the soil.
Line 22: deviations from non-steady state instead of non-steadiness.
Line 26: "was run" or simply "ran"
Line 28: please explain "native hydration". Is this potential water remaining within the soil from the field or elsewhere that has potentially mixed with the irrigation water?

The check for non-steady state was cursory and not all the assumptions were easily understood. While the exercise is interesting, it is not possible for the reader to determine how robust the results are. I think the authors need to decide how important this issue is to their results and either fully address the issue to the best extent possible or shorten the explanation and report that the non-steady state effect needs to be addressed by further experimentation and modeling.

Page 11

Conclusion- Can you bring out the larger relevance once more? How might the effect of pH influence our estimates of ecosystem carbon balance at different scales?

Figure 2. Please insert the step number within the top or bottom panel. Check the grammar within the caption, for example, I believe you want to say you measured the two calibration bottles in step 1.

Figure 3. I recommend that the use of a-CA in the experiments is clarified in the figure. Perhaps also how this figure is related to the experimental results.

Figure 4. It is probably worthwhile to report the results for all the soils or at least place them in the supplementary. Please reference where the K_{uncat} is presented in the text.

Figure 6. The caption does not reference a or b. In the text, I could not find a reference to a. Please explain what appears to be model uncertainty.