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Non-destructive measurement of carbonic anhydrase activity and the oxygen isotope composition of soil water

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Carbonic anhydrases are a group of metalloenzymes that catalyse the hydration of aqueous carbon dioxide (CO_2). The expression of carbonic anhydrase by bacteria, archaea and eukarya has been linked to a variety of important biological processes including pH regulation, substrate supply and biomineralisation. As oxygen isotopes are exchanged between CO_2 and water during hydration, the presence of carbonic anhydrase in plants and soil organisms also influences the oxygen isotope budget of atmospheric CO_2. Leaf and soil water pools have distinct oxygen isotope compositions, owing to differences in pool sizes and evaporation rates, which are imparted on CO_2during hydration. These differences in the isotopic signature of CO_2 interacting with leaves and soil can be used to partition the contribution of photosynthesis and soil respiration to net terrestrial CO_2 exchange. However, this relies on our knowledge of soil carbonic anhydrase activity and currently, the prevalence and function of these enzymes in soils is poorly understood.

Isotopic approaches used to estimate soil carbonic anhydrase activity typically involve the inversion of models describing the oxygen isotope composition of CO_2 fluxes to solve for the apparent, potentially catalysed, rate of oxygen exchange during hydration. This requires information about the composition of CO_2 in isotopic equilibrium with soil water obtained from destructive, depth-resolved soil water sampling. This can represent a significant challenge in data collection given the considerable potential for spatial and temporal variability in the isotopic composition of soil water and limited *a priori* information with respect to the appropriate sampling resolution and depth.

We investigated whether we could circumvent this requirement by constraining carbonic anhydrase activity and the composition of soil water in isotopic equilibrium with CO_2 by solving simultaneously the mass balance for two soil CO_2 steady states differing only in the oxygen isotope composition of ambient CO_2. This non-destructive approach was tested through laboratory incubations of air-dried soils that were re-wetted with water of known isotopic composition. Performance was assessed by comparing estimates of the soil water oxygen isotope composition derived from open chamber flux measurements with those measured in the irrigation water and soil water extracted following incubations. The influence of soil pH and bovine carbonic anhydrase additions on these estimates was also investigated.

Coherent values were found between the soil water composition estimates obtained from the dual steady state approach and those measured for irrigation waters. Estimates of carbonic anhydrase activity made using this approach also reflected well artificial increases to the concentration of carbonic anhydrase and indicated that this activity was sensitive to soil pH.