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Isotopically labelled water - A valuable tracer to track initiation and progress of bud dormancy in temperate trees

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Leaf-out of deciduous trees is regulated by a set of environmental factors such as cool temperatures during winter-dormancy (chilling), warm spring temperatures (forcing), and daylength (photoperiod), with complex interactions between these factors. Teasing apart these different factors in situ is challenging as no visible changes occurs during the dormancy phase. Manipulating these factors in climate chamber experiments may overcome this issue but may not reflect how they truly interact in natural conditions. Previous researches suggested that bud meristems are disconnected from the xylem flow during endodormancy and that the connection become progressively restored once exposed to a certain duration of chilling. Here we developed a new method using isotopically labelled water (D_2O) to quantify the amount of water that can reach buds during the whole dormancy till budburst for 5 different species (*Acer pseudoplatanus*, *Carpinus betulus*, *Fagus sylvatica*, *Quercus petraea*, *Tilia cordata*).

In detail, we harvested twig cuttings from leaf fall to budburst (~every two weeks, 12 times) of these species from two different sites (about 5°C of difference) and placed them into labelled water during 24 h at constant light and 20°C. Buds were then cut and water content extracted to quantify δD. Thus, tracing back the water flow into the buds by the amount of D_2O taken up. In parallel a subset of twigs was left in the room at 20°C to assess the time to budburst as a proxy for dormancy depth. Analyses of the data are ongoing and preliminary results show progressive increase of water uptake after induction of winter dormancy until budburst as chilling duration increased. Further, we also found distinct differences between species whereas *Carpinus betulus* showed the highest and *Tilia cordata* the lowest label uptake during winter dormancy. Furthermore, individuals growing at higher elevation took up less label indicating a stronger dormancy at lower winter temperatures. In summary, we think that our method seems a valuable tool to track quantitative changes in dormancy depth of temperate species especially, in combination with investigations on the molecular level such as sugars or hormones during winter-dormancy.