# Identification of Source-water Oxygen isotopes in trees Toolkit (ISO-Tool) for deciphering historical water use by forest trees

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**Key Points:**

- A toolkit for determining the oxygen isotopic signature of historical source water to trees is presented.
- The parameterization of ISO-Tool is subdivided based on data availability and output resolution.
- The tool is designed to retrospectively assess water source usage by plants at annual and sub-annual timescales.

**ABSTRACT**

Hydrological regimes are being perturbed under climate change due to the regional expression of the water cycle across the globe, leading to alterations in the spatial and temporal distribution of water near the Earth's surface. Water is a critical resource for forest ecosystems and hydrological limitations on vegetative health are particularly complex. To anticipate how subsurface water availability may evolve in the future and affect the dynamics of plant water-source usage, as well as the health and functioning of vegetation in various biomes, we need a robust, quantitative framework for linking water availability to past plant water-use, which is constrained by historical data. Here, we outline the Identification of Source-water Oxygen isotopes in trees Toolkit (ISO-Tool), designed to retrospectively

investigate the dynamics of tree-water uptake. ISO-Tool utilizes tree-ring isotopes ($\delta^{18}O$) combined with a biomechanistic fractionation model to predict the $\delta^{18}O$ of water utilized during any period of growth. Through comparisons with measured $\delta^{18}O$ in local water sources, climatic and hydrological variables, ISO-Tool can reconstruct and inform on past ecohydrological interactions. We provide an overview of the modeling components and data requirements necessary to constrain the retrodictions of source-water $\delta^{18}O$. We demonstrate the utility and efficacy of ISO-Tool for three riparian field sites characterized by differences in climatic, geomorphic and hydrologic complexity. However, ISO-Tool can be applied to a range of vegetated environments where distinct isotopic endmembers exist. We present a set of tool groups, which can be applied adaptively, ensuring that scientific progress in understanding retrospective ecohydrology can be made, even under varying degrees of data availability.

# 1 INTRODUCTION

## 1.1 Background

Forests worldwide are becoming increasingly vulnerable to variations in water availability as hydrological regimes respond to climate change (Allen et al., 2010, 2015; Choat et al., 2012; Clark et al., 2011; Hartmann et al., 2013). Yet, despite the fundamental role water plays in the health, productivity and distribution of tree species (Currie & Paquin, 1987; Hsiao, 1973; Schulze et al., 1987; Stephenson, 1990), there remain considerable uncertainties in how terrestrial water availability to forests will evolve under future climate (Allen & Ingram, 2002; Donat et al., 2016; IPCC, 2014; Sippel et al., 2017; Trenberth et al., 2014). Such shortcomings result from the incomplete characterization of moisture sources over a range of timescales, which is further complicated by the contribution of several potential sources to tree-available water. These sources include infiltrated precipitation in the vadose zone and shallow groundwater in the phreatic zone, where the latter can be derived from hyporheic streamflow contributions to shallow water tables (Busch et al., 1992; Evans et al., 2018; Singer et al., 2014; White & Smith, 2013). Therefore, for any interval of time, the particular water source used by a tree is a function of specific tree rooting depths as well as by the time-varying availability of root-zone water, which varies in response to climatic trends and fluctuations (Dawson & Pate, 1996; Snyder & Williams, 2000). The details surrounding these dynamic ecohydrological relationships are poorly understood, a knowledge gap which restricts our ability to anticipate how forests will respond to alterations in hydrological regimes that may affect one or both soil hydrological reservoirs (vadose and phreatic), particularly in regions where water is expected to become a limiting resource to tree growth (Bréda et al., 2006; García-Ruiz et al., 2011; Lindner et al., 2010).

To address this shortcoming, we take a retrospective approach and identify two fundamental questions: 1) What water source was used by a tree during a particular period of growth? 2) Does this water source utilization correspond to the dynamics of root-zone water availability? If these questions can be answered satisfactorily for a particular site, we believe this information on plant-water relationships could dramatically improve studies of ecohydrology, paleoclimate, and land surface dynamics within forest ecosystems.

Severe moisture deficits can reduce tree growth and biomass production (Berner et al., 2017; Charru et al., 2010; Sarris et al., 2011; Silva et al., 2010), increase the susceptibility to pathogen infestations and eventually lead to mortality from hydraulic failure and/or carbon starvation (Allen et al., 2010; Breshears et al., 2005; McDowell et al., 2011; Schlesinger et al., 2016). Through differential responses to moisture stress, individual tree and species' mortality rates can lead to altered stand demographics and composition, and thus ecosystem functioning (Clark et al., 2016; Hansen et al., 2001; Milad et al., 2011), whilst moisture limitations in the rooting zone may also determine whether particular tree species can get recruited and established at riparian sites (Mahoney & Rood, 1998; Singer & Dunne, 2004). These are key concerns, given the significant role trees play globally in carbon and water cycling (Bonan et al., 2008; Ciais et al., 2005; Ellison et al., 2017; Jasechko et al., 2013; Kurz et al., 2008; Settele et al., 2014) and in the provision of ecological services (Anderegg et al., 2013). Critically, forest vulnerability to drought conditions is not restricted to environments typically considered as moisture limited. Amazonian forest mortality was linked to drought conditions experienced in 2005 (Phillips et al., 2009) and 2010 (Lewis et al., 2011), with the later drought spanning 3.2 million $km^2$ (25 % greater than that of 2005). Furthermore, Chen et al., (2017) attributed trembling aspen die back in western Canadian boreal forests to insufficient water availability.

Retrospective insights into patterns of tree source water use (at seasonal and annual resolution) would be particularly useful for riparian zones, where phreatophytic forest species are highly dependent on the hydrological connectivity between the river and floodplain phreatic aquifer (Singer et al., 2014). Phreatophytes are sensitive to changes in shallow alluvial groundwater availability which can manifest as declines in healthy forest stands, reduced ecological services (Amoros & Bornette, 2002; Steiger et al., 2005; Stromberg et al., 2007) and shifts in the successional states of forest communities (e.g. Stromberg et al., 1996; Shafroth et al., 2000). Information about the timing and origin of tree source waters would also allow for accurate determinations of the ecohydrological impacts arising from river flow regulation practices (i.e. minimum flow requirements), river bed-level changes (e.g. gravel extraction / downstream

of dam construction) and to identify critical thresholds for groundwater abstraction. Such information would promote more sustainable water management strategies in forests where water resources are also under increasing population pressures (Jackson et al., 2001).

We believe that a simple, yet effective, methodology for determining historical sources of water used by trees, would be highly valuable in improving scientific enquiry in isotopic ecohydrologic investigations. Such an approach would need to be well defined, easily accessible and adaptable.

In this paper, we build on existing research by combining and enhancing a set of techniques to identify historical water-sources ($\delta^{18}O_{sw}$) used by trees, based on information from tree-ring cellulose ($\delta^{18}O_{cell}$), plant physiology, hydroclimate and a biomechanistic fractionation model (Roden et al., 2000; Barbour et al., 2004). This is presented in the form of a methodological 'toolkit' called the Identification of Source water Oxygen isotope Toolkit (ISO-Tool), for which we identify and explain the necessary data requirements and provide examples of its application. We distill these requirements and data sources and present them in the form of a tiered 'Tool Group' scheme, to provide a means to straightforwardly reconstruct the historical source water signatures of trees at annual or sub-annual scales. Our goal here is not to necessary develop a method for highly accurate characterization of past water usage by plants, but rather to enable researchers to better ascertain likely water sources to plants for particular periods of time, to distinguish between plant water sources within sites for different species, and to identify how plant water sources may vary along hydroclimatic gradients.

### 1.2 Isotope echoydrology

Stable isotope ratios of oxygen and hydrogen in water ($\delta^{18}O$ and $\delta^{2}H$) have been used to trace the sources of waters used by plants through the comparison of the isotopic signatures of potential endmember sources (e.g. soil, groundwater, surface waters) with those of waters extracted from the xylem (e.g. Bertrand et al., 2014; Dawson & Ehleringer, 1991; Dawson & Pate, 1996; Horton et al., 2003; Jackson et al., 1999; Schulze et al., 1996; White et al., 1985). This is based on broad evidence that no isotopic fractionation occurs between the soil water pool and the plant during root uptake (Allison et al., 1983; White et al., 1985), although $\delta^{2}H$ fractionation has been reported in some halophytes and woody xerophytes (Ellsworth & Williams, 2007; Lin & Sternberg, 1993). This means that the water taken up by a tree retains isotopic information specific to its origin and history within the hydrological cycle at the point of uptake. Characterizing plant source waters via isotope analysis is particularly powerful if potential endmember water sources, e.g. precipitation (*P)* or groundwater (*GW*) are isotopically distinct

and locally defined. This a fundamental principle when considering the use of $\delta^{18}O/\delta^{2}H$ for ecohydrological studies (Barbeta et al., 2018).

If combined measurements are made of $\delta^{18}O$ and $\delta^{2}H$ in xylem and potential endmember water sources, information relating to the evaporative history of the water (relative to local precipitation input) can be obtained, although such information is not necessary for identifying plant water sources (Sprenger et al., 2016).

Soil composition, texture and water status (e.g. Chen et al., 2017; Oshun et al., 2016; Vargas et al., 2017) have all been proposed causes of the fractionation of oxygen and hydrogen isotopes in plant-available water, prior to uptake, highlighting the need for careful consideration of these factors during investigations, as well as further research (for an overview, see Barbeta et al., 2018). Nevertheless, if such results are interpreted carefully, then stable isotope analyses remain a powerful method for understanding plant-water interactions. Herein, only $\delta^{18}O$ ecohydrology is discussed, since this isotope ratio functions as a better conservative tracer of water in ecosystems than $\delta^{2}H$ due to its higher mass and binding energy.

Commonly, the distinct hydrological reservoirs i.e. vadose (unsaturated) and phreatic (saturated) zones are characterized by differences in their water $\delta^{18}O$ content and water in these zones is preferentially used by different species (Singer et al., 2014). However, where potential source waters are isotopically similar, disentangling the water source use can be challenging (Drake & Franks, 2003).

Water in the vadose zone typically inherits an isotopic signature reflective of precipitation ($\delta^{18}O_{ppt}$) inputs (Robertson & Gazis, 2006) which varies seasonally as a function of temperature, air mass origin and history (Dansgaard, 1964; Gat, 1996). For temperate environments, this is exemplified by depleted and enriched $\delta^{18}O$ of precipitation in the winter and summer, respectively. The isotopic composition of soil moisture can also represent a time-varying mixture of isotopically separate precipitation events, which may also include the effects of near-surface evaporative enrichment, leading to the development of isotopic profiles in soil water with depth (Allison et al., 1983; Gazis & Feng, 2004; Hsieh et al., 1998; Sprenger et al., 2016).

In contrast, the $\delta^{18}O$ signature of phreatic water typically has low temporal variability, and it is often isotopically depleted in relation to vadose zone moisture, especially if *GW* is sourced from regional snowmelt runoff (colder percolated precipitation) (Gat, 1996) and/or is mixed with streamflow (*Q*) (Dawson & Ehleringer, 1991). However, there may be situations where isotopically distinct

groundwaters contribute to a rooting zone. Sargeant and Singer (2016) suggested that isotopically dissimilar waters, derived from different climatic regimes, may interact within a shallow alluvial aquifer receiving both a regionally derived *GW* and water derived from locally infiltrated precipitation. A pragmatic approach would be to isotopically characterize the potential water sources within a field site, ensuring that isotopic separation of plant available waters is possible (Barbareta et al., 2018).

### 1.3 Source water availability

In environments where both vadose and shallow phreatic waters are available, tree species can employ different physiological strategies for obtaining water. Deeply rooted phreatophytic species prefer phreatic water sources derived from shallow aquifers (Busch et al., 1992) and capillary rise, yet may also exhibit opportunistic behavior, switching to shallow soil moisture (or using combination of both) under periods of water deficit (Singer et al. 2013, 2014; Snyder & Williams, 2000; Sun et al., 2016). Shallowly rooted species are unable to access deeper phreatic water, so they rely on vadose moisture which is sensitive to balance between precipitation inputs and near-surface evaporation, although it is possible that capillary rise during a period of elevated water table can supply phreatic water to the unsaturated zone (Sánchez-Pérez et al., 2008). Additionally, plant roots can redistribute water across a soil-water potential gradient (hydraulic redistribution), both vertically and laterally (Brooks et al., 2002; Caldwell et al., 1998; Richards & Caldwell, 1987), allowing water from different depths and distances to be utilized by neighboring plants (Dawson, 1993), and complicating source-water identification. Studies which provide detailed ecohydrological information are typically based on comparisons between the contemporaneous measurements of tree xylem waters with those of local water sources (e.g. Plamboeck et al., 1999; Sánchez-Pérez et al., 2008). Whilst direct analysis of xylem $\delta^{18}O$ circumvents the leaf fractionation and exchange mechanisms which mask the source water $\delta^{18}O$ information stored in $\delta^{18}O_{cell}$ (McCarroll & Loader, 2004), studies of this sort are limited by the temporal domain of field work (typically 2-3 years), restricting the development of broader conclusions about ecohydrological interactions (Pettit & Froend, 2018). Long-term, seasonal reconstructions of tree source-waters $\delta^{18}O$ could yield potentially powerful new information about water availability to trees, and thus hydrological processes, whilst also providing historical context for real-time investigations.

### 1.4 Tree-ring oxygen isotopes

In order to gain retrospective insights into plant-water interactions over longer timescales, tree-ring isotopes represent an under-utilized excellent library of accurately dated isotopic information on tree water usage. The oxygen isotope ratio ($^{18}O/^{16}O$) contained within tree-ring cellulose ($\delta^{18}O_{cell}$) can be used to characterize the isotopic signature of the source water(s) ($\delta^{18}O_{sw}$) used during the time of ring formation, thereby providing an extended record of ecohydrological processes (McCarroll & Loader, 2004). Tree-ring $\delta^{18}O$ fractionation theory is largely well established (McCarroll & Loader, 2004), although some uncertainties remain regarding the timing and transfer of photosynthates from source to sink tissues, which is relevant for finer scale studies (Gessler et al., 2009; Offerman et al., 2011; Ogée et al., 2009). Whilst there is the possibility for the remobilization of stored carbohydrates, formed at an early period, this more evident in $\delta^{13}C$ measurements than those of $\delta^{18}O$ in tree rings (Hill et al., 1995)

The $\delta^{18}O_{cell}$ records an integrated isotopic signal of three components: (a) the signatures of trees' source water taken up from the rooting zone; (b) leaf-water enrichment as a function of evaporation (Dongmann et al., 1974; Flanagan et al., 1991); and (c) the biochemical fractionation that occurs during photosynthesis (Sternberg et al., 1986; Yakir & DeNiro, 1990). A final mechanism during cellulose synthesis allows for a proportion of oxygen atoms in sucrose to exchange with $\delta^{18}O_{sw}$, damping the leaf-level fractionation effects (Farquhar et al., 1998; Hill et al., 1995; Roden et al., 2000). While there is a consistent enrichment (27 ± 4 ‰) of photosynthate $\delta^{18}O$, compared with leaf water ($\delta^{18}O_{lw}$), leaf water enrichment is much more variable responding to atmospheric conditions and leaf level gas exchange relating to species' physiology (McCarroll & Loader, 2004; Roden et al., 2000).

Studies utilizing tree-ring $\delta^{18}O_{cell}$ have been able to provide insights into historical water availability to trees, although use of the 'raw' $\delta^{18}O_{cell}$ to infer water source assumes that climatic variables and tree-physiological responses are uniform across a study site or between sites, and the relative differences measured in $\delta^{18}O_{cell}$ are therefore solely a function of $\delta^{18}O_{sw}$ differences. For example, Marshall and Monserud (2006) suggested that differences observed in tree-ring $\delta^{18}O_{cell}$ of three co-located species over eight decades could be attributed to shifts in tree water source use, whilst Singer et al. (2013) suggested that fluctuations in floodplain hydrology strongly controlled the source water availability and utilization for two tree species with contrasting rooting profiles. However, in order to provide direct comparisons with endmember water sources, it is necessary to determine the $\delta^{18}O_{sw}$ from the $\delta^{18}O_{cell}$ value. Using detailed measurements of the primary controlling variables of $\delta^{18}O$ fractionation in leaves ($E$ - transpiration, $g_s$ - stomatal conductance, $\delta^{18}O_{wv}$ – atmospheric water vapor $\delta^{18}O$, $RH$ – relative humidity and $T$ – air temperature), it is possible to model the relationship between xylem water ($\delta^{18}O_{xyl}$) and $\delta^{18}O_{cell}$ ,to varying degrees, based on the quality of input data (Barbour et al.,

2004; Roden et al., 2000). This relationship can be used to inversely hindcast (model) the $\delta^{18}O$ of source waters used for the formation of each tree ring ($\delta^{18}O_{msw}$) (Bose et al., 2016; Sargeant & Singer, 2016; Singer et al., 2014). Earlier work by Anderson et al., (2002) demonstrated the potential for reconstructing historical $\delta^{18}O_{ppt}$ from $\delta^{18}O_{cell}$ by calibrating the model (incorporating climate variables and tree growth) against records of $\delta^{18}O_{ppt}$.

The model calculations of $\delta^{18}O_{msw}$ can then be verified against field measurements of the evolving isotopic composition of contributing water sources (*P*, *GW*). Using this approach, direct comparisons between $\delta^{18}O_{msw}$ with local endmember water source $\delta^{18}O$ have suggested that co-occurring tree species with distinct rooting depths (e.g. *Fraxinus* spp. and *Populus* spp.) often access and utilize different mixes of water sources at annual and sub-annual timescales, as a function of fluctuating hydro-climate and relative depths to local GW (Sargeant & Singer, 2016; Singer et al., 2014).

Identifying tree source water(s) requires information on how the potential local endmembers vary in their $\delta^{18}O$ signature on both annual and sub-annual timescales, which is particularly complex in situations where there may exist more than one potential water source available for tree growth (Sargeant & Singer, 2016; Singer et al., 2014). Furthermore, fluctuations in source water mixtures modify the isotopic signature of root-available water, thus challenging interpretations of water sources consistently available to trees (Busch et al., 1992; Dawson & Ehleringer, 1991; Sánchez-Pérez et al., 2008; Snyder & Williams, 2000). Despite these challenges, the identification of $\delta^{18}O_{sw}$ is critically important, especially considering that changes in soil water content may outpace the ability for new root growth to track such shifts (Plamboeck et al., 1999).

We demonstrate our methodology for three Mediterranean, riparian forest sites, at both annual and sub-annual resolutions, highlighting the ability of the ISO-Tool to discern valuable ecohydrological information at different timescales. Whilst our examples are based on riparian forests, ISO-Tool is also suitable for other forested environments where a distinct isotopic separation exists between potential water sources used for tree growth over different time periods. Our aim is to provide a new suite of methods to enable a greater understanding of forest ecohydrology in response to climatic fluctuations and trends in subsurface hydrology.

## 2 METHODS

### 2.1 Identification of Source water Oxygen isotope ratios in trees Toolkit (ISO-Toolkit)

The core principle of this research is that $\delta^{18}O_{cell}$ can be deconvolved to the $\delta^{18}O_{sw}$ taken up from the rooting zone during a particular growing season. The method utilizes an iterative modeling approach to model the source water $\delta^{18}O_{sw}$ signature ($\delta^{18}O_{msw}$), which is then directly comparable to the $\delta^{18}O$ of potential endmember water sources. In this section, we explore the calculations of $\delta^{18}O_{msw}$ and how the user defined input variables are used. We then analyze the sensitivity of the mechanistic model to these different input variables. Subsequently, we describe how key variables can be constrained in order to improve the accuracy of the $\delta^{18}O_{msw}$ calculations and we outline the data and methods that can be used to help interpret the model output. Combined, these form a methodological toolkit, which can be utilized for determining historical plant water use in a variety of forest ecosystems.

An overview of ISO-Tool is shown schematically in Figure 1. ISO-Tool, an open-source code available in two forms, enables the back-calculation of $\delta^{18}O$ of water sources used by plants based on measured $\delta^{18}O_{cell}$, climate and plant physiological variables. This code is based on the Barbour et al. (2004) model for plant $\delta^{18}O$ fractionation but modified to invert the problem.

The required user inputs of the toolkit are predefined in the following sections, but the manner in which these are constrained is governed by specific Tool Group selections. These data are added to the input file and must comprise of an average value with an estimate of standard deviation (SD) . From these, the model generates normal distributions that are repeatedly sampled by Monte-Carlo simulation, which produces an error margin of ±2 SD. From the mean input values, the model also calculates the mean $\delta^{18}O_{msw}$ value. The mean and standard deviation error bounds are produced as a plot and an associated output file is generated. The modeling script is editable to enable a 'bespoke' analysis through the modification of individual parameters (detailed below), but which are not currently included as requirements within the input file. Presently, the model uses an optimization routine to predict the $\delta^{18}O$ value of source water, $\delta^{18}O_{msw}$. The model results ($\delta^{18}O_{msw}$) can then be interpreted by the user from a selection of possible techniques contained within the Tool Groups necessary to characterize source water availability to trees.

The $\delta^{18}O_{msw}$ modeling script is provided as a Matlab code (doi: 10.5281/zenodo/1161221) with the input and output files in .csv (.txt) format. The model is provided at: https://github.com/blissville71/InverseBarbourModel. Here, the user will find downloadable files of the model as a Matlab code (includes Monte-Carlo simulations) and a Microsoft Excel version (excludes Monte-Carlo simulations), plus an example input file and accompanying instructions /overview for the model's rationale and use.

## 2.2 Modeling source water $\delta^{18}O$ ($\delta^{18}O_{msw}$)

The $\delta^{18}O_{msw}$ is predicted from a known $\delta^{18}O_{cell}$ by accounting for the tree-level fractionations and exchange processes which alter the $\delta^{18}O_{sw}$ signature until cellulose formation. In Figure 2a-d, the key components of the model are shown, as well as how required user input data is used in these equations, with an accompanying conceptualization in Figure 2e.

The extent to which transpiration enriches $\delta^{18}O_{sw}$ at the sites of evaporation ($\Delta_e$) in the leaf, is fundamental in determining the recorded variability in $\delta^{18}O_{cell}$ values. The $\Delta_e$ model is based on the work by Craig and Gordon (1965) and later modified for the leaf environment (Dongmann et al., 1974; Farquhar & Lloyd, 1993; Flannagan et al., 1991):

$$\Delta_e = (1 + \varepsilon^*)\left[(1 + \varepsilon_k)\left(1 - \frac{e_a}{e_i}\right) + \frac{e_a}{e_i}\,(1 + \Delta_v)\right] - 1 \quad (1)$$

which can be approximated as:

$$\Delta_e \approx \varepsilon^* + \varepsilon_k + (\Delta_v - \varepsilon_k)\frac{e_a}{e_i} \quad (2)$$

The evaporative enrichment of source water within the leaf is given in Figure 2a & e, and assumes an isotopic steady state, which is appropriate for our purposes since we rely on photosynthetically derived substrates ($\delta^{18}O_{cell}$), reflecting daytime leaf conditions during which an isotopic steady state is approached (Cernusak et al., 2016). Within the model, $T_l$ is assigned as $T$+1 (°C), as a simplifying assumption. An energy balance equation could be incorporated to compute a realistic $T_l$, but it would require additional data inputs (e.g. wind speed, leaf width, photosynthetically active radiation) that are typically unavailable for historical reconstruction (Barbour et al., 2000; Lorrey et al., 2016).

Although boundary layer conductance is an important component of $\varepsilon_k$, difficulties in determining time-varying values (Brenner & Jarvis, 1995) may necessitate the use of a fixed value of 1 mol $m^2s^{-1}$ (Barbour et al., 2004). In reality, boundary layer conductance varies as a function of leaf shape, size and thickness, stomatal density, wind velocities and energy balance (Buhay et al., 1996; Jarvis & McNaughton, 1986; Schuepp, 1993; Stokes et al., 2006). Constraining all of these parameters requires a much more detailed investigation beyond the scope of this study. However, both leaf temperature

(offset from air temperature) and boundary layer conductance values can be assigned at the user's discretion.

The magnitude of leaf water evaporative enrichment has been shown to be over-predicted by the equation in Figure 2a (Cernusak et al., 2016 and references therein). Farquhar and Lloyd, (1993) proposed that such discrepancies resulted from a Péclet effect, whereby the advection of unenriched source water via transpiration is opposed by the diffusion of enriched water to and from the sites of evaporation, respectively, operating over a given distance divided by the molar density of water and diffusivity of $H_2{}^{18}O$ in water (Figure 2b, c & e). As such, under high transpiration rates, the level of leaf water enrichment declines. Values of *L* (mm) can be calculated as a function of *E* based on the study of Song et al., (2013). They developed a regression between *L* and *E* for several different tree species ($L = 2.36 \times 10^{-5} E^{-1.20}$, $R^2 = 0.813$), where the regression function is expressed in meters (m). We included this relationship within our code so *L* is computed only based on *E* (obviating direct measurements of *L*). We also utilized the R-squared value to provide an error around these estimates. Barbour et al. (2004) showed that the inclusion of the Péclet effect improved their model estimates of the isotopic signature in cellulose (relative to source water), accounting for 89 % of the variability in their study.

During photosynthesis, sucrose molecules are enriched +27±4 ‰ above leaf water $\delta^{18}O$ as the result of carbonyl-bound oxygen atom exchange between triose phosphates and the medium water (Sternberg et al., 1986; Yakir & DeNiro, 1990). It is this sucrose which is utilized to form cellulose, a process by which a proportion of intermediary molecules are able to exchange with unenriched xylem water ($\delta^{18}O_{sw}$) (Farquhar et al., 1998; Hill et al., 1995) (Figure 2d & e). The fraction of oxygen atoms undergoing this exchange is controlled by the proportion of unenriched source water within the cell during cellulose synthesis. The fraction of exchanged oxygen atoms is reported as 0.42 for many tree species (Roden et al., 2000), whilst the source water content is assumed to be at unity for large, mature trees where the distance between leaf and sink tissue is considerable (Barbour et al., 2002). The overall effect of this process is to dampen evaporative enrichment signature transferred to the cellulose molecule at the leaf level.

### 2.3 Sensitivity analyses

We conducted a series of sensitivity analyses on the calculation of $\delta^{18}O_{msw}$ in response to variations in *T*, *RH*, $\delta^{18}O_{wv}$, as well as to paired values of $g_s$ and *E*. We began by varying each of these input variables independently and then advanced the analysis through a covariance of two variables

(Figure 3). During each sensitivity test, variables not under analysis were kept constant as indicated in Figure 3 'Default values'. Leaf gas exchange parameters used in all model runs are those from Singer et al., (2014) for *Fraxinus excelsior*.

### 2.3.1 $\delta^{18}O_{cell}$, $\delta^{18}O_{wv}$ and RH

The $\delta^{18}O_{msw}$ predictions respond primarily to changes in *RH*, $\delta^{18}O_{wv}$ and $\delta^{18}O_{cell}$ when each respective input is varied independently (Figure 3a). It should be noted that these results are only representative of the interactions occurring between changing variable and the default values of the remaining inputs indicated in Figure 3a.

We included $\delta^{18}O_{cell}$ to illustrate the positive response we expect on $\delta^{18}O_{msw}$ if all other variables are constant, thus, $\Delta_e$ is static so with increasing $\delta^{18}O_{cell}$ there must be a corresponding enrichment in $\delta^{18}O_{msw}$ (+1.5‰/‰). For $\delta^{18}O_{wv}$, a negative relationship exists with $\delta^{18}O_{msw}$ (-0.6‰/‰) (Figure 3a). The dependence of $\Delta_e$ on $\delta^{18}O_{wv}$ can be shown using equation (2). If $e_a/e_i = 1$ (saturated conditions), then $\Delta_e = \varepsilon^* + \Delta_v$ (Cernusak et al., 2016), because enrichment in $\delta^{18}O_{wv}$ causes depletion in $\delta^{18}O_{msw}$ in compensation. For *RH* there is a strong, positive, curvilinear relationship with $\delta^{18}O_{msw}$, so as *RH* increases, $\Delta_e$ decreases. This arises because $\delta^{18}O_{msw}$ must undergo enrichment to maintain a given $\delta^{18}O_{cell}$.

### 2.3.2 T, $g_s$ and E

When *T* (i.e. $T_l$) was varied from 2-40 °C, $\delta^{18}O_{msw}$ was shown to enrich by 3.3‰, a relatively weak effect on $\Delta_e$ over this large temperature range, compared to that of $\delta^{18}O_{wv}$ and *RH*. As $T_l$ increases, the equilibrium fractionation factor, $\varepsilon^*$, is reduced causing a decline in $\Delta_e$. This produces an enrichment of $\delta^{18}O_{msw}$ to maintain the static input $\delta^{18}O_{cell}$ value.

In order to test the sensitivity of $g_s$ (mol $m^{-2}s^{-1}$) and *E* (mmol $m^{-2}s^{-1}$), whilst also accounting for the physiological coupling between the two processes, these two variables were paired based on a simple expression derived from Fick's law of diffusion where $\Delta w$ is the leaf-to-air vapour pressure gradient (Dawson, 1996; Pearcy et al., 1989):

$$E = g_s \Delta w \quad (3)$$

Calculations were conducted using the Roden model of evaporative enrichment (Roden & Ehleringer, 2000) in an Excel file format available at ftp://ecophys.biology.utah.edu/tree_ring/ (Barbour

et al., 2004). Input values of $g_s$ ranged from 0.05 – 1.00 mol $m^{-2}s^{-1}$ at 101.3 kPa to generate paired estimates of $E$ with a subsequent range of 0.40 – 4.20 mmol $m^{-2}s^{-1}$. We recommend caution in interpreting the results on the $g_s/E$ pairing sensitivity test because we have forced the dependence between the two. In reality, these variables might be expected to behave distinctly with variations in climate, subsurface water availability and between species. The effect of $g_s$ and $E$ in the model is shown in Figure 3a. It indicates a negative relationship between $g_s$ / $E$ and $\delta^{18}O_{msw}$ leaf water. The level of leaf water enrichment declines (at a constant $e_a$) with increasing $g_s$ due to a reduction in leaf temperature and intercellular vapour pressure caused by increasing $E$ (Barbour et al., 2007).

### *2.3.3 Model sensitivity to covarying parameters*

We covaried input parameters to identify those which most strongly affect predictions (retrodictions) of $\delta^{18}O_{msw}$ (Figure 3b-g). The $\delta^{18}O_{cell}$ was kept constant at 30 ‰. The $\delta^{18}O_{cell}$ represents the starting point for the model calculations, and all other variables interact independently of it. The variance of air $T$ was shown to have very little interaction with other variables, as shifts in $\delta^{18}O_{msw}$ are predominantly controlled $RH$, $\delta^{18}O_{wv}$ and $g_s/E$ (Figure 3b, d & g, respectively). $RH$ had the greatest interaction with $\delta^{18}O_{wv}$ (Figure 3c) and $g_s/E$ (Figure 3f), producing a range in $\delta^{18}O_{msw}$ of 29.1 ‰ and 21.5 %, respectively for the range of plotted values. The interaction between $\delta^{18}O_{wv}$ and $g_s/E$ (Figure 3e) induced a 10.7 ‰ change in $\delta^{18}O_{msw}$ over the range values for each respective variable. Since the range in co-variables presented are unlikely to represent field conditions, we varied the input values ($T$, $RH$, $\delta^{18}O_{wv}$) using the range of variance observed for a study site in SE France for the growing seasons (May-Sept) of 2000-2010. The results are shown as grey shading in Figure 3b-d and indicate that under the growing season climatic conditions, $RH$ and $\delta^{18}O_{wv}$ remain the primary interacting controls of $\delta^{18}O_{msw}$, producing variations up to 6.1 ‰ over their respective ranges (Figure 3c). The effects were smaller for $T$-$RH$ and $T$-$\delta^{18}O_{wv}$, which produced $\delta^{18}O_{msw}$ ranges of 4.5 ‰ and 3.3 ‰, respectively (Figure 3b and d).

The sensitivity analyses allow for straightforward identification of how the input variables to the biomechanistic model affect the predictions of $\delta^{18}O_{msw}$. By constraining the relevant variables and increasing the accuracy of the input data, the reliability of model output obviously also increases, and thus the characterization of the source water utilized by a tree during a particular time period is improved. It is evident from the sensitivity analyses that $\delta^{18}O_{wv}$, $RH$ and $g_s/E$ require the greatest constraints since their variability can introduce significant shifts in back-calculated values of $\delta^{18}O_{msw}$.

Through sensitivity analyses, we identified the required environmental ($T$, $RH$, $\delta^{18}O_{wv}$) and physiological ($g_s$, $E$) input variables responsible for modulating leaf level $\delta^{18}O$ enrichment. By utilizing the model in inverse mode, for a known $\delta^{18}O_{cell}$, and driving it with time-dependent input variables, it is possible to calculate $\delta^{18}O_{msw}$ taken up by the tree.

## 2.4 Tool Groups – constraining input variables and interpretative techniques

In this section we outline how the necessary model input variables can be obtained and summarize the interpretative techniques required to explain the resulting $\delta^{18}O_{msw}$ values within a hydrological context. We begin by explaining the sampling and extraction procedures of tree-ring cellulose at annual and sub-annual resolutions, the essential input of the $\delta^{18}O_{msw}$ model. Following this, we present three different tiers of data or 'Tool Groups' within ISO-Tool (Figure 4), which highlight the various approaches that can be drawn on to obtain values of $T$, $RH$, $\delta^{18}O_{wv}$ , $g_s/E$ required for model calculations and the various ways in which these results can be interpreted. The calculated $\delta^{18}O_{msw}$ values are most useful if they can be compared to the potential water source endmembers which, in turn, require knowledge of the hydrological processes which control the availability of a particular water source to the tree's rooting zone.

The idea is that Tool Group A represents the optimal suite of techniques and its data inputs should be used whenever and wherever possible. However, when data limitations prevent the use of Tool Group A for particular variable, the second Tool Group (B) should be employed, where possible…and so on. Thus, in a real application, due to universal constraints on data, a researcher is likely to draw relevant data from all three tool groups. In the worst-case scenario, one could use data entirely from Tool Group C and still make progress in water source characterization from tree ring cellulose. The rationale for this classification is to highlight the potential to undertake research into historical ecohydrology even when the most desirable information is unavailable. Of course, the research question also governs which tool group is utilized. A critical point is that prior to conducting isotopic source water investigations, the location of study must exhibit isotopically distinct, potential endmember water sources (isotopic separation), which can be confirmed through exploratory $\delta^{18}O$ analyses or from wider literature/ database searches. We encourage researchers to make additions and refinements to the ISO-Tool.

For an overview of the techniques listed in Figure 4, please see the supporting information. A more detailed discussion of the methodologies for Tool Group A can be found in supporting text S1 (Barbour & Farquhar, 2000; Beyer et al., 2016; Böttcher et al., 2014; Busch et al., 1992; Cocozza et al., 2016; Cuny et al., 2015; David et al., 2013; Dawson et al., 2002; Delattre et al., 2015; Gröning et al., 2012; Hao et al., 2013; IAEA/GNIP precipitation sampling guide V2.02, 2014; Jarvis, 1995; Lambs et al., 2002; Meinzer et al., 1999; Parnell et al., 2008, 2010; Pearcy et al., 1989; Phillips & Gregg 2003; Robock et al., 2000; Rossi et al., 2006; Rundel & Jarrell,1989; S.U. et al., 2014; Scott et al., 2004; Snyder & Williams, 2000; Song et al., 2013; Soudant et al., 2016; Sprenger et al., 2016; Stokes, 2004; Volkmann & Weiler, 2014; West et al., 2006; White & Smith, 2013; Zencich et al., 2002; Zweifel et al., 2001), Tool Group B in supporting text S2 (Berkelhammer & Stott, 2009: Bowen & Revenaugh, 2003; Bowen & Wilkinson, 2002; Bowen et al., 2005; Bowen, 2017; Delatrre et al., 2015; Entekhabi et al., 2010; GNIP, IAEA/WMO, 2017; Granier et al., 1999; GSOD – 'Global Surface Summary of the Day', 2018; Hsieh et al., 1998; Jonard et al., 2011; Jones, 1998; McNaughton & Jarvis, 1991; Oren et al., 1999; Richardson et al., 2007; Schwartz et al., 2002; Sprenger et al., 2016; Srivastava, 2017; Vicente-Serrano et al., 2010; West et al., 2006; Yale University, 2017) and for Tool Group C, in supporting text S3 (Bowen et al., 2005; Bowen, 2017; Celle-Jeanton et al., 2001; Delattre et al., 2015; Horita & Wesolowski, 1994; GNIP/GNIR, IAEA/WMO, 2017; Jacob & Sonntag, 1991; Majoube, 1971; NOAA – 'National Oceanic Atmospheric Administration', 2018; Poyatos et al., 2016; Sargeant & Singer, 2016; Singer et al., 2014; Suni et al., 2003; Wen et al., 2010). This is not designed to be an exhaustive review of all possible techniques for the interpretation of $\delta^{18}O_{msw}$, but to broadly identify the tools which could be utilized in source-water characterization.

### *2.4.1 Tree-ring $\delta^{18}O_{cell}$*

The tree(s) chosen for isotopic analysis generally reflect particular research questions, but they would ideally be visually healthy individual specimens with distinguishable, annual growth rings in order to determine the associated climatic variables needed for modeling. Otherwise, it is challenging to constrain the timing of water use.

After tree selection, tree-rings can be collected the form of tree-cores, obtained by an increment borer (e.g. at least 5 mm diameter) at breast height. Extracted tree-cores are 'bladed' to expose a uniform surface to maximize ring visibility. Cores should not be mounted with adhesive to prevent contamination of wood material and ethanol should be used to clean equipment. The tree-rings

corresponding to the years of interest can be identified through visual cross-dating or standard dendrochronological methods whereby ring widths are measured under a microscope using a tree-ring measurement program such as 'Measure J2X Tree-Ring Measuring Programme'. Validation of cross-dating can be achieved using a cross-correlation software such as COFECHA (Holmes, 1983). Alternatively, the cores may be scanned combined and ring-widths measured digitally with the coordinate measuring and cross dating software programs CooRecoder and CDendro, respectively (Larsson, 2014). Individual rings of interest are dissected with a scalpel and homogenized in preparation α-cellulose extraction.

Sub-annual isotopic analysis of tree rings has been shown to contain additional information that is masked by annual (homogenized) whole-ring analyses and which may be important for understanding the seasonal dynamics of water availability (e.g. Roden et al., 2009; Sargeant & Singer, 2016). Tree-cores can be collected and processed as above but with each whole ring divided into sub-annual segments using a scalpel or microtome (e.g. Gärtner et al., 2014; Helle & Schleser, 2004). The number of divisions is determined by the researcher and we recommend conducting trials on surplus rings to ensure that each slice yields enough material cellulose mass for isotopic analysis.

Tree-ring α-cellulose is the preferred material for isotopic analysis since it retains the $\delta^{18}O$ information from the time of formation without undergoing any subsequent isotopic exchange (Wright, 2008). In comparison, other components of whole wood material (e.g. hemicellulose, lignin, lipids and waxes) can exchange oxygen and may also represent different periods of synthesis (Battipaglia et al., 2008; Gray & Thompson, 1977). The modified Brendel (MBrendel) method (Gaudinski et al., 2005), allows for batch processing with minimal laboratory equipment. McCarroll and Loader (2004) suggest that for softwoods, the high quantity of resins and extractions can be removed by a Soxhlet apparatus and a toluene-ethanol solution.

The α-cellulose can be analyzed for $^{18}O/^{16}O$ using an online, continuous flow system of a TC/EA (Temperature Conversion Elemental Analyzer) connected to an Isotope Ratio Mass Spectrometer (IRMS). Results are reported in per mil (‰) deviation from Vienna Standard Mean Ocean Water (VSMOW $\delta^{18}O = 2.0052 \times 10^{-3}$ ‰) (equation [3], where $R_s$ is the isotopic ratio of the sample and $R_{std}$ is that of the standard). This follows correction to the IAEA-CH3 (cellulose [Hunsinger et al., 2010]) standard, co-run with an internal lab standard and the effects of drift and linearity are accounted for to obtain $\delta^{18}O_{cell}$:

$$\delta^{18}O_{cell}\ (‰) = \left(\frac{R_s - R_{std}}{R_{std}}\right) \cdot 10^3 \quad (3)$$

480

481 **3 ISO-TOOL APPLICATIONS**

482 In this section, we present three example applications of ISO-Tool to demonstrate its
483 broad utility. Our examples illustrate water source characterizations drawing on components from each
484 Tool Group for constraining model inputs and the interpretation of results (Figure 4). The sites are all
485 located along the Rhône River, SE France (Table S1), and although we illustrate how ISO-Tool usage in
486 riparian environments, we emphasize that it can be employed in any site where isotopically distinct
487 water sources are present and where tree rings record annual growth. The $\delta^{18}O_{cell}$ results from which
488 $\delta^{18}O_{msw}$ were calculated are in Figure S1a-c. We refer the reader to Figure 4 for each of the following
489 case studies.

490 Statistical analyses were conducted using Minitab (version 17) and are reported relative to the
491 95% significance level. All statistical tests were conducted following the determination of sample set
492 normality with the Anderson-Darling (A-D) test and analysis and equal variance using the Levene test.
493 For the comparison of 2-sample means we used the T-test (*T*) or Mann-Whitney (Wilcoxon rank) test
494 (*U*), whilst for >2 datasets an ANOVA (*F*-test) and post-hoc test (Tukey-Kramer/Games-Howell) was used
495 to determine statistically similar groups.

496

497 **3.1 Example 1: Minimum flow restoration, Pierre-Bénite (PB)**

498 Riparian forests are often the focus of restoration schemes which aim to improve the level of
499 hydrological connectivity through the raising of minimum discharge levels below dams to benefit aquatic
500 ecology as well as riparian forests, but this has never been shown convincingly. This example is based on
501 data from the Pierre-Bénite (PB) site where water managers increased the minimum dry-season river
502 flow from 10 m/s to 100 m/s in the year 2000 to benefit aquatic ecology. This produced a mean water
503 table rise within the floodplain of ~0.5 m (Amoros et al., 2005; Singer et al., 2014). In Singer et al., (2014),
504 analysis of annual tree-ring $\delta^{18}O_{cell}$ records from two cohorts of *Populus nigra* situated at relatively 'high'
505 and 'low' floodplain elevations suggested that the flow restoration enabled phreatic water to become
506 available to *Populus* rooted at low floodplain elevations. Since $\delta^{18}O_{cell}$ cannot be directly compared with
507 local water source $\delta^{18}O$ signatures, we applied ISO-Tool to evaluate the conclusions of Singer et al.,
508 (2014).

Whole ring (annually resolved) $\delta^{18}O_{cell}$ was used in this example (Figure S1a), with climate records of *T* and *RH*, corresponding to the observed growing season of May-September, obtained from a local climate station (meteofrance.com, 2017). The $\delta^{18}O_{wv}$ values were calculated using an equilibrium fraction factor (Majoube, 1971) using records of *T* and GNIP (IAEA/WMO, 2017)-OIPC (Online Isotopes in Precipitation Calculator) (Bowen, 2017; Bowen et al., 2005) estimates of $\delta^{18}O_{ppt}$, assuming that $\delta^{18}O_{wv}$ was in isotopic equilibrium with $\delta^{18}O_{ppt}$. Monthly estimates of $\delta^{18}O_{ppt}$ were obtained from the records of two equidistant GNIP (IAEA/WMO, 2017) monitoring stations (Figure S2a-b), averaged and correlated to monthly $\delta^{18}O$ values for the site, obtained from the 'Online Isotopes in Precipitation Calculator' (Bowen, 2017; Bowen et al., 2005) (Figure S2c). Literature-derived values of $g_s/E$, were used. In situ measurements of $\delta^{18}O_{RW}$ were unavailable, thus necessitating the use of $\delta^{18}O_{RW}$ from 34 km downstream. Seasonal $\delta^{18}O_{ppt}$ for each year were computed by weighting the monthly $\delta^{18}O_{ppt}$ values by the corresponding *P* totals recorded at the climate station. Piezometric measurements of water table elevation were available for the study period (Figure S3).

In Figure 5a, the $\delta^{18}O_{msw}$ time series of each cohort is shown. Although there is a separation in cohort water use from 2000 onwards, with the 'high' cohort utilizing more enriched $\delta^{18}O_{msw}$ than the 'low' cohort (+0.9 ‰), it is not statistically significant ($T_{18}$ = 1.68, $p$ = 0.11). Neither the 'high' or 'low' poplar trees displayed a shift in $\delta^{18}O_{msw}$ following the flow restoration, drawing on an isotopically similar water source during both periods ('high': $T_{19}$ = -1.20, $p$ = 0.244; 'low': $T_{19}$ = -0.65, $p$ = 0.542). The implementation of a minimum river flow was seemingly insufficient to provide sustained phreatic water access to either cohort of trees. Instead, both cohorts seemingly relied on different mixtures of seasonal precipitation. It is possible that the water table elevation did make some portion of phreatic water available to *Populus* at low elevations, e.g. by capillary rise (Sánchez-Pérez et al., 2008), but its signature is masked by mixing with precipitation-sourced vadose moisture (Figure 5a). The lack of clear evidence of a phreatic water signature in $\delta^{18}O_{msw}$ of either cohort after the flow restoration may be due to a combination of insufficient river flow to raise the water table relative to the fixed rooting architecture for mature trees of this species. It is possible that these *Populus* trees previously developed extensive vadose zone roots at the expense of deeper, vertical roots during the period when phreatic water was unavailable (i.e. pre-2000). This dimorphic rooting characteristic is advantageous in environments where phreatic and vadose zone moisture are seasonally variable (Singer et al., 2014).

### 3.2 Example 2: Sub-annual water source use, Donzère-Mondragon (DM)

Seasonal variability in water sources may exert important controls on growing season water availability and corresponding tree health, which could be masked by whole tree-ring isotopic analysis (Sargeant & Singer, 2016). At the Donzère-Mondragon (DM) site we employed ISO-Tool to determine the sub-annual progression of $\delta^{18}O_{msw}$ of two co-located (<5 m apart), streamside individuals of *Fraxinus excelsior* (roots restricted to the vadose zone by a coarse gravel layer) and *Populus nigra* (roots capable of phreatic zone access). The two-species approach can be used to investigate the seasonal dynamics of water partitioning within different hydrological reservoirs.

We utilized the $\delta^{18}O_{cell}$ from Sargeant and Singer (2016) (Figure S1b), but employ a longer growing season (MJJAS vs. MJJA) and the use of update to date GNIP (IAEA/WMO, 2017) records and the latest version of the OIPC (Figure S4). Sub-annual patterns and inter-species differences in $\delta^{18}O_{msw}$ are evident wherein *Fraxinus* $\delta^{18}O_{msw}$ is enriched by +2.4 ‰ compared to *Populus* ($U$ = 17211, $p$ <0.001) (Figure 5b) but a $\delta^{18}O_{msw}$ convergence for both species from 2007-2010 ($T_{58}$ = 0.59, $p$ = 0.557).

Prior to 2007, *Fraxinus*' $\delta^{18}O_{msw}$ generally follows a seasonal pattern, shifting from depleted earlywood (EW) to enriched latewood (LW) (+1.5‰). This suggests the use of non-growing season (NGS) $\delta^{18}O_{ppt}$ for EW ($U$ = 1083, $p$ = 0.716) and growing season (GS) $\delta^{18}O_{ppt}$ LW ($U$ = 1318, $p$ = 0.053) growth, since phreatic water is typically unavailable to *Fraxinus* roots due to the presence of a local gravel layer in floodplain soils (Singer et al., 2013, 2014). These water sources correspond to the evolving isotopic signature of vadose zone water during the growing season. However, in 2001 and 2003 the $\delta^{18}O_{msw}$ and sub-annual pattern for *Fraxinus* is depleted compared with $\delta^{18}O_{ppt}$. This may result from overbank flooding and/or a very elevated water table in these years of high streamflow (Sargeant & Singer, 2016), which delivers isotopically depleted water to the vadose zone. For this same period (2000-2006), the *Populus* tree exhibits much smaller swings in seasonal water use and it appears to have used a mix of NGS $\delta^{18}O_{ppt}$ and phreatic water for both EW and LW.

During the water-use convergence period, *Fraxinus* $\delta^{18}O_{msw}$ is similar to NGS and GS $\delta^{18}O_{ppt}$, ($F_{3,35}$ = 2.96, $p$ = 0.047) but distinct from $\delta^{18}O_{RW}$ ($U$ = 737, $p$ <0.001). Notably, *Populus* underwent a shift in $\delta^{18}O_{msw}$ to that of $\delta^{18}O_{ppt}$ for both parts of the growing season ($F_{3,36}$ = 8.10, $p$ <0.001). This convergence water source use between these species indicates *Populus* switched from phreatic to vadose zone moisture uptake (e.g., use of NGS $\delta^{18}O_{ppt}$ in 2009, Figure 5b). The *Populus* water source shift suggests that the phreatic reservoir became inaccessible. An ancillary dataset provides evidence of local river incision of ~1.5 m (Figure S5) between 2003-2007 (Parrot, 2015). Downcutting of the riverbed reduces local river stage and by extension, lateral hyporheic flow into the floodplain, leading to a water table

decline. Thus, *Populus* roots apparently became stranded from the phreatic zone by river incision and were subsequently forced to rely on vadose zone water.

### 3.3 Example 3: Annual vs. sub-annual water source use, Mas Thibert (MT)

There are open questions as to whether gravel layers in river floodplains impede access to phreatic water with river floodplains for shallow-rooting species such as *Fraxinus* (Singer et al., 2014), and whether seasonal patterns of water use for such species are consistent across its distribution within the same region.

$\delta^{18}O_{wv}$ measurements taken from the nearby (7 km away) monitoring station of Delattre et al., (2015) to reconstruct historical $\delta^{18}O_{wv}$ and $\delta^{18}O_{ppt}$ for our study period (Figure S6) along with climate data (400 m away) sourced from MeteoFrance (meteofrance.com, 2017). This dataset and associated reconstruction enabled more accurate (nearly) site-based estimates of $\delta^{18}O_{wv}$ to the $\delta^{18}O_{msw}$ and seasonal $\delta^{18}O_{ppt}$, as opposed to those obtained from the nearest GNIP station (IAEA/WMO, 2017) at Avignon (~46 km north). We used reported values of leaf gas exchange ($g_s/E$) for each species in similar environmental conditions. The stomatal conductance / transpiration rates for *Fraxinus* were 0.145 ±0.065 mol $m^{-2}s^{-1}$ / 2.85 ±1.05 mmol $m^{-2}s^{-1}$ (Lemoine et al., 2001) and for *Populus* were 0.1625 ±0.1225 mol $m^{-2}s^{-1}$ /2.65 ±1.85 mmol $m^{-2}s^{-1}$ (Lambs et al., 2006).

Figure 5c demonstrates that a high variability in water source use ($\delta^{18}O_{msw}$) is dampened within the whole-ring series. The sub-annual $\delta^{18}O_{msw}$ within a single year of growth can range from 1.7 ‰ (2004) to 10.5 ‰ (2009), whilst the range in recorded annual $\delta^{18}O_{msw}$ values is 4.8 ‰. The year 2009 exhibits a substantial difference (8.5 ‰) between annual $\delta^{18}O_{msw}$ (-5.4 ‰) and the third LW value of $\delta^{18}O_{msw}$ (+3.1 ‰). This suggests that sub-annual information on water source uses can provide a more robust characterization of water use than annual data, and this information would enable better understanding plant-water relations.

The $\delta^{18}O_{msw}$ in EW is generally enriched by +2.0 ‰ compared to that in LW ($T_{52}$ = 2.91, $p$ = 0.005), but see 2006 and 2009 when this pattern is reversed. Interestingly, the general pattern of enriched EW and depleted LW contrasts to the seasonal pattern of water use by the same species at DM (Figure 5b). This suggests differential seasonal availability of water at particular rooting depths between the two sites. River water is indistinguishable from shallow phreatic water ($T_{16}$ = -1.00, $p$ = 0.333) at MT, indicating that hyporheic flow from the Rhône is the main source of water to the alluvial aquifer. There is also a clear separation between the means of NGS and GS $\delta^{18}O_{ppt}$ ($T_{20}$ = -2.61, $p$ = 0.017) at this site because of a markedly warmer growing season that delivers isotopically enriched precipitation. Once

603 infiltrated into the vadose zone, this water becomes further enriched by evaporation before uptake by
604 *Fraxinus*.

605 Despite the absence of a gravel layer impeding *Fraxinus* rooting depth at MT, it still apparently
606 relies on infiltrated precipitation. The greater enrichment in $\delta^{18}O_{msw}$ for EW growth compared with LW
607 may be driven by high spring-time evaporative enrichment of shallow vadose zone moisture arising from
608 the strong and dry, northerly 'Mistral' winds, whilst the LW $\delta^{18}O_{msw}$ may correspond to a relatively
609 lessened evaporative enrichment controlled by summer temperatures.

610

611 **4 DISCUSSION AND CONCLUSION**

612

613 The 'Identification of Source-water Oxygen isotopes in trees Toolkit' (ISO-Tool), presented here,
614 is designed to provide the necessary techniques to reconstruct the historical $\delta^{18}O_{sw}$ signatures used by
615 trees for annual or sub-annual periods and to interpret these signatures in terms of hydroclimatic
616 variability. ISO-Tool is designed to be used with three hierarchical 'Tool Groups' with different data
617 requirements (and thus output resolution). These 'Tool Groups' enable investigations of tree water
618 source use under various conditions of data availability. By bringing together techniques from multiple
619 research fields (e.g. dendrochronology, stable isotopes, hydrological data analysis, tree physiology,
620 ecohydrology) into a single and straightforward application framework, our aim is for ISO-Tool to be
621 easily accessible to scientific researchers interested to retrospectively identify water sources to plants
622 for a range of applications (e.g., to relate them to xylem water isotopes). We highlighted the utility of
623 the toolkit under varying scenarios of data availability and provided several applications: restoration
624 assessment (PB); seasonal water source use (DM) and a comparative analysis of low and high resolution
625 $\delta^{18}O_{cell}$ sampling (MT).

626 The underlying principle of ISO-Tool is that the $\delta^{18}O_{sw}$ utilized for any period can be obtained by
627 reversing the fractionation and exchange mechanisms responsible for producing the discrepancy
628 between the $\delta^{18}O$ of root zone moisture and that which is recorded in tree-ring in cellulose. Singer et al.,
629 (2014) and Sargeant and Singer (2016) utilized versions of this inverse modeling technique to determine
630 the isotopic signature of water sources used by riparian trees at annual and sub-annual resolution,
631 respectively. Recent work by Bose et al., (2016) reconstructed the isotopic value of soil water for trees
632 over a larger spatial scale using thirteen $\delta^{18}O_{cell}$ datasets. The present paper formalizes such methods
633 into a set of tools for assessing historical water use by plants.

The flexible modeling framework allows the user to define the values of specific parameters (e.g. leaf temperature and boundary layer conductance), circumventing the reliance on default values which may improve in accuracy as research in these topic areas advances. Additionally, if the researcher is able to provide more accurate estimates for such parameters, they can be easily incorporated. Advancement of the modeling component will necessitation the inclusion of post-photosynthetic metabolism and transport mechanisms (Gessler et al., 2014). As such, we acknowledge that the calculated $\delta^{18}O_{msw}$ values may not represent the *absolute* $\delta^{18}O$ values of water utilized by the tree, yet they remain valuable for interpreting ecohydrological interactions as long as isotopically distinct endmembers are present.

We have shown the ISO-Tool capability to add value to analyses of restoration efforts, wherein the back-calculated source waters from a particular tree can be used to assess whether it benefited from increased flow support of an elevated floodplain water table (PB). We also applied ISO-Tool to ascertain water use on sub-annual timescales, and this was demonstrated to be much more powerful than annual analyses, providing useful information on the seasonal variability of water sources during an annual growth cycle (DM and MT). This latter work pointed to strong seasonal differences in water usage for the same species at different locations along the Rhône River, a result that would not appear in annual data.

The back-calculation of tree $\delta^{18}O_{sw}$, is a more robust technique than $\delta^{18}O_{cell}$ for determining the water source utilized by trees. We suggest that ISO-Tool is well suited for site-based studies and those along climatic transects, as it is capable of shedding light on the nature of hydrological partitioning within the plant rooting zones.

We developed a new tool capable of providing estimates of the $\delta^{18}O$ of source-water(s) utilized by trees and incorporating a hierarchical data quality scheme for the constraint of model variables and interpretative techniques of model output, based on available data sources. We foresee ISO-Tool being useful in a wide range of ecohydrological applications with retrospective insights into source-water utilization by trees becoming an increasingly diagnostic tool under future hydroclimatic changes.

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## References

Allen, C. D., Breshears, D. D., & McDowell, N. G. (2015). On underestimation of global vulnerability to tree mortality and forest die-off from hotter drought in the Anthropocene. *Ecosphere*, *6*(8), 1-55.

Allen, C. D., Macalady, A. K., Chenchouni, H., Bachelet, D., McDowell, N., Vennetier, M., et al. (2010). A global overview of drought and heat-induced tree mortality reveals emerging climate change risks for forests. *Forest ecology and management*, *259*(4), 660-684.

Allen, M. R., & Ingram, W. J. (2002). Constraints on future changes in climate and the hydrologic cycle. *Nature*, *419*(6903), 224.

Allison, G. B., Barnes, C. J., Hughes, M. W., & Leaney, F. W. J. (1984). Effect of climate and vegetation on oxygen-18 and deuterium profiles in soils. *Isotopes Hydrology, IAEA, Vienna*, 105-122.

Amoros, C., & Bornette, G. (2002). Connectivity and biocomplexity in waterbodies of riverine floodplains. *Freshwater biology*, *47*(4), 761-776.

Amoros, C., Elger, A., Dufour, S., Grosprêtre, L., Piégay, H., & Henry, C. (2005). Flood scouring and groundwater supply in side-channel rehabilitation of the Rhône river, France. *Arch. Hydrobiol*, *155*, 147-167.

Anderegg, W. R., Kane, J. M., & Anderegg, L. D. (2013). Consequences of widespread tree mortality triggered by drought and temperature stress. *Nature climate change*, *3*(1), 30.

Anderson, W. T., Bernasconi, S. M., McKenzie, J. A., Saurer, M., & Schweingruber, F. (2002). Model evaluation for reconstructing the oxygen isotopic composition in precipitation from tree ring cellulose over the last century. *Chemical Geology*, *182*(2-4), 121-137.

695 Barbeta, A., Ogée, J., & Peñuelas, J. (2018). Stable-Isotope Techniques to Investigate Sources of Plant
696 Water. In *Advances in Plant Ecophysiology Techniques* (pp. 439-456). Springer, Cham.

697 Barbour, M. M. (2007). Stable oxygen isotope composition of plant tissue: a review. *Functional Plant*
698 *Biology*, *34*(2), 83-94.

699 Barbour, M. M., & Farquhar, G. D. (2000). Relative humidity-and ABA-induced variation in carbon and
700 oxygen isotope ratios of cotton leaves. *Plant, Cell & Environment*, *23*(5), 473-485.

701 Barbour, M. M., Fischer, R. A., Sayre, K. D., & Farquhar, G. D. (2000). Oxygen isotope ratio of leaf and
702 grain material correlates with stomatal conductance and grain yield in irrigated wheat. *Functional Plant*
703 *Biology*, *27*(7), 625-637.

704 Barbour, M. M., Roden, J. S., Farquhar, G. D., & Ehleringer, J. R. (2004). Expressing leaf water and
705 cellulose oxygen isotope ratios as enrichment above source water reveals evidence of a Péclet effect.
706 *Oecologia*, *138*(3), 426-435.

707 Barbour, M. M., Walcroft, A. S., & Farquhar, G. D. (2002). Seasonal variation in δ13C and δ18O of
708 cellulose from growth rings of Pinus radiata. *Plant, Cell & Environment*, *25*(11), 1483-1499.

709 Battipaglia, G., Jäggi, M., Saurer, M., Siegwolf, R. T., & Cotrufo, M. F. (2008). Climatic sensitivity of δ18O
710 in the wood and cellulose of tree rings: Results from a mixed stand of Acer pseudoplatanus L. and Fagus
711 sylvatica L. *Palaeogeography, Palaeoclimatology, Palaeoecology*, *261*(1-2), 193-202.

712 Beijing, China. *Journal of Geophysical Research: Atmospheres*, *115*(D1).

713 Berkelhammer, M., & Stott, L. D. (2009). Modeled and observed intra-ring δ18O cycles within late
714 Holocene Bristlecone Pine tree samples. *Chemical Geology*, *264*(1-4), 13-23.

715 Berner, L. T., Law, B. E., & Hudiburg, T. W. (2017). Water availability limits tree productivity, carbon
716 stocks, and carbon residence time in mature forests across the western US. *Biogeosciences*, *14*(2), 365-
717 378.

718 Bertrand, G., Masini, J., Goldscheider, N., Meeks, J., Lavastre, V., Celle-Jeanton, H., et al. (2014).
719 Determination of spatiotemporal variability of tree water uptake using stable isotopes (δ18O, δ2H) in an
720 alluvial system supplied by a high-altitude watershed, Pfyn forest, Switzerland. *Ecohydrology*, *7*(2), 319-
721 333.

722 Beyer, M., Koeniger, P., Gaj, M., Hamutoko, J. T., Wanke, H., & Himmelsbach, T. (2016). A deuterium-
723 based labeling technique for the investigation of rooting depths, water uptake dynamics and
724 unsaturated zone water transport in semiarid environments. *Journal of Hydrology*, *533*, 627-643.

725 Bonan, G. B. (2008). Forests and climate change: forcings, feedbacks, and the climate benefits of forests.
726 *science*, *320*(5882), 1444-1449.

727 Bose, T., Sengupta, S., Chakraborty, S., & Borgaonkar, H. (2016). Reconstruction of soil water oxygen
728 isotope values from tree ring cellulose and its implications for paleoclimate studies. *Quaternary*
729 *International*, *425*, 387-398.

730 Böttcher, K., Aurela, M., Kervinen, M., Markkanen, T., Mattila, O. P., Kolari, P., et al. (2014). MODIS time-
731 series-derived indicators for the beginning of the growing season in boreal coniferous forest—A
732 comparison with CO2 flux measurements and phenological observations in Finland. *Remote Sensing of*
733 *Environment*, *140*, 625-638.

734 Bowen, G. J. (2017) The Online Isotopes in Precipitation Calculator, version 3.1. Accessible at
735 http://www.waterisotopes.org. [Accessed May 2017].

736 Bowen, G. J., & Revenaugh, J. (2003). Interpolating the isotopic composition of modern meteoric
737 precipitation. *Water Resources Research*, *39*(10).

738 Bowen, G. J., & Wilkinson, B. (2002). Spatial distribution of δ18O in meteoric precipitation. *Geology*,
739 *30*(4), 315-318.

740 Bowen, G. J., Wassenaar, L. I., & Hobson, K. A. (2005). Global application of stable hydrogen and oxygen
741 isotopes to wildlife forensics. *Oecologia*, *143*(3), 337-348.

742 Bréda, N., Huc, R., Granier, A., & Dreyer, E. (2006). Temperate forest trees and stands under severe
743 drought: a review of ecophysiological responses, adaptation processes and long-term consequences.
744 *Annals of Forest Science*, *63*(6), 625-644.

745 Brenner, A. J., & Jarvis, P. G. (1995). A heated leaf replica technique for determination of leaf boundary
746 layer conductance in the field. *Agricultural and Forest Meteorology*, *72*(3-4), 261-275.

747 Breshears, D. D., Cobb, N. S., Rich, P. M., Price, K. P., Allen, C. D., Balice, R. G., et al.(2005). Regional
748 vegetation die-off in response to global-change-type drought. *Proceedings of the National Academy of*
749 *Sciences*, *102*(42), 15144-15148.

750 Brooks, J. R., Meinzer, F. C., Coulombe, R., & Gregg, J. (2002). Hydraulic redistribution of soil water
751 during summer drought in two contrasting Pacific Northwest coniferous forests. *Tree physiology*, *22*(15-
752 16), 1107-1117.

753 Buhay, W. M., Edwards, T. W. D., & Aravena, R. (1996). Evaluating kinetic fractionation factors used for
754 reconstructions from oxgen and hydrogen isotope ratios in plant water and cellulose. *Geochimica et*
755 *Cosmochimica Acta*, *60*(12), 2209-2218.

756 Busch, D. E., Ingraham, N. L., & Smith, S. D. (1992). Water uptake in woody riparian phreatophytes of the
757 southwestern United States: a stable isotope study. *Ecological Applications*, *2*(4), 450-459.

758 Caldwell, M. M., Dawson, T. E., & Richards, J. H. (1998). Hydraulic lift: consequences of water efflux from
759 the roots of plants. *Oecologia*, *113*(2), 151-161.

760 Celle-Jeanton, H., Travi, Y., & Blavoux, B. (2001). Isotopic typology of the precipitation in the Western

761 Cernusak, L. A., Barbour, M. M., Arndt, S. K., Cheesman, A. W., English, N. B., Feild, T. S., et al. (2016).
762 Stable isotopes in leaf water of terrestrial plants. *Plant, Cell & Environment*, *39*(5), 1087-1102.

763 Charru, M., Seynave, I., Morneau, F., & Bontemps, J. D. (2010). Recent changes in forest productivity: an
764 analysis of national forest inventory data for common beech (Fagus sylvatica L.) in north-eastern France.
765 *Forest Ecology and Management*, *260*(5), 864-874.

766 Chen, L., Huang, J. G., Alam, S. A., Zhai, L., Dawson, A., Stadt, K. J., & Comeau, P. G. (2017). Drought
767 causes reduced growth of trembling aspen in western Canada. *Global change biology*, *23*(7), 2887-2902.

768 Choat, B., Jansen, S., Brodribb, T. J., Cochard, H., Delzon, S., Bhaskar, R., et al. (2012). Global
769 convergence in the vulnerability of forests to drought. *Nature*, *491*(7426), 752.

770 Ciais, P., Reichstein, M., Viovy, N., Granier, A., Ogée, J., Allard, V., et al. (2005). Europe-wide reduction in
771 primary productivity caused by the heat and drought in 2003. *Nature*, *437*(7058), 529.

772 Clark, J. S., Bell, D. M., Hersh, M. H., & Nichols, L. (2011). Climate change vulnerability of forest
773 biodiversity: climate and competition tracking of demographic rates. *Global Change Biology*, *17*(5),
774 1834-1849.

775 Clark, J. S., Iverson, L., Woodall, C. W., Allen, C. D., Bell, D. M., Bragg, D. C., et al. (2016). The impacts of
776 increasing drought on forest dynamics, structure, and biodiversity in the United States. *Global Change*
777 *Biology*, *22*(7), 2329-2352.

778 Cocozza, C., Palombo, C., Tognetti, R., La Porta, N., Anichini, M., Giovannelli, A., et al. (2016). Monitoring
779 intra-annual dynamics of wood formation with microcores and dendrometers in Picea abies at two
780 different altitudes. *Tree physiology*, *36*(7), 832-846.Mediterranean region at three different time scales.
781 *Geophysical Research Letters*, *28*(7), 1215-1218.

782 Craig H, Gordon L. 1965. Deuterium and oxygen-18 variations in the ocean and marine atmosphere. In
783 Proceedings of a conference on Stable isotopes in Oceanographic studies and Paleotemperatures,
784 Spoleto, Italy.

785 Cuny, H. E., Rathgeber, C. B., Frank, D., Fonti, P., Mäkinen, H., Prislan, P., et al. (2015). Woody biomass
786 production lags stem-girth increase by over one month in coniferous forests. *Nature Plants*, *1*(11),
787 15160.

788 Currie, D. J., & Paquin, V. (1987). Large-scale biogeographical patterns of species richness of trees.
789 *Nature*, *329*(6137), 326.

790 Dansgaard, W. (1964). Stable isotopes in precipitation. *Tellus*, *16*(4), 436-468.

791 David, T. S., Pinto, C. A., Nadezhdina, N., Kurz-Besson, C., Henriques, M. O., Quilhó, T., et al. (2013). Root
792 functioning, tree water use and hydraulic redistribution in Quercus suber trees: A modeling approach
793 based on root sap flow. Forest Ecology and Management, 307, 136-146.

794 Dawson, T. E. (1993). Hydraulic lift and water use by plants: implications for water balance, performance
795 and plant-plant interactions. *Oecologia*, *95*(4), 565-574.

796 Dawson, T. E. (1996). Determining water use by trees and forests from isotopic, energy balance and
797 transpiration analyses: the roles of tree size and hydraulic lift. *Tree physiology*, *16*(1-2), 263-272.

798 Dawson, T. E., & Ehleringer, J. R. (1991). Streamside trees that do not use stream water. *Nature*,
799 *350*(6316), 335.

800 Dawson, T. E., & Pate, J. S. (1996). Seasonal water uptake and movement in root systems of Australian
801 phraeatophytic plants of dimorphic root morphology: a stable isotope investigation. *Oecologia*, *107*(1),
802 13-20.

803 Dawson, T. E., Mambelli, S., Plamboeck, A. H., Templer, P. H., & Tu, K. P. (2002). Stable isotopes in plant
804 ecology. *Annual review of ecology and systematics*, *33*(1), 507-559.

805 Delattre, H., Vallet-Coulomb, C., & Sonzogni, C. (2015). Deuterium excess in the atmospheric water
806 vapour of a Mediterranean coastal wetland: regional vs. local signatures. *Atmospheric Chemistry and*
807 *Physics*, *15*(17), 10167-10181.

808 Donat, M. G., Lowry, A. L., Alexander, L. V., O'Gorman, P. A., & Maher, N. (2016). More extreme
809 precipitation in the world's dry and wet regions. *Nature Climate Change*, *6*(5), 508.

810 Dongmann, G., Nürnberg, H. W., Förstel, H., & Wagener, K. (1974). On the enrichment of H 2 18 O in the
811 leaves of transpiring plants. *Radiation and environmental biophysics*, *11*(1), 41-52.

812 Drake, P. L., & Franks, P. J. (2003). Water resource partitioning, stem xylem hydraulic properties, and
813 plant water use strategies in a seasonally dry riparian tropical rainforest. *Oecologia*, *137*(3), 321-329.

814 Ellison, D., Morris, C. E., Locatelli, B., Sheil, D., Cohen, J., Murdiyarso, D., et al. (2017). Trees, forests and
815 water: Cool insights for a hot world. *Global Environmental Change*, *43*, 51-61.

816 Ellsworth, P. Z., & Williams, D. G. (2007). Hydrogen isotope fractionation during water uptake by woody
817 xerophytes. *Plant and Soil*, *291*(1-2), 93-107.

818 Entekhabi, D., Njoku, E. G., O'Neill, P. E., Kellogg, K. H., Crow, W. T., Edelstein, W. N., et al. (2010). The
819 soil moisture active passive (SMAP) mission. *Proceedings of the IEEE*, *98*(5), 704-716.

820 Evans, C. M., Dritschel, D. G., & Singer, M. B. (2018). Modeling subsurface hydrology in floodplains.
821 *Water Resources Research*, *54*(3), 1428-1459.

822 Farquhar, G. D., & Lloyd, J. (1993). Carbon and oxygen isotope effects in the exchange of carbon dioxide
823 between terrestrial plants and the atmosphere. In *Stable isotopes and plant carbon-water relations* (pp.
824 47-70).

825 Farquhar, G.D., Barbour, M.M., & Henry, B.K .(1998) Interpretation of oxygen isotope composition of
826 leaf material. In Griffiths, H. (1998). *Stable isotopes: integration of biological, ecological and geochemical*
827 *processes* (p. 438). Oxford: Bios scientific publishers.

828 Flanagan, L. B., & Ehleringer, J. R. (1991). Stable isotope composition of stem and leaf water:
829 applications to the study of plant water use. *Functional Ecology*, *5*(2), 270-277.

830 Flanagan, L. B., Comstock, J. P., & Ehleringer, J. R. (1991). Comparison of modeled and observed
831 environmental influences on the stable oxygen and hydrogen isotope composition of leaf water in
832 Phaseolus vulgaris L. *Plant Physiology*, *96*(2), 588-596.

833 García-Ruiz, J. M., López-Moreno, J. I., Vicente-Serrano, S. M., Lasanta–Martínez, T., & Beguería, S.
834 (2011). Mediterranean water resources in a global change scenario. *Earth-Science Reviews*, *105*(3-4),
835 121-139.

836 Gärtner, H., Lucchinetti, S., & Schweingruber, F. H. (2014). New perspectives for wood anatomical
837 analysis in dendrosciences: the GSL1-microtome. *Dendrochronologia*, *32*(1), 47-51.

838 Gat, J. R. (1996). Oxygen and hydrogen isotopes in the hydrologic cycle. *Annual Review of Earth and*
839 *Planetary Sciences*, *24*(1), 225-262.

840 Gaudinski, J. B., Dawson, T. E., Quideau, S., Schuur, E. A., Roden, J. S., Trumbore, S. E., ... & Wasylishen,
841 R. E. (2005). Comparative analysis of cellulose preparation techniques for use with 13C, 14C, and 18O
842 isotopic measurements. *Analytical Chemistry*, *77*(22), 7212-7224.

843 Gazis, C., & Feng, X. (2004). A stable isotope study of soil water: evidence for mixing and preferential
844 flow paths. *Geoderma*, *119*(1-2), 97-111.

845 Gessler, A., Brandes, E., Buchmann, N., Helle, G., Rennenberg, H., & Barnard, R. L. (2009). Tracing carbon
846 and oxygen isotope signals from newly assimilated sugars in the leaves to the tree-ring archive. *Plant,*
847 *Cell & Environment*, *32*(7), 780-795.

848 Gessler, A., Ferrio, J. P., Hommel, R., Treydte, K., Werner, R. A., & Monson, R. K. (2014). Stable isotopes
849 in tree rings: towards a mechanistic understanding of isotope fractionation and mixing processes from
850 the leaves to the wood. *Tree physiology*, *34*(8), 796-818.

851 Granier, A., Bréda, N., Biron, P., & Villette, S. (1999). A lumped water balance model to evaluate
852 duration and intensity of drought constraints in forest stands. *Ecological modelling*, *116*(2-3), 269-283.

853 Gray, J., & Thompson, P. (1977). Climatic information from 18O/16O analysis of cellulose, lignin and
854 whole wood from tree rings. *Nature*, *270*(5639), 708.

855 Gröning, M., Lutz, H. O., Roller-Lutz, Z., Kralik, M., Gourcy, L., & Pöltenstein, L. (2012). A simple rain
856 collector preventing water re-evaporation dedicated for δ18O and δ2H analysis of cumulative
857 precipitation samples. *Journal of hydrology*, *448*, 195-200.

858 GSOD – 'Global Surface Summary of the Day'. Accessible at: https://data.nodc.noaa.gov/cgi-
859 bin/iso?id=gov.noaa.ncdc:C00516 [Accessed Dec. 2018].

860 Hansen, A. J., Neilson, R. P., Dale, V. H., Flather, C. H., Iverson, L. R., Currie, D. J., et al. (2001). Global
861 change in forests: responses of species, communities, and biomes: interactions between climate change
862 and land use are projected to cause large shifts in biodiversity. *AIBS Bulletin*, *51*(9), 765-779.

863 Hao, X. M., Li, Y., & Deng, H. J. (2013). Assessment of hydraulic redistribution on desert riparian forests
864 in an extremely arid area. *Environmental monitoring and assessment*, *185*(12), 10027-10038.

865 Hartmann, D.L., A.M.G. Klein Tank, M. Rusticucci, L.V. Alexander, S. Brönnimann, Y. Charabi, F.J.
866 Dentener, E.J. Dlugokencky, D.R. Easterling, A. Kaplan, B.J. Soden, P.W. Thorne, M. Wild and P.M. Zhai,
867 2013: Observations: Atmosphere and Surface. In: Climate Change 2013: The Physical Science Basis.
868 Contribution of Working Group I to the Fifth Assessment Report of the Intergovernmental Panel on
869 Climate Change [Stocker, T.F., D. Qin, G.-K. Plattner, M. Tignor, S.K. Allen, J. Boschung, A. Nauels, Y. Xia,
870 V. Bex and P.M. Midgley (eds.)]. Cambridge University Press, Cambridge, United Kingdom and New York,
871 NY, USA.

872 Helle, G., & Schleser, G. H. (2004). Beyond CO2-fixation by Rubisco–an interpretation of 13C/12C
873 variations in tree rings from novel intra-seasonal studies on broad-leaf trees. *Plant, Cell & Environment*,
874 *27*(3), 367-380.

875 Hill, S. A., Waterhouse, J. S., Field, E. M., Switsur, V. R., & Ap Rees, T. (1995). Rapid recycling of triose
876 phosphates in oak stem tissue. *Plant, Cell & Environment*, *18*(8), 931-936.

877 Holmes, R. L. (1983). Computer-assisted quality control in tree-ring dating and measurement. *Tree-ring*
878 *bulletin*.

879 Horita, J., & Wesolowski, D. J. (1994). Liquid-vapor fractionation of oxygen and hydrogen isotopes of
880 water from the freezing to the critical temperature. *Geochimica et Cosmochimica Acta*, *58*(16), 3425-
881 3437.

882 Horton, J. L., Hart, S. C., & Kolb, T. E. (2003). Physiological condition and water source use of Sonoran
883 Desert riparian trees at the Bill Williams River, Arizona, USA. *Isotopes in Environmental and Health*
884 *Studies*, *39*(1), 69-82.

885 Hsiao, T. C. (1973). Plant responses to water stress. *Annual review of plant physiology*, *24*(1), 519-570.

886 Hsieh, J. C., Chadwick, O. A., Kelly, E. F., & Savin, S. M. (1998). Oxygen isotopic composition of soil water:
887 quantifying evaporation and transpiration. *Geoderma*, *82*(1-3), 269-293.

888 Hunsinger, G. B., Hagopian, W. M., & Jahren, A. H. (2010). Offline oxygen isotope analysis of organic
889 compounds with high N: O. *Rapid Communications in Mass Spectrometry*, *24*(21), 3182-3186.

890 IAEA/WMO (2017). Global Network of Isotopes in Precipitation. The GNIP Database. Accessible at:
891 http://www-naweb.iaea.org/napc/ih/IHS_resources_gnip.html [Accessed May 2017].

892 IAEA/WMO (2017). Global Network of Isotopes in Rivers. The GNIR Database. Accessible at: http://www-
893 naweb.iaea.org/napc/ih/IHS_resources_gnir.html [Accessed May 2017].

894 Jackson, P. C., Meinzer, F. C., Bustamante, M., Goldstein, G., Franco, A., Rundel, P. W., et al. (1999).
895 Partitioning of soil water among tree species in a Brazilian Cerrado ecosystem. *Tree physiology*, *19*(11),
896 717-724.

897 Jackson, R. B., Carpenter, S. R., Dahm, C. N., McKnight, D. M., Naiman, R. J., Postel, S. L., & Running, S.
898 W. (2001). Water in a changing world. *Ecological applications*, *11*(4), 1027-1045.

899 Jacob, H., & Sonntag, C. (1991). An 8-year record of the seasonal variation of 2H and 18O in atmospheric
900 water vapour and precipitation at Heidelberg, Germany. *Tellus B*, *43*(3), 291-300.

901 Jarvis, P. G. (1995). Scaling processes and problems. *Plant, Cell & Environment*, *18*(10), 1079-1089.

902 Jarvis, P. G., & McNaughton, K. G. (1986). Stomatal control of transpiration: scaling up from leaf to
903 region. In *Advances in ecological research* (Vol. 15, pp. 1-49). Academic Press.

904 Jasechko, S., Sharp, Z. D., Gibson, J. J., Birks, S. J., Yi, Y., & Fawcett, P. J. (2013). Terrestrial water fluxes
905 dominated by transpiration. *Nature*, *496*(7445), 347.

906 Jonard, F., André, F., Ponette, Q., Vincke, C., & Jonard, M. (2011). Sap flux density and stomatal
907 conductance of European beech and common oak trees in pure and mixed stands during the summer
908 drought of 2003. *Journal of hydrology*, *409*(1-2), 371-381.

909 Kurz, W. A., Dymond, C. C., Stinson, G., Rampley, G. J., Neilson, E. T., Carroll, A. L., ... & Safranyik, L.
910 (2008). Mountain pine beetle and forest carbon feedback to climate change. *Nature*, *452*(7190), 987.

911 Lambs, L., Loubiat, M., Girel, J., Tissier, J., Peltier, J. P., & Marigo, G. (2006). Survival and acclimatation of
912 Populus nigra to drier conditions after damming of an alpine river, southeast France. Annals of Forest
913 Science, 63(4), 377-385.

914 Lambs, L., Loudes, J., & Berthelot, M. (2002). The use of the stable oxygen isotope (18 O) to trace the
915 distribution and uptake of water in riparian woodlands. *Nukleonika*, *47*, 71-74.

916 Larsson, L. (2014). CooRecorder and Cdendro programs of the CooRecorder/Cdendro package version
917 7.7.

918 Lemoine, D., Peltier, J. P., & Marigo, G. (2001). Comparative studies of the water relations and the
919 hydraulic characteristics in Fraxinus excelsior, Acer pseudoplatanus and A. opalus trees under soil water
920 contrasted conditions. Annals of forest science, 58(7), 723-731.

921 Lewis, S. L., Brando, P. M., Phillips, O. L., van der Heijden, G. M., & Nepstad, D. (2011). The 2010 amazon
922 drought. *Science*, *331*(6017), 554-554.

923 Lin, G., & da SL Sternberg, L. (1993). Hydrogen isotopic fractionation by plant roots during water uptake
924 in coastal wetland plants. In *Stable isotopes and plant carbon-water relations* (pp. 497-510).

925 Lindner, M., Maroschek, M., Netherer, S., Kremer, A., Barbati, A., Garcia-Gonzalo, J., ... & Lexer, M. J.
926 (2010). Climate change impacts, adaptive capacity, and vulnerability of European forest ecosystems.
927 *Forest ecology and management*, *259*(4), 698-709.

928 Lorrey, A. M., Brookman, T. H., Evans, M. N., Fauchereau, N. C., Macinnis-Ng, C., Barbour, M. M., et al.
929 (2016). Stable oxygen isotope signatures of early season wood in New Zealand kauri (Agathis australis)
930 tree rings: Prospects for palaeoclimate reconstruction. *Dendrochronologia*, *40*, 50-63.

931 Luz, B., Kolodny, Y., & Horowitz, M. (1984). Fractionation of oxygen isotopes between mammalian bone-
932 phosphate and environmental drinking water. *Geochimica et Cosmochimica Acta*, *48*(8), 1689-1693.

933 Majoube, M. (1971). Oxygen-18 and deuterium fractionation between water and steam (in French).
934 *Journal de Chimie Physique et de Physico-Chimie Biologique*, *68*, 1423-1436.

935 Mahoney, J. M., & Rood, S. B. (1998). Streamflow requirements for cottonwood seedling recruitment-An
936 integrative model. *Wetlands*, *18*(4), 634-645.

937 Marshall, J. D., & Monserud, R. A. (2006). Co-occurring species differ in tree-ring δ18O trends. *Tree*
938 *physiology*, *26*(8), 1055-1066.

939 MATLAB (R2015b), The MathWorks, Inc., Natick, Massachusetts, United States. (n.d)

940 McCarroll, D., & Loader, N. J. (2004). Stable isotopes in tree rings. *Quaternary Science Reviews*, *23*(7-8),
941 771-801.

942 McDowell, N. G., Beerling, D. J., Breshears, D. D., Fisher, R. A., Raffa, K. F., & Stitt, M. (2011). The
943 interdependence of mechanisms underlying climate-driven vegetation mortality. *Trends in ecology &*
944 *evolution*, *26*(10), 523-532.

945 McNaughton, K. G., & Jarvis, P. G. (1991). Effects of spatial scale on stomatal control of transpiration.
946 *Agricultural and Forest Meteorology*, *54*(2-4), 279-302.

947 Meinzer, F. C., Andrade, J. L., Goldstein, G., Holbrook, N. M., Cavelier, J., & Wright, S. J. (1999).
948 Partitioning of soil water among canopy trees in a seasonally dry tropical forest. *Oecologia*, *121*(3), 293-
949 301.

950 Merlivat, L. (1978). Molecular diffusivities of H 2 16 O, HD 16 O, and H 2 18 O in gases. *The Journal of*
951 *Chemical Physics*, *69*(6), 2864-2871.

952 MeteoFrance – Accessible at: meteofrance.com [Accessed February 2017]

953 Milad, M., Schaich, H., Bürgi, M., & Konold, W. (2011). Climate change and nature conservation in
954 Central European forests: a review of consequences, concepts and challenges. *Forest ecology and*
955 *management*, *261*(4), 829-843.

956 Mintab (version 17), Minitab Inc. (n.d)

957 Mishra, A. K., & Singh, V. P. (2010). A review of drought concepts. *Journal of hydrology*, *391*(1-2), 202-
958 216.

959 NOAA – 'National Oceanic Atmospheric Administration' Accessible at:
960 https://www.esrl.noaa.gov/psd/data/gridded. [Accessed Dec. 2018].

961 Offermann, C., Ferrio, J. P., Holst, J., Grote, R., Siegwolf, R., Kayler, Z., & Gessler, A. (2011). The long way
962 down—are carbon and oxygen isotope signals in the tree ring uncoupled from canopy physiological
963 processes?. *Tree Physiology*, *31*(10), 1088-1102.

964 Ogée, J., Barbour, M. M., Wingate, L., Bert, D., Bosc, A., Stievenard, M., ... & Dewar, R. C. (2009). A
965 single-substrate model to interpret intra-annual stable isotope signals in tree-ring cellulose. *Plant, Cell &*
966 *Environment*, *32*(8), 1071-1090.

967 Oren, R., Sperry, J. S., Katul, G. G., Pataki, D. E., Ewers, B. E., Phillips, N., & Schäfer, K. V. R. (1999). Survey
968 and synthesis of intra-and interspecific variation in stomatal sensitivity to vapour pressure deficit. *Plant,*
969 *Cell & Environment*, *22*(12), 1515-1526.

970 Oshun, J., Dietrich, W. E., Dawson, T. E., & Fung, I. (2016). Dynamic, structured heterogeneity of water
971 isotopes inside hillslopes. *Water Resources Research*, *52*(1), 164-189.

972 Pachauri, R. K., Allen, M. R., Barros, V. R., Broome, J., Cramer, W., Christ, R., et al. (2014). *Climate change*
973 *2014: synthesis report. Contribution of Working Groups I, II and III to the fifth assessment report of the*
974 *Intergovernmental Panel on Climate Change* (p. 151). IPCC.

975 Parnell, A. C., Inger, R., Bearhop, S., & Jackson, A. L. (2010). Source partitioning using stable isotopes:
976 coping with too much variation. *PloS one*, *5*(3), e9672.

977 Parnell, A., Inger, R., Bearhop, S., & Jackson, A. L. (2008). Stable isotope analysis in R (SIAR).

978 Parrot, E. (2015). Analyse spatio-temporelle de la morphologie du chenal du Rhône du Léman a la
979 Méditerranéen (PhD Thesis), ENS, Lyon: 466

980 Pearcy, R.W., E.-D. Schulze and R. Zimmermann. (1989) Measurements of transpiration and leaf
981 conductance. In Pearcy, R.W., Ehleringer, J.R., Mooney, H. and Rundel, P.W. (Eds.), *Plant physiological*
982 *ecology: field methods and instrumentation* (pp. 137-160). Springer, Dordrecht.

983 Pettit, N. E., & Froend, R. H. (2018). How important is groundwater availability and stream perenniality
984 to riparian and floodplain tree growth?. *Hydrological Processes*, *32*(10), 1502-1514.

985 Phillips, D. L., & Gregg, J. W. (2003). Source partitioning using stable isotopes: coping with too many
986 sources. *Oecologia*, *136*(2), 261-269.

987 Phillips, O. L., Aragão, L. E., Lewis, S. L., Fisher, J. B., Lloyd, J., López-González, G., ... & Van Der Heijden,
988 G. (2009). Drought sensitivity of the Amazon rainforest. *Science*, *323*(5919), 1344-1347.

989 Plamboeck, A. H., Grip, H., & Nygren, U. (1999). A hydrological tracer study of water uptake depth in a
990 Scots pine forest under two different water regimes. *Oecologia*, *119*(3), 452-460.

991 Poyatos, R., Granda, V., Molowny-Horas, R., Mencuccini, M., Steppe, K., & Martínez-Vilalta, J. (2016).
992 SAPFLUXNET: towards a global database of sap flow measurements. *Tree physiology*, *36*(12), 1449-1455.

993 Richards, J. H., & Caldwell, M. M. (1987). Hydraulic lift: substantial nocturnal water transport between
994 soil layers by Artemisia tridentata roots. *Oecologia*, *73*(4), 486-489.

995 Richardson, A. D., Jenkins, J. P., Braswell, B. H., Hollinger, D. Y., Ollinger, S. V., & Smith, M. L. (2007). Use
996 of digital webcam images to track spring green-up in a deciduous broadleaf forest. *Oecologia*, *152*(2),
997 323-334.

998 Robertson, J. A., & Gazis, C. A. (2006). An oxygen isotope study of seasonal trends in soil water fluxes at
999 two sites along a climate gradient in Washington state (USA). *Journal of Hydrology*, *328*(1-2), 375-387.

1000 Robock, A., Vinnikov, K. Y., Srinivasan, G., Entin, J. K., Hollinger, S. E., Speranskaya, N. A., et al. (2000).
1001 The global soil moisture data bank. *Bulletin of the American Meteorological Society*, *81*(6), 1281-1300.

1002 Roden, J. S., & Ehleringer, J. R. (2000). Hydrogen and oxygen isotope ratios of tree ring cellulose for field-
1003 grown riparian trees. *Oecologia*, *123*(4), 481-489.

1004 Roden, J. S., Johnstone, J. A., & Dawson, T. E. (2009). Intra-annual variation in the stable oxygen and
1005 carbon isotope ratios of cellulose in tree rings of coast redwood (Sequoia sempervirens). *The Holocene*,
1006 *19*(2), 189-197.

1007 Roden, J. S., Lin, G., & Ehleringer, J. R. (2000). A mechanistic model for interpretation of hydrogen and
1008 oxygen isotope ratios in tree-ring cellulose. *Geochimica et Cosmochimica Acta*, *64*(1), 21-35.

1009 Rossi, S., Anfodillo, T., & Menardi, R. (2006). Trephor: a new tool for sampling microcores from tree
1010 stems. *Iawa Journal*, *27*(1), 89-97.

1011 Rundel, P.W., & Jarrell, W. M., (1989). Water in the Environment. In Pearcy, R.W., Ehleringer, J.R.,
1012 Mooney, H. and Rundel, P.W. (Eds.), *Plant physiological ecology: field methods and instrumentation*.
1013 Springer, Dordrecht.

1014 Sánchez-Pérez, J. M., Lucot, E., Bariac, T., & Trémolières, M. (2008). Water uptake by trees in a riparian
1015 hardwood forest (Rhine floodplain, France). *Hydrological Processes: An International Journal*, *22*(3), 366-
1016 375.

1017 Sargeant, C. I., & Singer, M. B. (2016). Sub-annual variability in historical water source use by
1018 Mediterranean riparian trees. *Ecohydrology*, *9*(7), 1328-1345.

1019 Sarris, D., Christodoulakis, D., & Körner, C. (2011). Impact of recent climatic change on growth of low
1020 elevation eastern Mediterranean forest trees. *Climatic Change*, *106*(2), 203-223.

1021 Schlesinger, W. H., Dietze, M. C., Jackson, R. B., Phillips, R. P., Rhoades, C. C., Rustad, L. E., & Vose, J. M.
1022 (2016). Forest biogeochemistry in response to drought. *Global change biology*, *22*(7), 2318-2328.

1023 Schuepp, P. H. (1993). Tansley review no. 59 leaf boundary layers. *New Phytologist*, *125*(3), 477-507.

1024 Schulze, E. D., Mooney, H. A., Sala, O. E., Jobbagy, E., Buchmann, N., Bauer, G., et al. (1996). Rooting
1025 depth, water availability, and vegetation cover along an aridity gradient in Patagonia. *Oecologia*, *108*(3),
1026 503-511.

1027 Schulze, E. D., Robichaux, R. H., Grace, J., Rundel, P. W., & Ehleringer, J. R. Plant Water Balance.
1028 *BioScience*, *37*(1).

1029 Schwartz, M. D., Reed, B. C., & White, M. A. (2002). Assessing satellite-derived start-of-season measures
1030 in the conterminous USA. *International Journal of Climatology*, *22*(14), 1793-1805.

1031 Scott, R. L., Edwards, E. A., Shuttleworth, W. J., Huxman, T. E., Watts, C., & Goodrich, D. C. (2004).
1032 Interannual and seasonal variation in fluxes of water and carbon dioxide from a riparian woodland
1033 ecosystem. *Agricultural and Forest Meteorology*, *122*(1-2), 65-84.*Measurement*, *54*, 92-105.

1034 Settele, J., Scholes, R., Betts, R. A., Bunn, S., Leadley, P., Nepstad, D., et al. (2015). Terrestrial and inland
1035 water systems. In *Climate Change 2014 Impacts, Adaptation and Vulnerability: Part A: Global and*
1036 *Sectoral Aspects*. Cambridge University Press.

1037 Shafroth, P. B., Stromberg, J. C., & Patten, D. T. (2000). Woody riparian vegetation response to different
1038 alluvial water table regimes. *Western North American Naturalist*, 66-76.

1039 Silva, L. C., Anand, M., & Leithead, M. D. (2010). Recent widespread tree growth decline despite
1040 increasing atmospheric CO2. *PloS one*, *5*(7), e11543.

1041 Singer, M. B., Sargeant, C. I., Piégay, H., Riquier, J., Wilson, R. J., & Evans, C. M. (2014). Floodplain
1042 ecohydrology: Climatic, anthropogenic, and local physical controls on partitioning of water sources to
1043 riparian trees. *Water resources research*, *50*(5), 4490-4513.

1044 Singer, M. B., Stella, J. C., Dufour, S., Piégay, H., Wilson, R. J., & Johnstone, L. (2013). Contrasting water-
1045 uptake and growth responses to drought in co-occurring riparian tree species. *Ecohydrology*, *6*(3), 402-
1046 412.

1047 Singer, M. B., & Dunne, T.(2004). An empirical-stochastic, event-based model for simulating inflow from
1048 a tributary network: Theoretical framework and application to the Sacramento River basin, California,
1049 *Water Resour. Res*., *40*(7), W07506, doi:07510.01029/02003WR002725.

1050 Sippel, S., Zscheischler, J., Heimann, M., Lange, H., Mahecha, M. D., van Oldenborgh, G. J., et al. (2016).
1051 Have precipitation extremes and annual totals been increasing in the world's dry regions over the last 60
1052 years?. *arXiv preprint arXiv:1609.00154*.

1053 Snyder, K. A., & Williams, D. G. (2000). Water sources used by riparian trees varies among stream types
1054 on the San Pedro River, Arizona. *Agricultural and Forest Meteorology*, *105*(1-3), 227-240.

1055 Song, X. I. N., Barbour, M. M., Farquhar, G. D., Vann, D. R., & Helliker, B. R. (2013). Transpiration rate
1056 relates to within-and across-species variations in effective path length in a leaf water model of oxygen
1057 isotope enrichment. *Plant, Cell & Environment*, *36*(7), 1338-1351.

1058 Soudant, A., Loader, N. J., Bäck, J., Levula, J., & Kljun, N. (2016). Intra-annual variability of wood
1059 formation and δ13C in tree-rings at Hyytiälä, Finland. *Agricultural and forest meteorology*, *224*, 17-29.

1060 Sprenger, M., Leistert, H., Gimbel, K., & Weiler, M. (2016). Illuminating hydrological processes at the
1061 soil-vegetation-atmosphere interface with water stable isotopes. *Reviews of Geophysics*, *54*(3), 674-704.

1062 Srivastava, P. K. (2017). Satellite soil moisture: Review of theory and applications in water resources.
1063 *Water Resources Management*, *31*(10), 3161-3176.

1064 Steiger, J., Tabacchi, E., Dufour, S., Corenblit, D., & Peiry, J. L. (2005). Hydrogeomorphic processes
1065 affecting riparian habitat within alluvial channel–floodplain river systems: a review for the temperate
1066 zone. *River Research and Applications*, *21*(7), 719-737.

1067 Stephenson, N. L. (1990). Climatic control of vegetation distribution: the role of the water balance. *The*
1068 *American Naturalist*, *135*(5), 649-670.

1069 Sternberg, L. D. S., Deniro, M. J., & Savidge, R. A. (1986). Oxygen isotope exchange between metabolites
1070 and water during biochemical reactions leading to cellulose synthesis. *Plant Physiology*, *82*(2), 423-427.

1071 Stokes, V. (2004). Assessing water use in plants: an introduction and guide to methods of measurement.
1072 *SCOTTISH FORESTRY.*, *58*(2), 13-21.

1073 Stokes, V. J., Morecroft, M. D., & Morison, J. I. (2006). Boundary layer conductance for contrasting leaf
1074 shapes in a deciduous broadleaved forest canopy. *Agricultural and Forest Meteorology*, *139*(1-2), 40-54.

1075 Stromberg, J. C., & Patten, D. T. (1996). Instream flow and cottonwood growth in the eastern Sierra
1076 Nevada of California, USA. *Regulated Rivers: Research & Management*, *12*(1), 1-12.

1077 Stromberg, J. C., Beauchamp, V. B., Dixon, M. D., Lite, S. J., & Paradzick, C. (2007). Importance of low-
1078 flow and high-flow characteristics to restoration of riparian vegetation along rivers in arid south-western
1079 United States. *Freshwater Biology*, *52*(4), 651-679.

1080 SU, S. L., Singh, D. N., & Baghini, M. S. (2014). A critical review of soil moisture measurement.

1081 Sun, Z., Long, X., & Ma, R. (2016). Water uptake by saltcedar (Tamarix ramosissima) in a desert riparian
1082 forest: responses to intra-annual water table fluctuation. *Hydrological Processes*, *30*(9), 1388-1402.

1083 Suni, T., Berninger, F., Markkanen, T., Keronen, P., Rannik, Ü., & Vesala, T. (2003). Interannual variability
1084 and timing of growing-season CO2 exchange in a boreal forest. *Journal of Geophysical Research:*
1085 *Atmospheres*, *108*(D9).

1086 Trenberth, K. E., Dai, A., Van Der Schrier, G., Jones, P. D., Barichivich, J., Briffa, K. R., & Sheffield, J.
1087 (2014). Global warming and changes in drought. *Nature Climate Change*, *4*(1), 17.

1088 Vargas, A. I., Schaffer, B., Yuhong, L., & Sternberg, L. D. S. L. (2017). Testing plant use of mobile vs
1089 immobile soil water sources using stable isotope experiments. *New Phytologist*, *215*(2), 582-594.

1090 Vicente-Serrano, S. M., Beguería, S., & López-Moreno, J. I. (2010). A multiscalar drought index sensitive
1091 to global warming: the standardized precipitation evapotranspiration index. *Journal of climate*, *23*(7),
1092 1696-1718.

1093 Volkmann, T. H., & Weiler, M. (2014). Continual in situ monitoring of pore water stable isotopes in the
1094 subsurface. *Hydrology and Earth System Sciences*, *18*(5), 1819-1833.

1095 Wen, X. F., Zhang, S. C., Sun, X. M., Yu, G. R., & Lee, X. (2010). Water vapor and precipitation isotope
1096 ratios in

1097 West, A. G., Patrickson, S. J., & Ehleringer, J. R. (2006). Water extraction times for plant and soil
1098 materials used in stable isotope analysis. *Rapid Communications in Mass Spectrometry: An International*
1099 *Journal Devoted to the Rapid Dissemination of Up-to-the-Minute Research in Mass Spectrometry*, *20*(8),
1100 1317-1321.

1101 White, J. C., & Smith, W. K. (2013). Water sources in riparian trees of the southern Appalachian foothills,
1102 USA: A preliminary study with stable isotope analysis. *Riparian Ecology and Conservation*, *1*, 46-52.

1103 White, J. W., Cook, E. R., & Lawrence, J. R. (1985). The DH ratios of sap in trees: Implications for water
1104 sources and tree ring DH ratios. *Geochimica et Cosmochimica Acta*, *49*(1), 237-246.

1105 Yakir, D., & DeNiro, M. J. (1990). Oxygen and hydrogen isotope fractionation during cellulose
1106 metabolism in Lemna gibba L. *Plant Physiology*, *93*(1), 325-332.

1107 Yale University (2017) The Stable Water Vapor Isotope Database (SWVID). Accessible at: http://vapor-
1108 isotope.yale.edu/ [Accessed 15 Jun 2017].

1109 Zencich, S. J., Froend, R. H., Turner, J. V., & Gailitis, V. (2002). Influence of groundwater depth on the
1110 seasonal sources of water accessed by Banksia tree species on a shallow, sandy coastal aquifer.
1111 *Oecologia*, *131*(1), 8-19.

1112 Zweifel, R., Item, H., & Häsler, R. (2001). Link between diurnal stem radius changes and tree water
1113 relations. *Tree Physiology*, *21*(12-13), 869-877.

1114

1115 **FIGURE CAPTIONS**

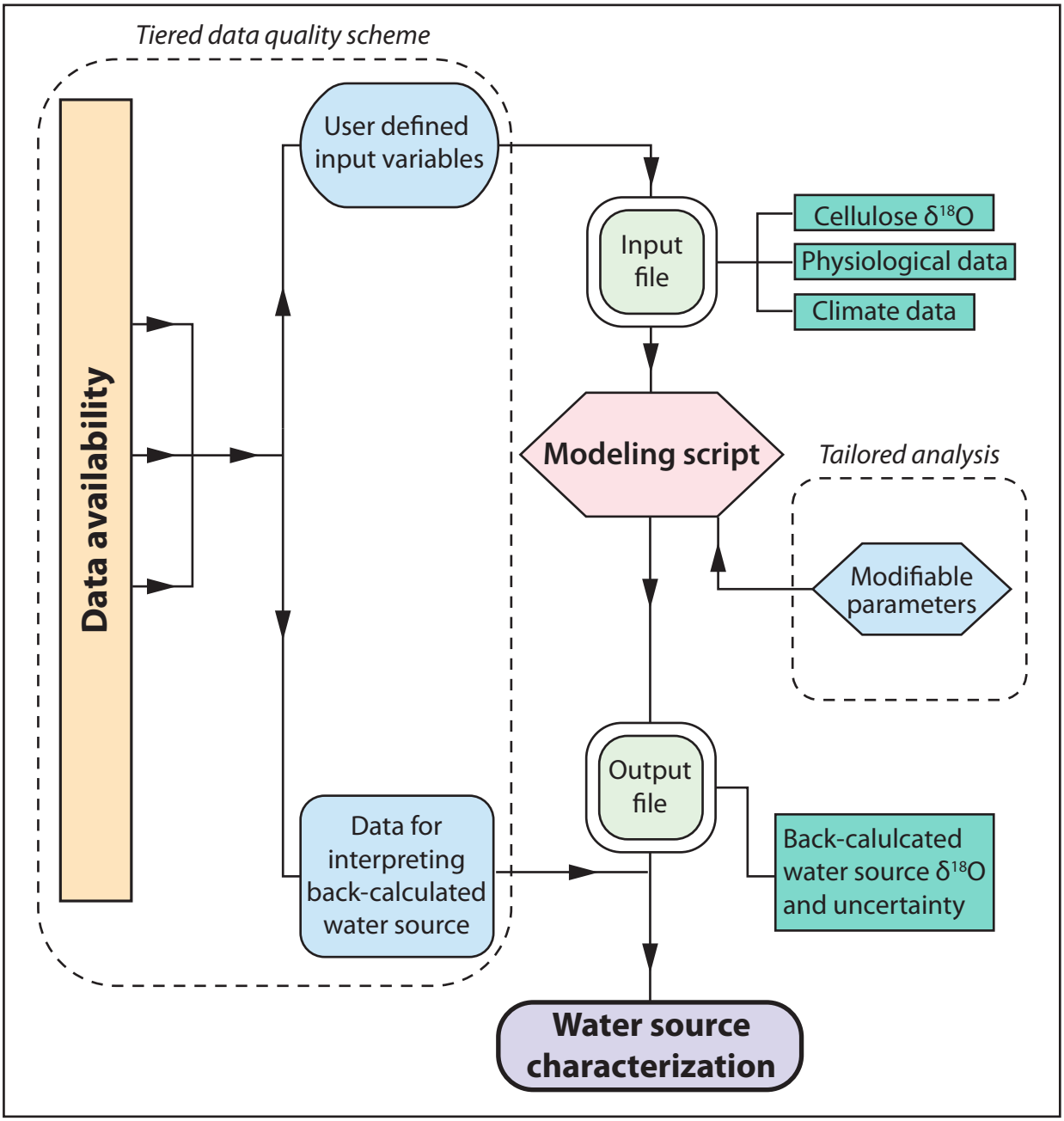
1116 **Figure 1**: Overview of ISO-Tool and the primary components.

1117 **Figure 2**: Key model components and how the required user input values are utilized (a-d), with
1118 associated terminology, and schematics of each process (e).

1119 **Figure 3:** Results of sensitivity analyses of independently (a) and co-varied (b-g) input variables on the
1120 modeled $\delta^{18}O_{sw}$ value. Sensitivity analyses of co-varied model inputs of (b )*T* and *RH*, (c) *RH* and $\delta^{18}O_{wv}$,
1121 (d) *T* and $\delta^{18}O_{wv}$, (e) $\delta^{18}O_{wv}$ with $g_s/E$ pairing, (f) *RH* with $g_s/E$ pairing and (g) *T* with $g_s/E$ pairing. Grey
1122 shading in b-d indicates the range of $\delta^{18}O_{msw}$ results produced by varying the respective input variables
1123 over their observed range of variance for the DM site (May-Sep: 2000-2010).

1124 **Figure 4:** The ISO-Toolkit components and interpretative techniques, divided into tool groups based on
1125 data availability and resolution, with Tool Group A being the most desirable set of data and
1126 interpretative techniques. A review of the mentioned datasets and interpretative techniques is given in
1127 the supplementary text material (S1-3).

1128 **Figure 5**: The mean annual $\delta^{18}O_{msw}$ for *Populus nigra* trees at site PB in relation to potential endmember
1129 water sources mean $\delta^{18}O$ (a), sub-annual $\delta^{18}O_{msw}$ for *Fraxinus excelsior* and *Populus nigra* at site DM
1130 shown in relation to potential endmember water source mean $\delta^{18}O$ (b) and the annual and sub-annual
1131 $\delta^{18}O_{msw}$ for *Fraxinus excelsior* at site MT (c). Colored banding around all reported $\delta^{18}O_{msw}$ is ± 1SD (based
1132 on the output of 1000 Monte Carlo simulations) and endmember water sources are shown as mean $\delta^{18}O$
1133 ± 1SD.


Tiered data quality scheme
Data availability
User defined input variables
Input file
Cellulose δ18O
Physiological data
Climate data
Modeling script
Tailored analysis
Modifiable parameters
Output file
Back-calulcated water source δ18O and uncertainty
Data for interpreting back-calculated water source
Water source characterization

## Model components

### a) *Leaf water $^{18}O$ evaporative enrichment above source water*

$$\Delta_e \approx \varepsilon^* + \varepsilon_k + (\Delta_v - \varepsilon_k)\frac{e_a}{e_i}$$

($\delta^{18}O_{wv}$, $RH$, $T$, $g_s$)

***where :***

$$\varepsilon^* = \exp\left[\frac{1137}{(T_l+273.15)^2} - \frac{0.4156}{(T_l+273.15)} - 2.0667 \times 10^{-3}\right] - 1$$

($T$)

$$\varepsilon_k = \frac{28r_s + 19r_b}{r_s + r_b}$$

($g_s$)

### b) *Péclet effect*

$$\wp = \frac{LE}{CD}$$

($E$)

### c) *Lamina water $^{18}O$ enrichment above source water*

$$\Delta_L = \frac{\Delta_e(1 - exp^{-\wp})}{\wp}$$

### d) *$\delta^{18}O_{cell}$ enrichment above $\delta^{18}O_{sw}$*

$$\Delta_{cell} = \Delta_L(1 - p_{ex}p_x) + \varepsilon_{wc}$$

($\delta^{18}O_{cell}$)

## Terminology

- $\Delta_e$ Enrichment above source water at sites of evaporation
- $\varepsilon^*$ Equilibrium fractionation factor between liquid water and vapor phase
- $\varepsilon_k$ Kinetic fractionation factors of $H_2{}^{18}O$ through the stomata and leaf boundary layer
- $\Delta_v$ Difference between $\delta^{18}O_{sw}$ and $\delta^{18}O_{wv}$
- $e_a$ Vapor pressure outside the leaf
- $e_i$ Vapor pressure inside the leaf
- $T_l$ Leaf temperature (˚C)
- $r_s$ Stomatal resistance (inverse of stomatal conductance [$g_s$])
- $r_b$ Boundary layer resistance (inverse of boundary layer conductance)
  - 28 ‰ kinetic fractionation factor through the stomata ($a_k$)
  - 19 ‰ Kinetic fractionation factor through the boundary layer ($a_{kb} = a_k^{2/3}$)

- $\wp$ Péclet effect
- $L$ Effective pathlength (m) ($L = 2.36 \times 10^{-5}E^{-1.20}$)
- $E$ Transpiration rate (mmol $m^{-2}$ $s^{-1}$)
- $C$ Molar density of water (55.5 x $10^3$ $m^{-3}$)
- $D$ Diffusivity of $H_2{}^{18}O$ in water (2.66 x $10^{-9}$ $m^2$ $s^{-1}$)

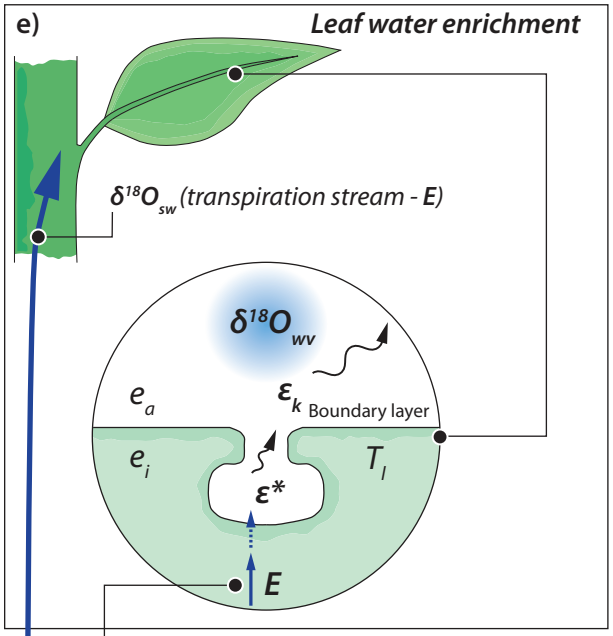
- $\Delta_L$ Leaf water enrichment above source water

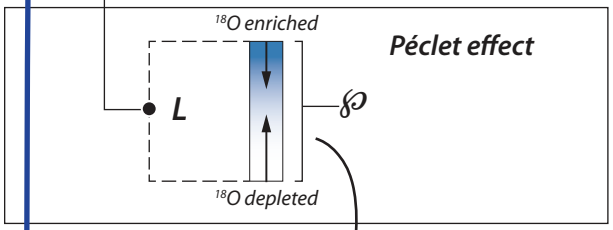
- $\Delta_{cell}$ Cellulose enrichment above $\delta^{18}O_{sw}$
- $p_{ex}$ Proportion of exchangeable oxygen in cellulose formed from sucrose (0.42)
- $p_x$ Proportion of uneriched water in cell (1)
- $\varepsilon_{wc}$ Equilibrium fractionation factor between water and carbonyl oxygen (+27 ± 4 ‰)

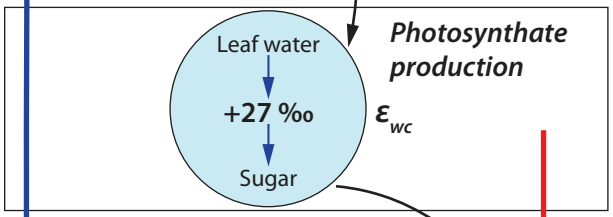
### Required user inputs

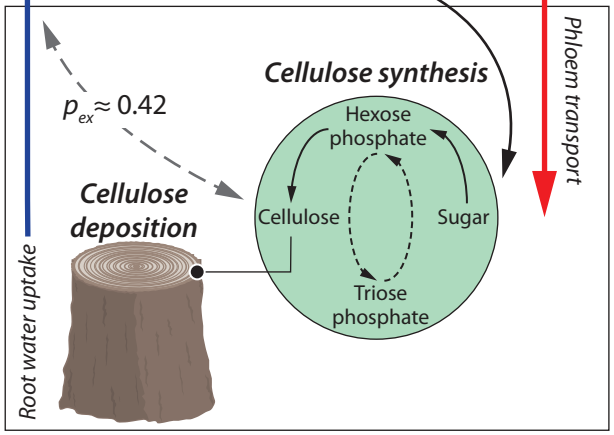
- $\delta^{18}O_{wv}$ Atmospheric water vapor $\delta^{18}O$ (‰)
- $T$ Air temperature (˚C)
- $RH$ Relative humidity (%)
- $g_s$ Stomatal conductance (mol $m^{-2}$ $s^{-1}$)
- $E$ Transpiration rate (mmol $m^{-2}$ $s^{-1}$)
- $\delta^{18}O_{cell}$ Cellulose $\delta^{18}O$ (‰)

## Conceptualization




## Model component references

*Craig & Gordon* (1965), *Dongmann et al.*, (1974), *Flanagan et al.*, (1991), *Farquhar & Lloyd* (1993)

*Majoube* (1971)

*Merlivat et al.*, (1978), *Flanagan & Ehleringer* (1991a & b), *Luz et al.*, (2009)

*Merlivatt et al.*, (1978), *Luz et al.*, (2009)

*Flanagan & Ehleringer* (1991a)

*Farquhar & Lloyd* (1993)
*Song et al.*, (2013)

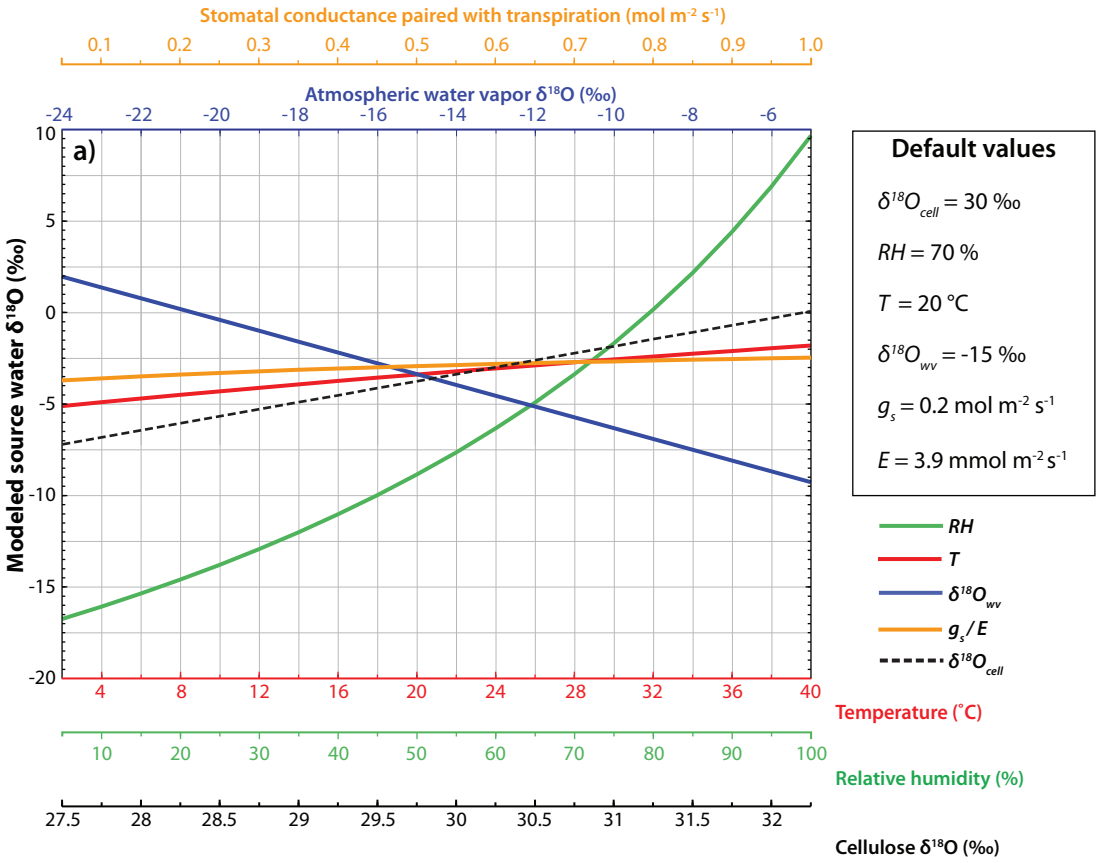
*Farquhar & Lloyd* (1993)

*Hill et al.*, (1995), *Farquhar et al.*, (1998); *Roden et al.*, (2000)

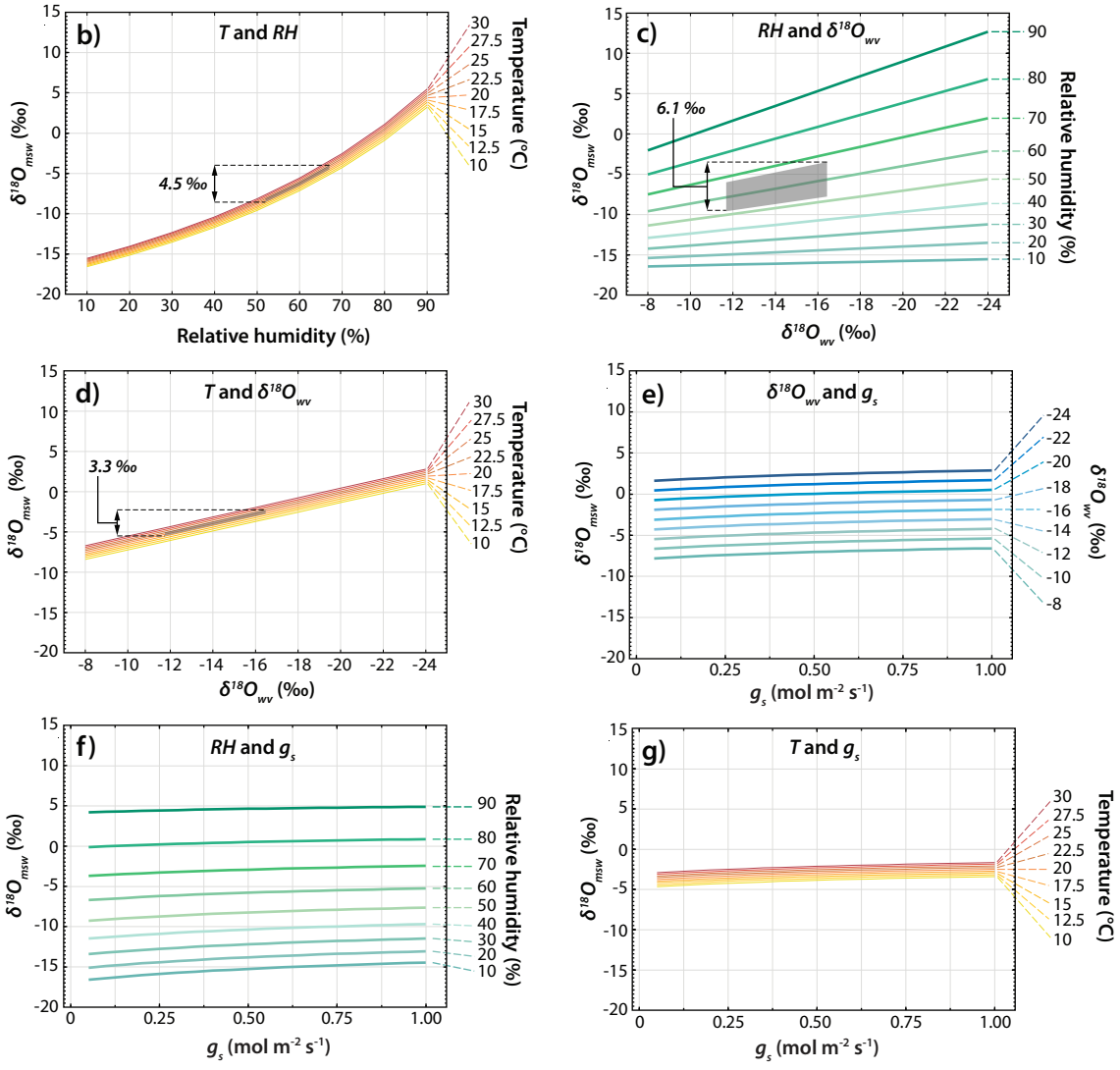
*Barbour et al.*, (2002)

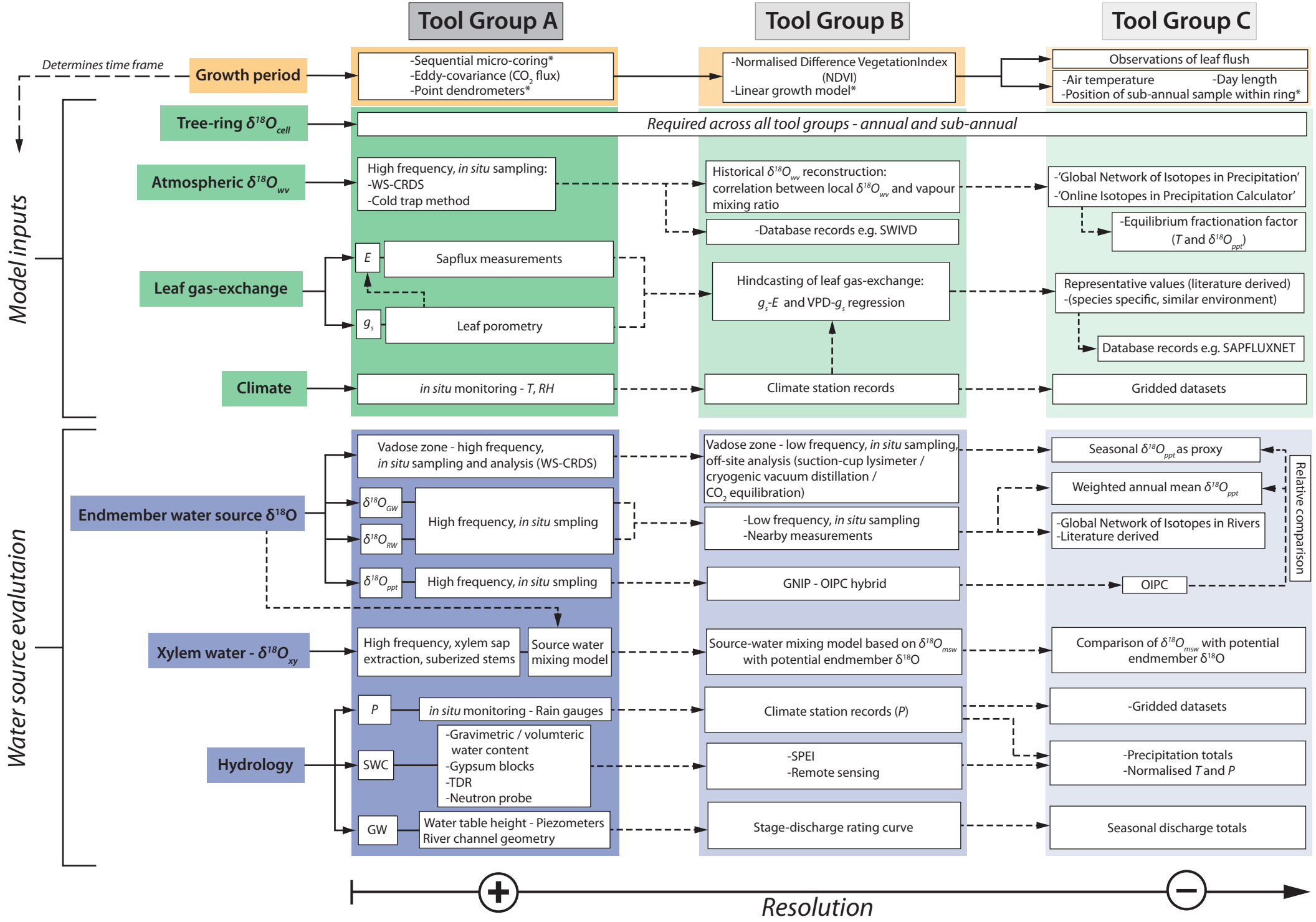
*Sternberg et al.*, (1986), *Yakir & DeNiro* (1990)

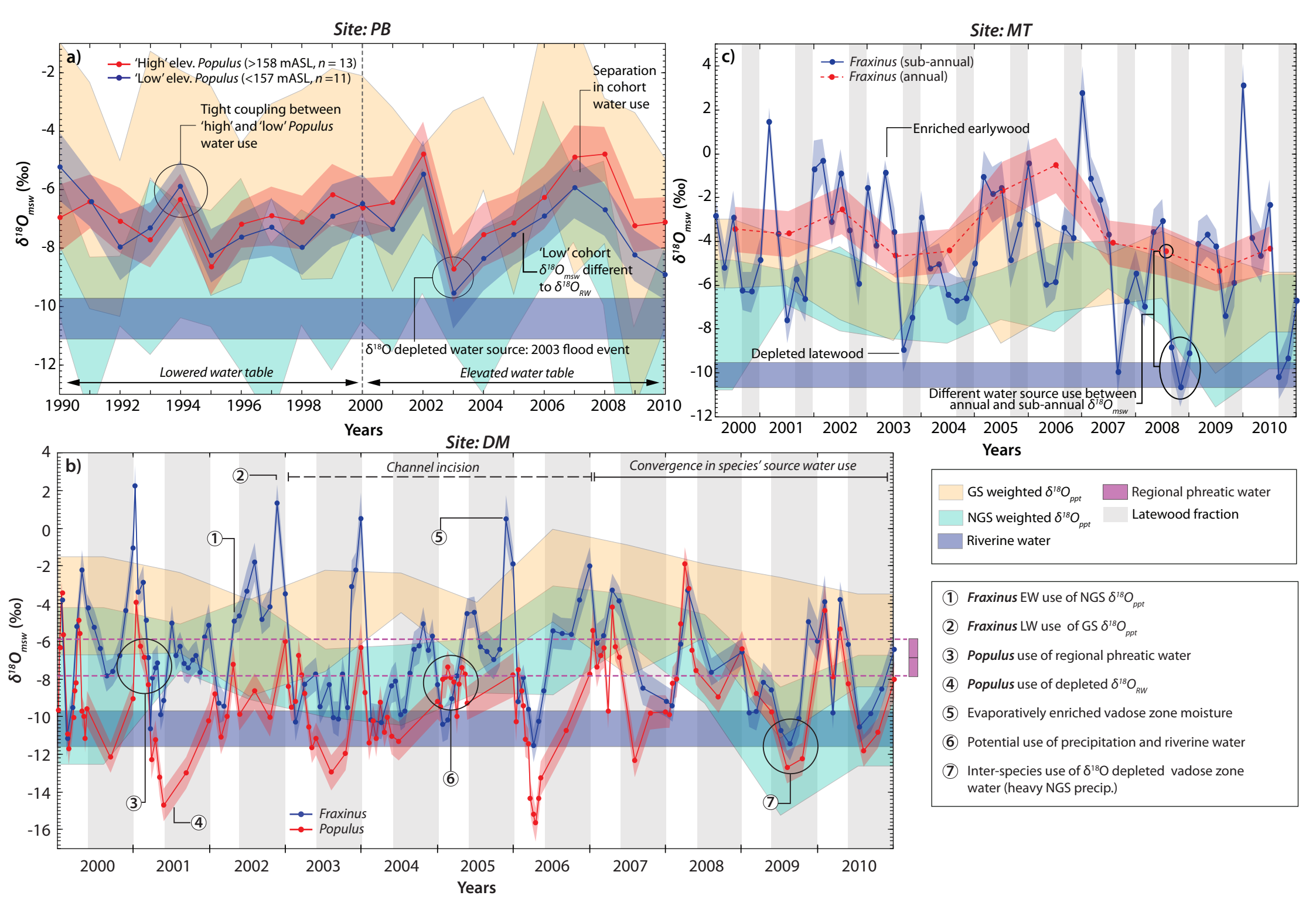
## Independent sensitivity analyses




## Co-varied sensitivity analyses




Tool Group A
Tool Group B
Tool Group C
Determines time frame
Growth period
-Sequential micro-coring*
-Eddy-covariance ($CO_2$ flux)
-Point dendrometers*
-Normalised Difference VegetationIndex (NDVI)
-Linear growth model*
Observations of leaf flush
-Air temperature
-Day length
-Position of sub-annual sample within ring*
Model inputs
Tree-ring $\delta^{18}O_{cell}$
Required across all tool groups - annual and sub-annual
Atmospheric $\delta^{18}O_{wv}$
High frequency, in situ sampling:
-WS-CRDS
-Cold trap method
Historical $\delta^{18}O_{wv}$ reconstruction: correlation between local $\delta^{18}O_{wv}$ and vapour mixing ratio
-Database records e.g. SWIVD
-'Global Network of Isotopes in Precipitation'
-'Online Isotopes in Precipitation Calculator'
-Equilibrium fractionation factor ($T$ and $\delta^{18}O_{ppt}$)
Leaf gas-exchange
$E$
Sapflux measurements
$g_s$
Leaf porometry
Hindcasting of leaf gas-exchange: $g_s$-$E$ and VPD-$g_s$ regression
Representative values (literature derived)
-(species specific, similar environment)
Database records e.g. SAPFLUXNET
Climate
in situ monitoring - T, RH
Climate station records
Gridded datasets
Water source evalutaion
Endmember water source $\delta^{18}O$
Vadose zone - high frequency, in situ sampling and analysis (WS-CRDS)
$\delta^{18}O_{GW}$
$\delta^{18}O_{RW}$
High frequency, in situ smpling
$\delta^{18}O_{ppt}$
High frequency, in situ smpling
Vadose zone - low frequency, in situ sampling, off-site analysis (suction-cup lysimeter / cryogenic vacuum distillation / $CO_2$ equilibration)
-Low frequency, in situ sampling
-Nearby measurements
GNIP - OIPC hybrid
Seasonal $\delta^{18}O_{ppt}$ as proxy
Weighted annual mean $\delta^{18}O_{ppt}$
-Global Network of Isotopes in Rivers
-Literature derived
OIPC
Relative comparison
Xylem water - $\delta^{18}O_{xy}$
High frequency, xylem sap extraction, suberized stems
Source water mixing model
Source-water mixing model based on $\delta^{18}O_{msw}$ with potential endmember $\delta^{18}O$
Comparison of $\delta^{18}O_{msw}$ with potential endmember $\delta^{18}O$
Hydrology
$P$
in situ monitoring - Rain gauges
SWC
-Gravimetric / volumteric water content
-Gypsum blocks
-TDR
-Neutron probe
GW
Water table height - Piezometers
River channel geometry
Climate station records ($P$)
-SPEI
-Remote sensing
Stage-discharge rating curve
-Gridded datasets
-Precipitation totals
-Normalised $T$ and $P$
Seasonal discharge totals
+
−
Resolution


Site: PB
a)
'High' elev. Populus (>158 mASL, n = 13)
'Low' elev. Populus (<157 mASL, n =11)
Tight coupling between 'high' and 'low' Populus water use
Separation in cohort water use
'Low' cohort δ18Omsw different to δ18ORW
δ18O depleted water source: 2003 flood event
Lowered water table
Elevated water table
δ18Omsw (‰)
Years
Site: MT
c)
Fraxinus (sub-annual)
Fraxinus (annual)
Enriched earlywood
Depleted latewood
Different water source use between annual and sub-annual δ18Omsw
Site: DM
b)
Channel incision
Convergence in species' source water use
Fraxinus
Populus
GS weighted δ18Oppt
NGS weighted δ18Oppt
Riverine water
Regional phreatic water
Latewood fraction
① Fraxinus EW use of NGS δ18Oppt
② Fraxinus LW use of GS δ18Oppt
③ Populus use of regional phreatic water
④ Populus use of depleted δ18ORW
⑤ Evaporatively enriched vadose zone moisture
⑥ Potential use of precipitation and riverine water
⑦ Inter-species use of δ18O depleted vadose zone water (heavy NGS precip.)