



# Controls on leaf water hydrogen and oxygen isotopes: a local investigation across seasons and altitude

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**Abstract.** The stable oxygen ( $\delta^{18}\text{O}_{\text{leaf}}$ ) and hydrogen ( $\delta^2\text{H}_{\text{leaf}}$ ) isotopes of leaf water act as a bridge that connects the hydroclimate to plant-derived organic matter. However, it remains unclear whether the source water (i.e., twig water, soil water, and precipitation) or meteorological parameters (i.e., temperature, relative humidity, and precipitation) are the dominant controls on  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$ . Here, we reported a seasonal analysis of  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  together with isotopes from potential source waters and meteorological parameters along an elevation transect on the Chinese Loess Plateau. We found that  $\delta^2\text{H}_{\text{leaf}}$  values were more closely correlated with source water isotopes than  $\delta^{18}\text{O}_{\text{leaf}}$  values, whereas  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values were similarly correlated with meteorological parameters along the elevation transect. Dual-isotope analysis showed that the  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values were closely associated because of their similar altitudinal and seasonal responses, generating a well-defined isotope line relative to the local meteoric water line (LMWL). We also compared the measured  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values with values predicted by the Craig–Gordon model and found no significant differences between them. We demonstrate that the first-order control on  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values was the source water, and the

second-order control was the enrichment associated with biochemical and environmental factors on the Loess Plateau.

## 1 Introduction

The stable isotope compositions of oxygen and hydrogen ( $\delta^{18}\text{O}$  and  $\delta^2\text{H}$ , respectively) are increasingly being used as powerful tracers to follow the path of water from its input as precipitation, movement through the soil, and ultimately to its release as soil evaporation and leaf transpiration (Penna and Van Meerveld, 2019). Leaf water transpiration plays a key role in regulating the water balance at scales ranging from catchment to global. Terrestrial plants can enrich heavier isotopes ( $^2\text{H}$  and  $^{18}\text{O}$ ) in leaf water via evaporative fractionation through the stoma (Helliker and Ehleringer, 2000; Liu et al., 2015; Cernusak et al., 2016), which is highly dependent on atmospheric conditions (e.g., temperature and relative humidity) and biophysiological processes (Farquhar et al., 2007; Kahmen et al., 2011; Cernusak et al., 2016). Subsequently, the isotopic signals from leaf water are integrated into plant organic matter, such as cellulose (e.g., Barbour, 2007; Lehman et al., 2017) and leaf wax (Liu et al., 2016, 2021b), as powerful proxies used for paleoclimate recon-

struction (Pagani et al., 2006; Schefuß et al., 2011; Hepp et al., 2020). However, although leaf water isotopes are the fundamental parameters in ecohydrology and organic biosynthesis, an adequate understanding of controls of leaf water isotopes and the role of source water and hydroclimate in determining leaf water isotopes is still lacking.

$\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values are influenced first by a plant's source water (mainly water taken up by roots from the soil; Cernusak et al., 2016; Barbour et al., 2017; Munksgaard et al., 2017; Liu et al., 2022) and second by the enrichment associated with transpiration (Munksgaard et al., 2017). Soil water for terrestrial plants generally originates from local precipitation, and precipitation isotopes vary spatially and temporally, being subject to controls including temperature, altitude, latitude, distance from the coast, and amount of precipitation (Bowen, 2010; Bowen and Good, 2015; Cernusak et al., 2016). More specifically, soil water isotopes are determined by a mixture of individual precipitation events with distinct isotopic signals and are also affected by evaporation, both of which lead to the development of isotopic gradients in soil water with depth (Allison et al., 1983; Liu et al., 2015). Many studies have shown that the  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values of root/xylem water can be used to characterize the water sources used by plants (Rothfuss and Javaux, 2017; Wu et al., 2018; Wang et al., 2019; Amin et al., 2020; Zhao et al., 2020; Liu et al., 2021a). These studies rested substantially on the assumption that no isotopic fractionation of  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values occurs during water uptake by plant roots (Dawson and Ehleringer, 1991; Ehleringer and Dawson, 1992; Chen et al., 2020), except in saline or xeric environments (Lin and Sternberg, 1993; Ellsworth and Williams, 2007). Some recent studies showed, however, that the occurrence of isotopic fractionation during root water uptake was probably more common than previously thought, especially with respect to  $\delta^2\text{H}$  values (Zhao et al., 2016; Wang et al., 2017; Barbeta et al., 2019; Poca et al., 2019; Liu et al., 2021a, 2022).

In addition to plant source water, leaf water is also isotopically enriched through the evaporative process during transpiration. The enrichment of  $^{18}\text{O}$  and  $^2\text{H}$  by leaf water transpiration can be predicted using the Craig–Gordon model (C–G model). This model was initially proposed to describe the evaporative enrichment of a freely evaporating water body (Craig and Gordon, 1965) and has been modified for plant leaves under steady-state conditions (Dongmann et al., 1974; Farquhar and Cernusak, 2005). However, the C–G model fails to explain the intra-leaf heterogeneity of  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  (Cernusak et al., 2016; Liu et al., 2021b), which is currently described using a two-pool model (Leaney et al., 1985; Song et al., 2015) and/or an advection–diffusion model, as the *Péclet* effect (Farquhar and Lloyd, 1993; Farquhar and Gan, 2003). Subsequently, more complicated models have been developed to cover non-steady-state conditions (Ogée et al., 2007). These models emphasize a mechanistic understanding of leaf water isotopic fractionation, but the relevant parameters cannot be strictly constrained or precisely

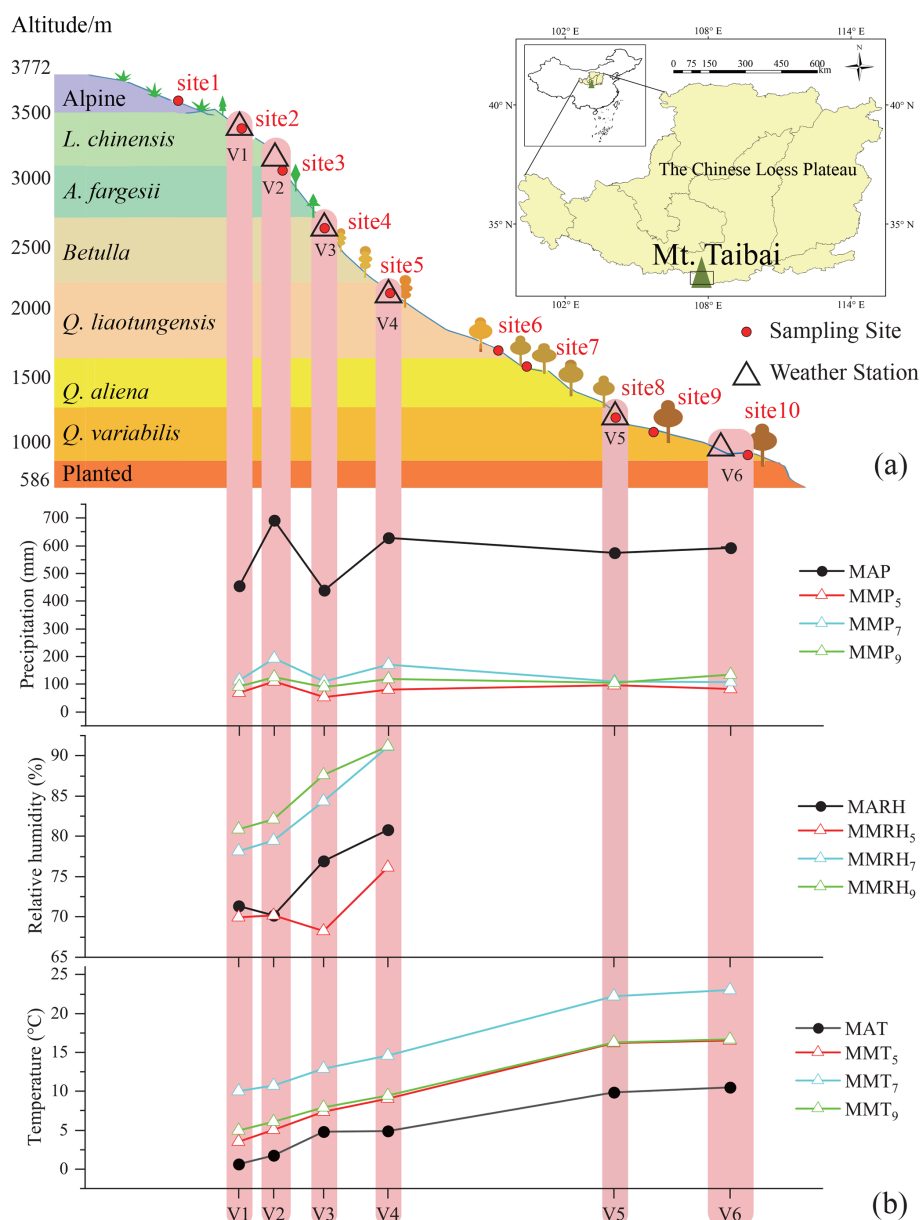
monitored, which hinders the use of these models under natural conditions (Plavcová et al., 2018).

This study combined the effects of measured source water isotopes and C–G model-predicted transpiration on  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values. Our objectives were to deepen the understanding of the controls on the  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  across different seasons. Based upon these objectives, we repeatedly sampled soils, twigs, and leaves in May, July, and September (representing spring, summer, and fall, respectively) from the same 10 plots distributed along an elevation transect. Simultaneously, we obtained the relevant meteorological parameters (e.g., temperature, relative humidity, and precipitation) from sites close to the sampling plots along the transect and used these to predict the  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values. The combined analysis of concurrent measurements of  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values in soil water, twig water, and leaf water with the predicted  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values of leaf water from the C–G model associated with the surrounding meteorological parameters will help to identify the factors that control  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values. Furthermore, we performed an isotope-based line analysis of the dual  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values of leaf water, associated with altitude and seasonality. This study will improve our understanding of the environmental signals preserved within the  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values extracted from plant organic biomarkers associated with leaf water.

## 2 Materials and methods

### 2.1 Study area

The Qinling Mountains form the dividing line between northern and southern China and mark the boundary between the watersheds of the Yellow and Yangtze rivers. Mt. Taibai (Fig. 1; 33.96° N, 107.77° E) rises to 3767 m a.s.l. (above sea level) and is the peak in the Qinling Mountains; it has a warm temperate ecosystem characterized by a rich diversity of flora and fauna. The mean annual temperature at the bottom of Mt. Taibai is 12.9 °C, and the mean annual precipitation is 609.5 mm (Zhang and Liu, 2010). The climate, soil, and vegetation vary significantly along our slope transect, exhibiting a remarkable vertical geo-ecological zonation (Fig. 1). The area contains a variety of climate zones: warm temperate (< 1300 m a.s.l.), temperate (1300–2600 m a.s.l.), cool temperate (2600–3350 m a.s.l.), and alpine (> 3350 m a.s.l.). The soil types vary from yellow loess soil at low elevations, spectacular rocky outcrops at middle elevations, and glacial remnants at high elevations. Vegetation along the transect is mainly coniferous and broadleaf forests, as well as alpine and subalpine vegetation (Fig. 1; Liu, 2021). The dominant species range from *Quercus variabilis*, *Q. aliena*, *Betula albosinensis*, *B. utilis*, *Abies fargessi*, and *Larix chinensis* forests to *Rhododendron clementinae* and *R. concinnum* alpine (Fig. S1 in the Supplement).



**Figure 1.** Sample sites (red dots) and weather stations (open triangles) that are distributed along vertical vegetation zones across the Mt. Taibai transect on the Chinese Loess Plateau (a). The meteorological parameters (precipitation, temperature, and relative humidity) vary with stations along the elevation transect (b). Mean annual (MAP, MAT, MARH) and monthly (MMP, MMT, MMRH) precipitation, temperature, and relative humidity. The subscripts refer to the month. The vertical vegetation distribution was adopted from Liu (2021).

## 2.2 Sampling strategy

Plants and soils were sampled in May, July, and September 2020, and samples were collected from 10 plots (3 × 3 m) covering all the vegetation zones along the northern slope of Mt. Taibai, extending from 608 to 3533 m a.s.l. (Fig. 1). Among the plots, six sites (i.e., sites 2, 3, 4, 5, 8, and 10; Fig. 1) were selected as being the closest to the weather stations along the elevation transect, and they were used in order to obtain the in situ meteorological data for analysis. For the

plants, one or two dominant deciduous and coniferous trees were chosen in each plot across the vegetation zone (Fig. S1). Several large leaves and suberized twigs were collected for each species. Between 3 to 10 large leaves were chosen for sampling, and a small number was collected in broadleaf forests and a large number in coniferous forests, depending on leaf size. The leaf samples were conducted in the context of the intact leaves because of the likely isotopic gradients within a leaf (Helliker and Ehleringer, 2000; Liu et al., 2016). Our sampling period was between 12:00 and 15:00 CST, be-

cause maximum diurnal enrichment of the leaf water isotopic composition occurs during this part of the day (Romero and Feakins, 2011; Liu et al., 2021). The twigs were collected simultaneously by cutting suberized twigs, and all of the twigs were cut into samples that were 3–4 cm long. The leaf and twig samples were immediately placed into glass vials with screw caps and sealed with polyethylene parafilm. For the soils, three surface soil samples (less than 10 cm deep) were collected from around the sampled plants using a small metal scoop at each plot. All sampling plots were located on slopes far from rivers and surface water bodies, which ensured that the soil water in each plot was derived exclusively from precipitation. Although the surface soil layers were collected only as the representatives of soil water in this study, these samples could provide a relatively good source of water for the plants, as supported by a prior study conducted along the same elevation transect (Zhang and Liu, 2010). The soil samples were tightly sealed in a polyethylene zipper bag on-site. All plant and soil samples were stored in a cool box ( $\sim 4^\circ\text{C}$ ) in the field and immediately transported to the laboratory. The altitude of each plot was determined using a handheld GPS unit with an error of  $\pm 5$  m.

### 2.3 Isotope analysis

The water in the plant and soil samples was extracted using an automatic cryogenic vacuum extraction system (LI-2100 Pro, LICA United Technology Limited, Beijing, China). The auto-extraction process was set for 3 h, and the extraction rate of water from the samples was more than 98 %. The isotopic composition of the soil water was measured using a Picarro L2130-i isotope water analyzer (Sunnyvale, CA, USA) at the State Key Laboratory of Loess and Quaternary Geology, Institute of Earth Environment, Chinese Academy of Sciences. The analytical accuracies were  $\pm 0.1\text{‰}$  for  $\delta^{18}\text{O}$  and  $\pm 1\text{‰}$  for  $\delta^2\text{H}$ . An isotope ratio mass spectrometer was coupled to a high-temperature conversion elemental analyzer (HT2000 EA-IRMS, Delta V Advantage; Thermo Fisher Scientific, Inc. USA) to take isotopic measurements of twig and leaf water at the Huake Precision Stable Isotope Laboratory on the campus of Tsinghua Shenzhen International Graduate School. The measurement precisions were  $\pm 0.2\text{‰}$  and  $\pm 1\text{‰}$  for  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$ , respectively. The isotopic composition of  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  is expressed as an isotopic ratio:

$$\delta_{\text{sample}}(\text{‰}) = \left( \frac{R_{\text{sample}} - R_{\text{standard}}}{R_{\text{standard}}} \right) \times 1000, \quad (1)$$

where  $\delta_{\text{sample}}$  represents  $\delta^{18}\text{O}$  or  $\delta^2\text{H}$ , and  $R_{\text{sample}}$  and  $R_{\text{standard}}$  indicate the ratio of  $^{18}\text{O}/^{16}\text{O}$  or  $^2\text{H}/^1\text{H}$  of the sample and standard, respectively. The  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values are reported relative to the Vienna Standard Mean Ocean Water (VSMOW). In addition, the mean monthly  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values of precipitation were determined using the Online Iso-

topes in Precipitation Calculator (Bowen and Revenaugh, 2003).

### 2.4 Modeling isotopes of leaf water

The C–G equation can be approximated as follows (Cernusak et al., 2022):

$$\delta_e = \delta_s + \varepsilon^+ + \varepsilon_k + (\delta_v - \delta_s - \varepsilon_k) \times \frac{e_a}{e_i}, \quad (2)$$

where  $\delta_e$  is the predicted  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values at the evaporative sites within leaves,  $\delta_s$  is the  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values of source water (equivalent to twig water in our study),  $\varepsilon^+$  is the equilibrium fractionation between liquid water and vapor, and  $\varepsilon_k$  is the kinetic fractionation during the diffusion of vapor through the stomata and the boundary layer.

In our analysis, we calculated  $\Delta_v$  (the enrichment of atmospheric vapor relative to source water) as  $\Delta_v = (\delta_v - \delta_s)/(1 + \delta_s)$ , and the values of  $\Delta_v$  are often close to  $-\varepsilon^+$  at the isotopic steady state (Barbour, 2007; Cernusak et al., 2016); therefore, we can calculate  $\delta_v$  as  $\delta_v = -\varepsilon^+ + (1 - \varepsilon^+)\delta_s$ . In addition,  $\frac{e_a}{e_i}$  is the ratio of the water vapor pressure fraction in the air relative to that in the intercellular spaces and is equal to the relative humidity (RH) in the air at steady state (Cernusak et al., 2022). Thus, Eq. (2) can be derived as

$$\delta_e = (1 - h)(\varepsilon^+ + \varepsilon_k) + (1 - \varepsilon^+h)\delta_s, \quad (3)$$

where  $\delta_s$  represents the isotopic values of twig water, and  $h$  is the mean annual or monthly RH (MARH or MMRH) in this study. The equilibrium fractionation ( $\varepsilon^+$ ) varies as a function of temperature (Bottinga and Craig, 1969) and can be equated to  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$ , as follows (Majoube, 1971):

$$\varepsilon_{\text{O}}^+(\text{‰}) = \left[ \exp \left( \frac{1.137}{(273 + T)^2} \times 10^3 - \frac{0.4156}{273 + T} - 2.0667 \times 10^{-3} \right) - 1 \right] \times 1000, \quad (4)$$

$$\varepsilon_{\text{H}}^+(\text{‰}) = \left[ \exp \left( \frac{24.844}{(273 + T)^2} \times 10^3 - \frac{76.248}{273 + T} + 52.612 \times 10^{-3} \right) - 1 \right] \times 1000. \quad (5)$$

The kinetic fractionation ( $\varepsilon_k$ ) can be calculated for  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  as (Farquhar et al., 2007; Cernusak et al., 2016)

$$\varepsilon_{\text{k}}^{\text{O}}(\text{‰}) = \frac{28r_s + 19r_b}{r_s + r_b}, \quad (6)$$

$$\varepsilon_{\text{k}}^{\text{H}}(\text{‰}) = \frac{25r_s + 17r_b}{r_s + r_b}, \quad (7)$$

where  $r_s$  and  $r_b$  are the resistances of the stomatal and boundary layers, respectively (i.e., the inverse of the conductance of the stomatal and boundary layers). Previous studies have found stomatal and boundary layer conductance values of 0.49 and  $2.85 \text{ mol m}^{-2} \text{ s}^{-1}$ , respectively (Cernusak et al., 2016; Munksgaard et al., 2017), resulting in  $\varepsilon_{\text{k}}^{\text{O}}$  and  $\varepsilon_{\text{k}}^{\text{H}}$  values of 26.7 and 23.8, respectively.

## 2.5 Statistical analysis

Statistical analysis (i.e., the mean, maximum, and minimum values, as well as the standard deviation) of the isotopes extracted from the precipitation, soil, twig, and leaf samples was performed to define the range and distribution of the  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values across the seasons. The Pearson correlation method was used to assess the correlations between the  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values among the different water types (i.e., precipitation, soil water, twig water, and leaf water). Hierarchical cluster analysis was used to show the relationships among  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values and potential source water isotopes ( $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values in precipitation, soil water, twig water, and leaf water), and meteorological parameters such as mean annual and monthly precipitation (MAP and MMP), mean annual and monthly temperature (MAT and MMT), and mean annual and monthly relative humidity (MARH and MMRH). A one-way analysis of variance (ANOVA) combined with a post hoc Tukey's least significant difference (LSD) test were performed to identify the significant differences in the isotopic compositions of precipitation, soil, twig, and leaf waters across the months. Comparisons of the relationships of  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  in the soil and leaf water were performed using covariance analysis (ANCOVA) to compare slopes across months. The structural equation model (SEM) was used to explain the respective effects of source waters (i.e., twig water, soil water, and precipitation) and meteorological parameters (i.e., temperature, precipitation, and RH) on  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values. The validated SEMs generated a good model fit, as indicated by a non-significant  $\chi^2$  test ( $p > 0.05$ ), a high comparative fit index ( $\text{CFI} > 0.95$ ), and a low root mean square error of approximation ( $\text{RMSEA} < 0.05$ ). A special SEM was constructed based on the Mantel  $R$  values in AMOS (version 24.0.0). Moreover, we used the Hybrid Single-Particle Lagrangian Integrated Trajectory (HYSPPLIT) model (Draxler and Rolph, 2003) to calculate air mass back trajectory for the central site ( $34.13^\circ\text{N}$ ,  $107.83^\circ\text{E}$ ,  $2270\text{ m a.s.l.}$ ) in the study area. These trajectories were initiated four times daily (at 00:00, 06:00, 12:00, and 18:00 CST), and their air parcel was released at  $2300\text{ m a.s.l.}$  for May, July, and September 2020 and moved backwards by winds for 120 h (5 d).

## 3 Results

### 3.1 Differing response of $\delta^{18}\text{O}$ and $\delta^2\text{H}$ values of leaf water

The measured  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values of leaf water responded differently to source water isotopes (Fig. 2a) and meteorological parameters (Fig. 2b) across the seasons. The leaf water  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values ( $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$ ) were clustered with those of the twig water ( $\delta^{18}\text{O}_{\text{twig}}$  and  $\delta^2\text{H}_{\text{twig}}$ ; Fig. 2a)

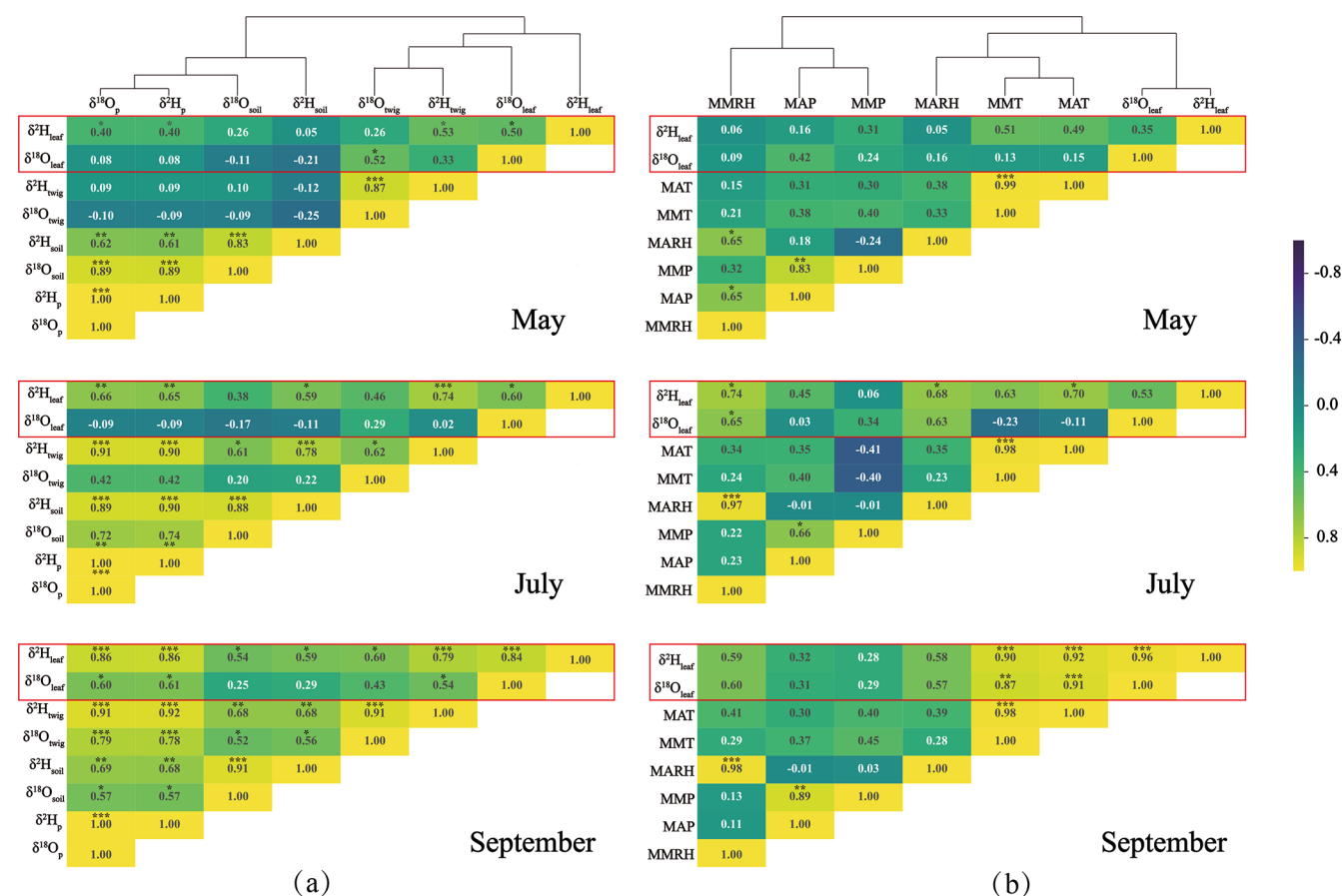
and with MARH, MAT, and MMT (Fig. 2b). The  $\delta^2\text{H}_{\text{leaf}}$  values were more closely correlated with isotopes of the potential source waters (e.g., twig water, soil water, and precipitation) than the  $\delta^{18}\text{O}_{\text{leaf}}$  values in different months (Fig. 2a). In contrast, leaf water  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values were correlated with meteorological parameters (Fig. 2b) throughout the study period. These correlations were more significant in summer (July) and autumn (September) than in spring (May).

### 3.2 Comparisons of measured and predicted $\delta^{18}\text{O}$ and $\delta^2\text{H}$ values of leaf water

The  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values predicted by the C–G model were compared with the measured  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values across all three months (Fig. 3). The C–G model explained 49 % and 70 % of the observed variations in the  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values, respectively (Fig. 3a and c). The slopes of the relationships for both  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values of leaf water were less than one, which suggests that part of the bulk leaf water is derived from unenriched vein water. However, there were no significant differences in  $\delta^{18}\text{O}_{\text{leaf}}$  ( $p = 0.54$ ; Fig. 3b) and  $\delta^2\text{H}_{\text{leaf}}$  values ( $p = 0.93$ ; Fig. 3d) between the C–G model predicted values and the measured values.

### 3.3 Variations of $\delta^{18}\text{O}$ and $\delta^2\text{H}$ values of different waters with seasons and altitude

There was a significant correlation between  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values ( $R^2 = 0.81$ ,  $p < 0.01$ ; Fig. 4), with significant clusters of  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values across the months, and values were higher in May, intermediate in July, and lower in September (Fig. 4). Within each month, the  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values were depleted in  $^2\text{H}$  and  $^{18}\text{O}$  at higher altitudes relative to lower altitudes. Likewise, the potential types of source water (i.e., twig water, soil water, and precipitation) exhibited consistent variations across the months, showing values that were relatively higher in May, intermediate in July, and lower in September (Fig. S1). The correlations between  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values among the source waters were also significant (Fig. S2). Nevertheless, the slopes and coefficients of determination ( $R^2$ ) between the  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values showed a decrease for precipitation, soil water, twig water, and leaf water from the three sampling months, except for soil water in May (Fig. S2). In addition, the ANCOVA showed no significant differences for the regression lines for precipitation ( $\text{df} = 0.47$ ,  $F = 2.49$ ,  $p = 0.11 > 0.05$ ), twig water ( $\text{df} = 53.2$ ,  $F = 0.42$ ,  $p = 0.66 > 0.05$ ), and leaf water ( $\text{df} = 437.3$ ,  $F = 2.78$ ,  $p = 0.08 > 0.05$ ) across the study months but a significant difference for soil water across the months ( $\text{df} = 308.8$ ,  $F = 10.9$ ,  $p < 0.05$ ).



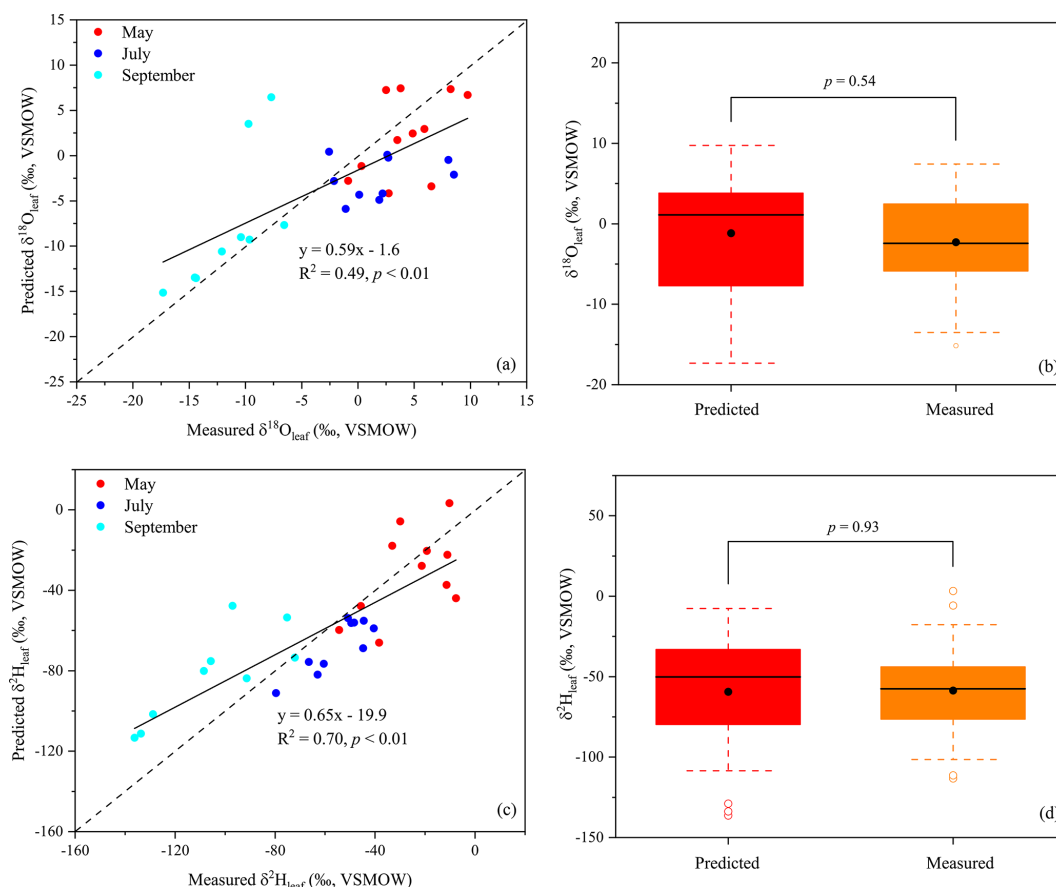
**Figure 2.** Heat maps of correlations ( $r$ ) between leaf water  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values and potential source water  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values (twig water, soil water, and precipitation  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values; a), and meteorological parameters (e.g., MAP, MMP, MAT, MMT, MARH, MMRH). The hierarchical cluster analysis of the isotopes of leaf water and source water (a) and meteorological parameters (b). The subscripts (p, soil, twig, leaf) refer to precipitation, soil water, twig water, and leaf water. \* Corrected significance at  $p < 0.05$ , \*\* corrected significance at  $p < 0.01$ , and \*\*\* corrected significance at  $p < 0.001$ .

## 4 Discussion

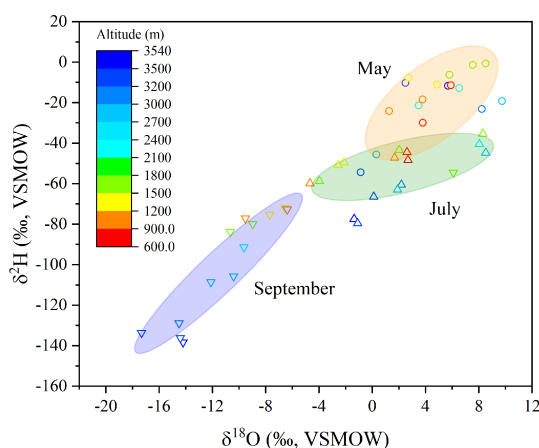
### 4.1 $\delta^{18}\text{O}$ and $\delta^2\text{H}$ values of leaf water

A recent global meta-analysis indicated that  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values reflect environmental drivers differently and showed that  $\delta^2\text{H}_{\text{leaf}}$  values more strongly reflect xylem water and atmospheric vapor  $\delta^2\text{H}$  values, whereas  $\delta^{18}\text{O}_{\text{leaf}}$  values more strongly reflect air relative humidity (Cernusak et al., 2022). Seasonal and localized observations along an elevation transect on the Chinese Loess Plateau supported these different responses of  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  to the isotopic composition of the source water and meteorological conditions (Fig. 2). This is likely due to the fact that variation in precipitation isotopic values compared with that in leaf water evaporative enrichment is larger for  $\delta^2\text{H}_{\text{leaf}}$  than  $\delta^{18}\text{O}_{\text{leaf}}$  (Cernusak et al., 2022). In addition, we found stronger correlations between  $\delta^2\text{H}_{\text{leaf}}$  and isotope values of the source water (twig water, soil water, and precipitation) than between  $\delta^{18}\text{O}_{\text{leaf}}$  values and the source water isotope values

(Fig. 2a). This is consistent with the global meta-analysis results (Cernusak et al., 2022). However, our localized study did not show a significantly different response of  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values to meteorological parameters, which responded at almost equivalent magnitudes (Fig. 2b). These observations suggest that plant organic isotopic proxies such as leaf wax (Sachse et al., 2012; Liu et al., 2016) and cellulose (Barbour, 2007; Lehman et al., 2017), which originate from  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values, can provide comparative information that indicates climatic signals (e.g., temperature, RH, and precipitation) in natural archives. These results argued with the recent global meta-analysis that  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values reflect climatic parameters (i.e., RH and temperature) differently (Cernusak et al., 2022). The stronger correlations for  $\delta^2\text{H}_{\text{leaf}}$  values than  $\delta^{18}\text{O}_{\text{leaf}}$  values with isotopic values of the source water were likely, because the  $\delta^2\text{H}_{\text{leaf}}$  values are ultimately determined only by precipitation  $\delta^2\text{H}$  (Sachse et al., 2012; Liu et al., 2016), whereas the  $\delta^{18}\text{O}_{\text{leaf}}$  values are affected by a mixture of precipitation



**Figure 3.** Measured leaf water isotopic composition for  $\delta^{18}\text{O}$  (a) and  $\delta^2\text{H}$  (c) values against values predicted by the C–G model. Boxplots show no significant differences for  $\delta^{18}\text{O}$  (b) and  $\delta^2\text{H}$  (d) values between measured and predicted leaf water. The dotted lines show one-to-one lines.



**Figure 4.** Correlation of leaf water  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values across months and altitude. Leaf water  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values were higher in May, intermediate in July, and lower in September, and while within each month, those isotopic values were relatively lower at high altitudes and higher in lower altitudes.

$\delta^{18}\text{O}$  and atmospheric factors ( $\text{O}_2$  and  $\text{CO}_2$ ) (Barbour, 2007; Cernusak et al., 2016). However, the comparative responses of both  $\delta^2\text{H}_{\text{leaf}}$  and  $\delta^{18}\text{O}_{\text{leaf}}$  values to climatic parameters were probably due to the same conditions surrounding the leaf.

The results of the cluster analysis showed that the isotope values of leaf water ( $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$ ) and twig water ( $\delta^{18}\text{O}_{\text{twig}}$  and  $\delta^2\text{H}_{\text{twig}}$ ) were clustered into one group, but those of soil water ( $\delta^{18}\text{O}_{\text{soil}}$  and  $\delta^2\text{H}_{\text{soil}}$ ) and precipitation ( $\delta^{18}\text{O}_{\text{p}}$  and  $\delta^2\text{H}_{\text{p}}$ ) were clustered into another (Fig. 2a). This indicates that the direct source water of  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  should be  $\delta^{18}\text{O}_{\text{twig}}$  and  $\delta^2\text{H}_{\text{twig}}$ , providing the source water isotope basis for the C–G model. In the C–G model (see Eq. 2), besides the source water isotopes, the equilibrium fractionation factor ( $\epsilon^+$ ) and atmospheric vapor enrichment ( $\Delta_{\text{v}}$ ) depend on the temperature at the isotopic steady state. Thus, the  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values were predicted to be associated primarily with temperature, RH, and source water, which is consistent with the results from the cluster analysis that the  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values were clustered with temperature (MAT and MMT) and RH (MARH; Fig. 2b). Based on the C–G model, we plotted the measured



and predicted  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values (Fig. 3a and c) and observed no significant differences between them (Fig. 3b and d). This is because our three-repeated samplings occur during the day when leaf water is generally near an isotopic steady state when chloroplasts are mostly located near the evaporative sites (Cernusak et al., 2016). The non-steady-state effects on leaf water isotopes were expected at night because of low stomatal conductance (Cernusak et al., 2005, 2016; Cuntz et al., 2007). Although the slopes of the predicted and measured  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values were less than one, the C–G model still provides a reasonable framework for guiding the analysis of the different controls on  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values.

#### 4.2 Dual $\delta^{18}\text{O}$ and $\delta^2\text{H}$ plots of leaf water

There was a significant linear correlation between the  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values, with remarkable clusters associated with the three months studied (Fig. 4). As is well known, the local meteoric water line (LMWL), generated by precipitation  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values at the local scale, serves as an important reference line for intercomparisons of different waters. Furthermore, the regression lines of the  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values from soil water, twig water, and leaf water (Fig. S2) suggest that the leaf water isotopes could well inherit isotopic signals of source waters that originate from twig water, soil water, and ultimately precipitation. The slopes and intercepts of the  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values decreased significantly from precipitation, soil water, twig water, and leaf water for each month, except for soil water in May (Fig. S2). Such patterns have been observed in many previous calibration studies (Brooks et al., 2010; Evaristo et al., 2015; Sprenger et al., 2016, 2017; Wang et al., 2017; Benettin et al., 2018; Barbeta et al., 2019; Penna and Van Meerveld, 2019; Liu et al., 2021a, 2022). The slopes of the LMWLs were lower in July (6.79) than in May (7.04) and September (6.85) but were not significantly different (ANCOVA test:  $\text{df} = 0.47$ ,  $F = 2.49$ ,  $p = 0.11 > 0.05$ ). This suggests that the local water vapor from precipitation was derived from the same source across the seasons but was subject to different intensities of evaporation as the temperature changed throughout the seasons (Li et al., 2019; Wu et al., 2019, 2021). The slopes of the  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values from the soil, twig, and leaf waters were much smaller than the LMWLs across the study months due to the secondary evaporation in the other water types.

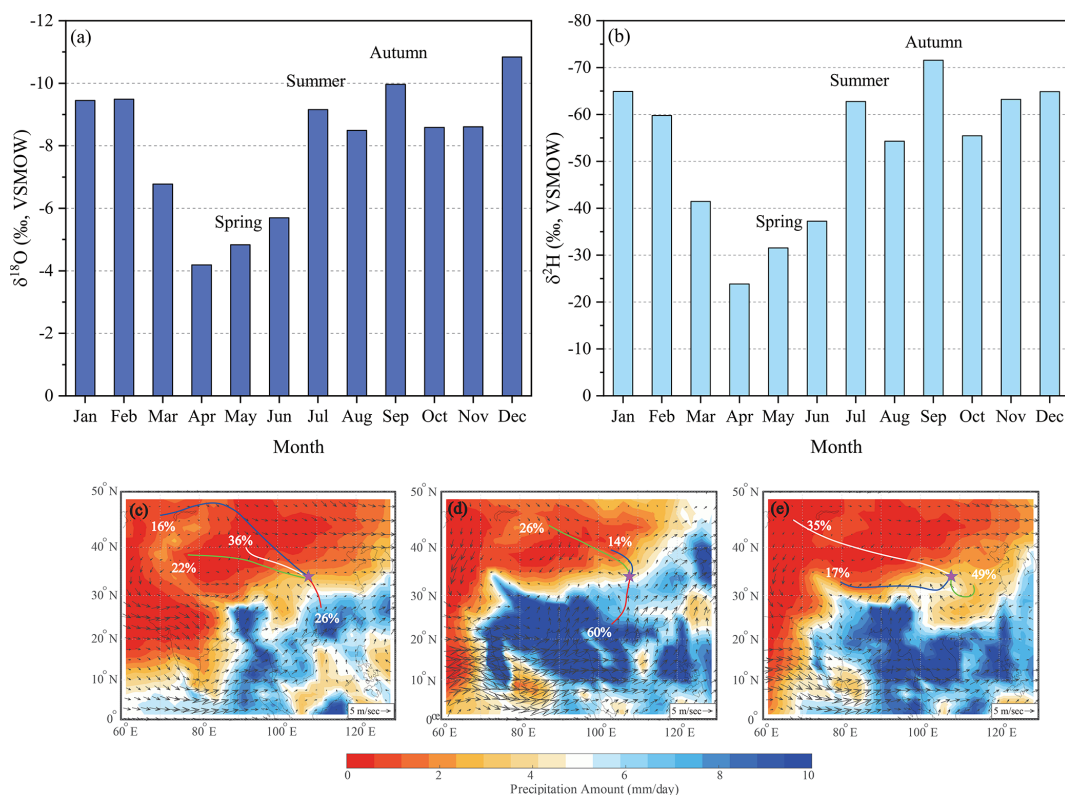
In the dual isotope plot of leaf water, there were well-defined clusters of  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values across the three months:  $^{18}\text{O}$  and  $^2\text{H}$  were depleted in September, there were intermediate values in July, and  $^{18}\text{O}$  and  $^2\text{H}$  were enriched in May (Fig. 4). When focusing on each month, relatively higher isotopic values occurred at low elevations, but lower isotopic values were present at high elevations despite there being no, or only weak, correlations between the  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values and altitude (Fig. S3). The correlations between the  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values and altitude,

and between the  $\delta^{18}\text{O}_{\text{twig}}$  and  $\delta^2\text{H}_{\text{twig}}$  values and altitude, were not significant across the three months; however, the  $\delta^{18}\text{O}_{\text{p}}$  and  $\delta^2\text{H}_{\text{p}}$ , and also the  $\delta^{18}\text{O}_{\text{soil}}$  and  $\delta^2\text{H}_{\text{soil}}$  values, were significantly correlated with altitude (Fig. S3), indicating that besides source water (precipitation and soil water), other factors associated with plants also affect  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values.

The dual isotope plot of  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values shows a significant isotope line,  $y = 4.52x - 50.7$  ( $R^2 = 0.81$ ,  $p < 0.01$ ; Fig. 4), but shallower slopes (3.53, 1.86, and 2.81 in May, July, and September, respectively) of  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values were observed across the seasons (Fig. S2). Such a correlation was supported by a recent study that conducted consecutive measurements of  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values in xylem and leaf water in Switzerland and indicated that leaf water provided great potential to determine the source water of plants (Benettin et al., 2021). Our study showed remarkable clusters in the measured (Fig. 4) and the C–G model-predicted (Fig. 3)  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values across the months and the consistencies of respective  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values with potential source water isotopes across months (Fig. S1). These findings of temporally consistent dynamics among the water types (i.e., precipitation, soil water, twig/stem water, and leaf water) have been observed in a number of previous studies (Phillips and Ehleringer, 1995; Cernusak et al., 2005; Sprenger et al., 2016; Berry et al., 2017; Liu et al., 2021a).

The isotopic inheritance from precipitation to leaf water indicates that seasonal variations of  $\delta^{18}\text{O}_{\text{p}}$  and  $\delta^2\text{H}_{\text{p}}$  values are the first-order control on the temporal patterns observed in leaf water. The seasonal dynamics of the  $\delta^{18}\text{O}_{\text{p}}$  and  $\delta^2\text{H}_{\text{p}}$  values reflect the combined effects of factors such as temperature, altitude, and precipitation amount, which are associated with orographic conditions, as well as sub-cloud evaporation, moisture recycling, and differences in the vapor source (Dansgaard, 1964; McGuire and McDonnell, 2007; Li et al., 2016; Penna and Van Meerveld, 2019; Wu et al., 2019). In this study, we used the HYSPLIT model to demonstrate the ultimate cause of the seasonal variations of  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values, that is, the monthly dynamics of the  $\delta^{18}\text{O}_{\text{p}}$  and  $\delta^2\text{H}_{\text{p}}$  values. The monthly variations of the  $\delta^{18}\text{O}_{\text{p}}$  and  $\delta^2\text{H}_{\text{p}}$  values from the Global Network for Isotopes in Precipitation (GNIP, <http://www.iaea.org/>, last access: 18 April 2021) at Xi'an station (AD 1985–1992), which is  $\sim 100$  km from our study transect, were enriched in  $^{18}\text{O}$  and  $^2\text{H}$  in May relative to July and September (Fig. 5a and b). The cluster mean of the moisture transport routes from HYSPLIT (Draxler and Rolph, 2003) and the climatological 850 hPa wind vectors showed that the primary moisture sources were from western China and central Asia in May, the China–India Peninsula and the Bay of Bengal, and local moisture recycling and convection (Fig. 5c–e). The seasonal variations in  $\delta^{18}\text{O}_{\text{p}}$  and  $\delta^2\text{H}_{\text{p}}$  values are consistently related to the onset, advancement, and retreat of the Asian summer monsoon and associated changes in the large-scale mon-





**Figure 5.** Variation of monthly mean precipitation  $\delta^{18}\text{O}$  (a) and  $\delta^2\text{H}$  (b) values at Xi'an station from the Global Network of Isotopes in Precipitation (GNIP) and cluster mean of moisture transport routes using the HYSPLIT model in May (c), July (d) and September (e), 2020. The background in (c)–(e) is the average precipitation ( $\text{mm d}^{-1}$ ) and 850 hPa wind vectors (arrows,  $\text{m s}^{-1}$ ) in May (c), July (d), and September (e) in AD 1979–2016 based on the database of the Global Precipitation Climatology Center (GPCC) (Becker et al., 2011) and the Modern-Era Retrospective analysis for Research and Applications (Rienecker et al., 2011).

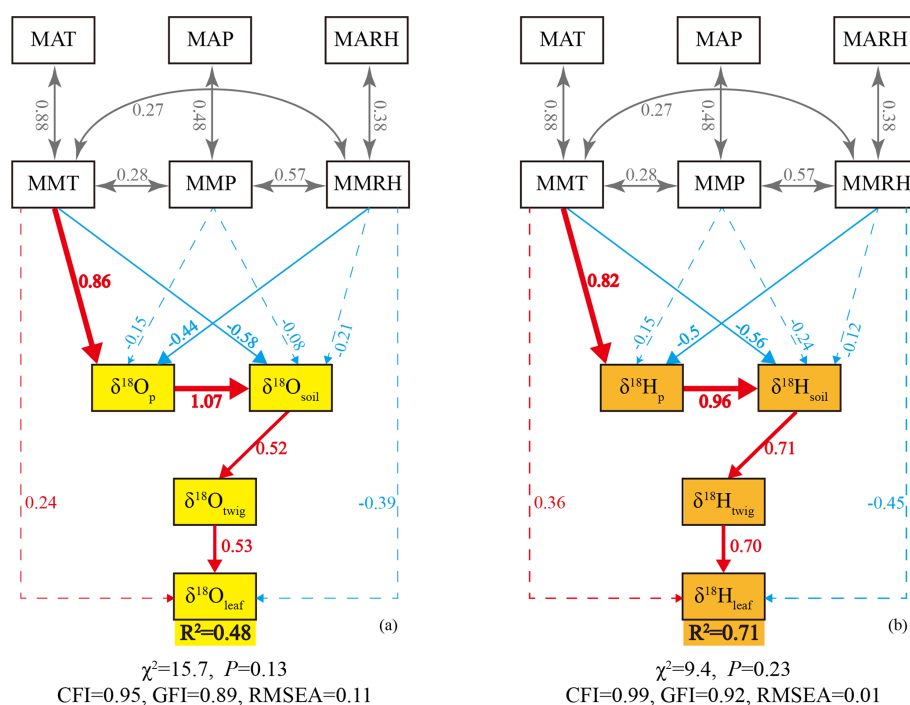
soon circulation (e.g., Zhang et al., 2020, 2021). As the summer monsoon starts in mid-May, the rainfall season starts in southern China; however, our study area is controlled mainly by moisture from the westerlies (Chiang et al., 2015) with relatively higher vapor,  $\delta^{18}\text{O}_\text{p}$ , and  $\delta^2\text{H}_\text{p}$  values (Fig. 5a–c). In July, the summer monsoon reaches its strongest phase, and the rainfall belt shifts to central and northern China, where the southerly wind brings plenty of moisture from the China–India Peninsula and the Bay of Bengal with lower vapor,  $\delta^{18}\text{O}_\text{p}$ , and  $\delta^2\text{H}_\text{p}$  values (Fig. 5a, b, and d). When the summer monsoon withdraws in September, the study area is controlled mainly by local moisture recycling and convection (Fig. 5e). Soil water, stored after the June–August monsoon rainfall with its lower  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values, results in even lower  $\delta^{18}\text{O}_\text{p}$  and  $\delta^2\text{H}_\text{p}$  values in September than in July (Fig. S1), causing significantly lower  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values of leaf water (Fig. 4).

#### 4.3 Framework of controls for $\delta^{18}\text{O}$ and $\delta^2\text{H}$ values of leaf water

To delineate the mechanisms that control the  $\delta^{18}\text{O}_\text{leaf}$  and  $\delta^2\text{H}_\text{leaf}$  values, we used the SEMs to quantify the complex

interactions among  $\delta^{18}\text{O}_\text{leaf}$  or  $\delta^2\text{H}_\text{leaf}$  values, source waters, and meteorological parameters (Fig. 6). The coefficients of determination ( $R^2$ ) were 0.48 and 0.71 for the  $\delta^{18}\text{O}_\text{leaf}$  and  $\delta^2\text{H}_\text{leaf}$  values, respectively, indicating that the models explained more variance for  $\delta^2\text{H}_\text{leaf}$  values than  $\delta^{18}\text{O}_\text{leaf}$  values (Fig. 6). The SEMs showed that potential source waters (i.e., twig water, soil water, and precipitation) had stronger effects on  $\delta^2\text{H}_\text{leaf}$  relative to  $\delta^{18}\text{O}_\text{leaf}$  values, while the meteorological parameters showed weak effects on both  $\delta^{18}\text{O}_\text{leaf}$  and  $\delta^2\text{H}_\text{leaf}$  values (a little larger for  $\delta^2\text{H}_\text{leaf}$  than  $\delta^{18}\text{O}_\text{leaf}$  values). This is consistent with our above correlation analysis (Fig. 2). Surprisingly, the MMT had significant effects on  $\delta^{18}\text{O}_\text{p}$  and  $\delta^2\text{H}_\text{p}$  values, suggesting that temperature plays a key role in determining  $\delta^{18}\text{O}_\text{p}$  and  $\delta^2\text{H}_\text{p}$  values, but this finding is not discussed further here. Collectively, the SEMs also showed that source water exerts the first-order control but affects  $\delta^{18}\text{O}_\text{leaf}$  and  $\delta^2\text{H}_\text{leaf}$  differently; the meteorological parameters had a weak control on  $\delta^{18}\text{O}_\text{leaf}$  and  $\delta^2\text{H}_\text{leaf}$ , with a more substantial effect on  $\delta^2\text{H}_\text{leaf}$  than  $\delta^{18}\text{O}_\text{leaf}$  values.

A schematic representation of the controls on  $\delta^{18}\text{O}_\text{leaf}$  and  $\delta^2\text{H}_\text{leaf}$  values (respective and dual) is shown in Fig. 7. It involves multiple processes associated with the hydroclimatic and biochemical factors that affect  $\delta^{18}\text{O}_\text{leaf}$  and  $\delta^2\text{H}_\text{leaf}$  val-



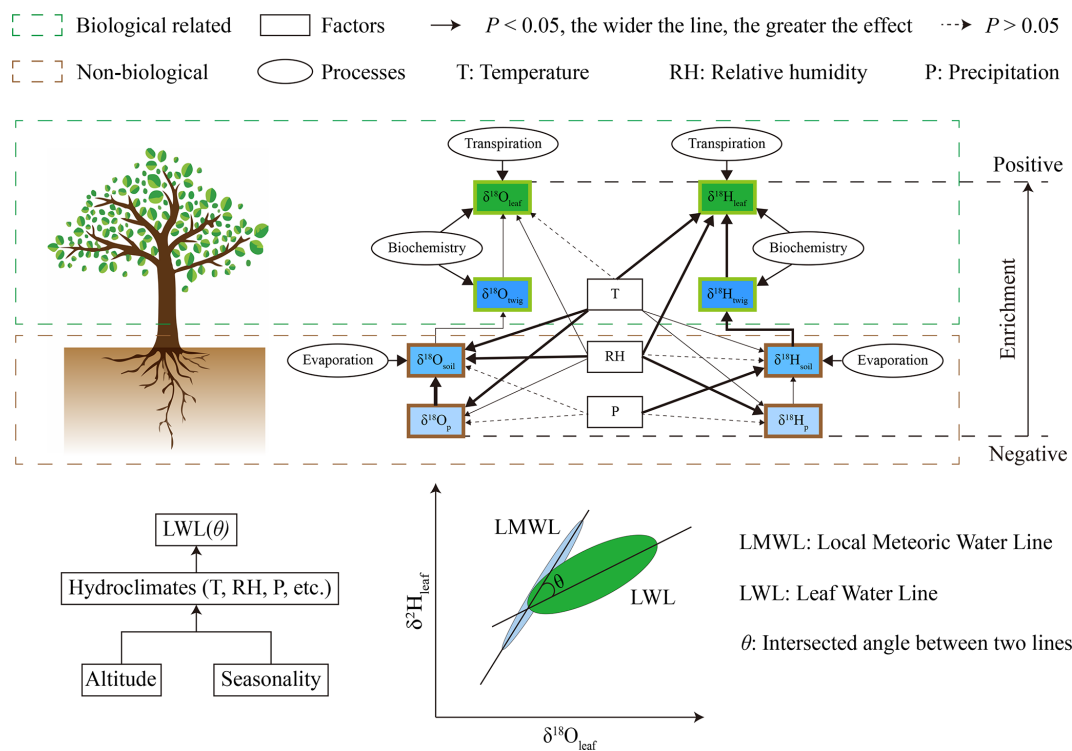
**Figure 6.** Structural equation model (SEM) of leaf water  $\delta^{18}\text{O}$  (a) and  $\delta^2\text{H}$  (b) values. The structural equation models considered all plausible pathways. Solid lines indicate significant positive (red) or negative (blue) effects, and dashed lines indicate non-significant effects. Grey lines indicate correlations between two variables. Numbers on the arrow indicate significant standardized path coefficients, proportional to the arrow width. The coefficients of determination ( $R^2$ ) represent the proportion of variance explained by the model.

ues. The meteorological parameters (temperature, RH, and precipitation) exerted distinct effects on the  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values of the source water and, thus, on the  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values, as demonstrated above by the SEM. Significant isotopic fractionation occurred mainly at two key locations across the vertical soil profiles and leaf architectures from precipitation to leaf water. First, an isotopic gradient across the vertical soil profile appeared because of evaporation from the surface soil layers (Ehleringer and Dawson, 1992; Goldsmith et al., 2012; Evaristo et al., 2015). This evaporative isotopic fractionation causes a linearly isotopic trajectory down the soil profile (Goldsmith et al., 2012; Rothfuss and Javaux, 2017; Wu et al., 2018; Wang et al., 2019; Amin et al., 2020; Zhao et al., 2020; Liu et al., 2021a). Second, there were significant isotopic heterogeneities because of transpiration associated with the  $\delta^{18}\text{O}_{\text{leaf}}$  (Helliker and Ehleringer, 2000; Farquhar and Gan, 2003; Gan et al., 2003; Song et al., 2015) and  $\delta^2\text{H}_{\text{leaf}}$  values (Šantrůček et al., 2007; Liu et al., 2016, 2021b) within a leaf, which depends substantially on veinal structures (Liu et al., 2021b). The within-leaf heterogeneity of the  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values can be explained using the *Péclet*-modified C–G model (Gan et al., 2003; Farquhar and Gan, 2003; Cernusak et al., 2005, 2016). Collectively, the soil evaporation and leaf transpiration produce isotopic enrichment above source water (precipitation or soil water). Soil evaporation leads to an isotopic gradi-

ent across the vertical soil profile, providing water sources for plant root uptake without isotope fractionation during the process (Dawson and Ehleringer, 1991; Ehleringer and Dawson, 1992; Chen et al., 2020). During water transport between roots and leaf petioles, isotopic compositions of xylem water remain unaltered from those in soils (i.e., soil immobile water), until it reaches the leaf, which undergoes water loss (Ehleringer and Dawson, 1992). Within the leaf, transpiration leads to significant isotopic enrichment (Helliker and Ehleringer, 2000; Liu et al., 2015; Cernusak et al., 2016), which is highly dependent on meteorological parameters (e.g., temperature and relative humidity). However, the meteorological parameters varied with altitude and seasonality, yielding an isotopic leaf water line (LWL) in the dual-isotope plot (Fig. 4). The LWL provides an important baseline for leaf-derived organic matter such as cellulose (e.g., Barbour, 2007; Lehman et al., 2017) and leaf wax (Liu et al., 2016, 2021). Overall, the LWL is controlled primarily by altitude and seasonality, as these are the main influencers of the hydroclimatic factors.

## 5 Conclusion

Along an elevation transect on the Chinese Loess Plateau, precipitation, soil water, twig water, and leaf water were repeatedly sampled to explore the controls on  $\delta^{18}\text{O}_{\text{leaf}}$  and



**Figure 7.** Schematics of the respective and dual isotopes of  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values from precipitation to leaf water, associated with physical (evaporation at soil profile and transpiration at leaf level) and biochemical processes. The dual isotopes of  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$  values yield an isotopic water line, the slope of which was lower than the LMWL. The intersected angle varied with hydroclimates, associated with altitude and seasonality.

$\delta^2\text{H}_{\text{leaf}}$  values associated with meteorological parameters and source water. The effects of meteorological parameters and source water on  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values were different, and the dual  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  plot generated an isotopic line. We found that  $\delta^2\text{H}_{\text{leaf}}$  values were more closely correlated with source water isotopes than  $\delta^{18}\text{O}_{\text{leaf}}$  values, whereas  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values were similarly correlated with meteorological parameters along the elevation transect. The observations suggest that plant organic isotopic proxies such as leaf wax and cellulose originating from  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values can provide comparative climatic information on the Loess Plateau of China. Additionally, the dual-isotope analysis showed that the  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values were closely correlated because of their similar altitudinal and seasonal responses. The first-order control on  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values was the source water (i.e., precipitation), and the meteorological parameters had a comparable effect on both  $\delta^{18}\text{O}_{\text{leaf}}$  and  $\delta^2\text{H}_{\text{leaf}}$  values, which varied with altitude and season across the transect on the Loess Plateau. In the future, we will investigate the relationships of intersection angle  $\theta$  with hydroclimatic and biochemical factors.

**Data availability.** The data can be made available by contacting the corresponding author.

**Supplement.** The supplement related to this article is available online at: <https://doi.org/10.5194/hess-27-599-2023-supplement>.

**Author contributions.** JL conceived the idea of the research and performed the data analysis. JL, HW, and HZ wrote the manuscript. LG and YZ edited the paper. JL and CJ performed the lab work. All authors contributed to discuss the results.

**Competing interests.** The contact author has declared that none of the authors has any competing interests.

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