Do Mature *Quercus aliena* Primarily Use Deep Soil Water?

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**Abstract:** Seasonal drought events induced by climate change have exacerbated the water deficit in trees. This is particularly pronounced in mature trees with considerable heights and extensive branches, where the long water transport distance leads to a reduction in hydraulic transportation efficiency. Therefore, investigating the water uptake patterns of mature trees is essential to enhance their growth and resilience to climate change. However, previous studies have predominantly focused on trees aged 10–60 years, with limited research on the water uptake patterns of trees over 60 years old. In this study, we employed hydrogen and oxygen stable isotopes coupled with the MixSIAR model to determine the water uptake patterns of mature *Quercus aliena* var. *acuteserrata* (40–60 years, 60–90 years, 120–150 years, >150 years) during the growing seasons of 2021 and 2022 in Baotianman Nature Reserve, Henan Province, China. Additionally, we utilized a random forest model to quantify the relative contributions of vegetation (fine root biomass) and soil properties (soil moisture, bulk density, total porosity, field capacity, and soil texture) to the water uptake patterns of *Q. aliena*. Our findings demonstrate that most mature *Q. aliena* predominantly extracted deep soil water (60–100 cm) in the early growing season, but shifted towards utilizing shallow soil water (0–40 cm) in the late growing season. The water uptake pattern of mature *Q. aliena* was primarily regulated by soil moisture. These results reveal that mature *Q. aliena* trees exhibit a flexible water use strategy, enabling them to cope with seasonal drought by altering the soil depth from which they extract water. In the future, in the process of the cultivation and conservation of mature oak trees, surface soil irrigation can be increased to prevent tree mortality resulting from water deficit during drought conditions.

**Keywords:** mature trees; stable isotopes; MixSIAR; water uptake; Quercus aliena

1. **Introduction**

Trees play a vital role in retaining water through root water uptake and trunk water storage [1–3]. Adequate water supply is essential for promoting tree growth during the growing season [4], particularly for mature trees with considerable heights and extensive branches, where the long water transport distance can reduce their hydraulic transportation efficiency [5]. In times of insufficient water, mature trees with lower hydraulic transport efficiency inevitably decrease stomatal conductance, ultimately inhibiting photosynthesis and growth [6,7]. Combined with the impact of frequent seasonal droughts triggered by climate change [8–11], the water deficit in mature trees is expected to become more pronounced. Therefore, exploring water uptake patterns in mature trees is crucial to enhance their growth and resilience to climate change.

Despite their significance, our understanding of water uptake patterns in mature trees is limited in three aspects. Firstly, previous studies predominantly focused on trees aged...
10–60 years [12,13]. However, trees over 60 or 100 years old have considerable economic and ecological value [14,15]. Therefore, studies on the water use patterns in trees aged over 60 years could provide a reliable basis for the conservation and restoration of mature trees. Unfortunately, experimental evidence in this domain is lacking.

Secondly, while studies have shown that young trees consistently absorb shallow soil water throughout the growing season [16–19], there is no consensus on the water use patterns in mature trees. Generally, as trees age, they develop deeper root systems to meet their growing water demands [20]. Mature trees primarily rely on deep soil water and groundwater due to their deep root systems [13,21,22]. For example, previous studies have illustrated that mature Populus euphratica (55–60 years, [23]), Pinus sylvestris (50–60 years, [17]), Quercus robur (80–85 years, [24]), Caragana korshins (8 years, [25]), and Tamarix chinensis (24 years, [26]) trees primarily took up deep soil water or groundwater.

In contrast, recent studies have reported that the water use patterns of some mature trees varied under different water conditions. Specifically, mature trees, such as P. sylvestris, Lithocarpus hancei, Castanopsis rufescens, and L. xylocarpus, mainly utilized shallow soil water in wet months, but changed to use deep soil water in dry months [19,27–29]. These inconsistent observations suggest that seasonal dynamics or water conditions may lead to differences in water uptake patterns in mature trees. Therefore, exploring water use patterns in mature trees on a seasonal scale is essential.

Lastly, the factors influencing water absorption in mature trees remain unclear. Soil properties and fine root biomass are possible factors influencing the water use patterns in tree species [30,31]. Soil with a low bulk density, high porosity, and field capacity can store more water for tree uptake [32,33], and potentially improve tree water absorption. In addition to the aforementioned factors, soil moisture is a crucial regulator of plant water uptake patterns, as plants prefer to take up water from water-saturated soil [34,35]. Soil texture is one of the most important physical attributes due to its influence on soil water movement and productivity [36]. Fine root biomass also exerts an important influence on plant water uptake, with a greater fine root biomass leading to a higher absorption rate of soil water by trees [37]. Particularly in arid or semi-arid conditions, the distribution of fine roots shifts to deeper layers, thereby increasing the absorption rate of deep soil water by trees [38,39]. However, it remains unclear whether these attributes influence water use patterns in mature trees.

To address the aforementioned issues, we selected mature Q. aliena forests (40–60 years, 60–90 years, 120–150 years, >150 years) in Baotianman Nature Reserve, Henan Province, China. Although the Nature Reserve preserves natural secondary forests with a long time span (40–150 years), no forest extinction phenomena have been observed. We employed hydrogen and oxygen stable isotopes coupled with the Mixture Stable Isotope Analysis in R (MixSIAR) model to analyze the water use pattern of Q. aliena in each forest. Furthermore, we evaluated soil factors and fine root biomass at each site to reveal the primary factors influencing the water use pattern of Q. aliena. This study aimed to test the following two hypotheses: (i) mature Q. aliena trees tend to take up deep soil water during the growing season; and (ii) fine root biomass and soil moisture jointly regulate the water use pattern of mature Q. aliena trees.

2. Materials and Methods

2.1. Study Area

The study area is situated in Baotianman National Nature Reserve in Neixiang County, Nanyang City, Henan Province, located on the southern slope of Mount Funiu (111°47′–112°04′ E, 33°20′–33°36′ N) (Figure 1). The area falls within a warm temperate to subtropical transition region with a continental monsoon climate. The mean annual temperature ranges from 11 °C to 14 °C. Precipitation occurs mainly from June to September, with the mean annual precipitation being 998 mm. The relative humidity and annual evaporation are 63% and 992 mm, respectively. The annual frost-free period lasts 160 days.
in high mountainous areas and 227 days in low mountainous areas. The dominant soil type is mountain brown soil [40–42].

Figure 1. Location of the study area, Baotianman forest, China.

The Baotianman forest is a typical natural secondary forest ecosystem in the transition zone from warm temperate to north subtropical. The vegetation zone is classified as a warm temperate deciduous broad-leaved forest. The growing season begins in May and ends in October. Oak trees, primarily distributed in the altitude range of 1300–1600 m, dominate the canopy. The sub-canopy layer of small trees at the study site includes *Cornus kousa* and *Cornus controversa*; the main shrub layer includes *Viburnum betulifolium*, and the herb layer includes *Carex siderosticta* and *Phlomis umbrosa*. We selected four *Q. aliena* forests of varying ages (40–60 years, 60–90 years, 120–150 years, >150 years) as study sites (Table 1).
Table 1. Basic overview of the four study sample sites.

<table>
<thead>
<tr>
<th>Sample Sites</th>
<th>Stand Age (Years)</th>
<th>Altitude (m)</th>
<th>Slope (°)</th>
<th>Stand Density (tree/hm²)</th>
<th>BDH (cm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>40–60</td>
<td>1386.0</td>
<td>17</td>
<td>1075</td>
<td>23.8</td>
</tr>
<tr>
<td>2</td>
<td>90–120</td>
<td>1369.6</td>
<td>12</td>
<td>692</td>
<td>27.2</td>
</tr>
<tr>
<td>3</td>
<td>120–150</td>
<td>1411.2</td>
<td>20</td>
<td>275</td>
<td>51.9</td>
</tr>
<tr>
<td>4</td>
<td>&gt;150</td>
<td>1365.9</td>
<td>15</td>
<td>62</td>
<td>87.0</td>
</tr>
</tbody>
</table>

Note: DBH represents diameter at breast height.

2.2. Samples Collection

The study was conducted during the growing season of 2021 and 2022 (May–October), including the early growing season (EGS, green value growth period; May–June), middle growing season (MGS, mature period; July–August), and late growing season (LGS, resulting period; September–October). In each month, samples of rainfall, stream water, groundwater, soil, and tree stem (xylem) were collected at each site.

Rainfall samples were collected after each rainfall event from three barrels randomly placed in open areas 200 m outside the forests. Subsequently, three rainfall samples were mixed completely for isotope analysis. Stream water and groundwater samples were collected simultaneously in the vicinity of the sample site (within 1–3 km) every 10 days.

For soil sampling, three 100 cm deep soil profiles were obtained at 0–20 cm, 20–40 cm, 40–60 cm, 60–80 cm, and 80–100 cm for hydrogen and oxygen isotope analysis.

Tree stem (xylem) samples were collected from three healthy trees with a similar ground diameter and crown width at each site. Tree stems (xylem) with diameters of 0.3–0.5 cm and lengths of 3–5 cm were collected from the sunny side of each tree. Then the epidermis and bast were removed from the tree stem.

All samples were collected between 8:00 a.m. and 10:00 a.m. and immediately placed into glass bottles, sealed with parafilm, and kept frozen (−20 °C) in the field.

2.3. Sample Pre-Treatment and Isotope Analysis

Soil and tree xylem water were extracted using a vacuum extraction apparatus (LICA, Beijing, China). An LGR instrument (Los Gatos Research, LWIA-45-EP, San Jose, CA, USA) with a measurement precision of <0.2‰ was employed for both δD and δ18O measurements [43]. The manufacturer’s post-processing software (LWIA Post-Analysis Software v4.5.0; LGR) was used for organic contaminant detection and spectral contamination correction in samples showing organic matter contamination [44]. Isotope ratios were expressed in per mil (‰) relative to V-SMOW (Vienna Standard Mean Ocean Water). The calculations were expressed as the following formula:

$$\delta = (\frac{R_{\text{sample}}}{R_{\text{standard}}} - 1) \times 1000\%$$

Where $R_{\text{sample}}$ is the the $^{2}$H/$^{1}$H and $^{18}$O/$^{16}$O molar ratios of the sample, and $R_{\text{standard}}$ is the V-SMOW.

2.4. Determination of Water Uptake Pattern

The Bayesian mixture model (MixSIAR), incorporating uncertainties associated with multiple sources and discrimination factors, was utilized to compute the proportion of the sources of water utilized by Q. aliena. The interface of the MixSIAR model consists of five modules, including "Read in data", "MCMC run length", "Error structure", "Specify prior", and "Diagnostics". In the "Read in data" module, the δD and δ18O values of tree stem (xylem) water were uploaded into the "Mixture data"; the mean and standard deviation values for δD and δ18O in each soil water layer (0–20 cm, 20–40 cm, 40–60 cm, 60–80 cm, and 80–100 cm) were introduced into "Source data"; and "Discrimination data" were set to 0, as these isotopes do not fractionate during water transport from root to xylem [45].
“MCMC (Markov chain Monte Carlo) run length”, “Error structure”, and “Specify prior” were set as “long”, “Residual only”, and “Uninformative prior”, respectively [37,46,47]. In the “Diagnostics” module, Gelman–Rubin and Geweke diagnostic tests were used to verify model convergence before accepting MixSIAR output. All analyses were conducted using the “MixSIAR” package in R software (version: 3.6.2).

2.5. Determination of Vegetation Biomass and Soil Physical Properties

To identify the factors influencing plant water uptake, we measured the fine root biomass (FB) of *Q. aliena* at each site. Fine roots (Ø ≤ 2 mm) were collected from 3–5 *Q. aliena* trees showing good growth in a 20 m × 20 m sample square, with roots obtained from each 20 cm soil depth segment of 100 cm. The roots were washed, dried to a constant weight, and their biomass was weighed [35]. Soil properties including soil moisture (SM), bulk density (BD), total porosity (TP), field capacity (FC), and soil texture were also measured to evaluate their impact on the water use pattern of *Q. aliena*. Soil samples were taken in situ at 20 cm intervals using 100 cm³ ring knives, sealed to prevent water loss, and analyzed in the laboratory for BD, TP, and FC. Another set of soil samples was collected in labeled aluminum boxes, and soil moisture was determined by oven drying at 105 °C for 24 h [48]. Soil texture was evaluated using a laser particle size analyzer (Microtrac S3500 SI, Microtrac Inc., Montgomeryville and York, PA, USA) [49].

2.6. Statistical Analysis

We employed One-way ANOVA and Duncan’s multiple comparison procedure to compare the soil moisture, fine root biomass, soil properties, and soil texture in each stand of *Q. aliena* (Figures S1 and S2; Table S1). Pearson’s correlation coefficient was used to determine the linear correlation between *Q. aliena* water uptake proportions and soil factors (soil moisture, soil properties, and soil texture) as well as plant factors (fine root biomass), the correlation coefficient for the sample is denoted by R², and the overall correlation coefficient is denoted by $P$. To identify the dominant factors influencing the percentage of water absorbed by *Q. aliena*, we conducted a random forest model using Python (version 3.11). The model solves the problem of multivariate covariance in general regression analysis, which is more conducive to calculating the nonlinear effects of variables. Meanwhile, the model also reflects the interactions between variables and is not sensitive to outliers. The percentage increases in the mean squared error (MSE) for each factor used to represent the relative importance of each predictor in the random forest model.

3. Results

3.1. Environmental Conditions

During the 2021 and 2022 growing seasons (May–October), the precipitation received totaled 1065.7 mm and 639.3 mm, respectively. Peak precipitation occurred in the middle growing season (MGE, July–August), representing 54.5% (580.4 mm) and 42.0% (268.5 mm) of the total rainfall for each respective year. Conversely, the lowest precipitation occurred in the early growing season (EGS, May–June), representing 14.3% (152 mm) and 24.5% (156.5 mm) of the total rainfall for each respective year. Additionally, the highest mean temperature was recorded in the MGE (Figure 2).
3.2. Relationship between Tree Xylem Water and Potential Water Sources

The isotopic signatures of $\delta D$ and $\delta^{18}O$ in groundwater and stream water were predominantly situated to the left of the local meteoric water line (LMWL), suggesting that they were mainly derived from precipitation and did not undergo evapotranspiration fractionation (Figure 3). In contrast, the $\delta D$ and $\delta^{18}O$ values of soil water and Q. aliena xylem water were situated on the right side of the LMWL, indicating that they were enriched by evaporation processes (Figure 3). Additionally, the $\delta D$ and $\delta^{18}O$ values of Q. aliena xylem water closely resembled those of soil water (Figure 3), suggesting that xylem water mainly originated from soil water.
3.3. Water Uptake Patterns of Q. aliena Trees

Based on MixSIAR results, mature Q. aliena trees primarily absorbed deep soil water (60–100 cm) in the EGS, but shifted to utilizing shallow soil water (0–40 cm) in the LGS, except for 40–60 year old Q. aliena trees in 2021 and 120–150 year old Q. aliena trees in 2022. Specifically, during the EGS of 2021, the proportions of water uptake from deep soil water were 66.5%, 64.0%, and 65.0% for 60–90, 120–150, and >150 year old Q. aliena trees, respectively (Figure 4). Similarly, in the EGS of 2022, the proportions of water uptake from deep soil water were 50.9%, 49.08%, and 69.3% for 40–60, 120–150, and >150 year old Q. aliena trees, respectively. However, during the LGS, 60–90, 120–150, and >150 year old Q. aliena trees mainly utilized shallow soil water (60.7%, 50.5%, and 46.8%, respectively) in 2021; and 40–60, 120–150, and >150 year old Q. aliena trees predominantly used shallow soil water (63.8%, 43.7%, and 55.0%, respectively) in 2022. Overall, most mature Q. aliena trees, especially in the late growing season, shifted towards utilizing shallow soil water (0–40 cm) in the late growing season. This finding is contrary to the conventional view that mature trees predominantly rely on deep soil water or groundwater.

![Proportion of water uptake at different growth stages in Q. aliena forests of different ages. EGS, early growing season (May–June); MGS, middle growing season (July–August); LGS, late-growing season (September–October).](image)

3.4. Relationship between Water Uptake Pattern of Q. aliena Trees and Plant or Soil Properties in the Growing Season

To determine the factors influencing the water use pattern of Q. aliena, we analyzed the relationship between its water uptake pattern and various soil/plant factors (Figures S1 and S2; Table S1; Figure 5). The results revealed that the proportion of water uptake by Q. aliena from the 0–60 cm layer was positively correlated with FB (p < 0.01), SM (p < 0.001), TP (p < 0.05), and FC (p < 0.05) and negatively correlated with BD (p < 0.01). In contrast, the proportion of water uptake by Q. aliena from the 60–100 cm layer was negatively correlated with FB (p < 0.05), FC (p < 0.05), SM (p < 0.01), and TP (p < 0.001), but positively correlated with BD (p < 0.01). Furthermore, the proportion of water uptake by Q. aliena showed no significant correlation with clay, silt, and sand (p > 0.05).
we employed a random forest analysis to assess the relative importance of plant and soil

Figure 3.5. Dominant Driver of Water Use Pattern

To identify the dominant factors influencing the water use pattern of *Q. aliena* trees, we employed a random forest analysis to assess the relative importance of plant and soil properties (Figure 6). The results shown indicate that all factors collectively explained 83.8% and 87.2% of the proportion of water uptake for *Q. aliena* from the 0–60 cm and 60–100 cm soil layers, respectively. Soil moisture was found to be a superior predictor for the proportion of water use by *Q. aliena* in the 0–60 cm (Figure 6a) and 60–100 cm (Figure 6b) layers, with relative importance values of 46.1% and 38.3%, respectively. Thus, soil moisture is a key driver influencing the water use patterns of *Q. aliena* forests.

Figure 6. Relative importance of influencing factors to proportion of water absorption based on random forest modeling. FB, fine-root biomass; SM, soil moisture; BD, bulk density; TP, total porosity; FC, field capacity.

4. Discussion

4.1. Water Use Strategies of Mature *Q. aliena* Trees

Our results revealed that mature *Q. aliena* trees primarily used deep soil water (60–100 cm) during the early growing season, but used shallow soil water (0–40 cm) during the late growing season, which partially supported our first hypothesis. This shift in the water source of mature trees from the deep to shallow soil layer during the growing season is a phenomenon observed in other tree species, such as *P. sylvestris* [28]. This can be attributed to the following two reasons. First, the difference in the soil moisture conditions influenced by the precipitation variations between the early and late growing seasons.
led to a distinct water source for the mature trees. Reduced rainfall in the early growing season resulted in lower soil moisture in the shallow layers [19], which could not meet the water consumption needs of mature trees with substantial evapotranspiration. Therefore, mature trees predominantly relied on deep soil water in the early growing season [50]. In the late growing season, adequate precipitation replenished shallow soil water, meeting the high water demand of mature trees. Conversely, precipitation recharge to deep soil water could not meet the water consumption requirements of mature trees. Consequently, mature trees shift to utilizing shallow soil water in the late growing season. Secondly, alterations in the root distribution of mature trees between the early and late growing seasons contributed to the observed shift in water uptake patterns. Typically, in the early growing season following winter, the roots in shallow soil need to regrow to absorb water, while the roots in deep soil can immediately transport water [51]. In contrast, Liu et al. [23] showed that mature *P. euphratica* mainly utilize deep soil water, which may be attributed to the difference in the environmental factors induced by various study areas. Specifically, this study area was located in the warm temperate-northern subtropical transition zone, while Liu et al. [23] was located in the arid zone; hence, the varied regional environment led to differences in the vertical distribution of soil water. In the climatic transition zone of the study area, the soil moisture in the shallow soil water was higher than that in the deep soil water, while the deep soil water was higher than the shallow soil water in the arid zone of Liu et al. [23]. Therefore, *Q. aliena* in our study mainly use the shallow soil water, while *P. euphratica* in Liu et al. [23] mainly use the deep soil water. Consequently, in the early growing season, mature trees predominantly extracted water from the deep soil layer. However, in the late growing season, the roots distributed across all soil layers are fully developed. Coupled with the higher soil moisture in the shallow soil, mature trees shift to using shallow soil water in the late growing season. Taken together, both soil moisture fluctuations and root distribution dynamics contribute to the variations in the water utilization patterns observed in mature trees at various stages of the growing season.

4.2. Key Drivers of Water Use Strategies for Mature *Q. aliena* Trees

The inquiry into the factors governing the water utilization patterns of mature *Q. aliena* trees led us to employ a random forest model, revealing that soil moisture predominantly dictated these patterns (Figure 5), partly supporting our second hypothesis. This could potentially be attributed to the fact that soil layers with a higher moisture content exhibit higher water availability [34,35]. Consequently, plants, especially mature trees with substantial transpiration, exhibit a preference for water from soil layers with high moisture levels [17,28,52–54]. Thus, the water use pattern of mature trees is regulated by soil moisture. In periods of ample precipitation, the adequate supply of soil water meets the water consumption demands of mature and tall trees [5]. Mature trees are more sensitive to shifts in soil moisture and adopt flexible water use strategies, indicating fewer constraints on water transport. Specifically, given that the precipitation in the early growing season was lower than that in the middle and late growing seasons in our study area (Figure 2), the shallow soil moisture in the early growing season was significantly lower than that in the middle and late growing seasons of mature *Q. aliena* trees (Figure 6). Consequently, mature *Q. aliena* trees primarily used deep soil water in the early growing season and shifted to shallow soil water in the late growing season.

Our results also indicated that fine root biomass had a small effect on the water use pattern of mature *Q. aliena* trees (Figure 5), which did not support our second hypothesis. This phenomenon may be attributed to the well-developed root system of mature trees, which is characterized by both horizontal and vertical growth. Prior research has demonstrated that the development of fine root biomass in oak trees reaches a steady state after 50 years [55]. Given that the age of mature *Q. aliena* trees in our study exceeded 60 years, the seasonal variation in fine root biomass was relatively small. Consequently, fine root biomass was not a key driver of the water uptake pattern of mature *Q. aliena* trees. Furthermore, the findings of this study indicated that soil properties and texture did not affect the water
use pattern of mature *Q. aliena* trees (Figure 5). This may be attributed to the fact that although soil physical properties can regulate tree water use patterns by modulating their hydraulic conductivity and water holding capacity \cite{35,56}, their regulatory impact becomes significant only when soil moisture becomes a limiting factor \cite{57}. In our study, the soil moisture levels for mature *Q. aliena* trees remained within the optimal threshold range for plant water supply (field capacity ranging from 0.5 to 0.8) \cite{58}. Therefore, soil physical properties did not directly affect the water use pattern of mature *Q. aliena* trees.

This study also found a difference in rainfall between the growing seasons of 2021 and 2022. Nevertheless, the water use pattern of mature *Q. aliena* exhibited no significant variation between these two periods (Figure 4). In other words, precipitation during the growing season had no effect on the water use pattern of mature *Q. aliena* trees. This may be attributed to the fact that precipitation changes were consistent during the growing seasons in both 2021 and 2022. Specifically, the maximum precipitation occurred in the MGS, followed by the LGS, with the lowest precipitation recorded in the EGS for both 2021 and 2022 (Figure 2). Consequently, this pattern resulted in lower soil moisture during the EGS and higher soil moisture during the MGS and LGS. As a result, our findings indicate that mature *Q. aliena* trees primarily used deep (60–100 cm) soil water during the EGS and shifted to utilizing shallow (0–40 cm) soil water during the LGS.

### 4.3. Implications and Uncertainties

Our study yields two key implications from the observed results. Firstly, contrary to the conventional view that mature trees predominantly rely on deep soil water or groundwater, our findings confirmed that mature *Q. aliena* trees had flexible water use strategies. Specifically, mature *Q. aliena* trees could adjust their water use pattern according to soil moisture levels. Consequently, when protecting or transplanting mature oak trees, we recommend ensuring appropriate soil moisture levels in addition to protecting the root system. Secondly, given that “a big tree strikes deep roots”, the traditional view holds that the root is the primary driver affecting the water uptake pattern of mature trees. However, our results indicated that soil moisture was the dominant factor influencing the water use pattern of mature *Q. aliena*. Therefore, future investigations on the factors influencing water absorption by mature trees should not solely focus on root biomass, but also consider the impact of various factors, including soil moisture.

Although this study elucidated the water use pattern of mature *Q. aliena* in the warm temperate-northern subtropical transition zone, there are still some uncertainties in our study. Firstly, given that the vegetation in the study area was mainly natural forest with more than 40 years of enclosure management, we only explored the water use pattern of mature *Q. aliena* (>40 years) in this study, but did not investigate the water use pattern of *Q. aliena* under 40 years. Therefore, we could not compare the water use strategies between young and mature forests. In the future, more research should be conducted to improve our understanding of the water uptake pattern of *Q. aliena* throughout its life cycle (young–mature–old). Second, due to the limitation of workload and funding, this study area was only established in the climate transition zone rather than other regions. However, there is a significant difference in the environmental conditions in various regions, and it is still unclear whether the difference affects the water use pattern of *Q. aliena*. Therefore, we should pay attention to the water use pattern of *Q. aliena* in different habitats in the future. Third, this study mainly focused on mature *Q. aliena* and did not involve other tree species; therefore, it is unclear whether the results on the water use pattern of *Q. aliena* in this study are applicable to other tree species. Accordingly, the water use patterns of other mature tree species should be further investigated to understand the water use strategies of more tree species during each growth period.

### 5. Conclusions

Using hydrogen and oxygen stable isotopes coupled with MixSIAR, we found that mature *Q. aliena* trees predominantly utilized deep soil water (60–100 cm) in the early
growing season, but shifted to shallow soil water (0–40 cm) in the late growing season. Furthermore, our results revealed that soil moisture played a pivotal role in regulating the water use patterns of mature Q. aliena trees throughout the growing season based on the random forest model. This indicated that mature Q. aliena trees exhibited flexible water use strategies. These findings carry significant implications, suggesting that the frequent seasonal drought events triggered by climate change have resulted in water scarcity, severely hindering tree growth, especially among mature trees with high water consumption. Mature Q. aliena trees adapted their water use pattern according to the soil moisture variations. Therefore, to help develop and protect mature oak trees, surface soil irrigation could be increased to prevent forest death due to drought or extreme drought.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/f15030402/s1, Figure S1: Fine root biomass and aboveground biomass of Q. aliena of different ages. Capital letters indicate significant different ages of Q. aliena in the same soil depth, and lowercase letters indicate significant differences for same age of Q. aliena in different soil depth (p < 0.05); Figure S2: Vertical profile (0–100 cm) of soil moisture (%) of Q. aliena at different stand ages. Error bars represent one standard deviation (n = 3). Lowercase letters indicate significant differences in soil moisture of the same soil depth at different growth stage (p < 0.05); Table S1: Soil physical properties in Q. aliena of different ages.

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Data Availability Statement: The datasets used and/or analyzed during this study are available from the corresponding author upon reasonable request.

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Conflicts of Interest: The authors declare no conflicts of interest.

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