Relationship between the Growth of Different Grades of Seedlings and Leaf Functional Traits in *Ormosia hosiei*

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Abstract

The wild resources of Ormosiahosiei are gradually being depleted, and its slow seedling growth constrains ex situ conservation in the field. This study focused on ex situ conserved Ormosiahosiei seedlings and, based on grading by ground diameter and seedling height, used one-way analysis of variance to analyze the growth and leaf functional traits of seedlings of different grades, aiming to enrich research data and provide a basis for further studies. The results showed that: (1) The growth of Ormosiahosiei seedlings followed an allometric growth pattern (P1.0<0.01), with priority given to vertical growth. (2) Among seedlings of different grades, there were no significant differences in leaf functional traits including leaf area, leaf thickness, carbon content, phosphorus content, stable carbon-13 isotope, intrinsic water use efficiency, and relative chlorophyll content; only leaf N content differed significantly (P<0.05). Leaf N:P<14, indicating that Ormosiahosiei is susceptible to N limitation during the early stage of growth. (3) Correlation analysis showed a highly significant positive correlation between ground diameter and intrinsic water use efficiency (P<0.01), and a significant positive correlation between seedling height and SPAD value (P<0.01). Leaf N content was significantly negatively correlated with $ $13C (P<0.01). In summary, during field reintroduction afforestation, Ormosiahosiei seedlings exhibited a resource allocation strategy that prioritized vertical growth, accompanied by low water use efficiency. Given the presence of nitrogen limitation during the early growth stage, nitrogen fertilizer management should be strengthened in ex situ conservation management.

Full Text

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Abstract: The wild resources of *Ormosia hosiei* are gradually being depleted, and its slow growth at the seedling stage constrains its ex-situ conservation in the wild. This study used *O. hosiei* seedlings under ex-situ conservation as the research object. Based on classification by ground diameter and seedling height, one-way analysis of variance was used to analyze the growth and leaf functional

traits of seedlings of different grades, with the aim of enriching research data and providing a basis for further studies. The results showed that: (1) The growth of *O. hosiei* seedlings followed an allometric growth pattern ($P_{1.0} < 0.01$) and preferentially emphasized vertical growth. (2) Among seedlings of different grades, there were no significant differences in leaf functional trait indicators including leaf area, leaf thickness, carbon content, phosphorus content, stable carbon-13 isotope, intrinsic water-use efficiency, and relative chlorophyll content; only leaf N content differed significantly ($P < 0.05$). Leaf N:P was < 14, indicating that the early growth of *O. hosiei* is susceptible to N limitation. (3) Correlation analysis showed that ground diameter was extremely significantly positively correlated with intrinsic water-use efficiency ($P < 0.01$), and seedling height was significantly positively correlated with SPAD value ($P < 0.01$). Leaf N content was significantly negatively correlated with $\delta^{13}C$ ($P < 0.01$). In summary, during reintroduction afforestation in the wild, *O. hosiei* seedlings exhibit a resource-allocation strategy that prioritizes vertical growth, while also being characterized by low water-use efficiency. Given that nitrogen limitation occurs during the early stage of growth, nitrogen fertilization management should be strengthened in ex-situ conservation management.

Keywords: *Ormosia hosiei*; wild reintroduction; seedling growth; leaf functional traits; intrinsic water-use efficiency

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Relationship between growth of different grade seedlings

and leaf functional traits of *Ormosia hosiei*

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Abstract: The wild resources of *Ormosia hosiei* are increasingly being depleted, and its slow seedling-stage growth poses significant constraints on ex-situ conservation efforts in wild habitats.

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This study focused on ex-situ conserved seedlings of *O. hosiei* and established a grading system based on diameter at ground level (DGL) and seedling height (SH). Through one-way ANOVA, we systematically analyzed the growth performance and leaf functional traits of seedlings across different grades. The research aims to enrich the scientific dataset of *O. hosiei* and establish a fundamental basis for subsequent studies on its population restoration and optimization of ex-situ conservation technologies. The results were as follows: (1) Seedlings of *O. hosiei* exhibited allometric growth patterns ($P_{1.0} < 0.01$), with preferential vertical growth. (2) Among seedlings of different grades, leaf functional traits including leaf area, leaf thickness, carbon content, phosphorus content, stable carbon-13 isotope (δ^{13}C), intrinsic water use efficiency (iWUE), and relative chlorophyll content (SPAD) showed no significant differences, except for leaf nitrogen (N) content ($P < 0.05$). The leaf N:P ratio < 14 suggested that *O. hosiei* is susceptible to N limitation during the early growth stage. (3) Correlation analysis revealed a highly significant positive correlation between DGL and iWUE ($P < 0.01$), as well as a significant positive correlation between SH and SPAD value ($P < 0.01$). However, there was a significant negative correlation between leaf N content and δ^{13}C ($P < 0.01$). In conclusion, during wild reintroduction planting, *O. hosiei* seedlings demonstrate a resource allocation strategy that prioritizes vertical growth, coupled with suboptimal water use efficiency. Ex-situ conservation management should therefore emphasize enhanced nitrogen fertilization practices to address the nitrogen limitation identified in early growth stages.

Key words: *Ormosia hosiei*, reintroduction in the wild, seedling growth, leaf functional traits, intrinsic water use efficiency

Ormosia hosiei, a tree species of the genus *Ormosia* in the family Leguminosae, is endemic to China. It is a national Class II key protected wild plant and is listed as Endangered (EN) on the IUCN Red List of Threatened Species. As a valuable native tree species and culturally significant tree with ornamental, medicinal, and economic value, its potential for development and utilization has attracted considerable attention (Qiu Haojie et al., 2020; Chu Xiuli et al., 2021). Since the 1950s and 1960s, affected by factors such as overharvesting and climate change, its habitat fragmentation has become pronounced. In addition, biological characteristics such as slow growth, unstable flowering and fruiting, brightly colored seeds that are readily eaten by animals, and difficulty in reproduction (thick seed coats that are dry and not easily water-absorbent, together with poor dispersal and reproductive capacity) (Gui Ping et al., 2018;

Qiu Haojie et al., 2020; Ren Wenling et al., 2023) have severely hindered natural regeneration, causing populations to show a marked declining trend (Zhang Qunfang et al., 2015; Wang Xiaodong et al., 2018). At present, both natural forests and plantation resources of *O. hosiei* are extremely scarce. Natural populations mostly occur around villages or near ancient temples (Chu Xiuli et al., 2021), while plantations have only been sporadically recorded in places such as Youxi, Fujian (You Genbiao et al., 2017). This state of resource depletion may severely constrain gene flow among populations. In practices of ex-situ conservation and reintroduction in the wild, there are few research reports on *O. hosiei* after establishment; the lack of such scientific data has impeded conservation efforts such as the optimization of wild reintroduction techniques, expansion of population size, and maintenance of population stability.

Reintroduction in the wild refers to the practice, based on ex-situ conservation, of reintroducing rare species that are on the brink of extinction for specific reasons—or have even become extinct in the wild—into their original ecosystems, or into environments suitable for their survival and reproduction, thereby helping them establish, survive over the long term, and form self-sustaining populations (Godefroid et al., 2011; Ren et al., 2014). At present, regarding

There have been relatively few studies on the field reintroduction of red bean tree, and reports on the growth and leaf functional traits of reintroduced seedlings in the field are even rarer. Existing studies have mainly focused on physiological and ecological characteristics (Han Hao et al., 2020), population quantitative structure dynamics (You Genbiao et al., 2017; Wang Mingbin et al., 2024), genetic diversity (Li Fengqing et al., 2017; Li Fengqing et al., 2018), chemical constituents (Qiu Yatie et al., 2018), diseases and insect pests (Han Senhui et al., 2021), seed germination (He Guanrong et al., 2017), seedling propagation (Tang Liangzhi et al., 2013; Zhu Min et al., 2024), drought stress (Rui Wenyi et al., 2012), and soil bacterial communities (Zhou Hongmin et al., 2022). Seedling growth can be divided into radial growth and vertical growth, represented respectively by ground diameter and seedling height; the former reflects mechanical support and water-absorption capacity, whereas the latter reflects light-acquisition capacity and spatial competition strategies (Aiba & Kohyama, 1996). Allometric growth mainly refers to unequal growth rates among different parts of an organism (Weiner & Thomas, 1992). Leaves are the principal sites where plants conduct photosynthesis and exchange matter and energy with the external environment, and leaf traits can best reflect a plant' s own growth status and survival strategies for adapting to the environment (Pan et al., 2020; Li et al., 2022). According to their functional attributes, leaf traits can be divided into morphological traits (specific leaf area, leaf mass per area, leaf vein density, leaf thickness, etc.), chemical traits (leaf carbon, nitrogen, and phosphorus contents, as well as secondary metabolites such as tannins and phenolics), and physiological traits (chlorophyll content, photosynthetic capacity, stomatal conductance, etc.) (Wright et al., 2004; Xiao Yihua et al., 2022). Taking seedling allometric growth and leaf functional traits as entry points, analyzing differences among red bean tree seedlings of different grades

is helpful for understanding their ecological adaptability and can provide scientific reference for the practice of field reintroduction.

This study used red bean tree seedlings planted for field reintroduction as the research object. A one-way analysis of variance was adopted, and seedlings were graded according to ground diameter and seedling height, with the aim of addressing the following questions: (1) the allometric growth characteristics of red bean tree seedlings planted in the field; (2) the leaf functional traits of seedlings of different grades; and (3) the relationship between growth characteristics and leaf functional traits of seedlings of different grades. The results of this study will help reveal the growth and life-history strategies of seedlings of different grades, clarify the relationship between growth and nutrient-use strategies during field reintroduction of red bean tree, enrich research materials on field reintroduction practice and ex situ conservation-based cultivation of robust seedlings for this species, and provide a foundation for further research.

1 Materials and Methods

1.1 Overview of the Study Area

The study site is located in Shaping Town, Lechang City, Shaoguan, Guangdong Province (113°2′ E, 25°5′ N), and is a demonstration area for ex situ conservation and field reintroduction of red bean tree. It has a subtropical monsoon climate. Warm and humid southerly airflows prevail in summer, rainfall is abundant, annual precipitation is 1,200 mm and is mostly concentrated from May to September, the annual mean temperature is 19.1 °C, and annual sunshine duration is 1,499.7 h. The terrain of Shaping Town is dominated by karst landforms, with many exposed rocky hills in the area, and the soils are mainly red limestone soil and red soil.

1.2 Research Methods

1.2.1 Plot Establishment

In February 2024, a 60-mu demonstration forest for ex situ conservation and reintroduction of red bean tree was established in Shaping Town, Lechang City. The planting stock consisted of 3-year-old seed-origin seedlings (ground diameter 8.74–9.86 mm, seedling height 0.55–0.61 m), with consistent growth vigor. The planting spacing was 2 m × 3 m. After tending in August 2024 (removal of weeds), nine 40 m × 30 m observation plots were established (distance between plots $ $10 m). To ensure that site conditions were basically consistent, plot elevation was controlled within 550–568 m, as were slope aspect (sunny slope to semi-sunny slope) and slope gradient (14°–25°). Red bean tree seedlings in each plot were tagged one by one; their ground diameter (DGL, diameter at ground level) was measured with a vernier caliper, and a tape

A ruler was used to measure seedling height (SH). Using the mean and standard

deviation (Niklas, 1995), the seedlings were divided into three grades: seedlings with diameter at ground level $DGL \geq \text{Mean} + 1/2SD$ and seedling height $SH \geq \text{Mean} + 1/2SD$ were Grade I; those with $M - 1/2SD \leq DGL < \text{Mean} + 1/2SD$ and $M - 1/2SD \leq SH < \text{Mean} + 1/2SD$ were Grade II; and those with DGL and $SH < \text{Mean} - 1/2SD$ were Grade III (Table 1).

Table 1 Grading of Seedlings

Seedling grade	Diameter at ground level	Seedling height
Seedling grade	Diameter at ground level (mm)	Seedling height (m)
I	$DGL \geq 15.43$	$SH \geq 1.05$
II	$14.07 \leq DGL < 15.43$	$0.91 \leq SH < 1.05$
III	$DGL < 14.07$	$SH < 0.91$

1.2.2 Allometric growth of seedlings

The nonlinear quantitative relationship of seedling allometric growth is usually expressed in the form of a power function (Enquist & Niklas, 2002). After logarithmic transformation, the linear relationship is as follows:

$$\log Y = \log \beta + \alpha \log X.$$

In the formula, Y and X represent seedling height (cm) and diameter at ground level (mm), respectively; β is the intercept, and α is the slope. When $\alpha = 1$, the relationship is isometric growth; when $\alpha \neq 1$, it is allometric growth (Gould, 1966). The standardized major axis (SMA) estimation method was used to estimate the parameters of the allometric growth equations between seedling height and diameter at ground level for different grades of *Ormosia hosiei* seedlings. Comparative analyses were conducted on differences among the slopes (α) of the treatments and on the differences between each slope and 1.0.

1.2.3 Measurement of leaf physicochemical and photosynthetic indices

For 10 *Ormosia hosiei* trees of different grades, three healthy, mature leaves free of diseases and insect pests were selected from each tree. A Pocket PEA chlorophyll fluorometer (Hansatech Instruments LTD, UK) was used to measure the SPAD value (soil and plant analyzer development, characterizing the relative chlorophyll content of leaves), the maximum photochemical efficiency of PS II (F_v/F_m), and the photosynthetic performance index (PI). Each leaf was measured three times and the mean was taken; the mean of the three leaves was used as the data for a single plant. The collected leaves were placed in a portable cooler and brought back to the laboratory. Leaf area (LA) was measured using an LI-3000C portable leaf area meter (LI-COR Biosciences, USA), and leaf thickness (LT) was measured with a vernier caliper. The leaves were

then dried in an oven at 60–80 °C to constant weight, ground into fine powder, and sieved to obtain homogeneous leaf powder samples. An elemental analyzer (ECS 4010, Costech Analytical Technologies, USA) was used to determine leaf carbon and nitrogen contents, and the molybdenum–antimony colorimetric method was used to determine leaf phosphorus content. Leaf N:P can be used as an effective indicator characterizing soil nutrient limitation status (Zhang et al., 2019): when N:P < 14, plant growth is limited by N; when N:P > 16, plant growth is limited by P; and when N:P is 14–16, growth is jointly limited by N and P (Chapin et al., 2011). The δ^{13}C of plant samples was measured using an isotope mass spectrometer (Isoprime 100, Isoprime LTD, UK). Because the $^{13}C/^{12}C$ value of plant leaves is relatively low and not conducive to analysis and comparison, it was expressed relative to the $^{13}C/^{12}C$ value of the international standard material (PDB). The calculation formula is as follows:

$$\delta^{13}\mathrm{C} = \frac{R_{\mathrm{P}} - R_{\mathrm{PDB}}}{R_{\mathrm{PDB}}}.$$

In the formula, δ^{13}C is the carbon isotope value of plant leaves; R_{P} is the $^{13}C/^{12}C$ value of plant leaves; and R_{PDB} is the standard material PDB.

of the $^{13}C/^{12}C$ value.

Intrinsic water-use efficiency (iWUE) was calculated based on the δ^{13}C value (Tang et al., 2022; Pan et al., 2024). The formula for calculating leaf stable carbon isotope discrimination (Δ^{13}C) is as follows:

$$\Delta^{13}\mathrm{C} = \left(\delta^{13}\mathrm{C}_a - \delta^{13}\mathrm{C}\right) / \left(1 + \frac{\delta^{13}\mathrm{C}}{1000}\right).$$

In the formula, δ^{13}C and $\delta^{13}\mathrm{C}_a$ represent the carbon isotope values of plant leaves and atmospheric CO_2, respectively. The concentration of CO_2 (C_a) and the carbon isotope value $\delta^{13}\mathrm{C}_a$ were calculated as follows (Feng, 1998):

$$C_a = 277.78 + 1.350\exp\left[0.01572(t - 1740)\right];$$

$$\delta^{13}\mathrm{C}_a = -6.429 - 0.006\exp[0.0217(t - 1740)].$$

In the formula, t is the year of sampling (2024).

Using the relative diffusion coefficient of water vapor to CO_2 in air (1.6), iWUE was solved by the equation (Pan et al., 2024). The calculation formula is as follows:

$$iWUE = \frac{C_a}{1.6} \times \frac{b - \Delta^{13}\mathrm{C} - f \times \frac{\Gamma^*}{C_a}}{b - a_s + \frac{g_{sc}}{g_m} \times (b - a_m)}$$

In the formula, a_s and a_m are the fractionations caused by diffusion of atmospheric CO_2 through stomata (4.4‰) and mesophyll (1.8‰), respectively; b (29‰) and f (11.2‰) are the fractionation constants associated with Rubisco carboxylation and photorespiration, respectively; g_{sc}/g_m is the conductance ratio for CO_2 diffusion through stomata and mesophyll (0.79); Γ^* is the CO_2 compensation point in the absence of mitochondrial respiration (Stocker et al., 2020). Based on temperature (the sampling temperature T in this study was 32 °C), $\Gamma^* = 42.7 + 1.68(T - 25) + 0.012(T - 25)^2$ was calculated.

1.3 Data Processing and Analysis

One-way analysis of variance (ANOVA) was used to test differences in the mean values of growth and leaf functional trait indicators among seedlings of different grades. Before data analysis, tests for normality and homogeneity of variance were performed, and multiple comparisons were conducted using Duncan' s method. Correlation analysis used Pearson' s correlation coefficient. The allometric growth relationship between ground diameter and seedling height of *Ormosia hosiei* seedlings was calculated using the "Smatr" package in R (4.4.3) (Warton et al., 2012). Intrinsic water-use efficiency was calculated using the "mesophyll" method in the "isocalcR" package (Mathias & Hudiburg, 2022). Data were visualized using Origin2021.

2 Results and Analysis

2.1 Allometric Growth of Seedlings

The results of standardized major axis analysis showed that the growth of *Ormosia hosiei* seedlings followed an allometric growth pattern rather than isometric growth ($P_{1.0} < 0.01$); that is, the allometric growth index differed significantly from 1.0 and did not conform to the predicted value of 1.0 under the geometric similarity model. The seedling allometric growth index was 1.62 (slope), indicating that seedling height increased faster than ground diameter. This reveals a resource-allocation strategy in the growth of *Ormosia hosiei* seedlings that prioritizes vertical growth to compete for light and spatial resources, and there was no significant linear growth relationship among seedlings of the same grade (Fig. 1).

Legend: Grade I seedling; Grade II seedling; Grade III seedling

Top-left panel:

y-axis: log(seedling height)
x-axis: log(diameter at ground level)

$$y = -1.90 + 1.62x$$

$$\text{SMA } R^2 = 0.64$$

$$P_{\text{Model}} < 0.01$$

$$P_{1.0} < 0.01$$

Top-right panel:

y-axis: log(seedling height)
x-axis: log(diameter at ground level)

$$y = 0.20 - 0.05x$$

$$\text{SMA } R^2 = 0.03$$

$$P_{\text{Model}} = 0.72$$

Bottom-left panel:

y-axis: log(seedling height)
x-axis: log(diameter at ground level)

$$y = 0.37 - 0.33x$$

$$\text{SMA } R^2 = 0.04$$

$$P_{\text{Model}} = 0.07$$

Bottom-right panel:

y-axis: log(seedling height)
x-axis: log(diameter at ground level)

$$y = -0.10 - 0.03x$$

$$\text{SMA } R^2 = 0.02$$

$$P_{\text{Model}} = 0.82$$

$P_{1.0}$ indicates the significance of the difference between the slope estimated by the standardized major axis and the theoretical value of 1.0.

Fig. 1 Allometric growth characteristics of seedlings

2.2 Functional Traits of Leaves

Among leaf morphological traits, the ranges of leaf area and leaf thickness among seedlings of different grades were 20.57–27.27 cm^2 and 0.25–0.26 mm, respectively, and for both traits the values of Grade I seedlings were higher than those of Grade II and Grade III seedlings. For leaf chemical traits, the values of C content, N content, P content, C:N, C:P, N:P, and $\delta^{13}C$ in seedlings of different grades were 480.26–480.80 $g \cdot kg^{-1}$, 23.32–23.52 $g \cdot kg^{-1}$, 1.79–1.81 $g \cdot kg^{-1}$, 20.42–20.60, 265.01–269.05, 12.97–13.06, and −28.40‰ to −28.23‰, respectively. Among these, only N content showed a significant difference: the N content of Grade II seedlings was significantly higher than that of Grade III seedlings ($P < 0.05$). Leaf N:P was < 14, indicating that, in this study, the growth of *Ormosia* plants was more likely limited by N. Among leaf physiological traits, intrinsic water-use efficiency (41.54–42.50 $\mu mol \cdot mol^{-1}$), SPAD value (29.48–30.66), photosynthetic performance index (0.70–0.95), and maximum photochemical efficiency (0.66–0.71) showed no significant differences among seedlings of different grades (Fig. 2). In summary, the differences among red bean tree seedlings of different grades were mainly reflected in N content, while most leaf functional traits showed no obvious differences.

[Figure 2 panels, top-to-bottom and left-to-right: Leaf area (cm^2), $P = 0.178$; Leaf thickness (mm), $P = 0.679$; C content ($g\ kg^{-1}$), $P = 0.294$; N content ($g\ kg^{-1}$), $P < 0.05$, with lowercase letters "ab" , "a" , and "b" ; P content ($g\ kg^{-1}$), $P = 0.110$; C:N, $P = 0.067$; C:P, $P = 0.117$; N:P, $P = 0.657$; $\delta^{13}C$ (‰), $P = 0.091$; iWUE ($\mu mol\ mol^{-1}$), $P = 0.091$; SPAD value, $P = 0.513$; PI, $P = 0.079$; F_v/F_m, $P = 0.081$. The x-axis in each panel is "Seedling grades"with grades I, II, and III. Legend: morphological traits of leaves; chemical characteristics of leaves; physiological traits of leaves.]

Different lowercase letters indicate that the results of statistical analysis are significant at the $P < 0.05$ level.

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Fig. 2 Functional traits of plant leaves

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2.3 Relationship between seedling growth and leaf functional traits

The correlation analysis showed that DGL and iWUE were extremely significantly positively correlated ($P < 0.01$), and this correlation was also found between SH and the SPAD value ($P < 0.01$). The SPAD value was significantly positively correlated with PI and LA, respectively ($P < 0.05$); F_v/F_m and C content, as well as LA and LT, all showed significant positive correlations ($P < 0.05$). By contrast, N content and $\delta^{13}C$, N content and N:P, and C content

and C:P were all extremely significantly negatively correlated ($P < 0.01$), while SPAD and iWUE, and F_v/F_m and C:P, were significantly negatively correlated ($P < 0.05$; Fig. 3).

[Figure 3 correlation heatmap. Variables shown: DGL, SH, SPAD, F_v/F_m, PI, P, N, C, C:N, C:P, N:P, δ^{13}C, iWUE, LA, and LT. Significance marks: $*$ $P < 0.05$; $**$ $P < 0.01$. Color scale ranges from 1.0 to −1.0.]

DGL and SH denote the diameter at ground level and seedling height, respectively; the SPAD value is an indicator characterizing chlorophyll content in leaves; F_v/F_m is the maximum photochemical efficiency of PS II; PI is the photosynthetic performance index; C:N, C:P, and N:P are the ratios of C, N, and P contents in leaves; δ^{13}C is the carbon isotope value of plant leaves; iWUE is the intrinsic water-use efficiency of plants; LA is leaf area; LT is leaf thickness.

DGL and SH denote the diameter at ground level and seedling height, respectively; the SPAD value serves as a crucial parameter for quantifying chlorophyll content in leaves; F_v/F_m represents the maximum photochemical efficiency of photosystem II (PS II); PI is the photosynthetic performance index; C:N, C:P, and N:P refer to the ratios of C, N, and P contents in leaves; δ^{13}C indicates the carbon isotope value of plant leaves; iWUE stands for

the intrinsic water use efficiency of plants; LA and LT represent leaf area and leaf thickness, respectively.

Fig. 3 Correlations between seedling growth and leaf functional traits

3 Discussion and Conclusions

3.1 Mechanisms of allometric growth in different grades of red bean tree seedlings

The allometric growth relationship between ground diameter and seedling height reflects the trade-off between vertical and radial growth of trees under environmental stress, and is the result of interactions between plants' own genetic traits and environmental factors (Wang Dan et al., 2021). In this study, the growth of both ground diameter and seedling height among different grades of red bean tree seedlings followed an allometric growth pattern. This differs from the findings of Chen Shi et al. (2023), possibly because of differences in tree species characteristics and environmental heterogeneity (Wang Dan et al., 2021). Different seedlings possess unique genetic characteristics, which determine their basic growth patterns and allometric growth rules. Seedlings of fast-growing tree species have relatively rapid growth rates and specific growth proportional relationships, whereas seedlings of slow-growing tree species grow more slowly and show different coordination among the growth of different parts (Zhu Miaomiao et al., 2022). Environmental heterogeneity, such as light, water, and soil nutrients, also affects individual size and thereby influences growth patterns (Dinnage et al., 2019). In this study, the allometric growth slope of red bean

tree seedlings was 1.62 (height growth was faster than ground diameter), indicating that in the allocation of photosynthates, the weight given to vertical growth (height) was higher than that given to radial growth (stem thickening). This reflects an allocation mechanism in which red bean tree seedlings prioritize vertical growth in order to capture light and spatial resources. The reason may be that the study site is located in a karst rocky desertification habitat, which is characterized by shallow soil and exposed rock; light distribution is uneven, and plant growth is clearly limited by light (Xiong Ling et al., 2024; Jiang Chaoyou et al., 2025). Different plants have trade-off strategies in resource acquisition and utilization (Wright et al., 2004). The strategy of red bean tree to prioritize vertical growth may be the result of comprehensive adaptation to the external environment in karst rocky desertification habitats in order to meet seedling growth requirements. Higher-grade seedlings have more developed root systems and thicker stems, and the coordination between aboveground and belowground growth is complex. They may preferentially allocate photosynthates to stem thickening and height growth to compete for more light resources, while also ensuring sufficient root growth to maintain water and nutrient uptake. By contrast, lower-grade seedlings, because of insufficient nutrient reserves and constraints imposed by the growth environment, grow slowly. In terms of allometric growth, root growth may be relatively slower than aboveground growth, or aboveground leaf growth may be sparse and unable to carry out photosynthesis effectively, thereby affecting overall growth and development (Liu Yao et al., 2024). Some studies have also indicated that the allometric growth relationships of plants differ among stand-age stages; that is, allometric relationships change with tree development. Because of plants' utilization of light resources, ground-diameter growth first decreases and then increases with increasing stand age (Wang Dan et al., 2021). This study did not observe the allometric growth relationships of red bean tree seedlings at different stages, and this can be further explored in future research.

3.2 Leaf functional traits and environmental adaptation mechanisms of red bean tree

Leaf morphological traits mainly include leaf attributes such as leaf size, shape, and accessory structural characteristics of the leaf surface (Zhang Zitan et al., 2021). In this study, only the leaf area (20.57–27.27 cm^2) and leaf thickness (0.25–0.26 mm) of red bean tree seedlings were measured. The former values were close to the leaf area (23.56 cm^2) reported by Chen Huanwei et al. for red bean tree, but leaf thickness was thinner (0.4 mm). The reasons for the differences in research results may be that the phenotypic expression of traits such as leaf area and leaf thickness is influenced by the external environment; under different environmental conditions, such as light, temperature, and moisture, the same traits may differ significantly (Zhang Zitan et al., 2021).

Plants'adaptation to the environment and changes in their own genetic structure during the process of environmental adaptation can be achieved through growth

and trait expression.

(Pigliucci et al., 2006). The nutrient-element contents of plant leaves reflect plant adaptation to environmental nutrient conditions (Ma et al., 2015). As key biogenic elements that constitute plant cell structures and maintain their functions, C, N, and P are closely linked to plant leaf elemental characteristics, their own structural properties, and the environments in which they occur (Sardans et al., 2012). In this study, the leaf C content of *Ormosia hosiei* ($480.26 \sim 480.80\ g \cdot kg^{-1}$) was higher than the mean leaf C contents of plants in China ($436.8\ g \cdot kg^{-1}$) and of global terrestrial plants ($464\ g \cdot kg^{-1}$) (Elser et al., 2000; Tang et al., 2018); its P content ($1.79 \sim 1.81\ g \cdot kg^{-1}$) was higher than the mean leaf P content of plants in China ($1.21\ g \cdot kg^{-1}$) (Tang et al., 2018), but lower than the global mean leaf P content ($1.99\ g \cdot kg^{-1}$) (Elser et al., 2000). Its N content ($23.32 \sim 23.52\ g \cdot kg^{-1}$) was higher than the mean leaf N contents of plants in China ($18.60\ g \cdot kg^{-1}$) and of global plants ($20.60\ g \cdot kg^{-1}$) (Elser et al., 2000; Han et al., 2005). Leaf C : N ($20.42 \sim 20.60$) was close to the global mean leaf C : N (22.50), whereas C : P ($265.01 \sim 269.05$) was lower than the global mean leaf C : P value (469.16) (Han et al., 2005). The range of leaf N : P values was $12.97 \sim 13.06$, higher than the global mean leaf N : P value of plants (12.7) (Reich and Oleksyn, 2004; Han et al., 2005). In addition, plant growth of *O. hosiei* in this study was more likely to be limited by N (Chapin et al., 2011). The $\delta^{13}C$ values in this study ($-28.40‰ \sim -28.23‰$) fell within the $\delta^{13}C$ range of terrestrial C_3 plants, from $-20‰$ to $-37‰$ (Kohn, 2010). Intrinsic water-use efficiency (iWUE), calculated based on $\delta^{13}C$ values, can serve as one of the objective evaluation indicators for water use and drought-resistance traits (Pan et al., 2024). The intrinsic water-use efficiency of *O. hosiei* was $41.54 \sim 42.50\ \ mol \cdot mol^{-1}$ (Fig. 2), lower than the mean value reported in existing studies for broad-leaved trees ($59.79\ \ mol \cdot mol^{-1}$) (Cai Jindong et al., 2024). This indicates that the intrinsic water-use efficiency of *O. hosiei* is not high, and the underlying causes remain to be further revealed. Considering the analysis that seedling growth is limited by nitrogen, future research should integrate genetic improvement—screening families with high intrinsic water-use efficiency—and habitat optimization, such as increasing nitrogen fertilization, to improve the success rate of field reintroduction.

Among leaf physiological traits, a higher SPAD value indicates a higher chlorophyll content in leaves and a stronger capacity of leaves to capture light energy, which helps improve the overall efficiency of photosynthesis and promotes carbon dioxide fixation and organic matter synthesis (Xiong et al., 2015). In this study, the SPAD value of *O. hosiei* leaves was $29.48 \sim 30.66$, which is lower than the SPAD value of seedlings (36.72) reported in existing studies (Chen Zhuokang et al., 2023). The reason is that SPAD values differ among tree species, and also vary with growth environment and developmental stage. For example, under sufficient light and favorable soil fertility, leaf SPAD values may be higher, whereas under the opposite conditions they may be lower. In addition, the SPAD values of young leaves are usually lower than those of mature leaves; as leaves grow and develop, SPAD values gradually increase and

then tend to stabilize. F_v/F_m is the maximum photochemical quantum yield of photosystem PS II, and its value reflects the potential maximum photochemical efficiency of the PS II reaction center (Materán et al., 2015). Leaves with a higher photosynthetic performance index (PI) usually have more reasonable chloroplast structure and pigment distribution, can capture light energy more effectively, and have an appropriate ratio of chlorophyll a to chlorophyll b, which facilitates light absorption at different wavelengths and provides an adequate energy source for photosynthesis (Rodríguez et al., 2023). In this study, PI and F_v/F_m showed a positive correlation, similar to results reported in previous studies, namely that a positive correlation exists between PI and F_v/F_m (Sun and Wang, 2018; Yue et al., 2023).

Considering the combination of leaf functional traits of *O. hosiei* and referring to data from the global leaf economics spectrum (Wright et al., 2004), its leaf economic spectrum tends toward a "quick return"strategy—high nitrogen content, thin leaves, and low water-use efficiency. This may be related to the coevolution of fluctuating light resources in its karst habitat and the nitrogen-fixing capacity of legumes, and remains to be further verified by research.

3.3 Correlation between seedling growth and leaf functional traits

In this study, ground diameter and intrinsic water-use efficiency showed an extremely significant positive correlation, while seedling height was significantly positively correlated with the SPAD value. The results of Li Fengqing et al. (2018) showed that, in the correlation analysis among traits of *Ormosia hosiei* seedlings, compared with seedling height, ground diameter was more closely related to leaf trait indicators of seedlings; however, a similar effect was not observed in the present study. In addition, studies have also indicated that ground-diameter growth of *O. hosiei* seedlings is jointly constrained by plant photosynthetic area and root growth, whereas seedling height is more strongly affected by root growth (Ren Wenling et al., 2023). Other studies have suggested that plant seedling height is likewise jointly influenced by plant photosynthetic area and root growth (Zhang Qunfang et al., 2015). This study did not explore in depth the specific mechanisms by which roots and photosynthesis affect seedlings; these mechanisms may be investigated further in future research. Some studies have also shown that *O. hosiei* prefers neutral and slightly acidic soils, and that planting sites should have deep, fertile soil layers, sufficient moisture, and no standing water (Chu Xiuli et al., 2021). Subsequent studies may combine soil properties and other factors to examine their effects on the establishment of reintroduced seedlings in the wild.

This study examined only the ecological adaptation strategies of *O. hosiei* and its ex situ conservation under macro-environmental conditions (karst landforms, subtropical monsoon climate, altitude, slope aspect, etc.). First, this study revealed the resource-allocation strategy of *O. hosiei* seedlings (an allometric growth pattern, with preferential vertical growth). During reintroduction afforestation in the wild, habitats with relatively weak light competition or am-

ple space (such as semi-sunny slopes and sparse forest gaps) may be preferentially selected to match their resource requirements for vertical growth and reduce growth limitation caused by insufficient light. Second, the study revealed that seedling growth is limited by nitrogen; therefore, in ex situ conservation, nitrogen-fertilization management should be strengthened in a targeted manner. For example, an appropriate amount of quick-acting nitrogen fertilizer may be applied (in view of the possible rhizobial symbiosis of leguminous plants, inoculation with efficient nitrogen-fixing bacteria may be combined) to alleviate nitrogen limitation and promote seedling growth. Finally, the study found that the intrinsic water-use efficiency of *O. hosiei* was relatively low. In ex situ conservation practice, habitats with stable water conditions (such as moderate soil moisture and no severe seasonal drought) should be selected, and afforestation in areas with extreme drought or poor drainage should be avoided. At the same time, measures such as mulching and moisture conservation may be used to maintain soil moisture and compensate for its low water-use efficiency. In addition, future research may combine genetic improvement (screening provenances with high intrinsic water-use efficiency) and habitat optimization (increasing nitrogen fertilizer) to improve the success rate of wild reintroduction. However, to comprehensively reveal the ecological adaptation strategies and ex situ conservation mechanisms of *O. hosiei*, key environmental-factor data should be supplemented on the basis of existing research, and multidimensional analytical frameworks should be incorporated, such as data on soil physicochemical properties (nutrient availability and physicochemical properties), climatic factors (light, moisture, and temperature), and biotic interaction factors (as a leguminous plant, *O. hosiei* may coexist symbiotically with rhizobia; rhizobial diversity and nitrogen-fixation activity should be supplemented). Adaptability analysis based on environmental factors and plant traits can provide an integrated scheme of "precise habitat screening–nutrient-oriented regulation–optimized community configuration" for ex situ conservation, thereby improving the success rate of wild reintroduction.

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