Changes in leaf functional traits of *Houttuynia cordata* in response to soil environmental factors in Anqing city of Anhui Province in China

Xiaopeng Wang¹, Meirong Ye², Xueping Zhang³, Rusong Xu⁴ and Dongqing Xu⁵

¹College of Life and Health Sciences, Anhui Science and Technology University, Fengyang, China; ²College of agriculture, Anhui Science and Technology University, Fengyang, China

ABSTRACT

Plant functional traits reflect the responses and adaptations of plants to their immediate environment. In this study, 10 different sources of the *Houttuynia cordata* population in Anhui Province, China, were examined in terms of their leaf responses and adaptive mechanisms towards soil characteristics. Our study observed the leaf weight ratio depicted a significant negative correlation with leaf potassium content ($r=0.652$, $P < 0.05$) and an extremely significant negative correlation with leaf saturated water content ($r=-0.720$, $P < 0.01$) in different *H. cordata* populations. The leaf mass per area, leaf weight ratio, leaf nitrogen content, leaf potassium content, and leaf saturated water content in *H. cordata* showed a significant correlation with soil available phosphorus, available potassium, soil total porosity, soil volume weight, and pH. Lastly, a significant relationship between leaf potassium content in plants and soil volume weight was observed ($R^2=0.448$, $P < 0.01$).

Plants have an intimate relationship with their habitat and environmental changes greatly affect plant functional traits, geographical distribution, community composition, and even ecosystem functions. During plant–environment interactions, changes in plant internal structure and external morphology results in co-evolution of different functions. This results in responses to external environmental changes and a generation of a series of plant traits that act on the ecosystem [1,2]. These traits are known as plant functional traits and are typically measurable. These traits are intimately associated with plant life processes such as growth, reproduction, and death [3]. These traits can reflect the capabilities of plants in resource acquisition, utilization, and storage, and can strongly affect ecosystem processes [4]. Hence, these traits are indicators of the mutual relationship between plants and the environment. For a long time, plant response and adaptive strategies towards the environment have been a focus of ecological research. Through self-adaptation, regulation and changing certain functional traits in response to environmental changes, plants form different growth, reproduction, defense, and other survival strategies. Therefore, studying the relationship between plant functional traits and the environment can better reveal their response characteristics and adaptation patterns towards the environment, the intrinsic relationship between plant traits and spatial heterogeneity towards the environment, and the results of adaptive evolution towards environmental changes [5]. Among plant functional traits, leaf functional traits are most intimately associated with plant growth and reproduction [6]. These traits can reflect the growth, metabolism, and development of plants, as well as reveal plant adaptive strategies in response to environmental changes [7,8]. Leaf area can be used to characterize the photosynthetic capacity of leaves, whereas leaf nitrogen levels directly determine the magnitude of plant photosynthetic capacity [9]. The size of leaf surface stomata and stomatal density control evapotranspiration and gaseous exchange between plants and the environment, and directly affect photosynthesis and transpiration in plants [10].

Ecologists have always been enthusiastic in studying the responses of plant functional traits to environmental changes. Examples include an increase in leaf area, leaf nitrogen, and phosphorus content with greater precipitation and a decrease in these traits with increasing air temperature [11,12]. Spatial heterogeneity due to varying terrain results in the redistribution of temperature and precipitation, which in turn affect soil temperature and soil moisture content, thereby changing plant functional traits [13]. Altitude and concavity are key terrain factors that affect plant functional traits in subtropical evergreen broad-leaf forests, and soil moisture content and nitrogen content are major soil factors that influence spatial variability in plant functional traits. Theoretically, soil moisture content is mainly determined by the canopy area, transmittance, litter quantity, and soil porosity [14,15]. However, research studies on the responses of plant functional traits to changes in soil characteristics are limited. In an ecosystem, reports on the plant
functional trait responses of a population to adapt to soil characteristics are scarce.

*Houttuynia cordata*, also known as fish mint, is an herbaceous perennial plant belonging to the genus *Houttuynia* and family Saururaceae. The entire herb can be used as a medicine, and its young leaves and rhizomes can be used as vegetables. This plant prefers moist and dark environments and is located on ubac slopes, field ridges, ditches, beside streams, wetland bushes, and forests. In China, *H. cordata* communities are located in the Yangtze River basin, Huai River basin, northwest China, northern China, and Tibet [16]. These plants reproduce and use their developed rhizomes to expand their territory and change their internal physiological levels to adapt to various environments [17–20]. In this study, we studied leaf functional traits and changes in soil physical and chemical properties of different *H. cordata* populations in Anhui Province to examine the relationship between leaf functional traits of *H. cordata* and soil characteristics. Understanding the response mechanisms of leaf functional traits to changes in soil characteristics has ecological significance.

1. Materials and methods

1.1. Overview of the study area

We chose Yaoluoping National Nature Reserve and Hongwu Village as the sampling sites. These two areas represent two kinds of topographic conditions in Anqing area of Anhui Province (Dabie Mountain area and the plain hilly area along the Yangtze River), which are representative topography of Anhui Province. And the *Houttuynia cordata* is widely distributed in these two areas.

The Yaoluoping National Nature Reserve is located in northwest Yuexi County of Anhui Province (N30° 40′–31°06′, E116°03′–116°33′). The main peak, Duozhijian, has the highest elevation (1,721.5 m asl), while Liyuwei has the lowest altitude (500 m asl). The climate of this nature reserve is considered subtropical monsoon. The region has zonal soil types: hilly yellow brown soil at altitudes below 800 m and hilly yellow brown soil and hilly meadow soil at altitudes above 800 m. The plants in the Yaoluoping Nature Reserve have an ancient origin with many endemic species and rich plant biodiversity. The zonal vegetation consists of mixed deciduous and evergreen broad-leaf forests. The Hongwu Village, Mamiao Town, Huaining County, Anhui Province, lies at the intersection of three county-level cities (Tongcheng, Huaining, and Qianshan). This village is located on the plains and low hills at the lower and middle reaches of the Yangtze River. The terrain is steep on the southwest and relatively flat to the northeast. The climate is subtropical humid monsoon; therefore, it is mild, has distinct seasons, and rainfall and high temperatures often occur simultaneously, with moderate precipitation and abundant sunshine. The soil consists mainly of red soil with high leaching characteristics. The soil parent material consists mainly of granite, syenite, shale, and quaternary-red clay.

1.2. Field survey and sampling

In June 2017, field survey and soil sampling were conducted at the *H. cordata* habitats in Yaoluoping Nature Reserve (sample plots 1–6) in Anhui Province and Hongwu Village, Mamiao Town, Huaining County (sample plots 7–10). Three 1 m × 1 m quadrats were randomly selected in each of the 10 sample plots and were divided into 20 cm × 20 cm small quadrats. Table 1 shows the environmental information for the study site.

1.3. Soil survey and sample analysis

Sampling was conducted in standard sample plots. Three sampling points were randomly selected within each quadrat, and 100 cm³ cutting rings were used to collect samples at the 0–10 and 10–20 cm soil layers. These samples were used to measure soil porosity (STP), soil volume weight (SVW), and soil moisture content (SMC). At the same time, five soil samples were collected in an ‘S’ shape. Subsequently, 1 kg of mixed soil samples was collected, and plant roots and rocks were removed. The soil samples were transported back to the laboratory, air-dried, and crushed. After passing through 1-mm and 0.25-mm sieves, the chemical characteristics of the soil samples were measured.

The cutting ring method was used to measure SVW, and total porosity was obtained by calculating the SVW and soil specific gravity (SSG). The soil specific gravity (SSG) was determined by specific gravity bottle method. Briefly, the following equations were also used: SSG = (g × d_w)/ (g + g_1 – g_2), g means the weight of dried soil; g_1 means the weight of specific gravity bottle and water at t °C; g_2 represents the weight of specific gravity bottle, water and soil sample at t °C; and d_wt represents the specific gravity of distilled water at t °C [21]. Soil organic carbon (SOC) was measured using the potassium dichromate solution heating method, available nitrogen (SNC) was measured using the Kjeldahl method, available phosphorus (SPC) was measured using molybdenum-antimonyl colorimetry, and soil available potassium (SKC) was measured using flame photometry [21,22].

1.4. Plants sampling and analysis

Three 1 m × 1 m quadrats were selected in each sample plot, and the adjacent lattice method was employed. The species name and the quantity of plants were recorded, and entire *H. cordata* were harvested. The fresh mass of the entire plant and leaves was measured.
A Jindongmei scanner (grayscale mode, resolution: 300 dpi) was used to measure and calculate leaf area (LA). The ratio of the total leaf area of the *H. cordata* and the number of leaves in a sample plot is the mean LA (cm²). Four to five leaves were randomly selected from each sample, and a Vernier caliper (accuracy: 0.01 mm) was used for measurement. Five leaves were measured each time in triplicate. The mean value is the mean leaf thickness (LT).

Four epidermis tissues (4–10 mm²) were selected from each leaf and fixed with fixation solution. The leaves were transported back to the laboratory for sectioning and observed using an Olympus CX22 (400×) optical microscope. A digital microscope interactive platform was used to acquire images (image processing software: MIE2.0), and the number of stomata was calculated. This was divided by the area of the selected region to obtain the mean stomatal density (number/mm²). Stomatal density (SD, num/mm²) was computed as follows: 

\[
SD = \frac{\text{number of stomata}}{\text{area in field (A)}}
\]

The samples were transported back to the laboratory and fixed at 105°C for 10 min and then dried to a constant weight at 50°C. The dry weights of the entire plant and leaves were obtained. An electronic balance (accuracy: 0.0001 g) was used to measure the leaf water-saturated fresh mass (g) of the samples. After drying at 60°C to a constant weight, the leaf dry weight (g) was measured and used to calculate saturated water content (LWC). The following equations were also used:

\[
\text{Saturated water content (LWC) } = \frac{\text{Leaf water} - \text{saturated fresh mass}}{\text{Leaf dry weight}}/\text{Leaf water} - \text{Saturated fresh mass}
\]

\[
\text{Leaf mass per area (LMA)} = \frac{\text{Leaf dry weight}}{\text{Dry weight of the entire plant}}
\]

Dried leaf samples were crushed and passed through a 100-micron sieve before use. Leaf nitrogen, potassium, and phosphorus content (LNC, LPC, and LKC, respectively) were measured using the same methods employed for soil measurements [21,22].

### 1.5. Statistical analyses

Excel 2003, SPSS 19.0, and R 2.13.1 were used for statistical analysis of the data. One-way ANOVA and LSD multiple comparison were used for the analysis of soil markers and leaf functional traits in different sample plots. Pearson correlation analysis, principal component analysis (PCA), and stepwise regression analysis were used to analyze the correlation between soil factors and leaf functional traits, and PCA sorting of soil factors and leaf functional traits was performed.

### Table 1. Environmental information of the study site.

| Site | Province | Annual precipitation (mm) | Annual temperature (°C) | Sample ID. Plot | Latitude | Longitude | Elevation (m) | Aspect | Gradient (°) | Transparence of community |
|------|----------|--------------------------|-------------------------|-----------------|-----------|-----------|--------------|--------|-------------|--------------------------|
| Yuexi County, Anhui Province | 1400–2000 | 1450–2000 | | 1 | Maoerlong (understory of a mixed broadleaf forest) | E116°04′527″ | N30°59′709″ | 995 | SW | 24.2 | 2.87 |
| | | | | 2 | Maoerlong (at the edge of a mixed broadleaf forest) | E116°04′734″ | N30°59′709″ | 991 | NW | 18.13 | 91.46 |
| | | | | 3 | Maoerlong (beside a bamboo forest) | E116°04′998″ | N30°59′709″ | 997 | SE | 5.3 | 5.70 |
| | | | | 4 | Meili village (understory next to a ditch) | E116°04′642″ | N30°59′709″ | 910 | SE | 25.7 | 65.37 |
| | | | | 5 | Waxiepai (adjacent slope) | E116°04′538″ | N30°59′709″ | 917 | SE | 17.7 | 70.81 |
| Huaining County, Anhui Province | 1425.3 | 16.4 | | 6 | Hongwu (understory at the roadside) | E116°46′292″ | N30°43′164″ | 49 | SE | 1.2 | 55.46 |
| | | | | 7 | Hongwu (behind the house, understory of a *Pinus massoniana* forest) | E116°46′166″ | N30°43′158″ | 38 | SW | 16 | 55.47 |
| | | | | 8 | Hongwu (understory next to a sweet potato field) | E116°46′642″ | N30°43′164″ | 52 | SE | 11 | 53.85 |
| | | | | 9 | Hongwu (beside cultivated land, below a reed bush) | E116°46′242″ | N30°43′164″ | 28 | SE | 28 | 72.69 |
The data in the figures and tables were presented as the mean ± standard error.

2. Results and analysis

2.1. Soil physical characteristics of Houttuynia cordata habitats

Figure 1 shows that there are significant differences in soil moisture content among different H. cordata habitats (P < 0.05). Among these habitats, soil moisture content was the lowest for Maoerlong sample plot 2 and the highest for Hongwu sample plot 8. SVW intuitively reveals the degree of soil compaction and reflects the resistance of moisture, air, heat, and root extension. SVW affects SSG and STP. There were significant differences in the SVWs of different H. cordata habitats (P < 0.05), of which the SVWs of sample plots 2, 3, and 7 were moderate (1.0–1.25 g/cm³) and demonstrate typical SVW characteristics. Overall, the SVW of the H. cordata habitats in Hongwu was large. An example is sample plot 8, which had the highest SVW with the least variation (1.45–1.55 g/cm³). Sample plots 9 and 10 also exhibited little variation (1.25–1.35 g/cm³). On the other hand, the SVW of H. cordata habitats in Yaoluoping were generally lower. Among these sample plots, the SVWs of sample plots 1, 4, 5, and 6 were less compact (<1.0 g/cm³). This shows that the understory environment is good and the degree of soil arability is high.

SSG refers to the mass of solid particles per unit volume of soil and is mainly determined by the mineral composition and the relative organic matter content of the soil. The SSGs of different H. cordata habitats were relatively higher (2.51-2.84 g/cm³). Among these sample plots, 2, 7, 8, and 9 have relatively higher SSGs and were significantly different from the other sample plots. This suggests that the degree of rainwater leaching in these sample plots is greater. Soil pores are channels formed when gaseous and liquid substances in soil migrate. The size, quantity, and spatial structure of soil pores determine the form and rate of material migration in soil, and STP is one of the important factors that are used to assess soil characteristics, such as soil water and air storage, and is one of the basic characteristics for assessing soil fertility. There were significant differences in STP between various H. cordata habitats (P < 0.05). Among these sample plots, the STPs of sample plots 1, 2, 4, 5, 6, and 7 were higher than 60%, suggesting abundant soil water and air storage. The STP of sample plots 3 and 9 were 50–60%, whereas the STP of sample plots 8 and 10 were slightly lower (45–50%). Overall, the STP of various Hongwu H. cordata
habitats was generally lower, of which sample plot 8 was the lowest. This shows that there is intense soil nutrient leaching and human interference.

### 2.2. Soil chemical characteristics of *H. cordata* habitats

Figure 2 shows that the soil pH levels of different *H. cordata* habitats were all lower than 6.5 and are therefore generally acidic. In addition, there are significant differences among sample plots ($P < 0.05$). Among these sample plots, sample plots 1, 2, 3, 6, 7, and 9 have slightly acidic soils (pH 5.5–6.5), and the soils of sample plots 4, 5, and 8 were acidic (pH 4.5–5.5). There were significant differences in the SKC among different sample plots ($P < 0.05$), which were all higher than 80 mg/kg. Soil in these plots was considered nutrient-rich, reaching levels of superior farmland (80–150 mg/kg, Grade 2) and above. Among these sample plots, the SKC of sample plots 5, 6, 8, and 10 were higher than 200 mg/kg (Grade 1), which belongs to the soil type with the highest potassium content. There were significant differences in SNC among different sample plots ($P < 0.05$) and SNCs showed variability. The SNC of sample plots 4, 5, and 6 were extremely high (>150 mg/kg), that of sample plots 1, 7, 8, and 10 were high (90–100 mg/kg), sample plot 9 was moderate (60–90 mg/kg), and soil in sample plots 2 and 3 were extremely low (<45 mg/kg). There were significant differences in SPC among different sample plots ($P < 0.05$), which were generally low. The SPC of sample plots 3, 4, and 6 was moderate (30–60 mg/kg), that of sample plots 1, 2, 5, 8, and 9 was lower (15–30 mg/kg), and sample plot 10 approached the lowest threshold for low soil grade. The SPC of sample plot 7 was extremely

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**Figure 2.** Differences in soil chemical characteristics in different *Houttuynia cordata* populations.
low (<15 mg/kg), a PSC value of only 5.662 mg/kg, which resulted in the highest nitrogen/phosphorus ratio (17.154). The SOC of different *H. cordata* habitats was generally higher, and the differences were significant (*P* < 0.05). Among all sample plots, sample plots 1, 7, and 10 had high SOC (3–4%), and sample plots 2, 3, 4, and 5 had moderate SOC (1–2%) levels. There were significant differences in the nitrogen/phosphorus ratios of different sample plots, and the differences were large.

### 2.3. Correlation analysis and principal component analysis of different soil factors in *H. cordata* habitats

Table 2 shows that the SPC in *H. cordata* habitats exhibits a significant negative correlation with SOC and the nitrogen/phosphorus ratio (<0.05). SVW shows an extremely significant positive correlation with SSG (*P* < 0.01), but has an extremely significant negative correlation with STP (*P* < 0.01). SSG shows a significant negative correlation with STP (*P* < 0.05). In addition, SKC is negatively correlated with SPC and STP but positively correlated with SNC, SOC, the nitrogen/phosphorus ratio, SMC, and SVW. SNC is positively correlated with the nitrogen/phosphorus ratio and STP but negatively correlated with SSG and SVW. SPC is negatively correlated with SVW. SOC is positively correlated with the nitrogen/phosphorus ratio and SMC. SMC is positively correlated with SSG and SVW, but negatively correlated with STP, although this was not significant (*P* < 0.05).

Table 3 shows that the communality of SVW was the highest for all soil physical characteristics in *Houttuynia cordata* habitats, followed by STP and SSG. Comparatively, SMC was the lowest at 0.36. Based on the principle of eigenvalue >1, one principal component (eigenvalue = 0.295) was extracted. The contribution of this principal component was 73.8%.

The principal component formula was: 

\[ y = 0.35Z_x + 0.46Z_x^2 + 0.11Z_x^3 - 0.55Z_x^4 + 0.45Z_x^5 \]

The principal component formula was: 

\[ y_1 = 0.09Z_x + 0.46Z_x^2 + 0.11Z_x^3 - 0.55Z_x^4 + 0.45Z_x^5 \]

\[ y_2 = 0.09Z_x + 0.46Z_x^2 + 0.11Z_x^3 - 0.55Z_x^4 + 0.45Z_x^5 \]

In the equation, \( y_1 \) represents principal component 1, \( y_2 \) indicates principal component 2, and \( Z_x \) is the standardized variable.

This shows that these two principal components are the main causes of changes in soil physical characteristics. The initial factor loading matrix of the principal components (Table 4) shows that SPC has a significant negative correlation with the first principal component, and the nitrogen/phosphorus ratio shows a significant positive correlation with the first principal component. SNC shows a significant positive correlation with the first principal component, while soil pH shows a significant negative correlation with the first principal component. PCA indicates that markers that show significant positive or negative correlations with the first principal component (i.e. SPC and nitrogen/phosphorus ratio) can be used as major markers for soil chemical characteristics, followed by SOC and SKC.

### 2.4. Leaf functional traits of different *H. cordata* populations and correlation analysis

Figure 3 shows that there are significant differences in LMA among *H. cordata* populations, with the LMA of *H. cordata* populations in Yaoluoping being higher than Hongwu. Sample plot 4 has the highest LMA,
followed by sample plot 6, while sample plots 7, 9, and 10 have the lowest LMA. There were significant differences in LWR among different *H. cordata* populations, and the LWR of Yaoluoping *H. cordata* populations is also higher than the Hongwu populations. Sample plots 4–6 have the highest LWR, followed by sample plots 1 and 3, whereas sample plot 10 has the lowest LWR. There were significant differences in SD among different *H. cordata* populations; sample plots 4 and 6 have the highest SD, followed by sample plots 3 and 10, whereas sample plots 1 and 5 have the lowest SD. There were no significant differences in LNC between sample plots 1–3 and 6–10, but these were significantly higher than sample plot 5 that had the lowest LNC. There were significant differences in LPC among different *H. cordata* populations, with sample plot 9 having the highest LPC and sample plots 2 and 4 having the lowest LPC. There were no significant differences among sample plots. There were significant differences in LKC among different *H. cordata* populations, with sample plot 9 having the highest LKC and sample plot 5 having the lowest LKC. There were no significant differences in LKC among sample plots 1, 4, and 7–10. There were significant differences in leaf-saturated water content among the different *H. cordata* populations, with sample plots 5 and 7 showing the highest leaf-saturated water content, followed by sample plots 1, 2, 8, and 9, whereas sample plots 3, 4, and 6 had the lowest leaf-saturated water content. There were significant differences in LA among different *H. cordata* populations; sample plot 1 had the highest LA, which was significantly higher than sample plots 2–4, 6, and 10, and there was no significant difference with the other sample plots. There were significant differences in leaf thickness among the different *H. cordata* populations; sample plot 7 had the greatest leaf thickness and sample plot 2 had a slightly thinner leaf thickness. A significant decrease was observed in leaf thickness for sample plots 3 and 6, and another significant decrease was observed in sample plots 4, 8, and 9; however, these plots were significantly thicker than sample plots 1 and 5, and sample plot 10 had the thinnest leaves.

Table 5 shows that among different *H. cordata* populations, LMW has a significant positive correlation with LWR and LA, and an extremely significant negative correlation with leaf-saturated water content. LWR shows a significant negative correlation with LKC and an extremely significant negative correlation with leaf-saturated water content. SD has an extremely significant negative correlation with LA. LNC shows a significant positive correlation with LPC and LKC. LPC content has a significant positive correlation with LA.

### 2.5. Correlation analysis of leaf functional traits and soil characteristics among different *H. cordata* populations

A correlation analysis of leaf functional diversity index and soil physiochemical characteristics was performed (Figure 4). The results showed that the first axis explained 99.99% of the *H. cordata* leaf functional trait-soil characteristics relationship. SPC, SVW, STP,
SKC, and SMC have a greater correlation with *H. cordata* leaf functional traits, followed by the correlations of SSG, soil pH, and SNC. SVW, SNC, SPC, and SKC were all positively correlated with the first axis, of which SNC has a close relationship with the first axis, while SMC, STP, and SSG were negatively correlated with the first axis. This shows that SNC is an important fertility marker for the functional trait of leaf growth in *H. cordata*, whereas SPC is a key factor that affects the growth of leaf functional traits in *H. cordata*. STP imparts a negative effect, suggesting that loose soil aids in the functional trait of leaf growth in *H. cordata*. STP, SNC, and SPC were positively correlated with the second axis, whereas SOC, SMC, SVW, SSG, and SKC were negatively correlated with the second axis. Stepwise regression analysis of *H. cordata* leaf functional traits and soil factors indicated that there is an extremely significant positive correlation between LMA, SPC and SKC, and between STP and LWR. Moreover, an extremely significant negative correlation exists between SPC and leaf saturated water content in *H. cordata*, and a significant negative correlation exists between SVW and LKC and between soil pH and LNC. Leaf functional traits, such as LWA, LWR, LNC, LKC, and leaf saturated water content have a significant correlation relationship with SPC, SKC, STP, SVW, and pH. Finally, there is a significant positive correlation between soil pH and LPC (Table 6).

Figure 3. Differences in leaf functional traits in different *Houttuynia cordata* populations. Different letters mean significant differences at the 0.05 level.

Figure 4. RDA sequence for influencing factors of soil characteristics in different *Houttuynia cordata* populations.
(Note: SMC: soil moisture content; STP: soil total porosity; SVW: soil volume weight; SSG: soil specific gravity; SOC: soil organic carbon; SNC: soil available nitrogen; SPC: soil available phosphorus; SKC: soil available potassium).
Table 5. Pearson correlation coefficient matrix among leaf functional traits of *Houttuynia cordata* populations.

| Indicators | LMF | SD | LNC | LPC | LKC | LT | LSWC | LA |
|------------|-----|----|-----|-----|-----|----|-------|----|
| LMA        | 0.739** | 0.585* | -0.392 | -0.551* | -0.443 | 0.28 | -0.903** | -0.606* |
| LMF        | 0.196 | 0.524 | -0.524 | -0.276 | -0.652* | 0.102 | -0.720** | -0.025 |
| SD         | 0.254 | 0.097 | 0.287 | 0.254 | 0.634 | -0.712** | |
| LNC        | 0.548* | 0.555* | 0.392 | 0.154 | 0.191 | 0.556* | |
| LPC        | 0.498 | 0.205 | -0.059 | 0.213 | 0.1 | 0.1 | |
| LKC        | 0.498 | 0.205 | -0.059 | 0.213 | 0.1 | 0.1 | |
| LT         | -0.283 | -0.361 | 0.423 | |
| LSWC       | 0.145 | 0.118 | |
| LA         | -0.118 | |

* P < 0.05; ** P < 0.01. LMA: Leaf mass per area; LMF: Leaf biomass fraction; SD: Stomata density; LNC: Leaf N content; LPC: Leaf P content; LKC: Leaf K content; LT: Leaf thickness; LSWC: Leaf saturated water content; LA: Leaf area.

Table 6. Stepwise regression analysis on soil factors and leaf functional traits of *Houttuynia cordata* among different population.

| Indicators | Regression equation | Standardized regression coefficient | R² | P |
|------------|---------------------|-------------------------------------|-----|---|
| LMA        | LMA = 0.00128 + 0.000185PC | B_{PC} = 0.7520 | 0.5655** | 0.0121 |
| LMA        | LMA = -0.00438 + 0.000265PC + 0.00002 SKC | B_{SPC} = 1.0506, B_{SKC} = 0.5378 | 0.7695** | 0.0020; 0.0445 |
| LMF        | LMF = -0.1471 + 0.0085 STP | B_{STP} = 0.8128 | 0.6606** | 0.0058 |
| LNC        | LNC = -0.0441 + 0.0399 SpH | B_{SpH} = 0.6887 | 0.4743* | 0.0276 |
| LKC        | LKC = -0.3124 + 0.5081 SW | B_{SW} = 0.6693 | 0.4480* | 0.0343 |
| LSWC       | LSWC = 94.6323–0.2852 SPC | B_{PC} = -0.7881 | 0.6212** | 0.0068 |

3. Discussion

Plants and soil are environmental factors, and the succession of plant communities involves mutual interactions among plants, climate, and soil [23]. The growth and development status of plants is determined by the plant’s own physiological characteristics, as well as various environmental factors. Environmental factors have indirect effects on plant growth because terrain properties affect plants through other ecological factors. In this study, we analyzed samples that significantly varied in SVW, STP, SMC, pH, and soil nutrients. The survey targets were mainly located in six regions: the understory with poor transmittance (1, 4), the understory with high transmittance (7, 8, 9, and 10), the edge of the forest (2), next to a cultivated field (3), adjacent to a water body (5), and at an adret slope (6). Sample plot 1 had the lowest transmittance, and sample plot 3 had the highest transmittance. The results of the study showed that SVW, SSG, SMC, and SOC were higher in the understory soil with high transmittance, whereas STP and SNC were lower. In this study, the Yaoluoping National Nature Reserve and Hongwu Village were chosen as the sampling sites. These two sites have a similar climate. Across all the samples, the change trends of soil volume weight (SVW), soil specific gravity (SSG) and soil moisture content (SMC) are generally similar in the two sites. Overall, the Hongwu *H. cordata* habitats in Mamiao Town, Anqing City, are located in transitional regions between low hills and plains, and the terrain is relatively flat. The soil contains more clay, has a strong water storage capacity, and therefore soil volume weight (SVW) and soil moisture content (SMC) are generally higher. However, the sample plots in Yaoluoping are located at high elevations, on steep gradients, and in regions with hilly yellow-brown soil that mainly consists of sand. The parent material mainly consists of granite and monzogranite. Therefore, SMC is generally lower. However, the measure soil porosity (STP) and available phosphorus (SPC) are higher in the Yaoluoping site. It indicates that there are significantly chemical differences between the hilly clay and sand soil, which further affect the distribution of plants. Despite the differences in soil nutrient content among locations, different plants maintain nutrient homeostasis in leaves through their own absorption and regulation mechanisms, which is a result of their adaptation to the environment [24].

In recent years, changes in plant traits and their relationship with the environment have become one of the primary research topics in plant ecology. During the long evolutionary and developmental process, plants interact with the environment and gradually form morphological and physiological structures that adapt to changes in the external environment. These structures are mainly manifested as differences in leaves, root systems, seeds, and other plant traits. Plant traits that can respond to environmental changes are known as plant functional traits [2,25]. Leaf functional traits are one of the most important characteristics of plants and are widely used in plant functional trait research [26]. Leaf functional traits are intimately associated with plant growth strategies and resource utilization abilities, and demonstrate the survival strategy of plants to obtain a maximum carbon harvest. LA is associated with the relative growth rate, photosynthetic rate, and LNC of plants, and can be used to reflect the carbon acquisition strategies of...
plants. LA has important effects on the relative growth rate of plants and is the best indicator for the trade-off of physiological processes [27]. Plants with larger LA have a larger area to acquire resources, and therefore have a higher net photosynthetic rate [28]. However, plants with low LA can better adapt to resource-poor and arid environments [29]. In this study, we observed higher specific leaf area in sample plot 1, which had the lowest transmittance, whereas LWR, LNC, LPC, and LKC were maintained at high levels. In sample plots with low transmittance (7, 8, 9, and 10), the specific leaf area, LNC, LPC, LKC, and leaf-saturated water content in *H. cordata* were relatively high. Studies have shown that leaf functional traits are influenced by environmental heterogeneity [30–33]. Plants adjust their leaf morphology and chemical composition in response to environmental changes [31]. Leaf thickness is an extremely valuable trait and is associated with resource acquisition, water storage and assimilation. In this study, we found that when SMC is low and transmittance is high, such as at the edge of forests (2), the edge of cultivated fields (3), and on adret slopes (6), the leaf thickness and specific leaf area of *H. cordata* are higher. An increase in leaf thickness or density helps to increase the distance or resistance of dispersion of internal moisture on the leaf surface, thereby decreasing internal moisture loss. However, leaf thickness is negatively correlated with specific leaf area and leaf-saturated water content, which agrees with the findings of Liu et al. [31]. In this study, we also found the leaf nitrogen, potassium, and phosphorus content (LNC, LPC, and LKC, respectively) were higher in Hongwu Village site, the results were consistent with leaf area (LA) and leaf-saturated water content (LSWC). The *H. cordata* is a typical medicine and edible homologous, which has been used for the treatment of human immunodeficiency virus, influenza virus, herpes simplex virus, and chronic sinusitis and nasal polyps [34]. Our previous study showed the moderate light and enough water could change the physiological property and biomass of *H. cordata*, which increased the contents of total flavonoids, vitamin C and protein in the leaves, but lowered light and increased water quantity could reduce the content of crude fiber in the leaves [20,35]. The chemical constituent of *H. cordata* were closely correlated with leaf functional traits. And the correlation analysis between the two factors need to be further studied. Changes in leaf functional traits in *H. cordata* in different habitats may be due to the plant's sensitivity to the environment. Plants adjust their leaf morphology and chemical composition in response to environmental changes, resulting in differences in resource utilization efficiency. This also shows that *H. cordata* have stronger leaf morphology plasticity to respond to changes in the geographical environment.

Soil nutrient content determines the resource utilization strategies of plants and shapes the phenotype of community functional traits that affect plants on different slopes [36]. This can also cause changes in plants by affecting traits within species. In this study, leaf functional traits, such as LWA, LWR, LNC, LKC, and leaf-saturated water content were significantly correlated with SPC, SKC, STP, SVW, and soil pH. Pan et al. [36] studied the leaf functional traits of plants in the Karst hills of Guilin. Their results showed that the specific leaf area of plants on an ubac slope was significantly correlated with SNC. The results of this study showed that SPC and SKC significantly affected the LMA of *H. cordata*. This may be because available phosphorus and potassium are the most important active states in soil and can be directly absorbed and utilized by plants. At the same time, phosphorus and potassium are activators of many enzymes in plants, and phosphorus and potassium absorbed from soil can promote photosynthesis and protein synthesis in plants, thereby increasing the LMW of plants. Soil pH was found to be positively correlated with LPC. This may be because the form of phosphorus that exists in the soil is greatly affected by pH, which agrees with the conclusion of Chen et al. [37] who studied the interactions between soil pH and phosphorus content. This study also observed a significant relationship between LKC and SVW, showing that plant elemental nutrient absorption is affected by different soil types, resulting in variations in plant functional traits. Therefore, leaf functional traits of the same plant species are controlled by variations in environmental factors in different environments. At the same time, these functional traits adapt to the surrounding habitat.

4. Conclusions

Plant functional traits reflect the responses and adaptations of plants to their immediate environment and environmental factors have indirect effects on plant growth. In this study, we found there were significant differences in LMA, LWR, SD, LPC, LKC, LA, leaf-saturated water content and leaf thickness among different *H. cordata* populations. The SPC, SVW, STP, SKC, and SMC have a greater correlation with *H. cordata* leaf functional traits. The STP depicted a significant negative correlation with functional trait of leaf growth in *H. cordatas*. Meanwhile, our result showed that SNC is an important fertility marker for the functional trait of leaf growth in *H. cordatas*, whereas SPC was a key factor that affects the growth of leaf functional traits in *H. cordatas*.
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Disclosure statement

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