Postprint: Phenotypic Variation in Quercus phillyraeoides Populations on Granite Castle-like Peaks in Southern Fujian

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Abstract

Castle-like peaks are special mountains that protrude above the surrounding environment with a castle-like appearance. The phenotype of Quercus phillyraeoides (Fagaceae) populations on the castle-like peak landforms in the Wushan Scenic Area of southern Fujian, China, has undergone variation, with its most significant difference from the original type being the presence of persistent yellowish-brown stellate hairs on the leaf abaxial surface. To explain this phenomenon, this study investigated the distribution of Q. phillyraeoides populations on the summit of the castle-like peak, including the mutant type, original type, and transitional type therein, as well as populations at the foot of the castle-like peak and another typical original type population from a different region. Based on leaf epidermal anatomy, using analysis of variance to statistically examine differences in microscopic traits, and combined with ecological factors, the study explored the causes of variation and possible evolutionary pathways. The results showed: (1) The mutant type formed an evergreen broad-leaved shrubland community where it was the dominant species on the west side of the summit, while the small shrubland patches on the east side were dominated by the original type, within which there were also a few transitional types. (2) The density of single-celled trichome bases (STB) and compound trichome bases (CTB) on the leaf adaxial epidermis was very low across all populations, even <1 mm-2, which macroscopically appeared as glabrous. The mean STB density on the leaf abaxial epidermis increased progressively from the original type to the transitional type and then to the mutant type. Compared with original populations, the mean density of CTB or typical stellate hairs (Tst) on the leaf abaxial epidermis in mutant populations varied from 0.481~1.122 mm-2 to 57.2 mm-2, which macroscopically appeared as dense pubescence. The stomatal length-to-width ratio increased from the original type (1.187~1.205) to the mutant type (1.258). (3) Analysis of variance results showed that STB and CTB densities on the leaf abaxial epidermis and

stomatal length-to-width ratio in the mutant type were significantly greater than those in the original type (P$ $0.01), but stomatal density and size showed no pattern or significant differences among populations ($P>0.05$). The soil on the summit of the Wushan castle-like peak is infertile with poor hydrothermal conditions. Integrating the habitat and statistical results, it can be concluded that the variation in leaf abaxial trichomes and stomatal length-to-width ratio in mutant Q. phillyraeoides has functions for resisting drought or extreme temperatures. Since pollen flow has not been interrupted, compared with geographic isolation, the special habitat promoting adaptive evolution in Q. phillyraeoides should be the main cause of this variation. This case provides direct evidence that special subtropical granite landforms promote population differentiation and speciation.

Full Text

Phenotypic Variation of *Quercus phillyreoides* Populations on Granite Castle Peaks in Southern Fujian, China

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Abstract

Castle peaks are distinctive mountains that resemble castles and protrude prominently above their surroundings. In the Wushan Scenic Area of southern Fujian, China, populations of *Quercus phillyreoides* (Fagaceae) inhabiting castle peak landforms exhibit phenotypic variation, most notably characterized by persistent yellow-brown stellate hairs on the leaf abaxial surface that differ significantly from the typical form. To explain this phenomenon, we investigated the distribution of *Q. phillyreoides* populations on the summit of castle peaks, including variant, original, and intermediate forms, as well as typical original populations at the foot of the castle peak and in another allopatric area. Based on leaf epidermal anatomy, we used analysis of variance to quantify differences in micromorphological traits and examined ecological factors to explore the causes of variation and potential evolutionary pathways. The results showed: (1) The variant form established an evergreen broad-leaved shrub community dominated by itself on the west side of the summit, while a small scrub patch on the east side was dominated by the original form, within which a few intermediate individuals occurred. (2) The density of simple trichome bases (STB) and compound trichome bases (CTB) on the leaf adaxial epidermis was extremely low ($<1\ mm^{-2}$) across all populations, appearing glabrous macroscopically. Mean STB density on the leaf abaxial epidermis increased progressively from original through intermediate to variant forms. Compared with original populations, mean CTB density or typical stellate trichome (Tst) density on the leaf abaxial epidermis in the variant population increased from 0.481–1.122 mm^{-2} to 57.2 mm^{-2}, macroscopically forming dense pubescence. Stomatal length-width ratio also increased progressively from original forms (1.187–1.205) to the variant form (1.258). (3) ANOVA revealed that STB and CTB densities on the leaf abaxial epidermis and stomatal length-width ratio were significantly greater in the variant form than in original forms ($P \leq 0.01$), whereas stomatal density and size showed no consistent patterns or significant differences among populations ($P > 0.05$). The soil on the summit of Wushan castle peak is impoverished with poor water and heat conditions. Integrating habitat characteristics with statistical results, we conclude that the variation in leaf abaxial trichomes and stomatal length-width ratio in variant *Q. phillyreoides* confers resistance to drought and extreme temperatures. Since pollen flow has not been interrupted, adaptive evolution driven by the special habitat, rather than geographical isolation, appears to be the primary cause of this variation. This case provides direct evidence that subtropical granite landforms with special characteristics can promote population differentiation and speciation.

Keywords: continental island system, *Quercus phillyreoides*, leaf micromorphological traits, natural selection, population differentiation

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Introduction

Granite castle peaks (hereafter "castle peaks") represent a distinctive granite landform type surrounded by steep vertical cliffs that create a castle-like appearance, sometimes forming continuous wall-like mountain ridges. The castle peaks along China's southeastern coast developed from moderate tectonic uplift after the Neogene, shaped by chemical weathering and fluvial incision (Cui et al., 2007). Castle peaks constitute a transitional granite landform type between stone forests and spheroidal weathering boulders. Their characteristics include precipitous surrounding cliffs with exposed rock faces and relatively flat summits where soil can accumulate. These peaks stand prominently among surrounding hills, creating ecological conditions fundamentally different from lowland areas (Porembski, 2007).

In tropical regions, another granite landform called "inselberg" resembles castle peaks but differs in that inselbergs experience only minor tectonic uplift and form isolated rock outcrops on plains through chemical weathering (Cui et al., 2007). Plant communities on inselbergs often differ from surrounding areas, an effect attributed to the unique habitats created by these isolated mountains (Porembski, 2007). Their degree of isolation depends on surrounding vegetation types, with ecological isolation compensating for relatively short geographic distances between inselbergs compared to true islands (Porembski, 2007). Ecological and distance isolation, combined with habitats distinct from foothill environments, collectively influence species evolution on inselbergs. Regarding species diversity, some studies suggest that factors such as area and environmental filtering show stronger associations with species richness than isolation distance (Conceição et al., 2007; Henneron et al., 2019), indicating that adaptive radiation occurs in heterogeneous habitats even at small spatial scales. Numerous studies have demonstrated that plant populations on inselbergs, whether distant or nearby, exhibit morphological and genetic structural variation with varying degrees of restricted gene flow, reduced within-population genetic diversity, and promoted population differentiation and speciation (Barbará et al., 2008; Byrne et al., 2019; Feliciano et al., 2022).

Inselbergs, as continental island systems, have long been used to observe ecological phenomena and speciation processes in both temperate and tropical regions (Porembski & Barthlott, 2012). China possesses a distinct geological history, and recent attention has focused on "sky islands" such as the Himalayas, Altai-Tianshan region, and Southwest China mountains, with numerous studies examining how topography and its changes, along with resulting climate shifts, influence phylogeographic structure (He & Jiang, 2014). Basins and gorges in Southwest China provided isolated microhabitat refugia for species during Pleistocene glacial periods, which became centers of genetic differentiation (Xie et al., 2012; Yang et al., 2012b). The uplift of the Qinghai-Tibet Plateau also enabled secondary contact and occasional hybrid speciation events between allopatrically differentiated species (Yang et al., 2012a; Zhang et al., 2019). Current evidence indicates that alpine plants on sky islands share a series of phylogeographic structures and evolutionary histories triggered by geological movements (Luo et al., 2016). However, evolutionary and ecological studies on granite castle peaks, peak clusters, peak walls, stone forests, and other landforms in China's subtropical regions remain rare.

The genus *Quercus* (sensu stricto) comprises approximately 300 species worldwide, with about 51 species, 14 varieties, and 1 form in China (Editorial Committee of Chinese Flora, Chinese Academy of Sciences, 1998). Due to complex reticulate evolutionary history, *Quercus* exhibits extremely high species diversity (Hipp et al., 2020), and has undergone adaptive evolution in various regions influenced by geography and paleoclimate (Yang et al., 2021). Leaf epidermal traits are often considered important characteristics for species delimitation (Deng et al., 2017). In November 2019, during a national survey of traditional Chinese medicine resources in the Wushan Scenic Area at the border of Yunxiao and Zhao'an counties in southern Zhangzhou, Fujian Province (Plate I: A, B), we discovered a *Quercus phillyreoides* population with dense, persistent yellow-brown stellate hairs covering the leaf abaxial surface (other traits showed no significant differences). After one year of continuous observation, we found that the trichomes on mature leaves did not abscise. *Quercus phillyreoides* is a widely distributed evergreen broad-leaved tree in the *Quercus* section *Ilex* (Fagaceae) south of the Qinling-Huaihe line in China, also occurring in Japan. Young leaves are sometimes sparsely hairy, while old leaves are glabrous or have only sparse pubescence on the midrib of the leaf abaxial surface (Editorial Committee of Chinese Flora, Chinese Academy of Sciences, 1998). We therefore hypothesized that leaf epidermal characteristics in this population had undergone some variation and conducted a comparative micromorphological study of *Q. phillyreoides* populations on the summit and foot of the castle peak in Wushan Scenic Area and in another allopatric area. Through leaf anatomy, we obtained epidermal traits and analyzed the trait data using ANOVA. This study addresses three questions: (1) Which leaf epidermal traits have varied in the variant population, and what are the patterns and trends of variation? (2) What is the relationship between this variation and the castle peak habitat, and does it have ecological rationality? (3) What is the historical process of population differentiation, and does it have evolutionary rationality? By exploring these questions, we aim to provide evidence that subtropical granite landforms with special characteristics can promote population differentiation.

1.1 Study Area Overview

The main body of Wushan in Zhangzhou is located at the border of Zhao'an and Yunxiao counties in southern Fujian, adjacent to Guangdong Province. It is surrounded by the southeastern coastal plain, the northeastern Zhangjiang River, and the southwestern Dongxi River. The mountain extends approximately 45 km north-south and 20 km east-west, covering an area of about 900 km^2. The Wushan Scenic Area is situated in the central part of Wushan, with its main peak reaching 1,050.6 m in elevation. The mountain is characterized by cliffs and precipices on all sides, with slightly angular features and a relatively flat summit that stands 215 m above nearby valleys. The bedrock is granite that experienced moderate local tectonic uplift after the Neogene (Plate I: B).

The Hongqiyan peak of Wushan represents a juvenile to adolescent stage of granite landform development (Chen et al., 2009) and is a typical castle peak (Plate I: C). The summit retains remnants of a unified Miocene planation surface, while the foothills accumulate thick weathering crusts that develop into red soils (Cui et al., 2007). The region has a subtropical monsoon climate with mean annual precipitation of 1,519.6 mm (624 mm in summer, 121 mm in winter) and daily mean temperatures of 20–27 °C (maximum summer temperature 31 °C, minimum winter temperature 12 °C). The zonal vegetation is evergreen broad-leaved forest, with Fagaceae, Lauraceae, and Theaceae as dominant groups in this forest zone (Wu & Jiang, 1997).

1.2 Research Samples

Before sample collection, we examined the type specimens of *Quercus phillyreoides* and all ordinary specimens (with photographs) in the Chinese Virtual Herbarium (CVH, https://www.cvh.ac.cn/). Both type specimens

(Cavalerie, J. 3626) were glabrous-leaf types, and among 209 ordinary specimens, one specimen from Guangxi (Chen Zhaozhou 50295) resembled our observations at the top of Hongqiyan (significantly hairy leaf abaxial surface) with similar habitat and life form recorded on the collection label. However, this trait does not appear at the northern boundary of *Q. phillyreoides* distribution in China, indicating that the stable hairy leaf abaxial surface trait is not a direct environmental effect nor can it be simply explained by phenotypic plasticity in *Q. phillyreoides*. Meanwhile, we found not only hairy-leaf populations but also typical original forms distributed on the summit of Hongqiyan.

To explain these phenomena, we collected leaves and voucher specimens of original, variant, and putative intermediate forms from the summit of Hongqiyan, as well as original forms from the foot of Hongqiyan (Table 1). The intermediate form observed in the field was defined relative to the nearly completely glabrous leaf abaxial surface of the original form and the nearly complete trichome coverage (leaf abaxial epidermis invisible) of the variant form. That is, although the intermediate form possesses persistent hairs, its leaf abaxial epidermis remains clearly visible throughout the plant, falling between the two extremes. Due to the stability of the original form, we could use any allopatric *Q. phillyreoides* population to represent all *Q. phillyreoides* groups across the full habitat range, thereby illustrating this variation at the summit of Hongqiyan (Table 1). We studied five (very small) populations, randomly selecting three adult individuals per population (individuals spaced $ $20 m apart or at maximum separation distance), with four leaves sampled from each individual in the four cardinal directions. Considering that trichome coverage would obscure stomatal traits and hinder subsequent observation and counting, we collected one additional leaf per direction for the variant form and removed the trichomes with a scalpel. Leaves were divided into tip, middle, and base sections and preserved in FAA fixative (38% formalin:glacial acetic acid:70% ethanol = 1:1:18, v/v/v). Sample collection times were nearly identical. Leaf epidermal section preparation followed Deng et al. (2017).

1.3 Statistical and Analytical Methods

We observed leaf upper and lower epidermal cell anticlinal walls, trichome bases, trichomes, and stomata using a BK5000 optical microscope. Leaf epidermal terminology and characteristic descriptions followed Deng et al. (2017). One field of view (200×) was photographed per section. We used ImageJ 1.53f51 (https://imagej.nih.gov/ij/index.html) to count all stomata within each field of view and measure their dimensions (length and width of guard cell outer contours) (Arena et al., 2017). Trichome/trichome base numbers were counted manually in each field of view. Stomatal and trichome/trichome base numbers were then converted to density, and stomatal length-width ratios were calculated (ratio of length to width). Data from leaf tip, middle, and base sections for each leaf were averaged to exclude random error. Since measurements from the four cardinal directions showed no obvious patterns, all were included in the original dataset to reduce potential systematic errors from small sample sizes. We performed F-tests and multiple comparisons (LSD) on each trait in the original dataset using SPSS 25 and created box plots for traits showing significant results. Differences were considered highly significant at $P \leq 0.01$ and significant at $P \leq 0.05$.

2.1 Phenotypic Variation and Spatial Distribution

The inflorescences, cupules, and nuts of the variant *Q. phillyreoides* on the summit of Hongqiyan (Plate II: H), as well as other leaf traits (shape, size, texture, color, and margin), were identical to the original form. The only difference was the dense, persistent yellowish or yellow-brown stellate hairs covering the leaf abaxial surface, with stellate hairs also being relatively dense on one-year-old branches (Plate II: F–G).

The variant population grows on granite weathering crust on the summit of Hongqiyan (Plate II: A–D), forming large clusters on the west side of the summit and creating an evergreen broad-leaved shrub community dominated by the variant form, with associated species including *Machilus phoenicis*, *Vaccinium zhangzhouense*, and *Rhododendron simiarum.* A small scrub patch on the east side is dominated by the original form, which occurs in smaller numbers and is also scattered sporadically around the variant population periphery. Variant individuals in the narrow, lower-elevation northern area likely dispersed from the west side of the summit. Within the original population, we also found individuals with slightly more developed leaf abaxial hairs, which can be considered intermediate forms.

2.2.1 Leaf Adaxial Epidermis (Plate III: A–E)

Anticlinal walls of leaf adaxial epidermal cells had uniform thickness and four morphological types: straight, curved, undulate, and sinuous (Plate III: A, D–E), with no patterns among population types or locations. Except for original form 2 (population 4) at the foot of Hongqiyan, where simple trichome bases (STB) were not observed in the field of view, all other populations had varying amounts of both simple (Plate III: A) and compound trichome bases (CTB) (Plate III: B–C, E), but mean densities were within 7 mm^{-2}. Residual capitate trichomes (Ca) derived from STB were observed in original form 3 (population 5) from Qingyun Mountain, but no trichome types corresponding to CTB were found. These results indicate that STB and CTB densities on the leaf adaxial epidermis were very low (<1 mm^{-2}) across all population types, and even when trichome bases were present, trichomes did not necessarily develop or were easily shed, resulting in a macroscopically glabrous appearance. Statistical summaries are presented in Table 2 .

2.2.2 Leaf Abaxial Epidermis (Plate III: F–L)

Anticlinal walls of leaf abaxial epidermal cells showed gradual transitions from straight to curved to undulate within each population, with uniform thickness (Plate III: H, J). Distinct capitate trichomes (Ca) and simple trichome bases (STB) were observed in all populations (Plate III: I–L), with mean STB densities ranging from 0–10.125 mm^{-2}. As shown in Table 3 , mean STB density increased progressively from original through intermediate to variant forms. Although compound trichome bases (CTB) were present in all populations (Plate III: F, H–I, K), typical stellate trichomes (Tst) derived from CTB were only observed in the variant (population 1) and intermediate (population 2) forms from the summit of Hongqiyan and in original form 3 (population 5) from Qingyun Mountain (Plate III: F–G, I, L). However, in original form 3 from Qingyun Mountain, stellate trichomes on the leaf abaxial epidermis were restricted to the base near the midrib, with corresponding mean CTB density <1 mm^{-2}. In contrast, Tst in the variant form from the summit of Hongqiyan occurred on the midrib, any lateral veins, or secondary veins, with mean CTB density reaching 57.2 mm^{-2}, and CTB area decreasing progressively from midrib to lateral to secondary veins. The intermediate form had mean CTB density between these extremes at 26.076 mm^{-2}. These results indicate that compared with other populations, the variant population showed a clear increase in CTB or Tst, macroscopically appearing densely stellate-pubescent, while the intermediate form had visible stellate trichomes but with the leaf abaxial epidermis still clearly visible, and the original form was nearly completely glabrous. Additionally, all populations had anomocytic stomata (Plate III: H, J). Mean stomatal density ranged from 453–516 mm^{-2}, mean stomatal width from 24.494–25.812 m, and mean stomatal length from 28.906–31.047 m, with no consistent patterns in these three traits between variant and original forms. However, mean stomatal length-width ratio increased progressively from original forms (1.187–1.205) through the intermediate form (1.240) to the variant form (1.258). Statistical summaries are presented in Table 3.

2.2.3 Significance Testing

ANOVA results (Tables 2, 3) revealed highly significant differences among populations in leaf adaxial STB density ($P = 0.002$), leaf abaxial STB ($P = 0.006$) and CTB ($P = 0.0005$) densities, and stomatal length-width ratio ($P = 0.002$) ($P \leq 0.01$). Although leaf adaxial STB density showed highly significant differences among populations, post-hoc tests revealed that these differences were contributed by the variant form versus original forms 2 and 3, and by original form 1 versus original forms 2 and 3, indicating that this trait occurred relatively randomly on the leaf adaxial epidermis. Post-hoc tests for leaf abaxial traits showed that differences in leaf abaxial STB and CTB densities and stomatal length-width ratio were primarily caused by the variant form (Figure 1 [Figure 1: see original paper]). Specifically, STB and CTB densities in the variant form were significantly greater than in original forms, regardless of whether original forms were located on the summit, foothill, or allopatric area. The mean stomatal length-width ratio in the variant form was 1.258 (maximum 1.333), whereas original forms had means $ $1.205 (maximum 1.274). This trait was thus substantially increased in the variant form, although not visually obvious in images. The intermediate form had intermediate values for all traits, reflecting its transitional status (Figure 1). These results demonstrate that the variant form has undergone significant variation ($P \leq 0.01$) in certain leaf abaxial epidermal traits (STB and CTB densities, stomatal length-width ratio) compared with original forms. However, stomatal density and size, which are highly sensitive to habitat conditions, showed no significant differences among populations on the summit, foothill, or allopatric area ($P > 0.05$) (Table 3).

3.1 Castle Peak Habitat Promotes Population Differentiation

Cases of population variation and speciation caused by inselbergs, another granite landform, are common (Mota et al., 2020; Ruas et al., 2020; Tavares et al., 2022), often attributed to genetic drift and inbreeding resulting from limited gene flow due to distance isolation (Barbará et al., 2007; Mota et al., 2020). However, heterogeneous habitats are sometimes considered to play a greater role in driving differentiation among populations of the same species (Milá et al., 2010; Mallet et al., 2014). Although inselberg habitats are generally less suitable for species survival, inselbergs in some regions of the world, such as southeastern Brazil, Madagascar, and southwestern Australia, have become centers of biodiversity, presumably due to isolation and long-term persistence promoting differentiation (Porembski, 2007). Consequently, inselbergs often harbor many endemic species.

Hongqiyan in Wushan is a typical castle peak landform. Similar to inselbergs, its summit habitat differs markedly from the south subtropical monsoon climate zone at the same latitude. Its uniqueness is manifested in: high light intensity and long sunshine duration in summer; low nighttime temperatures in winter; large diurnal temperature ranges; short water retention time in soil due to rainwater erosion; inherently poor and thin soil; and strong prevailing winds. Generally, the same species growing under moderate drought versus suitable conditions shows increased stomatal density, but when drought intensifies further, density decreases while stomatal size continues to decrease under drought conditions (Xu & Zhou, 2008). However, *Q. phillyreoides* in different habitats of Wushan did not show this trend, a phenomenon also observed in *Quercus* species at larger spatial scales (Sánchez-Acevedo et al., 2022). Since stomatal density and size are often correlated (Hetherington & Woodward, 2003), different *Q. phillyreoides* populations showed no significant differences in stomatal size, similar to density.

To cope with the challenges of the Wushan summit habitat, *Q. phillyreoides* in our study appears to rely on trichomes and adjust stomatal shape, as results showed significant differences among populations in leaf abaxial STB, CTB, and stomatal length-width ratio. Trichomes can adsorb water droplets from the air and reduce transpiration (Vitarelli et al., 2016; Moles et al., 2020) and provide some thermal insulation in winter (Peng et al., 2015). For desert plants, epidermal hair changes vary when responding to different abiotic stresses such as cold, drought, salinity, high temperature, and strong radiation; mechanical damage, drought, long daylight hours, high light intensity, and salt stress typically lead to increased trichome length or density (Ma et al., 2015). The leaf abaxial STB and CTB densities in the variant form from the Wushan summit were significantly greater than in all original forms (Table 3, Figure 1). Although trichomes may be induced in extreme environments, the presence of original form 1 on the summit and the lack of significant differences between it and original forms 2 and 3 (Table 3, Figure 1) confirm that short-term environmental effects have limited capacity to shape leaf abaxial trichomes in *Q. phillyreoides*. Significantly increased stellate trichomes (Tst) on the leaf abaxial surface would undoubtedly help adaptation to drought caused by poor water storage in summit soils and high evapotranspiration, as well as to cold and freezing damage resulting from the low specific heat and rapid heat dissipation of granite mountains in winter. Capitate trichomes can synthesize, store, and secrete numerous metabolites (Zhang et al., 2018), and increased capitate trichomes may enhance resistance to summit environmental stress. Regarding the mechanism of environmental effects on trichomes, studies on *Arabidopsis thaliana* have shown that mutants with abundant trichomes have significantly increased C32+ fatty acid mixture content in stem and leaf wax layers compared with trichome-deficient mutants, and the wax C32+ fatty acid content covering trichomes has been confirmed to be higher than in surrounding cells. Trichomes can cooperate with wax layer biosynthesis to reduce evaporation and regulate temperature (Hegebarth et al., 2016). Studies on cucumber (*Cucumis sativus*) trichome mutants indicate that *GL1* and *TTG1* are core genes for leaf epidermal initiation, while *GL3* and *EGL3* are also necessary. Transcripts of these genes constitute positive regulatory factors (Liu et al., 2016). However, transcription of negative regulatory factors such as *TRY* and *CPC* can inhibit trichome initiation (Schellmann et al., 2002). Research on trichome development mechanisms indicates they are regulated by multiple genes (Zhang et al., 2018). As a product of summit environmental stress response, the leaf abaxial trichomes in variant *Q. phillyreoides* may have undergone processes such as mutation of negative regulatory factors or transposition/enhancement of positive regulatory factors. The intrinsic connection between castle peak summit environmental factors and trichome initiation requires further research.

Stomatal length-width ratio actually reflects stomatal aperture, whose changes depend on the combined effects of subsidiary cell turgor controlled by ions and sugars, cytoskeletal organization, transmembrane transport, and gene expression. Substantial evidence shows that a single stimulus signaling pathway is insufficient to change stomatal aperture (Hetherington & Woodward, 2003). Among the *Q. phillyreoides* populations in this study, stomatal aperture in original form 1 on the summit did not appear to differ significantly from original forms 2 and 3 at the foothill or Qingyun Mountain at the same time point (Table 3, Figure 1), similar to the pattern for trichomes. However, in the variant form, stomatal length-width ratio was significantly increased, indicating variation in stomatal shape. Drought, excessively high or low leaf temperature can reduce stomatal aperture through signal transduction (Jiang et al., 2015; Duan et al., 2019), leading to the straightforward inference that under equivalent conditions, the variant form has reduced transpiration to cope with drought or extreme temperatures, and enhanced mobility to close stomata in windy environments.

In summary, the castle peak landform is the source of this variation. The habitat of this landform, particularly the poor and thin soil, results in inferior water and heat conditions compared with surrounding areas. Variation in STB, CTB, and stomatal length-width ratio has positive adaptive value for this habitat. These trait variations should be stable, as they are not rapidly and directly formed by environmental stress; after one year of observation, they remained stable across different developmental stages in both variant and original forms.

3.2 Possible Pathways of Population Differentiation

Wushan formed directly from varying degrees of uplift and weathering erosion along China's southeastern coast after the Neogene (Chen et al., 2009). The Last Glacial Period of the Pleistocene caused high-elevation plants to descend to nearby "refugia" (Chen et al., 2011), and *Q. phillyreoides* likely recolonized the summit during the interglacial or Holocene. Rodents are the main contributors to seed dispersal in Fagaceae, but initial colonization of the castle peak summit may have relied on birds (Oh, 2015). After original *Q. phillyreoides* reached the summit, adaptive evolution under habitat stress would have given adaptive individuals advantages, allowing them to expand into populations. However, small-scale distributions of original forms (Plate II: D) or individuals scattered around the variant population still exist on the summit. This first indicates that the original form itself can tolerate extreme habitats such as drought, and second can be viewed as a remnant profile of population persistence based on natural selection and competitive exclusion principles. Additionally, chance events such as bird dispersal may also have major impacts on summit community succession. Both pathways could lead to the current coexistence of original and variant forms and produce intermediate forms through hybridization. This means that intermediate forms may represent either residual random variation individuals from the speciation process or products of hybridization. In any case, as a wind-pollinated tree species, pollen flow from surrounding lowland original forms should never have been interrupted (Ashley, 2021). The geographic isolation effect of Hongqiyan castle peak may be quite limited, and the special habitat of the castle peak has clearly played a greater role in promoting variation than geographic isolation. The current presence of intermediate forms on the summit and the absence of variant forms at the foothill indicate that variant forms could fuse with original forms and disappear. However, at present, castle peaks do produce certain ecological island effects.

3.3 Ancestral Traits and Parallel Evolution

Compared with stomatal length-width ratio, trichome variation shows more evolutionary patterns within *Quercus*. First, it must be clarified that the variant *Q. phillyreoides* from Wushan and closely related species with similar leaf abaxial trichomes are separated by straight-line distances exceeding 300 km, excluding a hybrid origin.

Research shows that STB is an ancestral trait in *Quercus*, present to varying degrees in different evolutionary lineages and showing significant density differences among species (Deng et al., 2017), as seen in the leaf upper and lower epidermis of Wushan variant and original *Q. phillyreoides*. However, molecular phylogenetic results show that CTB has originated or been lost multiple times in different *Quercus* lineages (Yang et al., 2017; Hipp et al., 2020), with varying conditions among species in section *Ilex*. Therefore, density variation in CTB is possible in the variant form. Trichome development is regulated directly and indirectly by multiple genes (Pu et al., 2003), selection promotes gene interaction and rearrangement, and sorting of ancient ancestral polymorphisms can also cause similar traits to arise repeatedly. Molecular methods are needed to analyze the genetic relationships among different form populations.

Additionally, *Q. phillyreoides* trichome variation was found in the Guangxi specimen (Chen Zhaozhou 50295) and in 2 of 56 additional specimens examined from the Global Biodiversity Information Facility (GBIF, https://www.gbif.org/) from the Ryukyu Islands of Japan (Wright, s.n.; Type of *Quercus wrightii* = *Q. phillyreoides*). If this is not a single-plant mutation, it suggests that parallel evolution may have occurred among them.

In conclusion, the leaf abaxial simple trichome base (STB), compound trichome base (CTB), and stomatal length-width ratio of the *Q. phillyreoides* variant form from Hongqiyan, Wushan, Zhangzhou, Fujian, have undergone natural variation compared with original forms, i.e., STB and CTB increased significantly ($P \leq 0.01$) and stomatal length-width ratio increased significantly ($P \leq 0.01$). The special habitat of the Wushan castle peak promoted this variation, which has certain ecological and evolutionary rationality. This provides effective evidence that subtropical granite landforms with special characteristics can promote population differentiation. *Quercus phillyreoides*, widely distributed in southern China and Japan, is an ideal subject for studying trichome initiation and evolution, parallel speciation, and related topics. China has one of the world's most extensive granite distributions, particularly concentrated in Guangdong and Fujian provinces (accounting for 30–40% of China's total) (Cui et al., 2007). Tectonic movements accelerate granite exposure, creating extremely diverse geomorphological landscapes, with moderately to highly uplifted castle peaks, peak clusters, stone forests, and isolated giant peaks widely existing at various spatial scales. Plant flora, community ecology, extremely small populations, and species evolution on special granite landforms deserve further investigation.

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