Research paper

A new *Quercus* species from the upper Miocene of southwestern China and its ecological significance

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A R T I C L E   I N F O

A B S T R A C T

Quercus praedelavayi Xing Y.W. et Zhou Z.K. sp. nov. is reported from the upper Miocene of the Xianfeng flora in central Yunnan, southwestern China. The fossil species is identified based on the detailed leaf morphological and cuticular examinations. The primary venation is pinnate and the major secondary venation is craspedodromous with regular spacing. Stomata are anomocytic and occur on abaxial epidermis. Trichome bases are unicellular and multicellular. The new fossil species shows the closest affinity with *Quercus delavayi*, an extant species distributing in southwestern China. The responses of the functional leaf traits to the climate change were studied by comparing the leaf characters of fossil species and its nearest living relative. The stomatal density of *Q. praedelavayi* is higher than *Q. delavayi*, which suggests a lower palaeoatmospheric CO_2 concentration during the late Miocene. The trichome base density of *Q. praedelavayi* is higher than the extant *Q. delavayi*. Considering the palaeoclimatic reconstruction of Xianfeng flora, it rejected the hypothesis that increase in trichome density is an adaptation to the drier environment.

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1. Introduction

Quercus L. is the largest genus in the family Fagaceae, with more than four hundred species (Nixon, 1993; Huang et al., 1999). It occurs throughout temperate and subtropical montane regions of the Northern Hemisphere, extending into the Southern Hemisphere in northern South America and Indonesia (Camus, 1936–1954; Nixon, 1993; Huang et al., 1999; Deng et al., 2008). Consistent with the present broad distribution of *Quercus*, the fossil record of this genus is also abundant, preserving as pollen, leaves, acorns, cupules, wood remains and inflorescences (e.g., MacGinitie, 1941; Axelrod, 1956, 1964; MacGinitie, 1969; Writing Group of Cenozoic Plants of China [WGCPC], 1978; Manchester, 1981, 1983; Kvaček and Walther, 1989; Zhou, 1993; Borgardt and Pigg, 1999; Kvaček and Walther, 2004; Liu et al., 2007). Although the geological history of this genus could be traced back to the early Paleocene, even the Cretaceous, from the fossil records (Bell, 1957; Van Boskirk, 1998), *Quercus* fossils before the early Eocene are obscure due to a lack of specific morphological and anatomical characters (Zhou, 1993). The earliest reliable *Quercus* fossil in North America is from the middle Eocene Oregon flora (Bones, 1979; Manchester, 1983). Fossil leaves and wood, as well as nuts and cupules showing modern forms of *Quercus* were found in this flora (Bones, 1979; Manchester, 1983). In Europe, the earliest leaf fossils with cuticular characters of *Quercus* were found in the middle Eocene Aschersleben flora of Germany (Kvaček and Walther, 1989). *Quercus* fossils were abundant in Eurasia and North America from the Oligocene to Quaternary (Zhou, 1993).

The circumscription of modern *Quercus* has been highly controversial (Jones, 1986; Manos et al., 2001). The status of subgen. *Cyclobalanopsis* (Oersted) C. K. Schneider in *Quercus* has been the topic of much discussion (Jones, 1986; Deng et al., 2006). Many taxonomists treat subgen. *Cyclobalanopsis* as a subgenus of *Quercus* (e.g., Camus, 1936–1954; Barnett, 1944; Nixon, 1993), while others treat it as a separate genus (e.g., Schottky, 1912; Hjelmqvist, 1948; Huang et al., 1999). The latter viewpoint was supported by the phylogenetic study (Manos et al., 2001). In this study, we also consider *Cyclobalanopsis* as a subgenus of *Quercus*. The subgen. *Cyclobalanopsis* is mainly distributed in tropical and subtropical Asia (Huang et al., 1999). Members of subgen. *Cyclobalanopsis* are important evergreen trees in the subtropical to tropical forest ecosystems. They are often found as a dominant element in broad-leaved evergreen forests in

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East Asia (Huang et al., 1999; Luo and Zhou, 2001). The fossil history of subgen. *Cyclobalanopsis* is important to understand its biogeography and speciation history, as well as the vegetation change in East Asia. *Quercus* (*Cyclobalanopsis*) *naitoi* Huzioka et Takahasi is the earliest fossil species of this subgenus which was found in the Eocene sediments of Japan (Huzioka and Takahasi, 1970). China is a major center of diversity for subgen. *Cyclobalanopsis* and contains 69 species in its subtropical to tropical areas, of which 43 are endemic (Huang et al., 1999). The earliest reliable fossils of subgen. *Cyclobalanopsis* in China were reported from the Oligocene Jinggu flora in Yunnan Province, southwestern China (WGCPC, 1978; Zhou, 1993). During the Neogene, *Cyclobalanopsis* became common and were often found as main elements in many East Asian fossil floras (WGCPC, 1978; Wang et al., 1982; Zhou, 1993; Ge and Li, 1999; Tao, 2000). Accurate identification of fossils facilitates better understanding of the geological history and the evolution of subgen. *Cyclobalanopsis*.

Cupule features are used as key diagnostic feature in the taxonomy of the Fagaceae (Forman, 1966; Fey and Endress, 1983; Crepet and Nixon, 1989; Chen et al., 2008). However, only a few cupules were preserved as fossils. The leaf morphological and epidermal features also provide valuable taxonomic and systematic data both in fossil and living Fagaceae (Jones, 1984, 1986; Kvaček and Walther, 1989; Zhou et al., 1995; Luo and Zhou, 2002; Liu et al., 2009). Jones (1984) examined thoroughly the foliar characteristics of extant Fagaceae using leaf morphological and cuticular analysis. His study provided a basis for the interpretation of putatively fagaceous fossil leaves as well as useful information for testing classification schemes of modern Fagaceae. Kvaček and Walther (1989) investigated various Cenozoic Fagaceae leaves using leaf epidermal features, and their results also supported the efficiency of the leaf cuticular characteristics in identifying fagaceous fossil leaves. Numerous studies on leaf morphology and epidermis of extant *Quercus* have been carried out in China which indicated that foliar characters could be used successfully to distinguish Chinese oak species (Zhou et al., 1995; Luo and Zhou, 2002). However, identification only based on gross morphology without cuticle characteristics might be problematic when some species share similar gross morphology but differ in their cuticular features. For instance, *Quercus glauca* Thunberg and *Quercus schottkyana* Rehder et Wilson are vicariant species occurring between eastern and southeastern China and the Yunnan–Guizhou Plateau respectively (Luo and Zhou, 2001). They are very similar in leaf shape, sizes, even the venation types but differ in trichome types. Consequently, cuticular analysis appears very important for the precise identification of *Cyclobalanopsis* fossils.

Our present study focuses on two goals: (1) to precisely identify a new fossil *Quercus* species based on leaf morphological and cuticular features; (2) to explore the evolutionary and ecological significance of this new fossil species.

2. Materials and methods

Fossils were collected from the Xiaolongtan Formation which exposed in the Xianfeng basin (Xing, 2010). The Xianfeng basin is located about 60 km north of Kunming, Yunnan Province, southwestern China (25°25′ N, 102°51′ E, 2200 m alt.; Xing et al., 2010, Fig. 1). The geological age of the Xiaolongtan Formation has been considered to be the late Miocene based on mammal fauna (Zhang, 1974; Dong, 2001), plant and pollen assemblages (Zhou, 1985; Wang, 1996; Zhou, 2000; Xia et al., 2009) and regional stratigraphic correlations (BGMRYP, 1996). The lithological sequence of this formation has been previously described (Xing et al., 1999; Wu et al., 2006; Xing et al., 2010, 2012). It comprises four members, named as $N1^1x$–$N1^4x$. The present fossils were collected in the layer of $N1^3x^2$, which also yielded abundant plant macrofossils, shells and insect fragments (Fig. 2). A primary classification showed that Fagaceae (especially the genus *Quercus*) and Lauraceae are dominant in this flora (Xing, 2010; Xing et al., 2012).

The fossil leaves were numbered and photographed using a Canon PowerShot S5 IS digital camera. Fossil leaf fragments were physically lifted off from the bedding surface with dissecting needles. In order to remove the calcareous and siliceous materials, the leaf fragments were first macerated with 20% hydrochloric acid (HCl) for 6 h, then with 40% hydrofluoric acid (HF) for 12 h, ultimately with 20% HCl for 2 h. After being rinsed in water, the leaf fragments were then treated by 3.5% sodium hypochlorite (NaClO) solution for about 30 min until they became white and translucent. The bleached fragments were then immersed in water and the mesophyll material was soon dispersed, leaving the separated upper and lower cuticles. The clean cuticles were stained in 1% aqueous solution of Safranin O for 3 min. and rinsed in water to remove excess stain, after dehydrated in glycerin for 30 min, mounted in glycerin jelly for light microscopy examination. Seven slides were made in total. Stomata density were counted based on two relatively clear slides, HLT 450A-abaxial-005 and HLT 450A-abaxial-007.

The epidermis of the extant *Quercus delavayi* Franchet for comparison were prepared with a 1:1 solution of glacial acetic acid (CH3CO2H) and 30% hydrogen peroxide (H_2O_2) and then placed in a hot-water bath at 80 °C for 3–5 h. Once the leaves turned white and transparent, epidermis
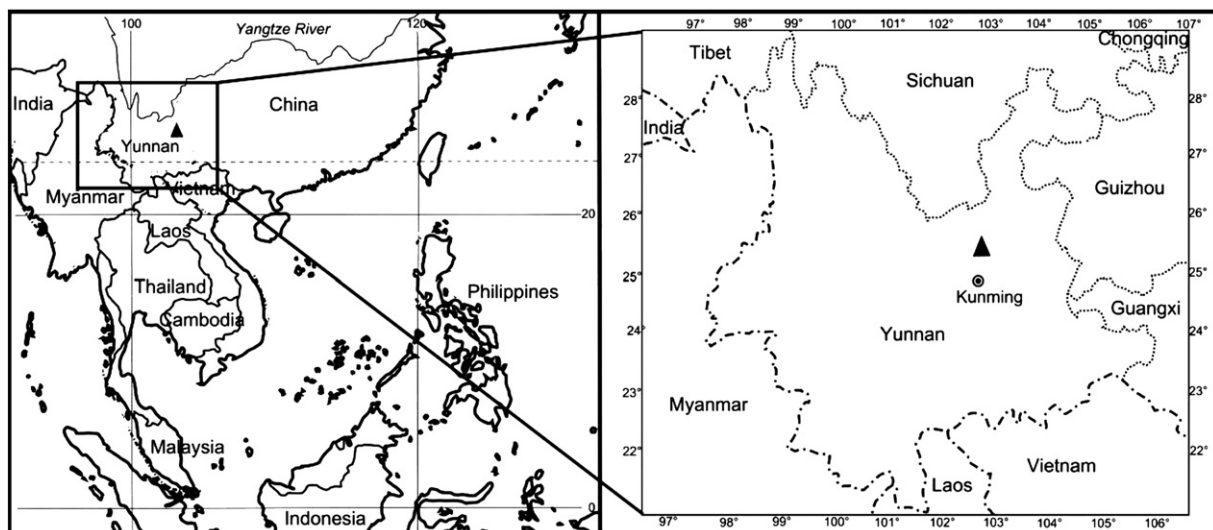

Fig. 1. Map showing the fossil locality (black triangle) of *Quercus praedelavayi* sp. nov.

Strata		Thickness /m	Steatigraphic column	Lithological characters
Quaternary	Q	6		Eluvium and alluvia.
Miocene — Xiaolongtan Formation	N_1^4x	100-200		Mudstone, fine sandstone and siltstone with 3 interlayered coal seams.
	$N_1^3x^2$	100-400		Muddy siltstone with diatom, yielding abundant fossils of plants and insect fragments.
	$N_1^3x^1$	50-280		Diatomite, yielding abundant fossils of plant, fish, snail and ostracod.
	N_1^2x	30-270		Coal-bearing member, comprising thick bedded lignite layers, carbonaceous mudstone and silty mudstone.
	N_1^1x	20-200		Sandy mudstone, muddy siltstone, with thin lignite layers.
Cambrian, Sinian		> 1000		Clasolite

Fig. 2. The sketch strata table of the Xianfeng coalmine. The member where the fossils were collected is marked as pentagram. After Xing et al., 2010.

was easily separated. The same procedures for light microscopy were then followed as those for fossil leaves. The epidermis was pictured by Leica DM1000. The extant specimens were collected from thirteen different localities in Yunnan by Deng M. and Hu J.J. in 2010. One leaf was chosen for each locality. Five different microscopic fields were photographed for each slide. The stomata and trichome bases were counted based on these pictures.

For the examination of the leaf morphology of extant subgen. *Cyclobalanopsis* leaves, we followed the method of Hickey and Wolfe (1975). Leaves were macerated in 5–20% sodium hydroxide (NaOH) firstly, then, rinsed in distilled water. After being bleached in 25% NaClO, leaves were immersed in 250% chloral hydrate ($C_2H_3Cl_3O_2$) for 24 h. The cleared leaves were then rinsed in distilled water, and dehydrated through an ethanol series. After being stained with 1% Safranine O, the cleared leaves were mounted on slides with neutral balsam.

All vouchers, clear leaves of extant species and cuticular slides are deposited at the Xishuangbanna Tropical Botanical Garden, Chinese Academy of Sciences (CAS). For descriptions of leaf morphology and cuticles, we followed the terms of Ellis et al. (2009).

3. Results

3.1. Systematics

Family: Fagaceae Dumortier
Genus: *Quercus* L.
Subgenus: *Cyclobalanopsis* (Oersted) Schneider

Species: *Quercus praedelavayi* Y.W. Xing et Z.K. Zhou sp. nov. (Plate I, 1–12; Plate II, 1–3; Plate III, 1–5).

Holotype: HLT 450 A, B (counterparts) (Plate I, 1, 2; Plate II, 1); slide No. HLT 450A-adaxial (Plate III, 1), HLT 450A-abaxial (Plate III, 2–5).

Paratypes: HLT003 A, B (Plate I, 3, 4; Plate II, 2).

Repository: Xishuangbanna Tropical Botanical Garden, Chinese Academy of Sciences (CAS).

Type locality: Xianfeng coalmine (25°25′ N, 102°51′ E), about 60 km north of Kunming, Yunnan Province, China.

Stratigraphic horizon: The upper bed ($N1^3x^2$) of the third member of the Xiaolongtan Formation.

Age: Late Miocene.

Etymology: The specific epithet refers to a close similarity with the extant species *Quercus delavayi*.

Other specimens: HLT022, HLT150, HLT161, HLT220, HLT302, HLT388, HLT429, HLT440, HLT458, HLT477, HLT483, HLT664, HLT849, HLT851, HLT1024, HLT1213.

Specific diagnosis: Leaves ovate or obovate, coriaceous, symmetrical, apex shape acuminate without drip tip, base shape convex. Leaf serrate, tooth spacing regular, tooth type CC-ST. Primary venation pinnate, midvein straight, robust. Secondaries in the toothed part craspedodromous, secondaries in the entire part camptodromous. Secondary angles to midvein 35°–60°, smoothly increasing proximally, interior secondaries absent. Intercostal tertiaries opposite or alternate percurrent, epimedial tertiaries alternate percurrent. Areolation complete, quadrangular or pentagonal. Free ending veinlets simple without branching. Stomata on abaxial epidermis, anomocytic. Trichome bases unicellular and multicellular.

3.2. Description

Gross morphology (Plates I, II, 1–3) — Fossil blades are marginal attached, unlobed, symmetrical and ovate or obovate in shape (Plates I, II, 1–3). The laminar size is notophyll, with 7–10 cm long, 2.5–3.5 cm wide. According to entire leaves, laminar length to width ratio (L:W) is about 3–3.5. Blade apex is acuminate without drip tip and apex angle is acute, with about 35°–50°. Blade base shape is convex to broadly cuneate and base angle is acute or occasionally obtuse, with about 70°–80°. The leaf margin is serrate or half toothed (the upper 2/3 part is serrate, and the lower 1/3 part is entire). Tooth spacing is regular and the tooth type is CC-ST, which means that the distal flank of the teeth is concave and the proximal flank is straight (Plates I, II, 1–3). The primary venation is pinnate with no naked basal veins and no agrophic veins. The midvein is straight and relatively robust. There are 13–15 pairs of secondary veins with regular spacing (Plates I, II, 1–3). The major secondaries in toothed part are craspedodromous (Plate II, 1–3). The secondaries in entire part are camptodromous (Plate II, 1–3). The secondary angles to midvein are smoothly increasing proximally with about 35°–60° from the apex to the base. The interior secondaries are absent. Tertiary veins have both opposite (majority of tertiaries cross between adjacent secondaries in parallel paths without branching) and alternate (majority of tertiaries cross between secondaries with regular offsets near the middle of the intercostal area) percurrent types, the outmost tertiaries looped, running along the margin (Plate II, 1–3); Areolation shows good development with quadrangular or pentagonal in shape. Veinlets are simple without branching.

Micromorphology (Plate III, 1–5) — The adaxial epidermis is composed of polygonal cells (Plate III, 1), 15–20 μm long, 10–18 μm wide. The epidermal cells have smooth and straight anticlinal walls. No trichome bases are found on the adaxial epidermis (Plate III, 2, 3, 5). The shape of cells on the abaxial epidermis is polygonal (Plate III, 2–4), 15–20 μm long and 10–15 μm wide. Anticlinal walls are straight or weakly undulate. The areoles are well developed which are composed of regular quadrangle cells and generally quadrangular or pentagonal (Plate III, 2). There are two different trichome base types on the lower epidermis. The multicellular trichome bases are round and compose of 5–7 cells with a diameter of 20–30 μm (Plate III, 3, 5), and the density is 140–152 (average 146) n/mm². The multicellular trichome bases usually correspond to stellate trichomes (Plate III, 8). The other one is composed of a single cell with a diameter of 10 μm (Plate III, 3), and the density of the unicellular bases is 127–146 (average 136) n/mm². The trichome bases are usually distributing among the areoles cells, sometimes in the areoles. The leaves are hypostomatic. The stomatal apparatuses are elliptical or nearly rounded and randomly oriented. The stomata are anomocytic, 11–23 μm long, 11–17 μm wide and the length

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Plate I. Selected fossils of *Quercus praedelavayi* sp. nov. Scale bars = 1 cm.

1–2, Holotype, HLT 450A, B (counterparts).
3–4, Paratype, HLT 003A, B (counterparts).
5, Specimen No., HLT388A.
6, Specimen No., HLT220A.
7, Specimen No., HLT851.
8, Specimen No. HLT429.
9–10, Specimen No. HLT366A, B (counterparts).
11, Specimen No. HLT161.
12, Specimen No. HLT664.

Plate II. Comparison of the leaf morphology of *Quercus praedelavayi* sp. nov. with its close related extant species. Scale bars = 1 cm. Black arrows show the outmost tertiaries running along the leaf margin. (see on page 104)

1–3, leaf morphology of *Quercus praedelavayi*.

4–6, leaf morphology of extant *Quercus*. 4, *Quercus delavayi*, 5, *Quercus schottkyana*, 6, *Quercus glauca*.

Plate III. Cuticular structures of *Quercus praedelavayi* sp. nov. and *Quercus delavayi* under light-microscope. (see on page 105)

1, Adaxial epidermis of *Quercus praedelavayi*. Holotype. Slide No. HLT450-adaxial. Scale bar = 100 μm.
2, Abaxial epidermis of *Quercus praedelavayi*. Holotype. Slide No. HLT450-abaxial. Scale bar = 100 μm.
3, Abaxial epidermis of *Quercus praedelavayi*. Holotype. Slide No. HLT450-abaxial. Black arrow shows the multicellular trichome base. White arrow shows the unicellular trichome base. Scale bar = 50 μm.
4, Anomocytic stomata of *Quercus praedelavayi*. Scale bar = 20 μm.
5, Multicellular trichome base of *Quercus praedelavayi*. Scale bar = 20 μm.
6, Adaxial epidermis of extant *Q. delavayi*. Slide No. DH020-3A-adaxial. The lower right graph shows the multicellular trichome base on the upper epidermis. Scale bar = 100 μm.
7, Abaxial epidermis of extant *Q. delavayi*. Slide No. DH020-3A-abaxial. Scale bar = 50 μm.
8, Abaxial epidermis of *Q. delavayi*. Slide No. DH020-3A-abaxial. Black arrow shows the stellate trichome. Scale bar = 50 μm.
9, Anomocytic stomata of *Q. delavayi*. Scale bar = 20 μm.
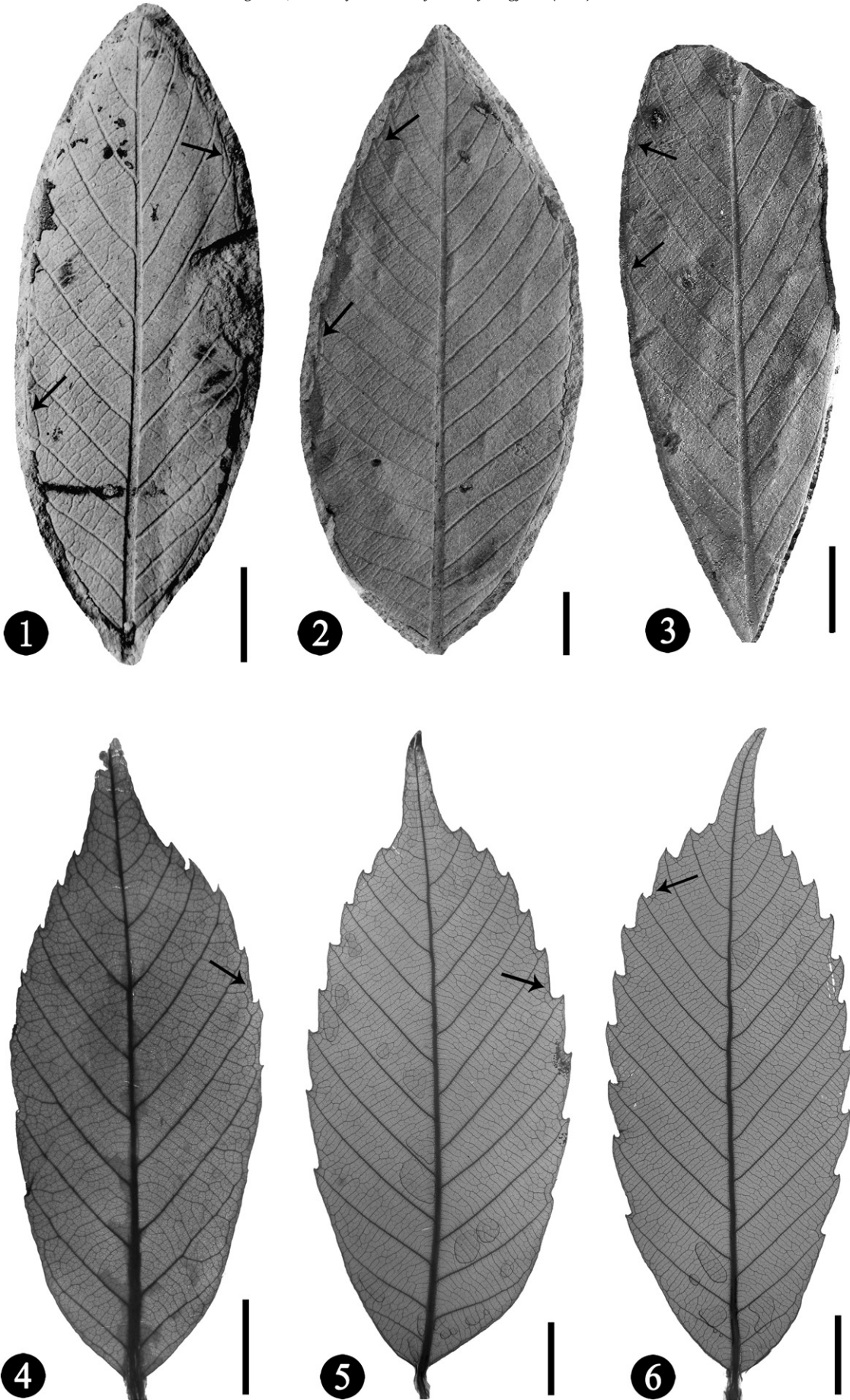10, Multicellular trichome base of *Q. delavayi*. Scale bar = 20 μm.
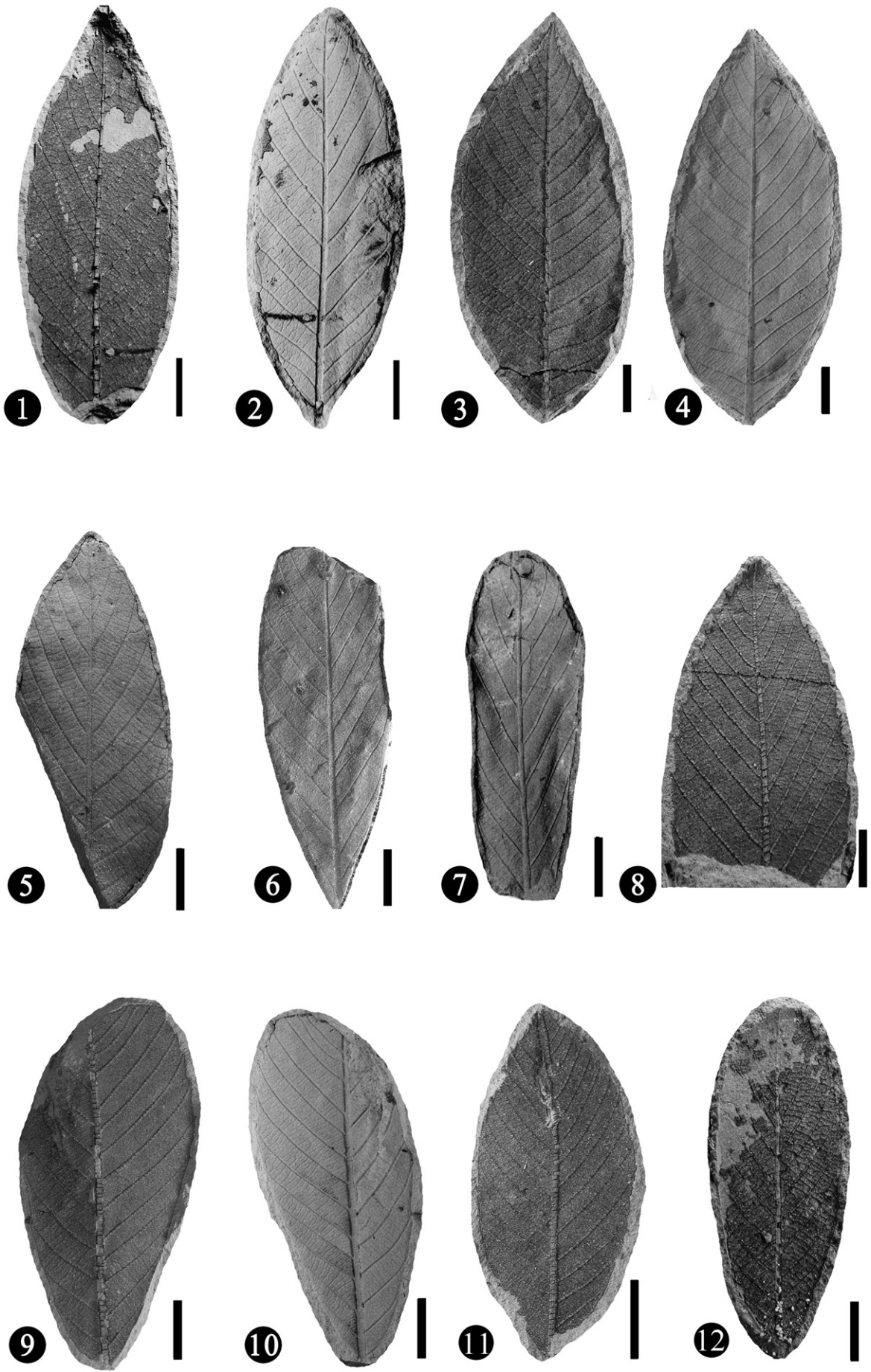
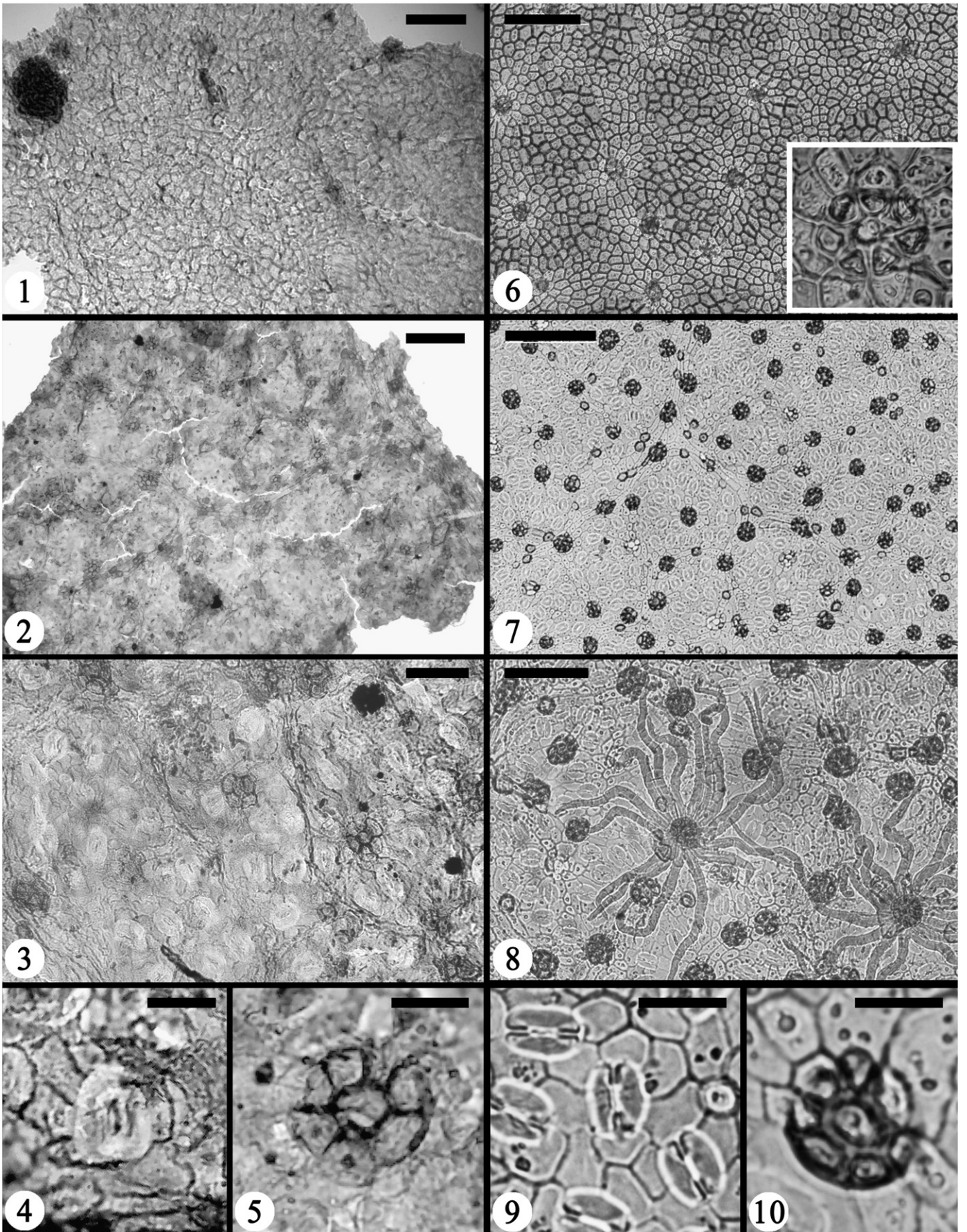**Plate II** *(caption on page 102).*

Plate III *(caption on page 102).*

to width ratio of stomata is 1 to 1.35 (Plate III, 3, 4). All of the stomata are randomly distributed in the areoles. The stomata density is 1123–1155 n/mm². As the epidermal cells are not clear enough, the sto-matal index is not calculated.

4. Discussion

4.1. Systematic position of the fossil species

Jones' (1986) study on the leaf morphology of fagaceous leaves pro-vided a framework to identify fagaceous fossils. According to his results, the synthetic morphological characters, such as symmetric leaf base, possessing unicellular trichome bases, serrate but not doubly serrate make our fossils a clear placement into Fagaceae. Currently, the family Fagaceae includes nine genera: *Fagus* L., *Castanea* Miller, *Castanopsis* (D. Don) Spach, *Chrysolepis* Hjelmquist, *Colombobalanus* Nixon & Crepet, *Formanodendron* Nixon & Crepet, *Lithocarpus* Blume, *Quercus* L., and *Trigonobalanus* Forman (Manos et al., 2001). Our fossils are distin-guished from *Trigonobalanus* s. l. (*Colombobalanus*, *Formanodendron*, *Trigonobalanus*), *Chrysolepis* and *Fagus* due to several diagnostic features, particularly leaf margin and trichome bases (Luo and Zhou, 2002). The fossils differ from *Castanopsis* by having anomocytic stomata (Liu et al., 2009). The characteristics of our fossils best match the genera *Castanea*, *Quercus* and *Lithocarpus*. *Castanea* mainly have CC-CC type tooth where-as our fossils have CC-ST type tooth. In general, *Lithocarpus* leaves show relatively little variability, being usually entire or only partly toothed (Jones, 1986). In China, only *Lithocarpus pachylepis* Camus and *Lithocarpus carolineae* (Skan) Rehder possess serrate leaves (Huang et al., 1999). The leaf blade of *L. pachylepis* is obovate-oblong to oblong, 20–35 cm long, 6–11 cm wide, which has a different shape and much larger size than our fossils. Furthermore, *L. pachylepis* possesses much more secondaries (25–30 pairs) than our fossils. *L. carolineae* also has different leaf shape and secondary venations with our fossils. Consequently, we assign our fossils to the genus *Quercus*, as the charac-teristics of our fossils are common in the *Quercus*. *Quercus* has large var-iation in leaf forms (Jones, 1986). Luo and Zhou (2002) stated that subgenus *Cyclobalanopsis* differs from subgenus *Quercus* by having semi-craspedodromous secondaries. However, the branched second-aries in their work are actually the outmost tertiaries running along leaf margin which are present in our fossils. Though they misused the terminology, the venation pattern is still diagnostic in subgenus *Cyclobalanopsis*. Therefore, our fossils are assigned to the subgenus *Cyclobalanopsis*. To better determine the fossil species, we compared our fossils with both extant and fossil species of subgen. *Cyclobalanopsis*.

4.2. Comparison of Quercus praedelavayi with extant Quercus subgenus Cyclobalanopsis species

In our study, we focused on the comparisons with the species possessing the same venation patterns. Selection of species and char-acteristics for comparison was based on previous work and our per-sonal observations (Huang et al., 1999; Luo and Zhou, 2002; Deng, 2007; Table 1). *Quercus austrocochinchinensis* Hickel & A. Camus, *Q. kiukiangensis* (Y. T. Chang) Y. T. Chang, *Quercus litoralis* Chun & P. C. Tam ex Y. C. Hsu & H. W. Jen, *Quercus oxyodon* MiQ, *Quercus sichouensis* (Hu) C. C. Huang & Y. T. Chang. Q. A. Wu and *Quercus thorelii* Hickel & A. Camus could be easily distinguished from our fossils by having much larger leaves. *Quercus augustinii* Skan, *Q. austrocochinchinensis*, *Q. thorelii*, *Q. litoralis*, *Q. sichouensis*, *Quercus subhinoidea* Chun & W. C. Ko, *Q. oxyodon* and *Quercus tiaoloshanica* Chun & W. C. Ko differ from the fossil species by having different sto-mata types (Deng, 2007). Previous study indicated that *Quercus delavayi*, *Quercus glauca* and *Quercus schottkyana* have cyclocytic sto-mata (Deng, 2007). However, our own observations indicated that they all possess anomocytic stomata. The fossil species cannot be dis-tinguished from *Q. delavayi*, *Q. glauca* and *Q. schottkyana* by gross morphology and stomata types (Plate II). However, the fossil species has both unicellular and multicellular trichome bases (Plate III, 2–4), while *Q. glauca* and *Q. schottkyana* only have unicellular trichome bases on abaxial epidermis (Table 1). Thus, the fossil species is most similar to the extant *Q. delavayi* by possessing similar blade shapes, sizes, venations and trichome bases. However, the fossil species has no trichomes on adaxial epidermis and the density of stomata is higher than that of *Q. delavayi* (Plate III; Table 2). It is better to consider these differences rather than simply assign the fossils to the extant species.

4.3. Comparison of Quercus praedelavayi with the subgenus Cyclobalanopsis fossil species

The identifications of the fossils assigned to subgen. *Cyclobalanopsis* were mainly based on such gross characteristics as blades shape, size and venation types rather than epidermal characters in previous work. Among these fossil species, *Quercus preglauca* Guo and *Quercus paraschottkyana* Wang and Liu possess similar leaf sizes, shapes, vena-tions and tooth type to extant *Quercus glauca* and *Quercus schottkyana* which are also very similar to our fossils (Table 1). As mentioned above, *Quercus delavayi*, *Q. glauca* and *Q. schottkyana*, are very diffi-cult to be distinguished by gross morphology. Therefore, we cannot

Table 1
Comparisons of *Quercus praedelavayi* sp. nov. with the extant and fossil species of subgenus *Cyclobalanopsis*.
Data sources: Huang et al., 1999; Luo and Zhou, 2002; Deng, 2007; fossil[1]: Jia et al., 2009; fossil[2]: Guo, 2011; fossil[3]: WGCPC, 1978.

Taxa	Size (long × wide)	L:W ratio	Shape	Number of secondaries	Stomata type	Trichome bases on abaxial epidermis	Trichome bases on adaxial epidermis
Q. praedelavayi (sp. nov.)	7–10 × 2.5–3.5	3	Elliptic to ovate-elliptic	13–15	Anomocytic	Unicellular, multicellular	Multicellular
Q. aff. delavayi (fossil[1])	6.2 × 1.6	4	Elliptic-lanceolate	10	Anomocytic	Unicellular, multicellular	Multicellular
Q. preglauca (fossil[2])	11 × 3.8	3	Elliptic	10–13	–	–	–
Q. paraschottkyana (fossil[3])	5.5–8 × 2.5–2.9	2.5	Elliptic	9–11	–	–	–
Q. augustinii	6–12 × 1–4	3–6	Ovate-lanceolate to elliptic-lanceolate	10–15	Cyclocytic	Unicellular	Unicellular
Q. austrocochinchinensis	10–17(−20) × 3–5	3.3–4	Oblong-elliptic to lanceolate	12–17	Cyclocytic	Multicellular	Multicellular
Q. delavayi	8–12 × 2–4.5	2.8–4	oblong to ovate-elliptic	10–14	Anomocytic	Unicellular, multicellular	Multicellular
Q. glauca	6–13 × 2–5.5	2.4–3	Obovate to oblong-elliptic	9–13	Anomocytic	Unicellular	Unicellular
Q. schottkyana	6–13 × 2–5	2.5–3	Long elliptic to obovate-oblanceolate	8–12	Anomocytic	Unicellular	Unicellular
Q. kiukiangensis	10–18 × 3.5–8.5	2.2–3	Oblong-elliptic	10–13	Anomocytic	Unicellular	Unicellular
Q. litoralis	10–20 × 5–10	2	Elliptic	7–9	Cyclocytic	Unicellular	Unicellular
Q. multinervis	7.5–15.5 × 2.5–5.5	2.5–3	Oblong-elliptic to elliptic-lanceolate	10–15	Cyclocytic	Unicellular	Unicellular
Q. oxyodon	12–24 × 3–8	3–4	Elliptic to oblong-lanceolate	16–24	Cyclocytic	Multicellular	Unicellular
Q. sichouensis	12–21 × 5–9	2.5	Oblong to ovate-elliptic	15–18	Cyclocytic	Multicellular	Multicellular
Q. subhinoidea	7–12 × 2.5–4	2.5–3	Oblong to lanceolate-elliptic	(15–)17–22	Cyclocytic	Multicellular	Multicellular
Q. thorelii	12–17 × 3.5–7	2.5–3.5	Ovate to oblong-elliptic	13–16	Cyclocytic	Multicellular	Multicellular
Q. tiaoloshanica	4–10 × 1.2–3	3.3	Oblong to obovate-elliptic	5–7	Cyclocytic	Unicellular	Multicellular

Table 2
Comparisons of the epidermis characters of *Quercus praedelavayi* sp. nov. with the extant *Q. delavayi*.

	Q. praedelavayi sp. nov.	*Q. delavayi*
Density of unicellular trichome bases on abaxial epidermis (n/mm^2)	127–146 (average 136)	12–134 (average 79)
Density of multicellular trichome bases on abaxial epidermis (n/mm^2)	140–152 (average 146)	73–219 (average 115)
Density of multicellular trichome bases on adaxial epidermis (n/mm^2)	–	24–43 (average 34)
Stomata density (n/mm^2)	1123–1155 (average 1139)	784–1080 (average 931)
Stomata size (length × width, μm)	11–23 × 11–17	15–30 × 10–21

simply assign our fossils to *Q. preglauca* and *Q. paraschottkyana*. To date, one fossil species has been reported based on epidermal characteristics in China. The fossil species which was also thought to have the closest affinity with extant *Q. delavayi* is from the upper Miocene of Zhejiang Province, eastern China (Jia et al., 2009). Based on the cuticular characteristics, the fossil from the upper Miocene of Zhejiang named *Q. aff. delavayi* is similar to our fossils and the extant *Q. delavayi* (Table 1). However, the tooth type and the third venation type of *Q. aff. delavayi* is not clear enough to be compared with our fossils and the extant *Q. delavayi*. Moreover, *Q. aff. delavayi* has elliptic-lanceolate leaves which are much narrower than the extant *Q. delavayi*.

Comparisons to both extant and fossil subgenus *Cyclobalanopsis* species indicate that our fossils have the closest affinity with extant *Quercus delavayi* by possessing similar blade shapes, sizes, tooth types, venations and trichome bases, but cannot be assigned to any extant or fossil species. Therefore, we describe them as a new species, *Quercus praedelavayi* Y.W. Xing et Z.K. Zhou.

4.4. Ecological and evolutionary significance of Quercus praedelavayi

The Neogene subgen. *Cyclobalanopsis* species are comparable with the extant species which makes them indicators of palaeoclimate and palaeovegetation. At present, *Quercus delavayi* is distributed in the evergreen forests with the elevation from 1000 to 2800 m in central to southwestern China (Huang et al., 1999). *Quercus preadelavayi* is the dominant species in the Xianfeng flora which indicates a warm and humid subtropical evergreen forest. This also matches the palaeoclimatic reconstructions in southwestern China (Xia et al., 2009; Jacques et al., 2011; Sun et al., 2011; Xing et al., 2012).

It has been well demonstrated that CO_2 concentration is an important factor that drives the plant evolution (Woodward, 1987; Franks and Beerling, 2009). As an important gateway for gas exchange and transpiration of plants, stomata density and index are sensitive to the CO_2 concentration change. Plants increase or reduce the stomata to adapt lower or higher CO_2 concentrations (Woodward, 1987; Van der Burgh et al., 1993; Woodward and Kelly, 1995; Kürschner et al., 1996; Beerling et al., 1998; Sun et al., 2003; McElwain, 2004; Kürschner et al., 2008). The stomata density of *Q. praedevayi* is 1123–1155 (average 1139) n/mm^2 which is higher than the present value (784–1080 (average 931) n/mm^2). This suggests that the palaeoatmospheric CO_2 concentration during the late Miocene might be lower than the present which also matches with previous findings (Beerling and Royer, 2011; Fig. 3). However, as the response of stomatal density and index to the CO_2 concentration is highly species-specific (Kürschner et al., 1997; Royer, 2001; Kouwenberg et al., 2007), further work are needed to reconstruct the palaeoatmospheric CO_2 concentration of Xianfeng flora.

Epidermal trichomes, sunken stomata and stomatal furrows are the "xeromorphic" cuticular features (Haworth and McElwain, 2008). These features are commonly considered as adaptations to aridity, increasing boundary layer resistance and consequently limiting transpiration (Wuenscher, 1970; Schuepp, 1993; Rocas et al., 1997; Aronne and De Micco, 2001). We hypothesize that the *Quercus* species in dry environment has a higher trichome density than in humid region. To test the hypothesis, we counted the density of trichome bases of the *Quercus praedelavayi* and *Quercus delavayi*. The trichome base density of *Q. praedelavayi* is higher than the extant values. According to the palaeoclimatic reconstructions of Xianfeng flora (Xing et al., 2012), the precipitation of Xianfeng flora in the late Miocene (1206.0–1613.0 mm) was higher than the present value (1038 mm). The results
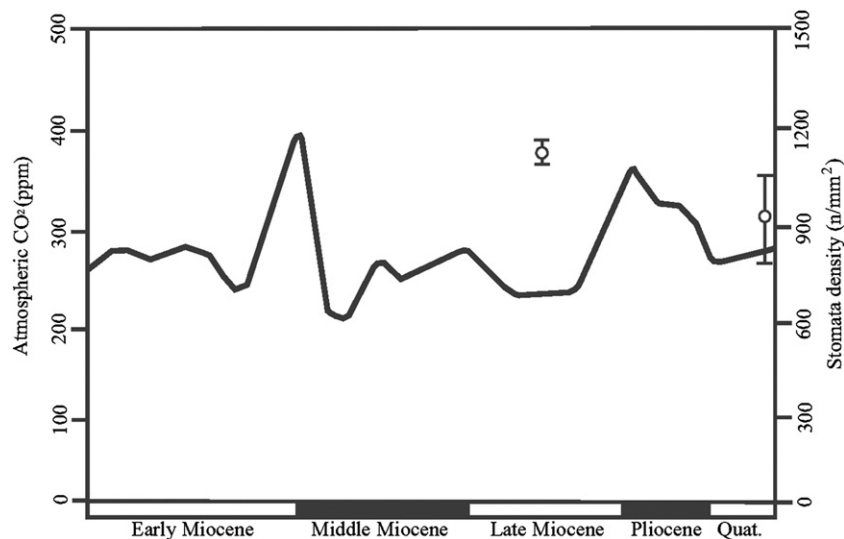
Fig. 3. The Neogene atmospheric CO2 concentration and the stomata density of *Quercus praedelavayi* and *Quercus delavayi* (circles with error bars). The curve is after Beerling and Royer, 2011.

indicate that the evolution of trichome density of *Q. delavayi* is not driven by the decrease in precipitation.

Acknowledgments

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