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Pollen morphology and ultrastructure of *Quercus* with focus on Group Ilex (=*Quercus* subgenus *Heterobalanus* (Oerst.) Menitsky): implications for oak systematics and evolution

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Abstract
It has been suggested that pollen ornamentation can be used to distinguish infrageneric groups in *Quercus*. Here, we document pollen morphology of nearly all species of *Quercus* Group Ilex and show that they share distinctive microrugulate pollen ornamentation. Furthermore, pollen ultrastructure of all six infrageneric groups of *Quercus* was studied comparatively and evaluated using a phylogenetic framework for Fagaceae. Characteristics of the foot layer, such as its thickness and continuity, and the ratio foot layer to tectum are not stable within infrageneric groups of *Quercus*. The foot layer in derived groups (Groups Cerris, Quercus, Lobatae) is irregular, discontinuous and thin (type 2 foot layer). Members of Groups Cyclobalanopsis and Ilex either have a regular, continuous, and comparatively thick foot layer (type 1), or a type 2 foot layer. Only Group Protobalanus consistently has a type 1 foot layer and is similar to *Trigonobalanus* and *Colombobalanus* among trigonobalanoids, and to castanoids. Because (I) all except for one species of *Castanea* and *Nothofagus* have a type 1 foot layer and (II) basal groups in *Quercus* may have a type 1 foot layer, we suggest that this is the ancestral state in Fagaceae, and that the type 2 foot layer of *Fagus* is derived.

Keywords: Quercus *Group Ilex, evolution, pollen ornamentation, foot layer*

A major attempt of phylogenetic and evolutionary studies has been to explore morphological characters or combinations of characters that are suitable to define natural groups of plants (Walker & Doyle 1975; Doyle & Endress 2000, 2010; Doyle 2005). Existing phylogenies for a large number of plant groups based on molecular data can provide a framework for assessing the evolution and phylogenetic significance of morphological characters (see e.g. Doyle & Endress 2000; Endress & Doyle 2009). At the same time, conflict in molecular phylogenetic reconstructions can be investigated using morphological characters (Denk et al. 2005; Friis et al. 2007; Denk & Grimm 2009; Mathews 2009). Pollen characters have been playing a key role in identifying natural groups and in understanding plant evolution (e.g. Kuprianova 1962; Doyle & Le Thomas 1997; Denk 2003; Huysmans et al. 2003; Dettmann & Jarzen 2005; Denk & Tekleva 2006; Wortley et al. 2007, 2008; Denk & Grimm 2009; Nagels et al. 2009; Wang et al. 2010).

Quercus is a large genus (*c.* 400–500 species) of monoecious trees and shrubs distributed across the Northern Hemisphere with a small extension into South America. Six infrageneric groups are recognised within *Quercus*: (I) Group Cyclobalanopsis (*c.* 150 spp.; estimated by Flora of China Editorial Committee 1999) occurs in subtropical and tropical regions in southeast Asia, while the Groups Lobatae (II) (*c.* 195 spp.; estimated by Flora of North America Editorial Committee 1997) and Protobalanus (III) (five species) at present are found in the New World only. The Groups Cerris (IV) and Ilex (V) (11 and *c.* 30 spp.; Denk & Grimm 2009, 2010) are confined to

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Eurasia, while the Group Quercus (VI) (*c.* 160 spp.; estimated in Flora of North America Editorial Committee 1997 and Flora of China Editorial Committee 1999) occurs both in the Old and New World. The majority of molecular phylogenetic studies have resolved two major clades within *Quercus*, one consisting of the infrageneric groups Ilex, Cerris and Cyclobalanopsis, and the other consisting of the groups Protobalanus, Lobatae and Quercus (Manos & Stanford 2001; Manos et al. 2001, 2008; Oh & Manos 2008; Denk & Grimm 2010).

In traditional classification schemes of *Quercus* (e.g. Camus 1936–1938, 1938–1939, 1952–1954; Schwarz 1936; Nixon 1993), the systematic placement of Groups Cerris and Ilex (the 'Cerris oaks' and the 'Ilex oaks') has been remarkably unstable (cf. Denk & Grimm 2010 for the taxonomic history of these groups) because of the lack of diagnostic morphological characters.

Recently, it has been shown that pollen ornamentation is highly diagnostic for the six infrageneric groups of *Quercus* (Denk & Grimm 2009). Furthermore, it has earlier been suggested that Ilex oaks are distinctive among oaks by having a relatively thicker and more homogeneous foot layer than other groups of oaks (Crepet & Daghlian 1980). The latter study was based on very limited material (two specimens of *Quercus* Group Ilex).

At present, there is no comprehensive study documenting pollen morphology in the European and western and eastern Asian members of *Quercus* Group Ilex. A number of detailed accounts on pollen morphology and ultrastructure of genera of Fagales are available (*Alfaroa*, *Carya*, *Castanea*, *Castanopsis*, *Chrysolepis*, *Engelhardia*, *Fagus*, *Juglans*, *Lithocarpus*, *Nothofagus*, *Oreomunnea*, *Platycarya*, *Pterocarya*; Stone & Broome 1975; Praglowski 1982; 1984) but not of *Quercus*. The work of Wang and Pu (2004) on pollen morphology of Fagaceae covers many of the Chinese native genera and species of Fagaceae, but, unfortunately, the scanning electron microscopy (SEM) images are of insufficient quality to evaluate the systematic and phylogenetic value of pollen ornamentation.

The present paper aims at filling this gap. Most of the 31 species of the Eurasian *Quercus* Group Ilex were investigated using light microscopy (LM, 27 spp.) and electron microscopy (SEM, 27 spp., and transmission electron microscopy (TEM), 12 spp.). A further 23 species of the other five infrageneric groups of *Quercus* were sectioned for TEM investigations and studied with SEM (groups Cerris, 7 spp., Cyclobalanopsis, 2 spp., Protobalanus, 3 spp., Quercus, 9 spp., Lobatae, 2 spp.). Our investigation had two main objectives: first, to document the variability of the tectum ornamentation within and among species of *Quercus* Group Ilex, and second, to evaluate the evolutionary significance of pollen ultrastructure encountered in members of all six infrageneric groups of *Quercus* and its relevance for Fagaceae evolution.

Material and methods

Pollen material investigated for the present study (Table I) originates from herbarium material stored at the herbaria Edinburgh (E), Istanbul (ISTO), Stockholm (S) and Berkeley (UC). LM investigations were made on material from the palynological slide collection at the Swedish Museum of Natural History, Stockholm. A full list of herbarium vouchers for LM, SEM and TEM preparations is provided in the 'Specimens investigated' section. For SEM investigations, pollen was removed from anthers and mounted on SEM stubs. Non-acetolysed pollen was used for SEM observations, because previous studies using acetolysed and non-acetolysed pollen of Fagaceae showed the same details of the pollen surface (cf. Praglowski 1982, 1984; Solomon 1983a, 1983b; Denk 2003; Denk & Grimm 2009).

For TEM investigations, individual anthers or parts of anthers were fixed with 1% osmium tetroxide (OsO_4), dehydrated in an ethanol series, stained with uranyl acetate, dehydrated in acetone, and embedded in epoxy resin. Pollen grains were sectioned with an ultramicrotome LKB-3 and ultrathin sections were then post-stained with lead citrate, and examined using a Jeol 100 B and a Jeol 1011 transmission electron microscopes.

Pollen terminology follows Nilsson and Praglowski (1992), Punt et al. (2007), Denk and Grimm (2009) and Hesse et al. (2009). For the foot layer, the term continuous was used when the foot layer was uninterrupted in the mesocolpium region, discontinuous when the foot layer was disrupted. Regular denotes a foot layer that is of even thickness in the mesocolpium region, irregular a foot layer that is unevenly thick.

Phylogenetic framework

For a phylogenetic framework, the studies by Manos et al. (2001) and Denk and Grimm (2009, 2010) were used. These studies identified six infrageneric groups and showed that the classical subdivision into *Quercus* subgenus *Quercus* and *Quercus* subgenus *Cyclobalanopsis* is unnatural. We follow the concept of Denk and Grimm (2010) who introduced informal names for

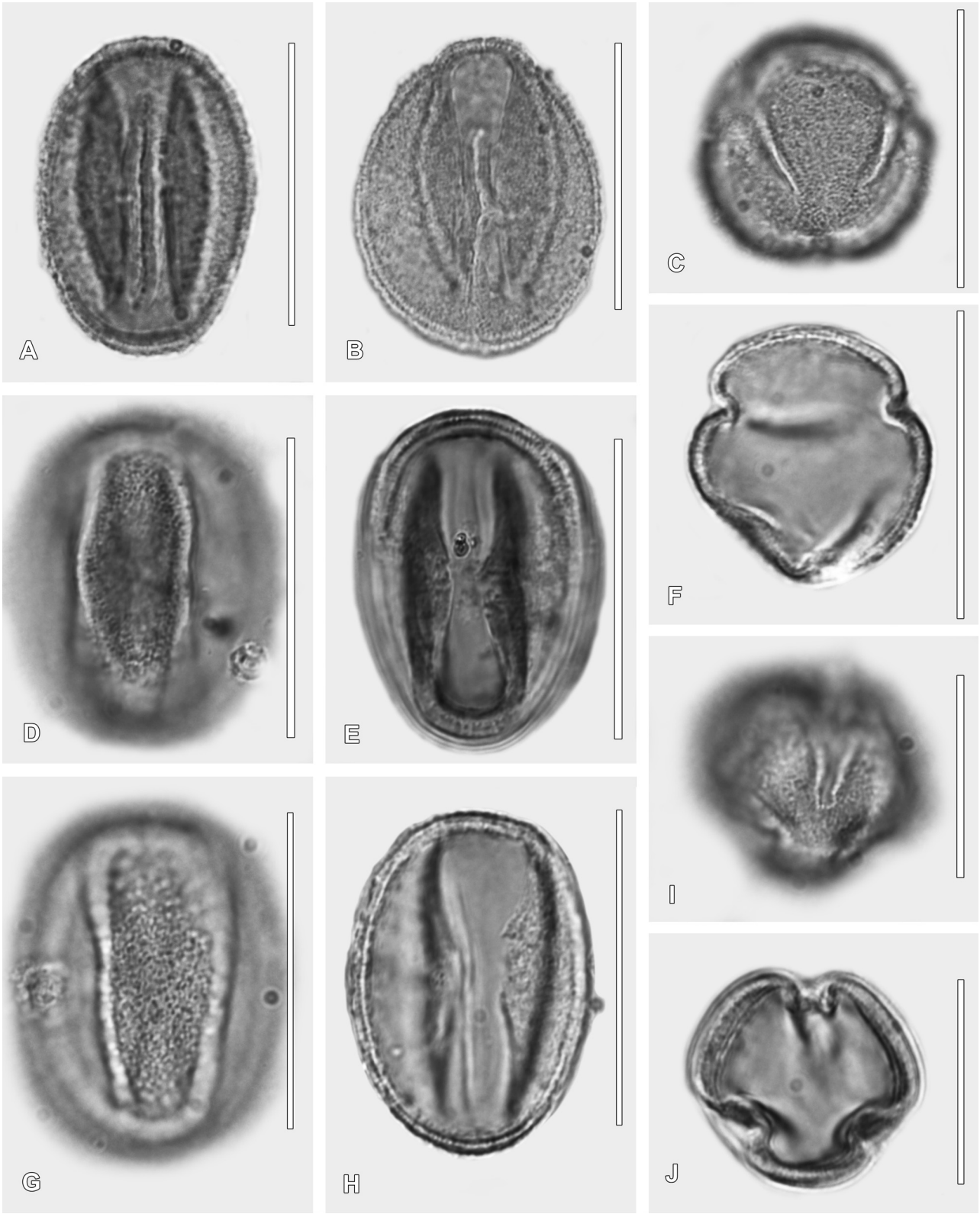

Figure 1. Pollen of *Quercus* Group Ilex, light micrographs. **A, B, D, E, G, H.** Equatorial view. **C, F, I, J.** polar view. **A.** *Quercus coccifera*, Erdtman 1444 (S). **B.** *Quercus ilex*, Firenze 1873 (S). **C.** *Quercus ilex* subsp. *ballota* (S). **D, E.** *Quercus lanata*, Garrett 1175 (S); **D.** High focus; **E.** Middle low focus. **F.** *Quercus phillyraeoides*, M. Takeoka s. n. (S), low focus. **G, H.** *Quercus phillyraeoides*, Murata & Koyama (S); **G.** High focus; **H.** Low focus. **I, J.** *Quercus semecarpifolia*, J. E. T. Aitchison 24 (S); **I.** High focus; **J.** Low focus. Scale bars – 30 μm (A–H), 20 μm (I, J).

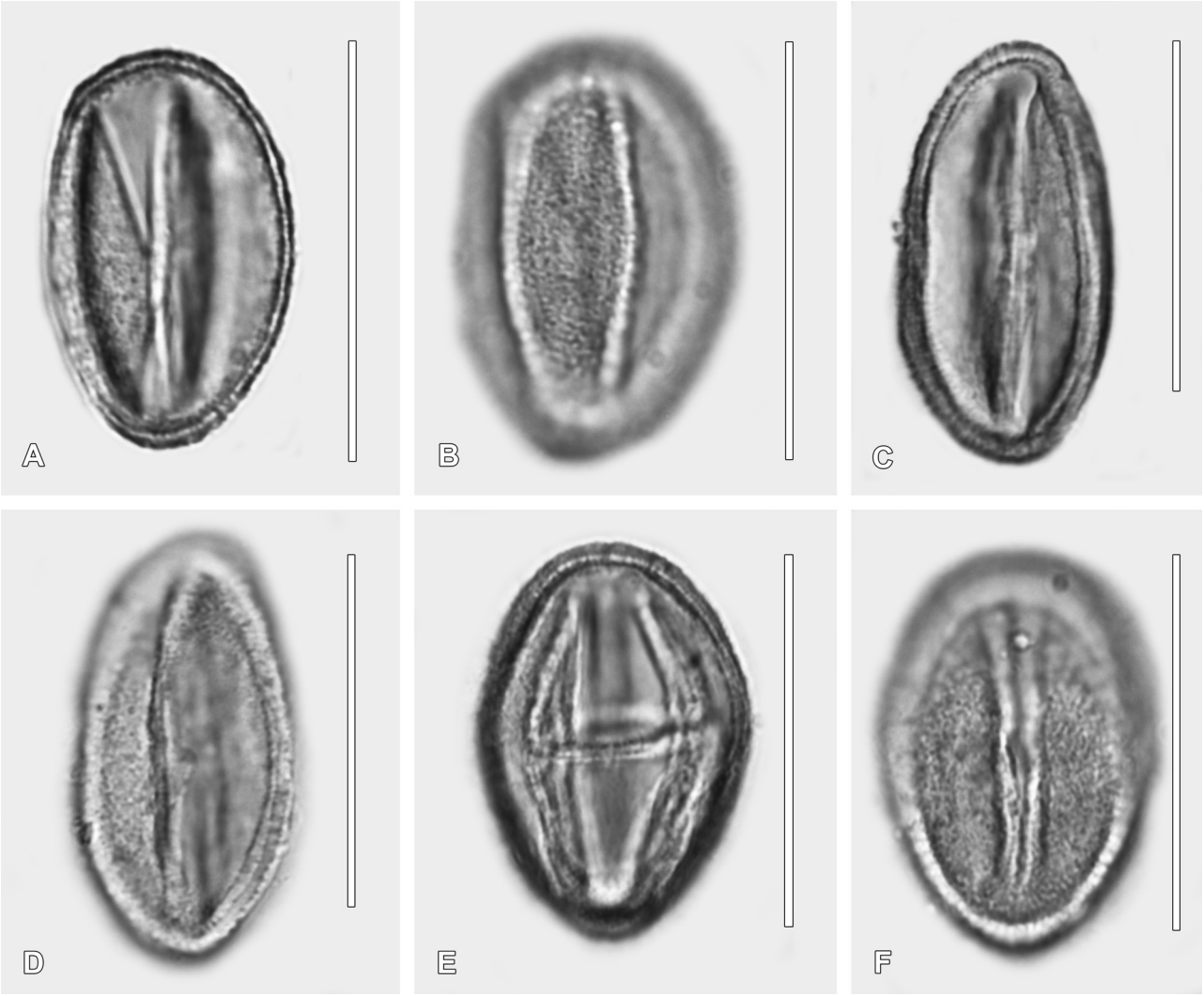

Figure 2. Pollen of *Quercus* Group Ilex, light micrographs, equatorial view. **A, B.** *Quercus phillyraeoides*, Murata & Koyama (S); **A.** Low focus; **B.** High focus. **C–F.** *Quercus semecarpifolia*, J. E. T. Aitchison (S); **C.** Low focus; **D.** High focus; **E.** Low focus; **F.** High focus. Scale bars – 30 μm.

the six (monophyletic) groups within *Quercus*. The nomenclature for these groups is as follows: 1. *Quercus* Group Cerris, 2. *Quercus* Group Cyclobalanopsis, 3. *Quercus* Group Ilex, 4. *Quercus* Group Lobatae, 5. *Quercus* Group Quercus, and 6. *Quercus* Group Protobalanus. For previously used partly synonymous names, see Denk and Grimm (2010).

For practical reasons, we use the subfamilial name Castaneoideae Oersted to denote the genera *Castanea*, *Castanopsis*, *Lithocarpus*, *Chrysolepis* and *Notholithocarpus*. For *Trigonobalanus*, *Formanodendron* and *Colombobalanus*, we use the informal term trigonobalanoids.

Results (comparative pollen morphology)

Light microscopy (Figures 1, 2)

Pollen is dispersed as monads. The shape is prolate, the outline in polar view is trilobate to rounded, in equatorial view elliptic to oval. Pollen is small to medium sized (Table II), tricolpate with long and narrow colpi. Tricolporoidate grains have been reported in the literature (e.g. Colombo et al. 1983; Van Benthem et al. 1984), but unambiguous endoapertures were not observed in the present study. The pollen is tectate, columellate; pollen ornamentation is scabrate.

Table I. Taxa investigated for the present study. For specimen details see 'Specimens investigated'.

Infrageneric group, species	Distribution
***Quercus* Group Ilex**	
Q. acrodonta Seemen [incl. *Q. handeliana* A.Camus]	Central, South China
Q. alnifolia Poech	Cyprus
Q. aquifolioides Rehder et E.H.Wilson	Tibet, Southwest China
Q. aucheri Jaub. et Spach	Kos (Greece), Southwest Turkey
Q. baloot Griff.	Southwest Himalayas
Q. baronii Skan	Central, South China
Q. coccifera L.	South Europe, North Africa
Q. cocciferoides Hand.-Mazz.	Sichuan, Yunnan
Q. dilatata Royle	Afghanistan, Pakistan
Q. dolicholepis A.Camus [incl. *Q. spathulata* Seemen]	China
Q. engleriana Seemen	Assam, Tibet, South China
Q. franchetii Skan	Thailand, China
Q. gilliana Rehder et E.H.Wilson	Tibet, South and Central China
Q. guyavifolia H.Lév.	Sichuan, Yunnan
Q. ilex L.	South Europe, North Africa
Q. kingiana Craib	Thailand, Burma, Southwest China
Q. lanata Sm.	Himalayas to Southeast Asia
Q. leucotrichophora A.Camus [incl. *Q. incana* Roxb.]	North Pakistan to North Burma, North Thailand, Vietnam
Q. lodicosa E.F.Warb.	Tibet, Assam, Burma
Q. longispica (Hand.-Mazz.) A.Camus	Sichuan, Yunnan
Q. monimotricha (Hand.-Mazz.) Hand.-Mazz.	North Burma Southwest China
Q. pannosa Hand.-Mazz.	Sichuan, Yunnan
Q. phillyreoides A.Gray	East Asia
Q. rehderiana Hand.-Mazz.	Tibet, China
Q. semecarpifolia Sm.	Southwest Himalayas to East Asia
Q. senescens Hand.-Mazz.	East Himalayas, Southwest China, Tibet
Q. spinosa David	Taiwan, China, Burma
***Quercus* Group Cerris**	
Q. acutissima Carruth.	China, Korea, Japan
Q. afares Pomel	North Africa
Q. brantii Lindl. subsp. *brantii*	Asia Minor
Q. cerris L.	South Europe, Asia Minor
Q. ithaburensis Decne. subsp. *macrolepis* (Kotschy) Hedge et Yalt.	Italy to Asia Minor
Q. libani Oliv.	Asia Minor
Q. suber L.	South Europe, north-western Africa
***Quercus* Group Cyclobalanopsis**	
Q. acuta Thunb.	East Asia
Q. augustinii Skan	Southeast Asia
Q. championii Benth.	China, Taiwan
Q. fleuryi Hickel et A.Camus	Southeast Asia
Q. glauca Thunb.	India to East Asia
Q. longinux Hayata	Taiwan
***Quercus* Group Protobalanus**	
Q. chrysolepis Liebm.	South-western USA
Q. tomentella Engelm.	California, Baja California
Q. vacciniifolia Kellogg	Western USA
***Quercus* Group Quercus**	
Q. arizonica Sarg.	Southern USA, Mexico
Q. brandegeei Goldmann	Mexico
Q. bumelioides Liebm.	Honduras, Costa Rica, Panama, Guatemala, Nicaragua
Q. dentata Thunb.	Japan, Korea, China
Q. douglasii Hoo. et Arn.	California
Q. fabri Hance	East and Southeast China, Korea
Q. faginea Lam.	Southwestern Europe, North Africa
Q. frainetto Ten.	South Europe, Asia Minor
Q. garryana Douglas ex Hook.	Western USA
Q. griffithii Hook.f. et Thomson ex Miq.	India to Southeast Asia
Q. infectoria subsp. *veneris* (A.Kern.) Meikle	South-eastern Europe, Asia Minor
Q. kelloggii Newb.	Western North America

(*Continued*)

Table I. (*Continued*).

Infrageneric group, species	Distribution
Q. macranthera Fisch. et C.A.Mey.	Asia Minor, Caucasus, Iran
Q. michauxii Nutt.	Southern and Central USA
Q. petraea (Matt.) Liebl.	Europe, Asia Minor
Q. pyrenaica Willd.	Southwestern Europe, northwestern Africa
Q. robur L.	Europe, Asia Minor
Q. tuberculata Liebm.	Mexico
Q. virginiana Mill.	Southeastern USA
***Quercus* Group Lobatae**	
Q. canbyi Trel.	Texas, Mexico
Q. humboldtii Bonpl.	Texas to northern South America
Q. laevis Walter	South-eastern USA
Q. nigra L.	South-eastern USA

Scanning electron microscopy (Figures 3–10)

Pollen ornamentation of *Quercus* Group Ilex shows a consistent pattern in all specimens investigated. The tectum is rugulate to microrugulate, the ornamentation may appear granulate or microverrucate in low magnification overviews (e.g. Figs 3J, 4B, F, 5C, G,). The microrugulae agglomerate and form desert-rose-like structures (Denk & Grimm 2009; conical [tepee]-shaped according to Rowley & Gabarayeva 2004; Figures 7–10). These structures may be clearly defined (Figures 7F, K, 8J, 9I) or their borders not distinctly defined (Figure 8N). Moreover, the portions of individual rugulae which fuse may be of markedly different length. Within the same species, rugulae can be > 1 μm long in one collection and not more than *c.* 0.5 μm in another (e.g. Figure 7L, N, *Q. baloot* Griff.). Other species consistently appear to have short and thin rugulae (Figure 8K, L, *Q. cocciferoides* Hand.-Mazz.; Figure 9B–D, L, *Q. franchetii* Skan, *Q. kingiana* Craib; Figure 10L, M, *Q. senescens* Hand.-Mazz.). In some cases, the rugulae are more or less horizontal and fuse to form a rugulate pattern with weak relief (Figures 7L, 10N, *Q. baloot*, *Q. spathulata* Seemen). The tectum commonly is perforate (Figures 3B, 4A, 6A, 7–10). The tectum ornamentation is the same in the mesocolpium, polar and apertural regions. The colpus membrane is covered with granular elements in *Q. senescens* (Figure 6F, G), while it is smooth in other samples (Figure 3I, *Q. baloot*; Figure 6E, J, *Q. semecarpifolia* Sm., *Q. coccifera* L.).

Transmission electron microscopy Quercus *Group Ilex* (Figures *11*, *12*)

All species investigated with TEM showed essentially the same type of exine ultrastructure (endexine, foot layer, columellae, and tectum). However, while some elements show little variation (columellae), others are variable and potentially important for understanding phylogenetic relationships within Fagaceae (foot layer).

The pollen wall comprises an endexine in the mesocolpium and apertural region. The endexine of the non-apertural region may be homogeneous and of even thickness (e.g. Figure 11B, F, M, O), or coarsely laminated (Figure 11A, I) to fragmented (Figure 11, L). Towards the apertural region, the endexine becomes considerably thicker and coarsely laminated (Figure 11C, G, H) and forms the colpus membrane. The thickness of the endexine ranges from < 0.1 to 0.3 μm in the mesocolpium and from 0.4 to 0.6 μm in the apertural region (Table III). In the apertural region, the endexine may be compacted and then displaying white lines (Figure 12).

The foot layer has the same electron density, and hence appears in the same colour in the TEM micrographs as the columellae and the tectum. It may be discontinuous or continuous and of even or uneven thickness. The foot layer commonly becomes discontinuous and patchy towards the apertural region (Figure 11G, H). In the non-apertural region, the foot layer is continuous and regular in some of the investigated taxa (e.g. *Quercus aucheri* Pomel, Figure 11A, B; 'type 1' foot layer) and markedly disrupted and irregular in others (e.g. *Q. lanata* Sm.,

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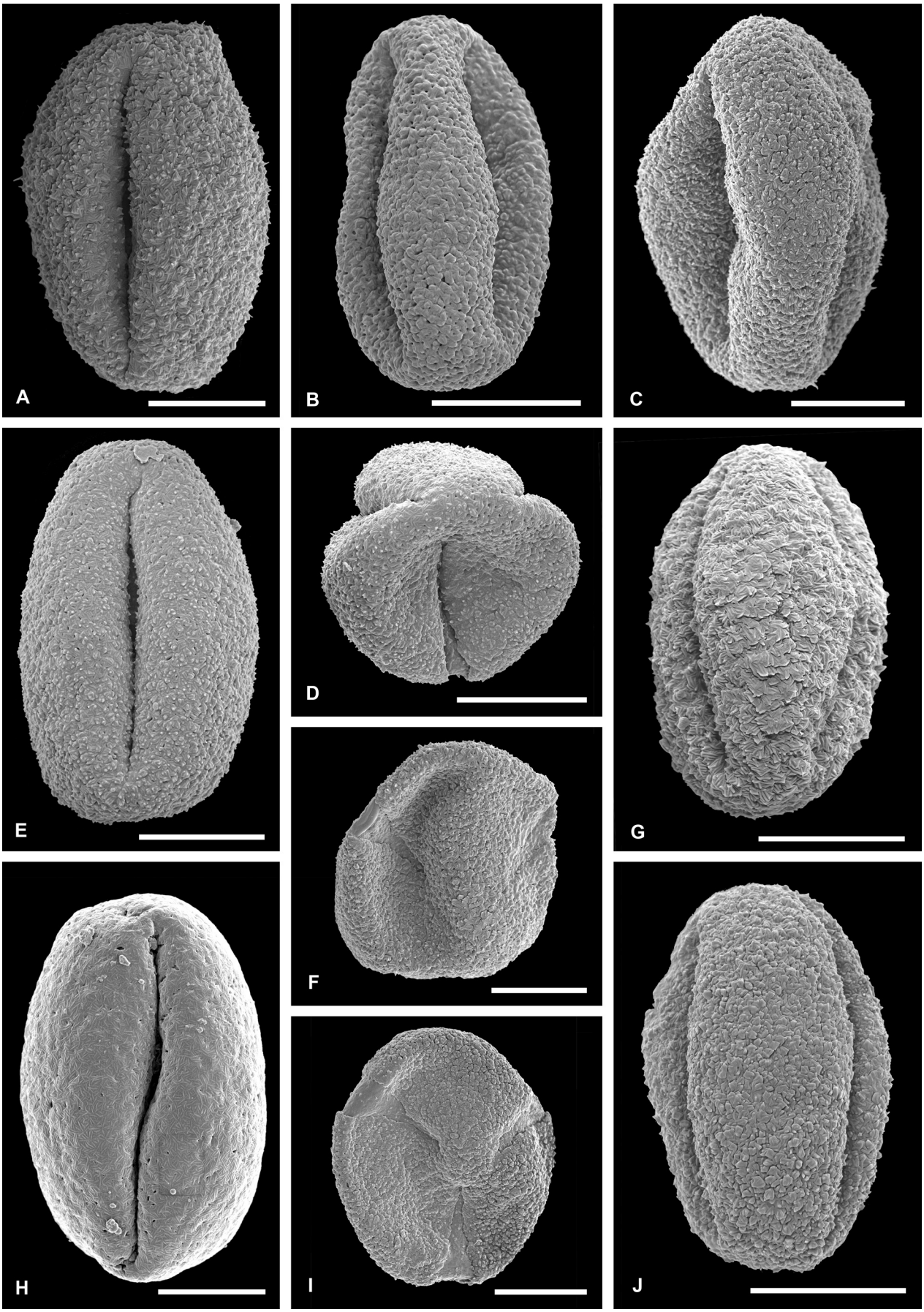
Figure 3. Pollen of *Quercus* Group Ilex, scanning electron micrographs. **A, B, C, E, G, H, J.** Equatorial view. **D, F, I.** Polar view. **A.** *Quercus acrodonta*, G. Forrest 16522 (E). **B.** *Quercus alnifolia*, Davis s. n. (E). **C.** *Quercus alnifolia*, Foggie 215 (E). **D.** *Quercus alnifolia*, s. n. (E). **E.** *Quercus aucheri*, ISTO 8007. **F.** *Quercus aucheri*, ISTO 19257. **G–I.** *Quercus baloot*; **G.** Koetz 8222 (E); **H.** J. E. T. Aitchison (S); **I.** Bowes Lyon 616 (E). **J.** *Quercus coccifera*, Lenander 23.4.36 (S). Scale bars – 10 μm.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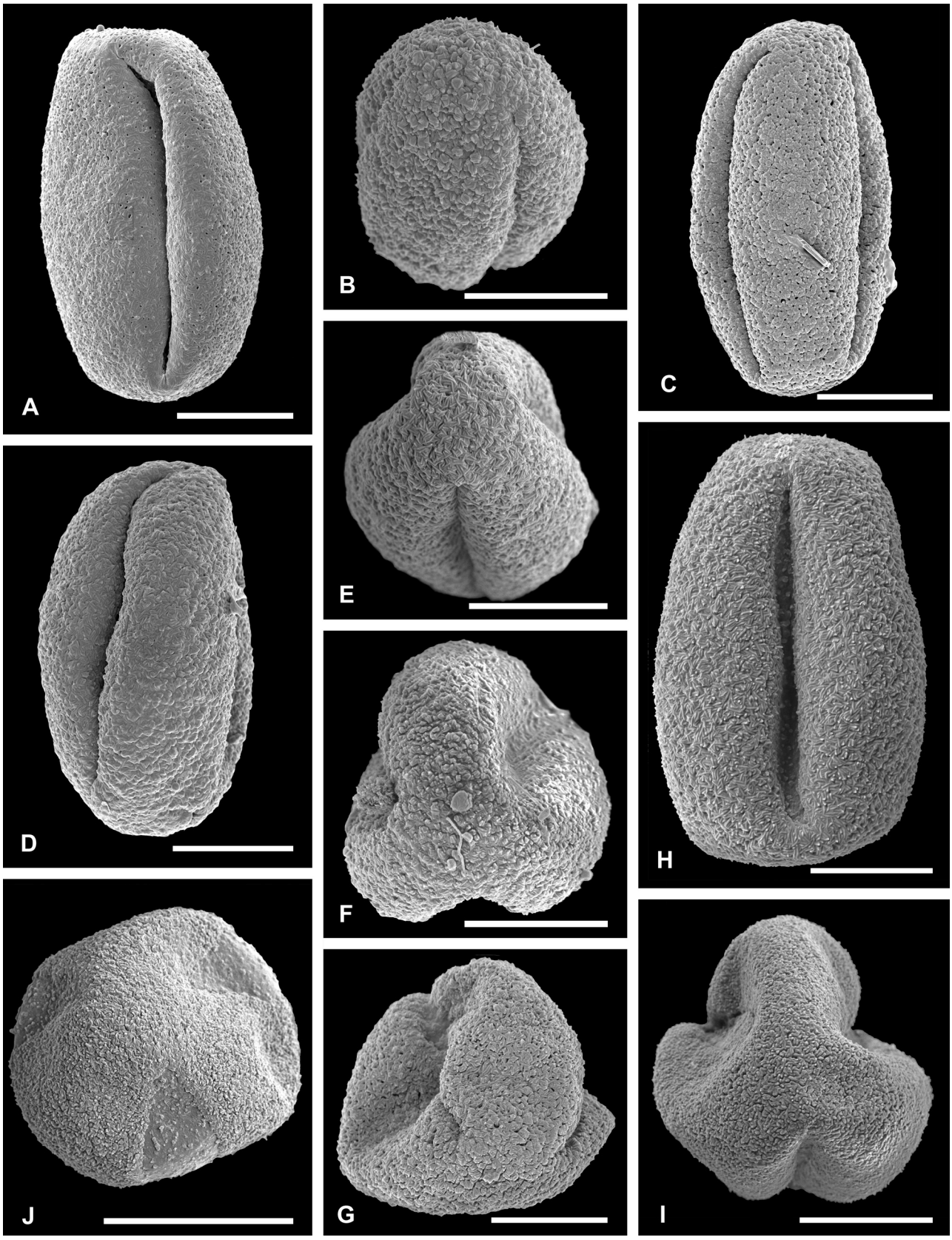
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Table II. Pollen dimensions of members of *Quercus* Group Ilex.

Taxon	Polar axis (μm)	Equatorial diameter (μm)
Quercus acrodonta Seemen [incl. *Q. handeliana* A.Camus]	32–35	20-22[a]
Q. alnifolia Poech	23–35	15–25[a]
Q. aquifolioides Rehder et E.H.Wilson	23.1–25.2	23.1–24[b]
Q. aucheri Jaub. et Spach	27–31	17–27[a]
Q. baloot Griff.	27–37	14–25[a]
Q. baronii Skan	21–23.1	22–23.1[b]
Q. coccifera L.	19–33	14–23[a]
	20.2–31	16.4–22.8[c]
Q. cocciferoides Hand.-Mazz.	25–33	17–24[a]
	21.2–27.8	19.8–23.1[b]
Q. dilatata Royle	32–36	18–22[a]
Q. dolicholepis A.Camus [incl. *Q. spathulata* Seemen]	18–31	13–21[a]
	27.3–33.5	21–27.5[b]
Q. engleriana Seemen	25.2–37.8	21–35.4[b]
Q. franchetii Skan	22–32	14–22[a]
	25.2–27.3	23.2–26.3[b]
Q. gilliana Rehder et E.H.Wilson	24–32	17–23[a]
	29.4–37.5	25.2–31.5[b]
Q. guyavifolia H.Lév.	–	24–32[a]
	31.5–37.8	25.2–31.3[b]
Q. ilex L.	(21) 25–38	(15) 17–28[a]
	21.2–30.2	15.2–24.7[c]
Q. kingiana Craib		23–26[a]
Q. lanata Sm.	30–34	18–25[a]
Q. leucotrichophora A.Camus [incl. *Q. incana* Roxb.]	31–33	19–27[a]
Q. lodicosa E.F.Warb.	26–29	19–22[a]
Q. longispica (Hand.-Mazz.) A.Camus	29–35	21–23[a]
	27.3–31.5	23.1–29.4[b]
Q. monimotricha (Hand.-Mazz.) Hand.-Mazz.	27.3–33.1	16.8–23.1[b]
Q. pannosa Hand.-Mazz.	27–28	22–26[a]
	27.3–31.5	21–29.4[b]
Q. phillyreoides A.Gray	24–33	17–22[a]
	24–29.6	16–23.1[b]
Q. rehderiana Hand.-Mazz.	22–29	20–23[a]
	23.1–27.3	19–24.1[b]
Q. semecarpifolia Sm.	22–36	21–25[a]
	25.2-35.7	23.1–33.6[b]
Q. senescens Hand.-Mazz.	23–30	20–25[a]
	29.4–33.6	21–25.2[b]
Q. spinosa David	25–28	20–25[a]
	23.1–26.2	21–25.2[b]
Q. fimbriata Chun et C.C.Huang ex Y.C.Hsu et H.W.Jen[d]	27.3–33.6	22–26.8[b]
Q. marlipoensis Hu et W.C.Cheng	No data	No data
Q. oxyphylla (E.H.Wilson) Hand.-Mazz.	No data	No data
Q. setulosa Hickel et A.Camus	No data	No data
Q. tarokoensis Hayata	No data	No data
Q. utilis Hu et W.C.Cheng	No data	No data
Quercus Group Ilex	18–38	14–34
Quercus Group Cerris	26–40	24–38
Quercus Group Cyclobalanopsis	19–33	17–33
Quercus Group Protobalanus (SEM)	20–30 (40)	14–23
Quercus Group Quercus	23–50	21–46
Quercus Group Lobatae (SEM)	36–42	22–26

[a]This study; [b]Wang & Pu (2004); [c]Beug (2004); [d]Probably a hybrid between *Q. aquifolioides* and *Q. dolicholepis*; Taxonomy follows IOPI (1996–2007).

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Figure 4. Pollen of *Quercus* Group Ilex, scanning electron micrographs. **A, C, D, H.** Equatorial view. **B, E, F, J, G, I.** Polar view. **A.** *Quercus coccifera*, Wall 9.5.33 (Palestina) (S); Microrugulate, perforate grain. **B.** *Quercus coccifera*, Lenander 23.4.36 (S). **C.** *Quercus cocciferoides*, P. Simeon Tenore 246 (E); Distinctly perforate. **D.** *Quercus dilatata*, Duthie 12949 (E). **E.** *Quercus dilatata*, A. Andersson 4_1915. **F.** *Quercus engleriana*, E. H. Wilson 3633 (E). **G.** *Quercus franchetii*, Henry 9298A (E). **H.** *Quercus gilliana*, Forrest 2056 (E); Note ornamentation of colpus membrane. **I.** *Quercus gilliana*, H. Mazzetti 1556 (E). **J.** *Quercus guyavifolia*, Chamberlain et al. 004 (E). Scale bars – 10 μm.

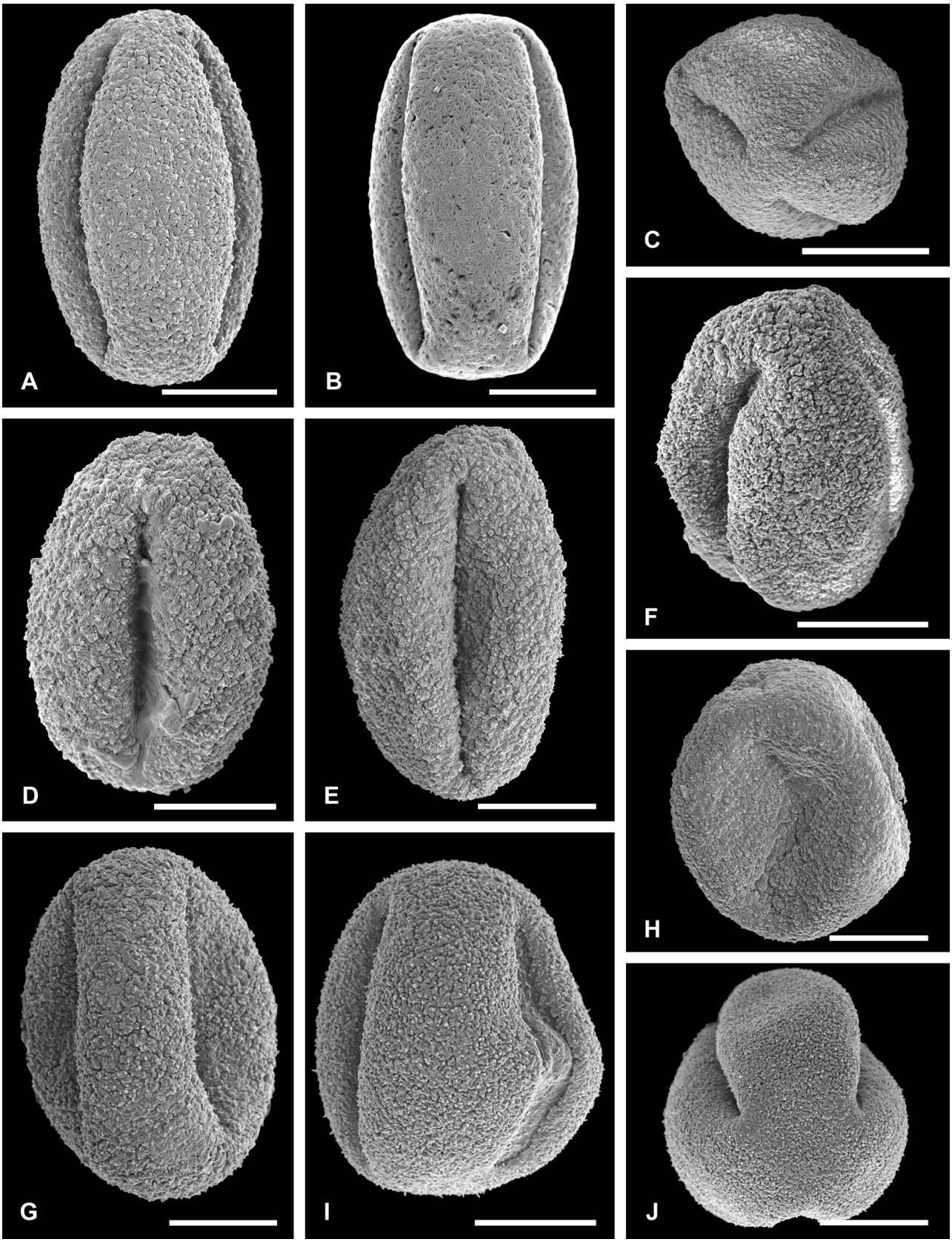


Figure 5. Pollen of *Quercus* Group Ilex, scanning electron micrographs. **A, B, D, E, G, I.** Equatorial view, **C, H, J.** Polar view. **F.** Oblique polar view. **A.** *Quercus ilex*, A. Uggla (S). **B.** *Quercus ilex*, I. Segelberg 19.5.1964 (S). **C.** *Quercus kingiana*, Hosseus 300_282 (E). **D.** *Quercus lanata*, Kumrun s. n. (E). **E.** *Quercus leucotrichophora*, Stainton 207 (E). **F.** *Quercus lodicosa*, G. Forrest 26611 (E). **G.** *Quercus longispica*, G. Forrest 21318 (E). **H.** *Quercus monimotricha*, E. H. Wilson 3626 (E). **I, J.** *Quercus pannosa*, ACE 506 (E). Scale bars – 10 μm.

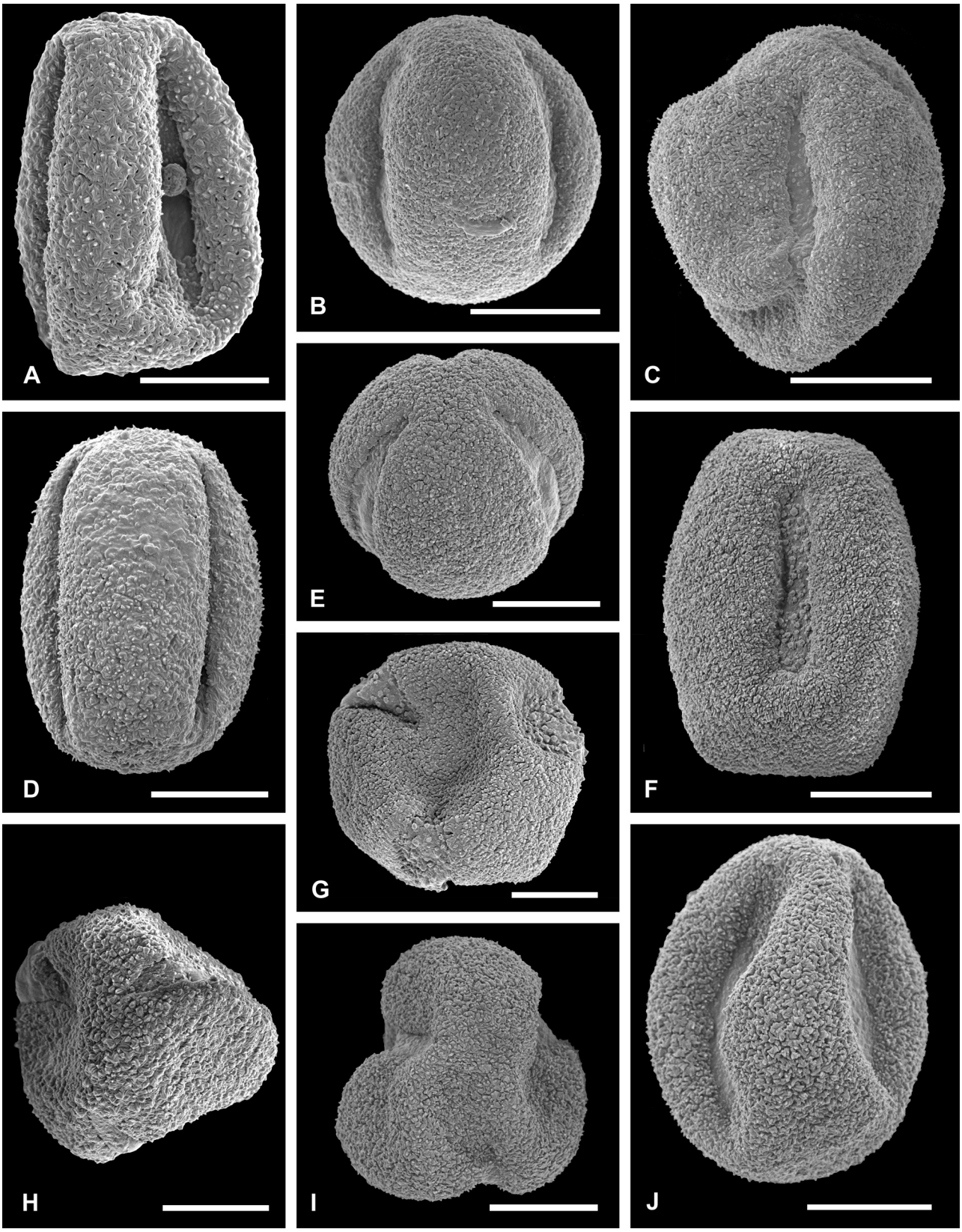


Figure 6. Pollen of *Quercus* Group Ilex, scanning electron micrographs. **A, B, C, D, F, J.** Equatorial view. **E, G, H, I.** Polar view. **A.** *Quercus phillyreoides*, 27.4.1911 (S). **B.** *Quercus rehderiana*, E. E. Maire s. n. (E). **C.** *Quercus rehderiana*, J. F. Rock 24264 (E). **D.** *Quercus semecarpifolia*, J. E. T. Aitchison 394 (S). **E.** *Quercus semecarpifolia*, Parkinson 4363 (S). **F.** *Quercus senescens*, Ducloux 1265 (E). Note verrucate ornamentation of colpus membrane. **G.** *Quercus senescens*, Chamberlain 004 (E). **H.** *Quercus spathulata*, E. H. Wilson 3625 (E). **I.** *Quercus spinosa*, E. H. Wilson 1493 (E). **J.** *Quercus spinosa*, E. H. Wilson 1493 (E). Scale bars – 10 μm.

Table III. Pollen ultrastructure of members of *Quercus* Group Ilex.

Species	Ectexine (μm)	Tectum (μm)	Columella		Foot layer (μm)	
			Hight (μm)	Width (μm)		
Q. alnifolia	0.58 (0.45–0.86)	0.26 (0.13–0.35) [0.51 (0.45–0.6)][a]	0.16 (0.1–0.2)	0.18 (0.13–0.275)	0.13 (0.05–0.2)	ILE
Q. aucheri	0.81 (0.545–1.0) [0.82]	0.265 (0.125–0.41) [0.49 (0.45–0.68)][a]	0.18 (0.125–0.27)	0.24 (0.125–0.375)	0.205 (0.09–0.32)	
Q. baloot	0.91 (0.6–1.47)	0.44 (0.2–0.73) [0.76][a]	0.2 (0.13–0.3)	0.22 (0.13–0.37)	0.15 (0.07–0.2)	
Q. cocciferoides	0.85 (0.5–1.67)	0.31 (0.19–0.55)	0.25 (0.17–0.38)	0.25 (0.14–0.45)	0.22 (0.17–0.25)	
Q. dilatata	0.9 (0.58–1.23)	0.4 (0.19–0.69)	0.22 (0.15–0.375)	0.2 (0.08–0.38)	0.17 (0.08–0.31)	
Q. engleriana	0.85 (0.64–1.0) [1.14–1.32]	0.4 (0.27–0.545) [0.73 (0.64–0.82)][a]	0.14 (0.11–0.2)	0.23 (0.14–0.45)	0.13 (0.02–0.23)	
Q. franchetii	0.45 (0.38–0.54)	0.22 (0.19–0.23)	0.11 (0.08–0.15)	0.12 (0.06–0.15)	0.09 (0.06–0.17)	
Q. guyavifolia	1.06 (0.67–1.55)	0.48 (0.28–0.8)	0.22 (0.14–0.28)	0.205 (0.13–0.33)	0.23 (0.11–0.44)	
Q. ilex	0.8 (0.6–1.0)	0.33 (0.2–0.5)	0.22 (0.15–0.275)	0.21 (0.175–0.27)	0.2 (0.13–0.27)	
Q. lanata	0.85 (0.45–1.2)	0.46 (0.08–0.92)	0.225 (0.16–0.4)	0.21 (0.115–0.48)	0.11 (0.08–0.18)	
Q. semecarpifolia	1.07 (0.75–1.6)	0.575 (0.31–1.0)	0.15 (0.1–0.33)	0.21 (0.125–0.57)	0.21 (0.125–0.3)	
Q. spathulata	0.98 (0.625–1.625)	0.54 (0.27–1.06)	0.21 (0.125–0.31)	0.21 (0.13–0.31)	0.2 (0.125–0.375)	
Q. acutissima	0.68 (0.44–1.04) [0.79 (0.66–0.91)]	0.3 (0.2–0.45) [0.51 (0.4–0.69)][a]	0.22 (0.09–0.4)	0.3 (0.13–0.53)	0.08 (0.02–0.23)	CER
Q. afares	1.16 (0.69–1.56) [1.35 (0.94–1.73)]	0.39 (0.19–0.61) [0.71 (0.5–0.82)][a]	0.34 (0.23–0.5)	0.27 (0.18–0.41)	0.25 (0.18–0.41)	
Q. brantii subsp. *brantii*	1.27 (0.56–1.76)	0.67 (0.47–0.88)	0.36 (0.24–0.52)	0.24 (0.13–0.4)	0.23 (0.08–0.4)	
Q. cerris	0.95 (0.67–1.33) [1.35 (0.93–2.0)]	0.43 (0.33–0.6) [0.95 (0.65–1.66)][a]	0.19 (0.1–0.35)	0.27 (0.17–0.47)	0.1 (0.05–0.2)	
Q. ithaburensis subsp. *macrolepis*	0.68 (0.44–0.875)	0.27 (0.16–0.5) [0.6 (0.44–0.94)][a]	0.18 (0.125–0.28)	0.21 (0.125–0.375)	0.06 (0.03–0.125)	
Q. libani	0.69 (0.36–1.23)	0.38 (0.18–0.86)	0.13 (0.045–0.19)	0.21 (0.14–0.305)	0.08 (0.045–0.11)	
Q. suber	0.87 (0.6–1.25)	0.47 (0.3–0.75)	0.19 (0.15–0.25)	0.19 (0.125–0.275)	0.09 (0.05–0.15)	
Q. glauca	1.04 (0.875–1.23)	0.44 (0.375–0.59)	0.28 (0.22–0.32)	0.15 (0.125–0.25)	0.25 (0.12–0.36)	CYC
Q. longinux	0.83 (0.8–0.87)	0.49 (0.4–0.55)	0.21 (0.15–0.27)	0.2 j(0.125–0.3)	0.12 (0.07–0.15)	
Q. chrysolepis	1.315 (0.77–1.6)	0.57 (0.33–0.82)	0.24 (0.09–0.33)	0.28 (0.16–0.6)	0.34 (0.09–0.5)	
Q. tomentella	1.25 (1.0–1.67)	0.48 (0.33–0.83)	0.33 (0.17–0.47)	0.25 (0.17–0.4)	0.33 (0.17–0.5)	PRO
Q. vacciniifolia	0.99 (0.85–1.3)	0.435 (0.25–0.7)	0.2 (0.19–0.225)	0.24 (0.19–0.4)	0.25 (0.19–0.35)	

Q. infectoria subsp. *veneris* [*Q. boissieri*]	0.88 (0.5–1.7)	0.39 (0.13–0.7)	0.31 (0.2–0.5)	0.265 (0.17–0.47)	0.1 (0.07–0.2)
Q. dentata	0.51 (0.4–0.6) [0.92 (0.87–1.0)]	0.19 (0.1–0.33) [0.565 (0.53–0.67)][a]	0.18 (0.13–0.33)	0.23 (0.1–0.47)	0.09 (0.03–0.13)
Q. macranthera	0.71 (0.44–1.19) [1.24 (0.89–1.54)]	0.24 (0.11–0.42) [0.675 (0.5–0.96)][a]	0.23 (0.15–0.38)	0.38 (0.08–0.58)	0.17 (0.04–0.31)
Q. michauxii	1.01 (0.77–1.33) [1.35 (1.17–1.6)]	0.41 (0.2–0.73) [0.8 (0.67–1.0)][a]	0.31 (0.2–0.47)	0.27 (0.2–0.33)	0.225 (0.13–0.33)
Q. robur [*Q. pedunculiflora*]	1.0 (0.9–1.1)	0.41 (0.2–0.67)	0.24 (0.13–0.4)	0.27 (0.2–0.4)	0.15 (0.07–0.2)
Q. pyrenaica	0.99 (0.73–1.155) [1.395 (1.23–1.545)]	0.41 (0.23–0.64) [0.715 (0.59–0.82)][a]	0.24 (0.11–0.41)	0.25 (0.14–0.42)	0.21 (0.045–0.36)
Q. robur	0.94 (0.47–1.45)	0.33 (0.13–0.75)	0.23 (0.13–0.3)	0.2 (0.13–0.3)	0.11 (0.07–0.225)
Q. virginiana	0.74 (0.56–0.97) [0.84 (0.8–0.93)]	0.31 (0.16–0.53) [0.425 (0.37–0.47)][a]	0.21 (0.16–0.27)	0.19 (0.13–0.27)	0.14 (0.03–0.2)
Q. nigra	0.51 (0.3–1.0) [0.6–0.67]	0.2 (0.05–0.43) [0.48 (0.4–0.67)][a]	0.19 (0.1–0.27)	0.23 (0.13–0.37)	0.11 (0.05–0.2)
Q. laevis	0.65 (0.45–0.92) [0.93 (0.65–1.12)]	0.22 (0.1–0.4) [0.46 (0.3–0.6)][a]	0.235 (0.13–0.33)	0.25 (0.18–0.425)	0.13 (0.05–0.24)

Note: Tectum: values in rectangular brackets refer to local thickenings of the tectum, such as verrucae.
[a]Average height of foot layer/average height of tectum with verrucae [without verrucae].
Note: ILE = Group Ilex. CER = Group Cerris. CYC = Group Cyclobalanopsis. PRO = Group Protobalanus. QUE = Group Quercus. LOB = Group Lobatae

Figure 11N; 'type 2' foot layer). The thickness of the foot layer ranges from < 0.1 to 0.2 µm and may increase to 0.4 µm locally. The ratio between foot layer and tectum (based on average thickness in the mesocolpium) approaches one in some species (*Q. aucheri*, *Q. cocciferoides*), in which the foot layer is relatively thick (type 1 foot layer). Other species have a comparatively thin foot layer, resulting in foot layer/tectum ratios of 0.25 to 0.35 (*Q. baloot*, *Q. engleriana* Seemen, *Q. lanata*, *Q. semecarpifolia*; Table IV). The height of the columellae is between 0.1 and 0.2 µm and does not show much variation. In contrast, the thickness of the tectum is considerably variable among species. Average thickness of the tectum ranges from 0.3 to 0.6 µm; locally, the tectum may be as thick as 1 µm (e.g. *Q. semecarpifolia*, Figure 11O).

Quercus Groups Cerris, Cyclobalanopsis, Protobalanus, Lobatae and Quercus (Figures 13–15)

Group Cerris. — The tectum ornamentation is verrucate (Figure 13A, C; see Denk & Grimm 2009, for details). As in Group Ilex, the endexine in non-apertural regions may be homogeneous and of even thickness (e.g. Figure 13G) or coarsely laminated (Figure 13C, E). Towards the apertural region, the endexine becomes coarsely laminated (Figure 13D). The thickness of the endexine ranges from 0.1 to 0.2 µm in the non-apertural region and from 0.3 to 0.65 µm in the apertural region (Table III).

The foot layer may be discontinuous or continuous and of even or uneven thickness. The foot layer commonly becomes discontinuous and patchy towards the apertural region (Figure 13D). In the non-apertural region, the foot layer usually is markedly disrupted and irregular (e.g. *Quercus suber* L., Figure 13A, B; type 2 foot layer) or, rarely, thicker, more regular and nearly continuous in a few of the investigated taxa (e.g. *Q. brantii* Lindl., Figure 13C; approaching type 1 foot layer). As in Group Ilex, the thickness of the foot layer ranges from < 0.1 to 0.2 µm and may increase to 0.4 µm locally. The ratio between foot layer and tectum (based on average thickness in the mesocolpium) ranges from 0.19 (*Q. suber*) to 0.34 (*Q. brantii*) and is generally less than in Group Ilex (Table IV). The height of the columellae is between 0.1 and 0.5 µm and hence is more variable than in Group Ilex. Also the thickness of the tectum is considerably variable among species (0.2 to 1 µm), which is due to the irregular presence of verrucae on the tectum (Figure 13A–G).

Table IV. Pollen ultrastructure of members of the six infrageneric Groups of *Quercus*: foot layer in mesocolpium region.

Species	Properties of the foot layer in the genus *Quercus*			
	Continuous/discontinuous	Regular/irregular	Ratio foot layer/tectum (based on averages)	
Quercus alnifolia	Continuous. occasionally disrupted	Regular	0.5 (locally 0.6-0.75)	ILE
Q. aucheri	Continuous	Regular	0.8	
Q. baloot	Discontinuous	Irregular	0.35	
Q. cocciferoides	Continuous. occasionally disrupted	Regular	0.7	
Q. dilatata	Discontinuous	Irregular	0.4	
Q. engleriana	Continuous/discontinuous	Regular/irregular	0.3	
Q. franchetii	Continuous	Regular	0.4	
Q. guyavifolia	Continuous. occasionally disrupted	Regular	0.5	
Q. ilex	Continuous. occasionally disrupted	Regular/irregular	0.6	
Q. lanata	Markedly discontinuous	Irregular	0.25	
Q. semecarpifolia	Continuous. occasionally disrupted	Regular/irregular	0.35	
Q. spathulata	Continuous. occasionally disrupted	Irregular	0.4	
Q. acutissima	Discontinuous	Irregular	0.15 [0.26][a]	CER
Q. brantii subsp. *brantii*	Nearly continuous	Regular	0.34	
Q. cerris	Discontinuous	Irregular	0.23	
Q. ithaburensis subsp. *macrolepis*	Discontinuous	Irregular	0.1 [0.22][a]	
Q. libani	Discontinuous	Irregular	0.21	
Q. suber	Discontinuous	Irregular	0.19	
Q. glauca	Nearly continuous	Regular	0.56	CYC
Q. longinux	Discontinuous	Irregular	0.24	
Q. chrysolepis	Nearly Continuous	Regular	0.6	PRO
Q. tomentella	Nearly continuous	Regular	0.7	
Q. vacciniifolia	Nearly continuous	Regular	0.6	
Q. infectoria subsp. *veneris*			0.25	QUE
Q. dentata	Discontinuous	Irregular	0.16 [0.47][a]	
Q. macranthera	Discontinuous	Irregular	0.25 [0.7][a]	
Q. michauxii	Discontinuous	Irregular	0.28 [0.5][a]	
Q. robur [as *Q. pedunculiflora*]	Discontinuous	Irregular	0.36	
Q. petraea	Discontinuous	Irregular	0.35	
Q. pyrenaica	Nearly continuous	Regular/irregular	0.29 [0.51][a]	
Q. robur	Discontinuous	Irregular	0.33	
Q. virginiana	Discontinuous	Irregular	0.33 [0.45][a]	
Q. nigra	Discontinuous	Irregular	0.2 [0.55][a]	LOB
Q. laevis	Discontinuous	Irregular	0.28 [0.59][a]	

[a]Average height of foot layer/average height of tectum with verrucae [without verrucae].
Note: ILE = Group Ilex. CER = Group Cerris. CYC = Group Cyclobalanopsis. PRO = Group Protobalanus. QUE = Group Quercus. LOB = Group Lobatae

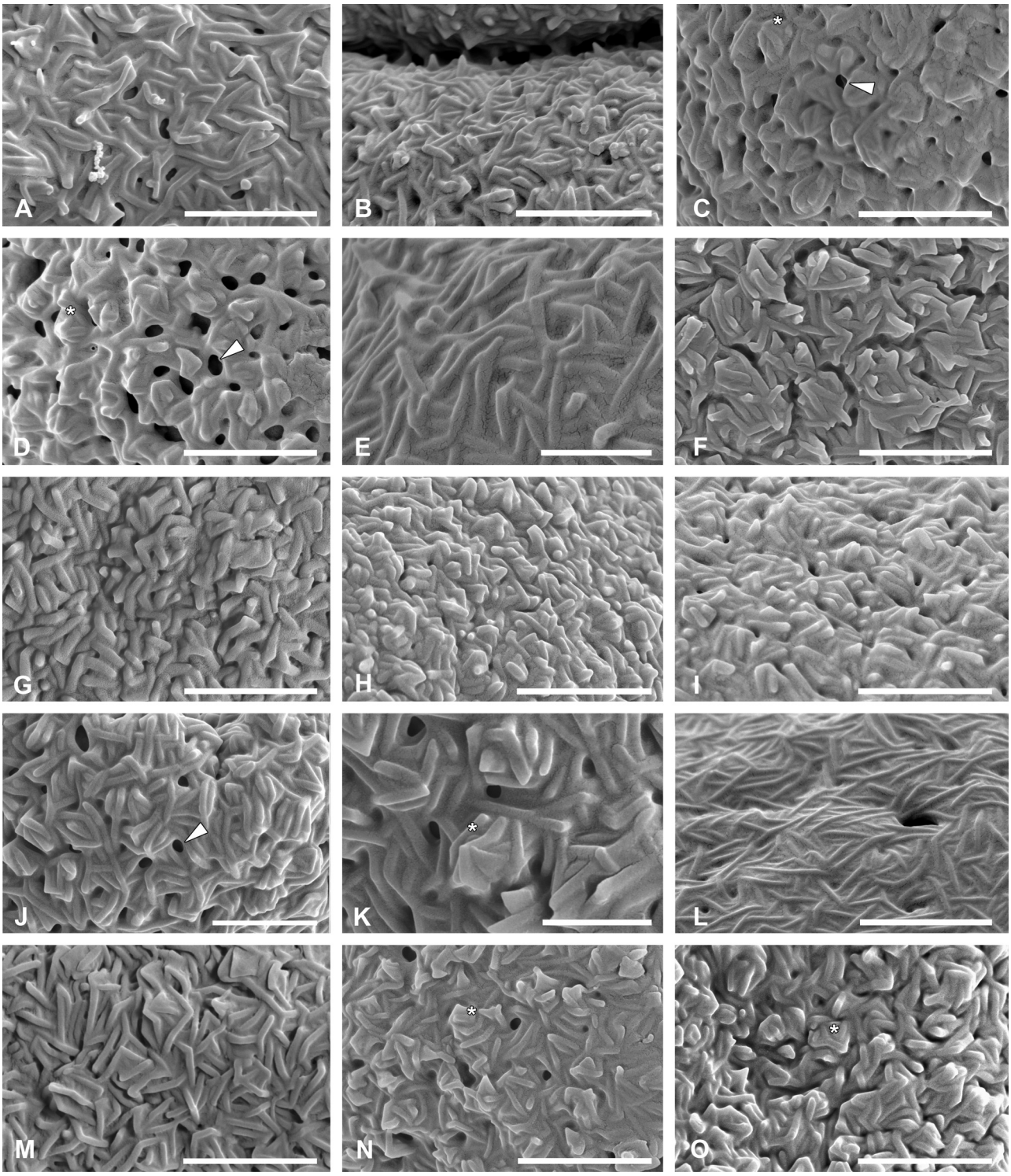


Figure 7. *Quercus* Group Ilex, details of pollen surface, scanning electron micrographs. **A.** *Quercus acrodonta*, G. Forrest 16522 (E). **B–F.** *Quercus alnifolia*; **B–E.** Davis s. n. (E); Note agglomerated rugulae and perforations (*white arrowheads*); **F.** Foggie 215 (E). **G.** *Quercus aquifolioides*, G. Forrest 10026 (E). **H–K.** *Quercus aucheri*; **H, I.** ISTO 8007; **J, K.** ISTO 19257; Note perforations (*arrowhead*) and desert-rose-like structures of rugulae (*asterisk*). **L–O.** *Quercus baloot*; **L.** Aitchison s. n. (S); **M.** Koeltz 222 (E); **N, O.** Bowes Lyon 616 (E). Scale bars – 2 µm (A–D, F–J, L–O), 1µm (E, K).

Group Cyclobalanopsis. — The tectum ornamentation is microrugulate with vertically oriented rugulae (rod-like vertical according to Denk & Grimm 2009). The endexine and foot layer are variable (Table III). The ratio between foot layer and tectum (based on average thickness in the mesocolpium

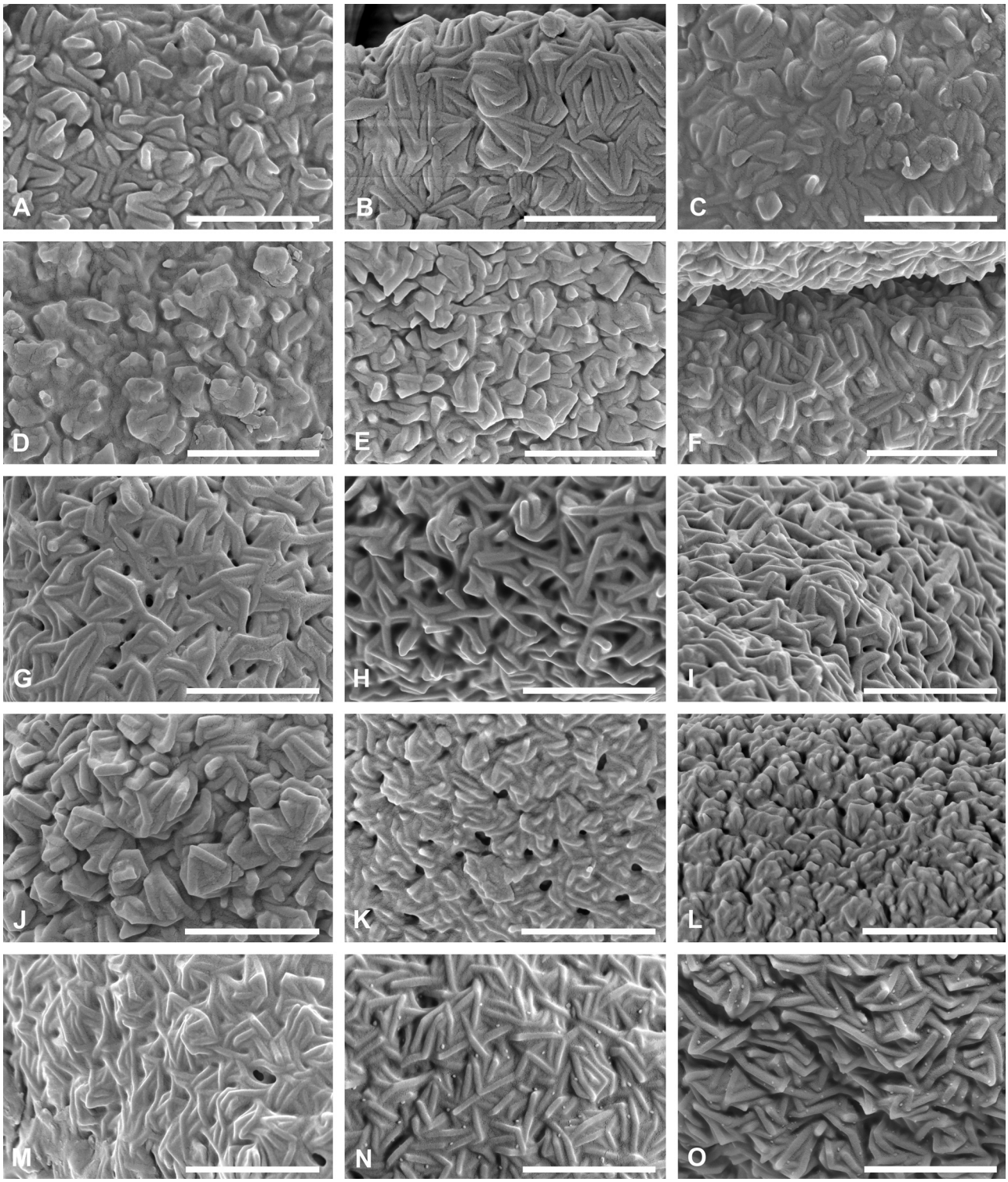


Figure 8. *Quercus* Group Ilex, details of pollen surface, scanning electron micrographs. **A, B.** *Quercus baronii*, G. H. Yang 58623 (PE). **C–J.** *Quercus coccifera*; **C.** Dinsmore (Sherafat) (S); **D–F.** J. Lenander 23.4.36 (S); **G, I.** E.H. Wall 9.5.33 (Palestine) (S); **H.** G. Een 22.4.52 (S). **K, L.** *Quercus cocciferoides*; **K.** Handel Mazzetti 3693 (E); **L.** P. Simeon Tenore 45 (E). **M–O.** *Quercus dilatata*, A. Andersson 4_1915 (E). Scale bars – 2 μm.

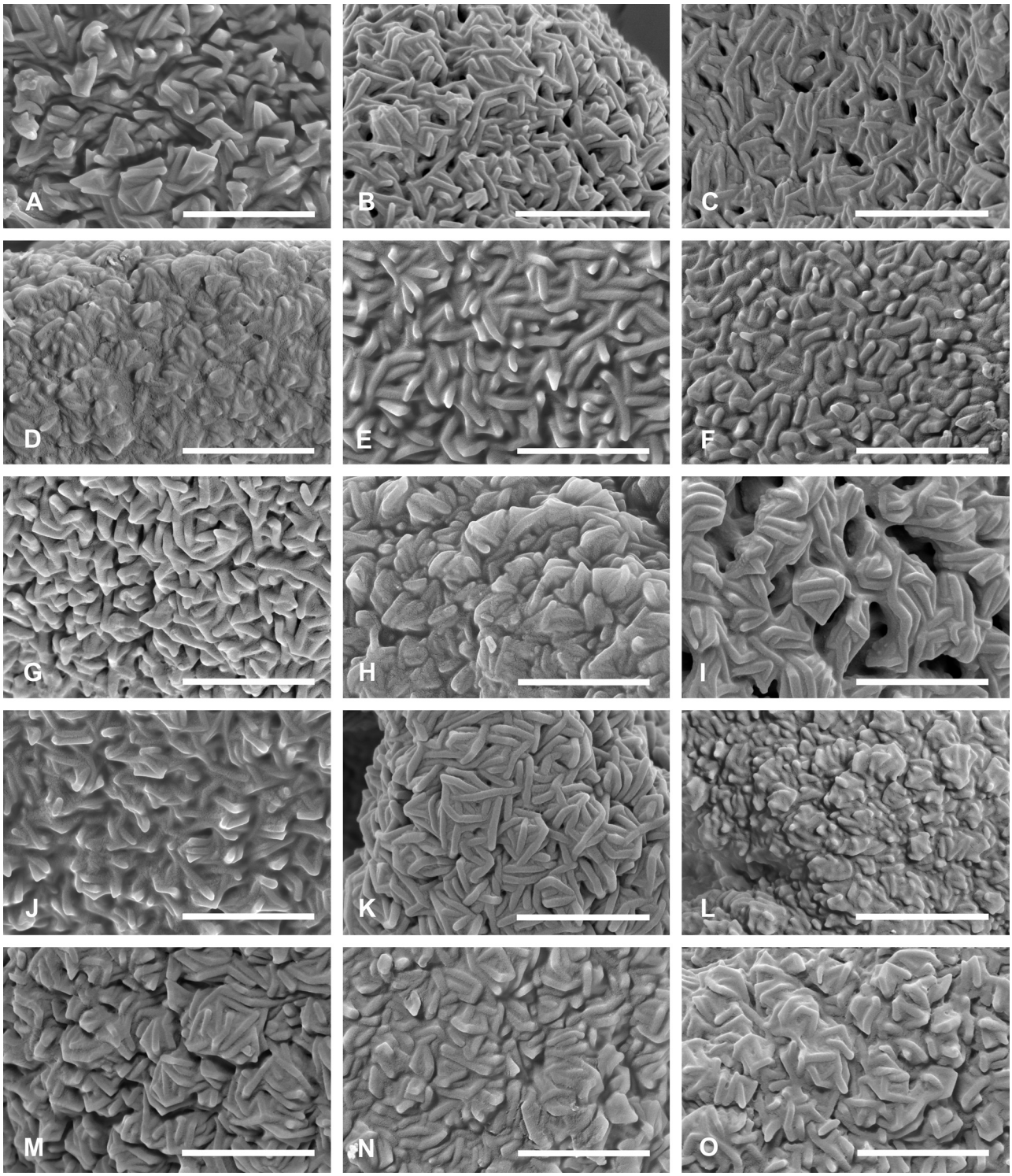
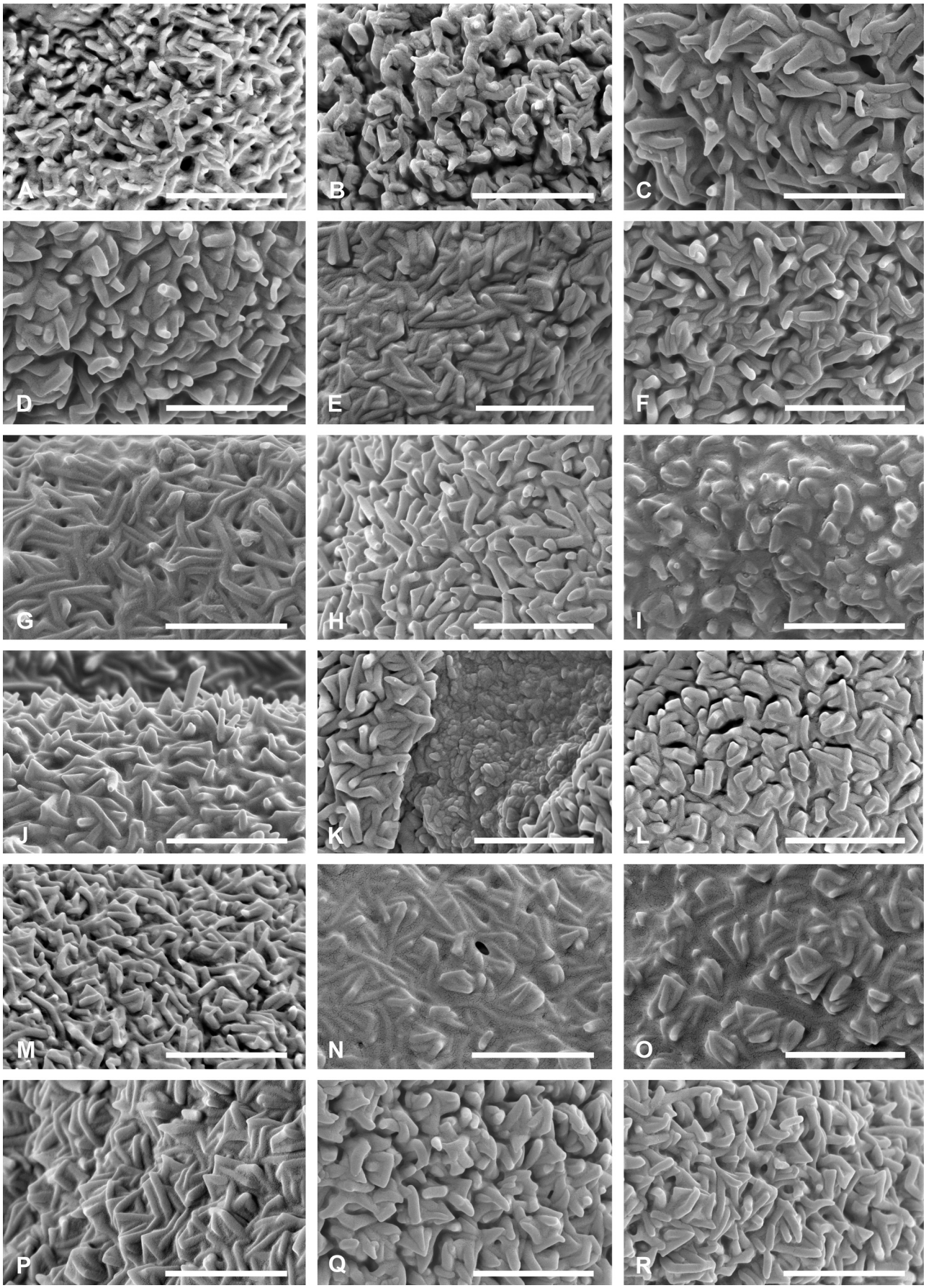


Figure 9. *Quercus* Group Ilex, details of pollen surface, scanning electron micrographs. **A.** *Quercus engleriana*, E. H. Wilson 3633 (E). **B–D.** *Quercus francheti;* **B, C.** Henry 9298A (E); **D.** Smitinand 6781_24519 (E). **E, F.** *Quercus gilliana*; **E.** Forrest 2056 (E); **F.** H. Mazzetti 1556 (E). **G.** *Quercus guyavifolia*, Chamberlain et al. 004 (E). **H–K.** *Quercus ilex*; **H.** ISTF s. n.; **I.** Anisits 30.5.1911 (S); **J.** H. Lenander 18.5.1933 (S); **K.** A. Uggla s. n. 20.4.1936 (S). **L.** *Quercus kingiana*, Hosseus 300_282 (E). **M, N.** *Quercus lanata*; **M.** Kumrun s. n. (E); **N.** Dickason 8435 (E). **O.** *Quercus leucotrichophora*, Stainton 207 (E) [=*Q. lanata*]. Scale bars – 2 μm.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area) is 0.24 (*Quercus longinux* Hayata) and 0.56 (*Q. glauca* Thunb.; Figure 13H, I) and is within the variability encountered among members of Group Ilex (Table IV). The height of the columellae is between 0.1 and 0.2 µm; the thickness of the tectum ranges from 0.5 to 0.8 µm in the two species investigated.

Group Protobalanus. — The tectum ornamentation is rod-like masked according to Denk and Grimm (2009) without distinct verrucae; Figure 14A–C). While the endexine is fragmentary in the non-apertural region and characteristically thickened in the apertural region, all investigated species of this group have a continuous, homogeneous and relatively thick foot layer (type 1 foot layer; Table III). The ratio between foot layer and tectum ranges from 0.6 to 0.7 (Table IV). The height of the columellae is between 0.2 and 0.3 µm (occasionally to 0.4 µm) long; the thickness of the tectum ranges from 0.4 to 0.8 µm.

Groups Quercus and Lobatae. — The tectum ornamentation is verrucate (Figure 15). The endexine is irregular and coarsely laminated, its thickness varies considerably (Figure 15C; Table III). In the apertural region, it is markedly thicker and coarsely laminated. The foot layer is generally thin, irregular and discontinuous (Figure 15A, B, F; type 2 foot layer), but in some cases (*Quercus pyrenaica* Willd.), it may be thicker, regular and only occasionally disrupted (Figure 15C, D). The ratio between foot layer and tectum ranges from 0.16 to 0.36. The height of the columellae is between 0.18 (*Q. dentata* Thunb.) and 0.31 µm (*Q. infectoria* Oliv., *Q. michauxii* Nutt.), but locally varies from 0.1 to 0.4 µm; the thickness of the tectum ranges from 0.2 to 0.4 µm in areas without supratectal verrucae to 0.3 to 1 µm in areas with verrucae.

Discussion

Variability in pollen ornamentation

Pollen ornamentation is consistent among all species of *Quercus* Group Ilex. This has previously been suggested by Denk and Grimm (2009). Nevertheless, there is a substantial intraspecific variation in members of Group Ilex. This has been documented for other infrageneric groups of *Quercus* (Group Quercus: Solomon 1983a; Group Lobatae: Solomon 1983b; Group Cyclobalanopsis: Makino et al. 2009). It is noteworthy that pollen ornamentation in Group Quercus (white oaks) and Group Lobatae (red oaks) is conservative to the extent that morphologically and ecologically conspicuously different species have virtually indistinguishable pollen ornamentation. For example, nearly identical pollen is found in the North American white oaks *Q. alba* L. and *Q. virginiana* Mill. (Solomon 1983a), of which the former has deciduous lobed leaves while the latter has evergreen, coriaceous, entire-margined leaves. Borsch and Wilde (2000) investigated the variability of tectum ornamentation below species level and found considerable variation of pollen ornamentation in pollen grains from the same inflorescence of *Q. ilex* L. (Group Ilex). According to these authors, to Rowley (1996) and Rowley and Gabarayeva (2004), microrugulae and aggregations of microrugulae are masked to varying degrees by sporopollenin, which accounts for some of this morphological variability. For *Nelumbo*, Borsch and Wilde (2000) documented pollen morphological variability in a single flower and found a substantial degree of variation in size, degree of branching and fusion of basic sculpture elements. Individuals and populations showed a certain morphological variability, while more distinct pollen morphotypes were encountered in different populations. Borsch and Wilde (2000) stated that although highly similar pollen grains may be encountered in different populations, 'there is no regularity'. From the present data of species of *Quercus* Group Ilex, similar conclusions can be drawn. Although a much larger sample size would be desirable and potentially would reveal a pattern within the encountered variability, at the moment, we do not expect to find a geographic or taxonomic pattern in pollen ornamentation of Ilex oaks. Within the range of variability encountered for *Quercus* Group Ilex, certain types are also found in other genera of Fagaceae. For example,

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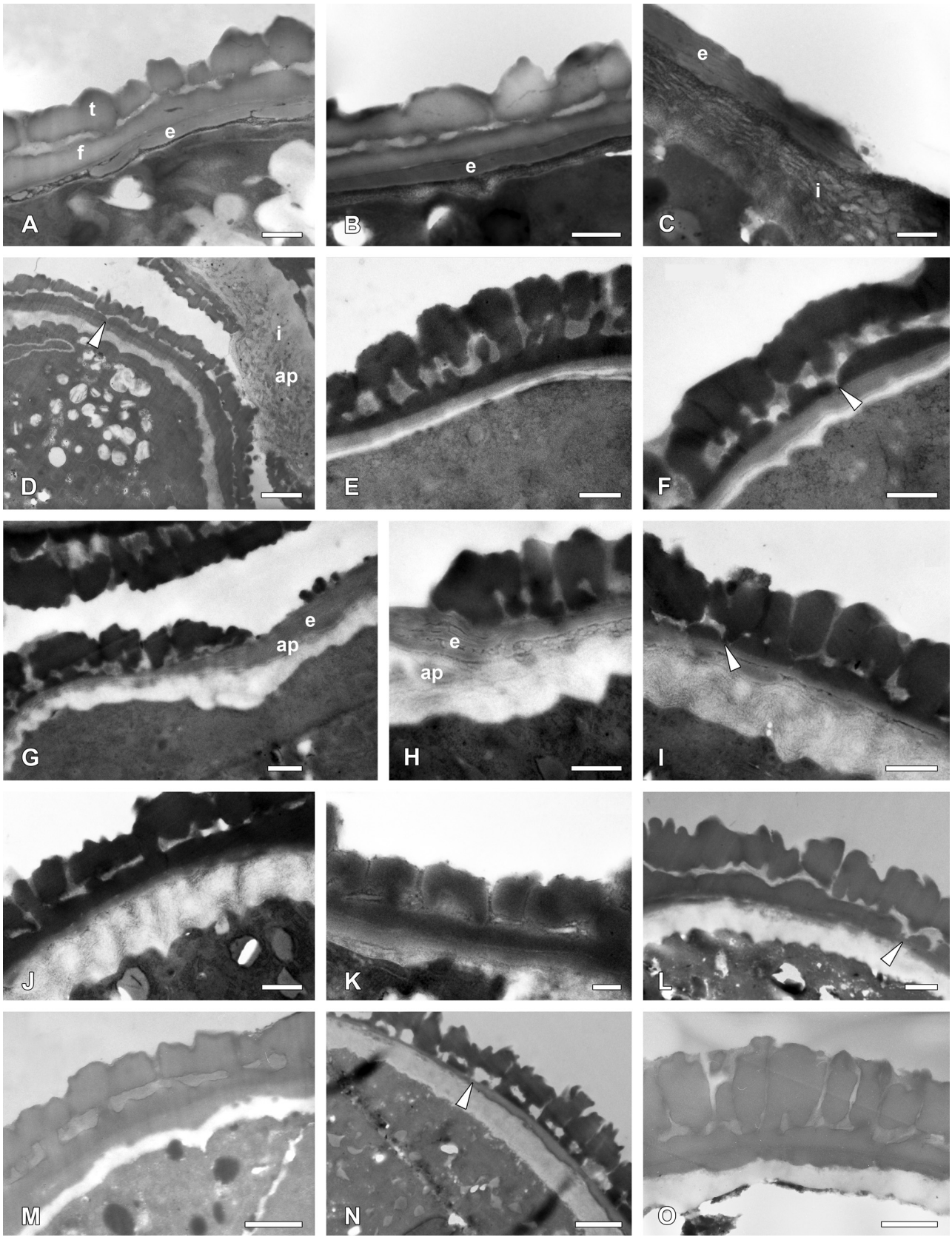
Figure 10. *Quercus* Group Ilex, details of pollen surface, scanning electron micrographs. **A, B.** *Quercus lodicosa*, G. Forrest 26611 (E). **C, D.** *Quercus longispica*, G. Forrest 21318 (E). **E.** *Quercus monimotricha*, E. H. Wilson 3626 (E). **F.** *Quercus pannosa*, ACE 506 (E). **G.** *Quercus phillyreoides*, s. n. 27.4.1911 (S). **H.** *Quercus rehderiana*, J. F. Rock 24264 (E). **I–K.** *Quercus semecarpifolia*; **I, J.** J. E. T. Aitchison 394 (S); **K.** Parkinson 4363 (S). **L, M.** *Quercus senescens*; **L.** Chamberlain 004 (E); **M.** Ducloux 1265 (E). **N–P.** *Quercus spathulata*; **N, O.** G. Forrest 12397 (E); **P.** E. H. Wilson 3625 (E). **Q, R.** *Quercus spinosa*, E. H. Wilson 1493 (E). Scale bars – 2 µm.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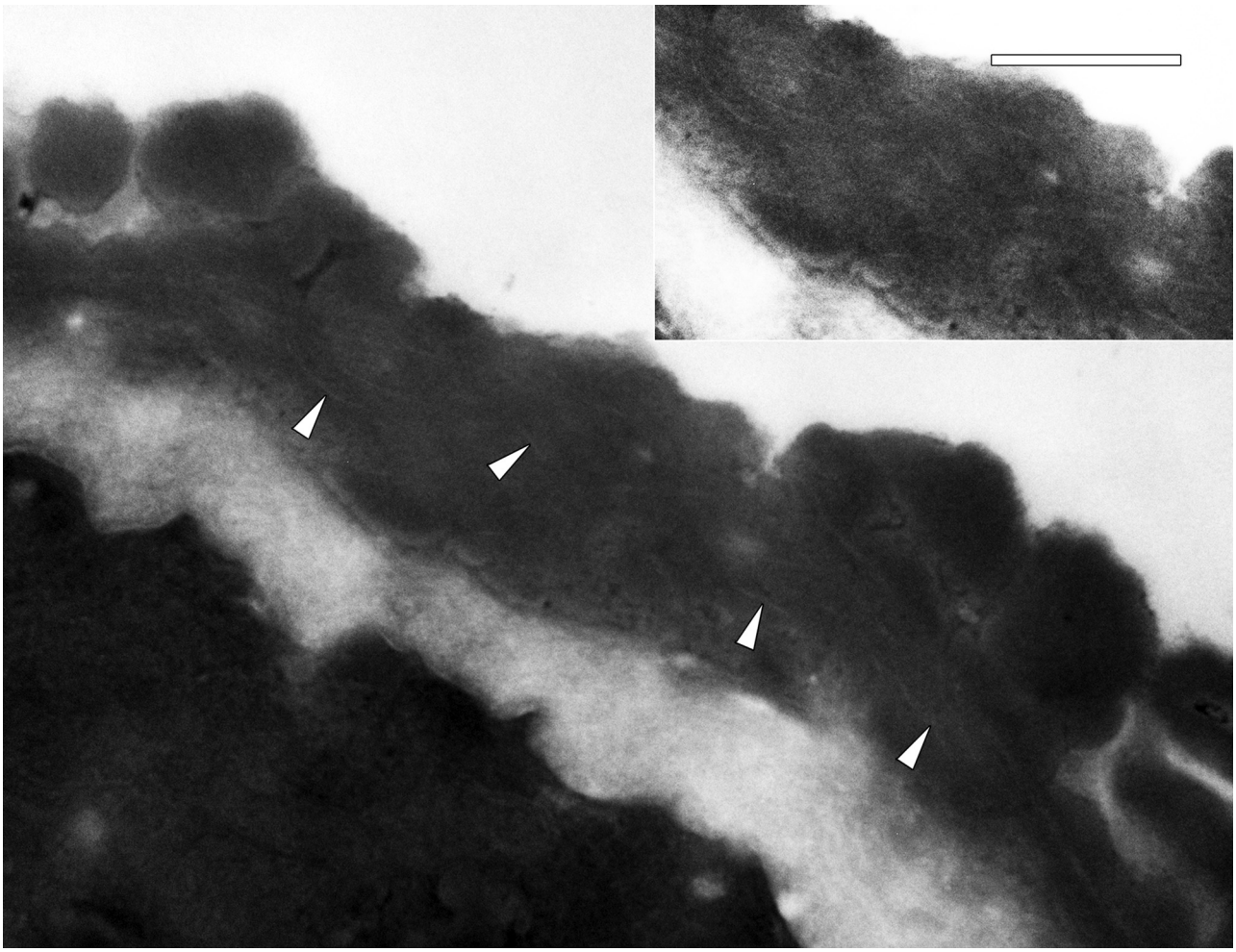

Figure 12. *Quercus* Group Ilex, pollen ultrastructure, transmission electron micrographs. *Quercus franchetii*, A. Henry 9298A (E). Apertural region. Compacted endexine displaying white lines (*arrowheads*); shown with normal and enhanced (inset) contrast to highlight white lines. Scale bar – 500 nm.

rugulae are occasionally in the same plane as the pollen surface and fuse to form a rugulate pattern with weak relief (Figure 7L). This pattern is reminiscent of the tectum sculpturing in Castaneoideae (e.g. *Castanopsis cuspidata* [Thunb.] Schottky; Praglowski 1984, fig. 4C).

Furthermore, a similar variability of tectum ornamentation as in *Quercus* Group Ilex is seen in *Fagus* (Praglowski 1982; Denk 2003) although the basic elements in *Fagus* are substantially larger than in *Quercus* (rugulate pattern in *Fagus* as opposed to predominantly microrugulate in *Quercus*).

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Figure 11. *Quercus* Group Ilex, pollen ultrastructure, transmission electron micrographs. **A–C.** *Quercus aucheri*, ISTO 19257; **A.** Section in mesocolpium region; foot layer approximately of same thickness as tectum; fragmented endexine; **B.** Same as (**A**), but homogeneous endexine; **C.** Section through aperture region; coarsely laminated endexine and intine. **D.** *Quercus cocciferoides*, Handel-Mazzetti 3632 (E); two neighbouring pollen grains sectioned in mesocolpium and aperture region; foot layer and endexine more or less homogeneous, continuous; absent in aperture region. **E–H.** *Quercus dilatata*, Duthie 12949 (E); **E.** Section through mesocolpium; continuous foot layer and endexine; **F.** Mesocolpium; discontinuous foot layer; **G.** Aperture region; thickened endexine with remnants of the ectexine on the colpus membrane; **H.** Coarsely laminated endexine in aperture region. **I–J.** *Quercus engleriana*, E. H. Wilson 3633 (E); **I.** Section through mesocolpium; discontinuous foot layer; **J.** Mesocolpium; continuous foot layer. **K.** *Quercus franchetii*, A. Henry 9298A (E); mesocolpium; continuous foot layer. **L.** *Quercus guyavifolia*, D. Chamberlain et al. 004 (E); mesocolpium; discontinuous foot layer and fragmented endexine. **M.** *Quercus ilex*, I. Segelberg 19.5.1964 (S); mesocolpium. continuous foot layer. **N.** *Quercus leucotrichophora* (as *Q. lanata*), J. D. A. Stainton 207 (E); mesocolpium; markedly discontinuous foot layer. **O.** *Quercus semecarpifolia*, J. E. T. Aitchison 394 (S); mesocolpium; continuous foot layer. Arrowheads indicate gaps in foot layer; e, endexine; i, intine; ap, aperture region; f, foot layer; t, tectum. Scale bars – 500 nm (A–C, E–J, L); 1 μm (D, N); 200 nm (K); 667 nm (M, O).

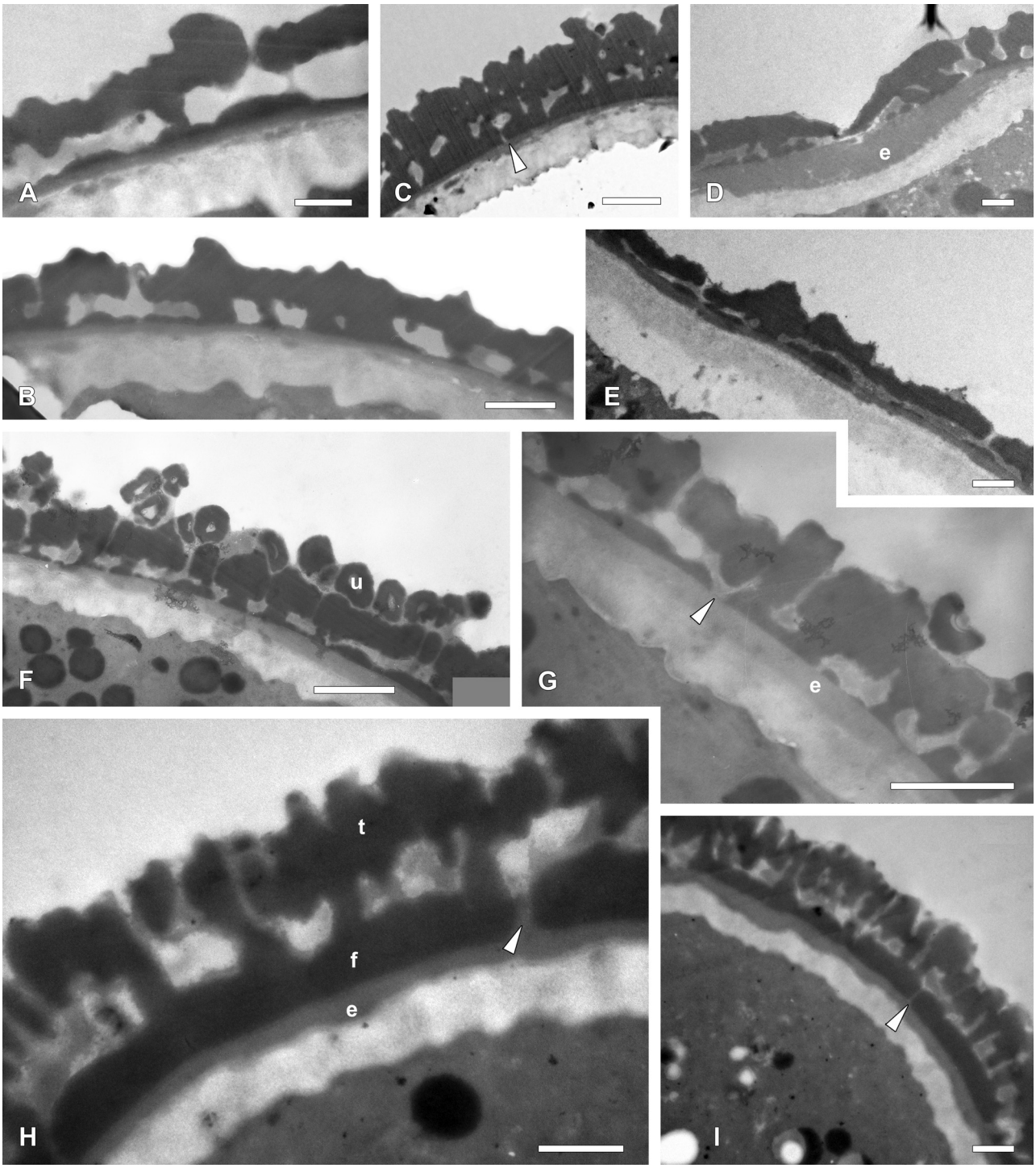

Figure 13. *Quercus* Group Cerris (**A–G**) and Group Cyclobalanopsis (**H, I**), pollen ultrastructure, transmission electron micrographs. **A, B.** *Quercus acutissima*, J. F. Rock 2451 (E); mesocolpium; irregular and markedly discontinuous foot layer. **C.** *Quercus brantii*, H. E. Wright et al. 6323 (E); mesocolpium; foot layer thick, regular, discontinuous. **D, E.** *Quercus ithaburensis* subsp. *macrolepis*, K. H. Rechinger 22299 27/4/1964 (S); **D.** Apertural region with thickened endexine; **E.** Mesocolpium; fragmented, irregular foot layer. **F, G.** *Quercus suber*, H. Lenander 9.5.1928 (S); mesocolpium; fragmented, irregular foot layer; note Ubisch bodies on top of tectum. **H, I.** *Quercus glauca*, H. Togasi 1040 (S); mesocolpium; foot layer thick, regular, disrupted at some places. e, endexine; f, foot layer; t, tectum; u, Ubisch body. Scale bars – 500 nm (A, D, E, H, I), 667 nm (B), 1 μm (C, F, G).

Pollen ornamentation in extinct Fagaceae and fossil members of modern groups

Among extinct members of Fagaceae, the pollen ornamentation of *Quercus* Group Ilex is not common. Oldest fossils ascribed to Fagales with a pollen type strongly reminiscent of Fagaceae are from the early Coniacian of Japan (Takahashi et al. 2008). Pollen from male flowers of *Archaefagacea* clearly falls within the morphological variability of pollen of extant Castaneoideae. From the late Cretaceous of eastern North America, pollen similar to modern Castaneoideae is associated to staminate flowers of *Antiquacupula* (Sims et al. 1998), while *Protofagacea* (Herendeen et al. 1995) has small, reticulate-foveolate pollen. Herendeen et al. (1995) considered this pollen unlike any modern Fagaceae and stated that although resembling extant Fagaceae-Nothofagaceae in many respects, the reticulate to micro-foveolate exine pattern and small size in pollen of *Protofagacea* do not occur in modern Fagaceae and Nothofagaceae. In this context, it is noteworthy that pollen of extant Castaneoideae are the smallest in Fagaceae and are only slightly larger than the (*in situ*) grains of *Protofagacea* and *Archaefagacea*. Both Castaneoideae and *Protofagacea* have tricolporate pollen. Pollen of Castaneoideae are foveolate-perforate; the perforations commonly are much larger in the mesocolpium than along the apertures and at the poles (Praglowski 1984). This is also seen in *Protofagacea*, in which an essentially rugulate exine (see Herendeen et al. 1995, fig. 33) with loosely arranged rugulae appears reticulate when the rugulate pattern is masked by sporopollenin (e.g. fig. 34 in Herendeen et al. 1995). Therefore, pollen of *Protofagacea* may well correspond with pollen of fossil and extant Castaneoideae.

From the Paleocene/Eocene and Eocene/Oligocene of North America, *Castanopsoidea* and *Fagopsis* have castaneoid pollen grains, which closely resemble pollen of Castaneoideae in their pollen wall ultrastructure and sculpture patterns (Manchester & Crane 1983; Crepet et al. 1989a). In contrast, extinct trigonobalanoids from the Oligocene of Texas (Crepet et al. 1989b) have pollen that is similar to the Paleogene European *Eotrigonobalanus* and unlike any modern pollen in Fagaceae (Hofmann 2010; Hofmann et al. 2011; Denk et al. 2012). Extinct trigonobalanoids from younger Neogene sediments of Eurasia (*Trigonobalanopsis*) typically have pollen of the Castaneoideae type (Walther & Zetter 1993; Denk et al. 2012).

From the middle Eocene of western Greenland, dispersed pollen grains virtually identical to modern Group Ilex pollen have recently been discovered (F. Grímsson, unpublished data) and from the early Oligocene of Central Europe, this type of pollen has been reported (Denk et al. 2012).

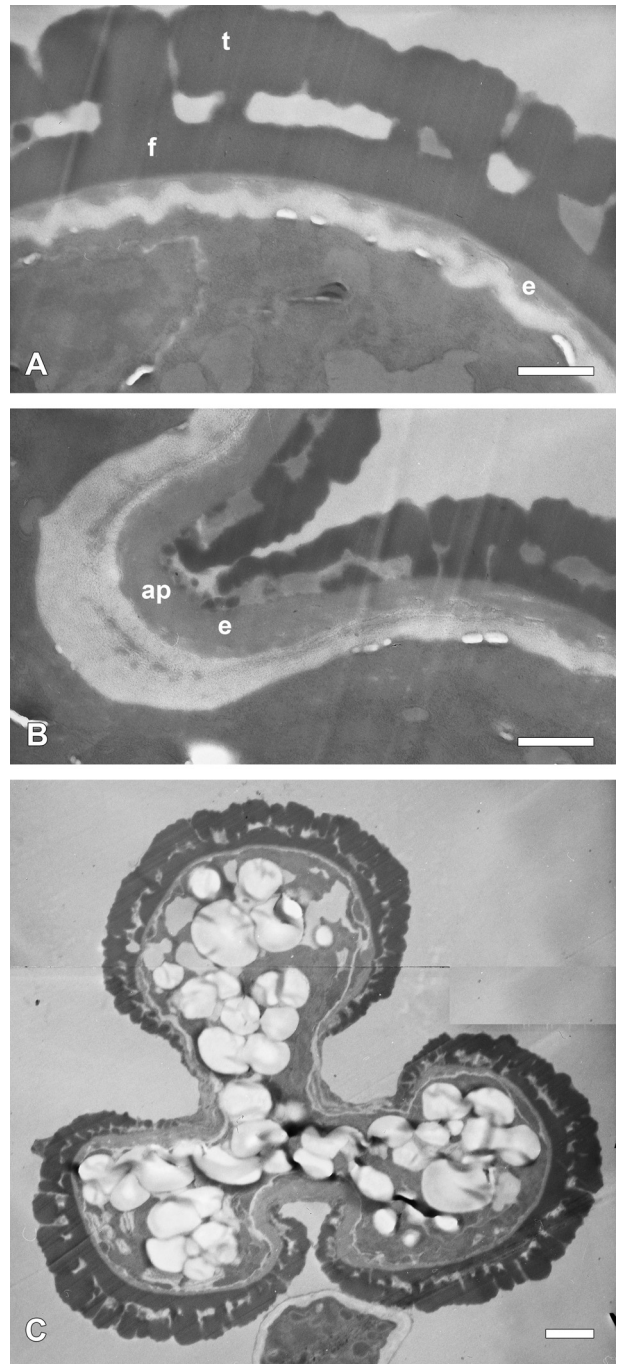

Figure 14. *Quercus* Group Protobalanus, pollen ultrastructure, transmission electron micrographs. **A, B.** *Quercus chrysolepis*, A. D. E. Elmer 4539 (E); **A.** Mesocolpium; continuous, thick, and regular foot layer and fragmented endexine; **B.** Apertural region; thickened endexine and discontinuous foot layer. **C.** *Quercus tomentella*, Blanche 317 (E); equatorial section showing the thick foot layer in the mesocolpium. e, endexine; ap, aperture region; f, foot layer; t, tectum. Scale bars – 667 nm (A, B), 1 μm (C).

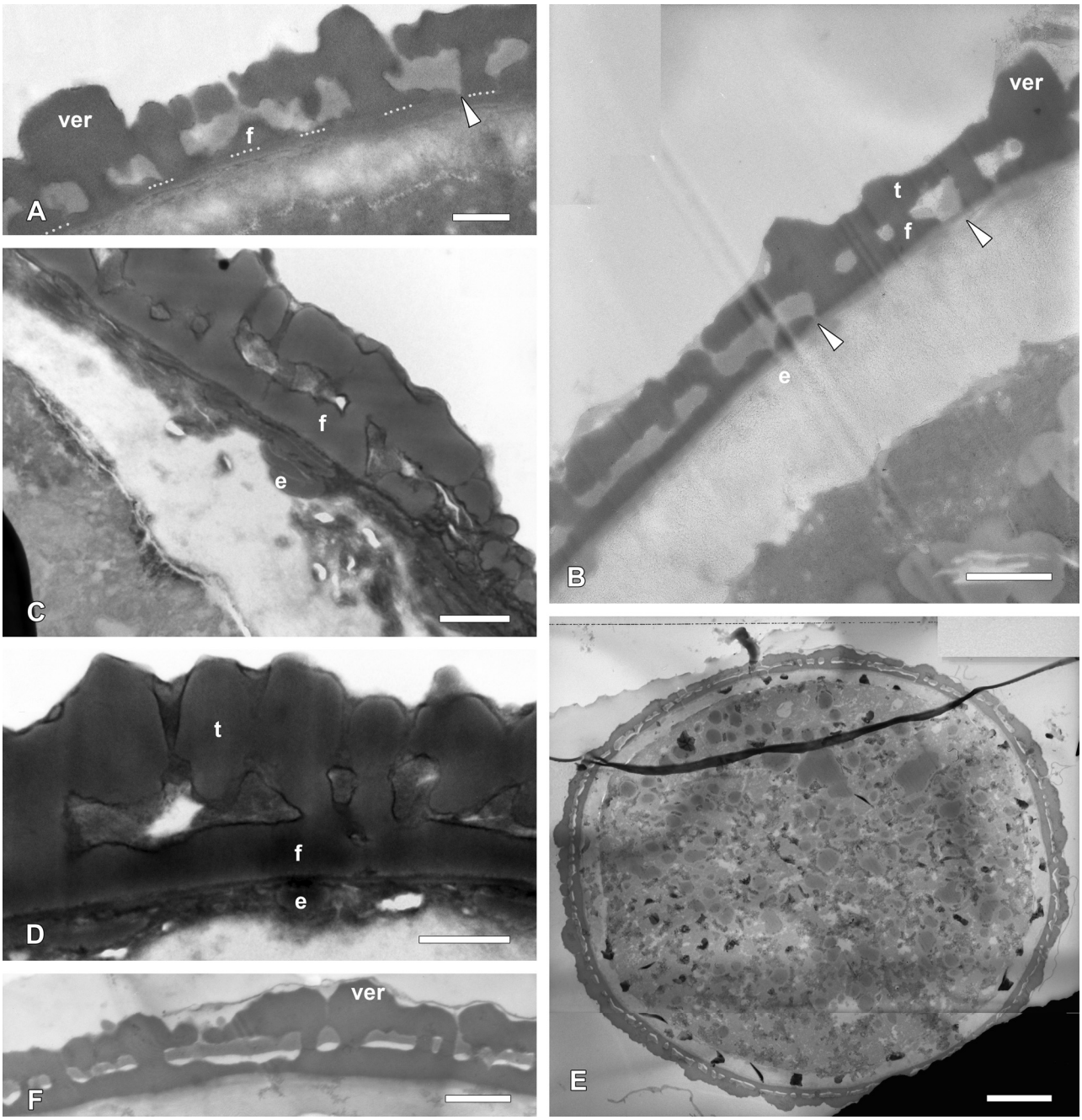

Figure 15. *Quercus* Group Quercus, pollen ultrastructure, transmission electron micrographs. **A.** *Quercus petraea*, Sochi Botanical Garden; mesocolpium; indistinct boundary between endexine and foot layer (indicated with dotted line), discontinuous, irregular foot layer. **B.** *Quercus dentata*; mesocolpium; relatively thin, discontinuous foot layer. **C, D.** *Quercus pyrenaica*, O. Cyren 4/6/1928 (S); **C.** Mesocolpium towards apertural region (bottom right); lamellate endexine with white lines becoming thicker in apertural region; **D.** Mesocolpium; nearly continuous, relatively thick foot layer. **E, F.** *Quercus virginiana*, E. J. Palmer 9199 (S); **E.** Oblique section through pollen grain; **F.** Mesocolpium; discontinuous, irregular foot layer. e, endexine; f, foot layer; t, tectum; ver, verrucae; arrowhead, disruption in foot layer. Scale bars – 500 nm (A, C, D), 667 nm (B, F), 2.5 µm (E).

Possibly coeval with the Greenland record are pollen grains from the Eocene Changchang Basin of south China (Hainan) that show similarities to modern *Quercus* Group Ilex and Group Cyclobalanopsis (Hofmann 2010).

Evolutionary significance of the foot layer

The importance of the foot layer for evolutionary studies in angiosperms has previously been emphasised for modern and extinct genera in the basal eudicot family Platanaceae, in which Cretaceous extinct lineages

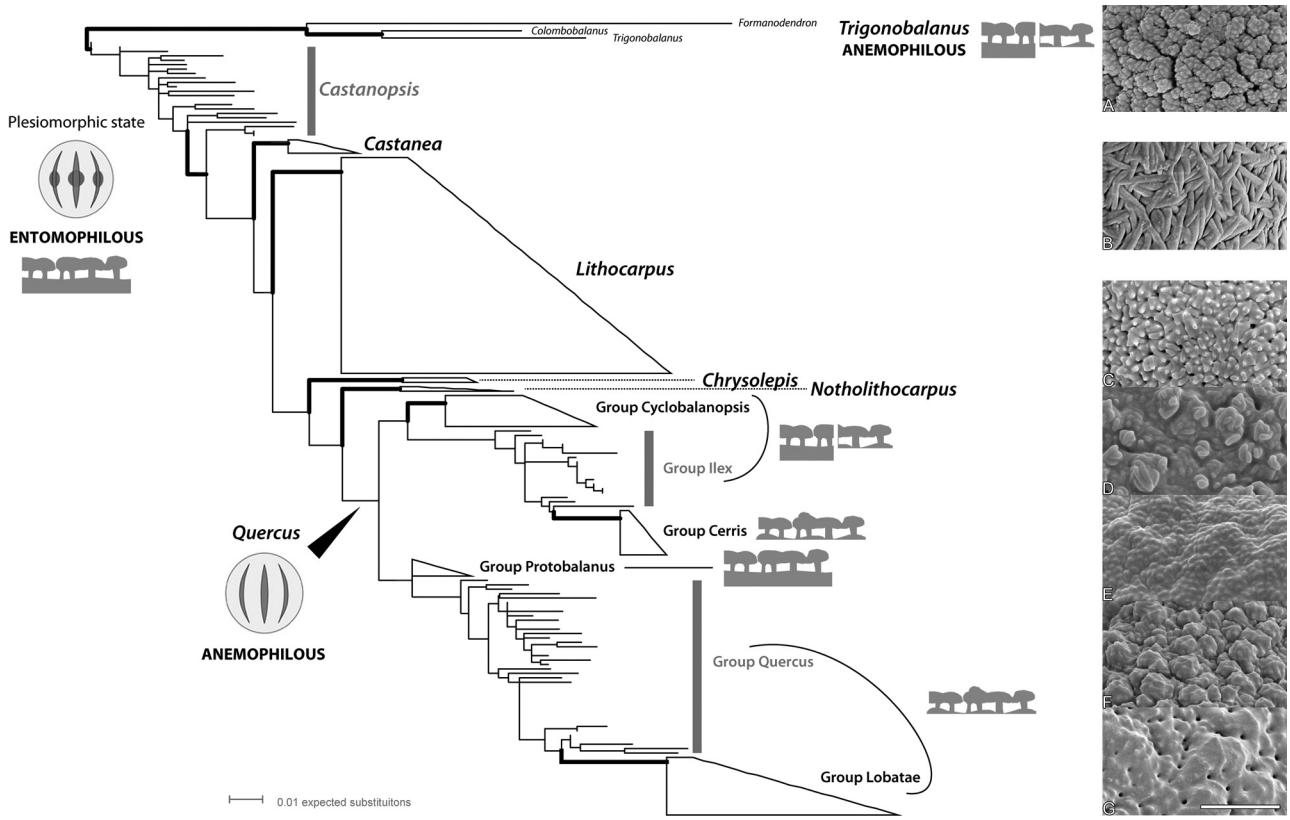

Figure 16. Phylogenetically important pollen characteristics mapped on a molecular tree for Fagaceae. Maximum likelihood tree from Denk and Grimm (2010). Note that all members of Castaneoideae (*Castanopsis*, *Castanea*, *Lithocarpus*, *Chrysolepis*, *Notholithocarpus*) have tricolporate, striate pollen with a homogeneous and thick foot layer. In contrast, pollen in the genus *Quercus* is remarkably diverse. The trigonobalanoids (Trigonobalanus s.l.) have a *Quercus* type pollen ornamentation. **A–G.** Typical pollen ornamentation in Fagaceae; **A.** *Trigonobalanus verticillata* Forman, Chew et al. 2732C (P); **B.** *Castanopsis cuspidata* Schottky, Fumihiro Konta 35502 (S); **C.** *Quercus* Group Cyclobalanopsis, *Q. acuta*, C. Wilford s. n. (S); **D.** *Quercus* Group Cerris, *Q. libani*, R. Wheeler Haines 18-7-1961 (E); **E.** *Quercus* Group Protobalanus, *Q. chrysolepis*, Lewis S. Rose 41236 (S); **F.** *Quercus* Group Quercus, *Q. frainetto*, Wirzbicki 917 (S); **G.** *Quercus* Group Lobatae, *Q. marylandica*, E. J. Palmer 9141 (S). Scale bars – 2 µm.

typically possess a markedly thicker foot layer than modern taxa (Friis et al. 1988; Denk & Tekleva 2006; Tekleva & Denk 2012). Although there seems not to be a causal relationship between a thick foot layer and insect pollination, Cretaceous Platanaceae were most likely entomophilous, while modern types are consistently anemophilous (Friis et al. 1988). In Fagaceae, a relatively thick foot layer, expressed by a high ratio between foot layer thickness and tectum thickness, is consistently found in members of the five genera of Castaneoideae (Praglowski 1984; Table V). Members of Castaneoideae also commonly have a regular and continuous foot layer (type 1 foot layer). In contrast, in *Fagus*, the trigonobalanoids and *Quercus*, the relative thickness of the foot layer is highly variable and the foot layer may be regular or irregular and continuous or discontinuous (Figure 16, Table V). Within *Quercus*, the foot layer in putatively derived groups (Group Cerris, Quercus and Lobatae; Denk & Grimm 2010; Figure 16) is irregular, discontinuous and comparatively thin (type 2 foot layer). Members of Groups Cyclobalanopsis and Ilex either have a regular, continuous and comparatively thick foot layer (type 1) or a type 2 foot layer. From the present data, there appears not to be any regularity (geographical, ecological or taxonomic) in the morphological variability encountered among species of Group Ilex. For example, the highest foot layer to tectum ratios along with the thickest foot layer were encountered in the East Mediterranean *Q. aucheri* and the East Asian *Q. cocciferoides* (Table IV). Intra-group relationships for *Quercus* Group Ilex have been proposed by Menitsky (1984, 2005). The taxa investigated for the present study cover all sections, subsections and cycles recognised by Menitsky (1984, 2005) for subgenus *Heterobalanus* Oersted (corresponding to *Quercus* Group Ilex). However, *Q. baloot* and *Q. cocciferoides*, both in subsection *Ilex* (Loudon) Guerke, and Cycle *Q. ilex* according to Menitsky (1984, 2005), have markedly different sporoderm characteristics (Table IV).

Only Group Protobalanus consistently has a type 1 foot layer and is similar to *Trigonobalanus* and

Table V. Evolutionary significance of pollen ultrastructural features in Fagaceae and *Nothofagus*.

Taxon	Foot layer		Foot layer/tectum
	Regular/irregular	Continuous/discontinuous	
Nothofagus[a]	Regular	Continuous, occasionally interrupted	*c.* 1.0
Fagus[a, b]	Irregular	Continuous	0.2–0.4
Colombobalanus[c]	Irregular	Continuous	0.6–0.8
Trigonobalanus[c]	Regular	Continuous	0.8–1.2
Formanodendron[b, c]	Irregular	Continuous (?)	0.35–0.5
Chrysolepis[d]	Regular	Continuous	*c.* 0.6
***Lithocarpus* s.l.**[b, d]	Regular	Continuous	0.5–0.8
Castanopsis[b, d]	Regular	Continuous	*c.* 0.8–1.3
Castanea[b, d]	Regular/irregular	Continuous/discontinuous	*c.* 1.0 [0.2–0.3][d]
Quercus	Regular/irregular	Continuous/discontinuous	0.2–0.7
Group Quercus	Irregular	Discontinous	0.2–0.4
Group Lobatae	Irregular	Discontinous	0.2–0.3
Group Protobalanus	Regular	Continuous	0.6–0.7
Group Cyclobalanopsis	Regular/irregular	Continuous/discontinuous	0.2–0.6
Group Ilex	Regular/irregular	Continuous/discontinuous	0.2–0.7
Group Cerris	Irregular	Discontinous	0.1–0.3

[a]Praglowski (1982); [b]Wang and Pu (2004); [c]Wang et al. (1998); [d]Praglowski (1984) [two markedly distinct types are found in *Castanea*: foot layer disrupted and in places vestigial, e.g. *C. crenata* and foot layer continuous and thick, e.g. *C. vesca*].
Colour coding: Dark grey = foot layer/tectum ±1, medium grey = 0.5–0.8, light grey = 0.2–0.4.
Note: Crepet and Daghlian (1980) reported an extraordinary thick foot layer for the species *Trigonobalanus verticillata*, resulting in a ratio foot layer to tectum of 1.85 (Crepet & Daghlian 1980, table 2). Based on the TEM section accompanying this study, the foot layer and endexine did not stain differently and hence the border between them is difficult to tell. Measurements and illustrations in Wang et al. (1998) are more useful and clearly show the foot layer/tectum ration being in the range 0.8–1.2.]

Colombobalanus among the trigonobalanoids, and to castanoids. Based on the observations that (I) all except for one species of *Castanea* and *Nothofagus* have a type 1 foot layer and (II) that putatively basal groups in *Quercus* may have a type 1 foot layer, we suggest that this is the ancestral state in Fagaceae, and that the type 2 foot layer of *Fagus* is derived. In a similar way, Manos et al. (2001) argued that although ancestral character state reconstruction suggested insect pollination to be secondarily derived in Fagaceae, the ancestral state for Fagaceae actually is insect pollination and not wind pollination. Our results show that there is a direct link in Fagaceae between foot layer thickness and insect pollination.

Acknowledgements

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Specimens investigated

Group Ilex: *Quercus acrodonta*, G. Forrest 16522 (E); *Q. alnifolia*, P. H. Davis s. n. (E), Foggie 215 (E), A. Foggie s. n. (E); *Q. aquifolioides*, G. Forrest 100026 (E); *Q. aucheri*, P. H. Davis 8007 (ISTO), E. Özhatay 19257 (ISTO); *Q. baloot*, J. E. T. Aitchison 262:128:25:87 (S), S. A. Bowes Lyon 616 (E), W. Koelz 8222 (E); *Q. baronii*, G. H. Yang 58623 (PE), Forestry School of Mei County, Shaanxi Province 05104572 (NF); *Q. coccifera*, F. S. Meyers, J. E. Dinsmore B7123/3123 (S), J. E. Dinsmore 9122, B3122 (S), J. E. Dinsmore 7124C (S), H. Lenander 23-4-1936 (S), G. Een 22-4-1952 (S), P. H. Davis 7959 (ISTO), E. Özhatay 19820 (ISTO), E. Wall 9-5-1933 (S), G. Erdtman 8.5.49 (S), Valbonne 1891 (S), Lorch 1953 (S); *Q. cocciferoides*, Handel-Mazzetti 3632 (E), P. Simeon Ten. 45 (E), P. Simeon Ten. 246 (E); *Q. dilatata*, A. Andersson 4–1915 (E), Duthie 12949 (E); *Q. engleriana*, E. H. Wilson 3633 (E); *Q. franchetii*, A. Henry 9298A (E), T. Smitinand, I. Alsterlund 6781_24519 (E); *Q. gilliana*, G. Forrest 2056 (E), Handel-Mazzetti 1556 (E); *Q. guyavifolia*, D. Chamberlain et al. 004 (E); *Q. ilex*, A. Uggla 20-4-1936 (S), Anisits 30-5-1911 (S), I. Segelberg 19-5-1964 (S), ISTF 17026 (ISTF), H. Lenander 18-5-1933 (S), Torrelodones 1949 (S), Firenze 1873 (S), de Wilde & Dorgelo 2592 (S), J. H. Vijk s. n. (S); *Q. kingiana*, C. C. Hosseus 300_282 (E); *Q. lanata*, F. G. Dickason 8435 (E), Kumrun s.n. (E), H. B. Garrett 1175 (S); *Q. leucotrichophora*, J. D. A. Stainton 207 (E); *Q. lodicosa*, G. Forrest 26611 (E); *Q. longispica*, G. Forrest 21318 (E); *Q. monimotricha*, E. H. Wilson 3626 (E); *Q. pannosa*, ACE 506 (E); *Q. phillyreoides*, Det. O. Schwarz 27-4-1911 (S), M. Takeoka s. n. 26/4/1976 and 4/5/1977 (S), Murata & Koyama s. n. 25/4/1965; *Q. rehderiana*, J. F. Rock 24264 (E), E. E. Maire s. n. (E); *Q. semecarpifolia*, J. E. T. Aitchison 394 (S), Parkinson 4363 (S); *Q. senescens*, D. Chamberlainet al. 159a (E), F. Ducloux 1265 (E); *Q. spathulata* (syn. *Q. dolicholepis* A. Camus), G. Forrest 12397 (E), E. H. Wilson 3625 (E); *Q. spinosa*, E. H. Wilson 1493 (E).

Group Cerris: *Quercus acutissima*, J. F. Rock 2451 (E), S. K. Lau 3173 (E); *Q. brantii*, H. E. Wright et al. 6323 (E), O. Polunin, Khudairi 16 (S); *Q. castaneifolia*, P. Sintensis 1485a (E); *Q. cerris*, Jerusalem Botanical Garden, M. J. E. Coode, B. M. G. Jones 6426 (ISTO), N. Lundqvist 14364 (S); *Q. libani*, R.

Wheeler Haines 18-7-1961 (E); *Q. ithaburensis* subsp. *macrolepis*, K. H. Rechinger 22299 (S); *Q. suber*, Fernanes & Sousa 1590, 5/5/1947, E. Wall 5 (S), H. Lenander 9-5-1928 (S); *Q. variabilis*, E. Dahlström 5-4-1947 (S).

Group Quercus: *Quercus arizonica*, E. Palmer 77 (UC); *Q. brandegei*, H. S. Gentry 4361 (UC); *Q. bumelioides*, R. L. Rodgriquez 346 (UC), A. Smith s. n. 19/02/1940 (UC); *Q. dentata*, E. E. Maire 1475_1913 (E); *Q. douglasii*, P. S. Woolsey & G. M. Tucker 1898 (UC); *Q. fabri*, Kingdon Ward 5094 (E), Xiao Bai Zhong 3393 (E); *Q. faginea*, P. H. Davis 50823 (E); *Q. frainetto*, F. Schultz 917 (S), Pancic 14-4-1985 (S), Wierzbicki 917 (S); *Q. garryana*, J. P. Tracy 6008 (UC), J. P. Tracy 15915 (UC); *Q. griffithii*, F. G. Dickason 7385 (E); *Q. infectoria* subsp. *veneris*, Jerusalem Botanical Garden, *Q. kelloggii*, J. P. Tracy 4895; *Q. macranthera*, ISTO 29084 11/6/1974 (ISTO), A. H. & V. F. Brotherus 775 (S), Kozlowski (S), P. Sintensis 4197 (S); *Q. michauxii*, fresh material, Gainesville USA; *Q. petraea*, Sochi Botanical Garden, E. Rastan 1880 (S), B. Wallnöfer 13566 (E); *Q. pontica*, E. K. Balls 412 (E); *Q. pyrenaica*, O. Cyren 4-6-1928 (S), G. Lidman 1806 (S); *Q. robur*, Jerusalem Botanical Garden, J. Vetter 8-5-1932 (S), T. Symonowiczówna (S); *Q. tuberculata*, A. Carter & J. Reese 4566 (UC), T. T. Marshall. Jr. 118; *Q. virginiana*, E. J. Palmer 9199 (S).

Group Lobatae: *Quercus canbyi*, R. M. King 4543 (UC), *Q. humboldtii*, J. L. Zarucchi, G. McPherson, J. Koldán 7293 (UC), *Q. laevis*, fresh material, Gainesville USA; *Quercus nigra*, fresh material, Gainesville USA.

Group Cyclobalanopsis: *Quercus acuta*, C. Wilford s. n. (S); *Q. augustinii*, F. Ducloux 993 (E), G. Forrest 24033 (E), G. Forrest 26600 (E); *Q. championii*, T. Ying 0243 (E); *Q. fleuryi*, W. T. Tsang 26676 (E); *Q. glauca*, H. Togasi 1040 (S); *Q. helferiana*, J. F. Rock 2662 (E); *Q. longinux*, fresh material, Sochi Botanical Garden

Group Protobalanus: *Quercus chrysolepis*, A. D. E. Elmer 4539 (E), Lewis S. Rose 41236 (S), H. E. & S. T. Parks 24129 (S); *Q. tomentella*, Blanche 317 (E), E. K. Balls 11863 (S); *Q. vaccinifolia*, E. K. Balls 11107 (S), A. A. Heller 6980 (E), A. A. Heller 10938 (E).

References

Beug H-J. 2004. Leitfaden der Pollenbestimmung für Mitteleuropa und angrenzende Gebiete. München: Friedrich Pfeil.

Borsch T, Wilde V. 2000. Pollen variability within species, populations, and individuals, with particular reference to *Nelumbo nucifera*. In: Harley M, Blackmore S, Morton C, eds. Pollen and spores: Morphology and biology, 285–299. Kew: Royal Botanic Gardens.

Camus A. 1936–1938. Les Chênes. Monographie du genre *Quercus*. Tome I. Genre *Quercus*, sous-genre *Cyclobalanopsis*, sous-genre *Euquercus* (sections *Cerris* et *Mesobalanus*). Texte. Paris: Paul Lechevalier.

Camus A. 1938–1939. Les Chênes. Monographie du genre *Quercus*. Tome II. Genre *Quercus*, sous-genre *Euquercus* (sections *Lepidobalanus* et *Macrobalanus*). Texte. Paris: Paul Lechevalier.

Camus A. 1952–1954. Les Chênes. Monographie du genre *Quercus*. Tome III. Genre *Quercus*, sous-genre *Euquercus* (sections *Protobalanus* et *Lobatae*) et genre *Lithocarpus*. Texte. Paris: Paul Lechevalier.

Colombo PM, Lorenzoni FC, Grigoletto F. 1983. Pollen grain morphology supports the taxonomical discrimination of Mediterranean oaks (*Quercus*, Fagaceae). Plant Systematics and Evolution 141: 273–284. doi:10.1007/BF00989007

Crepet WL, Daghlian CP. 1980. Castaneoid inflorescences from the Middle Eocene of Tennessee and the diagnostic value of pollen (at the subfamily level) in the Fagaceae. American Journal of Botany 67: 739–757. doi:10.2307/2442667

Crepet WL, Nixon KC. 1989a. Earliest megafossil evidence of Fagaceae: Phylogenetic and biogeographic implications. American Journal of Botany 76: 842–855. doi:10.2307/2444540

Crepet WL, Nixon KC. 1989b. Extinct transitional Fagaceae from the Oligocene and their phylogenetic implications. American Journal of Botany 76: 1493–1505. doi:10.2307/2444437

Denk T. 2003. A phylogeny of *Fagus* L. (Fagaceae) based on morphological data. Plant Systematics and Evolution 240: 55–81. doi:10.1007/s00606-003-0018-x

Denk T, Grimm GW. 2009. Significance of pollen characteristics for infrageneric classification and phylogeny in *Quercus* (Fagaceae). International Journal of Plant Sciences 170: 926–940. doi:10.1086/600134

Denk T, Grimm GW. 2010. The oaks of western Eurasia: Traditional classifications and evidence from two nuclear markers. Taxon 59: 351–366.

Denk T, Grimm GW, Hemleben V. 2005. Patterns of molecular and morphological differentiation in Fagus (Fagaceae): Phylogenetic implications. American Journal of Botany 92: 1006–1016. doi:10.3732/ajb.92.6.1006

Denk T, Grímsson F, Zetter R. 2012. Fagaceae from the Early Oligocene of Central Europe: Persisting New World and emerging Old World biogeographic links. Review of Palaeobotany and Palynology 169: 7–20. doi:10.1016/j.revpalbo.2011.09.010

Denk T, Tekleva MV. 2006. Comparative pollen morphology and ultrastructure of *Platanus*, implications for phylogeny and evaluation of the fossil record. Grana 45: 195–221. doi:10.1080/00173130600873901

Dettmann ME, Jarzen DM. 1991. Pollen evidence for Late Cretaceous differentiation of Proteaceae in southern polar forests. Canadian Journal of Botany 69: 901–906. doi:10.1139/b91-116

Doyle JA. 2005. Early evolution of angiosperm pollen as inferred from molecular and morphological phylogenetic analyses. Grana 44: 227–251. doi:10.1080/00173130500424557

Doyle JA, Endress PK. 2000. Morphological phylogenetic analysis of basal angiosperms: Comparison and combination with molecular data. International Journal of Plant Sciences 161: SS121–SS153. doi:10.1086/317578

Doyle JA, Endress PK. 2010. Integrating Early Cretaceous fossils into the phylogeny of living angiosperms: Magnoliidae and eudicots. Journal of Systematics and Evolution 48: 1–35. doi:10.1111/j.1759-6831.2009.00058.x

Doyle JA, Le Thomas A. 1997. Significance of palynology for phylogeny of Annonaceae: Experiments with removal of pollen characters. Plant Systematics and Evolution 206: 133–159. doi:10.1007/BF00987945

Endress PK, Doyle JA. 2009. Reconstructing the ancestral angiosperm flower and its initial specializations. American Journal of Botany 96: 22–66. doi:10.3732/ajb.0800047

Flora of China Editorial Committee. 1999. Flora of China, Cycadaceae through Fagacaeae, Vol. 4. St. Louis: Missouri Botanical Garden Press.

Flora of North America Editorial Committee. 1997. Flora of North America North of Mexico, Magnoliophyta: Magnoliidae and Hamamelidae, Vol. 3. New York: Oxford University Press.

Friis EM, Crane PR, Pedersen KR. 1988. Reproductive structures of Cretaceous Platanaceae. Biologiske Skrifter 31: 1–55.

Friis EM, Crane PR, Pedersen KR, Bengtson S, Donoghue PCJ, Grimm GW, Stampanoni M. 2007. Phase-contrast X-ray microtomography links Cretaceous seeds with Gnetales and Bennettitales. Nature 450: 549–552. doi:10.1038/nature06278

Herendeen PS, Crane PR, Drinnan AN. 1995. Fagaceous flowers, fruits, and cupules from the Campanian (Late Cretaceous) of

Central Georgia, USA. International Journal of Plant Sciences 156: 93–116. doi:10.1086/297231

Hesse M, Halbritter H, Zetter R, Weber M, Buchner R, Frosch-Radivo A, Ulrich S. 2009. Pollen terminology – An illustrated handbook. Wien, New York: Springer.

Hofmann C-C. 2010. Microstructure of Fagaceae pollen from Austria (Palaeocene/Eocene boundary) and Hainan Island (?Middle Eocene). Abstracts of the 8th European Palaeobotany-Palynology Conference, Budapest, Hungary, 6–10 July 2010, 119.

Hofmann C-C, Mohamed O, Egger H. 2011. A new terrestrial palynoflora from the Palaeocene/Eocene boundary in the northwestern Tethyan realm (St. Pankraz, Austria). Review of Palaeobotany and Palynology 166: 295–310. doi:10.1016/j.revpalbo.2011.06.003

Huysmans S, Dessein S, Smets E, Robbrecht E. 2003. Pollen morphology of NW European representatives confirms monophyly of Rubieae (Rubiaceae). Review of Palaeobotany and Palynology 127: 219–240. doi:10.1016/S0034-6667(03)00121-0

International Organization for Plant Information (IOPI). 1996–2007. Provisional global plant checklist. International Organization for Plant Information, online database. http://www.bgbm.org/iopi/gpc/default.asp; accessed 19 February 2009.

Kuprianova AL. 1962. Palynological data to the systematics of the Fagales and Urticales. Soviet Reports from the First International Palynological Conference, Tucson, AZ, USA, 23–27 April 1962, 17–25. Moscow: Academy of Sciences.

Makino M, Hayashi R, Takahara H. 2009. Pollen morphology of the genus Quercus by scanning electron microscope. Scientific Reports of Kyoto Prefectural University, Life and Environmental Sciences 61: 53–81.

Manchester SR, Crane PR. 1983. Attached leaves, inflorescences, and fruits of *Fagopsis*, an extinct genus of fagaceous affinity from the Oligocene Florissant flora of Colorado, U.S.A. American Journal of Botany 70: 1147–1164. doi:10.2307/2443285

Manos PS, Cannon CH, Oh S-H. 2008. Phylogenetic relationships and taxonomic status of the paleoendemic Fagaceae of western North America: Recognition of a new genus, *Notholithocarpus*. Madroño 55: 181–190. doi:10.3120/0024-9637-55.3.181

Manos PS, Stanford AM. 2001. The historical biogeography of Fagaceae: Tracking the Tertiary history of temperate and subtropical forests of the Northern Hemisphere. International Journal of Plant Sciences 162: SS77–SS93. doi:10.1086/323280

Manos PS, Zhou Z-K, Cannon CH. 2001. Systematics of Fagaceae: Phylogenetic tests of reproductive trait evolution. International Journal of Plant Sciences 162: 1361–1379. doi:10.1086/322949

Mathews S. 2009. Phylogenetic relationships among seed plants: Persistent questions and the limits of molecular data. American Journal of Botany 96: 228–236. doi:10.3732/ajb.0800178

Menitsky ÛL. 1984. Duby asii. Leningrad: Nauka.

Menitsky ÛL. 2005. Oaks of Asia. Enfield: Science Publishers.

Nagels A, Muasya AM, Huysmans S, Vrijdaghs A, Smets E, Vinckier S. 2009. Palynological diversity and major evolutionary trends in Cyperaceae. Plant Systematics and Evolution 277: 117–142. doi:10.1007/s00606-008-0111-2

Nilsson S, Praglowski J. 1992. Erdtmann's handbook of palynology. 2nd edition. Copenhagen: Munksgaard.

Nixon KC. 1993. Infrageneric classification of *Quercus* (Fagaceae) and typification of sectional names. Annales des Sciences Forestières 50: 25s–34s. doi:10.1051/forest:19930701

Oh S-H, Manos PS. 2008. Molecular phylogenetics and cupule evolution in Fagaceae as inferred from nuclear CRABS CLAW sequences. Taxon 57: 434–451.

Praglowski J. 1982. Fagaceae L., Fagoideae. World Pollen and Spore Flora 11: 1–28.

Praglowski J. 1984. Fagaceae Dumort. Castaneoideae Oerst. World Pollen and Spore Flora 13: 1–21.

Punt W, Hoen PP, Blackmore S, Nilsson S, Le Thomas A. 2007. Glossary of pollen and spore terminology. Review of Palaeobotany and Palynology 143: 1–81. doi:10.1016/j.revpalbo.2006.06.008

Rowley JR. 1996. Exine origin, development and structure in pteridophytes, gymnosperms and angiosperms. In: Jansonius J, McGregor DC, eds. Palynology, principles and applications, 443–462. Dallas: American Association of Stratigraphic Palynologists Foundation.

Rowley JR, Gabarayeva NI. 2004. Microspore development in *Quercus robur* (Fagaceae). Review of Palaeobotany and Palynology 132: 115–132. doi:10.1016/j.revpalbo.2004.05.003

Schwarz O. 1936. Entwurf zu einem naturlichen System der Cupuliferen und der Gattung *Quercus* L. Notizblatt des Königlichen botanischen Gartens und Museums zu Berlin 13 (116): 1–22. doi:10.2307/3994908

Sims HJ, Herendeen PS, Crane PR. 1998. New genus of fossil Fagaceae from the Santonian (Late Cretaceous) of Central Georgia, U.S.A. International Journal of Plant Sciences 159: 391–404. doi:10.1086/297559

Solomon AM. 1983a. Pollen morphology and plant taxonomy of white oaks in eastern North America. American journal of botany 70: 481–494. doi:10.2307/2443159

Solomon AM. 1983b. Pollen morphology and plant taxonomy of red oaks in eastern North America. American Journal of Botany 70: 495–507. doi:10.2307/2443160

Stone DE, Broome CR. 1975. Juglandaceae A. Rich. ex Kunth. World Pollen and Spore Flora 4: 1–35.

Takahashi M, Friis EM, Herendeen PS, Crane PR. 2008. Fossil flowers of Fagales from the Kamikitaba locality (Early Coniacian; Late Cretaceous) of northeastern Japan. International Journal of Plant Sciences 169: 899–907. doi:10.1086/589933

Tekleva MV, Denk T. 2012. Sporoderm ultrastructure of *Platanus quedlinburgensis* Pacltová emend. Tschan, Denk & von Balthazar from the Late Cretaceous of Germany. Acta Palaeobotanica 52: 177–191.

Van Benthem F, Clarke GCS, Punt W. 1984. The northwest European pollen flora, 33. Fagaceae. Review of Palaeobotany and Palynology 42: 87–110. doi:10.1016/0034-6667(84)90063-0

Walker JW, Doyle JA. 1975. The bases of angiosperm phylogeny: Palynology. Annals of the Missouri Botanical Garden 62: 664–723. doi:10.2307/2395271

Walther H, Zetter R. 1993. Zur Entwicklung der paläogenen Fagaceae Mitteleuropas. Palaeontographica B 230: 183–194.

Wang H, He H-J, Chen J-Q, Lu L. 2010. Palynological data on Illiciaceae and Schisandraceae confirm phylogenetic relationships within these two basally-branching angiosperm families. Flora - Morphology, Distribution, Functional Ecology of Plants 205: 221–228. doi:10.1016/j.flora.2009.02.004

Wang P, Pu F. 2004. Pollen morphology and biogeography of Fagaceae. Guangzhou: Guangdong Science and Technology Press.

Wang P, Pu F, Zheng Z-H. 1998. Palynological evidence for taxonomy of *Trigonobalanus* (Fagaceae). Acta Phytotaxonomica Sinica 36: 238–241.

Wortley AH, Funk VA, Robinson H, Skvarla JJ, Blackmore S. 2007. A search for pollen morphological synapomorphies to classify rogue genera in Compositae (Asteraceae). Review of Palaeobotany And Palynology 146: 169–181. doi:10.1016/j.revpalbo.2007.03.003

Wortley AH, Funk VA, Skvarla JJ. 2008. Pollen and the evolution of Arctotideae (Compositae). The Botanical Review 74: 438–466. doi:10.1007/s12229-008-9014-8