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EVOLUTION OF ANGIOSPERM POLLEN: 4. BASAL EUDICOTS¹

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ABSTRACT

In this paper, the fourth in a series documenting palynological characters across angiosperms using a contemporary phylogenetic framework, we deal with the basal eudicots, a group which includes Buxales, Proteales, Ranunculales, Sabiaceae, and Trochodendrales. Using available molecular sequences of *matK* and *rbcL* from previous studies, we reconstructed a maximum likelihood tree for a total of 196 genera (including nine outgroups), representing 13 families and all four orders of basal eudicots. Across the 196 genera, 20 pollen characters were documented from prior publications and new observations. These were coded using two strategies and optimized onto the reconstructed phylogenetic tree using Fitch parsimony, maximum likelihood, and hierarchical Bayesian inference to infer ancestral states. Pollen samples of 24 species from 24 genera in eight families were imaged under LM and SEM to illustrate the diversity of basal eudicot pollen. In addition, we tested for correlated evolution between plant growth form and pollen shape class, and between anemophily and pollen aperture number, using maximum likelihood and Markov chain Monte Carlo analyses. Basal eudicot pollen showed high morphological diversity, especially in characters including tectum sculpture, aperture number, and ectoaperture shape. Depending upon the method of reconstruction, 14 to 18 plesiomorphic palynological states were unequivocally inferred, and a total number of 357 character state transitions were found at or above tribal level. These provide palynological support for at least 58 of the clades discovered by molecular phylogenetic estimation. For example, using hierarchical Bayesian inference with comprehensive data coding, 222 state changes were inferred. Pollen size, tectum sculpture, and pollen shape class changed the most frequently among the 20 studied characters. The most concentrated character state changes in basal eudicot pollen are estimated to have occurred around the Barremian to Albian stages of the Early Cretaceous. Tests of correlated evolution suggest that the herbaceous growth form is significantly associated with spheroidal pollen shape and the arborescent growth form with oblate pollen shape. However, no significant correlations were found between anemophily and aperture number. Patterns of evolutionary change in pollen size and tectum sculpture, and their adaptive functions, are discussed. Based on previous evidence and our data, pollination syndrome is the most likely factor associated with the high frequency of state changes in these two characters.

Key words: Basal eudicots, character evolution, correlation, pollen morphology, Proteales, Ranunculales, systematic significance.

Eudicots, or Eudicotyledoneae (Cantino et al., 2007), are considered a strongly supported monophyletic group in the Angiosperm Phylogeny Group (APG) system (APG III, 2009; APG IV, 2016), including over 200,000 species, comprising about 75% of extant angiosperm species and encompassing enormous morphological, biochemical, anatomical, and ecological diversity (Drinnan et al., 1994; Judd & Olmstead, 2004; Furness et al., 2007; Moore et al., 2010). Eudicots were first recognized and defined

morphologically as the tricolpate clade based on the significant synapomorphy of tricolpate pollen apertures or types derived from this (Donoghue & Doyle, 1989; Doyle & Hotton, 1991). Within eudicots, a few lineages, including Ranunculales, Sabiaceae, Proteales, Trochodendrales, and Buxales, have been revealed by phylogenetic studies to represent the earliest branches and are termed “basal eudicots” or “first/early diverging eudicots” (Chase et al., 1993; Savolainen et al., 2000a, 2000b; Soltis et al., 2000,

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2003, 2007, 2011; Jansen et al., 2007; APG III, 2009; Moore et al., 2010; APG IV, 2016).

Much attention and effort have been devoted to understanding the origin, diversity, evolution, and phylogeny of basal eudicots (Endress & Igersheim, 1999; Hoot et al., 1999; S. Kim et al., 2004; Anderson et al., 2005; Worberg et al., 2007; Hilu et al., 2008; Forest & Chase, 2009; Barniske et al., 2012; Sun et al., 2016). Since the earliest broad-scale molecular phylogenetic study of Chase et al. (1993), the delimitation of basal eudicots has changed significantly. In the APG III classification (2009), and as defined here, basal eudicots comprise Buxales, Proteales, Ranunculales, Sabiaceae (note that in the more recent APG IV classification [2016], published since our work was conducted, Sabiaceae has been included under Proteales), and Trochodendrales, together including about 293 genera, 14 families (note that in APG IV [2016], published since our work was conducted, Haptanthaceae is included in Buxaceae, giving only 13 families in basal eudicots), and ca. 6000 species, accounting for ca. 2% of angiosperms. Phylogenetic relationships among lineages within basal eudicots have been gradually elucidated: Ranunculales are now considered to have diverged first, followed by the sister clades of Sabiaceae–Proteales, then Trochodendrales and Buxales, successively, with Buxales sister to core eudicots (Moore et al., 2010; Soltis et al., 2011; Barniske et al., 2012; Magallón et al., 2015). However, alternative topologies have been proposed, particularly regarding the position of Sabiaceae in relation to Proteales (as sister clades or successive branches), and of Buxales or Trochodendrales as sister to the core eudicots (Soltis et al., 2003, 2007; S. Kim et al., 2004; Worberg et al., 2007; Hilu et al., 2008; Sun et al., 2016).

At lower taxonomic levels, phylogenetic relationships within Ranunculales remain partly unresolved (W. Wang et al., 2009). Ranunculales, the largest order of basal eudicots, containing two thirds of the species (4445) in 199 genera within seven families, has been strongly supported as monophyletic (W. Wang et al., 2009; Soltis et al., 2011; Barniske et al., 2012). Within Ranunculales, the phylogenetic positions of the two earlier-diverging families Papaveraceae and Eupteleaceae are controversial, although the remaining families, i.e., Berberidaceae, Circaeasteraceae, Lardizabalaceae, Menispermaceae, and Ranunculaceae, form a well-supported core clade (Hilu et al., 2003; S. Kim et al., 2004; Soltis et al., 2007, 2011; Worberg et al., 2007; W. Wang et al., 2009; Barniske et al., 2012; Sun et al., 2016). Proteaceae is the largest family within Proteales,

comprising 80 genera and over 1700 species of mostly woody angiosperms (Hoot & Douglas, 1998; Sauquet & Cantrill, 2007), within which relationships are still not completely understood (e.g., Johnson & Briggs, 1963, 1975; Douglas, 1995; Weston & Barker, 2006; Barker et al., 2007; Sauquet et al., 2009a, 2009b).

The taxonomic and systematic value of pollen morphology has long been recognized. Palynology is unique within plant morphology: no other discipline can provide so great an amount of character state information from so little material with such relative ease (Walker & Doyle, 1975). The importance of pollen morphological variation in basal eudicots has been shown in numerous studies, for example, of Berberidaceae (Nowicke & Skvarla, 1981; Chang & Wang, 1983), Buxaceae (Brückner, 1993), Menispermaceae (Ferguson, 1975; Harley & Ferguson, 1982; Harley, 1985), Proteaceae (Dettmann, 1998; Milne & Martin, 1998; Sauquet & Cantrill, 2007), and Ranunculales (Pragłowski, 1975; Nowicke & Skvarla, 1982; Blackmore et al., 1995). Indeed, Proteaceae was the first family in which pollen morphology was considered in relation to plant classification, by Robert Brown (1811; reviewed in Blackmore, 2007). The close phylogenetic relationships of *Berberis* L. and *Mahonia* Nutt. in Berberidaceae (Meacham, 1980; Wang et al., 2007a) and of *Faurea* Harv. and *Protea* L. in Proteaceae (Barker et al., 2002; Sauquet et al., 2009b) are well supported by spiraperturate pollen (Nowicke & Skvarla, 1981; Chang & Wang, 1983) and slit-shaped [sic] apertures (Sauquet & Cantrill, 2007), respectively. Similarly, the monophyly of clade Leucadendrinae (Proteaceae) in the phylogeny of Weston and Barker (2006) is supported by small pollen size, frequently ellipsoid-lolate apertures, and an apertural region marked by a thin and foliated [sic] foot layer (Sauquet & Cantrill, 2007).

The adaptive significance of pollen characters has also been of interest for decades, with discussions including the functional role of pollen apertures in the exchange of intine-bound recognition substances and in the regulation of harmomegathy (Heslop-Harrison, 1976; Blackmore & Crane, 1998; Katifori et al., 2010). Furness et al. (2007) suggested that endoapertures might have originated in eudicots for their harmomegathic role. Pollen ectoaperture shape has been suggested to be associated with pollination syndrome in both monocots (Luo et al., 2015) and the nitrogen-fixing clade (including Cucurbitales, Fabales, Fagales, and Rosales) (He et al., in prep.). Tectum sculpture has also been found to correlate

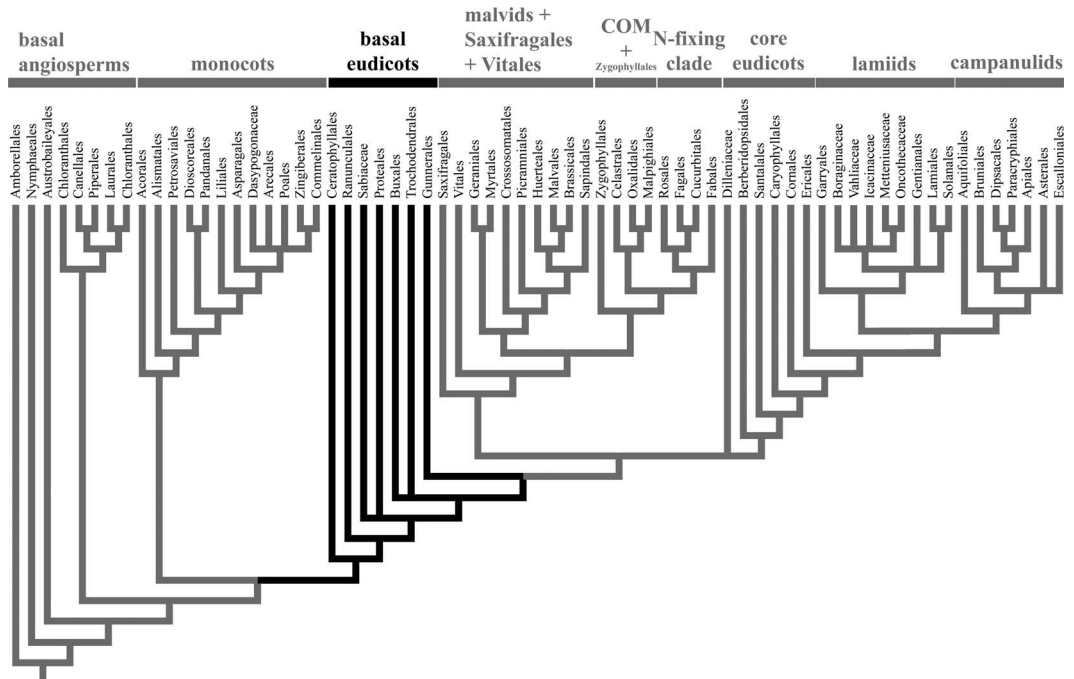


Figure 1. Phylogenetic framework for angiosperms, divided into groups following Wortley et al. (2015). The highlighted taxa indicate the basal eudicots, studied in this paper. COM, Celastrales, Oxalidales, and Malpighiales; N-fixing, nitrogen-fixing.

with pollination syndrome in the nitrogen-fixing clade (He et al., in prep.).

The basal eudicots are a critical group of intermediate origins, having possible sister-group relationships to Ceratophyllales, monocots, or magnoliids, and forming a paraphyletic grade at the base of the core eudicots (APG III, 2009; Burleigh et al., 2009; Moore et al., 2010; Soltis et al., 2011; Magallón et al., 2015). The time that the eudicots originated is inferred to be a significant turning point in the evolution of angiosperm pollen characters such as aperture number, aperture position, basic pollen grain shape, and symmetry, as well as the presence of endexine and endoapertures (Blackmore et al., 1995; Furness & Rudall, 2004; Furness et al., 2007; Wortley et al., 2015). In addition, basal eudicots are suggested to have experienced a rapid diversification during the latter part of the Early Cretaceous (Anderson et al., 2005; Sun et al., 2011; W. Wang et al., 2014), and all lineages of the grade are estimated to have emerged by the end of the Early Cretaceous (Anderson et al., 2005; Magallón et al., 2015; Tank et al., 2015). Integrating pollen character evolution with a molecular phylogenetic framework may provide some clues to understanding the origin and subsequent explosive radiation of the eudicots.

Recent, increasingly robust phylogenies of basal eudicots provide a foundation for investigating the

distribution of pollen features and the potential underlying patterns in their transformations. Utilizing Fitch parsimony (FP), maximum likelihood (ML), and hierarchical Bayesian (HB) inference, we reconstruct the possible evolution of 20 basal eudicot pollen characters. Moving on from the basal angiosperms examined in Lu et al. (2015; see Fig. 1), here we aim to investigate the pollen diversity of the basal eudicots; to infer pollen character state transitions, diagnostic states, and to assess their significance in systematics; to infer the possible plesiomorphic pollen morphology of basal eudicots; and to explore possible evolutionary patterns in selected pollen characters in the context of a molecular phylogeny. We also conduct tests of correlated evolution for specific hypotheses between pollen characters and ecological traits.

MATERIALS AND METHODS

OBSERVATION OF POLLEN MORPHOLOGY

Pollen grains from 24 species representing 24 genera, eight families, and four orders of basal eudicots were observed using LM and SEM to exemplify the pollen morphological diversity of this group and to supplement the available palynological information in the main analysis with new data. Pollen samples were collected from the living

collections of Kunming Botanical Garden and herbarium collections of the Southwest China Germplasm Bank of Wild Species and the Kunming Institute of Botany (KUN); Royal Botanic Garden Edinburgh (E); Australian National Herbarium (CANB); Australian National Botanic Garden Herbarium (CBG); California Academy of Sciences (CAS); and Missouri Botanical Garden (MO) (for voucher information see Appendix 1). Pollen grains were prepared for observation using the acetolysis method of Erdtman (1960). For some samples, where we failed to get satisfactory images using this method, unacetolyzed pollen grains were mounted directly onto specimen stubs and sputter-coated with gold-palladium (Behnke, 1982; Sauquet & Cantrill, 2007) (images obtained without acetolysis are shown in boldface in Appendix 1). Pollen grains were observed under a Hitachi S-4800 SEM (Hitachi, Tokyo, Japan) at 10.0 KV (KUN).

RECONSTRUCTION OF PHYLOGENETIC RELATIONSHIPS

A robust resolution of phylogenetic relationships with adequate taxon sampling is a prerequisite for secure reconstruction of character evolution (Endress & Doyle, 2009; Soltis et al., 2013). Although many previous studies have focused on basal eudicot phylogeny (S. Kim et al., 2004; Soltis et al., 2007, 2011; Worberg et al., 2007; Jacques & Bertolino, 2008; Sauquet et al., 2009a; W. Wang et al., 2009), none have performed the wide-ranging taxon sampling across the whole group necessary to fulfill our study objectives. We therefore reconstructed an ML tree using *matK* and *trnK* data from a combination of the three latest, relatively well-resolved phylogenetic frameworks (Sauquet et al., 2009a; W. Wang et al., 2009; Soltis et al., 2011) (sequences downloaded with permission from the National Center for Biotechnology Information), to cover a broad range of taxa both phylogenetically and palynologically. Our analysis comprises 196 generic terminals, representing all orders and families of this group (except Haptanthaceae). Six genera (*Ceratophyllum* L. [Ceratophyllales], and five monocots: *Acorus* L. [Acoraceae], *Tofieldia* Huds. [Tofieldiaceae], *Orotium* L. [Araceae], *Alisma* L. [Alismataceae], and *Potamogeton* L. [Potamogetonaceae]), inferred to be the closest groups to the basal eudicots in Soltis et al. (2011), were included as outgroups, and three genera (*Gunnera* L., *Myrothamnus* Welw., and *Dillenia* L.) of core eudicots were also included to represent the nested higher taxa of angiosperms. Phylogenetic analysis was conducted with RAxML (Stamatakis, 2006). A bootstrap analysis (1000 replicates) was performed simultaneously with the ML analysis

(option “-f a”), under the model GTRGAMMA. The resulting topology differed from the latest published phylogenies (i.e., Sauquet et al., 2009a; W. Wang et al., 2009; Moore et al., 2010; Soltis et al., 2011; Barniske et al., 2012; Magallón et al., 2015) for this group regarding the positions of Sabiaceae, Trochodendraceae, and some lower taxa in Menispermaceae, Proteaceae, and Ranunculaceae. Since the support values for these conflicting relationships were lower in our phylogeny than in existing studies using greater numbers of loci (Wang et al., 2007b; Sauquet et al., 2009a; W. Wang et al., 2009; Moore et al., 2010; Soltis et al., 2011; Barniske et al., 2012; Magallón et al., 2015), we repeated our analysis, implementing constraints to ensure a match to these studies. Names of orders and families follow APG III (2009) and APG IV (2016); those of lower taxa (tribes, subtribes, and genera) are consistent with the three source studies.

POLLEN CHARACTERS AND CODING STRATEGY

Pollen morphological data for all 196 genera in the phylogenetic tree were compiled from previous publications (LM, SEM, and transmission electron microscopy [TEM] data; see Appendix 2), online databases (PalDat: A Palynological Database [<https://www.paldat.org/>]) and the Australasian Pollen and Spore Atlas [<http://apsa.anu.edu.au/>]), and new observations. Twenty pollen characters were selected and coded for analysis: dispersal unit, polarity, symmetry (in polar view), basic shape, shape class, outline in polar view (amb), size (diameter of longest axis), aperture number, aperture position, aperture structure, ectoaperture shape, endoaperture shape, operculum presence/absence, supracteal element presence/absence, supracteal element shape, tectum presence/absence, tectum sculpture, infratectum structure, foot layer presence/absence, and endexine presence/absence (see Table 1). These characters were chosen on the basis of their variability and taxonomic significance at interordinal, interfamilial, and intrafamilial levels as previously identified (Erdtman, 1952; Walker & Doyle, 1975; Nowicke & Skvarla, 1979, 1981, 1982; Ferguson, 1985; Blackmore et al., 1995; Wortley et al., 2015), and for good coverage of data across taxa. Character state delimitation followed Wortley et al. (2015), themselves following previous palynological researchers (including Erdtman, 1952; Fægri, 1956; Walker & Doyle, 1975; Fægri & Iversen, 1989; Punt et al., 2007). Within genera, pollen data from multiple species were recorded where possible, to provide best coverage of character state variation; thus, polymorphic states were observed for some characters in

Table 1. The 20 pollen morphological characters and two ecological traits and their states defined in the two matrices in this study. C, comprehensive method of coding; D, democratic method of coding.

Character	Character states
1. Dispersal unit	C, D: 0, monad; 1, dyad; 2, tetrad.
2. Polarity	C, D: 0, apolar; 1, heteropolar; 2, isopolar (including subisopolar).
3. Symmetry (in polar view)	C, D: 0, bilateral; 1, radial; 2, asymmetrical.
4. Basic shape	C, D: 0, boat-shaped; 1, globose.
5. Shape class	C, D: 0, oblate (P/E: 0.5–0.75); 1, suboblate (P/E: 0.75–0.88); 2, spheroidal (P/E: 0.88–1.14); 3, subprolate (P/E: 1.14–1.33); 4, prolate (P/E: 1.33–2).
6. Outline in polar view (amb)	C, D: 0, circular; 1, polygonal (including concave or convex); 2, elliptic.
7. Size (diameter of longest axis)	C: 0, small (10–24 µm); 1, medium (25–49 µm); 2, large (50–99 µm); 3, very large (100–199 µm). D: 0, small (10–24 µm); 1, medium (25–49 µm); 2, large (50–99 µm).
8. Aperture number	C, D: 0, zero; 1, one; 2, two; 3, three; 4, four to six; 5, more than six.
9. Aperture position	C, D: 0, equatorial; 1, global; 2, distal polar; 3, inapplicable.
10. Aperture structure	C, D: 0, simple; 1, compound (including colpoidate and colpoidiate); 2, inapplicable.
11. Ectoaperture shape	C: 0, colpate; 1, porate; 2, spiral; 3, syncolpate; 4, zonate; 5, inapplicable. D: 0, colpate; 1, porate; 2, spiral; 3, syncolpate; 4, inapplicable.
12. Endoaperture shape	C, D: 0, circular; 1, lalongate; 2, lolongate; 3, inapplicable.
13. Operculum presence/absence	C, D: 0, absent; 1, present; 2, inapplicable.
14. Supratectal element presence/absence	C, D: 0, absent; 1, present.
15. Supratectal element shape	C: 0, gemmate (including granulate and scabrate); 1, echinate; 2, pilate (including baculate and with rodlets); 3, rugulate; 4, striate; 5, verrucate; 6, angular; 7, inapplicable. D: 0, gemmate (including granulate and scabrate); 1, echinate; 2, pilate (including baculate and with rodlets); 3, verrucate; 4, angular; 5, inapplicable.
16. Tectum presence/absence	C, D: 0, absent; 1, present.
17. Tectum sculpture	C: 0, areolate (including verruco-areolate); 1, fossulate; 2, perforate (including foveolate); 3, imperforate; 4, striate; 5, striato-reticulate; 6, reticulate (including microreticulate and semitectate); 7, rugulate; 8, crotonoid; 9, inapplicable. D: 0, areolate (including verruco-areolate); 1, perforate/foveolate; 2, imperforate; 3, striate; 4, striato-reticulate; 5, reticulate (including microreticulate and semitectate); 6, rugulate; 7, crotonoid; 8, inapplicable.
18. Infratectum structure	C, D: 0, columellate (including clavate); 1, granulate; 2, protocolumellate (sensu Nowicke & Skvarla, 1981); 3, granulate/columellate (granulate predominant); 4, columellate/granulate (columellate predominant).
19. Foot layer presence/absence	C, D: 0, absent; 1, present.
20. Endexine presence/absence	C, D: 0, absent; 1, present.
21. Pollination syndrome	C: 0, insects (entomophily); 1, mammals (chiropterophily and therophily); 2, birds (ornithophily); 3, wind (anemophily); 4, water (hydrophily); 5, self-pollination.
22. Plant growth form	C: 0, herbaceous; 1, shrub; 2, vine; 3, arborescent.

P/E, polar and equatorial axes.

some taxa. Two different coding strategies were employed, i.e., comprehensive (including all polymorphic states of each character appearing within a terminal taxon) and democratic (selecting the most common state for each character in each terminal taxon), following Wortley et al. (2015) and Lu et al. (2015). Inapplicable data (no state possible) were coded as an extra state, to avoid confusion with missing data (any state possible), which may produce unexpected and undesirable results in computer-based phylogenetic analyses (Maddison, 1993; Zar-

agüeta-Bagils & Bourdon, 2007). See Appendices 3 and 4 for data matrices.

ANALYSES OF CHARACTER EVOLUTION

The 20 pollen characters for 196 genera were optimized on the reconstructed phylogenetic tree using three methods of inference: FP, which specifies character states as unordered and equally weighted and minimizes the number of character state changes (Fitch, 1971; Cunningham et al., 1998); ML, which

uses an explicit model of character evolution to estimate the probabilities of all possible character state reconstructions at every node on the tree (Cunningham et al., 1998); and HB Markov chain Monte Carlo (MCMC) modeling, calculating the relative probability for each possible character state at each node on a set of trees sampled from a distribution (Huelsenbeck & Bollback, 2001). Data coded using the comprehensive strategy were reconstructed using FP and HB inference (hereafter abbreviated as CFP and CHB, respectively; ML was not usable for data with polymorphic states). Data coded using the democratic strategy were reconstructed under all three methods (hereafter abbreviated as DFP, DML, and DHB). FP and ML (using the Mk-1 model) optimizations were conducted with Mesquite 2.75 (Maddison & Maddison, 2011); HB inference was implemented using BayesTraits 1.0 (available from <<http://www.evolution.rdg.ac.uk/>>). For CHB and DHB analyses, parameters were set as follows: rate variations ranging from four to 74 across the 20 characters; sampling frequency of 300 to 800 generations; and a burn-in of 10,000 generations, for a total of 5,000,000 generations. For ML and HB analyses, the state with the highest likelihood or probability at each node was taken to be the state inferred for this node.

TESTS OF CORRELATED EVOLUTION

We conducted tests of correlation between pollen features and pollination syndrome and plant growth form. Pollination syndrome, which has previously been suggested to correlate with pollen features (Muller, 1979; Linder, 2000; Luo et al., 2015), and plant growth form were coded using the comprehensive method (see Appendix 3) and optimized on the phylogeny using both FP and HB methods. A visual inspection of the results of these analyses suggested a correlation between anemophily and aperture number (when aperture number is greater than or equal to four), and between plant growth form and pollen shape class (arborescent forms with oblate pollen; herbaceous forms with spheroidal pollen). No correlations between other pollen characters and ecological traits were suggested based on this preliminary inspection. To investigate whether these observed correlations were significant, ML tests were conducted under a discrete model using BayesTraits (Pagel, 1994). Each polymorphic trait was recoded as a binary character (i.e., pollen shape class was recoded as spheroidal or non-spheroidal and oblate or non-oblate; plant growth form was recoded as herbaceous or non-herbaceous and arborescent or non-arborescent; pollination syndrome was recoded

as anemophilous or non-anemophilous; aperture number was recoded as less than four or greater than or equal to four) to meet the requirements of the Bayes discrete test, which only works for binary characters at present (Pagel, 2006). We allowed BayesTraits to fit two models to the phylogeny for each pair of traits, i.e., an independent model (I) and dependent model (D), implying, respectively, that one trait varies independently from, or dependently on, the other. BayesTraits was then used to calculate a likelihood score for both of these two models, representing the fitness of the data to the models. The likelihood ratio (LR) was determined by the equation $LR = 2[\text{likelihood (D)} - \text{likelihood (I)}]$. When L (D) is significantly greater than likelihood (I), we infer a correlation between the two traits, which can be tested against a χ^2 distribution with four degrees of freedom (following Pagel, 1994).

In addition, MCMC analysis was conducted for both dependent and independent models using a reversible-jump hyperprior model and an exponential prior distribution to test the correlation. Rate variations were set from nine to 14 with a sampling frequency of 300 and a burn-in of 10,000 generations, for a total of 5,000,000 generations. The log Bayes factor (LBF) value was determined with a similar equation: $LBF = 2[\text{harmonic mean (D)} - \text{harmonic mean (I)}]$. An LBF value greater than two indicates positive correlation; a value greater than five indicates a strong correlation; and a value greater than 10 suggests a very strong correlation (Pagel & Meade, 2006).

Each pair of binary traits can yield four different combinations of states under the dependent model (Fig. 2; for detailed interpretation of this figure see Pagel & Meade, 2006). Transition rates for correlated characters were estimated following Pagel and Meade (2006). Specific hypotheses of contingent evolution were tested by restricting two of the transition rates to equal (e.g., for herbaceous plant growth form and spheroidal pollen shape class, $q_{12} = q_{34}$ indicates that the rate of evolution of the shape class from spheroidal to non-spheroidal is constant, no matter what growth form is observed). This 7-parameter restricted model was then compared to a full dependent model using the likelihood ratio test $LR = 2[\text{likelihood (D)} - \text{likelihood (} q_{12} = q_{34}\text{)}]$ with one degree of freedom, following Pagel (1994). A significant LR suggests that the parameters are significantly different from each other, indicating (for instance) that the state of the growth form influences the evolutionary direction of the shape class character (Pagel, 1994).

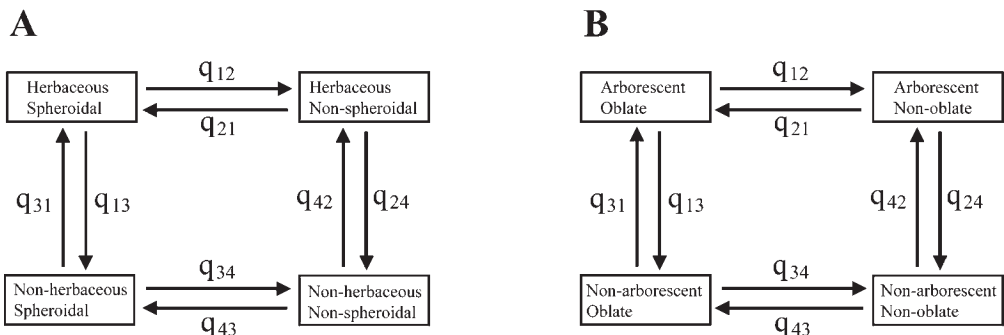


Figure 2. The eight possible transitions for four combinations of two pairs of binary traits. —A. Herbaceous growth form and spheroidal shape. —B. Arborescent growth form and oblate shape.

Since the transition rates q_{24} and q_{42} for the pair comprising herbaceous growth form and spheroidal pollen shape were both zero, we also restricted each of the others to zero (e.g., $q_{24} = 0$ indicates that the plant growth form will not change from herbaceous to non-herbaceous when the pollen shape class is non-spheroidal), recalculated the LR under the dependent model, and compared the 7-parameter restricted model to the unrestricted dependent model using a χ^2 test with one degree of freedom to test whether our hypotheses are correct. A significant LR would indicate that the transition rate is significantly different from zero (Friedman & Barrett, 2008), and the assumption is invalid.

RESULTS

OBSERVATION OF POLLEN MORPHOLOGY

Corydalis linarioides Maxim. (Ranunculales, Papaveraceae, Fig. 3A–C) has pollen dispersed as monads; grains isopolar, radially symmetrical, globose (usually spheroidal); amb circular; size small (ca. 20–24 μm); apertures six, colpate, globally arranged, without endoapertures, aperture membrane granular to echinate; tectum perforate-fossulate; supracteal elements scattered, small, granular; infratectum structure and foot layer not seen.

Eschscholzia caespitosa Benth. (Ranunculales, Papaveraceae, Fig. 3D–F) has pollen dispersed as monads; grains isopolar, radially symmetrical, globose (usually spheroidal); amb circular; size small to medium (ca. 19–26 μm); apertures five, colpate, located at the equator, without endoapertures, aperture membrane granular to echinate; tectum perforate-reticulate; supracteal elements echinate; infratectum structure and foot layer not seen.

Hypecoum procumbens L. (Ranunculales, Papaveraceae, Fig. 3G–I) has pollen dispersed as monads; grains isopolar, radially symmetrical, globose (usually

spheroidal); amb circular; size small (ca. 12–15 μm); apertures two, colpate, located at the equator, without endoapertures, aperture membrane echinate; tectum with scattered microperforations; supracteal elements echinate, more or less uniformly distributed on the tectum surface; infratectum structure and foot layer not seen.

Stephania cephalantha Hayata (Ranunculales, Menispermaceae, Fig. 3J–L) has pollen dispersed as monads; grains isopolar, radially symmetrical, globose; amb triangular to circular; size small (ca. 12–15 μm); apertures three, porate, located at the equator, without endoapertures, aperture membrane ornamentation not seen; tectum reticulate; without supracteal elements; infratectum columellate; foot layer present.

Berberis yunnanensis Franch. (Ranunculales, Berberidaceae, Fig. 4A–C) has pollen dispersed as monads; grains usually apolar, asymmetrical, globose (usually spheroidal); amb circular; size medium (ca. 39–42 μm); aperture one or variable, spiral and/or syncolpate, globally arranged, without endoapertures, aperture membrane smooth to granular; tectum perforate; without supracteal elements; infratectum protocolumellate (sensu Nowicke & Skvarla, 1981); foot layer absent.

Caulophyllum robustum Maxim. (Ranunculales, Berberidaceae, Fig. 4D–F) has pollen dispersed as monads; grains isopolar, radially symmetrical, globose (usually subprolate); amb circular; size medium (ca. 31–45 μm); apertures three, colpate, located at the equator, without endoapertures, aperture membrane granulate; tectum reticulate; without supracteal elements; infratectum columellate; foot layer present.

Diphylleia sinensis H. L. Li (Ranunculales, Berberidaceae, Fig. 4G–I) has pollen dispersed as monads; grains isopolar, radially symmetrical, globose (usually suboblate); amb circular; size small to medium (ca.

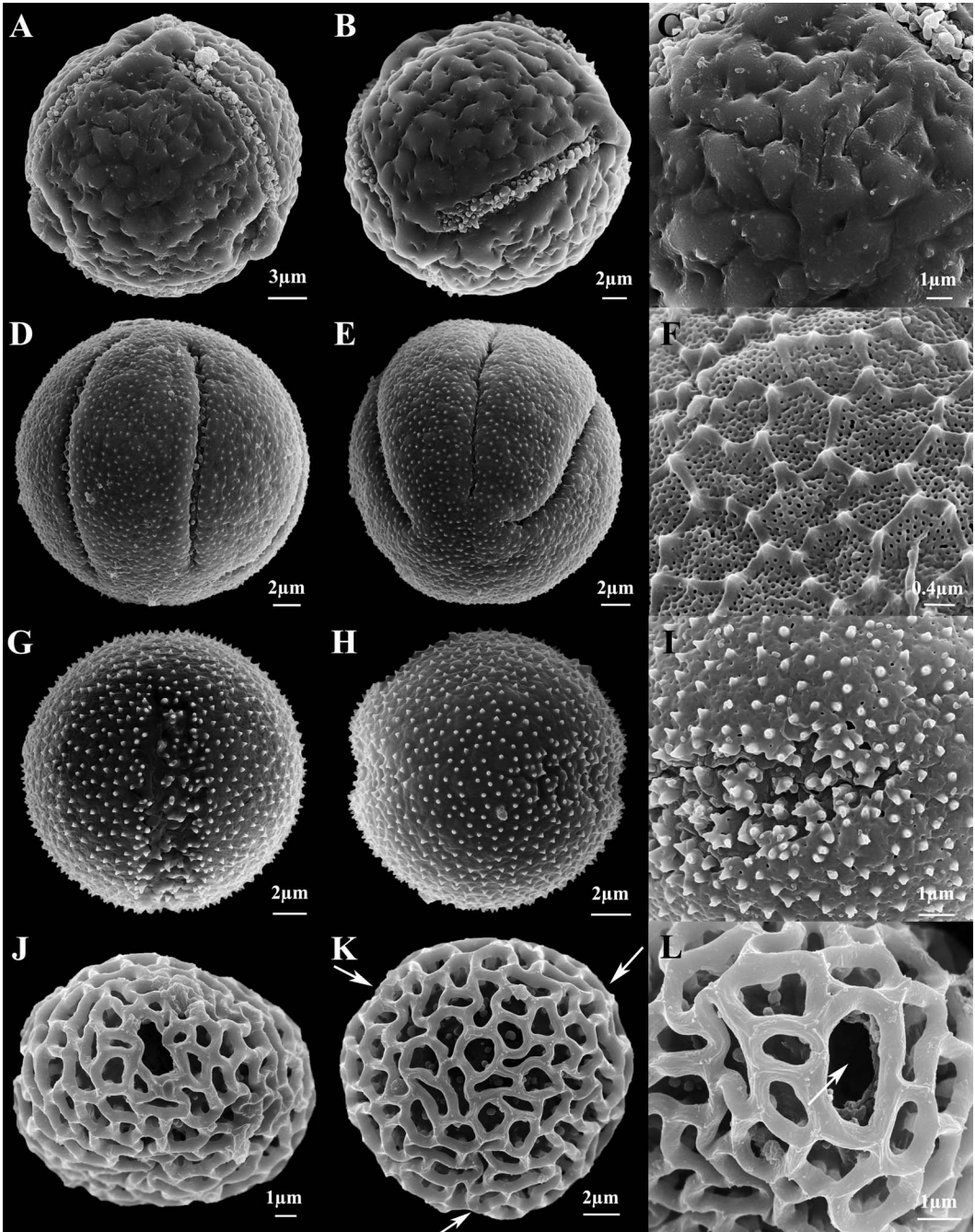


Figure 3. Scanning electron micrographs of basal eudicot pollen. A–C. *Corydalis linarioides* Maxim. —A. Equatorial view, showing single free, globose pollen grain with six short, globally distributed, colpate apertures and granular to echinate aperture membrane. —B. Polar view. —C. Detail of areolate tectum with sparse microperforations. D–F. *Eschscholzia caespitosa* Benth. —D. Equatorial view, showing single free, globose pollen grain with multiple equatorial, colpate apertures. —E. Oblique polar view. —F. Detail of perforate-reticulate tectum and echinate suprategate elements. G–I. *Hypecoum procumbens* L. —G. Equatorial view, showing single free, globose pollen grain with two equatorial, colpate apertures and echinate aperture membrane. —H. Polar view. —I. Detail of tectum with sparse microperforations and echinate suprategate elements. J–L. *Stephania cephalantha* Hayata —J. Equatorial view, showing single free, globose pollen grain with porate aperture and columellate infrategate. —K. Polar view, showing three equatorial apertures (arrowed) and reticulate tectum. —L. Detail showing porate aperture (arrowed).

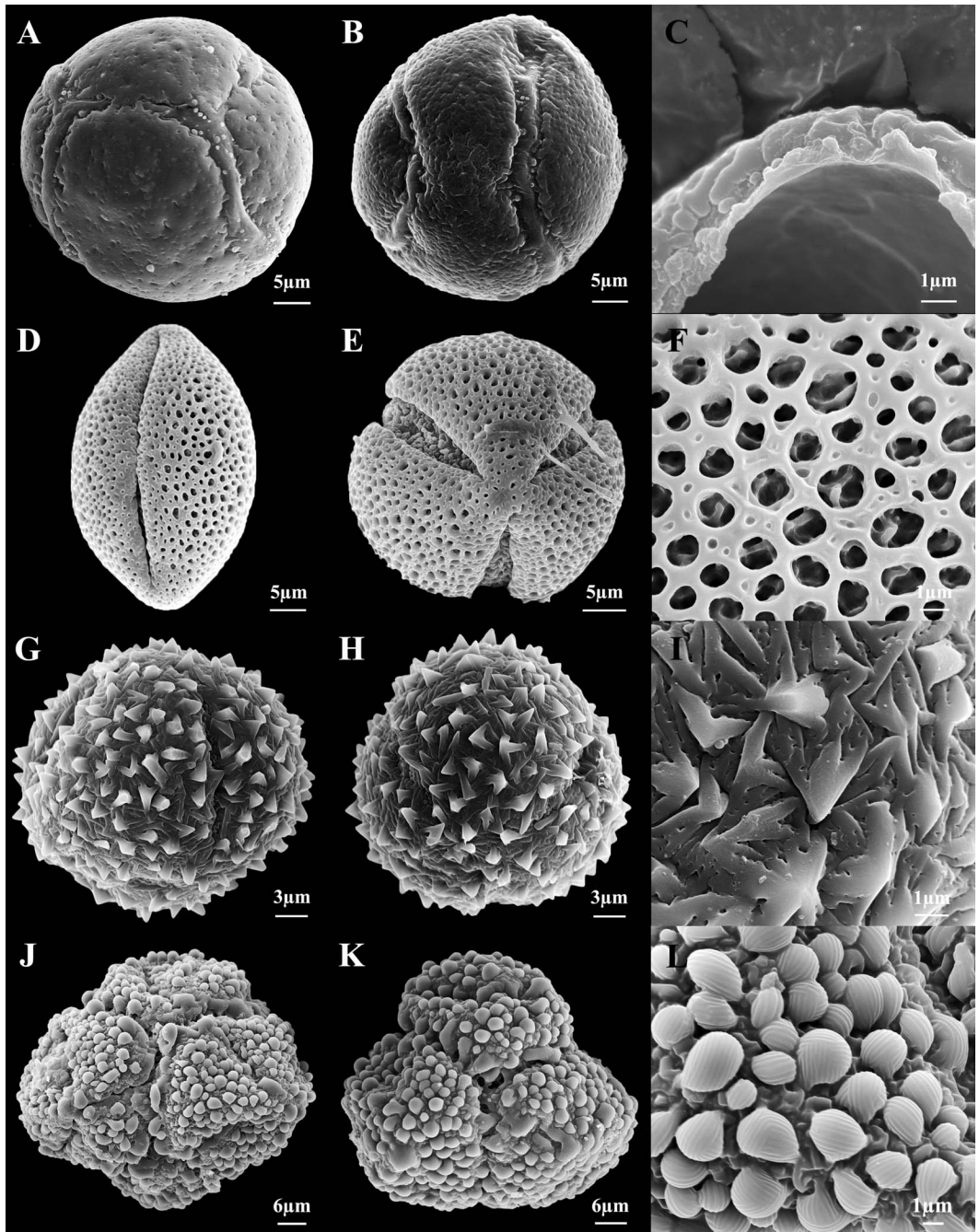


Figure 4. Scanning electron micrographs of basal eudicot pollen. A–C. *Berberis yunnanensis* Franch. —A. Single free, globose, synaperturate pollen grain with smooth to granular aperture membrane. —B. Single free, globose, synaperturate pollen grain. —C. Detail of protocolumellate infratectum. D–F. *Caulophyllum robustum* Maxim. —D. Equatorial view. —E. Polar view, showing single free, globose pollen grain with three equatorial, colpate apertures and granular aperture membrane. —F. Detail of reticulate tectum. G–I. *Diphylleia sinensis* H. L. Li. —G. Equatorial view, showing single free, globose pollen grain with three equatorial, colpate apertures. —H. Polar view. —I. Detail of rugulate tectum with scattered perforations and echinate supratectal elements. J–L. *Podophyllum emodi* Wall. ex Hook. f. & Thomson (\equiv *P. hexandrum* Royle). —J, K. Tetrads. —L. Detail of verrucate supratectal elements with striate ornamentation.

23–29 μm); apertures three, colpate, located at the equator, without endoapertures, aperture membrane ornamented; tectum rugulate with perforations; supratectal elements echinate; infratectum structure and foot layer not seen.

Podophyllum emodi Wall. ex Hook. f. & Thomson (\equiv *P. hexandrum* Royle) (Ranunculales, Berberidaceae, Fig. 4J–L) has pollen dispersed as tetrads; single grains isopolar, radially symmetrical, globose (usually spheroidal); amb circular; size medium (ca. 33–40 μm); apertures three, colpate, located at the equator, without endoapertures, aperture membrane ornamented; tectum rugulate; supratectal elements verrucate, variable in size, with striate ornamentation; infratectum structure and foot layer not seen.

Ranzania japonica (T. Itô ex Maxim.) T. Itô. (Ranunculales, Berberidaceae, Fig. 5A–C) has pollen dispersed as dyads and/or monads; single grains isopolar, radially symmetrical, globose (usually spheroidal); amb circular; size medium (ca. 35–44 μm); apertures more than six, colpate, globally arranged, without endoapertures, aperture membrane ornamented; tectum fossulate-areolate; without supratectal elements; infratectum columellate (Nowicke & Skvarla, 1981); foot layer present (Nowicke & Skvarla, 1981).

Anemone vitifolia Buch.-Ham. ex DC. (Ranunculales, Ranunculaceae, Fig. 5D–F) has pollen dispersed as monads; grains isopolar, radially symmetrical, globose (usually subprolate); amb circular; size small to medium (ca. 20–26 μm); apertures three (although this is atypical: many *Anemone* L. have pantoporate grains; cf. Baladehi et al., 2013), colpate, located at the equator, without endoapertures, aperture membrane echinate; tectum almost imperforate; supratectal elements echinate; infratectum columellate; foot layer present.

Ranunculus japonicus Thunb. (Ranunculales, Ranunculaceae, Fig. 5G–I) has pollen dispersed as monads; grains isopolar, radially symmetrical, spheroidal; amb circular; size medium (ca. 26–28 μm); apertures more than six (although many *Ranunculus* L. have tricolpate pollen grains; cf. Perveen & Qaiser, 2006), colpate, globally arranged, without endoapertures, aperture membrane echinate; tectum undulate with scattered perforations; supratectal elements echinate; infratectum structure and foot layer not seen.

Thalictrum delavayi Franch. (Ranunculales, Ranunculaceae, Fig. 5J–L) has pollen dispersed as monads; grains apolar, radially symmetrical, spheroidal; amb circular; size small (ca. 18–20 μm); apertures more than six, porate, globally arranged, without endoapertures, aperture membrane echinate;

tectum imperforate; supratectal elements echinate; infratectum structure and foot layer not seen.

Trollius ranunculoides Hemsl. (Ranunculales, Ranunculaceae, Fig. 6A–C) has pollen dispersed as monads; grains isopolar, radially symmetrical, globose (usually spheroidal); amb circular; size small (ca. 17–19 μm); apertures three, colpate, located at the equator, without endoapertures, aperture membrane echinate; tectum striate with perforations between the striae; without supratectal elements; infratectum structure and foot layer not seen.

Meliosma arnotiana (Wight) Walp. (Sabiaceae, Fig. 6D–F) has pollen dispersed as monads; grains isopolar, radially symmetrical, globose (usually subprolate); amb circular; size small (ca. 18–22 μm); apertures three, colpate, located at the equator, endoapertures present, aperture membrane granulate; tectum foveolate; without supratectal elements; infratectum columellate; foot layer present.

Grevillea baueri R. Br. (Proteales, Proteaceae, Fig. 6G–I) has pollen dispersed as monads; grains isopolar, radially symmetrical, oblate; amb polygonal; size large (ca. 60–65 μm); apertures three, porate, located at the equator, without endoapertures, aperture membrane smooth; tectum perforate-(micro-)undulate; supratectal elements verrucate; infratectum structure and foot layer not seen.

Hakea mitchellii Meisn. (Proteales, Proteaceae, Fig. 6J–L) has pollen dispersed as monads; grains isopolar, radially symmetrical, oblate; amb polygonal; size large (ca. 53–69 μm); apertures three, porate, located at the equator, without endoapertures, aperture membrane ornamented; tectum sparsely perforate; supratectal elements gemmate; infratectum structure and foot layer not seen.

Macadamia rousseii (Vieill.) Sleumer (Proteales, Proteaceae, Fig. 7A–C) has pollen dispersed as monads; grains isopolar, radially symmetrical, oblate; amb polygonal; size medium (ca. 31–37 μm); apertures three, porate, located at the equator, without endoapertures, aperture membrane ornamented; tectum around the aperture area perforate and intercolpium foveolate; without supratectal elements; infratectum short-columellate; foot layer present.

Stenocarpus umbelliferus Druce (Proteales, Proteaceae, Fig. 7D, E) has pollen dispersed as monads; grains isopolar, radially symmetrical, oblate; amb polygonal; size medium (ca. 27–35 μm); apertures three, porate, located at the equator, without endoapertures, aperture membrane ornamented; tectum perforate to reticulate; without supratectal elements; infratectum columellate; foot layer present.

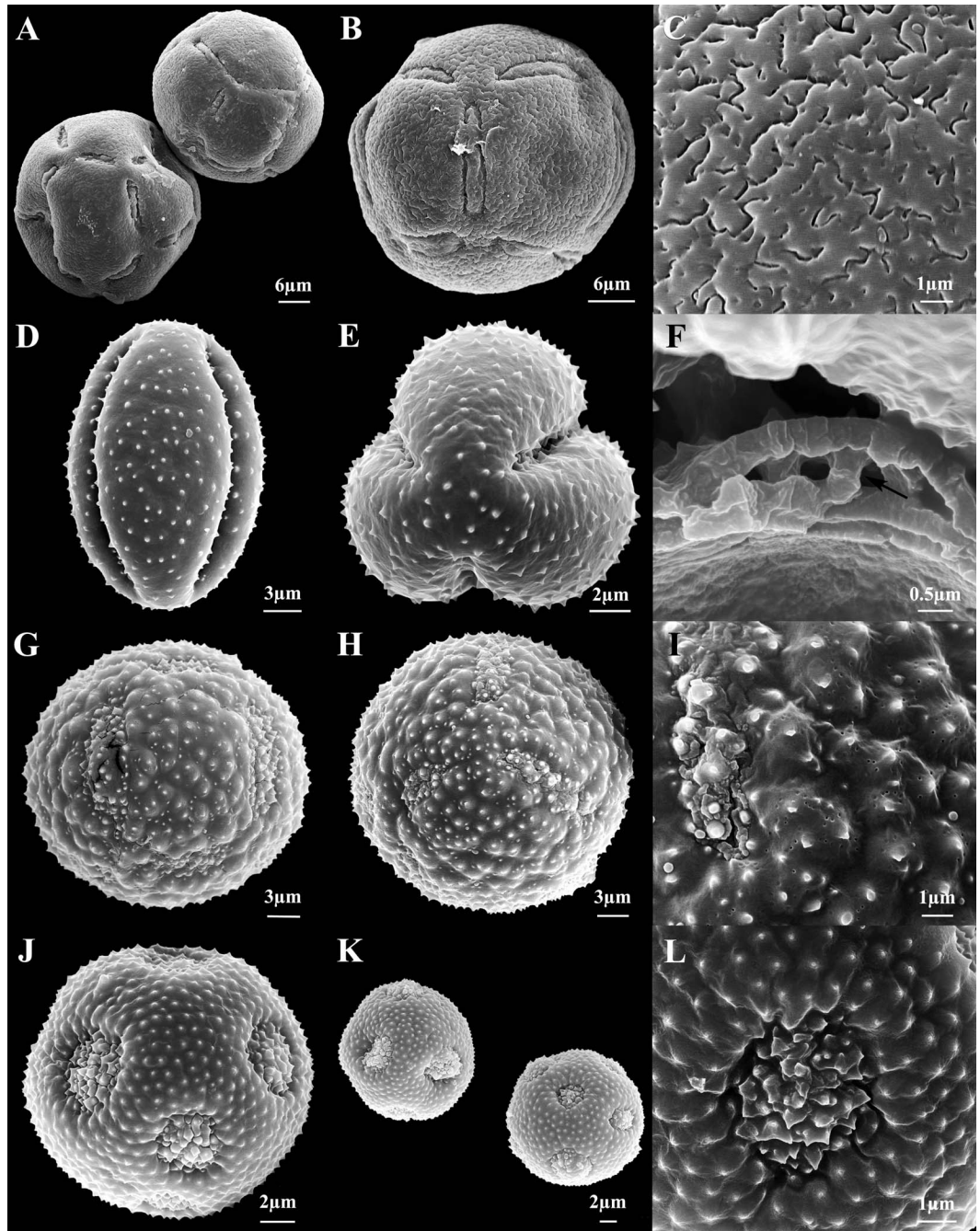


Figure 5. Scanning electron micrographs of basal eudicot pollen. A–C. *Ranzania japonica* (T. Itô ex Maxim.) T. Itô. —A. Dyad. —B. Single free, globose pollen grain with multiple globally distributed, colpate apertures and ornamented aperture membrane. —C. Detail of perforate-areolate tectum. D–F. *Anemone vitifolia* Buch.-Ham. ex DC. —D. Equatorial view, showing single free, globose pollen grain with three equatorial, colpate apertures and imperforate tectum. —E. Polar view. —F. Detail of columellate infratectum (arrowed). G–I. *Ranunculus japonicus* Thunb. —G. Equatorial view, showing single free, spheroidal pollen grain with globally distributed, colpate apertures and echinate aperture membrane. —H. Polar view. —I. Detail of tectum with scattered small perforations and echinate suprategular elements. J–L. *Thalictrum delavayi* Franch. —J. Single free, spheroidal pollen grain with globally distributed, porate apertures and echinate aperture membrane. —K. Two free, spheroidal pollen grains. —L. Detail showing porate aperture with coarsely ornamented membrane and imperforate tectum.

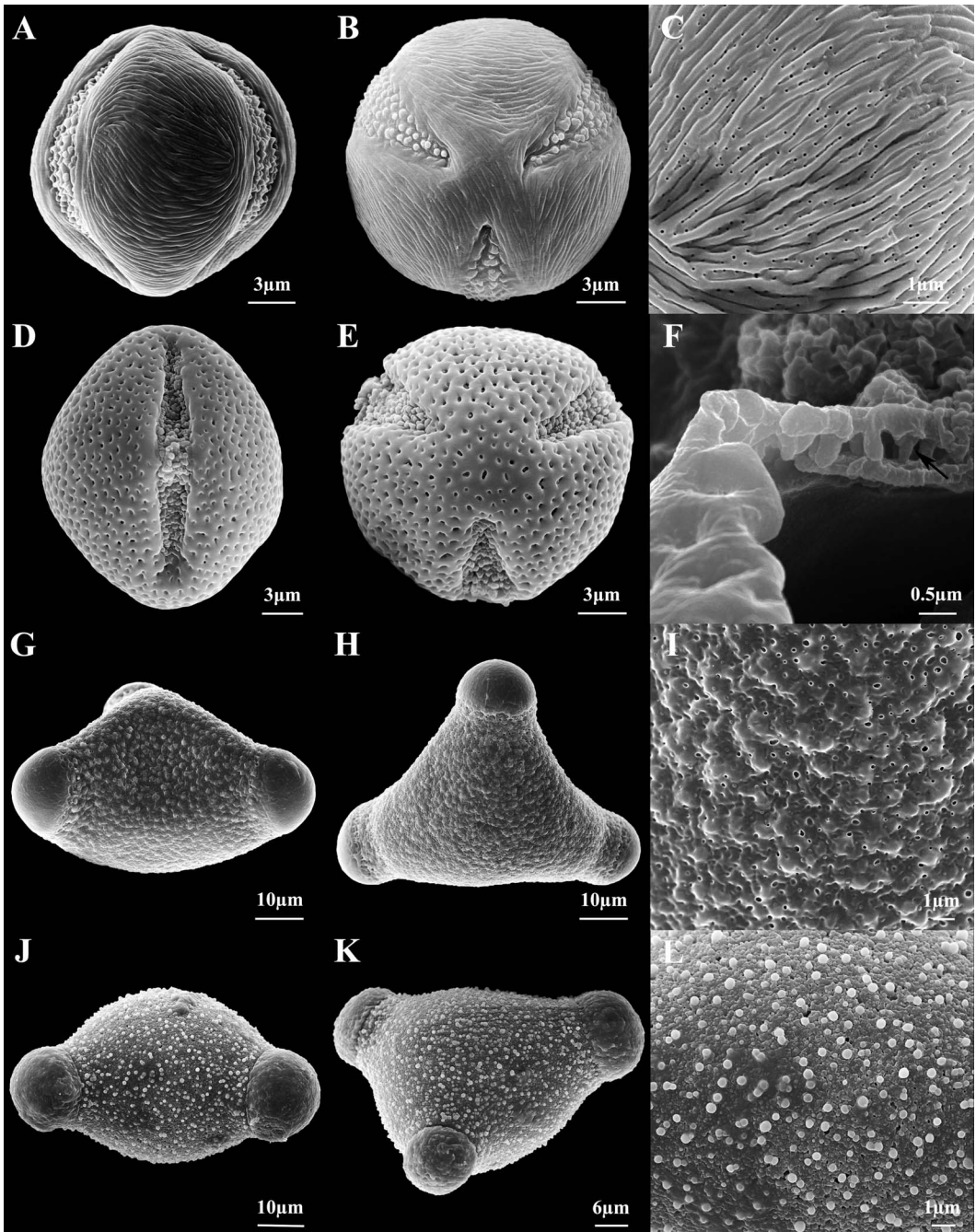


Figure 6. Scanning electron micrographs of basal eudicot pollen. A–C. *Trolius ranunculoides* Hemsl. —A. Equatorial view, showing single free, spheroidal pollen grain with three equatorial, colpate apertures and echinate aperture membrane. —B. Polar view. —C. Detail of striate tectum with perforations. D–F. *Meliosma arnottiana* (Wight) Walp. —D. Equatorial view, showing single free, globose pollen grain with three equatorial, colpate apertures and granular aperture membrane. —E. Polar view. —F. Detail of columellate infratectum (arrowed). G–I. *Grevillea baueri* R. Br. —G. Equatorial view, showing single free pollen grain with three equatorial, porate apertures and granular aperture membrane. —H. Polar view. —I. Detail showing undulate tectum with perforations. J–L. *Hakea mitchellii* Meisn. —J. Equatorial view, showing single free pollen grain with three equatorial, porate apertures and ornamented aperture membrane. —K. Oblique polar view. —L. Detail showing gemmate supracteal elements.

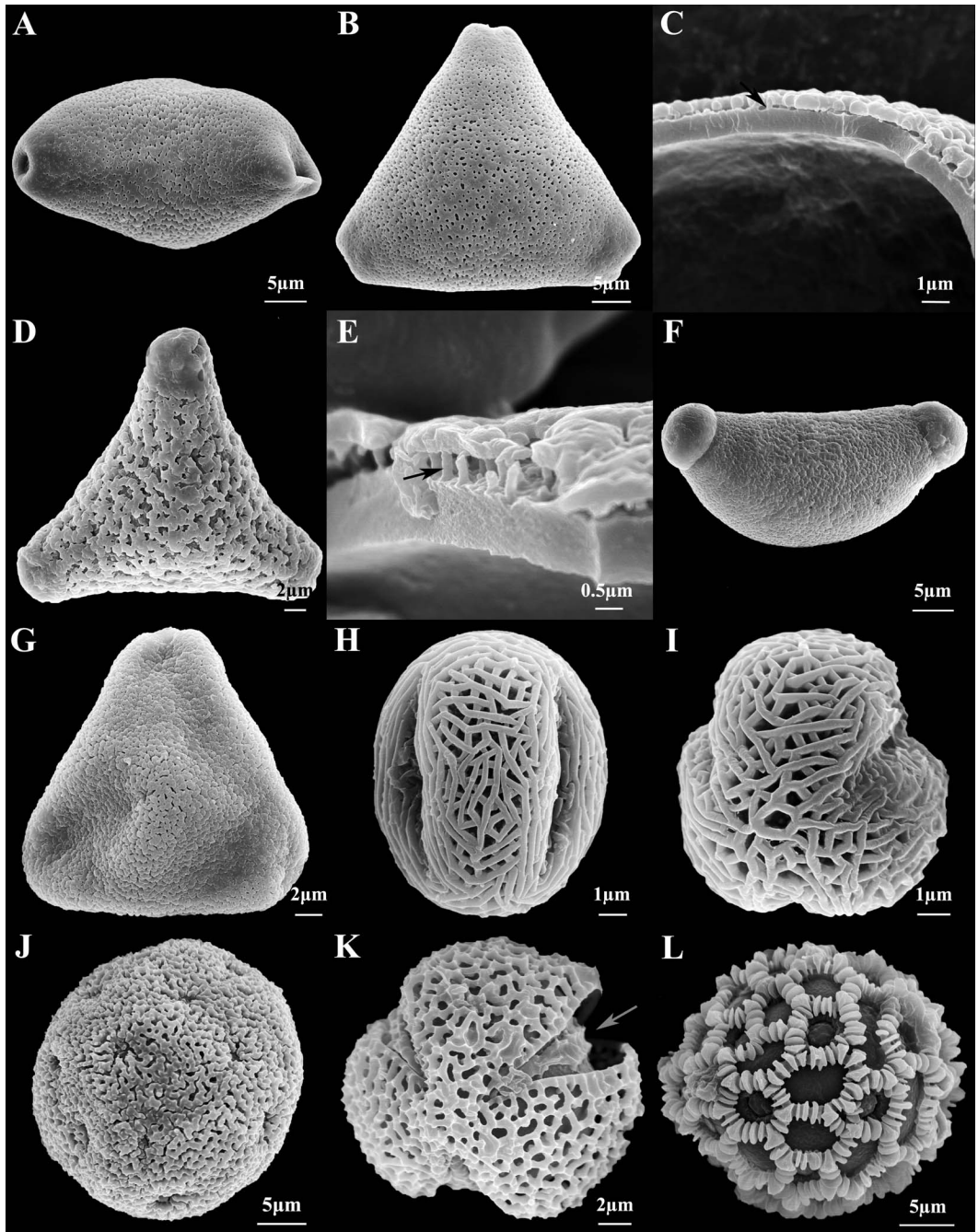


Figure 7. Scanning electron micrographs of basal eudicot pollen. A–C. *Macadamia roussetii* (Vieill.) Sleumer. —A. Equatorial view. —B. Polar view, showing single free pollen grain with three equatorial, porate apertures. —C. Detail showing shortly columellate infratectum (arrowed). D, E. *Stenocarpus umbelliferus* Druce —D. Equatorial view, showing single free pollen grain with three equatorial, porate apertures. —E. Detail showing columellate infratectum (arrow). —F. *Banksia acanthopoda* (A. S. George) A. R. Mast & K. R. Thiele, single free pollen grain with two equatorial, porate apertures and ornamented aperture membrane. —G. *Helicia macrostachya* Lauterb., polar view, showing single free pollen grain with three equatorial, porate apertures. H, I. *Tetracentron sinense* Oliv. —H. Equatorial view, showing single free spheroidal pollen grain with three equatorial, colpate apertures and striato-reticulate tectum. —I. Polar view. —J. *Buxus sempervirens* L., single free, spheroidal pollen grain with multiple globally distributed, porate apertures. —K. *Didymelea integrifolia* J. St.-Hil., polar view, showing single free, spheroidal pollen grain with three equatorial, colpate apertures and operculum present (arrow). —L. *Pachysandra axillaris* Franch., single free, spheroidal pollen grain with multiple globally distributed, porate apertures, ornamented aperture membrane, crotonoid tectum, and angular supracteal elements.

Banksia acanthopoda (A. S. George) A. R. Mast & K. R. Thiele (Proteales, Proteaceae, Fig. 7F) has pollen dispersed as monads; grains subisopolar, bilaterally symmetrical, boat-shaped; amb elliptic; size medium (ca. 35–37 μm); apertures two, porate, located at the equator, without endoapertures, aperture membrane ornamented; tectum rugulate with perforations; without supracteal elements; infratectum structure and foot layer not seen.

Helicia macrostachya Lauterb. (Proteales, Proteaceae, Fig. 7G) has pollen dispersed as monads; grains isopolar, radially symmetrical, oblate, amb polygonal; size small to medium (ca. 21–26 μm); apertures three, porate, located at the equator, without endoapertures, aperture membrane ornamented; tectum rugulate-reticulate; without supracteal elements; infratectum structure and foot layer not seen.

Tetractron sinense Oliv. (Trochodendrales, Trochodendraceae, Fig. 7H, I) has pollen dispersed as monads; grains isopolar, radially symmetrical, globose (usually spheroidal); amb circular; size small (ca. 9–12 μm); apertures three, colpate, located at the equator, without endoapertures, aperture membrane ornamented; tectum striato-reticulate; without supracteal elements; infratectum columellate; foot layer present.

Buxus sempervirens L. (Buxales, Buxaceae, Fig. 7J) has pollen dispersed as monads; grains apolar, radially symmetrical, spheroidal; amb circular; size medium (ca. 28–31 μm); apertures more than six, porate, globally arranged, without endoapertures, aperture membrane ornamented; tectum foveolate to reticulate; supracteal elements echinate; infratectum columellate; foot layer present.

Didymeles integrifolia J. St.-Hil. (Buxales, Buxaceae, Fig. 7K) has pollen dispersed as monads; grains isopolar, radially symmetrical, globose (usually spheroidal); amb circular; size small (ca. 17–23 μm); apertures three, colpate, located at the equator, without endoapertures, aperture membrane ornamented; operculum present; tectum reticulate; supracteal elements echinate; infratectum columellate; foot layer present.

Pachysandra axillaris Franch. (Buxales, Buxaceae, Fig. 7L) has pollen dispersed as monads; grains apolar, radially symmetrical, spheroidal; amb circular; size medium (ca. 25–27 μm); apertures more than six, porate, globally arranged, without endoapertures, aperture membrane ornamented; tectum appearing crotonoid; supracteal elements angular; infratectum columellate; foot layer present.

grains isopolar (Character 2), radially symmetrical (Character 3), and globose (Character 4), with three equatorially distributed (Characters 8 and 9), simple apertures (Character 10), tectum (Character 16), foot layer (Character 19), and endexine (Character 20) all present and operculum (Character 13) absent. In contrast to these generally consistent features, others are rather diverse, including pollen shape class (Character 5), outline in polar view (Character 6), pollen size (Character 7), ectoaperture shape (Character 11), supracteal element shape (Character 15), and tectum sculpture (Character 17). For instance, tectum sculpture (Character 17) was found to exhibit nine states within the basal eudicots: areolate, fossulate, perforate (including foveolate), imperforate, striate, striato-reticulate, reticulate, rugulate, and crotonoid. Of these, the crotonoid state was found to be unique in this study, occurring only in *Pachysandra* Michx. (Buxaceae).

Anemone, *Buxus* L., *Nelumbo* Adans., and *Ranunculus* displayed comparatively higher pollen diversity than the other genera studied in this paper. For instance, within *Anemone* alone, we found at least five states of ectoaperture shape (Character 11, viz., porate, colpate, spiral, syncolpate, and zonate) and four states of aperture number (Character 8, viz., one, three, six, and multiple). The morphological diversity of pollen within *Nelumbo*, despite having only two species, encompasses a variety of ectoaperture types, including globose-spheroidal pollen grains with tricolpate, spiral, or zonate apertures and boat-shaped pollen grains with monosulcate apertures (Appendix 3). Other genera, such as *Batrachium* (DC.) Gray and *Cimicifuga* L., are more conservative in pollen morphology, with only one or two states found for each of the 20 characters studied (albeit with not all characters documented in the literature).

RECONSTRUCTION OF POLLEN CHARACTER EVOLUTION

In reconstructing the plesiomorphic states for eudicots, 16 of the 20 characters studied displayed congruent states across all the five methods of inference (DFP, DML, DHB, CFP, and CHB), counting ambiguous states as congruent. These uniform plesiomorphic states were as follows: grains released as monads, isopolar (state ambiguous under DFP and CFP), radially symmetrical, globose, spheroidal, and circular in polar view, size medium, aperture position equatorial (ambiguous under DFP), aperture structure simple, ectoaperture shape colpate, tectum sculpture reticulate (ambiguous under DFP and CFP), infratectum structure columellate (ambiguous under DML), operculum and supracteal elements absent, tectum present, and foot layer

POLLEN MORPHOLOGICAL DIVERSITY

The majority of basal eudicot taxa are characterized by pollen released as monads (Character 1), with

present. For the remaining four characters, two were resolved inconsistently across the five analyses: aperture number (Character 8; ambiguous under DFP and CFP, three under DML and CHB, and more than six under DHB) and endexine presence/absence (Character 20; present under CFP, DPF, DML, and CHB; absent under DHB). The plesiomorphic states for endoaperture shape (Character 12) and supra-rectal element shape (Character 15) were both inferred as inapplicable, which is consistent with the observed plesiomorphies of simple apertures and absent supra-rectal elements.

A total of 357 unambiguous pollen character state changes were inferred at or above tribal level across all the five analyses (DFP, DML, DHB, CFP, and CHB) and are listed in Table 2. The unambiguous changes inferred under CHB are shown in Figure 8, along with changes in the two ecological traits studied. Under this method of inference, 222 unambiguous pollen character state changes were inferred and found to be distributed irregularly, concentrated on 117 internodes of the phylogenetic tree. Four internodes were each inferred to bear six state changes (the root of Buxales, the root of Sabiaceae, the root of Fumarioideae, and the root of the clade comprised of *Meconopsis* Vig. and *Papaver* L., both in Papaveraceae). Three further internodes were inferred to bear five state changes (the root of Papaveraceae, the root of Adonideae, and the root of Coptidoideae within Ranunculaceae); and five internodes, four state changes (Proteaceae, three clades within Proteaceae [the clade comprising Petrophileae and *Franklandia* R. Br., the root of Petrophileae, and the clade comprising Banksieae and *Sphalmium* B. G. Briggs, B. Hyland & L. A. S. Johnson], and the clade composed of *Aspidocarya* Hook. f. & Thomson and *Parabaena* Miers [Menispermaceae]). Eleven further internodes were inferred to have three changes and 33 to have two changes. A majority of internodes were associated with only one character state change. No character state changes were found at the other 76 nodes. The states for the ecological characters pollination syndrome and plant growth form changed 17 and 22 times respectively.

TESTS FOR EVOLUTIONARY CORRELATIONS

According to the ML correlated evolution test (Table 3), *P* values for the correlations between herbaceous growth form and spheroidal pollen shape class and between arborescent growth form and oblate pollen shape class were 0.002 and 0.008, respectively. The correlation between anemophily and aperture number greater than or equal to four had

a *P* value of 0.071. In the MCMC test, the LBF values for the former two pairs were 15.052 and 9.399, and for the latter pair was less than zero (−0.758458). Transition rates for the two pairs of correlated characters are shown in Table 4. The results of LR tests for specific hypotheses of contingent evolution are shown in Table 5. When restricted such that $q_{31} = q_{42}$, for the correlation between herbaceous growth form and spheroidal pollen shape, the *P* value was 0.0085, and for the correlation between arborescent growth form and oblate pollen shape, 0.0082. When each of the transition rates in Table 4 was restricted to zero (q_{24} and q_{42} for herbaceous growth form and spheroidal pollen shape were not restricted since they were resolved as zero in the ML correlated evolution test), *P* values were all less than 0.01 (Table 5), with the exception of q_{21} and q_{43} for arborescent growth form and oblate shape class.

DISCUSSION

SYSTEMATIC SIGNIFICANCE OF POLLEN MORPHOLOGY IN BASAL EUDICOTS

For clades of basal eudicots estimated using molecular data, at or above tribal level, at least 58 were characterized by unambiguous pollen character state changes in at least one of the five analyses conducted (Table 2; Fig. 8). This to us suggests that pollen characters have great potential significance to provide independent support for phylogenetic relationships estimated from other (e.g., molecular) evidence in basal eudicot lineages. Furthermore, several taxa, for instance Banksieae (Proteaceae), Circaeasteraceae, Cissampelinae (Menispermaceae), and Sabiaceae, may be identified by their unique pollen features (Table 2, Fig. 8), implying that pollen characters may also provide evidence for classification and diagnosis of taxa in this group.

An inferred character state transition to three apertures (Character 8; under DFP and CFP) on the branch leading to eudicots supports their monophyly, as proposed through molecular evidence, and agrees with previous studies that tricolpate pollen is a synapomorphy of this clade (Chase et al., 1993; Furness & Rudall, 2004; APG III, 2009; Qiu et al., 2010; Soltis et al., 2011). Within eudicots, the close affinity between Buxales and core eudicots (indicated by Worberg et al., 2007; Moore et al., 2010; Soltis et al., 2011; Barniske et al., 2012) is supported by transitions to globally distributed apertures (Character 9; DHB), supra-rectal elements that are present and angular in shape (Character 14 and 15; both

Table 2. States of 18 of the 20 studied pollen characters for lineages at or above tribal level inferred from Fitch parsimony (FP), maximum likelihood (ML), and hierarchical Bayesian inference (HB) on comprehensive (C) and democratic (D) datasets. Pollen characters and state numbers follow Table 1. a, DFP; b, DML; c, DHB; d, CFP; e, CHB.

Clades	Pollen characters																	
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	17	18	20
Ceratophyllales– eudicots				1 ^{ce}		0 ^{abde}		3 ^{bc} ; 5 ^c	0 ^{ce}									
Eudicots								3 ^{ad}										
Trochodendrales– Buxales–core eudicots	0 ^c						0 ^{acde}	5 ^c								4 ^c		1 ^{ce}
Trochodendrales– Buxales–core eudicots–Sabiaceae– Proteales								3 ^c										
Buxales–core eudicots									1 ^c					1 ^{ce}	4 ^c ; 6 ^c	5 ^c		
Ranunculales	1 ^c													1 ^c	1 ^c			1 ^{ce}
Core Ranunculales– Eupteleaceae	2 ^c			3 ^c				4 ^c						0 ^c				
Core Ranunculales Menispermaceae– Ranunculaceae– Berberidaceae					3 ^{acd}			3 ^c									1 ^{bc} ; 2 ^c	
Ranunculaceae– Berberidaceae				2 ^c														
Ranunculaceae Ranunculaceae (excluding <i>Glaucidium</i>)				2 ^a											1 ^{ce}	1 ^{ce}		
Ranunculaceae (excluding <i>Hydrastis</i> and <i>Glaucidium</i>)	1 ^c			2 ^d				5 ^c							1 ^{abd}	1 ^{abde}	1 ^c	
Thalictroideae				3 ^c														
Ranunculeae– Anemoneae	0 ^c							5 ^c	1 ^c									
Anemoneae	0 ^c							5 ^c	1 ^c									
Ranunculeae	2 ^c							3 ^c	0 ^c									
Delphinieae– Adonideae				3 ^{ce}														
Delphinieae				3 ^b														
Adonideae				2 ^{ce}			0 ^{ce}							0 ^{ce}			4 ^c	
Coptidoideae	0 ^{abde}						0 ^{ce}	5 ^{abde}	1 ^{abde}		1 ^{abde}							
Berberidaceae				2 ^{ad} ; 3 ^c													5 ^c	
Berberidoideae– Nandinoideae				2 ^c				5 ^c	1 ^c								6 ^c	2 ^{ce}
Berberidoideae	1 ^c	0 ^c						5 ^{abde}	1 ^{abde}		2 ^c						1 ^c ; 2 ^c	
Nandinoideae				3 ^{ce}			2 ^{ce}	3 ^c									5 ^a	0 ^{ce}
Podophylloideae																	3 ^c ; 4 ^c	
Menispermaceae				3 ^{bc}			0 ^{abde}			1 ^{abde}				2 ^b ; 0 ^c				
Tinosporoideae				4 ^{ce}										2 ^{acd}				
Cissampelinae				2 ^{abde}												1 ^{abde}		
Circaeasteraceae– Lardizabaloideae				3 ^b														
Circaeasteraceae				2 ^c													3 ^{abc} ; 4 ^{de}	
Lardizabaloideae				4 ^c			0 ^c											
Akebieae				3 ^c			0 ^{abc}											
Papaveraceae	0 ^{ce}							5 ^c	1 ^{ce}						1 ^{bde}	1 ^b ; 0 ^{ce}		
Fumarioideae	2 ^{ce}						0 ^c	3 ^c ; 2 ^c	0 ^{ce}						3 ^c ; 5 ^c	1 ^{bc} ; 2 ^c		
Papaveroideae															1 ^a	1 ^{ad}		

Table 2. Continued.

Clades	Pollen characters																	
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	17	18	20
Sabiaceae–Proteales					3 ^c ; 4 ^c											1 ^c		
Proteales					4 ^c													
Proteaceae– <i>Platanus</i>						1 ^c										5 ^c	4 ^c	
Proteaceae					0 ^{abcde}	1 ^{abcde}					1 ^{abcde}					1 ^{bc} ; 2 ^c	4 ^c	0 ^b
Proteoideae							2 ^c									1 ^a	1 ^c	0 ^{ad}
Persoonioideae														1 ^{ce}	0 ^{ce}			
Persoonieae					1 ^{bce}		2 ^c							1 ^{abd}	0 ^{abd}			0 ^{ad}
Bellendenoideae– Persoonioideae																1 ^a	0 ^{ce}	
Symphionematoideae					1 ^c									1 ^{ce}	2 ^c ; 5 ^c			0 ^{ce}
Symphionematoideae– Proteoideae																		1 ^c
Grevilleoideae																5 ^{bc} ; 6 ^c	0 ^{ce}	1 ^{bc}
Petrophileae					0 ^{ce}		1 ^c								3 ^c ; 5 ^c	5 ^c ; 6 ^c		
Proteaceae							0 ^{abce}											1 ^{abd}
Conospermeae							2 ^{abc}											
Embothriaceae																	1 ^c ; 2 ^c	3 ^{ce}
Leucadendreae																	1 ^c	1 ^{ce}
Banksiaeae			0 ^{abd}			2 ^{abd}	0 ^c	2 ^{abd}										
Roupaleae														1 ^{ce}	3 ^c ; 5 ^c			
Sabiaceae					3 ^{ab} ; 2 ^c		0 ^c			1 ^{abcde}		0 ^c ; 1 ^{abc}				5 ^c ; 2 ^c		1 ^{ce}
Buxaceae	0 ^c						1 ^c	5 ^c	1 ^c			2 ^c		1 ^{abd}	1 ^{ab}	7 ^c ; 8 ^c		
Buxaceae (excluding Didymelaceae)	0 ^{ab}							5 ^{ab}	1 ^{ab}		1 ^{abce}							
Trochodendraceae		2 ^c						0 ^b	3 ^c									

Note: Characters 16 and 19 are not listed here because they displayed no unambiguous state changes at or above tribal level across the studied taxa.

under DHB and CHB), and reticulate tectum sculpture (Character 17; DHB).

Ranunculales

Within Ranunculales, relationships among Eupteleaceae, Papaveraceae, and core Ranunculales have remained an open question since the early molecular analysis of Chase et al. (1993). Some studies have provided support for the woody Eupteleaceae diverging first and Papaveraceae forming the sister group to core Ranunculales (Hilu et al., 2003; S. Kim et al., 2004; Worberg et al., 2007; W. Wang et al., 2009; Soltis et al., 2011; Sun et al., 2016). Conversely, recent results have suggested Papaveraceae diverged first, and Eupteleaceae is sister to core Ranunculales (Hoot et al., 1999; Soltis et al., 2000, 2003, 2007; Hilu et al., 2008; Barniske et al., 2012). In our optimizations, based on the latter topology, the following states were found to be shared by Eupteleaceae and core Ranunculales: polarity isopolar (Character 2; DHB), shape subprolate (Character 5; CHB), apertures four to six (Character 8; DHB),

and suprategal elements absent (Character 14; DHB).

Within Papaveraceae, the systematic positions of *Pteridophyllum* Siebold & Zucc. and *Hypecoum* L. have been subject to controversy (W. Wang et al., 2009; Pérez-Gutiérrez et al., 2012; Hoot et al., 2015; Sauquet et al., 2015). *Pteridophyllum* has been indicated as the earliest-diverging lineage in Papaveraceae (Hoot et al., 2015) or as sister to Papaveroideae (Sauquet et al., 2015), but both with poor support. Based on molecular evidence and morphological traits, W. Wang et al. (2009) suggested *Pteridophyllum* and *Hypecoum* are sister groups. In our study, their sister-group relationship is supported by transitions to suprategal elements present (Character 14; under DFP) and echinate (Character 15; under DFP, DML, CFP, and CHB). This is in agreement with Sauquet et al. (2015), who suggest that *Pteridophyllum* and *Hypecoum* are similar in perianth symmetry, number of floral spurs, and number of stamens. Furthermore, W. Wang et al. (2009) estimated that *Pteridophyllum* and *Hypecoum* are close to *Dicentra* Bernh. and *Corydalis* DC., these four together composing Fumarioideae. Based on our

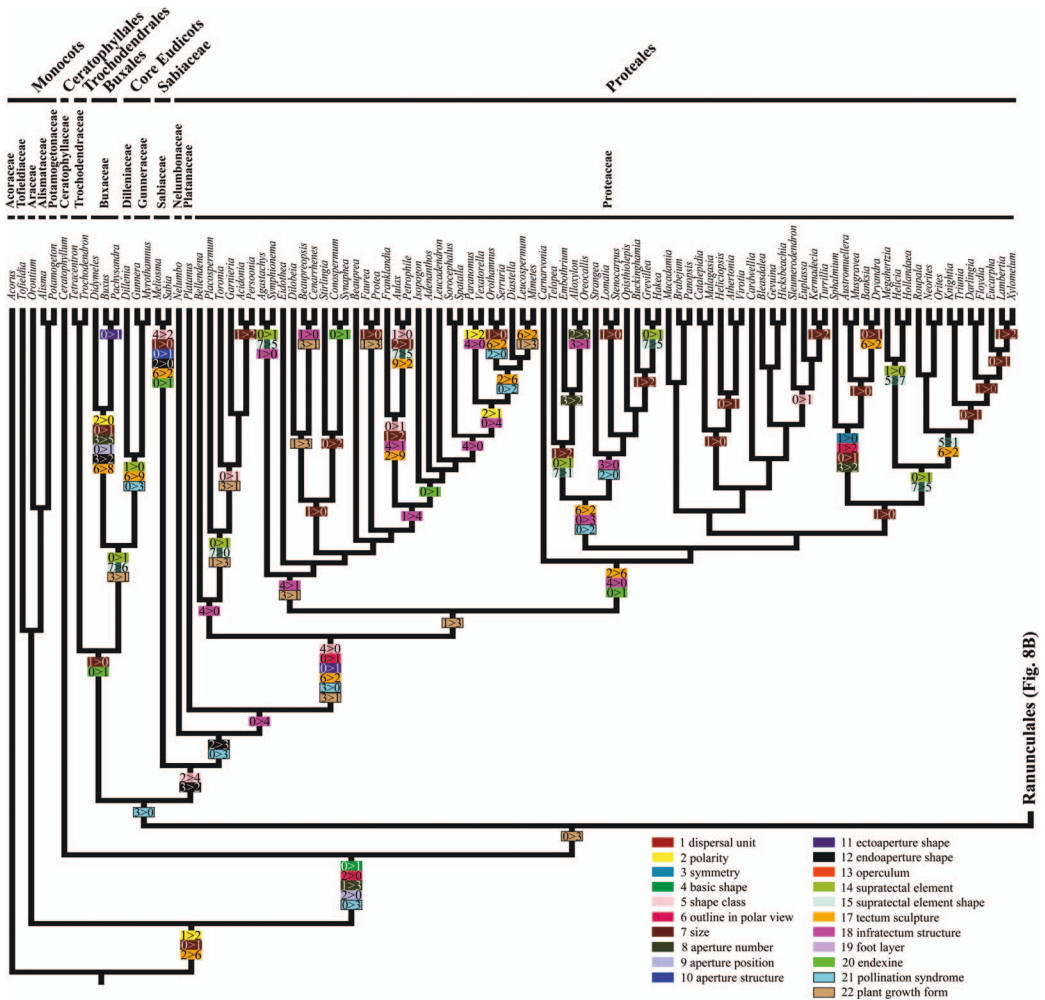


Figure 8A. The unambiguous character state changes inferred under hierarchical Bayesian inference using the comprehensive dataset (CHB) on a maximum likelihood (ML) phylogeny of basal eudicots. Changes in each character (except Character 16, in which no change occurred) are labeled assuming a single transition between different states at adjacent nodes. Numbers on bars indicate state transformations from the lower to upper node. Characters and states follow Table 1. Buxales, Proteales, Sabiaceae, Trochodendrales, and outgroups.

results, six character states, viz., polarity isopolar (Character 2; DHB and CHB), pollen grain size small (Character 7; CHB), apertures (Character 8) two (CHB) or three (DHB) and equatorially distributed (Character 9; DHB and CHB), supratectal elements verrucate (Character 15; DHB and CHB), and tectum sculpture perforate (Character 17; DML, DHB, and CHB), were inferred to be gained on the lineage leading to Fumarioideae, providing palynological evidence for the treatment by W. Wang et al. (2009).

Molecular phylogenies have strongly supported that *Kingdonia* Balf. f. & W. W. Sm. and *Circaeaster* Maxim. form a monophyletic clade, Circaeasteraceae (Hoot et al., 1999; Soltis et al., 2007; W. Wang et al.,

2009). In our study, these two genera were inferred together to have gained spheroidal pollen grains (Character 5; CHB) with a striate tectum sculpture (Character 17; all five analyses). The striate tectum is relatively rare in basal eudicots, and may therefore distinguish Circaeasteraceae from other families in Ranunculales.

Berberidaceae has been divided into three subfamilies based on molecular phylogenetic results, a division which also corresponds to chromosome base numbers, i.e., Podophylloideae ($x = 6$), Berberidoideae ($x = 7$), and Nandinoideae ($x = 8/10$) (Kim & Jansen, 1998; Y. D. Kim et al., 2004; Wang et al., 2007a; W. Wang et al., 2009). Each of these

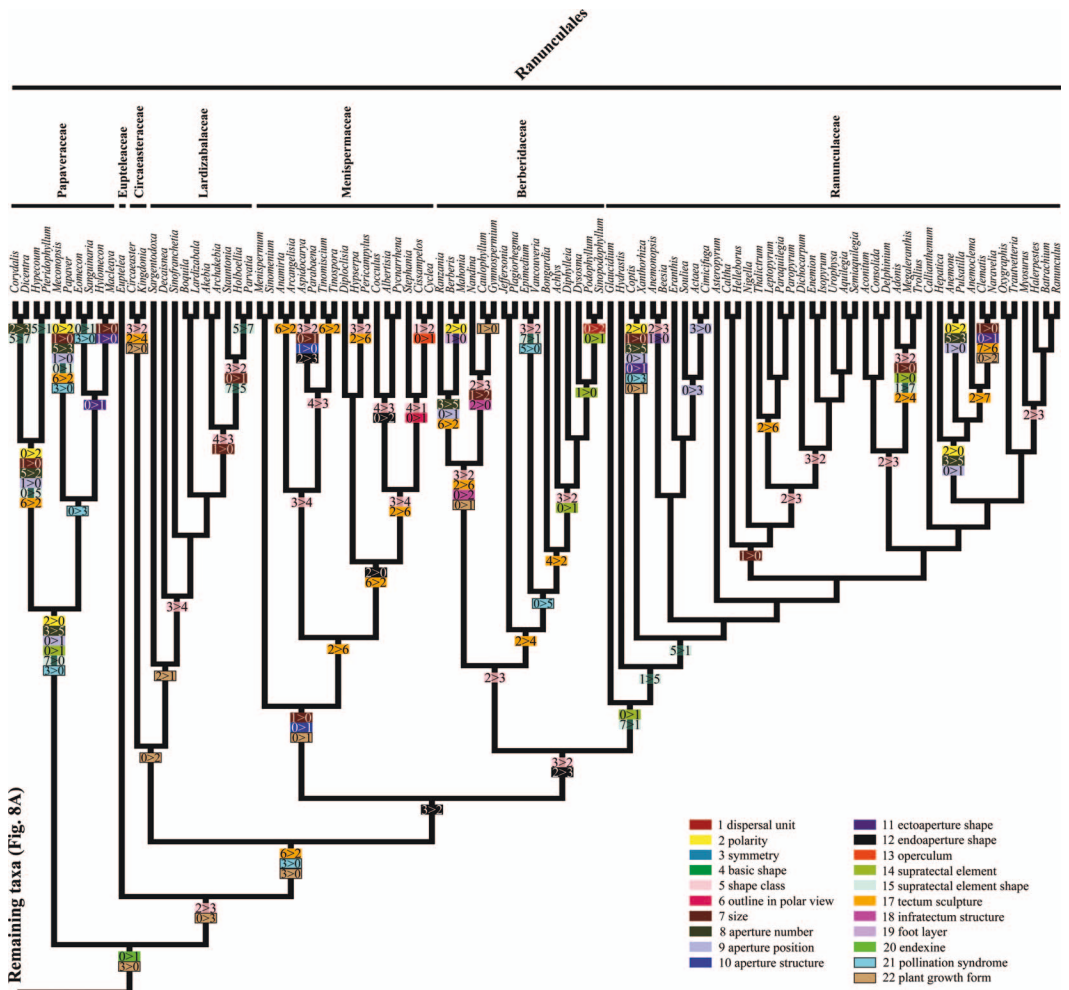


Figure 8B. The unambiguous character state changes inferred under hierarchical Bayesian inference using the comprehensive dataset (CHB) on a maximum likelihood (ML) phylogeny of basal eudicots. Changes in each character (except Character 16, in which no change occurred) are labeled assuming a single transition between different states at adjacent nodes. Numbers on bars indicate state transformations from the lower to upper node. Characters and states follow Table 1. Order Ranunculales.

subfamilies is supported by pollen morphological character state changes in our investigation (Table 2, Fig. 8). Within Berberidoideae, *Berberis* and *Mahonia* share multiple similar features and have long been regarded as sister taxa (Meacham, 1980; Peng et al., 2006; Wang et al., 2007a; Furness, 2008), or *Mahonia* as a segregate genus of a paraphyletic *Berberis* (Adhikari et al., 2015), in which case the two should be united under *Berberis* (Laferriere, 1997; Y. D. Kim et al., 2004). Six shared pollen character state transitions inferred in this study confirm the close relationship between *Berberis* and *Mahonia*: to apolar pollen (Character 2; DFP, DML, CFP, and CHB), asymmetrical pollen (Character 3; DHB), single

apertures (Character 8; DHB), spiral ectoapertures (Character 11; DML), protocolumellate infractum structure (Character 18; DFP, DML, and CFP), and foot layer absent (Character 19, all five analyses).

Ranunculaceae was divided by Tamura (1993) into five subfamilies based mainly on fruit and ovule characters, viz., Helleboroideae, Isopyroideae, Ranunculoideae, Thalictroideae, and Hydrastidoideae, a treatment subsequently challenged by W. Wang et al. (2009) based on evidence from DNA sequences and morphological features. W. Wang et al. (2009) proposed an updated classification, dividing Ranunculaceae into five different subfamilies (Glaucidioideae, Hydrastidoideae, Coptidoideae, Thalictroideae,

Table 3. Tests of correlated evolution between ecological and pollen characters under maximum likelihood (ML) and Markov chain Monte Carlo (MCMC) methods.

Trait pair (ecological character and pollen character)	ML				MCMC		
	Likelihood				Harmonic mean of log likelihood		
	Dependent model	Independent model	Likelihood ratio (LR)	<i>P</i> value	Dependent model	Independent model	Log Bayes factor (LBF)
Anemophily/aperture number ≥ 4	-132.71669	-137.025171	8.616962	0.071	-143.451908	-143.072679	-0.758458
Herbaceous/spheroidal shape	-122.12716	-130.630644	17.006968	0.002**	-130.539705	-138.065634	15.051858**
Arborescent/oblate shape	-85.741066	-92.675025	13.867918	0.008*	-96.086758	-100.786388	9.39926*

* $P < 0.01$, LBF > 5 .** $P < 0.005$, LBF > 10 .

and Ranunculioideae). Of the groups defined by W. Wang et al. (2009), the monophyly of Coptidoideae is supported by five pollen character state changes in our results: transitions to apolarity (Character 2), globally distributed apertures (Character 9), and ectoapertures porate (Character 11), each under all five methods; small-sized pollen grains (Character 7; DHB and CHB); and more than six apertures (Character 8; DFP, DML, CFP, and CHB).

Proteales and Sabiaceae

Within Proteaceae, tribe Banksieae is recognized as a clade from morphological and molecular evidence (Johnson & Briggs, 1975; Douglas, 1995; Weston & Barker, 2006; Sauquet et al., 2009a). The pollen morphology of this clade is unusual compared with other genera in Proteaceae (see Appendices 3 and 4), and it is defined by four pollen character state changes: to bilateral symmetry (Character 3), elliptical outline in polar view (Character 6), and double apertures (Character 8), all under DFP, DML, and CFP inference; and small size (Character 7) under CHB.

Until recently, support for the placement of Sabiaceae remained weak (Moore et al., 2010; Soltis et al., 2011; Barniske et al., 2012). However, in APG IV (2016) it has been classified in Proteales on the basis of strong molecular support found by Sun et al.

(2016). Based on our optimization of pollen characters, the monophyly of Sabiaceae is supported by six character states: pollen shape class (Character 5) spheroidal (CHB) or subprolate (DFP and DML), pollen small (Character 7; CHB), aperture structure (Character 10) compound (DFP, DML, CFP, DHB, and CHB), endoapertures (Character 12) alonate (DFP, DML, and DHB) or circular (CHB), tectum sculpture (Character 17) reticulate (DHB) or perforate (CHB), and endexine present (Character 20; DHB and CHB). Two pollen character state transitions were inferred on the branch leading to Sabiaceae-Proteales, viz., pollen shape class to subprolate (DHB) or prolate (CHB) (Character 5) and tectum sculpture to perforate (Character 17; DHB). However, Sabiaceae may be distinguished from Proteales by pollen grains with compound apertures (Character 10; DFP, DML, CFP, DHB, and CHB).

Buxales

In APG III (2009), Buxales was treated as two families, Buxaceae (including Didymelaceae) and Haptanthaceae. Didymelaceae (von Balthazar et al., 2003; Köhler, 2007b) has been estimated as sister to Buxaceae using both morphological and molecular evidence (von Balthazar & Endress 2002; von Balthazar et al., 2003; Worberg et al., 2007), while APG III (2009) did not recognize the family

Table 4. Transition rates for states of each of the two pairs of correlated traits from the maximum likelihood analysis under a dependent model.

Trait pair	Transition rate							
	q12	q13	q21	q24	q31	q34	q42	q43
Herbaceous/spheroidal shape	15.47	2.84	62.35	0	8.34	9.49	0	12.32
Arborescent/oblate shape	1.83	19.86	3.25	12.67	76.07	15.20	0.77	0.30

Table 5. Likelihood ratio tests of specific hypotheses for each of the two pairs of correlated traits.

Trait pair	Restriction	Hypothesis	<i>P</i> value
Herbaceous/ spheroidal shape	q12 = q34	shape class changing from spheroidal to non-spheroidal does not depend on the state of plant growth form	0.5424
	q21 = q43	shape class changing from non-spheroidal to spheroidal does not depend on the state of plant growth form	0.0714
	q13 = q24	plant growth form changing from herbaceous to non-herbaceous does not depend on the state of shape class	0.1507
	q31 = q42	plant growth form changing from non-herbaceous to herbaceous does not depend on the state of shape class	0.0085*
	q12 = 0	shape class changing from spheroidal to non-spheroidal does not occur when plant growth form is herbaceous	$9.478 \times 10^{-6**}$
	q21 = 0	shape class changing from non-spheroidal to spheroidal does not occur when plant growth form is herbaceous	0.0035**
	q13 = 0	plant growth form changing from herbaceous to non-herbaceous does not occur when shape class is spheroidal	$2.781 \times 10^{-5**}$
	q31 = 0	plant growth form changing from non-herbaceous to herbaceous does not occur when shape class is spheroidal	0.0068*
	q34 = 0	shape class changing from spheroidal to non-spheroidal does not occur when plant growth form is non-herbaceous	$9.507 \times 10^{-5**}$
	q43 = 0	shape class changing from non-spheroidal to spheroidal does not occur when plant growth form is non-herbaceous	$4.904 \times 10^{-14**}$
Arborescent/ oblate shape	q12 = q34	shape class changing from oblate to non-oblate does not depend on the state of plant growth form	0.0938
	q21 = q43	shape class changing from non-oblate to oblate does not depend on the state of plant growth form	0.2448
	q13 = q24	plant growth form changing from arborescent to non-arborescent does not depend on the state of shape class	0.5237
	q31 = q42	plant growth form changing from non-arborescent to arborescent does not depend on the state of shape class	0.0082*
	q12 = 0	shape class changing from oblate to non-oblate does not occur when plant growth form is arborescent	$9.627 \times 10^{-5**}$
	q21 = 0	shape class changing from non-oblate to oblate does not occur when plant growth form is arborescent	0.0619
	q13 = 0	plant growth form changing from arborescent to non-arborescent does not occur when shape class is oblate	0.0004**
	q31 = 0	plant growth form changing from non-arborescent to arborescent does not occur when shape class is oblate	$2.752 \times 10^{-6**}$
	q24 = 0	plant growth form changing from arborescent to non-arborescent does not occur when shape class is non-oblate	0.0003**
	q42 = 0	plant growth form changing from non-arborescent to arborescent does not occur when shape class is non-oblate	0.0019**
q34 = 0	shape class changing from oblate to non-oblate does not occur when plant growth form is non-arborescent	0.0062*	
q43 = 0	shape class changing from non-oblate to oblate does not occur when plant growth form is non-arborescent	0.5293	

* *P* < 0.01.

** *P* < 0.005.

Didymelaceae but placed it in a broadly circumscribed Buxaceae. This Buxaceae s.l. is supported by eight pollen character state transitions: to apolar (Character 2) and medium-sized (Character 7) grains with more than six apertures (Character 8), globally distributed (Character 9), and lolongate endoaperture shape (Character 12), all under CHB; suprategal elements present (Character 14; DFP, DML, and CFP) and echinate (Character 15; DFP and DML);

and tectum crotonoid (Character 17) under DHB and CHB. Meanwhile, the clade of Buxaceae with Didymelaceae excluded is supported by pollen character states including apolar (Character 2) pollen grains with more than six apertures (Character 8) and globally distributed (Character 9), all under DFP and DML; and ectoapertures porate (Character 11; DFP, DML, DHB, and CHB). Since Haptanthaceae was not able to be included in our study, we cannot comment

on the palynological support (or otherwise) for its recent placement within Buxaceae.

The relationship between Ceratophyllales and eudicots

The systematic significance of pollen morphology regarding the various alternative phylogenetic relationships of Ceratophyllales, monocots, basal angiosperms, and basal eudicots has been discussed in previous studies (reviewed by Lu et al., 2015; Luo et al., 2015). In the present study, four pollen character state transitions, including to basic shape globose (Character 4; DHB and CHB), outline in polar view circular (Character 6; DML, DFP, CFP, DHB, and CHB), aperture number (Character 8) three (DML and CHB) or more than six (DHB), and aperture position equatorial (Character 9; DHB and CHB), were inferred at the root of the branch leading to Ceratophyllales and basal eudicots, consistent with the results of Lu et al. (2015).

POLLEN MORPHOLOGICAL EVOLUTION IN BASAL EU DICOTS

Of the 20 characters studied, optimized using CHB inference (taken, as in previous papers, as an example for this discussion; see Fig. 8), several were highly conserved: basic shape (Character 4) and foot layer presence/absence (Character 19) both changed twice, and dispersal unit (Character 1), symmetry (Character 3), and operculum presence/absence (Character 13) changed only once across all basal eudicot taxa studied. With the exception of operculum presence/absence, the above characters have also been found to be conservative elsewhere in angiosperms (Wortley et al., 2015), including dispersal unit and foot layer in monocots (Luo et al., 2015) and basal angiosperms (Lu et al., 2015), and symmetry within eudicots (Wortley et al., 2015). By contrast, in the present study, large numbers of state changes were observed in pollen size (Character 7, changed 37 times), shape class (Character 5, changed 32 times), and tectum sculpture (Character 17, changed 32 times). Pollen size and tectum sculpture have also been found to display the most frequent changes in the other groups of angiosperms studied in this series so far (e.g., basal angiosperms [Lu et al., 2015] and monocots [Luo et al., 2015]).

General evolutionary trends in angiosperm pollen characters (reviewed by Punt, 1975) have previously been considered to include increasing grain size; increasing aperture number, accompanied by a shift in aperture position from equatorial to global (reviewed by van Campo, 1976; Furness & Rudall, 2004) and a change in aperture shape from colpate to porate; and increasing complexity of tectum ornamentation (reviewed by Walker & Doyle, 1975). In

the present study, these evolutionary trends were confirmed in basal eudicots, with the exception of pollen size (Fig. 8).

In terms of pollen size (Character 7), Walker and Doyle (1975) noted that early-diverging angiosperm taxa produce pollen mainly of the large size class, and that pollen size is undoubtedly an easily reversible feature. Across our five analyses, medium size was inferred to be the plesiomorphic state for basal eudicots, with small and large size both evolving many times in different lineages within the group (Fig. 8), i.e., a similar evolutionary pattern to that within angiosperms as a whole (Wortley et al., 2015). Pollen size as a phenotype has been found to be affected by chromosome ploidy level, environmental factors, and flower characters, among others (Muller, 1979; Stroo, 2000). It therefore might not be feasible to seek a general evolutionary trend in pollen size across angiosperms as a whole.

Our optimization of aperture number (Character 8) indicated a transformation from one to three occurred at the root of the eudicot clade, with the three apertures having an equatorial distribution across the pollen grain surface. It has been agreed that the monophyletic eudicots are characterized by three equatorially arranged apertures (Furness & Rudall, 2004), while the pollen of basal angiosperms and monocots is predominantly characterized by monosulcate or monosulcate-derived apertures situated at the distal pole (Walker, 1974; Furness & Rudall, 2004; Harley, 2004; Lu et al., 2015; Luo et al., 2015). Although the tri-aperturate type also occurs sporadically in monocots and some magnoliid dicots (Harley, 2004), these types do not appear to be homologous with those of eudicots, their derivation following Garside's rule (in the development of tetrahedral tetrads of tri-aperturate pollen grains, apertures are formed in groups of three at four points in the developing tetrad [Garside, 1946]) compared to those of eudicots, which are commonly arranged according to Fischer's rule (apertures formed in pairs at six points in the developing tetrad [Fischer, 1890; Doyle et al., 1990]). Garside's rule has also, interestingly, been observed in Proteaceae (Blackmore & Barnes, 1995) and Olacaceae (Maguire et al., 1974).

The shift in pollen aperture features from distal monosulcate to equatorial tri-aperturate has long been regarded as a key innovation in the evolutionary history of seed plants (Muller, 1970; Walker & Doyle, 1975; Blackmore & Crane, 1998; Furness & Rudall, 2004). It still remains obscure how the transition from bilaterally symmetrical, distally monosulcate pollen grains to radially symmetrical, equatorially arranged

tri-aperturate pollen grains was effected: these two types differ greatly and no transitional forms have been found (Kuprianova, 1979; Harley, 2004). Blackmore and Crane (1998) proposed that the transformation within the angiosperms from distal to equatorial apertures appears to be correlated with evolutionary changes in the timing and processes of meiosis. Blackmore and Crane (1998), followed by Ressayre et al. (2002) and Ressayre et al. (2005), indicated that aperture pattern is determined by the sites where cytokinesis following meiosis is completed and primexine is not deposited, and these sites themselves are determined by a number of events (including cytokinesis type, orientation of the second meiotic axis that affects tetrad shape, and pattern of callose deposition following cell plate formation) that individually appear to be controlled by at least one gene. Further studies of ontogenesis might provide some clues to understanding this transformation.

Pollen with simple apertures was consistently inferred as the plesiomorphic state for basal eudicots under all five methods of inference, while compound apertures evolved in Buxaceae, Menispermaceae, and Sabiaceae (Appendix 3; Fig. 8). The basal eudicots are the first branch of the angiosperm tree in which we see the occurrence of endoapertures. Harley (2004) and Furness et al. (2007) agree that endoapertures do not exist either in monocots or in the basal angiosperms but occur commonly in eudicots, and Furness et al. (2007) proposed that endoapertures may have been selected in eudicots because they favor harmomegathy by providing areas of more flexible exine. Blackmore and Crane (1998) described endoapertures as being formed during development where the presence of onci disrupts endexine formation. Onci were revealed to be related to germination and early pollen tube growth (Heslop-Harrison et al., 1986). Thus, it is possible that endoapertures are associated with pollen tube germination and emergence, along with a harmomegathic role.

ADAPTIVE EVOLUTION IN BASAL EUDICOT POLLEN CHARACTERS

The adaptive evolution of angiosperm pollen characters has been repeatedly discussed, several hypotheses have been proposed, and possible relationships have been observed between pollination strategies and pollen features (Muller, 1979; Linder, 1998, 2000; G. Wang et al., 2014; Luo et al., 2015). Regarding pollen size, for example, Muller (1979) suggested that, typically, wind-pollinated grains fall into a 20–40 μm range, with larger grains mostly transported by animal pollinators and very small ones often associated with highly specialized pollination

systems. According to our CHB results (Fig. 8), pollen size (Character 7) showed the most frequent state changes (37) of all 20 studied characters, and these changes occurred more frequently in Proteaceae than in other basal eudicot lineages (21 of the 37 times). Interestingly, pollination syndrome is also much more diverse in Proteaceae than in other basal eudicot taxa, with pollinators including birds, such as honeyeaters, lorikeets, silvereyes, sugarbirds, and sunbirds (Ford et al., 1979; Collins & Rebelo, 1987; Midgley, 1987; Maynard, 1995); mammals, such as bats, marsupials, and rodents (Collins & Rebelo, 1987; Maynard, 1995); and insects, such as bees, beetles, moths, butterflies, and flies (Collins & Rebelo, 1987; Maynard, 1995). Anemophily has also been reported (e.g., in *Leucadendron* R. Br.; Midgley, 1987). Our results therefore suggest a possible hypothesis that different-sized pollen grains correspond to different pollination strategies, as suggested by Muller (1979). Within Proteaceae, our results indicate that large-size pollen is a synapomorphy of the well-supported monophyletic clade Embothriinae, all four genera of which are reported to be mainly bird-pollinated (Maynard, 1995; Barker et al., 2007). Furthermore, the estimated date of the radiation of Embothriinae is 52.8 ± 6.8 million years ago (Ma) (around the Early Eocene; Barker et al., 2007), corresponding approximately with the origin of the Meliphagoidea, the order containing the nectar-feeding Meliphagidae, which may have diverged in Australia in the Early Eocene (Barker et al., 2004). However, it should be noted that next-generation DNA data have recently suggested the Meliphagoidea instead originated around the Early Oligocene (Prum et al., 2015). Due to these disputed dates, it is difficult to draw strong conclusions regarding the relationship between variation in pollination syndrome and pollen size in Proteaceae. However, our results suggest possible avenues for further research to test this correlation within Proteaceae.

Tectum sculpture (Character 17) has also been highlighted as associated with pollination strategy. For example, entomophilous (or zoophilous) flowers often appear to produce pollen grains with an elaborate, ornamented exine surface, while psilate pollen grains might be characteristic of anemophilous and hydrophilous plants (Whitehead, 1969; Hu et al., 2008). Sannier et al. (2009) suggested that the relationship between pollen grain ornamentation and pollination may vary between taxonomic groups. An association between pollen tectum sculpture and pollination syndrome has been confirmed in the nitrogen-fixing clade (He et al., in prep.). In the present study, tectum sculpture not only displays

frequent state transformations but also the most numerous (nine) character states of all the 20 characters studied (Appendix 3; Fig. 8). The exine plays a vital role in the interaction of pollen grains with both pollinators and the receptive part of the stigma (Heslop-Harrison, 1976; Muller, 1979). The diversity of tectum sculpture and frequency of state changes seen in basal eudicots may be attributed to the variety of pollination strategies in the group (Appendix 3) (Kubitzki et al., 1993; Bosch et al., 1997; Kubitzki, 2007; Hu et al., 2008). However, although such an association has been widely accepted, we found no positive statistical correlation between tectum sculpture and pollination strategy.

Pollen shape class (Character 5) showed high variability throughout basal eudicot evolution (Fig. 8). In a correlation test between pollen shape class and plant growth form, both ML and MCMC inference indicated a very strong correlation between herbaceous form and spheroidal pollen grain shape ($P = 0.002$, LBF value = 15.052), and between arborescent form and oblate pollen ($P = 0.008$, LBF value = 9.399). Based on a further test, transitions in plant growth form from non-herbaceous to herbaceous and from non-arborescent to arborescent were both inferred to depend on pollen shape class. Analysis showed that, except for q21 and q43 for the pair “arborescent growth form and oblate pollen shape class,” all transition rates were significantly different from zero. This suggests that, in general, transition rates in the characters pollen shape class and plant growth form might be affected by one another, although the directions of the transitions were less closely linked. To our knowledge, this is the first time such a result has been found. A direct explanation for the correlation between pollen shape class and plant growth form remains unclear; however, various hypotheses are plausible: plant life form has previously been found to be correlated with breeding system features such as pollen grain number, ovule number, and pollen to ovule ratio (Jürgens et al., 2002). Pollen to ovule ratios are also suggested to be associated with pollination mode (anemophily and entomophily; Michalski & Durka, 2009). Thus, breeding system features might provide a link between plant growth form and pollination mode, which is considered a potential factor that may influence pollen morphological evolution of the angiosperms (Muller, 1979).

Both Sun et al. (2011) and W. Wang et al. (2014) have suggested that the basal eudicots were already present and might have experienced an accelerated evolution and diversification during the latest

Barremian and earliest Aptian stages of the Early Cretaceous. In our CHB analysis, the most concentrated pollen character state changes were estimated to occur mainly at the roots of Buxales, Papaveraceae, Proteaceae, and Sabiaceae, and within Papaveraceae (Fig. 8). The origins of these taxa all lie close to the Barremian to Albian stage (100.5 Ma to ~129.4 Ma; International Chronostratigraphic Chart, 2015, <<http://www.stratigraphy.org>>), with the stem ages of Buxales estimated at about 117–128.6 Ma, Papaveraceae at about 114–132.88 Ma, Proteaceae at about 94.81–110 Ma, and Sabiaceae at about 118–130.76 Ma (Anderson et al., 2005; Magallón et al., 2015; Tank et al., 2015), while Hoot and Douglas (1998) suggested that the Proteaceae might have begun to diverge in the Albian and to diversify by the Middle Cretaceous. Thus, many pollen state changes within basal eudicots may correspond to times of rapid taxonomic diversification.

The unstable climate of the Cretaceous (with temperature changes, sea level fluctuations, and an ocean anoxic event in the middle of the period) may be one of the main factors affecting the early evolution of angiosperms (Coiffard et al., 2007). It is thought that the core angiosperms arose on Albian floodplains under warmer and drier conditions, and tricolpate pollen became regularly present and more diverse in the Middle and Late Albian (Hickey & Doyle, 1977; Coiffard et al., 2007). It has also been shown in monocots that the most frequent state changes in pollen characters occurred during the Albian stage (Luo et al., 2015). We consider it likely that the dramatically changing climate and geography of the Early Cretaceous might also have influenced the pollen morphology of basal eudicots.

CONCLUSIONS AND FUTURE PROSPECTS

In this paper, using three different methods of inference on two differently coded datasets, we reconstruct the distribution of pollen character states for basal eudicots on a molecular phylogenetic tree. We identify evolutionary patterns, infer possible plesiomorphic and diagnostic character states, and note the state changes that are congruent with key branches in the molecular phylogeny. We reconfirm some previously postulated evolutionary trends, including an increase in aperture number, increase in pollen exine ornamentation complexity, and change in aperture shape from colpate to porate. In addition, the basal eudicots represent the first angiosperm lineage to display endoapertures. We infer that plant growth form is significantly associated with pollen shape class. Ecological factors such as past climate, habitat, and pollinator shifts are likely

to exert selective pressures on pollen characters and favor certain character state transformations. Future studies may elucidate the underlying mechanisms of these transformations from the respect of evolution of development (Hall, 2003). Further investigation may also provide more detailed interpretations of the morphological diversity in basal eudicots and variation in angiosperm pollen.

Literature Cited

- Adhikari, B., R. Milne, R. T. Pennington, T. Särkinen & C. A. Pendry. 2015. Systematics and biogeography of *Berberis* s.l. inferred from nuclear ITS and chloroplast *ndhF* gene sequences. *Taxon* 64: 39–48.
- Allen, C. R., A. S. Garmestani, J. A. LaBram, A. E. Peck & L. B. Prevost. 2006. When landscaping goes bad: The incipient invasion of *Mahonia bealei* in the southeastern United States. *Biol. Invasions* 8: 169–176.
- Anderson, C. L., K. Bremer & E. M. Friis. 2005. Dating phylogenetically basal eudicots using *rbcL* sequences and multiple fossil reference points. *Amer. J. Bot.* 92: 1737–1748.
- APG (Angiosperm Phylogeny Group) III. 2009. An update of the Angiosperm Phylogeny Group classification for the orders and families of flowering plants: APG III. *Bot. J. Linn. Soc.* 161: 105–121.
- APG (Angiosperm Phylogeny Group) IV. 2016. An update of the Angiosperm Phylogeny Group classification for the orders and families of flowering plants: APG IV. *Bot. J. Linn. Soc.* 181: 1–20.
- Argue, C. L. 1974. Pollen studies in the Alismataceae (Alismaceae). *Bot. Gaz.* 135: 338–344.
- Armstrong, J. A. 1979. Biotic pollination mechanisms in the Australian flora—A review. *New Zealand J. Bot.* 17: 467–508.
- Bailey, I. W. & C. G. Nast. 1945. Morphology and relationships of *Trochodendron* and *Tetracentron*, I. Stem, root, and leaf. *J. Arnold Arbor.* 26: 143–154.
- Baladehi, M. H., M. Habibi & D. Azizian. 2013. Pollen morphology of Iranian species of *Anemone* L. & *Pulsatilla* Mill. (Ranunculaceae) and its implications. *Acta Bot. Gallica* 160: 19–26.
- Banks, H., P. Stafford & P. R. Crane. 2007. Aperture variation in the pollen of *Nelumbo* (Nelumbonaceae). *Grana* 46: 157–162.
- Barker, F. K., A. Cibois, P. Schikler, J. Feinstein & J. Cracraft. 2004. Phylogeny and diversification of the largest avian radiation. *Proc. Natl. Acad. Sci. U.S.A.* 101: 11040–11045.
- Barker, N. P., P. H. Weston, J. P. Rourke & G. Reeves. 2002. The relationships of the southern African Proteaceae as elucidated by internal transcribed spacer (ITS) DNA sequence data. *Kew Bull.* 57: 867–883.
- Barker, N. P., P. H. Weston, F. Rutschmann & H. Sauquet. 2007. Molecular dating of the ‘Gondwanan’ plant family Proteaceae is only partially congruent with the timing of the break-up of Gondwana. *J. Biogeogr.* 34: 2012–2027.
- Barniske, A. M., T. Borsch, K. Müller, M. Krug, A. Worberg, C. Neinhuis & D. Quandt. 2012. Phylogenetics of early branching eudicots: Comparing phylogenetic signal across plastid introns, spacers, and genes. *J. Syst. Evol.* 50: 85–108.
- Behnke, H.-D. 1982. Sieve-element plastids, exine sculpturing and the systematic affinities of the Buxaceae. *Pl. Syst. Evol.* 139: 257–266.
- Bhattacharya, A., S. Mondal & S. Mandal. 1999. Entomophilous pollen incidence with reference to atmospheric dispersal in eastern India. *Aerobiologia* 15: 311–315.
- Blackmore, S. 2007. Pollen and spores: Microscopic keys to understanding the earth’s biodiversity. *Pl. Syst. Evol.* 263: 3–12.
- Blackmore, S. & G. L. A. Heath. 1984. Berberidaceae. The northwest European pollen flora, 30. *Rev. Palaeobot. Palynol.* 42: 7–21.
- Blackmore, S. & S. H. Barnes. 1995. Garside’s rule and the microspore tetrads of *Grevillea rosmarinifolia* A. Cunningham and *Dryandra polycephala* Benth (Proteaceae). *Rev. Palaeobot. Palynol.* 85: 111–121.
- Blackmore, S. & P. Crane. 1998. The evolution of apertures in the spores and pollen grains of embryophytes. Pp. 159–182 in S. J. Owens & P. J. Rudall (editors), *Reproductive Biology*. Royal Botanic Gardens, Kew.
- Blackmore, S., P. Stafford & V. Persson. 1995. Palynology and systematics of Ranunculiflorae. *Pl. Syst. Evol.* 9 (suppl.): 71–82.
- Blattner, F. R. & J. W. Kadereit. 1999. Morphological evolution and ecological diversification of the forest-dwelling poppies (Papaveraceae: Chelidonioideae) as deduced from a molecular phylogeny of the ITS region. *Pl. Syst. Evol.* 219: 181–197.
- Bogner, J. & S. J. Mayo. 1998. Acoraceae. Pp. 7–11 in K. Kubitzki (editor), *The Families and Genera of Vascular Plants*, Vol. IV. Springer, Berlin.
- Bosch, M. & N. M. Waser. 1999. Effects of local density on pollination and reproduction in *Delphinium nuttallianum* and *Aconitum columbianum* (Ranunculaceae). *Amer. J. Bot.* 86: 871–879.
- Bosch, M., J. Simon, C. Blanché & J. Molero. 1997. Pollination ecology in tribe Delphineae (Ranunculaceae) in W Mediterranean area: Floral visitors and pollinator behaviour. *Lagascalia* 19: 545–562.
- Brett, J. F. & U. Posluszny. 1982. Floral development in *Caulophyllum thalictroides* (Berberidaceae). *Canad. J. Bot.* 60: 2133–2141.
- Brown, R. 1811. On the Proteaceae of Jussieu. *Trans. Linn. Soc. London* 18: 36–64.
- Brückner, P. 1993. Pollen morphology and taxonomy of Eurasian species of the genus *Buxus* (Buxaceae). *Grana* 32: 65–78.
- Burleigh, J. G., K. W. Hilu & D. E. Soltis. 2009. Inferring phylogenies with incomplete data sets: A 5-gene, 567-taxon analysis of angiosperms. *BMC Evol. Biol.* 9: 61.
- Buzgo, M. & P. K. Endress. 2000. Floral structure and development of Acoraceae and its systematic relationships with basal angiosperms. *Int. J. Pl. Sci.* 161: 23–41.
- Cantino, P. D., J. A. Doyle, S. W. Graham, W. S. Judd, R. G. Olmstead, D. E. Soltis, P. S. Soltis & M. J. Donoghue. 2007. Towards a phylogenetic nomenclature of *Tracheophyta*. *Taxon* 56: 822–846.
- Canto, A., C. M. Herrera, M. Medrano, R. Pérez & I. M. García. 2008. Pollinator foraging modifies nectar sugar composition in *Helleborus foetidus* (Ranunculaceae): An experimental test. *Amer. J. Bot.* 95: 315–320.
- Carthew, S. M. 1994. Foraging behaviour of marsupial pollinators in a population of *Banksia spinulosa*. *Oikos* 133–139.
- Chalcoff, V. R., M. A. Aizen & C. Ezcurra. 2012. Erosion of a pollination mutualism along an environmental gradient

- in a south Andean treelet, *Embothrium coccineum* (Proteaceae). *Oikos* 121: 471–480.
- Chanda, S., S. Nilsson & S. Blackmore. 1988. Phylogenetic trends in the Alismatales with reference to pollen grains. *Grana* 27: 257–272.
- Chang, K.-T. & P.-L. Wang. 1983. Study on the pollen morphology of the family Berberidaceae. *Acta Phytotax. Sin.* 21: 130–142.
- Chase, M. W., D. E. Soltis, R. G. Olmstead, D. Morgan, D. H. Les, B. D. Mishler, M. R. Duvall, R. A. Price, H. G. Hills, Y.-L. Qiu, K. A. Kron, J. H. Rettig, E. Conti, J. D. Palmer, J. R. Manhart, K. J. Sytsma, H. J. Michaels, W. J. Kress, K. G. Karol, W. D. Clark, M. Hedrén, B. S. Gaut, R. K. Jansen, K. J. Kim, C. F. Wimpee, J. F. Smith, G. R. Furnier, S. H. Strauss, Q.-Y. Xiang, G. M. Plunkett, P. S. Soltis, S. M. Swensen, S. E. Williams, P. A. Gadek, C. J. Quinn, L. E. Eguarte, E. Golenberg, G. H. Learn, S. W. Graham, S. C. H. Barrett, S. Dayanandan & V. A. Albert. 1993. Phylogenetics of seed plants: An analysis of nucleotide sequences from the plastid gene *rbcl*. *Ann. Missouri Bot. Gard.* 80: 528–580.
- Christenhusz, M. J. M. 2012. An overview of Lardizabala-ceae. *Curtis's Bot. Mag.* 29: 235–276.
- Clarke, G. C. S., W. Punt & P. P. Hoen. 1991. Ranunculaceae. The northwest European pollen flora, 51. *Rev. Palaeobot. Palynol.* 69: 117–271.
- Coiffard, C., B. Gomez & F. Thévenard. 2007. Early Cretaceous angiosperm invasion of western Europe and major environmental changes. *Ann. Bot. (Oxford)* 100: 545–553.
- Collins, B. G. & T. Rebelo. 1987. Pollination biology of the Proteaceae in Australia and southern Africa. *Austral. J. Ecol.* 12: 387–421.
- Cunningham, C. W., K. E. Omland & T. H. Oakley. 1998. Reconstructing ancestral character states: A critical reappraisal. *Trends Ecol. Evol.* 13: 361–366.
- Dahl, Å. E. 1989. Taxonomic and morphological studies in *Hypecoum* sect. *Hypecoum* (Papaveraceae). *Pl. Syst. Evol.* 163: 227–280.
- Davis, S. L. 1997. Stamens are not essential as an attractant for pollinators in females of cryptically dioecious *Thalictrum pubescens* Pursch. (Ranunculaceae). *Sexual Pl. Reprod.* 10: 293–299.
- Denisow, B. & M. Wrzesień. 2006. The study of blooming and pollen efficiency of *Adonis vernalis* L. in xerothermic plant communities. *J. Apic. Sci.* 50: 25–32.
- Denk, T. & M. V. Tekleva. 2006. Comparative pollen morphology and ultrastructure of *Platanus*: Implications for phylogeny and evaluation of the fossil record. *Grana* 45: 195–221.
- Dettmann, M. E. 1998. Pollen morphology of Eidotheoi-deae: Implications for phylogeny in the Proteaceae. *Austral. Syst. Bot.* 11: 605–612.
- Dettmann, M. E. & D. M. Jarzen. 1998. The early history of the Proteaceae in Australia: The pollen record. *Austral. Syst. Bot.* 11: 401–438.
- Díaz Lifante, Z., M. J. Díez & I. Fernández. 1990. Morfología polínica de las subfamilias Melanthioideae y Asphodeloideae (Liliaceae) en la Península Ibérica y su importancia taxonomica. *Lagascalia* 16: 211–225.
- Díez, M. J., S. Talavera & P. García-Murillo. 1988. Contributions to the palynology of hydrophytic, non-entomophilous angiosperms. I. Studies with LM and SEM. *Candollea* 43: 147–158.
- Dohzono, I. & K. Suzuki. 2002. Bumblebee-pollination and temporal change of the calyx tube length in *Clematis stans* (Ranunculaceae). *J. Plant Res.* 115: 355–359.
- Donoghue, M. J. & J. A. Doyle. 1989. Phylogenetic analysis of angiosperms and the relationships of Hamamelidaceae. Pp. 17–45 in P. R. Crane & S. Blackmore (editors), *Evolution, Systematics, and Fossil History of the Hamamelidaceae*, Vol. 1. Clarendon, Oxford.
- Douglas, A. W. 1995. Affinities. Pp. 6–14 in P. McCarthy (editor), *Flora of Australia*, Vol. 16. Elaeagnaceae, Proteaceae I. Australian Biological Resources Study/CSIRO Publishing, Melbourne.
- Doyle, J. A. & C. L. Hotton. 1991. Diversification of early angiosperm pollen in a cladistic context. Pp. 169–195 in S. Blackmore & S. H. Barnes (editors), *Pollen and Spores: Patterns of Diversification*. Clarendon Press, Oxford.
- Doyle, J. A., C. L. Hotton & J. V. Ward. 1990. Early Cretaceous tetrads, zonulculate pollen, and Winteraceae. I. Taxonomy, morphology, and ultrastructure. *Amer. J. Bot.* 77: 1544–1557.
- Drinnan, A. N., P. R. Crane & S. B. Hoot. 1994. Patterns of floral evolution in the early diversification of non-magnoliid dicotyledons (eudicots). *Pl. Syst. Evol.* 8 (suppl.): 93–122.
- Eckert, C. & A. Schaefer. 1998. Does self-pollination provide reproductive assurance in *Aquilegia canadensis* (Ranunculaceae)? *Amer. J. Bot.* 85: 919–924.
- Endress, P. K. 1989. Aspects of evolutionary differentiation of the Hamamelidaceae and the Lower Hamamelidaceae. *Pl. Syst. Evol.* 162: 193–211.
- Endress, P. K. 1997. Relationships between floral organization, architecture, and pollination mode in *Dillenia* (Dilleniaceae). *Pl. Syst. Evol.* 206: 99–118.
- Endress, P. K. 2010. Flower structure and trends of evolution in Eudicots and their major subclades. *Ann. Missouri Bot. Gard.* 97: 541–583.
- Endress, P. K. & A. Igersheim. 1999. Gynoecium diversity and systematics of the basal eudicots. *Bot. J. Linn. Soc.* 130: 305–393.
- Endress, P. K. & J. A. Doyle. 2009. Reconstructing the ancestral angiosperm flower and its initial specializations. *Amer. J. Bot.* 96: 22–66.
- Erdtman, G. 1952. Pollen Morphology and Plant Taxonomy. Angiosperms. An Introduction to Palynology I. Almqvist & Wiksell, Stockholm.
- Erdtman, G. 1960. The acetolysis method, a revised description. *Svensk Bot. Tidskr.* 54: 561–564.
- Fægri, K. 1956. Recent trends in palynology. *Bot. Rev. (Lancaster)* 22: 639–664.
- Fægri, K. & J. Iversen. 1989. *Textbook of Pollen Analysis*. Munksgaard, Copenhagen.
- Fay, M. F. & M. J. Christenhusz. 2012. Ranunculales—Buttercups, poppies and their relatives. *Curtis's Bot. Mag.* 29: 222–234.
- Fenster, C. B. & S. Martén-Rodríguez. 2007. Reproductive assurance and the evolution of pollination specialization. *Int. J. Pl. Sci.* 168: 215–228.
- Ferguson, I. K. 1975. Pollen morphology of the tribe Triclisieae of the Menispermaceae in relation to its taxonomy. *Kew Bull.* 30: 49–75.
- Ferguson, I. K. 1978. Pollen morphology of the tribe Coscinieae of the Menispermaceae in relation to its taxonomy. *Kew Bull.* 32: 339–346.
- Ferguson, I. K. 1985. The role of pollen morphology in plant systematics. *Anales Asoc. Palin. Langua Esp.* 2: 5–18.

- Fernández, I. 1986a. Contribución al conocimiento palinológico de la familia Ranunculaceae en Andalucía. I Subf. Helleboroideae. *Lagascalia* 14: 13–23.
- Fernández, I. 1986b. Contribución al conocimiento palinológico de la familia Ranunculaceae en Andalucía. II: Subfam. Ranunculoidea. *Candollea* 41: 75–85.
- Feuer, S. 1986. Pollen morphology and evolution in the Persoonioideae, Sphalmioideae and Carnarvonioideae (Proteaceae). *Pollen & Spores* 28: 123–155.
- Feuer, S. 1989. Pollen morphology of Embotriaceae (Proteaceae). *Grana* 28: 225–242.
- Feuer, S. 1990. Pollen morphology of the Embotriaceae (Proteaceae) II. Embotriinae (*Embothrium*, *Oreocallis*, *Telopea*). *Grana* 29: 19–36.
- Fischer, H. 1890. Beiträge zur vergleichenden Morphologie der Pollenkörner. J. U. Kern, Breslau.
- Fitch, W. M. 1971. Toward defining the course of evolution: Minimum change for a specific tree topology. *Syst. Zool.* 20: 406–416.
- Fleming, P. A. & S. W. Nicolson. 2002. How important is the relationship between *Protea humiflora* (Proteaceae) and its non-flying mammal pollinators? *Oecologia* 132: 361–368.
- Ford, H. A., D. C. Paton & N. Forde. 1979. Birds as pollinators of Australian plants. *New Zealand J. Bot.* 17: 509–519.
- Forest, F. & M. W. Chase. 2009. Eudicots. Pp. 169–176 in S. B. Hedges & S. Kumar (editors), *The Timetree of Life*. Oxford University Press, New York.
- Friedman, J. & S. C. H. Barrett. 2008. A phylogenetic analysis of the evolution of wind pollination in the angiosperms. *Int. J. Pl. Sci.* 169: 49–58.
- Fukuda, I. 1967. The biosystematics of *Achlys*. *Taxon* 16: 308–316.
- Furness, C. A. 2007. Why does some pollen lack apertures? A review of inaperturate pollen in eudicots. *Bot. J. Linn. Soc.* 155: 29–48.
- Furness, C. A. 2008. Successive microsporogenesis in eudicots, with particular reference to Berberidaceae (Ranunculales). *Pl. Syst. Evol.* 273: 211–223.
- Furness, C. A. & P. J. Rudall. 2004. Pollen aperture evolution—A crucial factor for eudicot success? *Trends Pl. Sci.* 9: 154–158.
- Furness, C. A. & H. Banks. 2010. Pollen evolution in the early-divergent Monocot order Alismatales. *Int. J. Pl. Sci.* 171: 713–739.
- Furness, C. A., S. Magallón & P. J. Rudall. 2007. Evolution of endoapertures in early-divergent eudicots, with particular reference to pollen morphology in Sabiaceae. *Pl. Syst. Evol.* 263: 77–92.
- Garside, S. 1946. The developmental morphology of the pollen of Proteaceae. *S. African J. Bot.* 12: 27–34.
- Ghorbani, N. M., D. Azizian, M. Sheydaei & M. Khatamsaz. 2008. Pollen morphology of some *Adonis* L. species (Ranunculaceae) from Iran. *Iran. J. Bot.* 14: 165–170.
- Goldingay, R. L. & R. J. Whelan. 1993. The influence of pollinators on fruit positioning in the Australian shrub *Telopea speciosissima* (Proteaceae). *Oikos* 68: 501–509.
- Grayum, M. H. 1992. Comparative external pollen ultrastructure of the Araceae and putatively related taxa. *Monogr. Syst. Bot. Missouri Bot. Gard.* 43: 1–167.
- Guan, B.-C., C.-X. Fu, Y.-X. Qiu, S.-L. Zhou & H. P. Comes. 2010. Genetic structure and breeding system of a rare understory herb, *Dyosma versipellis* (Berberidaceae), from temperate deciduous forests in China. *Amer. J. Bot.* 97: 111–122.
- Guo, B.-L., P.-G. Xiao & S.-Z. He. 1998. The pollen exine ornamentation of *Epimedium* and its taxonomic significance. *Acta Bot. Yunnan.* 20: 220–224.
- Halbritter, H. & M. Hesse. 1995. The convergent evolution of exine shields in angiosperm pollen. *Grana* 34: 108–119.
- Hall, B. K. 2003. Evo-Devo: Evolutionary developmental mechanisms. *Int. J. Developm. Biol.* 47: 491–496.
- Hannan, G. L. & H. A. Prucher. 1996. Reproductive biology of *Caulophyllum thalictroides* (Berberidaceae), an early flowering perennial of eastern North America. *Amer. Midl. Naturalist.* 136: 267–277.
- Harley, M. M. 1985. Pollen morphology and taxonomy of the tribe Fibraureae (Menispermaceae). *Kew Bull.* 40: 553–565.
- Harley, M. M. 2004. Triaperturate pollen in the monocotyledons: Configurations and conjectures. *Pl. Syst. Evol.* 247: 75–122.
- Harley, M. M. & I. K. Ferguson. 1982. Pollen morphology and taxonomy of the tribe Menispermeae (Menispermaceae). *Kew Bull.* 37: 353–366.
- He, H.-X., X.-L. Zhang & Y. Ren. 2006. Floral variation in tepals, sterile and fertile stamens of *Kingdonia uniflora* (Ranunculaceae) with reference to pollinators and pollination. *Acta Bot. Yunnan.* 28: 371–377.
- Heard, T. A. 1993. Pollinator requirements and flowering patterns of *Macadamia integrifolia*. *Austral. J. Bot.* 41: 491–497.
- Henderson, D. M. 1965. The pollen morphology of *Meconopsis*. *Grana* 6: 191–209.
- Heslop-Harrison, J. 1976. The adaptive significance of the exine. Pp. 27–37 in I. K. Ferguson & J. Muller (editors), *The Evolutionary Significance of the Exine*. Academic Press, London.
- Heslop-Harrison, Y., J. S. Heslop-Harrison & J. Heslop-Harrison. 1986. Germination of *Corylus avellana* L. (hazel) pollen: Hydration and the function of the oncus. *Acta Bot. Neerl.* 35: 265–284.
- Hickey, L. J. & J. A. Doyle. 1977. Early Cretaceous fossil evidence for angiosperm evolution. *Bot. Rev.* 43: 3–104.
- Hilu, K. W., T. Borsch, K. Müller, D. E. Soltis, P. S. Soltis, V. Savolainen, M. W. Chase, M. P. Powell, L. A. Alice, R. Evans, H. Sauquet, C. Neinhuis, T. A. B. Slotta, J. C. Rohwer, C. S. Campbell & L. W. Chatrou. 2003. Angiosperm phylogeny based on *matK* sequence information. *Amer. J. Bot.* 90: 1758–1776.
- Hilu, K. W., C. Black, D. Diouf & J. G. Burleigh. 2008. Phylogenetic signal in *matK* vs. *trnK*: A case study in early diverging eudicots (angiosperms). *Molec. Phylogen. Evol.* 48: 1120–1130.
- Hoot, S. B. & A. W. Douglas. 1998. Phylogeny of the Proteaceae based on *atpB* and *atpB-rbcL* intergenic spacer region sequences. *Austral. Syst. Bot.* 11: 301–320.
- Hoot, S. B., S. Magallón & P. R. Crane. 1999. Phylogeny of basal eudicots based on three molecular data sets: *atpB*, *rbcL*, and 18S nuclear ribosomal DNA sequences. *Ann. Missouri Bot. Gard.* 86: 1–32.
- Hoot, S. B., K. M. Wefferling & J. A. Wulff. 2015. Phylogeny and character evolution of Papaveraceae s.l. (Ranunculales). *Syst. Bot.* 40: 474–488.
- Horovitz, A. 1976. Edaphic factors and flower colour distribution in the Anemoneae (Ranunculaceae). *Pl. Syst. Evol.* 126: 239–242.

- Howell, G. J., A. T. Slater & R. B. Knox. 1993. Secondary pollen presentation in angiosperms and its biological significance. *Austral. J. Bot.* 41: 417–438.
- Hu, S., D. L. Dilcher, D. M. Jarzen & D. W. Taylor. 2008. Early steps of angiosperm-pollinator coevolution. *Proc. Natl. Acad. Sci. U.S.A.* 105: 240–245.
- Huang, S.-Q., Y. Takahashi & A. Dafni. 2002. Why does the flower stalk of *Pulsatilla cernua* (Ranunculaceae) bend during anthesis? *Amer. J. Bot.* 89: 1599–1603.
- Huelsensbeck, J. P. & J. P. Bollback. 2001. Empirical and hierarchical Bayesian estimation of ancestral states. *Syst. Biol.* 50: 351–366.
- Husain, T., R. R. Rao & A. Garg. 1998. *Berberis lycium* Royle complex in India. *Feddes Repert.* 109: 385–392.
- Jacques, F. M. B. & P. Bertolino. 2008. Molecular and morphological phylogeny of Menispermaceae (Ranunculales). *Pl. Syst. Evol.* 274: 83–97.
- Jang, M. Y. & K. Heo. 2005. Reproductive morphology of *Megaleranthis saniculifolia* Ohwi (Ranunculaceae) and its systematic implications. *J. Pl. Biol.* 48: 28–135.
- Jansen, R. K., Z.-Q. Cai, L. A. Raubeson, H. Daniell, C. W. de Pamphilis, J. Leebens-Mack, K. F. Müller, M. Guisinger-Bellian, R. C. Haberle, A. K. Hansen, T. W. Chumley, S. B. Lee, R. Peery, J. R. McNeal, J. V. Kuehl & J. L. Boore. 2007. Analysis of 81 genes from 64 plastid genomes resolves relationships in angiosperms and identifies genome-scale evolutionary patterns. *Proc. Natl. Acad. Sci. U.S.A.* 104: 19369–19374.
- Jeong, J. H., E. H. Kim, W.-H. Guo, K. O. Yoo, D. G. Jo & Z. S. Kim. 2010. Genetic diversity and structure of the endangered species *Megaleranthis saniculifolia* in Korea as revealed by allozyme and ISSR markers. *Pl. Syst. Evol.* 289: 67–76.
- Johnson, L. & B. G. Briggs. 1963. Evolution in the Proteaceae. *Austral. J. Bot.* 11: 21–61.
- Johnson, L. & B. G. Briggs. 1975. On the Proteaceae: The evolution and classification of a southern family. *Bot. J. Linn. Soc.* 70: 83–182.
- Jorgensen, T. H., T. Petanidou & S. Andersson. 2006. The potential for selection on pollen colour dimorphisms in *Nigella degenii*: Morph-specific differences in pollinator visitation, fertilisation success and siring ability. *Evol. Ecol.* 20: 291–306.
- Judd, W. S. & R. G. Olmstead. 2004. A survey of tricolpate (eudicot) phylogenetic relationships. *Amer. J. Bot.* 91: 1627–1644.
- Jürgens, A. & S. Dötterl. 2004. Chemical composition of anther volatiles in Ranunculaceae: Genera-specific profiles in *Anemone*, *Aquilegia*, *Caltha*, *Pulsatilla*, *Ranunculus*, and *Trollius* species. *Amer. J. Bot.* 91: 1969–1980.
- Jürgens, A., T. Witt & G. Gottsberger. 2002. Pollen grain numbers, ovule numbers and pollen-ovule ratios in Caryophylloideae: Correlation with breeding system, pollination, life form, style number, and sexual system. *Sexual Pl. Reprod.* 14: 279–289.
- Kalis, A. J. 1979. Papaveraceae. The northwest European pollen flora: 20. *Rev. Palaeobot. Palynol.* 28: 209–260.
- Katifori, E., S. Alben, E. Cerda, D. R. Nelson & J. Dumais. 2010. Foldable structures and the natural design of pollen grains. *Proc. Natl. Acad. Sci. U.S.A.* 107: 7635–7639.
- Kato, M. 2005. Ecology of traplining bees and understory pollinators. Pp. 128–133 in D. W. Roubik, S. Sakai & A. A. H. Karim (editors), *Pollination Ecology and the Rain Forest*. Springer, New York.
- Kawagoe, T. & N. Suzuki. 2002. Floral sexual dimorphism and flower choice by pollinators in a nectarless monoecious vine *Akebia quinata* (Lardizabalaceae). *Ecol. Res.* 17: 295–303.
- Kim, S., D. E. Soltis, P. S. Soltis, M. J. Zanis & Y. Suh. 2004. Phylogenetic relationships among early-diverging eudicots based on four genes: Were the eudicots ancestrally woody? *Molec. Phylog. Evol.* 31: 16–30.
- Kim, Y. D. & R. K. Jansen. 1998. Chloroplast DNA restriction site variation and phylogeny of the Berberidaceae. *Amer. J. Bot.* 85: 1766–1778.
- Kim, Y. D., S. H. Kim & L. R. Landrum. 2004a. Taxonomic and phylogeographic implications from ITS phylogeny in *Berberis* (Berberidaceae). *J. Pl. Res.* 117: 175–182.
- Kim, Y. D., S. H. Kim, C. H. Kim & R. K. Jansen. 2004b. Phylogeny of Berberidaceae based on sequences of the chloroplast gene *ndhF*. *Biochem. Syst. Ecol.* 32: 291–301.
- Köhler, E. 2006. Three new *Buxus* species (Buxaceae) from eastern Cuba. *Willdenowia* 36: 479–489.
- Köhler, E. 2007a. Buxaceae. Pp. 40–47 in K. Kubitzki (editor), *The Families and Genera of Vascular Plants, Vol. IX. Flowering Plants. Eudicots: Berberidopsidales, Buxales, Crossosomatales, Fabales p.p., Geraniales, Gunnerales, Myrtales p.p., Proteales, Saxifragales, Vitales, Zygophyllales, Clusiaceae Alliance, Passifloraceae Alliance, Dilleniaceae, Huaceae, Picramniaceae, Sabiaceae*. Springer, Berlin.
- Köhler, E. 2007b. Didymelaceae. Pp. 129–131 in K. Kubitzki (editor), *The Families and Genera of Vascular Plants, Vol. IX. Flowering Plants. Eudicots: Berberidopsidales, Buxales, Crossosomatales, Fabales p.p., Geraniales, Gunnerales, Myrtales p.p., Proteales, Saxifragales, Vitales, Zygophyllales, Clusiaceae Alliance, Passifloraceae Alliance, Dilleniaceae, Huaceae, Picramniaceae, Sabiaceae*. Springer, Berlin.
- Köhler, E. & P. Brückner. 1989. The genus *Buxus* (Buxaceae): Aspects of its differentiation in space and time. *Pl. Syst. Evol.* 162: 267–283.
- Kreunen, S. S. & J. M. Osborn. 1999. Pollen and anther development in *Nelumbo* (Nelumbonaceae). *Amer. J. Bot.* 86: 1662–1676.
- Kubitzki, K. 2007. *The Families and Genera of Vascular Plants, Vol. IX. Flowering Plants. Eudicots: Berberidopsidales, Buxales, Crossosomatales, Fabales p.p., Geraniales, Gunnerales, Myrtales p.p., Proteales, Saxifragales, Vitales, Zygophyllales, Clusiaceae Alliance, Passifloraceae Alliance, Dilleniaceae, Huaceae, Picramniaceae, Sabiaceae*. Springer, Berlin.
- Kubitzki, K., J. G. Rohwer & V. Bittrich. 1993. *The Families and Genera of Vascular Plants II. Flowering Plants. Dicotyledons. Magnoliid, Hamamelid and Caryophyllid Families*. Springer, Berlin.
- Kuprianova, L. A. 1979. On the possibility of the development of tricolpate pollen from monosulcate. *Grana* 18: 1–4.
- Ladd, P. G. & S. W. Connell. 1994. Andromonoecy and fruit set in three genera of the Proteaceae. *Bot. J. Linn. Soc.* 116: 77–88.
- Laferriere, J. E. 1997. Transfer of specific and infraspecific taxa from *Mahonia* to *Berberis* (Berberidaceae). *Acta Bot. Indica* 25: 243–246.
- Lee, C. H., S. Lee, Y. Suh, S. H. Yeau & N. S. Lee. 2004. A palynotaxonomic study of Korean *Adonis* (Ranunculaceae). *J. Pl. Biol.* 47: 383–390.

- Lee, S. & S. Blackmore. 1992. A palynotaxonomic study of the genus *Trollius* (Ranunculaceae). *Grana* 31: 81–100.
- Les, D. H. 1991. Genetic diversity in the monoecious hydrophile *Ceratophyllum* (Ceratophyllaceae). *Amer. J. Bot.* 78: 1070–1082.
- Li, T.-Q., H.-J. Cao, M.-S. Kang, Z.-X. Zhang, N. Zhao & H. Zhang. 2011. Pollen flora of China woody plants by SEM, 1st ed. Science Press, Beijing.
- Liang, H.-R. & W.-M. Yan. 1991. Study on the pollen morphology of *Epimedium* in China. *Bull. Bot. Res.*, Harbin 11: 81–87.
- Lieux, M. H. 1980. An atlas of pollen of trees, shrubs, and woody vines of Louisiana and other southeastern states, Pt. II. Platanaceae to Betulaceae. *Pollen & Spores* 22: 191–243.
- Linder, H. P. 1998. Morphology and the evolution of wind pollination. Pp. 123–135 in S. T. Owens & P. J. Rudall (editors), *Reproductive Biology in Systematics, Conservation and Economic Botany*. Royal Botanic Gardens, Kew.
- Linder, H. P. 2000. Pollen morphology and wind pollination in angiosperms. Pp. 73–88 in M. M. Harley, C. M. Morton & S. Blackmore (editors), *Pollen and Spores: Morphology and Biology*. Royal Botanic Gardens, Kew.
- Lu, J.-C., Q.-S. Sun, Z.-M. Li, W.-Q. Li & J.-P. Li. 1999. A study on pollen morphology of *Anemone* from northeast of China. *J. Shenyang Pharm. Univ.* 16: 32–35.
- Lu, L., A. H. Wortley, D.-Z. Li, H. Wang & S. Blackmore. 2015. Evolution of angiosperm pollen. 2. The basal angiosperms. *Ann. Missouri Bot. Gard.* 100: 227–269.
- Luo, Y., L. Lu, A. H. Wortley, D.-Z. Li, H. Wang & S. Blackmore. 2015. Evolution of angiosperm pollen. 3. Monocots. *Ann. Missouri Bot. Gard.* 101: 406–455.
- Ma, S.-B. & Z.-H. Hu. 1997. A contribution to the geographical distribution and phylogeny of Podophylloideae (Berberidaceae). *Acta Bot. Yunnan.* 1: 48–56.
- Maddison, W. P. 1993. Missing data versus missing characters in phylogenetic analysis. *Syst. Biol.* 42: 576–581.
- Maddison, W. P. & D. R. Maddison. 2011. Mesquite: A modular system for evolutionary analysis. Vers. 2.75. <<http://mesquiteproject.org>>, accessed 10 December 2014.
- Magallón, S., S. Gómez-Acevedo, L. L. Sánchez-Reyes & T. Hernández-Hernández. 2015. A metacalibrated time-tree documents the early rise of flowering plant phylogenetic diversity. *New Phytol.* 207: 437–453.
- Maguire, B., J. J. Wurdack & Y.-C. Huang. 1974. Pollen grains of some American Olacaceae. *Grana* 14: 26–38.
- Mast, A. R., E. F. Milton, E. H. Jones, R. M. Barker, W. R. Barker & P. H. Weston. 2012. Time-calibrated phylogeny of the woody Australian genus *Hakea* (Proteaceae) supports multiple origins of insect-pollination among bird-pollinated ancestors. *Amer. J. Bot.* 99: 472–487.
- Maynard, G. V. 1995. Pollinators of Australian Proteaceae. Pp. 30–36 in P. McCarthy (editor), *Flora of Australia*, Vol. 16, *Eleagnaceae, Proteaceae* 1. Australian Biological Resources Study/CSIRO Publishing, Melbourne.
- Mayo, S. J., J. Bogner & P. C. Boyce. 1998. Araceae. Pp. 26–74 in K. Kubitzki (editor), *The Families and Genera of Vascular Plants*, Vol. VI. Flowering Plants. Monocotyledons: Alismatanae and Commelinanae (except Gramineae). Springer, Berlin.
- Meacham, C. A. 1980. Phylogeny of the Berberidaceae with an evaluation of classifications. *Syst. Bot.* 5: 149–172.
- Melampy, M. N. & A. M. Hayworth. 1980. Seed production and pollen vectors in several nectarless plants. *Evolution* 34: 1144–1154.
- Memon, H. R. 1985. Implication of pollen surface pattern for species and generic-level classification in Proteaceae. *Pakistan J. Bot.* 17: 87–106.
- Meng, A.-P., Z.-G. Zhang, J.-Q. Li, L. R. De Craene & H.-C. Wang. 2012. Floral development of *Stephania* (Menispermaceae): Impact of organ reduction on symmetry. *Int. J. Pl. Sci.* 173: 861–874.
- Michalski, S. G. & W. Durka. 2009. Pollination mode and life form strongly affect the relation between mating system and pollen to ovule ratios. *New Phytol.* 183: 470–479.
- Midgley, J. J. 1987. The derivation, utility and implications of a divergence index for the fynbos genus *Leucadendron* (Proteaceae). *Bot. J. Linn. Soc.* 95: 137–152.
- Milne, L. A. & A. R. H. Martin. 1998. Conospermeae (Proteoideae) pollen morphology and its phylogenetic implications. *Austral. Syst. Bot.* 11: 503–552.
- Momose, K., T. Yumoto, T. Nagamitsu, M. Kato, H. Nagamasu, S. Sakai, R. D. Harrison, T. Itioka, A. A. Hamid & T. Inoue. 1998. Pollination biology in a lowland dipterocarp forest in Sarawak, Malaysia. I. Characteristics of the plant-pollinator community in a lowland dipterocarp forest. *Amer. J. Bot.* 85: 1477–1501.
- Mondal, M. & K. Mitra. 1982. Pollen morphology and systematic relationship of Sabiaceae. *Geophytology* 12: 166–180.
- Moore, J. P., G. G. Lindsey, J. M. Farrant & W. F. Brandt. 2007. An overview of the biology of the desiccation-tolerant resurrection plant *Myrothamnus flabellifolia*. *Ann. Bot. (Oxford)* 99: 211–217.
- Moore, M. J., P. S. Soltis, C. D. Bell, J. G. Burleigh & D. E. Soltis. 2010. Phylogenetic analysis of 83 plastid genes further resolves the early diversification of eudicots. *Proc. Natl. Acad. Sci. U.S.A.* 107: 4623–4628.
- Motten, A. F. 1982. Autogamy and competition for pollinators in *Hepatica americana* (Ranunculaceae). *Amer. J. Bot.* 69: 1296–1305.
- Muller, J. 1970. Palynological evidence on early differentiation of angiosperms. *Biol. Rev.* 45: 417–450.
- Muller, J. 1979. Form and function in angiosperm pollen. *Ann. Missouri Bot. Gard.* 66: 593–632.
- Myerscough, P. J., R. J. Whelan & R. A. Bradstock. 2000. Ecology of Proteaceae with special reference to the Sydney region. *Cunninghamia* 6: 952–1015.
- Nowicke, J. W. & J. J. Skvarla. 1979. Pollen morphology: The potential influence in higher order systematics. *Ann. Missouri Bot. Gard.* 66: 633–700.
- Nowicke, J. W. & J. J. Skvarla. 1981. Pollen morphology and phylogenetic relationships of the Berberidaceae. *Smithsonian Contr. Bot.* 50: 1–83.
- Nowicke, J. W. & J. J. Skvarla. 1982. Pollen morphology and the relationships of *Circaeaster*, of *Kingdonia*, and of *Sargentodoxa* to the Ranunculales. *Amer. J. Bot.* 69: 990–998.
- Nowicke, J. W. & J. J. Skvarla. 1983. A palynological study of the genus *Helleborus* (Ranunculaceae). *Grana* 22: 129–140.
- Pagel, M. 1994. Detecting correlated evolution on phylogenies: A general method for the comparative analysis of discrete characters. *Proc. Roy. Soc. London, Ser. B, Biol. Sci.* 255: 37–45.
- Pagel, M. 2006. BayesTraits. v1.0. University of Reading. <www.evolution.rdg.ac.uk>, accessed 16 January 2016.

- Pagel, M. & A. Meade. 2006. Bayesian analysis of correlated evolution of discrete characters by reversible-jump Markov chain Monte Carlo. *Amer. Naturalist* 167: 808–825.
- Paton, D. C. & V. Turner. 1985. Pollination of *Banksia ericifolia* Smith: Birds, mammals and insects as pollen vectors. *Austral. J. Bot.* 33: 271–286.
- Payne, W. W. & J. L. Seago. 1968. The open conduplicate carpel of *Akebia quinata* (Berberidales: Lardizabalaceae). *Amer. J. Bot.* 55: 575–581.
- Pellmyr, O. 1984. The pollination ecology of *Actaea spicata* (Ranunculaceae). *Nordic J. Bot.* 4: 443–456.
- Pellmyr, O. 1985. The pollination biology of *Actaea pachypoda* and *A. rubra* (including *A. erythrocarpa*) in northern Michigan and Finland. *Bull. Torrey Bot. Club* 112: 265–273.
- Pellmyr, O. 1986. The pollination ecology of two nectarless *Cimicifuga* sp. (Ranunculaceae) in North America. *Nordic J. Bot.* 6: 713–723.
- Pellmyr, O. 1988. Bumble bees (Hymenoptera: Apidae) assess pollen availability in *Anemonopsis macrophylla* (Ranunculaceae) through floral shape. *Ann. Entomol. Soc. Amer.* 81: 792–797.
- Peng, Y., S.-B. Chen, Y. Liu, S.-L. Chen & P.-G. Xiao. 2006. A pharmacophylogenetic study of the Berberidaceae (s.l.). *Acta Phytotax. Sin.* 44: 241–257.
- Pérez-Gutiérrez, M. A., A. T. Romero-García, M. J. Salinas, G. Blanca, M. C. Fernández & V. N. Suárez-Santiago. 2012. Phylogeny of the tribe Fumariaceae (Papaveraceae s.l.) based on chloroplast and nuclear DNA sequences: Evolutionary and biogeographic implications. *Amer. J. Bot.* 99: 517–528.
- Perveen, A. 1999. A palynological survey of aquatic flora of Karachi-Pakistan. *Turkish J. Bot.* 23: 309–317.
- Perveen, A. & M. Qaiser. 2004. Pollen flora of Pakistan—XL. Fumariaceae. *Pakistan J. Bot.* 36: 467–474.
- Perveen, A. & M. Qaiser. 2006. Pollen flora of Pakistan—L. Ranunculaceae. *Pakistan J. Bot.* 38: 499–509.
- Perveen, A. & M. Qaiser. 2010. Pollen flora of Pakistan—LXV. Berberidaceae. *Pakistan J. Bot.* 42: 1–6.
- Pragowski, J. 1970. The pollen morphology of the Haloragaceae with reference to taxonomy. *Grana* 10: 159–239.
- Pragowski, J. 1975. The pollen morphology of the Trochodendraceae, Tetracentraceae, Cercidiphyllaceae and Eupteleaceae with reference to taxonomy. *Pollen & Spores* 16: 449–467.
- Prum, R. O., J. S. Berv, A. Dornburg, D. J. Field, J. P. Townsend, E. M. Lemmon & A. R. Lemmon. 2015. A comprehensive phylogeny of birds (Aves) using targeted next-generation DNA sequencing. *Nature* 526: 569–573.
- Punt, W. 1975. Pollen morphology of the Dichapetalaceae with special reference to evolutionary trends and mutual relationships of pollen types. *Rev. Palaeobot. Palynol.* 19: 1–97.
- Punt, W. & A. Marks. 1991. Buxaceae. The northwest European pollen flora, 50. *Rev. Palaeobot. Palynol.* 69: 113–115.
- Punt, W., P. P. Hoen, S. Blackmore, S. Nilsson & A. Le Thomas. 2007. Glossary of pollen and spore terminology. *Rev. Palaeobot. Palynol.* 143: 1–81.
- Qiu, Y.-L., L. Li, B. Wang, J.-Y. Xue, T. A. Hendry, R.-Q. Li, J. W. Brown, Y. Liu, G. T. Hudson & Z.-D. Chen. 2010. Angiosperm phylogeny inferred from sequences of four mitochondrial genes. *J. Syst. Evol.* 48: 391–425.
- Rachele, L. D. 1974. Pollen morphology of the Papaveraceae of the northeastern United States and Canada. *Bull. Torrey Bot. Club* 101: 152–159.
- Rawal, R. S. & Y. P. S. Pangtey. 1991. Distribution and phenology of climbers of Kumaun in Central Himalaya, India. *Vegetatio* 97: 77–87.
- Ressayre, A., B. Godelle, C. Raquin & P. H. Gouyon. 2002. Aperture pattern ontogeny in angiosperms. *J. Exp. Zool.* 294: 122–135.
- Ressayre, A., L. Dreyer, S. Triki-Teurtroy, A. Forchioni & S. Nadot. 2005. Post-meiotic cytokinesis and pollen aperture pattern ontogeny: Comparison of development in four species differing in aperture pattern. *Amer. J. Bot.* 92: 576–583.
- Romero, A. T., M. J. Salinas & M. C. Fernández. 2003. Pollen wall development in *Hypocoum imberbe* Sm. (Fumariaceae). *Grana* 42: 91–101.
- Ronse De Craene, L. P. & L. Wanntorp. 2008. Morphology and anatomy of the flower of *Meliosma* (Sabiaceae): Implications for pollination biology. *Pl. Syst. Evol.* 271: 79–91.
- Rysiak, K. & B. Żuraw. 2011. The biology of flowering of winter aconite (*Eranthis hyemalis* (L.) Salisb.). *Acta Agrobot.* 64: 25–32.
- Salinas, M. J., A. T. Romero, G. Blanca, R. de la Herrán, M. Garrido-Ramos, C. Ruíz-Rejón, C. Morales, M. Ruíz-Rejón & V. Suárez. 2003. Contribution to the taxonomy and phylogeny of *Sarcocapnos* DC. (Fumariaceae). *Pl. Syst. Evol.* 237: 153–164.
- Sannier, J., W. J. Baker, M.-C. Anstett & S. Nadot. 2009. A comparative analysis of pollinator type and pollen ornamentation in the Araceae and the Arecaceae, two unrelated families of the monocots. *BMC Res. Notes* 2: 145.
- Santisuk, T. 1979. A palynological study of the tribe Ranunculaceae. *Opera Bot.* 48: 1–74.
- Sauquet, H. & D. J. Cantrill. 2007. Pollen diversity and evolution in Proteoideae (Proteales: Proteaceae). *Syst. Bot.* 32: 271–316.
- Sauquet, H., P. H. Weston, C. L. Anderson, N. P. Barker, D. J. Cantrill, A. R. Mast & V. Savolainen. 2009a. Contrasted patterns of hyperdiversification in Mediterranean hotspots. *Proc. Natl. Acad. Sci. U.S.A.* 106: 221–225.
- Sauquet, H., P. H. Weston, N. P. Barker, C. L. Anderson, D. J. Cantrill & V. Savolainen. 2009b. Using fossils and molecular data to reveal the origins of the Cape proteas (subfamily Proteoideae). *Molec. Phylog. Evol.* 51: 31–43.
- Sauquet, H., L. Carrive, N. Poullain, J. Sannier, C. Damerval & S. Nadot. 2015. Zygomorphy evolved from disymmetry in Fumarioideae (Papaveraceae, Ranunculales): New evidence from an expanded molecular phylogenetic framework. *Ann. Bot. (Oxford)* 115(6): 895–914. doi: 10.1093/aob/mcv020.
- Savolainen, V., M. W. Chase, S. B. Hoot, C. M. Morton, D. E. Soltis, C. Bayer, M. F. Fay, A. Y. De Bruijn, S. Sullivan & Y.-L. Qiu. 2000a. Phylogenetics of flowering plants based on combined analysis of plastid *atpB* and *rbcL* gene sequences. *Syst. Biol.* 49: 306–362.
- Savolainen, V., M. F. Fay, D. C. Albach, A. Backlund, M. van der Bank, K. M. Cameron, S. A. Johnson, M. D. Lledó, J.-C. Pintaud, M. Powell, M. C. Sheahan, D. E. Soltis, P. S. Soltis, P. Weston, W. M. Whitten, K. J. Wurdack & M. W. Chase. 2000b. Phylogeny of the eudicots: A nearly complete familial analysis based on *rbcL* gene sequences. *Kew Bull.* 55: 257–309.

- Sharifnia, F., S. Heydarian & F. Salimpour. 2010. Biosystematic study of some *Papaver* (Papaveraceae) species from Iran. *Iranian J. Bot.* 16: 54–68.
- Smith, B. H., M. L. Ronsleben & K. R. Swartz. 1986. Reproductive ecology of *Jeffersonia diphylla* (Berberidaceae). *Amer. J. Bot.* 73: 1416–1426.
- Sohmer, S. H. & D. F. Sefton. 1978. The reproductive biology of *Nelumbo pentapetala* (Nelumbonaceae) on the Upper Mississippi River. II. The insects associated with the transfer of pollen. *Brittonia* 30: 355–364.
- Soltis, D. E., P. S. Soltis, M. W. Chase, M. E. Mort, D. C. Albach, M. Zanis, V. Savolainen, W. H. Hahn, S. B. Hoot, M. F. Fay, M. Axtell, S. M. Swensen, L. M. Prince, W. J. Kress, K. C. Nixon & J. S. Farris. 2000. Angiosperm phylogeny inferred from 18S rDNA, *rbcL*, and *atpB* sequences. *Bot. J. Linn. Soc.* 133: 381–461.
- Soltis, D. E., A. E. Sinters, M. J. Zanis, S. Kim, J. D. Thompson, P. S. Soltis, L. P. R. De Craene, P. K. Endress & J. S. Farris. 2003. Gunnerales are sister to other core eudicots: Implications for the evolution of pentamery. *Amer. J. Bot.* 90: 461–470.
- Soltis, D. E., M. A. Gitzendanner & P. S. Soltis. 2007. A 567-taxon data set for angiosperms: The challenges posed by Bayesian analyses of large data sets. *Int. J. Pl. Sci.* 168: 137–157.
- Soltis, D. E., S. A. Smith, N. Cellinese, K. J. Wurdack, D. C. Tank, S. F. Brockington, N. F. Refulio-Rodriguez, J. B. Walker, M. J. Moore, B. S. Carlswald, C. D. Bell, M. Latvis, S. Crawley, C. Black, D. Diouf, Z.-X. Xi, C. A. Rushworth, M. A. Gitzendanner, K. J. Sytsma, Y.-L. Qiu, K. W. Hilu, C. C. Davis, M. J. Sanderson, R. S. Beaman, R. G. Olmstead, W. S. Judd, M. J. Donoghue & P. S. Soltis. 2011. Angiosperms phylogeny: 17 genes, 640 taxa. *Amer. J. Bot.* 98: 704–730.
- Soltis, D. E., M. E. Mort, M. Latvis, E. V. Mavrodiev, B. C. O'Meara, P. S. Soltis, J. G. Burleigh & R. de Casas. 2013. Phylogenetic relationships and character evolution analysis of Saxifragales using a supermatrix approach. *Amer. J. Bot.* 100: 916–929.
- Stamatakis, A. 2006. RAxML-VI-HPC: Maximum likelihood-based phylogenetic analyses with thousands of taxa and mixed models. *Bioinformatics* 22: 2688–2690.
- Steenhuisen, S.-L. & S. D. Johnson. 2012. Evidence for beetle pollination in the African grassland sugarbushes (*Protea*: Proteaceae). *Pl. Syst. Evol.* 298: 857–869.
- Steenhuisen, S.-L., H. Van der Bank & S. D. Johnson. 2012. The relative contributions of insect and bird pollinators to outcrossing in an African *Protea* (Proteaceae). *Amer. J. Bot.* 99: 1104–1111.
- Stern, K. R. 1962. The use of pollen morphology in the taxonomy of *Dicentra*. *Amer. J. Bot.* 49: 362–368.
- Stroo, A. 2000. Pollen morphological evolution in bat pollinated plants. *Pl. Syst. Evol.* 200: 225–242.
- Su, Y.-J. & Q.-H. Liu. 1994. A study on the pollen morphology of *Dyosma* from Hubei. *J. Wuhan Bot. Res.* 12: 217–219.
- Sun, G., D. L. Dilcher, H.-S. Wang & Z.-D. Chen. 2011. A eudicot from the Early Cretaceous of China. *Nature* 471: 625–628.
- Sun, Y., M. J. Moore, S. Zhang, P. S. Soltis, D. E. Soltis, T. Zhao, A. Meng, X. Li, J. Li & H. Wang. 2016. Phylogenomic and structural analyses of 18 complete plastomes across nearly all families of early-diverging eudicots, including an angiosperm-wide analysis of IR gene content evolution. *Molec. Phylog. Evol.* 96: 93–101.
- Suzuki, K. 1983. Breeding system and crossability in Japanese *Epimedium* (Berberidaceae). *Bot. Mag. (Tokyo)* 96: 343–350.
- Suzuki, K. 1984. Pollination system and its significance on isolation and hybridization in Japanese *Epimedium* (Berberidaceae). *Bot. Mag. (Tokyo)* 97: 381–396.
- Takahashi, M. 1995. Development of structure-less pollen wall in *Ceratophyllum demersum* L. (Ceratophyllaceae). *J. Pl. Res.* 108: 205–208.
- Takahashi, M. & S. Kawano. 1989. Pollen morphology of the Melanthiaceae and its systematic implications. *Ann. Missouri Bot. Gard.* 76: 863–876.
- Tamura, M. 1993. Ranunculaceae. Pp. 563–583 in K. Kubitzki, J. G. Rohrer & V. Bittrich (editors), *The Families and Genera of Vascular Plants II. Flowering Plants. Dicotyledons. Magnoliid, Hamamelid and Caryophyllid Families*. Springer, Berlin.
- Tank, D. C., J. M. Eastman, M. W. Pennell, P. S. Soltis, D. E. Soltis, C. E. Hinchliff, J. W. Brown, E. B. Sessa & L. J. Harmon. 2015. Nested radiations and the pulse of angiosperm diversification: Increased diversification rates often follow whole genome duplications. *New Phytol.* 207: 454–467.
- Tatlidil, S., A. Bıçakcı, H. Malyer & K. H. Can Baser. 2005. Pollen morphology of *Thalictrum* L., species (Ranunculaceae) in Turkey. *Pakistan J. Bot.* 37: 203–212.
- Thanikaimoni, G. 1968. Morphologie des pollens des Ménispermacées. *Trav. Sect. Sci. Techn. Inst. Franç. Pondichéry* 5: 1–57.
- Tolgor, H.-Y. Bao, S.-M. Zhang & X.-L. Nie. 1997. Pollen morphology of *Aconitum* L. and its taxonomic significance. *J. Jinlin Agric. Univ.* 19: 59–64.
- Van Campo, M. 1976. Patterns of pollen morphological variation within taxa. Pp. 125–137 in I. K. Ferguson & J. Muller (editors), *The Evolutionary Significance of the Exine*. Academic Press, London.
- Vaughton, G. 1996. Pollination disruption by European honeybees in the Australian bird-pollinated shrub *Grevillea barklyana* (Proteaceae). *Pl. Syst. Evol.* 200: 89–100.
- Vishnu-Mittre & B. D. Sharma. 1963. Studies of Indian pollen grains 2. Ranunculaceae. *Pollen & Spores* 5: 285–296.
- Von Balthazar, M. & P. K. Endress. 2002. Reproductive structures and systematics of Buxaceae. *Bot. J. Linn. Soc.* 140: 193–228.
- Von Balthazar, M., G. E. Schatz & P. K. Endress. 2003. Female flowers and inflorescences of Didymelaceae. *Pl. Syst. Evol.* 237: 199–208.
- Walker, J. W. 1974. Aperture evolution in the pollen of primitive angiosperms. *Amer. J. Bot.* 61: 1112–1137.
- Walker, J. W. & J. A. Doyle. 1975. The bases of angiosperm phylogeny: Palynology. *Ann. Missouri Bot. Gard.* 62: 664–723.
- Wallace, H. M., G. V. Maynard & S. J. Trueman. 2002. Insect flower visitors, foraging behaviour and their effectiveness as pollinators of *Persoonia virgata* R. Br. (Proteaceae). *Austral. J. Entomol.* 41: 55–59.
- Wang, F. & D.-Z. Li. 2002. Cladistic analysis of the Lardizabalaceae based on morphology data. *Acta Bot. Yunnan.* 24: 445–454.
- Wang, F., N. Chien, Y. Zhang & H. Yang. 1995. *Pollen Flora of China*, 2nd ed. Science Press, Beijing.
- Wang, F., D.-Z. Li & J.-B. Yang. 2002. Molecular phylogeny of the Lardizabalaceae based on *trnL-F*

- sequences and combined chloroplast data. *Acta Bot. Sin.* 44: 971–977.
- Wang, G., J. Chen, Z.-B. Li, F.-P. Zhang & D.-R. Yang. 2014. Has pollination mode shaped the evolution of *Ficus* pollen? *PLoS One* 9: e86231.
- Wang, H.-F., B. K. Kirchoff, H.-N. Qin & Z.-X. Zhu. 2009. Reproductive morphology of *Sargentodoxa cuneata* (Lardizabalaceae) and its systematic implications. *Pl. Syst. Evol.* 280: 207–217.
- Wang, J.-Q. 1990. A study on pollen morphology of *Potamogeton*, *Zannichellia* and *Triglochin* from China. *Acta Phytotax. Sin.* 28: 372–378.
- Wang, W. & N.-X. Zhao. 2002. Pollen morphology of the Araceae from China. *Acta Phytotax. Sin.* 40: 517–529.
- Wang, W.-T. & L. Xie. 2007. A revision of *Clematis* sect. *Tubulosae* (Ranunculaceae). *Acta Phytotax. Sin.* 45: 425–457.
- Wang, W., Z.-D. Chen, Y. Liu, R.-Q. Li & J.-H. Li. 2007a. Phylogenetic and biogeographic diversification of Berberidaceae in the Northern Hemisphere. *Syst. Bot.* 32: 731–742.
- Wang, W., H.-C. Wang & Z.-D. Chen. 2007b. Phylogeny and morphological evolution of tribe Menispermaceae (Menispermaceae) inferred from chloroplast and nuclear sequences. *Perspect. Pl. Ecol. Evol. Syst.* 8: 141–154.
- Wang, W., A.-M. Lu, Y. Ren, M. E. Endress & Z.-D. Chen. 2009. Phylogeny and classification of Ranunculales: Evidence from four molecular loci and morphological data. *Perspect. Pl. Ecol. Evol. Syst.* 11: 81–110.
- Wang, W., D. L. Dilcher, G. Sun, H.-S. Wang & Z.-D. Chen. 2014. Accelerated evolution of early angiosperms: Evidence from ranunculalean phylogeny by integrating living and fossil data. *J. Syst. Evol.* 53: 1–7.
- Wang, X.-Q., D.-Y. Hong & Z.-Y. Li. 1993. A study on pollen and seed coat in the tribe *Cimicifugeae* and some allied genera (Ranunculaceae). *Cathaya* 5: 131–149.
- Wang, Y.-H., J.-M. Chen, C. Xu, X. Liu, Q.-F. Wang & T. J. Motley. 2010. Population genetic structure of an aquatic herb *Batrachium bungei* (Ranunculaceae) in the Hengduan Mountains of China. *Aquatic Bot.* 92: 221–225.
- Wanntorp, L., M. E. Dettmann & D. M. Jarzen. 2004a. Tracking the Mesozoic distribution of *Gunnera*: Comparison with the fossil pollen species *Tricolpites reticulatus* Cookson. *Rev. Palaeobot. Palynol.* 132: 163–174.
- Wanntorp, L., J. Praglowski & E. Grafström. 2004b. New insights into the pollen morphology of the genus *Gunnera* (Gunneraceae). *Grana* 43: 15–21.
- Wei, Z.-X. & D.-Z. Li. 1995. Pollen morphology and wall ultrastructure of Sargentodoxaceae. *Acta Bot. Yunnan.* 17: 197–200.
- Weston, P. H. & N. P. Barker. 2006. A new suprageneric classification of the Proteaceae, with an annotated checklist of genera. *Telopea* 11: 314–344.
- Whitehead, D. R. 1969. Wind pollination in the angiosperms: Evolutionary and environmental considerations. *Evolution* 23: 28–35.
- Wilkinson, H. & L. Wanntorp. 2007. Gunneraceae. Pp. 177–183 in K. Kubitzki (editor), *The Families and Genera of Vascular Plants*, Vol. IX. Flowering Plants. Eudicots: Berberidopsidales, Buxales, Crossosomatales, Fabales p.p., Geraniales, Gunnerales, Myrtales p.p., Proteales, Saxifragales, Vitales, Zygophyllales, Clusiaceae Alliance, Passifloraceae Alliance, Dilleniaceae, Huaceae, Picramniaceae, Sabiaceae. Springer, Berlin.
- Worberg, A., D. Quandt, A.-M. Barniske, C. Löhne, K. W. Hilu & T. Borsch. 2007. Phylogeny of basal eudicots: Insights from non-coding and rapidly evolving DNA. *Organisms Diversity Evol.* 7: 55–77.
- Wortley, A. H., H. Wang, L. Lu., D.-Z. Li & S. Blackmore. 2015. Evolution of angiosperm pollen. 1. Introduction. *Ann. Missouri Bot. Gard.* 100: 177–226.
- Xi, Y.-Z. 1993. Investigation on pollen morphology of *Aconitum* L. *Acta Bot. Sin.* 53: 674–686.
- Xia, Q. & Z.-X. Peng. 1989. A study on the pollen morphology of Lardizabalaceae, Sargentodoxaceae and its significance in taxonomy. *Bull. Bot. Res.*, Harbin 9: 99–114.
- Xia, Q. & J. Kong. 1991. A study on the ultrastructure of pollen wall of Lardizabalaceae and Sargentodoxaceae. *Bull. Bot. Res.*, Harbin 11: 93–98.
- Yang, D.-K., H. Yang & Z. Chen. 2006. Studies on the pollen morphology of *Ranunculus* L. in Shandong. *J. Shandong Normal Univ.*, Nat. Sci. 21: 123–125.
- Ye, Q. G., E. Bunn & K. W. Dixon. 2012. A ballistic pollen dispersal system influences pollination success and fruit-set pattern in pollinator-excluded environments for the endangered species *Synaphea stenoloba* (Proteaceae). *Bot. J. Linn. Soc.* 170: 59–68.
- Yildiz, K., S. Gücel & M. Y. Dadandi. 2009. A palynological investigation of endemic taxa from northern Cyprus. *Pakistan J. Bot.* 41: 991–1007.
- Ying, T.-S. 1979. On *Dysosma* Woodson and *Sinopodophyllum* Ying, gen. nov. of the Berberidaceae. *Acta Phytotax. Sin.* 17: 15–23.
- Yu, X.-S. & Q.-H. Liu. 1991. A study on the pollen morphology of *Epimedium* Hubei Province. *J. Wuhan Bot. Res.* 9: 295–297.
- Yuan, Q. & Q.-E. Yang. 2006. Cytology, palynology, and taxonomy of *Asteropyrum* and four other genera of Ranunculaceae. *Bot. J. Linn. Soc.* 152: 15–26.
- Zaragüeta-Bagils, R. & E. Bourdon. 2007. Three-item analysis: Hierarchical representation and treatment of missing and inapplicable data. *Compt. Rend. Palevol.* 6: 527–534.
- Zavada, M. S. & D. L. Dilcher. 1986. Comparative pollen morphology and its relationship to phylogeny of pollen in the Hamamelidae. *Ann. Missouri Bot. Gard.* 73: 348–381.
- Zhang, Y.-L. 1983. Pollen morphology of *Kingdonia uniflora* and its taxonomic significance. *Acta Phytotax. Sin.* 21: 441–447.
- Zhou, Y., Y.-Z. Guo & X.-S. Meng. 1990. A morphological study of pollen of five *Thalictrum* plants. *J. Shenyang Coll. Pharm.* 7: 274–276.

Appendix 1. Voucher details for pollen samples shown in Figures 3 through 7. Images obtained without acetolysis are shown in boldface.

Taxon	Collection	Locality	Herbarium	Figure
<i>Corydalis linarioides</i> Maxim. (Papaveraceae)	LJQ-QLS-2008-0113	Qinghai, China	KUN	3A-C
<i>Eschscholzia caespitosa</i> Benth. (Papaveraceae)	<i>J. T. Howell 37282</i>	Tulare Co., California, U.S.A.	CAS	3D-F
<i>Hypecoum procumbens</i> L. (Papaveraceae)	<i>L. S. Rose s.n.</i>	s. loc.	CAS	3G-I
<i>Stephania cephalantha</i> Hayata (Menispermaceae)	<i>B. Xiong 2352</i>	Jiangxi, China	MO	3J-L
<i>Berberis yunnanensis</i> Franch. (Berberidaceae)	09CS1052	Shangri-La, Yunnan, China	KUN	4A
<i>Berberis yunnanensis</i> Franch. (Berberidaceae)	BEP06	Kunming Botanical Garden, China (cultivated)	KUN	4B, C
<i>Caulophyllum robustum</i> Maxim. (Berberidaceae)	03-0077	Yunnan, China	KUN	4D, E, F
<i>Diphylleia sinensis</i> H. L. Li (Berberidaceae)	6709	Yunnan, China	KUN	4G, H, I
<i>Podophyllum emodi</i> Wall. ex Hook. f. & Thomson (= <i>P. hexandrum</i> Royle) (Berberidaceae)	1022	Diqing Prefecture, Yunnan, China	E	4J-L
<i>Ranzania japonica</i> (T. Itô ex Maxim.) T. Itô (Berberidaceae)	Cult, W03	Royal Botanic Garden, Edinburgh (cultivated)	E	5A-C
<i>Anemone vitifolia</i> Buch.-Ham. ex DC. (Ranunculaceae)	BEP01	Kunming Botanical Garden, China (cultivated)	KUN	5D, E, F
<i>Ranunculus japonicus</i> Thunb. (Ranunculaceae)	SCSB-HN-1577	Hunan, China	KUN	5G
<i>Ranunculus japonicus</i> Thunb. (Ranunculaceae)	SCSB-HN-1416	Hunan, China	KUN	5H, I
<i>Thalictrum delavayi</i> Franch. (Ranunculaceae)	ZhangDC-07ZX-1287	Tibet, China	KUN	5J-L
<i>Trollius ranunculoides</i> Hemsl. (Ranunculaceae)	SunH-07ZX-0232	Tibet, China	KUN	6A, B, C
<i>Meliosma arnottiana</i> (Wight) Walp. (Sabiaceae)	09CS1264	Yunnan, China	KUN	6D, E, F
<i>Grevillea baueri</i> R. Br. (Proteaceae)	BEP04	Kunming Botanical Garden, China (cultivated)	KUN	6G, H, I
<i>Hakea mitchellii</i> Meisn. (Proteaceae)	1402	South Australia	CBG	6J, K, L
<i>Macadamia rousseilii</i> (Vieill.) Sleumer (Proteaceae)	26624	New Caledonia	CANB	7A-C
<i>Stenocarpus umbelliferus</i> Druce (Proteaceae)	722	New Caledonia	CANB	7D, E
<i>Banksia acanthopoda</i> (A. S. George) A. R. Mast & K. R. Thiele (Proteaceae)	9496	Western Australia	CANB	7F
<i>Helicia macrostachya</i> Lauterb. (Proteaceae)	LAE 55386	Papua New Guinea	CANB	7G
<i>Tetracentron sinense</i> Oliv. (Trochodendraceae)	DZL-063	Yunnan, China	KUN	7H, I
<i>Buxus sempervirens</i> L. (Buxaceae)	13376	Arnold Arboretum, Boston, Massachusetts, U.S.A.	KUN	7J
<i>Didymeles integrifolia</i> J. St.-Hil. (Buxaceae)	18.088SF	s. loc.	MO	7K
<i>Pachysandra axillaris</i> Franch. (Buxaceae)	574469	Yunnan, China	KUN	7L

Appendix 2. Literature sources for palynological and ecological data, listed alphabetically by taxon (asterisks indicate ecological literature).

OUTGROUPS

Acorales/Acoraceae/Acorus L. (Grayum, 1992; Bogner & Mayo, 1998*; Buzgo & Endress, 2000*; Wang & Zhao, 2002; Furness & Banks, 2010).

Alismatales/Alismataceae/Alisma L. (Argue, 1974; Chanda et al., 1988; Furness & Banks, 2010). **Araceae/Orontium** L. (Mayo et al., 1998*; Furness & Banks, 2010; <<http://www.paldata.org/>>). **Potamogetonaceae/Potamogeton** L. (Wang, 1990; Perveen, 1999; Furness & Banks, 2010). **Tofieldiaceae/Tofieldia** Huds. (Takahashi & Kawano, 1989; Díaz Lifante et al., 1990; Furness & Banks, 2010).

Ceratophyllales/Ceratophyllaceae/Ceratophyllum L. (Erdtman, 1952; Díez et al., 1988; Les, 1991*; Takahashi, 1995).

Dilleniales/Dilleniaceae/Dillenia L. (Erdtman, 1952; Endress, 1997*; Bhattacharya et al., 1999; Li et al., 2011; <<http://apsa.anu.edu.au/>>).

Gunnerales/Gunneraceae/Gunnera L. (Pragowski, 1970; Wanntorp et al., 2004a, 2004b; Furness et al., 2007;

Kubitzki, 2007*; Wilkinson & Wanntorp, 2007; Endress, 2010*; <<http://www.paldata.org/>>); **Myrothamnus** Welw. (Erdtman, 1952; Zavada & Dilcher, 1986; Kubitzki et al., 1993*; Wanntorp et al., 2004b; Furness et al., 2007; Moore et al., 2007*; Endress, 2010*).

BASAL EUDICOTS

Buxales/Buxaceae/Buxus L. (Erdtman, 1952; Behnke, 1982; Köhler & Brückner, 1989, 1993; Punt & Marks, 1991; Wang et al., 1995; Köhler, 2006, 2007a; Furness et al., 2007; Kubitzki, 2007*; Li et al., 2011); **Pachysandra** Michx. (Behnke, 1982; Von Balthazar & Endress, 2002*; Köhler, 2007a; Kubitzki, 2007*; <<http://www.paldata.org/>>); **Didymeles** Thouars (Erdtman, 1952; Zavada & Dilcher, 1986; Furness et al., 2007; Köhler, 2007b; Kubitzki, 2007*).

Proteales/Nelumbonaceae/Nelumbo Adans. (Sohmer & Sefton, 1978*; Blackmore et al., 1995; Kreunen & Osborn, 1999; Banks et al., 2007; Furness et al., 2007; <<http://www.paldata.org/>>). **Platanaceae/Platanus** L. (Lieux, 1980; Kubitzki et al., 1993*; Wang et al., 1995; Denk & Tekleeva, 2006; Furness et al., 2007; Endress, 2010*; Li et al., 2011; <<http://www.paldata.org/>>). **Proteaceae/Acidonia** L. A. S.

- Johnson & B. G. Briggs (Memon, 1985; Feuer, 1986; Kubitzki, 2007*); *Adenanthos* Labill. (Erdtman, 1952; Ford et al., 1979*; Memon, 1985; Collins & Rebelo, 1987*; Maynard, 1995*; Dettmann & Jarzen, 1998; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Agastachys* R. Br. (Dettmann & Jarzen, 1998; Milne & Martin, 1998; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Alloxyylon* P. H. Weston & Crisp (Weston & Barker, 2006; Kubitzki, 2007*); *Athertonia* L. A. S. Johnson & B. G. Briggs (Memon, 1985; Weston & Barker, 2006; Kubitzki, 2007*); *Atulax* P. J. Bergius (Collins & Rebelo, 1987*; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Austromuellera* C. T. White (Memon, 1985; Weston & Barker, 2006; Kubitzki, 2007*); *Banksia* L. f. (Erdtman, 1952; Ford et al., 1979*; Memon, 1985; Paton & Turner, 1985*; Collins & Rebelo, 1987*; Carthew, 1994*; Weston & Barker, 2006; Kubitzki, 2007*; <<http://www.paldat.org/>>); *Beauprea* Brongn. & Gris (Erdtman, 1952; Dettmann & Jarzen, 1998; Milne & Martin, 1998; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Beaupreopsis* Virot (Milne & Martin, 1998; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Bellendena* R. Br. (Erdtman, 1952; Memon, 1985; Feuer, 1986; Dettmann & Jarzen, 1998; Kubitzki, 2007*); *Bleasdalea* F. Muell. ex Domin (Weston & Barker, 2006; Kubitzki, 2007*); *Brabejum* L. (Erdtman, 1952; Collins & Rebelo, 1987*; Ladd & Connell, 1994*; Weston & Barker, 2006; Kubitzki, 2007*); *Buckinghamia* F. Muell. (Memon, 1985; Feuer, 1989; Kubitzki, 2007*); *Cardwellia* F. Muell. (Erdtman, 1952; Weston & Barker, 2006; Kubitzki, 2007*); *Carnarvonia* F. Muell. (Memon, 1985; Feuer, 1986; Dettmann & Jarzen, 1998; Kubitzki, 2007*); *Catalepidia* P. H. Weston (Weston & Barker, 2006; Kubitzki, 2007*); *Cenarrhenes* Labill. (Erdtman, 1952; Milne & Martin, 1998; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Conospermum* Sm. (Erdtman, 1952; Collins & Rebelo, 1987*; Maynard, 1995*; Milne & Martin, 1998; Wallace et al., 2002*; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Darlingia* F. Muell. (Erdtman, 1952; Memon, 1985; Weston & Barker, 2006; Kubitzki, 2007*); *Diastella* Salisb. ex J. Knight (Erdtman, 1952; Collins & Rebelo, 1987*; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Dilobeia* Thouars (Erdtman, 1952; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Dryandra* R. Br. (Erdtman, 1952; Ford et al., 1979*; Memon, 1985; Collins & Rebelo, 1987*; Howell et al., 1993*; Weston & Barker, 2006; Kubitzki, 2007*); *Eidothea* A. W. Douglas & B. Hyland (Dettmann & Jarzen, 1998; Kubitzki, 2007*); Sauquet & Cantrill, 2007); *Embothrium* J. R. Forst. & C. Forst. (Memon, 1985; Feuer, 1990; Kubitzki, 2007*; Chalcoff et al., 2012*); *Eucarpha* Spach (Memon, 1985; Weston & Barker, 2006; Kubitzki, 2007*); *Euplassa* Salisb. ex Knight (Erdtman, 1952; Weston & Barker, 2006; Kubitzki, 2007*); *Faurea* Harv. (Erdtman, 1952; Memon, 1985; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Floydia* L. A. S. Johnson & B. G. Briggs (Memon, 1985; Weston & Barker, 2006; Kubitzki, 2007*); *Franklandia* R. Br. (Erdtman, 1952; Dettmann & Jarzen, 1998; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Garnieria* Brongn. & Gris (Erdtman, 1952; Feuer, 1986; Kubitzki, 2007*); *Gevuina* Molina (Dettmann & Jarzen, 1998; Weston & Barker, 2006; Kubitzki, 2007*); *Grevillea* R. Br. ex Knight (Erdtman, 1952; Ford et al., 1979*; Collins & Rebelo, 1987*; Maynard, 1995*; Voughton, 1996*; Weston & Barker, 2006; Kubitzki, 2007*; Li et al., 2011; Mast et al., 2012*; <<http://www.paldat.org/>>); *Hakea* Schrad. (Memon, 1985; Collins & Rebelo, 1987*; Kubitzki, 2007*; Mast et al., 2012*; <<http://www.paldat.org/>>); *Helicia* Lour. (Erdtman, 1952; Myerscough et al., 2000*; Kubitzki, 2007*; Li et al., 2011); *Heliciopsis* Sleumer (Memon, 1985; Momose et al., 1998*; Weston & Barker, 2006; Kubitzki, 2007*); *Hicksbeachia* F. Muell. (Memon, 1985; Weston & Barker, 2006; Kubitzki, 2007*; <<http://apsa.anu.edu.au/>>); *Hollandaea* F. Muell. (Weston & Barker, 2006; Kubitzki, 2007*; <<http://apsa.anu.edu.au/>>); *Isopogon* R. Br. ex Knight (Armstrong, 1979*; Collins & Rebelo, 1987*; Kubitzki, 2007*; Sauquet & Cantrill, 2007; <<http://www.paldat.org/>>); *Kermadecia* Brongn. & Gris (Erdtman, 1952; Memon, 1985; Weston & Barker, 2006; Kubitzki, 2007*); *Knighitia* R. Br. (Erdtman, 1952; Dettmann & Jarzen, 1998; Kubitzki, 2007*); *Lambertia* Sm. (Erdtman, 1952; Memon, 1985; Collins & Rebelo, 1987*; Maynard, 1995*; Kubitzki, 2007*); *Leucadendron* R. Br. (Collins & Rebelo, 1987*; Kubitzki, 2007*; Sauquet & Cantrill, 2007); Friedman & Barrett, 2008b*; <<http://www.paldat.org/>>); *Leucospermum* R. Br. (Collins & Rebelo, 1987*; Dettmann & Jarzen, 1998; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Lomatia* R. Br. (Feuer, 1989; Myerscough et al., 2000*; Kubitzki, 2007*); *Macadamia* F. Muell. (Erdtman, 1952; Memon, 1985; Heard, 1993*; Dettmann & Jarzen, 1998; Weston & Barker, 2006; Kubitzki, 2007*); *Malagasia* L. A. S. Johnson & B. G. Briggs (Memon, 1985; Weston & Barker, 2006; Kubitzki, 2007*); *Mimetes* Salisb. (Erdtman, 1952; Collins & Rebelo, 1987*; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Megahertzia* A. S. George & B. Hyland (Weston & Barker, 2006; Kubitzki, 2007*); *Musgravea* F. Muell. (Weston & Barker, 2006; Kubitzki, 2007*; <<http://apsa.anu.edu.au/>>); *Neorites* L. S. Sm. (Memon, 1985; Weston & Barker, 2006; Kubitzki, 2007*); *Opisthiolepis* L. B. Smith (Memon, 1985; Feuer, 1989; Kubitzki, 2007*); *Oreocallis* R. Br. (Johnson & Briggs, 1963*; Feuer, 1990; Kubitzki, 2007*); *Orites* R. Br. (Erdtman, 1952; Kubitzki, 2007*); *Orothamnus* Pappe ex Hook. (Memon, 1985; Collins & Rebelo, 1987*; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Panopsis* Salisb. ex Knight (Erdtman, 1952; Memon, 1985; Weston & Barker, 2006; Kubitzki, 2007*); *Paranomus* Salisb. (Collins & Rebelo, 1987*; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Personia* Sm. (Feuer, 1986; Collins & Rebelo, 1987*; Wallace et al., 2002*; Kubitzki, 2007*); *Petrophile* R. Br. ex Knight (Armstrong, 1979*; Memon, 1985; Collins & Rebelo, 1987*; Maynard, 1995*; Dettmann & Jarzen, 1998; Milne & Martin, 1998; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Placospermum* C. T. White (Memon, 1985; Feuer, 1986; Dettmann & Jarzen, 1998; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Protea* L. (Erdtman, 1952; Collins & Rebelo, 1987*; Fleming & Nicolson, 2002*; Kubitzki, 2007*; Sauquet & Cantrill, 2007); Steenhuisen & Johnson, 2012*; Steenhuisen et al., 2012*); *Roupala* Aubl. (Erdtman, 1952; Memon, 1985; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Serruria* Burm. ex Salisb. (Collins & Rebelo, 1987*; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Sleumerodendron* Virot (Memon, 1985; Weston & Barker, 2006; Kubitzki, 2007*); *Sorocephalus* R. Br. (Collins & Rebelo, 1987*; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Spatalla* Salisb. (Collins & Rebelo, 1987*; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Sphalmium* B. G. Briggs, B. Hyland & L. A. S. Johnson (Memon, 1985; Feuer, 1986; Dettmann & Jarzen, 1998; Kubitzki, 2007*); *Stenocarpus* R. Br. (Erdtman, 1952; Memon, 1985; Feuer, 1989; Myerscough et al., 2000*; Kubitzki, 2007*); *Stirlingia* Endl. (Memon, 1985; Collins & Rebelo, 1987*; Milne & Martin, 1998; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Strangea* Meisn. (Feuer, 1989; Kubitzki, 2007*); *Symphionema* R. Br. (Milne & Martin, 1998; Myerscough et al., 2000*; Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Synaphea* R. Br. (Collins & Rebelo, 1987*; Milne & Martin, 1998; Kubitzki, 2007*; Sauquet & Cantrill, 2007; Ye et al., 2012*); *Telopea* R. Br. (Feuer, 1990; Goldingay & Whelan, 1993*; Maynard, 1995*; Kubitzki, 2007*); *Toronia* L. A. S. Johnson & B. G. Briggs (Memon, 1985; Feuer, 1986;

Kubitzki, 2007*); *Triunia* L. A. S. Johnson & B. G. Briggs (Memon, 1985; Weston & Barker, 2006; Kubitzki, 2007*); *Turillia* A. C. Sm. (Weston & Barker, 2006; Kubitzki, 2007*); *Vexatorella* Rourke (Collins & Rebelo, 1987*); Kubitzki, 2007*; Sauquet & Cantrill, 2007); *Virotia* L. A. S. Johnson & B. G. Briggs (Memon, 1985; Weston & Barker, 2006; Kubitzki, 2007*); *Xylomelum* Sm. (Erdtman, 1952; Ladd & Connell, 1994*; Myerscough et al., 2000*; Weston & Barker, 2006; Kubitzki, 2007*).

Ranunculales/Berberidaceae*Achlys* DC. (Erdtman, 1952; Fukuda, 1967*; Nowicke & Skvarla, 1981, 1982; Blackmore et al., 1995; Fay & Christenhusz, 2012*); *Berberis* L. (Nowicke & Skvarla, 1981; Chang & Wang, 1983; Blackmore & Heath, 1984; Kubitzki et al., 1993*; Blackmore et al., 1995; Husain et al., 1998; Furness, 2008; Perveen & Qaiser, 2010; <<http://www.paldat.org/>>; <<http://www.efloras.org/>>*); *Bongardia* C. A. Mey. (Nowicke & Skvarla, 1981; Kubitzki et al., 1993*; Blackmore et al., 1995; Fay & Christenhusz, 2012*); *Caulophyllum* Michx. (Nowicke & Skvarla, 1981; Brett & Posluszny, 1982*; Chang & Wang, 1983; Kubitzki et al., 1993*; Hannan & Prucher, 1996*; Furness, 2007; Endress, 2010*); <<http://www.efloras.org/>>*); *Diphylleia* Michx. (Erdtman, 1952; Nowicke & Skvarla, 1979, 1981; Chang & Wang, 1983; Kubitzki et al., 1993*; Ma & Hu, 1997*; Guan et al., 2010*); Fay & Christenhusz, 2012*); <<http://www.efloras.org/>>*); *Dysosma* Woodson (Ying, 1979; Nowicke & Skvarla, 1981; Chang & Wang, 1983; Su & Liu, 1994; Ma & Hu, 1997*; Guan et al., 2010; Guan et al., 2010*); <<http://www.efloras.org/>>*); *Epimedium* L. (Nowicke & Skvarla, 1981, 1982; Suzuki, 1983, 1984*; Chang & Wang, 1983; Blackmore & Heath, 1984; Liang & Yan, 1991; Yu & Liu, 1991; Kubitzki et al., 1993*; Guo et al., 1998; Guan et al., 2010; <<http://www.efloras.org/>>*); *Gymnospermium* Spach (Nowicke & Skvarla, 1981; W. Wang et al., 2009; <<http://www.efloras.org/>>*); *Jeffersonia* Barton (Nowicke & Skvarla, 1979, 1982; Chang & Wang, 1983; Smith et al., 1986*; Fenster & Martén-Rodríguez, 2007*; Fay & Christenhusz, 2012*); *Mahonia* Nutt. (Nowicke & Skvarla, 1981; Chang & Wang, 1983; Blackmore & Heath, 1984; Kubitzki et al., 1993*; Blackmore et al., 1995; Halbritter & Hesse, 1995; Allen et al., 2006*; Furness, 2007, 2008; <<http://www.paldat.org/>>; <<http://www.efloras.org/>>*); *Nandina* Thunb. (Erdtman, 1952; Nowicke & Skvarla, 1981; Chang & Wang, 1983; Kubitzki et al., 1993*; <<http://www.paldat.org/>>; <<http://www.efloras.org/>>*); *Plagiorhegma* Maxim. (Nowicke & Skvarla, 1981; W. Wang et al., 2009; <<http://www.efloras.org/>>*); *Podophyllum* L. (Erdtman, 1952; Ying, 1979; Nowicke & Skvarla, 1981; Chang & Wang, 1983; Blackmore et al., 1995; Ma & Hu, 1997*; Guan et al., 2010*); Fay & Christenhusz, 2012*); *Ranzania* T. Ito (Erdtman, 1952; Nowicke & Skvarla, 1981; Kubitzki et al., 1993*; Blackmore et al., 1995; Furness, 2007); *Sinopodophyllum* T. S. Ying (Ying, 1979; Chang & Wang, 1983; Ma & Hu, 1997*; W. Wang et al., 2009; <<http://www.efloras.org/>>*); *Vancouveria* C. Morren & Decne. (Erdtman, 1952; Nowicke & Skvarla, 1981, 1982; Kubitzki et al., 1993*; W. Wang et al., 2009; Fay & Christenhusz, 2012*); **Circaeasteraceae***Circaeaster* Maxim. (Nowicke & Skvarla, 1982; Kubitzki et al., 1993*; Furness et al., 2007; Fay & Christenhusz, 2012*); *Kingdonia* Balf. f. & W. W. Sm. (Nowicke & Skvarla, 1982; Zhang, 1983; He et al., 2006*; Furness et al., 2007; Fay & Christenhusz, 2012*); **Eupteleaceae***Euptelea* Siebold & Zucc. (Walker, 1974; Pragłowski, 1975; Zavada & Dilcher, 1986; Kubitzki et al., 1993*; Wang et al., 1995; Furness et al., 2007; Endress, 2010*; Li et al., 2011; Fay & Christenhusz, 2012*); **Lardizabalaceae***Akebia* Decne. (Erdtman, 1952; Nowicke & Skvarla, 1982; Xia &

Peng, 1989; Kubitzki et al., 1993*; Blackmore et al., 1995; Wang et al., 1995; Wei & Li, 1995; Kawagoe & Suzuki, 2002*; Furness et al., 2007; Li et al., 2011; Christenhusz, 2012*); *Archakebia* C. Y. Wu, T. C. Chen & H. N. Qin (Wei & Li, 1995; Wang & Li, 2002; W. Wang et al., 2009; W. Wang et al., 2009*); *Boquila* Decne. (Nowicke & Skvarla, 1982; Kubitzki et al., 1993*; Wang & Li, 2002; W. Wang et al., 2009; Christenhusz, 2012*); Fay & Christenhusz, 2012*); *Decaisnea* Hook. f. & Thomson (Nowicke & Skvarla, 1982; Xia & Peng, 1989; Xia & Kong, 1991; Kubitzki et al., 1993*; Wei & Li, 1995; Furness et al., 2007; Christenhusz, 2012*; <<http://www.paldat.org/>>); *Holboellia* Wall. (Nowicke & Skvarla, 1982; Xia & Peng, 1989; Kubitzki et al., 1993*; Wang et al., 1995; Wei & Li, 1995; <<http://www.paldat.org/>>); *Lardizabala* Ruiz & Pav. (Erdtman, 1952; Nowicke & Skvarla, 1982; Christenhusz, 2012*); Fay & Christenhusz, 2012*); *Parvatia* Decne. (Payne & Seago, 1968*; Wang & Li, 2002; W. Wang et al., 2009; W. Wang et al., 2009*); *Sargentodoxa* Rehder & E. H. Wilson (Nowicke & Skvarla, 1982; Kubitzki et al., 1993*; Wang et al., 1995; Wei & Li, 1995; Furness et al., 2007; H.-F. Wang et al., 2009; Christenhusz, 2012*); *Sinofranchetia* (Diels) Hemsl. (Xia & Peng, 1989; Xia & Kong, 1991; Kubitzki et al., 1993*; Wang et al., 1995; Wang & Li, 2002; Furness et al., 2007; Endress, 2010*; Christenhusz, 2012*); *Stauntonia* DC. (Xia & Peng, 1989; Xia & Kong, 1991; Kubitzki et al., 1993*; Wang et al., 1995; Christenhusz, 2012*); **Menispermaceae***Albertisia* Becc. (Ferguson, 1975; Kubitzki et al., 1993*; W. Wang et al., 2009*); *Anamirta* Colebr. (Thanikaimoni, 1968; Ferguson, 1978; Kubitzki et al., 1993*; W. Wang et al., 2009*); *Arcangelisia* Becc. (Thanikaimoni, 1968; Ferguson, 1978; W. Wang et al., 2009*); *Aspidocarya* Hook. f. & Thomson (Thanikaimoni, 1968; Kubitzki et al., 1993*; W. Wang et al., 2009*); *Cissampelos* L. (Thanikaimoni, 1968; Harley & Ferguson, 1982; Kubitzki et al., 1993*; W. Wang et al., 2009*); *Cocculus* DC. (Thanikaimoni, 1968; Harley & Ferguson, 1982; Kubitzki et al., 1993*; Li et al., 2011); *Cyclea* Arn. ex Wight (Harley & Ferguson, 1982; Kubitzki et al., 1993*; W. Wang et al., 2009*); *Diploclisia* Miers (Harley & Ferguson, 1982; Kubitzki et al., 1993*; Kato, 2005*; W. Wang et al., 2009*); *Hypserpa* Miers (Thanikaimoni, 1968; Harley & Ferguson, 1982; Kubitzki et al., 1993*; W. Wang et al., 2009*); *Menispermum* L. (Thanikaimoni, 1968; Harley & Ferguson, 1982; Kubitzki et al., 1993*; Furness et al., 2007; W. Wang et al., 2009*; Li et al., 2011); *Parabaena* Miers (Thanikaimoni, 1968; Kubitzki et al., 1993*; W. Wang et al., 2009*); *Pericampylus* Miers (Thanikaimoni, 1968; Harley & Ferguson, 1982; Kubitzki et al., 1993*; W. Wang et al., 2009*); *Pycnarrhena* Miers ex Hook. f. & Thomson (Thanikaimoni, 1968; Ferguson, 1975; Kubitzki et al., 1993*; W. Wang et al., 2009*); *Sinomenium* Diels (Thanikaimoni, 1968; Harley & Ferguson, 1982; Kubitzki et al., 1993*; W. Wang et al., 2009*; Endress, 2010*); *Stephania* Lour. (Thanikaimoni, 1968; Harley & Ferguson, 1982; Kubitzki et al., 1993*; Blackmore et al., 1995; Meng et al., 2012*); *Tinomiscium* Miers ex Hook. f. & Thomson (Thanikaimoni, 1968; Harley & Ferguson, 1982; Kubitzki et al., 1993*; W. Wang et al., 2009*); *Tinospora* Miers (Thanikaimoni, 1968; Kubitzki et al., 1993*; Blackmore et al., 1995; Furness et al., 2007; W. Wang et al., 2009*; Li et al., 2011); **Papaveraceae***Corydalis* DC. (Rachele, 1974; Kalis, 1979; Kubitzki et al., 1993*; Blackmore et al., 1995; Romero et al., 2003; Perveen & Qaiser, 2004; Fay & Christenhusz, 2012*); *Dicentra* Bernh. (Stern, 1962; Rachele, 1974; Kubitzki et al., 1993*; <<http://www.paldat.org/>>); *Eomecon* Hance (Erdtman, 1952; Kubitzki et al., 1993*; Blattner & Kadereit, 1999*; W. Wang et al., 2009);

- Hylomecon* Maxim. (Erdtman, 1952; Kubitzki et al., 1993*; Blattner & Kadereit, 1999*; W. Wang et al., 2009); *Hypocoum* L. (Rachele, 1974; Dahl, 1989; Kubitzki et al., 1993*; Blackmore et al., 1995; Salinas et al., 2003); *Maclaya* R. Br. (Rachele, 1974; Kubitzki et al., 1993*; Blattner & Kadereit, 1999*; W. Wang et al., 2009; Endress, 2010*); *Meconopsis* Vig. (Henderson, 1965; Kubitzki et al., 1993*; W. Wang et al., 2009); *Papaver* L. (Rachele, 1974; Kalis, 1979; Kubitzki et al., 1993*; Sharifnia et al., 2010; <<http://www.paldata.org/>>); *Pteridophyllum* Siebold & Zucc. (Erdtman, 1952; Rachele, 1974; Kubitzki et al., 1993*; W. Wang et al., 2009); *Sanguinaria* L. (Erdtman, 1952; Kubitzki et al., 1993*; Blattner & Kadereit, 1999*; W. Wang et al., 2009). **Ranunculaceae**/*Aconitum* L. (Vishnu-Mittre & Sharma, 1963; Clarke et al., 1991; Kubitzki et al., 1993*; Xi, 1993; Wang et al., 1995; Bosch et al., 1997*; Tolgor et al., 1997; Bosch & Waser, 1999*; Perveen & Qaiser, 2006; <<http://www.paldata.org/>>); *Actaea* L. (Pellmyr, 1985*; Clarke et al., 1991; Kubitzki et al., 1993*; Wang et al., 1993; Wang et al., 1995; <<http://www.paldata.org/>>); *Adonis* L. (Santisuk, 1979; Nowicke & Skvarla, 1982; Kubitzki et al., 1993*; Lee et al., 2004; Denisow & Wrzesień, 2006*; Perveen & Qaiser, 2006; Ghorbani et al., 2008; <<http://www.paldata.org/>>); *Anemoclema* (Franch.) W. T. Wang (Wang et al., 1995; W. Wang et al., 2009*); *Anemone* L. (Horowitz, 1976*; Nowicke & Skvarla, 1979, 1982; Santisuk, 1979; Fernández, 1986b; Clarke et al., 1991; Kubitzki et al., 1993*; Wang et al., 1995; Lu et al., 1999; Perveen & Qaiser, 2006; Baladehi et al., 2013; <<http://www.paldata.org/>>); *Anemonopsis* Siebold & Zucc. (Pellmyr, 1988*; Kubitzki et al., 1993*; Wang et al., 1993); *Aquilegia* L. (Clarke et al., 1991; Kubitzki et al., 1993*; Wang et al., 1995; Eckert & Schaefer, 1998*; Jürgens & Dötterl, 2004*; Perveen & Qaiser, 2006; Fay & Christenhusz, 2012*; <<http://www.paldata.org/>>); *Asteropyrum* J. R. Drumm. & Hutch. (Kubitzki et al., 1993*; Wang et al., 1995; Yuan & Yang, 2006); *Batrachium* (DC.) Gray (Nowicke & Skvarla, 1981; Kubitzki et al., 1993*; W. Wang et al., 2009*; Wang et al., 2010*); *Beesia* Balf. f. & W. W. Sm. (Pellmyr, 1984*; Kubitzki et al., 1993*; X. Wang et al., 1993; F. Wang et al., 1995); *Callianthemum* C. A. Mey. (Santisuk, 1979; Kubitzki et al., 1993*; <<http://www.paldata.org/>>); *Caltha* L. (Santisuk, 1979; Clarke et al., 1991; Kubitzki et al., 1993*; Wang et al., 1993; Wang et al., 1995; Jürgens & Dötterl, 2004*; Perveen & Qaiser, 2006; Furness et al., 2007; <<http://www.paldata.org/>>); *Cimicifuga* L. ex Wernisch. (Pellmyr, 1986*; Kubitzki et al., 1993*; Wang et al., 1993, 1995); *Clematis* L. (Vishnu-Mittre & Sharma, 1963; Nowicke & Skvarla, 1979, 1981; Clarke et al., 1991; Rawal & Pangtey, 1991*; Dohzono & Suzuki, 2002*; Perveen & Qaiser, 2006; Wang & Xie, 2007; Wang et al., 2010*; Li et al., 2011; <<http://www.paldata.org/>>); *Consolida* (DC.) Gray (Clarke et al., 1991; Bosch et al., 1997*; <<http://www.paldata.org/>>); *Coptis* Salisb. (Nowicke & Skvarla, 1979; Kubitzki et al., 1993*; Furness et al., 2007); *Delphinium* L. (Fernández, 1986a; Clarke et al., 1991; Kubitzki et al., 1993*; Bosch et al., 1997*; Bosch & Waser, 1999*; Perveen & Qaiser, 2006; Yildiz et al., 2009; <<http://www.paldata.org/>>); *Dichocarpum* W. T. Wang & P. G. Xiao (Kubitzki et al., 1993*; W. Wang et al., 2009); *Enemion* Raf. (Kubitzki et al., 1993*; W. Wang et al., 2009); *Eranthis* Salisb. (Nowicke & Skvarla, 1979; Clarke et al., 1991; Kubitzki et al., 1993*; Furness et al., 2007; W. Wang et al., 2009*; Rysiak & Żuraw, 2011*; <<http://www.paldata.org/>>); *Glaucidium* Siebold & Zucc. (Nowicke & Skvarla, 1981; Furness et al., 2007; W. Wang et al., 2009*); *Halerpestes* Greene (Kubitzki et al., 1993*; W. Wang et al., 2009; W. Wang et al., 2009*); *Helleborus* L. (Nowicke & Skvarla, 1983; Fernández, 1986a; Clarke et al., 1991; Kubitzki et al., 1993*; Canto et al., 2008*; <<http://www.paldata.org/>>); *Hepatica* Mill. (Nowicke & Skvarla, 1981; Motten, 1982*; Clarke et al., 1991; Kubitzki et al., 1993*; W. Wang et al., 2009*); *Hydrastis* J. Ellis (Nowicke & Skvarla, 1981, 1982; Kubitzki et al., 1993*; Furness et al., 2007; W. Wang et al., 2009*); *Isopyrum* L. (Melampy & Hayworth, 1980*; Clarke et al., 1991; Kubitzki et al., 1993*; <<http://www.paldata.org/>>); *Leptopyrum* Rehb. (Kubitzki et al., 1993*; W. Wang et al., 2009); *Megaleranthis* Ohwi (Kubitzki et al., 1993*; Blackmore et al., 1995; Jang & Heo, 2005; Jeong et al., 2010*); *Myosurus* L. (Santisuk, 1979; Clarke et al., 1991; W. Wang et al., 2009*; <<http://www.paldata.org/>>); *Naravelia* Adans. (Vishnu-Mittre & Sharma, 1963); *Nigella* L. (Fernández, 1986a; Clarke et al., 1991; Kubitzki et al., 1993*; Wang et al., 1995; Jørgensen et al., 2006*; <<http://www.paldata.org/>>); *Oxygraphis* Bunge (Santisuk, 1979; Kubitzki et al., 1993*; Wang et al., 1995; W. Wang et al., 2009*); *Paraquilegia* J. R. Drumm. & Hutch. (Vishnu-Mittre & Sharma, 1963; Kubitzki 1993*; Wang et al., 1995); *Paropyrum* Ulbr. (W. Wang et al., 2009; W. Wang et al., 2009*); *Pulsatilla* Mill. (Clarke et al., 1991; Huang et al., 2002*; Jürgens & Dötterl, 2004*; Baladehi et al., 2013; <<http://www.paldata.org/>>); *Ranunculus* L. (Nowicke & Skvarla, 1979, 1981; Santisuk, 1979; Clarke et al., 1991; Kubitzki et al., 1993*; Jürgens & Dötterl, 2004*; Perveen & Qaiser, 2006; Yang et al., 2006; Furness et al., 2007; <<http://www.paldata.org/>>); *Semiaquilegia* Makino (Kubitzki et al., 1993*; W. Wang et al., 2009); *Souliea* Franch. (Erdtman, 1952; Kubitzki et al., 1993*; Wang et al., 1993; Wang et al., 1995; Furness et al., 2007; W. Wang et al., 2009*); *Thalictrum* L. (Fernández, 1986b; Zhou et al., 1990; Clarke et al., 1991; Kubitzki et al., 1993*; Blackmore et al., 1995; Davis, 1997*; Tatlidil et al., 2005; Perveen & Qaiser, 2006; Friedman & Barrett, 2008*; W. Wang et al., 2009*; Fay & Christenhusz, 2012*; <<http://www.paldata.org/>>); *Trautvetteria* Fisch. & C. A. Mey. (Santisuk, 1979; W. Wang et al., 2009*); *Trollius* L. (Nowicke & Skvarla, 1979, 1982; Clarke et al., 1991; Lee & Blackmore, 1992; Kubitzki et al., 1993*; Wang et al., 1993; Blackmore et al., 1995; Jürgens & Dötterl, 2004*; Perveen & Qaiser, 2006; <<http://www.paldata.org/>>); *Urophyssa* Ulbr. (Kubitzki et al., 1993*; W. Wang et al., 2009); *Xanthorhiza* Marshall (Kubitzki et al., 1993*; Furness et al., 2007; W. Wang et al., 2009; Endress, 2010*).
- Sabiaceae**/*Meliosma* Blume (Mondal & Mitra, 1982; Furness et al., 2007; Kubitzki, 2007*; Ronse De Craene & Wanntorp, 2008*; Li et al., 2011); *Sabia* Colebr. (Mondal & Mitra, 1982; Furness et al., 2007; Kubitzki, 2007*; Ronse De Craene & Wanntorp, 2008*; Li et al., 2011).
- Trochodendrales/Trochodendraaceae**/*Tetracentron* Oliv. (Bailey & Nast, 1945; Praglowski, 1975; Zavada & Dilcher, 1986; Endress, 1989; Kubitzki et al., 1993*; Wang et al., 1995; Furness et al., 2007; Li et al., 2011); *Trochodendron* Siebold & Zucc. (Bailey & Nast, 1945; Praglowski, 1975; Zavada & Dilcher, 1986; Kubitzki et al., 1993*; Wang et al., 1995; Furness et al., 2007; Endress, 2010*).

Appendix 3. Comprehensively coded matrix of pollen morphological and ecological data for basal eudicots (characters are ordered as in Table 1).

<i>Achlys</i>	021130{0,1}300030071{4,5}011{0,3}0
<i>Acidonia</i>	0211{1,2}1{1,2}300130{0,1}{0,2,7}1{2,6}010?1
<i>Aconitum</i>	0211{2,3,4}0{0,1}{3,2,4}00030{0,1}{0,1,7}1{2,6}01100
<i>Acorus</i>	0100?20120030071{2,3}01100
<i>Actaea</i>	0211{1,2,3}01{3,4}00030111201100
<i>Adenanthos</i>	0{1,2}1101{0,1}30013?071{2,4,6}011{0,1,2}{1,3}
<i>Adonis</i>	0211{2,3,4}0{0,1}{3,4}00030111201100
<i>Agastachys</i>	0211{0,1}11{2,3}001301{2,5}120???{1,3}
<i>Akebia</i>	0211{3,4}0{0,1}30{0,1}0?0071201102
<i>Albertisia</i>	0211200{3,4}000300716011?2
<i>Alisma</i>	001120{0,1}{5,2}{0,1}0130111{2,6}011{0,3}0
<i>Alloxylon</i>	0211?1?30013???????23
<i>Anamirta</i>	0211200301020071{2,6}011?2
<i>Anemoclema</i>	0011201{4,5}100301117???00
<i>Anemone</i>	0{0,2}11{1,2,3,4}0{0,1}{3,4,5}{0,1}0{0,1,2,3,4}301{0,1}1201100
<i>Anemonopsis</i>	021130?300030111{2,3}00100
<i>Aquilegia</i>	0211{2,3,4}00300030111{2,3}011{0,2}0
<i>Arcangelisia</i>	0211400301020071{2,6}011?2
<i>Archakebia</i>	0211?0030003???1201102
<i>Aspidocarya</i>	021120130{0,1}0{2,3}0??160?1?2
<i>Asteropyrum</i>	0011201{4,5}10{0,1}301113???00
<i>Athertonia</i>	0211?1130013???????23
<i>Aulax</i>	0211111300130111{2,6}11001
<i>Austromuelleria</i>	020102020013???????23
<i>Banksia</i>	020102{0,1}2001300712???{0,1,2}{1,3}
<i>Batrachium</i>	02113013000301112011{0,5}0
<i>Beauprea</i>	021101{1,2}300130{0,1}{0,5,7}1{0,2,6,7}010?{1,3}
<i>Beaupeopsis</i>	0{1,2}1101{0,1}300130071{2,7}??0?1
<i>Beesia</i>	0211201{3,4}00030111{2,3}00100
<i>Bellendenia</i>	0211{0,1}1{0,1}{3,4}00130071{2,6}010?1
<i>Berberis</i>	00{1,2}1{2,3}0{0,1,2}{1,5}10{0,2,3}30071{2,6}20101
<i>Bleasdalea</i>	0211?1?30013???????23
<i>Bongardia</i>	0211302300030071{2,6}011?0
<i>Boquila</i>	0211?01300030071201102
<i>Brabejum</i>	021101130013???????0{1,3}
<i>Buckinghamia</i>	021101{1,2}300130071{2,3}011?3
<i>Buxus</i>	0{0,2}11{1,2}0{0,1}{3,5}{0,1}{0,1}{0,1}{0,3}0{0,1}{1,2,5}1{2,6}011{0,3}{1,3}
<i>Callianthemum</i>	0{0,2}11201{3,4,5}{0,1}0030111201100
<i>Caltha</i>	0211{1,2,3}0{0,1}{3,4}00030111{2,6}01100
<i>Cardwellia</i>	0211{0,1}1130013???16????3
<i>Carnarvonia</i>	0211{0,1}1{0,1}300130071{2,6}011?3
<i>Catalepidia</i>	0211?1?30013???????23
<i>Caulophyllum</i>	021130{1,2}3000300716011{0,5}0
<i>Cenarrhenes</i>	021101{0,1}30013?071{0,1,2,6,7}0???{1,3}
<i>Ceratophyllum</i>	0011{1,2}0{0,1}0325320713?0040
<i>Cimicifuga</i>	0211201300030111201100
<i>Circaeaster</i>	0211{2,3}003000300714??100
<i>Cissampelos</i>	021121{0,1}301001071601102
<i>Clematis</i>	0{0,2}11{2,3,4}0{0,1}{3,4,5}{0,1}0{0,1}30111{2,3}011{0,3}{0,1,2}
<i>Cocculus</i>	0211{3,4}0030100007120110{1,2,3}
<i>Conospermum</i>	0{1,2}11{0,1,2}1{1,2}30013?071{2,6}0110{1,3}
<i>Consolida</i>	0211{3,4}01300030111201100
<i>Coptis</i>	001120?510130111201130
<i>Corydalis</i>	0211{2,3,4}0{0,1}{3,4,5}{0,1}0{0,3}30{0,1}{5,7}1{1,2,7}0??00
<i>Cyclea</i>	02112103010010716011?2
<i>Darlingia</i>	021101{0,1}30013?????0??23
<i>Decaisnea</i>	0211{2,3,4}01300030071201101
<i>Delphinium</i>	0211{2,3,4}0{0,1}3000301112011{0,2}0
<i>Diastella</i>	0{1,2}1101{0,1}300130071601101

Appendix 3. Continued.

<i>Dicentra</i>	0211{2,3}0{0,1}{2,3,4,5}00{0,3}30{0,1}{5,7}1{1,2,6,7}01100
<i>Dichocarpum</i>	0211?0?{3,4,5}0003?11???1?00
<i>Didymeles</i>	0211{1,2}003010011{1,2}16011?3
<i>Dillenia</i>	021120{0,1}{2,3,4}0{0,1}0?007160110{1,3}
<i>Dilobeia</i>	0211010300130{0,1}01{0,2}111?3
<i>Diphylleia</i>	0211{1,2}01300030111{2,7}011?0
<i>Diploclisia</i>	0211{3,4}003010{0,2}007120??02
<i>Dryandra</i>	020102{1,2}200130???????{0,1,2}{1,3}
<i>Dyosma</i>	0211{2,3,4}0{1,2}300030071{2,4,5,6,7}011{0,5}0
<i>Eidothea</i>	021101{1,2}300130071{2,6}010?3
<i>Embothrium</i>	020102{1,2}200130111{1,2}311{0,2}{1,3}
<i>Enemion</i>	0211?0?30003?11???1?00
<i>Eomecon</i>	001120?51013???16?1100
<i>Epimedium</i>	0211{2,3,4}0{0,1}300030{0,1}{0,1,5}1{1,2,4,5,6,7}011{0,5}0
<i>Eranthis</i>	0211{2,3}01300030111201100
<i>Eucarpha</i>	0211?1{0,1}30013????????{1,3}
<i>Euplassa</i>	0211011{3,4}0013???16????{1,3}
<i>Euptelea</i>	0{0,1,2}{1,2}1{2,3,4}01{3,4}{0,1}0030{0,1}{1,7}1601133
<i>Faurea</i>	0{1,2}1101{0,1,2}30013?071{0,2}110?3
<i>Floydia</i>	0211?1030013?????????3
<i>Franklandia</i>	0211{1,2}{0,1}{2,3}3001300709010?1
<i>Garnieria</i>	02110113001301012310?{1,3}
<i>Gevuina</i>	021101130013?071{2,6}????3
<i>Glaucidium</i>	02113013000301{0,1}1201100
<i>Grevillea</i>	0211{0,2}1{1,2}{3,4}001301512???{0,1,2}{1,3}
<i>Gunnera</i>	0211{0,1,2}0{0,1}300030071601130
<i>Gymnospermium</i>	02113023000300716????00
<i>Hakea</i>	021101{1,2,3}3001301{0,5}1{2,6}???{0,1,2}{1,3}
<i>Halerpestes</i>	0211?0?30003?11???1?00
<i>Helicia</i>	021101{0,1}3001300717???0{1,3}
<i>Heliciopsis</i>	0211?1{1,2}30013????????03
<i>Helleborus</i>	0211{1,2,3}0{0,1}300030{0,1}{0,1,7}1{2,6}01100
<i>Hepatica</i>	0{0,2}11{2,3}01{2,3,5}{0,1}00301112011{0,5}0
<i>Hicksbeachia</i>	021101{0,1,2}300130??16????3
<i>Holboellia</i>	0211{2,3,4}0{0,1}30{0,1}030071201102
<i>Hollandaea</i>	021101{0,1}300130??16????{1,3}
<i>Hydrastis</i>	0211{2,3}01300030071{4,5}011{0,5}0
<i>Hylomecon</i>	021120130003??16????00
<i>Hypecoum</i>	020120{0,1}200030111201100
<i>Hypserpa</i>	0211{2,3,4}003010{0,2}0071201102
<i>Isopogon</i>	021101{0,1,2}300130071{2,6}411{0,1}{1,3}
<i>Isopyrum</i>	0211200300030111201100
<i>Jeffersonia</i>	0211{3,4}01300030071{4,5}01100
<i>Kermadecia</i>	0211111{1,2}30013???16????3
<i>Kingdonia</i>	0211{2,3}01300030071{4,5}01100
<i>Knightia</i>	0211011300130??1{2,6}??1?3
<i>Lambertia</i>	021101{0,1,2}300130111?0??{0,2}{1,3}
<i>Lardizabala</i>	021140130{0,1}0?0071201102
<i>Leptopyrum</i>	0211?0?30003?11????00
<i>Leucadendron</i>	0{1,2}1101{1,2}3001301{0,1}1{0,1,2,6,7}411{0,3}{1,3}
<i>Leucospermum</i>	011101{1,2}300130{0,1}{0,7}1{2,6}411{0,1,2}{1,3}
<i>Lomatia</i>	02{0,1}1{0,1}1{0,1}{2,3}00130071{2,6}0110{1,3}
<i>Macadamia</i>	0211?1{0,1}300130071201103
<i>Macleaya</i>	001120051013?1016?1?30
<i>Mahonia</i>	00{1,2}120{1,2}{1,5}10{0,2,3}30071{2,6}20101
<i>Malagasia</i>	0211?1030013????????03
<i>Meconopsis</i>	0{0,2}11{1,2,3}0{0,1}{0,3,4,5}{0,1,3}{0,2}{0,5}3?111601?00
<i>Megahertzia</i>	0211?1?30013?????????3
<i>Megaleranthis</i>	021120?30003??14????00

Appendix 3. Continued.

<i>Meliosma</i>	0211{2,3,4}0{0,1}3010{0,1}0071{2,6}01103
<i>Menispermum</i>	0211{2,3}003010000712011?2
<i>Mimetes</i>	0{1,2}11011300130071{2,6}4112{1,3}
<i>Musgravea</i>	0201?2020013???16????3
<i>Myosurus</i>	021120030003011120????0
<i>Myrothamnus</i>	2211{1,2}0030013?0709011{0,3}1
<i>Nandina</i>	0211{2,3}01300030071{2,6}011?1
<i>Naravelia</i>	001120051013???16????2
<i>Nelumbo</i>	0{1,2}{0,1}{0,1}2{0,2}{1,2}{1,2,3}{0,2}0{0,2,4}30071{0,2,6}01100
<i>Neorites</i>	0211?1030013????????3
<i>Nigella</i>	0211{2,3}01{3,4}00030111201100
<i>Opisthiolepis</i>	0211{0,1}1{0,1}3001300712011?3
<i>Oreocallis</i>	02{1,2}1{0,1,2}1{1,2,3}{2,3,4}001301{0,1,4,5}1{2,6}{1,3}112{1,3}
<i>Orites</i>	021101{0,1}30013????????{1,3}
<i>Orontium</i>	0100?2{1,2}120030071201100
<i>Orothamnus</i>	011101{0,1}300130071{2,6}411?1
<i>Oxygraphis</i>	021120130003011?????00
<i>Pachysandra</i>	00112015101301618?110{0,1}
<i>Panopsis</i>	021101{0,1}30013???16????{1,3}
<i>Papaver</i>	0{0,2}11{2,3,4}0{0,1}{3,4,5}{0,1}0{0,1}301{1,5}1201100
<i>Parabaena</i>	0211{1,2,3,4}0030003???160?102
<i>Paranomus</i>	0{1,2}1101{0,1}300130071{2,6,7}01101
<i>Paraquilegia</i>	021130{0,1}30003011160??00
<i>Paropyrum</i>	0211?0?30003?11???1?0
<i>Parvatia</i>	0211?0130003???12?11??
<i>Pericampylus</i>	0211{2,3}003010000?160??02
<i>Persoonia</i>	0211{0,1}1{1,2}300130{0,1}{0,3,4,5}12{0,4}100{1,3}
<i>Petrophile</i>	021101{0,1,2}300130071{2,6}010{0,1}1
<i>Placospermum</i>	0211{0,1}1{1,2}{3,4}001300712011?3
<i>Plagiorhegma</i>	0211{3,4}01300030071{4,5}011?0
<i>Platanus</i>	0211{2,3,4}0{0,1}{3,4}00030071601133
<i>Podophyllum</i>	{0,2}21120{0,1}300030{0,1}{0,1,5}1{2,4,5,7}01100
<i>Potamogeton</i>	0011{0,1,2}0{0,1}{0,1}32532111601130
<i>Protea</i>	0{1,2}1101{0,1}300130071{0,2,7}110{0,1,2}{1,3}
<i>Pteridophyllum</i>	0211{2,3}0{0,1}{2,3,4}0003?111?0??00
<i>Pulsatilla</i>	0211{1,2,3}01{3,4,5}{0,1}0030111201100
<i>Pycnarrhena</i>	0211{3,4}00301020071{2,6}01102
<i>Ranunculus</i>	0{0,2}11{1,2,3}0{0,1,2}{3,4,5}{0,1}0{0,1,3}301{1,5}1201100
<i>Ranzania</i>	{0,1}211201{0,4,5}{1,3}{0,2}{0,5}30071{0,1,2}011?0
<i>Roupala</i>	021101{0,1,2}30013????????{1,3}
<i>Sabia</i>	0211{2,3,4}0{0,1}3010{0,1}0071{2,6}011?{1,2}
<i>Sanguinaria</i>	00112015101301116????00
<i>Sargentodoxa</i>	0211{3,4}0{0,1}{2,3}00{0,3}30071201102
<i>Semiaquilegia</i>	0211?0?30003?111??1?00
<i>Serruria</i>	0{1,2}1101{0,1}300130071{2,4,6}01101
<i>Sinofranchetia</i>	0211{2,3}0{0,1}30{0,1}0{1,3}0??1201102
<i>Sinomenium</i>	021130030102007120?102
<i>Sinopodophyllum</i>	2211201300030151??1150
<i>Sleumerodendron</i>	0211?1130013????????3
<i>Sorocephalus</i>	0211011300130071{2,4}01101
<i>Souliea</i>	0011201032532111501100
<i>Spatalla</i>	021101{0,1}300130071{2,6}01101
<i>Sphalmium</i>	0211{0,1}1{1,2}{3,4}00130071{2,6}011?3
<i>Stauntonia</i>	0211{2,3,4}0{0,1}30{0,1}0{1,3}0{0,1}{0,5}1{2,6}01102
<i>Stenocarpus</i>	02{1,2}1{0,1}1{0,1,2}{3,4}00130071{2,6}{0,4}110{1,3}
<i>Stephania</i>	0211110{3,4}0{0,1}1{0,2}0071601102
<i>Stirlingia</i>	021101{0,1,2}300130071{0,2,6,7}01001
<i>Strangea</i>	021101{1,2}300130071{2,6}011?1
<i>Symphionema</i>	021101{0,1}300130071{4,6}01?01

Appendix 3. Continued.

<i>Synaphea</i>	0211012300130071{2,6}0??{0,3}1
<i>Telopea</i>	0211{0,1}12300130{0,1}{1,5,7}1{2,6}{1,3}11{0,2}{1,3}
<i>Tetracentron</i>	02{1,2}1{2,3}00300030071{4,5,6}01103
<i>Thalictrum</i>	001120{0,1}{4,5}101301{0,1}12011{0,3}0
<i>Tinomiscium</i>	0211{2,3}003010200712011?2
<i>Tinospora</i>	0211{3,4}0{0,1}30{0,1}{0,3}2107160?102
<i>Tofieldia</i>	0211{2,3,4}2{0,1}200030{0,1}{0,5,7}1{2,6}01?00
<i>Toronia</i>	0211{0,1}1{1,2}300130101{2,6}010?{1,3}
<i>Trautvetteria</i>	021120?300030111201??0
<i>Triunia</i>	0211?1?30013????????{1,3}
<i>Trochodendron</i>	0211{1,2}0{0,1}300030071{5,6}011{0,3}{1,3}
<i>Trollius</i>	0211{2,3,4}0{0,1}{2,3,4}00030071{2,4}01100
<i>Turrillia</i>	0211?1?30013????????3
<i>Urophysa</i>	0211?0?30003?11??1?00
<i>Vancouveria</i>	0211201300030071{4,5}011?0
<i>Vexatorella</i>	0{1,2}11011300130071{2,7}01101
<i>Vrotia</i>	0211?1{0,1}30013????????{1,3}
<i>Xanthorhiza</i>	00112005101301112?11?1
<i>Xylomelum</i>	0211{0,1}1{1,2}30013?11?????0{1,3}

Appendix 4. Democratically coded matrix of pollen morphological data for basal eudicots (characters are ordered as in Table 1).

<i>Achlys</i>	02113013000300513011
<i>Acidonia</i>	02111123001301211010
<i>Aconitum</i>	02113013000301111011
<i>Acorus</i>	0100?201200300511011
<i>Actaea</i>	02112013000301111011
<i>Adenanthos</i>	021101130013?0511011
<i>Adonis</i>	02112013000301111011
<i>Agastachys</i>	021111130013012110??
<i>Akebia</i>	02113003010?00511011
<i>Albertisia</i>	02112003000300515011
<i>Alisma</i>	00112015101301111011
<i>Alloxylon</i>	0211?1?30013?????????
<i>Anamirta</i>	02112003010200511011
<i>Anemoclema</i>	00112015100301116???
<i>Anemone</i>	02112013000301111011
<i>Anemonopsis</i>	021130?3000301111001
<i>Aquilegia</i>	02112003000301111011
<i>Arcangelisia</i>	02114003010200511011
<i>Archakebia</i>	0211?0030003???11011
<i>Aspidocarya</i>	0211201301020??150?1
<i>Asteropyrum</i>	00112015101301112???
<i>Athertonia</i>	0211?1130013?????????
<i>Aulax</i>	02111113001301111110
<i>Austromuelleria</i>	020102020013?????????
<i>Banksia</i>	02010212001300511???
<i>Batrachium</i>	02113013000301111011
<i>Beauprea</i>	02110113001300511010
<i>Beaupreopsis</i>	01110103001300511?0
<i>Beesia</i>	02112013000301111001
<i>Bellendenia</i>	02110113001300511010
<i>Berberis</i>	00212011102300511201
<i>Bleasdalea</i>	0211?1?30013?????????
<i>Bongardia</i>	02113023000300515011
<i>Boquila</i>	0211?013000300511011
<i>Brabejum</i>	021101130013?????????
<i>Buckinghamia</i>	02110123001300511011
<i>Buxus</i>	00112015101301115011
<i>Callianthemum</i>	02112013000301111011
<i>Caltha</i>	02112013000301111011
<i>Cardwellia</i>	021101130013???15???
<i>Carnarvonia</i>	02110113001300515011
<i>Catalpidia</i>	0211?1?30013?????????
<i>Caulophyllum</i>	02113023000300515011
<i>Cenarrhenes</i>	021101130013?05110??
<i>Ceratophyllum</i>	00112010324320512?00
<i>Cimicifuga</i>	02112013000301111011
<i>Circaeaster</i>	02113003000300513??1
<i>Cissampelos</i>	02112103010010515011
<i>Clematis</i>	02112003000301111011
<i>Cocculus</i>	02113003010000511011
<i>Conospermum</i>	021111230013?0511011
<i>Consolida</i>	02113013000301111011
<i>Coptis</i>	001120?5101301111011
<i>Corydalis</i>	021120141003005110??
<i>Cyclea</i>	02112103010010515011
<i>Darlingia</i>	021101130013?????0??
<i>Decaisnea</i>	02113013000300511011

Appendix 4. Continued.

<i>Delphinium</i>	02113013000301111011
<i>Diastella</i>	02110113001300515011
<i>Dicentra</i>	02112013000300511011
<i>Dichocarpum</i>	0211?0?30003?11??1?
<i>Didymeles</i>	02112003010011115011
<i>Dillenia</i>	02112003000300515011
<i>Dilobeia</i>	02110103001300510111
<i>Diphylleia</i>	02111013000301116011
<i>Diploclisia</i>	021130030100005110??
<i>Dryandra</i>	0201022200130???????
<i>Dysosma</i>	02112013000300515011
<i>Eidothea</i>	02110123001300511010
<i>Embothrium</i>	02010222001301111311
<i>Enemion</i>	0211?0?30003?11??1?
<i>Eomecon</i>	001120?51013???15?11
<i>Epimedium</i>	02113013000301014011
<i>Eranthis</i>	02112013000301111011
<i>Eucarpha</i>	0211?1?30013?????????
<i>Euplassa</i>	021101130013???15???
<i>Euptelea</i>	0111201400301115011
<i>Faurea</i>	021101030013?05111110
<i>Floydia</i>	0211?1030013?????????
<i>Franklandia</i>	02112023001300508010
<i>Garnieria</i>	02110113001301011310
<i>Gevuina</i>	021101130013?0515???
<i>Glaucidium</i>	02113013000301111011
<i>Grevillea</i>	02110123001301311???
<i>Gunnera</i>	02111013000300515011
<i>Gymnospermium</i>	02113023000300515???
<i>Hakea</i>	02110123001301311???
<i>Halerpestes</i>	0211?0?30003?11??1?
<i>Helicia</i>	02110103001300516???
<i>Heliciopsis</i>	0211?1?30013?????????
<i>Helleborus</i>	02112013000301015011
<i>Hepatica</i>	00112015100301111011
<i>Hicksbeachia</i>	0211012300130?15???
<i>Holboellia</i>	02113003000300511011
<i>Hollandaea</i>	0211010300130??15???
<i>Hydrastis</i>	02112013000300514011
<i>Hylomecon</i>	021120130003???15???
<i>Hypocoum</i>	02012012000301111011
<i>Hypserpa</i>	02113003010000511011
<i>Isopogon</i>	02110113001300515411
<i>Isopyrum</i>	02112003000301111011
<i>Jeffersonia</i>	02113013000300513011
<i>Kermadecia</i>	021111130013???15???
<i>Kingdonia</i>	02113013000300513011
<i>Knightia</i>	0211011300130??11??1
<i>Lambertia</i>	0211012300130111?0??
<i>Lardizabala</i>	02114013000300511011
<i>Leptopyrum</i>	0211?0?30003?11?????
<i>Leucadendron</i>	02110113001301011411
<i>Leucospermum</i>	01110113001300515411
<i>Lomatia</i>	02110113001300515011
<i>Macadamia</i>	0211?113001300511011
<i>Macleaya</i>	001120051013?1015?1?
<i>Mahonia</i>	00112015103300511201
<i>Malagasia</i>	0211?1030013?????????
<i>Meconopsis</i>	021120030003?111501?

Appendix 4. Continued.

<i>Megahertzia</i>	0211?1?30013????????
<i>Megaleranthis</i>	021120?30003???13???
<i>Meliosma</i>	02113013010100515011
<i>Menispermum</i>	02113003010000511011
<i>Mimetes</i>	02110113001300511411
<i>Musgravea</i>	0201?2020013???15???
<i>Myosurus</i>	021120030003011110??
<i>Myrothamnus</i>	221120030013?0508011
<i>Nandina</i>	02112013000300515011
<i>Naravelia</i>	001120051013???15???
<i>Nelumbo</i>	02112013000300511011
<i>Neorites</i>	0211?1030013????????
<i>Nigella</i>	02112013000301111011
<i>Opisthiolepis</i>	02111113001300511011
<i>Oreocallis</i>	02110123001301111111
<i>Orites</i>	021101130013????????
<i>Orontium</i>	0100?221200300511011
<i>Orothamnus</i>	01110113001300511411
<i>Oxygraphis</i>	021120130003011?????
<i>Pachysandra</i>	00112015101301417?11
<i>Panopsis</i>	021101130013???15???
<i>Papaver</i>	02112013000301311011
<i>Parabaena</i>	021130030003???150?1
<i>Paranomus</i>	02110113001300511011
<i>Paraquilegia</i>	021130130003011150??
<i>Paropyrum</i>	0211?0?30003?11????1?
<i>Parvatia</i>	0211?0130003???11?11
<i>Pericampylus</i>	021120030100005150??
<i>Persoonia</i>	02111113001301011010
<i>Petrophile</i>	02110113001300515010
<i>Placospermum</i>	02110113001300511011
<i>Plagiorhegma</i>	02113013000300513011
<i>Platanus</i>	02113013000300515011
<i>Podophyllum</i>	02112013000301111011
<i>Potamogeton</i>	00111010324321115011
<i>Protea</i>	02110103001300511110
<i>Pteridophyllum</i>	021120130003?111?0??
<i>Pulsatilla</i>	02112013000301111011
<i>Pycnarrhena</i>	02114003010200515011
<i>Ranunculus</i>	02112013000301111011
<i>Ranzania</i>	12112015100300511011
<i>Roupala</i>	021101130013????????
<i>Sabia</i>	02113013010100515011
<i>Sanguinaria</i>	00112015101301115???
<i>Sargentodoxa</i>	02113013000300511011
<i>Semiaquilegia</i>	0211?0?30003?111??1?
<i>Serruria</i>	02110113001300511011
<i>Sinofranchetia</i>	0211200300030?11011
<i>Sinomenium</i>	021130030102005110?1
<i>Sinopodophyllum</i>	2211201300030131??11
<i>Sleumerodendron</i>	0211?1130013????????
<i>Sorocephalus</i>	02110113001300511011
<i>Souliea</i>	00112010324321114011
<i>Spatalla</i>	02110113001300515011
<i>Sphalmium</i>	02110123001300515011
<i>Stauntonia</i>	02113003000301315011
<i>Stenocarpus</i>	02110113001300511011
<i>Stephania</i>	02111103001300515011
<i>Stirlingia</i>	02110123001300511010

Appendix 4. Continued.

<i>Strangea</i>	02110113001300511011
<i>Symphionema</i>	0211011300130051501?
<i>Synaphea</i>	021101230013005110??
<i>Telopea</i>	02111123001301111111
<i>Tetracentron</i>	02112003000300514011
<i>Thalictrum</i>	00112005101301111011
<i>Tinomiscium</i>	02113003010200511011
<i>Tinospora</i>	021140030102105150?1
<i>Tofieldia</i>	0211320200030051501?
<i>Toronia</i>	02111123001301011010
<i>Trautvetteria</i>	021120?300030111101?
<i>Triunia</i>	0211?1030013????????
<i>Trochodendron</i>	02112003000300515011
<i>Trollius</i>	02113003000300513011
<i>Turrillia</i>	0211?1?30013????????
<i>Urophysa</i>	0211?0?30003?11??1?
<i>Vancouveria</i>	02112013000300514011
<i>Vexatorella</i>	02110113001300511011
<i>Virotia</i>	0211?1?30013????????
<i>Xanthorrhiza</i>	00112005101301111?11
<i>Xylomelum</i>	021101130013?11?????