# REINTERPRETATION OF THE MID-CRETACEOUS FOSSIL FLOWER *ENDOBEUTHOS PALEOSUM* AS A CAPITULAR, UNISEXUAL INFLORESCENCE OF PROTEACEAE

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## ABSTRACT

The Myanmar amber fossil *Endobeuthos paleosum* was originally described as composed of an individual flower with a calyx of numerous, helically arranged sepals, a whorl of petals, and 60+ stamens each bearing a single bisporangiate anther. The 6 flowers, embedded together in a single block of amber, were described as varying in their calyx pubescence and length of corolla segments. The numerous stamens, with their single anther, led to a hypothesized relationship with certain members of family Dilleniaceae. We now propose a complete reinterpretation of this fossil as being an involucrate capitulum of family Proteaceae, in which the numerous "stamens" are identified instead as staminate flowers, although of reduced and highly modified morphology. Organs previously called the calyx and corolla are instead a series of helically-arranged bracts that surround the tight cluster of flowers. The Proteaceae being a diverse and significant element in Southern Hemisphere floras, the reinterpretation of *Endobeuthos* is important in providing the first Cretaceous fossil flower identified for the family, dated at some 20 my younger than the proposed Proteaceae crown group age of 119 Mya.

## RESUMEN

El fósil de ámbar de Myanmar *Endobeuthos paleosum* se describió originalmente como compuesto por una flor individual con un cáliz de numerosos sépalos dispuestos helicoidalmente, un verticilo de pétalos y más de 60 estambres, cada uno con una única antera bisporangiada. Las 6 flores, incrustadas juntas en un único bloque de ámbar, se describieron como variables en cuanto a la pubescencia del cáliz y la longitud de los segmentos de la corola. Los numerosos estambres, con su única antera, llevaron a hipotetizar una relación con ciertos miembros de la familia Dilleniaceae. Ahora proponemos una reinterpretación completa de este fósil como un capítulo involucrado de la familia Proteaceae, en el que los numerosos "estambres" se identifican en cambio como flores estaminadas, aunque de morfología reducida y muy modificada. Los órganos anteriormente denominados cáliz y corola son en cambio una serie de brácteas dispuestas helicoidalmente que rodean el apretado racimo de flores. Siendo las Proteáceas un elemento diverso y significativo en las floras del Hemisferio Sur, la reinterpretación de *Endobeuthos* es importante al proporcionar la primera flor fósil del Cretácico identificada para la familia, datada en unos 20 my más joven que la edad propuesta para el grupo de la corona de las Proteáceas de 119 Mya.

## INTRODUCTION

At the time of publication of *Endobeuthos paleosum* (Poinar & Chambers 2018), the authors were puzzled by its head-like flower having the combination of multiple helical rows of "sepals" together with numerous "stamens" whose anther consisted of a thickened connective and seemingly a single pair of microsporangia (Figs. 1, 4). The fossils were interpreted as bearing some similarities to members of the subfamily Doliocarpoideae of family Dilleniaceae (op. cit., p. 138). Being familiar with Proteaceae only through cultivated genera like *Grevillea*, with racemose flowers, we did not include this family in our analysis. On recently becoming acquainted with the capitular inflorescences in such genera of Proteaceae as *Protea* (Fig. 2), *Leucospermum,* and *Leucadendron* (Welsford et al. 2014), we now wish to transfer attention to this family as the most likely taxonomic group to which *Endobeuthos* belongs. Even so, the necessary new interpretation of its flowers requires postulating an extreme reduction in tepal length as well as in the number of stamens and perianth lobes, all in combination with unisexuality through the almost complete loss of gynoecia in the capitula available for study. A dioecious or andromonoecious breeding system must be postulated for the fossil species as a whole.

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## MATERIALS AND METHODS

Fossils of *Endobeuthos paleosum* occupy a block of amber from the well-known Noije Bum 2001 Summit Site mine in the Hukawng Valley, SW of Maingkhwan, Myanmar. The amber-containing deposits, which originated as near-shore marine sediments, were carefully described by Cruickshank and Ko (2003). Using data from palynology and fossil ammonites, these authors assigned an age of 97–110 Mya. A more recent study involving U-Pb dating of *in situ* zircons in volcanic clasts, determined the age to be 98.79 ± 0.62 Mya, placing the fossils at the mid-Cretaceous Albian/Cenomanian boundary (Shi et al. 2012). Other recent authors who have described fossils from this site (e.g. Crepet et al. 2016; Liu et al. 2018; Shi et al. 2022) agree in their use of this date. It now seems well established that the Myanmar amber originated in araucariaceous forests in Gondwana and were carried north during the Late Cretaceous Period as part of the India Block, until its collision with SE Asia in the early Cenozoic (Poinar et al. 2007; Poinar 2018; Poinar et al. 2020; Shi et al. 2022).

## RESULTS AND DISCUSSION

Figures 1 and 3 to 5 are taken from the original publication (Poinar & Chambers 2018), with modified labeling that befits our new interpretation. In figure 1A, the involucre of the capitulum is not obscured by dense trichomes, as in the left-hand capitulum, and its numerous helically-arranged bracts are evident. The arrow in figure 1B points to a staminate flower after all the pollen has been shed. Figure 2 shows, for comparison, the capitate inflorescence of *Protea mundii*, a modern species of Proteaceae. The helical arrangement of involucral bracts is evident, as in figure 1A. In figure 3, the holotype capitulum of *Endobeuthos* (Poinar & Chambers 2022) shows a distinct switch between the outer series of shorter involucral bracts (Fig. 3C) and a longer inner series (Fig. 3A), a combination not so evident on the 5 accompanying capitula. A similar difference in length between inner and outer involucral bracts occurs in various modern species of capitulate Proteaceae, as well (Fig. 2). The staminate flowers (Fig. 4) are much modified, the perianth having a slender, tubular lower portion and a single, much abbreviated tepal blade that bears one nearly sessile stamen (Fig. 4A–C). Careful inspection of the anther (Fig. 4B, C) reveals that it is tetrasporangiate, as in modern Proteaceae (Weston 2006; Heywood et al. 2007), not a bisporangiate theca as was proposed in the original paper. The flowers exhibit extreme reduction from the usually 4-lobed perianth of the family, in which each lobe bears a stamen (Ladd & Bowen 2020). However, it is clear that the stamen possesses a complete anther, not a half-anther as was originally proposed for this organ. Also figured are flowers with the anther newly dehisced (Fig. 4D) and with pollen fully dispersed (Fig. 4E). Pistils are almost entirely absent from the heads, although on one capitulum there are what appear to be 3 pubescent style-tips projecting from among the mass of flowers (Fig. 5), revealing the occasional presence of hermaphrodite or pistillate flowers.

In Proteaceae, the perianth is typically 4-lobed, the lobes being designated as tepals (Weston 2006). The perianth may be actinomorphic, with the lobes elongated and separate as in *Persoonia* (Citerne et al. 2017, fig. 1A), *Aulax* (Rourke 1987, fig. 1), and *Stirlingia* (Ladd & Bowen 2020, fig. 2D), or it may be zygomorphic and with lobes united, often cup-shaped, and terminal on an elongated perianth blade arising from one side of a tubular base (Citerne et al. 2017, fig. 1C, E). Each perianth lobe bears a single, usually subsessile stamen with a bilocular anther and 4 microsporangia. Usually all 4 anthers are fertile, but in the Western Australian genus *Conospermum*, only one complete and 2 half-anthers are fertile, the sterile loculi of the lateral 2 anthers being fused to the abaxial sterile anther (Ladd & Bowen 2020, fig. 3A–C). Another genus with partly sterile anthers is *Synaphea* (Ladd & Bowen 2020, fig. 3D, E), but the pattern is the reverse of *Conospermum,* with the adaxial anther sterile and fused to the style during bud development. In some species of *Adenanthos* and *Protea,* the adaxial anther is sterile and may remain adnate to the style apex (Ladd & Bowen 2020, p. 6). In the genus *Placospermum* of subfamily Persoonioideae, only the adaxial stamen is fertile, the other three being reduced to staminodia (Stevens 2001 and onwards).

Pollen release in *Endobeuthos* was apparently passive, there being no obvious adaptation for explosive release and no pollen-presenting style (Ladd & Bowen 2020). Pollen may have been dispersed by insects, as has been hypothesized for some other flowers in Myanmar amber (Poinar 2022), but only rarely is there clear

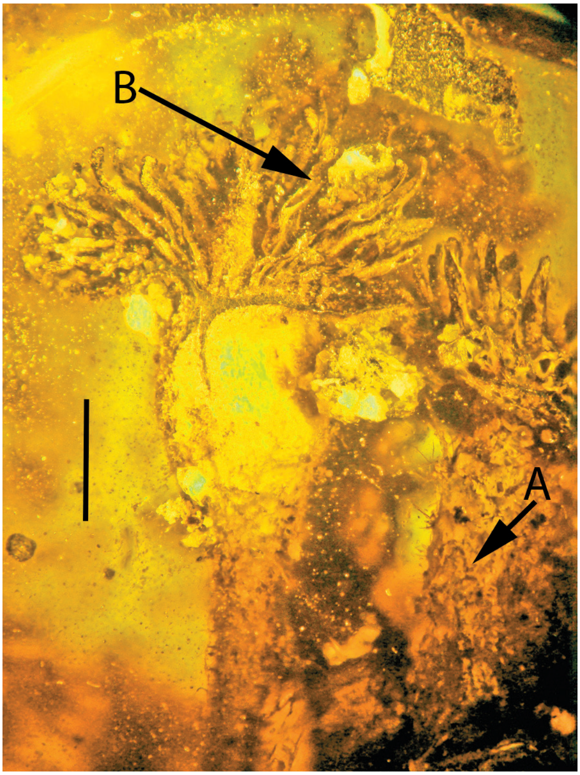

**Fig. 1.** ***Endobeuthos paleosum*****. Lateral view of 2 capitula. A. Involucre of helically arranged bracts. B. Individual staminate flower after release of pollen. Scale bar = 1.1 mm.**

evidence for the pollination syndromes of fossil plants (Stevens 2001 onwards). In the dioecious genus *Leucadendron,* endemic to South Africa, Welsford et al. (2015) present evidence for insect pollination in some species and for wind pollination in others, the latter considered to be the derived condition. Passive pollen release is characteristic of subfamily Persoonioideae, a sister group to the remainder of the family (Ladd & Bowen 2020, fig. 1, table 1; Barker et al. 2007, fig. 2), as well as 3 sampled genera in subfamily Proteoideae.

In the genus *Leucospermum*, there is a modification of perianth structure that is suggestive of how the simplified perianth of *Endobeuthos* may have evolved. In the treatment of Proteaceae by Heywood et al. (2007, fig. 1), the illustrated flower of *Leucospermum conocarpodendron* shows that one of its slender perianth segments is positioned separately and abaxial to the opposing adaxial segment, which carries the other 3 tepals and their attached stamens, fused together as a 3-toothed terminal blade. The displaced perianth segment bears a single fertile stamen on its expanded apex, and except for this structure being terminal on a stalk-like base, it resembles the single tepal lobe of a staminate *Endobeuthos* flower. In the genus *Protea,* as described and illustrated by Weston (2006, pg. 388, fig. 136), the perianth is similar to this in having the abaxial tepal, with its single stamen, set separate from and opposite to the 3 fused, adaxial tepals. The anther of this tepal in both *Leucospermum* and *Protea* faces adaxially, however, rather than abaxially as in *Endobeuthos*. Nonetheless, the divided perianths of these modern genera are models of how simplification of the *Endobeuthos* flower could have evolved, with 3 of the perianth segments having been lost to produce flowers with just one fertile tepal.

Reexamination of the pollen of *Endobeuthos* shows that the grains are spherical and tricolpate (Fig. 6). The illustrated grain, being imbedded in the amber, does not display the character of the colpi and echinate exine as well as the grain pictured in the original publication, which, however, is oblong and uncharacteristically

**Fig. 2.** ***Protea mundii*. Capitulum showing helically arranged bracts increasing in size towards the apex. Floral tepals have long apical bristles. Photograph courtesy of G.D. Carr.**

distorted (Poinar & Chambers 2018, fig. 6). Pollen like that of *Endobeuthos* is unusual among modern genera of Proteaceae, the closest approach being some examples from *Beauprea*, a *genus insertae sedis* in subtribe Proteoideae found in New Caledonia (Weston 2006, p. 386). Studies of this genus (Pocknall & Crosbie 1988) illustrate some species with triangular grains and others with tricolpate grains having a convexly rounded but apparently not completely spherical shape. Cretaceous fossil pollen attributed to *Beauprea,* known by the name *Beaupreadites*, is recorded from the Maastrichtian Stage (Pocknall & Crosbie 1988). According to Sauquet and Cantrill (2007), the pollen of *Beauprea* is "unique in Proteaceae in having clearly defined, strongly angular (in polar view) colpoid apertures." These authors point to the pollen of *Franklandia*, another *genus incertae sedis* in the Proteoideae, as having a spheroidal shape "found nowhere else in the family." This pollen is triporate, however, and is over 80µ in diameter, the largest in the family (Sauquet & Cantrill 2007).

Fossil pollen from the Cretaceous that resembles *Endobeuthos* in being round in cross-section and tricolpate is often assigned to the catch-all genus *Tricolpites*. Palynological studies in Australia (Dettmann & Playford 1969) show a stage in sediments of the Otway Basin, Victoria, termed the *Tricolpites pannosus* Zone, which spans the contact between the Albian and Cenomanian Stages (op. cit., tab. 9.2, p. 179), the same time period as our *Endobeuthos* fossils. A pollen grain of *Tricolpites pannosus* is illustrated in microscopic cross-section

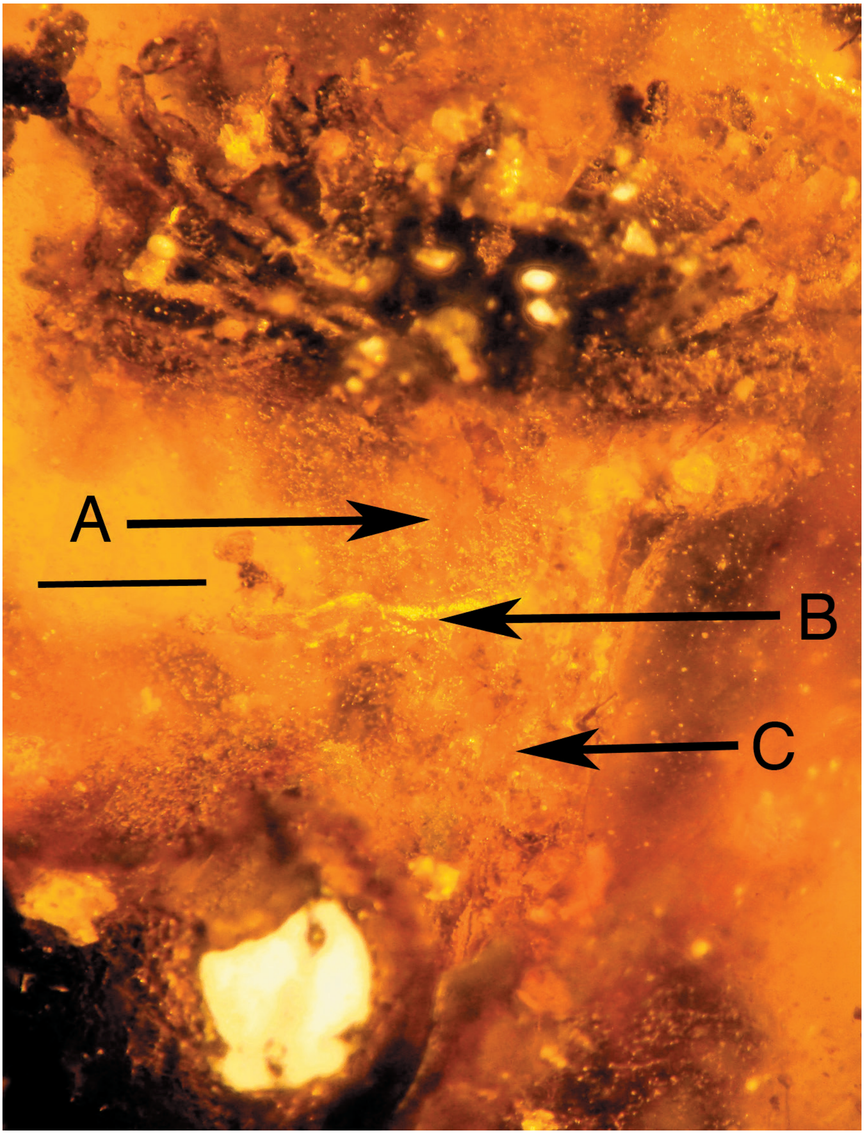

**Fig. 3.** ***Endobeuthos paleosum*****.** Capitulum of holotype, lateral view. **A.** Series of elongate inner involucral bracts. **B.** Juncture between outer and inner bracts. **C.** Series of short outer involucral bracts. Scale bar = 1.0 mm.

(op. cit., plate 13, figs. 15–16), fitting the description of that fossil genus. Higher up in the Cretaceous is the *Tricolpites pachyexinus* Zone (op. cit., tab. 9.2), representing the Upper Coniacian/Lower Santonian Stages. The pollen of this taxon (op. cit., plate 13, fig. 14) resembles *T. pannosus* and, like it, is round in cross-section and tricolpate. In this paper, the authors make no assignment of these *Tricolpites* fossils to a modern family. Neither are they mentioned in a later paper (Dettmann 2017, fig. 86), in which several types of fossil pollen are attributed to Proteaceae, but none are older than the Santonian. We mention them because of their similarity, in shape and aperture form, to *Endobeuthos*, with its putative assignment to Proteaceae, whereas such grains are rare in modern genera of the family.

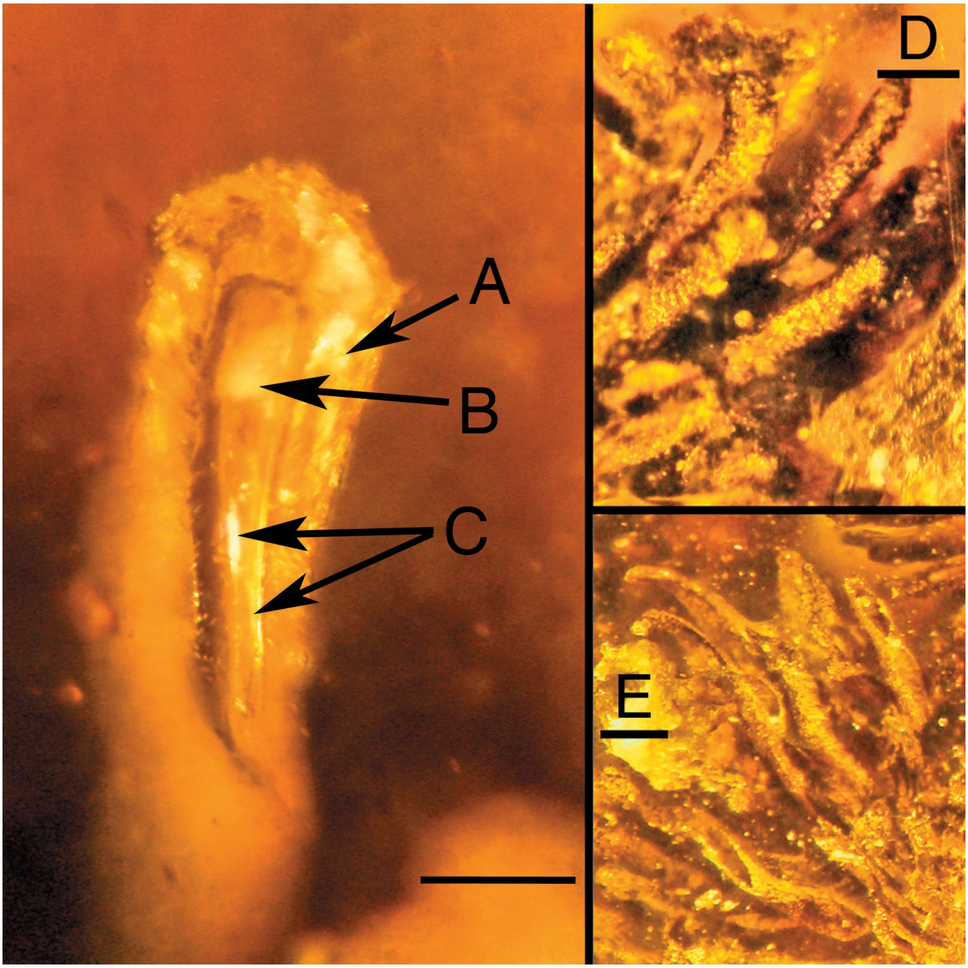

Fig. 4. ***Endobeuthos paleosum*****. Flowers. A. Blade of tepal. B. Anther of stamen. Filament is short or lacking. C. Two locules, each with a pair of linear microsporangia. Scale bar = 0.05 mm. D. Flowers with dehisced stamens and abundant pollen. Scale bar = 0.2 mm. E. Flowers after dispersal of pollen. Note tubular portion of perianth and slender, terete pedicel. Scale bar = 0.2 mm.**

Barker et al. (2007) used chloroplast DNA sequence data to estimate the crown group age of Proteaceae as 118.5 ± 8.2 Mya. An earlier report by Anderson et al. (2005), using both penalized likelihood and nonparametric rate smoothing to analyze *rbcL* sequences, date the Proteaceae stem group as 110 Mya (PL) and 119 Mya (NPRS), and the crown group as 85 Mya (PL) and 96 Mya (NPRS). Their oldest referenced Proteaceae pollens were *Beaupreapopsis*-type (Proteoideae) and *Macadamia/ Helicia*-type (Grevilleoideae), both appearing in the Santonian of Australia. If *Endobeuthos* is correctly placed in Proteaceae, it is the first known Cretaceous fossil flower of the family and extends the record of *Tricolpites*-type pollen in the family to approximately 99 Mya.

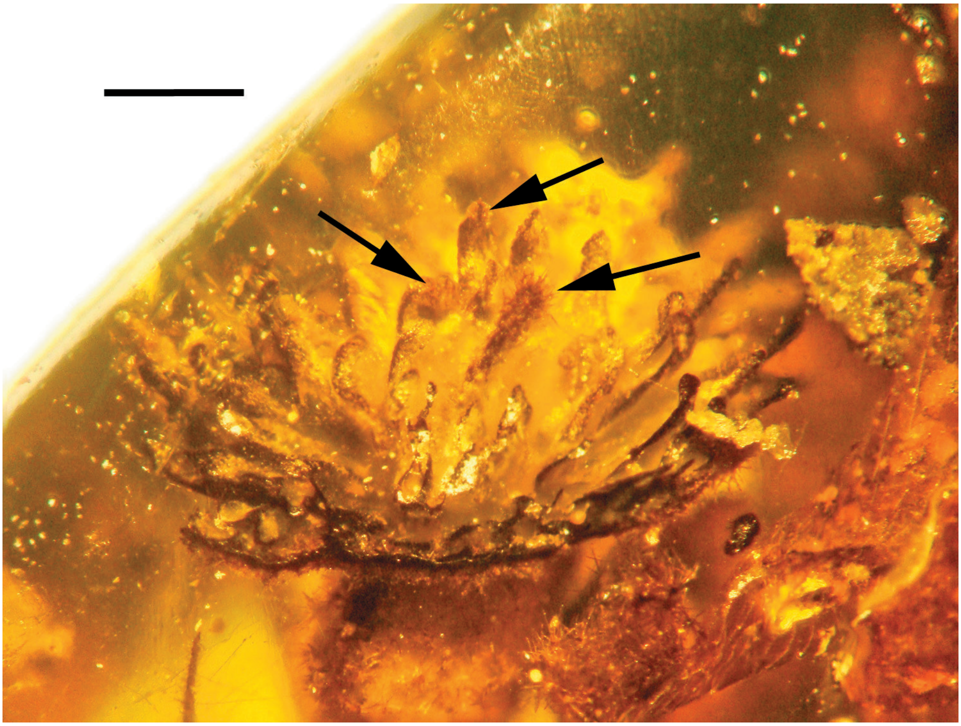

Fig. 5. *Endobeuthos paleosum*. Capitulum, apical view. Arrows show 3 styles partially exserted from among the staminate flowers. Note the additional series of inner involucral bracts around the flower cluster. Scale bar = 0.9 mm.

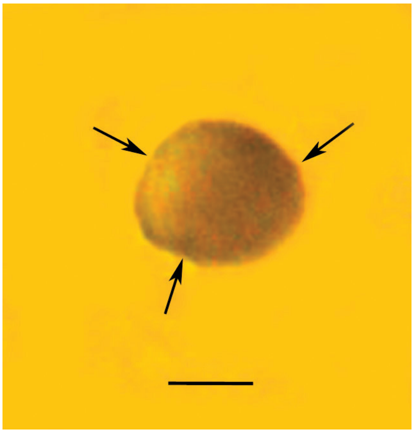

Fig. 6. *Endobeuthos paleosum*. Tricolpate pollen grain. Arrows show colpi. Scale bar = 16 µ.

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## REFERENCES

Anderson, D.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.

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.

Citerne, H.L. et al. 2017. Characterization of *CYCLOIDEA*-like genes in Proteaceae, a basal eudicot family with multiple shifts in floral symmetry. Ann. Bot. 119:367–378.

Crepet, W.L., K.C. Nixon, D. Grimaldi, & M. Riccio. 2016. A mosaic Lauralean flower from the Early Cretaceous of Myanmar. Amer. J. Bot. 103:290–297.

Cruickshank, R.D. & K. Ko. 2003. Geology of an amber locality in the Hukawng Valley, northern Myanmar. J. Asian Earth Sci. 21:441–455.

Dettmann, M.E. 2017. Cretaceous vegetation: The microfossil record. In: Hill, R.S., ed. History of the Australian vegetation. Cretaceous to Recent. University of Adelaide Press, Adelaide, Australia. Pp. 143–170.

Dettmann, M.E. & G. Playford. 1969. Palynology of Australian Cretaceous: A review. In: Campbell, K.S.W., ed. Stratigraphy and palaeontology. Essays in honor of Dorothy Hill. Australian National University Press, Canberra, Australia. Pp. 174–210.

Heywood, V.H., R.K. Brummitt, A. Culham, & O. Seberg. 2007. Flowering plants of the world. Firefly Books, Ltd., London, UK. Pp. 268–269.

Ladd, P.G. & B.J. Bowen. 2020. Pollen release in the Proteaceae. Pl. Syst. Evol. https://doi.org/10.1007/s00606-020-01707-2

Liu, Z-J., D. Huang, C. Cai, & A. Wang. 2018. The core eudicot boom registered in Myanmar amber. Sci. Rep. 8:16765.

Pocknall, D. & Y.M. Crosbie. 1988. Pollen morphology of *Beauprea* (Proteaceae): Modern and fossil. Rev. Palaeobot. Palynol. 53:305–327.

Poinar, G.O., Jr. 2018. Burmese amber: Evidence of Gondwanan origin and Cretaceous dispersion. Hist. Biol. 2018:1–6.

Poinar, G.O., Jr. 2022. Flowers in amber. Springer Nature, Switzerland AG.

Poinar, G.O., Jr. & K.L. Chambers. 2018. *Endobeuthos paleosum* gen. et sp. nov., fossil flowers of uncertain affinity from mid-Cretaceous Myanmar amber. J. Bot. Res. Inst. Texas 12:133–139.

Poinar, G.O., Jr. & K.L. Chambers. 2022. Validation of the previously published name *Endobeuthos paleosum*, a fossil flower in mid-Cretaceous Myanmar amber. J. Bot. Res. Inst. Texas 16:73–74.

Poinar, G.O., Jr., G.J.B. Lambert, & Y. Wu. 2007. Araucarian source of fossiliferous Burmese amber: Spectroscopic and anatomical evidence. J. Bot. Res. Inst. Texas 1:449–455.

Poinar, G.O., Jr., K.L. Chambers, U.T. Iwaniec, & F.E. Vega. 2020. *Valviloculus pleristaminis* gen. et sp. nov.*,* a Lauralean fossil flower with valvate anthers from mid-Cretaceous Myanmar amber. J. Bot. Res. Inst. Texas 14:359–366.

Rourke, J.P. 1987. The inflorescence morphology and systematics of *Aulax* (Proteaceae). S. Afr. Tydskr. Plantk. 53:464–480.

Sauquet, H. & D.J. Cantrill. 2007. Pollen diversity and evolution in Proteoideae (Proteales: Proteaceae). Syst. Bot. 32:271–316.

Shi, C., et al. 2022. Fire-prone Rhamnaceae with South African affinities in Cretaceous Myanmar amber. Nature Pl. https://doi.org/10.1038/s41477-021-01091-w

Shi, G., D.A. Grimaldi, G.E. Harlow, Ji Wang, Ju Wang, M. Yang, W. Lei, O. Li, & X. Li. 2012. Age constraint on Burmese amber based on U-Pb dating of zircons. Cretaceous Res. 37:155–163.

Stevens, P.F. 2001 onwards. Angiosperm Phylogeny Website, Version 14, 2017 [and more or less continuously updated since]. https://www.mobot.org/MOBOT/research/APweb [Accessed on 20 April 2023].

Welsford, M.R., J.J. Midgley, & S.D. Johnson. 2014. Experimental evaluation of insect pollination versus wind pollination in *Leucadendron* (Proteaceae). Int. J. Pl. Sci. 175:296–306.

Welsford, M.R., N. Hobbhahn, J.J. Midgley, & S.D. Johnson. 2015. Floral trait evolution associated with shifts between insect and wind pollination in the dioecious genus *Leucadendron* (Proteaceae). Evol. 70(1):126–139.

Weston, P.H. 2006. Proteaceae. In: Kubitzki, K., ed. The families and genera of vascular plants. IX. Flowering plants—Eudicots. Springer-Verlag, Berlin, Germany. Pp. 364–404.