

A eudicot from the Early Cretaceous of China

Ge Sun^{1,2}, David L. Dilcher^{1,2,3}, Hongshan Wang^{1,4} & Zhidian Chen⁵

The current molecular systematics of angiosperms¹ recognizes the basal angiosperms and five major angiosperm lineages: the Chloranthaceae, the magnoliids, the monocots, *Ceratophyllum* and the eudicots, which consist of the basal eudicots and the core eudicots². The eudicots form the majority of the angiosperms in the world today. The flowering plants are of exceptional evolutionary interest because of their diversity of over 250,000 species and their abundance as the dominant vegetation in most terrestrial ecosystems, but little is known of their very early history. In this report we document an early presence of eudicots during the Early Cretaceous Period. Diagnostic characters of the eudicot fossil *Leefructus* gen. nov. include simple and deeply trilobate leaves clustered at the nodes in threes or fours, basal palinactinodromous primary venation, pinnate secondary venation, and a long axillary reproductive axis terminating in a flattened receptacle bearing five long, narrow pseudo-syncarpous carpels. These morphological characters suggest that its affinities are with the Ranunculaceae, a basal eudicot family. The fossil co-occurs with *Archaeo-*fructus sinensis*³ and *Hyr-*cantha decussata*⁴ whereas *Archaeo-*fructus liaoningensis*⁵ comes from more ancient sediments. Multiple radiometric dates of the Lower Cretaceous Yixian Formation place the bed yielding this fossil at 122.6–125.8 million years old^{6–8}. The earliest fossil records of eudicots are 127 to 125 million years old, on the basis of pollen^{9,10}. Thus, *Leefructus* gen. nov. suggests that the basal eudicots were already present and diverse by the latest Barremian and earliest Aptian.***

The evolutionary history of the angiosperms is still being revealed since their Early Cretaceous or possible earlier origin and their rapid radiations during the mid-Cretaceous^{2,11,12,13}. Here we present a new fossil plant from the Jehol Biota. The Jehol Biota is preserved mainly in

the Yixian Formation of northeast China (Figs 1 and 2), which has yielded rich assemblages of animals and plants. Early birds, feathered dinosaurs, placental mammals and a variety of invertebrate fossils have been found^{14–16} as well as several angiosperms, including *Archaeo-*fructus*^{3–5} and *Hyr-*cantha*^{3–5}. The presence of *Archaeo-*fructus*^{3–5} at about 125 Myr ago¹⁷ clearly documents the presence of basal angiosperms in the megafossil record of the Early Cretaceous and now we also document the presence of basal eudicots from the same stratigraphic horizon. These early angiosperms are followed by diverse and abundant fossils***



Figure 1 | Fossil locations. The map shows the Dawangzhangzi locality in Lingyuan where the fossil *Leefructus mirus* gen. et sp. nov. was collected (1) and the Huangbanjigou locality in Beipiao where *Archaeo-*fructus liaoningensis* was collected (2). The inset map shows the location of Liaoning Province in northeast China (star indicates Beijing).*

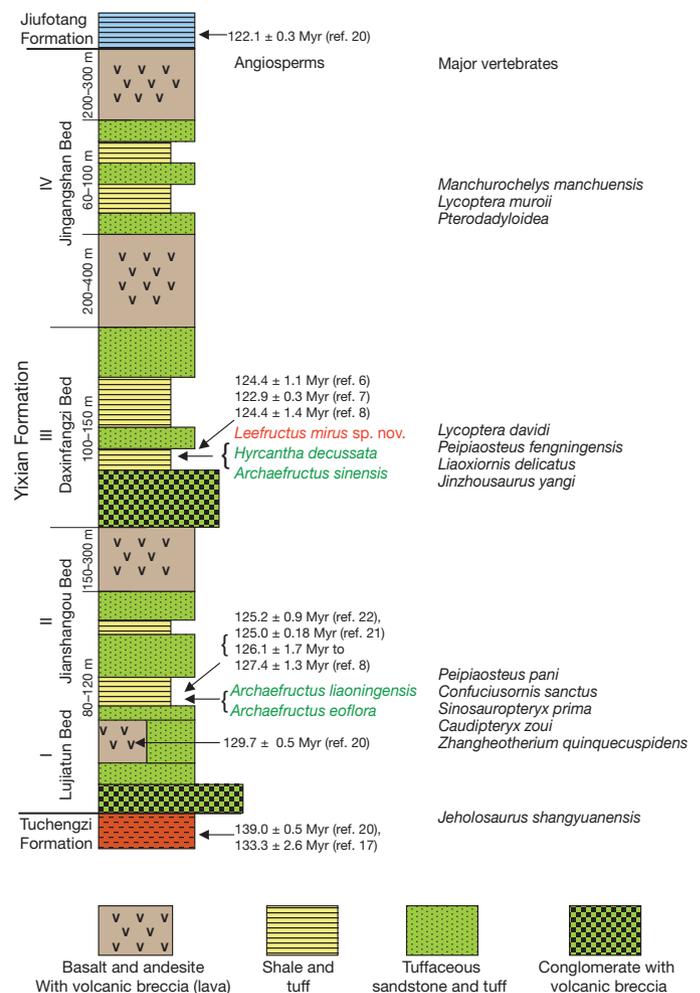


Figure 2 | The ages of the Yixian Formation, showing the fossil angiosperms and major vertebrates found there. The Yixian Formation is subdivided into four beds. I, the Lujiatun Bed (about 129.7 Myr old²⁰, recently dated as about 133.3 Myr old¹⁷). II, the Jianshangou Bed (about 125.2 Myr old²² and 125.0 Myr old²¹, recently dated as about 127.4–126.1 Myr old⁸). III, the Daxinfangzi Bed (about 124.4 Myr old⁶ and 122.9 Myr old⁷, recently dated as about 124.4 Myr old⁸). IV, the Jingangshan Bed (about 123–122.1 Myr old²⁰).

¹Paleontological Institute of Shenyang Normal University, Shenyang 110034, China. ²Research Center of Paleontology, Jilin University, Changchun 130026, China. ³Department of Biology, Indiana University, Bloomington, Indiana 47405, USA. ⁴Florida Museum of Natural History, University of Florida, Gainesville, Florida 32611-7800, USA. ⁵State Key Laboratory of Systematic and Evolutionary Botany, Institute of Botany, Chinese Academy of Sciences, Beijing 100093, China.

of all major angiosperm taxa in sediments of the late Aptian and the Albian ages (112 to 100 Myr ago). Much of the record of early angiosperm diversity and their subsequent middle to late Albian radiations come to us from fossils that encompass a complete range of preserved plant organs such as pollen, mesofossils and megafossils^{11,12}. We follow the Angiosperm Phylogeny Group¹⁸ to classify the fossil.

Angiosperms
Eudicots

Ranunculales (compare Family stem lineage to the Ranunculaceae)

Leefructus Sun, Dilcher, Wang et Chen, gen. nov.

Leefructus mirus Sun, Dilcher, Wang et Chen, sp. nov.

Generic diagnosis: The plant consists of a shoot bearing leaves at distinct nodes. The leaf is simple, petiolate, and deeply trilobate with each lobe further dissected. Leaves clustered at the nodes in threes or fours are attached in a tight spiral at each node. Primary venation is basal palinactinodromous. Secondary venation is pinnate, with secondary vein branches bracing the leaf sinuses and then producing conspicuous looping veins near the leaf margin. The long reproductive axis is axillary, terminating in a flattened receptacle bearing five long, narrow, pseudo-syncarpous carpels.

Etymology: *Lee* is given in honour of Shiming Li of Shenyang, China; *fructus* for fruiting. The species epithet *mirus* is from the Latin word *mira*, meaning beautiful.

Specific diagnosis: The specific diagnosis is the same as for the generic diagnosis because of limited material at this time.

Holotype: PISNU-0701 (Fig. 3), deposited in the Paleontological Institute of Shenyang Normal University, Shenyang, China.

Age and horizon: Early Cretaceous, Daxin角度 Bed, Yixian Formation.

Description and interpretation: The whole shoot is 16 cm long, consisting of a stem with two nodes, each bearing leaves and one fertile branch. The major stem appears herbaceous (see Fig. 4 for a reconstruction, and Figs 3 and 5c), robust, multistranded, with a prominent node, 55 mm long by 2–2.5 mm wide, bearing leaves, fruit and a vegetative shoot. The leaf scar and leaf attachment at the node appear to be helical. Three or four leaves are clustered at each node. Proximal leaves



Figure 3 | Holotype specimen of *Leefructus mirus* Sun, Dilcher, Wang et Chen. The fossil shows multistranded stems, several leaves clustered at two distinct nodes, three-lobed leaves, and a single reproductive auxiliary shoot. Scale bar, 1 cm. Photograph by C. T. Li.



Figure 4 | Reconstruction of *Leefructus mirus* Sun, Dilcher, Wang et Chen. Diagram by S. Trammel and D.L.D.

are larger with lateral lobes about 20–35 mm long by 11–12 mm wide and medial lobes are about 35–40 mm long by 15–22 mm wide. Leaves are simple, estipulate, and deeply trilobate with each lobe further pinnately lobed with non-glandular lobules gradually decreasing in

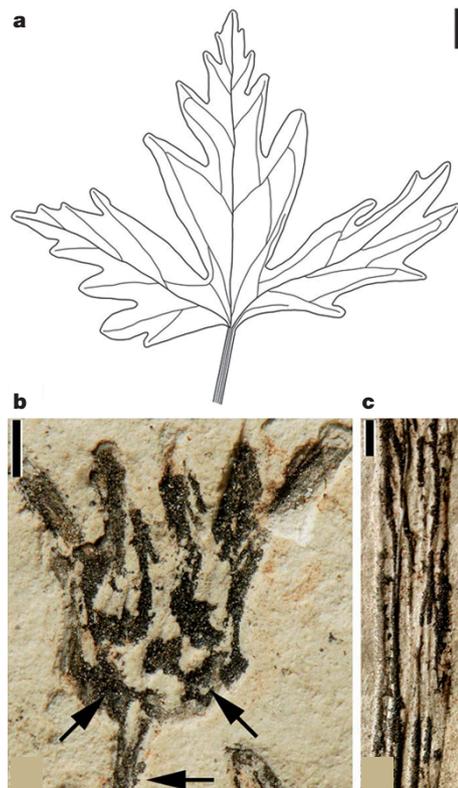


Figure 5 | Details of *Leefructus mirus* Sun, Dilcher, Wang et Chen. a, Line diagram of the leftmost leaf seen in Fig. 3 to show venation. Scale bar, 5 mm. (Diagram by D.L.D.) b, Enlargement of the fruit in Fig. 3 to show five pseudo-syncarpous elongate carpels loosely fused and slightly enclosed by a flat receptacle that has faint oval basal scars (indicated by arrows), and a faint scar 3 mm below the receptacle (indicated by the horizontal arrow). Scale bar, 1 mm. c, Enlargement of the main axis in Fig. 3 to show the multistranded stem. Scale bar, 1 mm.

size distally, resulting in a toothed appearance on the distal leaf margin (Fig. 5a). The petioles of proximal leaves are elongate, about 37–40 mm long by 1.3–1.5 mm wide, multistranded and broadly attached at the node. There may be one abscission scar present at the node. The petioles of distal leaves are short, 10–15 mm long. Primary venation is basal palinactinodromous with three major and two minor primary veins diverging at the extreme base of the leaf lamina from the petiole. One major primary vein forms the medial vein in each major lobe; each of the minor secondary veins form sub-marginal veins in the basal portion of the medial lobes and extend into the basal marginal lobes, then loop to join superadjacent secondary veins and successive marginal lobes (Fig. 5a). Secondary venation is pinnate with an irregular branching pattern. Secondary veins branch and produce conspicuous looping secondary or tertiary veins near the leaf margin; these branches form intramarginal veins extending to and fusing with the next distal secondary veins, or extending into the apex of the lobules but diminishing before reaching the leaf margin. An axillary solitary inflorescence terminates an elongate pedicel 50 mm long by 0.5 mm wide with a faint scar 1 mm below the receptacle. The fruit (Fig. 5b) is 6 mm long by 4 mm wide, pseudo-syncarpous with the basal two-thirds of the five elongate carpels loosely fused, the carpels sitting on and slightly enclosed by a flat receptacle. Elongate tips are present on each carpel, suggesting long stigmatic tips. The receptacle has several faint oval scars (Fig. 5b) that may represent scars of deciduous stamens.

The geological age of the Yixian Formation has received considerable attention over the past two decades¹⁹. The main focus has been on the two horizons (beds) yielding early angiosperms: (1) the Daxinfangzi Bed in Lingyuan and (2) the Jianshangou Bed in Beipiao (Fig. 2).

The Daxinfangzi Bed of the Yixian Formation in Lingyuan is an important horizon for early angiosperms, including *Archaeofructus sinensis*, *Hyracantha decussata* and *Leeffructus mirus* gen. et sp. nov. This bed has been dated as 122.9 ± 0.3 Myr old⁷, 124.4 ± 1.4 Myr old⁸ and 124.4 ± 1.1 Myr old⁶. We consider the Daxinfangzi Bed to range in age from 122.6 to 125.8 Myr. The lowest volcanic beds of the Lujiatun Bed have been dated at 133.3 ± 2.6 Myr (ref. 17) and 129.7 ± 0.5 Myr (ref. 20). The Jiufotang Formation, which conformably overlies the Yixian Formation, was dated as 122.1 ± 0.3 Myr old^{17,20} (Fig. 2). These dates indicate that the entire Yixian Formation ranges in age from 121.8 to 135.9 Myr, lasting a total of about 7 to 14 Myr (refs 17 and 20).

The Jianshangou Bed in Beipiao is also an important horizon for the age of early angiosperms including *Archaeofructus liaoningensis* and *A. eoflora*. Since 2001 this bed is dated as 125.0 ± 0.18 Myr old²¹ or 125.2 ± 0.9 Myr old²², which is close to the Barremian–Aptian boundary. Recently, further dating using both ⁴⁰Ar/³⁹Ar and U–Pb methods yielded ages of 127.4 ± 1.3 to 126.1 ± 1.7 Myr (ref. 8). According to these radiometric dates, the age of the Jianshangou Bed can be considered to range from 124.3 to 128.7 Myr.

Wang *et al.*²³ have examined basal eudicot phylogeny and evolution with a large generic and species sampling including all seven families of Ranunculales (105 taxa including 42 new sequences, 129 genera of Ranunculales and 99 genera of Ranunculaceae) using both a 65-character morphological data set and molecular data from four genomic regions—the plastids *rbcL*, *matK*, *trnL-F* and nuclear ribosomal 26S ribosomal DNA. They present their results using both maximum parsimony and Bayesian inference, which give strong support for the monophyletic nature of three main clades, the family Eupteleaceae (two species), the family Papaveraceae (750 species) and the core Ranunculales, the latter consisting of the five families Circaeasteraceae (two species), Lardizabalaceae (50 species), Menispermaceae (450), Berberidaceae (650 species) and Ranunculaceae (2,000 species). The number of species in these families is very uneven, but the combination of molecular and morphological character states resulted in well-resolved cladograms²³.

The morphological characters of *Leeffructus mirus* gen. et sp. nov. are well preserved. These characters include multistranded and possible herbaceous upright stems, several leaves clustered at two distinct nodes,

three-lobed leaves with the lobe margins further lobed and a unique venation pattern. The fossil leaves resemble the venation patterns and forms of *Delphinium* leaves, typical of the Ranunculaceae. The fossil shoot has a simple reproductive auxiliary long pedicel terminating in a collection of five basally fused follicles sitting on a flattened receptacle (Figs 3–5), which appears to have several small scars around its base. Several genera of the Ranunculaceae have two to five multilobed leaves and carpels borne on flattened receptacles at the ends of long axillary pedicels. The small circular scars on the receptacle may represent attachment scars of stamens (Fig. 5b). Because many of the fossil characters occur in extant genera of the Ranunculaceae, we suggest the fossil *Leeffructus mirus* gen. et sp. nov. to be an extinct taxon along the stem lineage of this extant family.

The term eudicot was proposed in 1991 as a “putatively monophyletic group” using tricolpate pollen to define the clade⁹. This clade was recognized at the Barremian–Aptian boundary from sediments in Gabon²⁴ and from the early Albian of the Potomac Group in North America²⁵. Hughes¹⁰ recognized tricolpate pollen from Bed 35 at the base of the Vectis Formation that Hughes considered to be “Phase 4”, which is at the Barremian–Aptian boundary. This current understanding of the fossil record places the earliest fossils of the eudicots at this Barremian–Aptian boundary age of about 125 Myr. The distribution of this pollen type from cores taken off the coast of Gabon and from England suggests that eudicot plants were already distributed widely by about 125 Myr ago and probably had an earlier origin.

The early fossil record of basal eudicot evolution is incomplete, so each fossil that can be placed in this clade provides new and important information about early angiosperms. Previous fossil angiosperms reported from the Yixian Formation include *Archaeofructus* and *Hyracantha*^{3–5}. It is possible that *Hyracantha* could also be considered within the stem lineage of the Ranunculaceae^{4,26}, and *Archaeofructus* is considered to be basal to all angiosperms^{3,26}.

Molecular studies during the past decade have helped to push the age of the first angiosperms earlier than the Early Cretaceous^{27–30}. The basalmost ANITA (*Amborella*, *Nymphaea*, *Illicium*, *Trimenia* and *Austrobaileya*) grade must have existed before mesangiosperms (magnoliids, Chloranthaceae, *Ceratophyllum*, monocots and eudicots). The presence of the megafossils, *Leeffructus mirus* gen. et sp. nov. and *Hyracantha decussata*^{4,26}, which also has possible Ranunculales/Ranunculaceae affinities, documents eudicot evolution early in angiosperm history. Both the fossil record and molecular data suggest the early presence of basal eudicots, which encourages us to consider seriously the probable pre-Cretaceous evolution of the angiosperms.

METHODS SUMMARY

Leeffructus mirus gen. et sp. nov. was collected from the Daxinfangzi Bed (previously known as Dawangzhangzi Bed) of the middle part of the Yixian Formation exposed in Dawangzhangzi Village (41° 09′ 961″ N, 119° 16′ 298″ E) about 15 km south of Lingyuan City in western Liaoning Province (Figs 1 and 2). Lithologically the Daxinfangzi bed is mainly composed of yellowish grey and grey sandstone intercalated with grey silt stone and tuffaceous silt and fine-grained sandstone. The Daxinfangzi bed is dated as 122.6–125.8 Myr old^{6–8,14,17}. *Leeffructus mirus* gen. et sp. nov. is preserved as an impression in the intercalated yellowish grey siltstone. Only one part of the impression was collected and studied. On the same slab, *Leeffructus mirus* gen. et sp. nov. co-occurs with the fish, *Lycoptera davidi* Sauvage. Several small samples were removed from the same slab and macerated in hopes of finding pollen, spores or cuticular material but we had no success. Small dark pigmented flakes were removed and examined with the scanning electron microscope for organic remains. These were found to consist of iron-stained matrix and no original organic material is preserved.

Received 14 April 2010; accepted 6 January 2011.

1. Qiu, Y.-L. *et al.* The earliest angiosperms: evidence from mitochondrial, plastid and nuclear genomes. *Nature* **402**, 404–407 (1999).
2. Moore, M. J. *et al.* Using plastid genome-scale data to resolve enigmatic relationships among basal angiosperms. *Proc. Natl Acad. Sci. USA* **104**, 19363–19368 (2007).

3. Sun, G. *et al.* Archaeofractaceae, a new basal angiosperm family. *Science* **296**, 899–904 (2002).
4. Dilcher, D. L. *et al.* An early infructescence *Hyracantha decussata* (comb. nov.) from the Yixian Formation in northeastern China. *Proc. Natl Acad. Sci. USA* **104**, 9370–9374 (2007).
5. Sun, G. *et al.* In search of the first flower: a Jurassic angiosperm, *Archaeofructus*, from Northeast China. *Science* **282**, 1692–1695 (1998).
6. Zhang, H. *et al.* U-Pb isotopic age of the Lower Yixian Formation in Lingyuan of western Liaoning and its significance. *Geol. Rev.* **52**, 63–71 (2006).
7. Swisher, C. C. *et al.* Cretaceous age for the feathered dinosaurs of Liaoning, China. *Nature* **400**, 58–61 (1999).
8. Meng, F. X., Gao, S. & Liu, X. M. U-Pb zircon geochronology and geochemistry of volcanic rocks of the Yixian Formation in the Lingyuan area, western Liaoning, China. *Geol. Bull. China* **27**, 364–373 (2008).
9. Doyle, J. A. & Hotton, C. L. in *Pollen and Spores: Patterns of Diversification* (eds Blackmore, S. & Barnes, S. H.) 169–195 (Clarendon Press, 1991).
10. Hughes, N. F. *The Enigma of Angiosperm Origins* (Cambridge University Press, 1994).
11. Crepet, W. L., Nixon, K. C. & Gandolfo, M. A. Fossil evidence and phylogeny: the age of major angiosperm clades based on mesofossil and microfossil evidence from Cretaceous deposits. *Am. J. Bot.* **91**, 1666–1682 (2004).
12. Anderson, C. L., Bremer, K. & Friis, E. M. Dating phylogenetically basal eudicots using rbcL sequences and multiple fossil reference points. *Am. J. Bot.* **92**, 1737–1748 (2005).
13. Taylor, T. N., Taylor, E. L. & Krings, M. *Paleobotany: The Biology and Evolution of Fossil Plants* (Elsevier Science and Technology, 2008).
14. Chang, M. M. (ed). *Jehol Biota* [in Chinese] (Shanghai Sci. Tech. Press, 2001).
15. Ren, D. *et al.* Flower-associated brachycera flies as fossil evidence for Jurassic angiosperm origins. *Science* **280**, 85–88 (1998).
16. Zhou, Z., Barrett, P. M. & Hilton, J. An exceptionally preserved Lower Cretaceous ecosystem. *Nature* **421**, 807–814 (2003).
17. Peng, Y. D. *et al.* $^{40}\text{Ar}/^{39}\text{Ar}$ and K-Ar dating of the Yixian Formation volcanic rocks, western Liaoning Province, China. *Geochimica* **32**, 427–435 (2003).
18. Angiosperm Phylogeny Group. 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 (2009).
19. Barrett, P. M. Evolutionary consequences of dating the Yixian Formation. *Trends Ecol. Evol.* **15**, 99–103 (2000).
20. Chang, S.-C. *et al.* High-precision $^{40}\text{Ar}/^{39}\text{Ar}$ age for the Jehol Biota. *Palaeogeogr. Palaeoclimatol. Palaeoecol.* **280**, 94–104 (2009).
21. Swisher, C. C. *et al.* Further support for a Cretaceous age for the feathered-dinosaur beds of Liaoning, China: New dating of the Yixian and Tuchengzi Formations. *Chin. Sci. Bull.* **47**, 136–139 (2002).
22. Wang, S. S. *et al.* Further discussion on the geological age of Sihetun vertebrate assemblage in western Liaoning, China: evidence from Ar-Ar dating. *Acta Petrol. Sin.* **7**, 663–668 (2001).
23. Wang, W. *et al.* Phylogeny and classification of Ranunculales: evidence from four molecular loci and morphological data. *Perspect. Plant Ecol. Evol. Syst.* **11**, 81–110 (2009).
24. Doyle, J. A. *et al.* Angiosperm pollen from the pre-Albian Lower Cretaceous of equatorial Africa. *Bull. Centres Recherches Exploration-Production Elf-Aquitaine* **1**, 451–473 (1977).
25. Doyle, J. A. Cretaceous angiosperm pollen of the Atlantic Coastal Plain and its evolutionary significance. *J. Arnold Arbor.* **50**, 1–35 (1969).
26. Leng, Q. & Friis, E. M. *Sinocarpus decussates* gen. et sp. nov., a new angiosperm with basally syncarpous fruits from the Yixian Formation of Northeast China. *Plant Syst. Evol.* **241**, 77–88 (2003).
27. Wikström, N., Savolainen, V. & Chase, M. W. Evolution of the angiosperms: calibrating the family tree. *Proc. R. Soc. Lond.* **268**, 2211–2220 (2001).
28. Bell, C. D., Soltis, D. E. & Soltis, P. S. The age of the angiosperms: a molecular timescale without a clock. *Evolution* **59**, 1245–1258 (2005).
29. Bell, C. D., Soltis, D. E. & Soltis, P. S. The age and diversification of the angiosperms re-revisited. *Am. J. Bot.* **97**, 1296–1303 (2010).
30. Smith, S. A., Beaulieu, J. M. & Donoghue, M. J. An uncorrelated relaxed-clock analysis suggests an earlier origin for flowering plants. *Proc. Natl. Acad. Sci. USA* **107**, 5897–5902 (2010).

Acknowledgements We acknowledge the support of the Key Lab of Evolution of Past Life and Environment in Northeast Asia, Ministry of Education, China, and Project “111” of China, NSFC project number 40842002, and the President Special Fund of Shenyang Normal University to carry out this research project during 2008–2009. Many thanks to S. M. Li and L. X. Wang for their help in collecting the fossil specimen. We thank Y. Duan, C. T. Li, Y. S. Liu, D. M. Jarzen, T. Lott, S. Trammel and W. Wang for their assistance in analysis, computer work, photography and artwork. We also thank P. and D. Soltis, K. Nixon, M. Moore and J. Doyle for suggestions.

Author Contributions G.S. and D.L.D. designed the research plan. G.S., D.L.D., H.W. and Z.C. performed analysis. G.S., D.L.D. and H.W. wrote the manuscript. All authors discussed and commented on the manuscript.

Author Information Reprints and permissions information is available at www.nature.com/reprints. The authors declare no competing financial interests. Readers are welcome to comment on the online version of this article at www.nature.com/nature. Correspondence and requests for materials should be addressed to G.S. (sunge@synu.edu.cn) and D.L.D. (dilcher@indiana.edu).