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New insights into the Phylogeny of *Angelica* and its Allies (Apiaceae) with Emphasis on East Asian Species, Inferred from nrDNA, cpDNA, and Morphological Evidence

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Abstract—Classification of *Angelica* and its allies is complex and controversial, with previous phylogenetic studies restricted to examination of small numbers of taxa using only nrDNA ITS sequences. In this study, phylogenetic analyses of nrDNA ITS and ETS sequences, cpDNA sequences (*rps16* intron, *rps16-trnK*, *rpl32-trnL*, and *trnL-trnT*), and morphological data, supplemented by observations of fruit anatomy and micromorphology, were used to ascertain evolutionary relationships and confirm generic boundaries within *Angelica* s.l. (including *Angelica*, *Archangelica*, *Coelopleurum*, *Czernaevia*, and *Ostericum*), with emphasis on its East Asian members. Most species of *Angelica* s.l. fall into two major, disparate clades, the *Ostericum* clade and the *Angelica* group, with the latter comprising five major lineages that are distinguished both molecularly and morphologically: *Angelica* s.s. (including *Czernaevia*), *Archangelica*, *Coelopleurum*, *Glehnia*, and a newly identified Littoral *Angelica* clade. A North American *Angelica* clade was also distinguished in the ITS trees. Taxonomic realignments will be required, as many species of *Angelica* fall outside of *Angelica* s.s. and four species of *Angelica* occur outside of the *Angelica* group (*A. hirsutiflora*, *A. oncosepala*, *A. paeonifolia*, and *A. sinensis*). The *Angelica* s.s. clade contains predominantly East Asian species and comprises five major lineages, two of which represent plants exclusively from the eastern Himalayas; each of these five lineages can also be defined morphologically. The results obtained are significantly different from traditional treatments of *Angelica* s.l. and provide new insights into the phylogeny and classification of the group.

Keywords—Classification, fruit anatomy, ITS and ETS sequences, plastid DNA intron and intergenic spacer sequences, Umbelliferae.

Angelica sensu lato (s.l.; Apiaceae subfamily Apioideae) is a taxonomically complex and controversial group, exhibiting much morphological diversity and problematic generic limits. It comprises about 110 species, with major genera including *Angelica* L., *Archangelica* Hoffm., *Coelopleurum* Ledeb., *Czernaevia* Turcz. ex Ledeb., and *Ostericum* Hoffm. (Mathias 1965; Pimenov 1968b; Tutin 1968; Vasiléva and Pimenov 1991; Pimenov and Leonov 1993; Sheh et al. 2005). The group is distributed on all continents of the Northern Hemisphere, with its greatest number of species (about 55) concentrated in East Asia (Hiroe and Constance 1958; Siro and Gen 1978; Wen 1999; Sheh et al. 2005; Liao and He 2012; Liao et al. 2012a). Some of these East Asian species (e.g. *A. dahurica*, *A. decursiva*, and *A. sinensis*) are of great economic value, having been used in traditional Chinese medicine for hundreds of years (Shan 1992; Sheh et al. 2005; taxonomic authorities for all species considered in this investigation are presented in Appendix 1).

Fruit characters have long played a key role in the higher-level classification of subfamily Apioideae. The widely used system of Drude (1898), for example, emphasized the orientation of fruit compression and the number of ribs and vittae. *Angelica* s.l. is typically characterized by dorsally compressed mericarps with prominent dorsal ribs and broad lateral wings (Theobald 1971; Sheh et al. 2005). However, its fruits are polymorphic for these characters, resulting in differing treatments of poorly circumscribed taxa. Emphasizing fruit morphology, Qin et al. (1995) divided *Angelica* s.l. from East Asia and North America into taxa corresponding approximately to the five aforementioned genera. However, the most significant characters differentiating these genera (i.e. the degree of fruit compression, width of wings, and vittae number) appear homoplastic. Based solely upon these variable fruit characters, it has been difficult to infer evolutionary relationships within *Angelica* s.l. Heretofore, there has been no published morphological phylogenetic study of East Asian *Angelica* and its relatives.

To date, molecular phylogenetic analyses of *Angelica* s.l. have been based almost exclusively upon examination of small numbers of taxa using only nuclear ribosomal DNA (nrDNA) internal transcribed spacer (ITS) sequences. Xue et al. (2007), in their study of *Angelica* sensu stricto (s.s.) and allies from East Asia using 44 ITS sequences, proposed that *Angelica* was polyphyletic. With a slightly greater sampling, Feng et al. (2009) suggested a monophyletic *Angelica* s.s. upon the inclusion of *Coelopleurum*, *Czernaevia*, and *Ostericum koreanum*, but with the exclusion of several other species once recognized in *Angelica* s.l. As examples of the latter, *A. oncosepala* allied with the genus *Heracleum* in tribe Tordylieae, *A. sinensis* occurred in the *Sinodielsia* clade, and *Ostericum grosseserratum* was placed in the *Acronema* clade. In each of these studies, the sampling of East Asian *Angelica* s.l. included no more than half out of a total of approximately 110 known species and inference of phylogeny was based solely upon the use of ITS sequences.

Additional molecular tools, such as nrDNA external transcribed spacer (ETS) and chloroplast DNA (cpDNA) sequences, can be used to bolster phylogenetic hypotheses inferred using just ITS data. Logacheva et al. (2010) showed that the ETS region is a valuable phylogenetic marker in Apiaceae for low level analysis, as it had a higher percentage of parsimony informative characters than ITS across a comparable group of taxa. Plastid markers, while more conservatively evolving than either ITS or ETS, have already demonstrated their utility in phylogenetic analysis of the Apiaceae across a broad range of hierarchical levels (Downie et al. 2010). Other than our concurrent and related study of the historical biogeography of the East Asian *Angelica* group (Liao et al. 2012a), no other study of *Angelica* s.l. has incorporated evidence from ITS, ETS, and plastid DNA markers, or considered results of both molecular and morphological phylogenetic analyses simultaneously.

The objective of this study was to estimate the phylogeny of *Angelica* s.l. and infrageneric relationships in *Angelica* s.s.,

with an emphasis on its East Asian members. These relationships were inferred using phylogenetic analyses of nrDNA, cpDNA, and morphological data, and supplemented with evidence obtained from additional observations of fruit anatomy and micromorphology. The biogeographical history of *Angelica* s.l., with emphasis on the East Asian *Angelica* group, has been treated in a separate publication (Liao et al. 2012a).

MATERIALS AND METHODS

Molecular Study—Because some members of *Angelica* s.l. have a complicated relationship with other taxa of Apiioideae, a broad sampling of tribe Selineae was included in the molecular study, as well as additional representatives of other tribes and major clades within the subfamily. Sampling of East Asian *Angelica* included 44 of its approximately 55 known species, with the majority of specimens collected from the wild. Voucher information for all accessions included in the molecular study is presented in Appendix 1.

For the molecular analyses, four datasets were constructed. Dataset I included the ITS1 and ITS2 spacer regions from 116 representatives (33 genera) of subfamily Apiioideae in order to ascertain the major lineages of *Angelica* s.l. and their allied taxa. The intervening 5.8S region was excluded from this dataset because it is missing from many Apiaceae sequences already deposited in GenBank. *Notopterygium forbesii* and *Pleurospermum franchetianum* were used to root the trees because they occupied basal branches in phylogenies of Chinese Apiioideae (Zhou et al. 2008, 2009). Datasets II, III, and IV each comprised 46 representatives of the *Angelica* group, plus three outgroups (*Cnidium monnieri* and two species of *Peucedanum* L.) whose selection was based on results of phylogenetic analyses of dataset I. Dataset II included sequences from both the ITS (including 5.8S) and ETS regions, whereas dataset III included sequences from four noncoding cpDNA loci: *rps16* intron, *rps16-trnK* intergenic spacer, *rpl32-trnL* intergenic spacer, and *trnL-trnT* intergenic spacer. Dataset IV represented combined nrDNA and cpDNA data for these 49 representatives. These four datasets, as well as the trees derived from them, are available in TreeBASE (No. S12434).

Total genomic DNA was extracted from 15–20 mg of leaves obtained from silica-gel-dried, field-collected material or herbarium specimens following a modification of the CTAB protocol of Doyle and Doyle (1987). The primer sequences for ITS amplification were published previously in White et al. (1990), the ETS primer sequences were obtained from Wright et al. (2001), the primers for *rps16-trnK* and *rpl32-trnL* were those of Steane et al. (2005), and those for amplification of the *rps16* intron and *trnL-trnT* intergenic spacer regions were first reported by Shaw et al. (2005, 2007). PCR amplification conditions for the ITS and ETS regions were as follows: an initial denaturation at 94°C (3 min), followed by 30 cycles of 94°C denaturation (45 s), 55°C annealing (45 s), and 72°C extension (45 s), with a final extension for 10 min at 72°C. The PCR parameters for amplification of each plastid locus were an initial denaturation at 94°C (3 min), followed by 35 cycles of 94°C denaturation (1 min), 52°C annealing (1 min), and 72°C extension (1 min), with a final extension for 10 min at 72°C. PCR products were separated in a 1.5% (w/v) agarose TAE gel and purified using the Wizard PCR Preps DNA Purification System (Promega Corporation, Madison, Wisconsin) following the manufacturer's instructions. Cycle sequencing reactions were performed using the purified PCR product, AmpliTaq DNA polymerase (Life Technologies Corporation, Carlsbad, California), and fluorescent BigDye terminators. The sequencing products were resolved using an ABI Prism 310 DNA sequencer (Applied Biosystems; now Life Technologies Corporation).

DNA sequences were initially aligned using the default pairwise and multiple alignment parameters in Clustal X (Jeanmougin et al. 1998), then rechecked and adjusted manually as necessary using MEGA4 (Tamura et al. 2007). Gaps were positioned to minimize nucleotide mismatches. Uncorrected pairwise nucleotide differences were determined using PAUP* version 4.0b10 (Swofford 2003).

Phylogenetic analyses of dataset I were conducted by employing maximum parsimony (MP) and Bayesian Inference (BI) methods, using the programs PAUP* and MrBayes version 3.1.2 (Ronquist and Huelsenbeck 2003), respectively. For the MP analysis, heuristic searches were carried out with 1000 random addition sequence replicates. One tree was saved at each step during stepwise addition and tree-bisection-reconnection (TBR) was used to swap branches; the maximum number of trees was set to 20,000. All characters were unordered and equally weighted. Gaps were treated as missing data. Bootstrap support (BS)

values were calculated from 1,000,000 replicate analyses using “fast” stepwise-addition of taxa and only those values compatible with the majority-rule consensus tree were recorded. Prior to the BI analysis, MrModeltest version 2.2 (Nylander 2004) was used to determine a best-fit model of nucleotide substitution and the GTR+I+G model under the Akaike Information Criterion (AIC; Akaike 1974) was selected. From a random starting tree, the BI analysis was run for 6 million generations and the trees were saved to a file every 100 generations. Ten simultaneous Markov chain Monte Carlo (MCMC) chains were run, and the temperature was adjusted to 0.1 in order to keep an appropriate heat range. Branch lengths of the trees were saved. Variation in likelihood scores to determine apparent stationarity was examined graphically for each independent run using the program Tracer 1.4 (Drummond and Rambaut 2007). The first 12,000 trees were discarded as “burn-in” and a majority-rule consensus tree was calculated based upon the remaining 48,000 trees. Datasets II to IV were each analyzed using MP, BI, and maximum likelihood (ML) methods. The MP analyses were the same as those described for dataset I. The BI analyses were run for 8 million generations and the trees were saved to a file every 100 generations. Ten simultaneous MCMC chains were run and the temperature was adjusted to 0.1. The first 16,000 trees were discarded as “burn-in” and a majority-rule consensus tree was calculated based upon the remaining 64,000 trees. The model of nucleotide substitution for the ML analysis was selected using Modeltest version 3.06 (Posada and Crandall 1998). The GTR+I+G model was chosen based on the AIC. Maximum likelihood searches were performed using a heuristic search method implemented using PHYML version 2.4 (Guindon and Gascuel 2003), each with 100 random sequence additions. Bootstrap values were not calculated for the ML trees because of the large amount of time required to obtain these data in an initial run.

To examine the extent of conflict between the nrDNA and cpDNA data partitions (datasets II and III), the incongruence length difference test (ILD; Farris et al. 1994) was carried out using PAUP*. This test was implemented with 1000 partition-homogeneity test replicates, using a heuristic search option with simple addition of taxa, TBR branch swapping, and MaxTrees set to 1000.

Fruit Anatomical and Micromorphological Study—Mature fruits from 42 species of *Angelica* s.l. and its allies (including 29 *Angelica* species, six *Ostericum* species, two *Archangelica* species, *Coelopleurum saxatile*, *Czernaevia laevigata*, *Glehnia littoralis*, *Heraclium xiaojinense*, and *Ligusticum angelicifolium*) were included in the fruit anatomical and micromorphological study (Appendix 1). For most of these species, fruits of at least three individuals from each of several populations were examined.

For observation of anatomical features, mature fruits were subjected to traditional paraffin sectioning methods: (1) dehydration for paraffin embedding (subsequent changes of 50%, 75%, and 95% ethanol/water, and then two changes of 100% ethanol, 1 hr each step; changes of 30%, 50%, and 80% xylene/ethanol, and then two changes of 100% xylene, 2 hrs each step; paraffin wax at 60°C, two changes, 4 hrs each step); (2) embedding of tissues into paraffin blocks; (3) slicing paraffin blocks at 10–15 μ m and expansion of paraffin ribbons on glass at 45–50°C; (4) deparaffinization and rehydration of sections in 100% xylene, 50% xylene/ethanol and gradient ethanol, respectively; and (5) staining with Safranin O/Fast Green.

For micromorphological observations, mature fruits were mounted on stubs using double-sided adhesive tape after ultrasonic cleaning in 95% ethanol/water and drying. Each sample was coated with a thin layer of gold and examined at 800 \times using a Hitachi-SX-450 scanning electron microscope. Terminology to describe surface ornamentation of these fruits follows that of Liu et al. (2004).

Morphological Study—In total, 48 taxa were examined for morphological character variation, including 45 species from the *Angelica* group and their subspecific taxa. Three of these taxa were not included in the cpDNA study (*Angelica decursiva* f. *albiflora*, *A. genuflexa*, and *Archangelica officinalis*) and two taxa were not included in any molecular analyses [*Angelica cartilaginosmarginata* (Makino ex Y. Yabe) Nakai var. *cartilaginosmarginata* and *Coelopleurum nakaianum* (Kitag.) Kitag.] because of technical difficulties in obtaining DNA.

The characters examined included features of the stem, leaf, inflorescence, flower, pollen, and fruit, with those of the mericarp given emphasis because they are most variable and have been considered important in traditional treatments of Apiioideae (e.g. Qin et al. 1995; Pimenov et al. 2000; Sheh et al. 2005). Data were obtained from the aforementioned fruit anatomical study, field-based observations, and by examination of herbarium specimens from several major Chinese herbaria (Appendix 1). Pollen data were obtained mainly from Sheh et al. (1997) and Shu and Sheh (2004). For a few taxa and characters, data were obtained directly

from the literature (Hiroe and Constance 1958; Tutin 1968; Shan 1992; Kao 1993; Sheh et al. 2005). Fifty-three characters were considered, of which 20 character states were treated as ordered (Table 1). For the continuous quantitative characters, character states were delimited by detecting discontinuities in character variation (Stevens 1991). Phylogenetic analysis of the morphological dataset was carried out using MP. Heuristic searches were replicated 1000 times using random stepwise addition of taxa and TBR branch swapping. Bootstrap support values were calculated from one million replicate analyses using "fast" stepwise-addition of taxa. The

morphological dataset, as well as the consensus tree derived from these data, is available in TreeBASE (No. S12434).

RESULTS

Sequence Characteristics—Sequence characteristics of each of the four molecular datasets, including partitioned data for

TABLE 1. Morphological characters and states used in the phylogenetic analysis of the *Angelica* group. An asterisk (*) indicates that the character state was treated as ordered. ^asub-rectangular: length / width \leq 2.3; super-rectangular: length / width \geq 2.4 (Sheh et al. 1997), ^bsize index = the square root of (length \times width) (Shu and Sheh 2004)

No.	Character	States
1	Habit	biennial (0), perennial (1)
2	Plant odor	slight (0), strong (1)
3	Root diameter (cm)*	< 1 (0), 1–2.5 (1), > 2.5 (2)
4	Plant height (m)*	< 0.3 (0), 0.3–2 (1), > 2 (2)
5	Stem presence	absent (0), present (1)
6	Stem diameter (cm)*	< 0.8 (0), 0.8–2 (1), > 2 (2)
7	Stem internal structure	hollow (0), solid (1)
8	Stem indumentum	glabrous (0), villous (1), densely pubescent (2)
9	Basal blade size (cm)*	< 30 \times 20 (0), 30 \times 20–60 \times 40 (1), > 60 \times 40 (2)
10	Form of blade division*	once-pinnate (0), 2–3-pinnate (1), 4-pinnate (2)
11	Blade texture*	papery (0), thinly leathery (1), leathery (2)
12	Blade indumentum	glabrous (0), villous (1), densely pubescent (2)
13	Leaflet shape	elliptic (0), lanceolate (1), subrhombic (2)
14	Leaflet size (cm)*	< 10 \times 5 (0), > 10 \times 5 (1)
15	Form of leaflet margin*	densely serrulate (0), irregularly biserrate (1), incised-serrate (2)
16	Form of leaflet base	cuneate (0), decurrent (1)
17	Presence of white-cartilaginous leaflet margin	absent (0), present (1)
18	Sheath shape	undeveloped (0), tubular (1), saccate (2)
19	Sheath indumentum	glabrous (0), villous (1), densely pubescent (2)
20	Petiole internal structure	hollow (0), solid (1)
21	Petiole indumentum	glabrous (0), villous (1), densely pubescent (2)
22	Form of rachis and petiolules	not geniculate (0), geniculate (1)
23	Umbel diameter (cm)*	< 8 (0), 8–20 (1), > 20 (2)
24	Number of rays*	< 20 (0), 20–40 (1), > 40 (2)
25	Umbel shape	upwards-open (0), subglobose (1)
26	Bract shape	lanceolate (0), saccate (1)
27	Bract number*	absent or 1–2 (0), more than five (1)
28	Bracteole number*	absent (0), many (1)
29	Flower color	white (0), yellowish-green or greenish-white (1), dark purple (2)
30	Petal shape	ellipsoid-lanceolate (0), obcordate (1)
31	Outer petal size	not enlarged (0), enlarged (1)
32	Stylopodium shape	conical (0), flattened (1)
33	Stylopodium color	green (0), purple (1)
34	Pollen shape (outline in equatorial view) ^a	elliptic (0), sub-rectangular (1), super-rectangular (2), equatorially-constricted (3)
35	Pollen size [size index ^b]*	17–21 (0), 21–25 (1), 25–29 (2)
36	Fruit shape	subround (0), obovate/rectangular (1), narrow-elliptic (2)
37	Fruit size (mm)*	< 3 \times 4 (0), 3 \times 4–6 \times 8 (1), > 6 \times 8 (2)
38	Fruit indumentum	glabrous (0), pubescent (1)
39	Fruit rib development	equal-developed (0), unequal-developed (1)
40	Dorsal ribs form*	low keeled (0), narrow-winged (1), corky-winged (2)
41	Lateral rib width*	narrower than half of the body (0), as wide as the body (1), much wider than the body (2)
42	Lateral ribs thickness	corky (0), thick (1), thin (2)
43	Endosperm shape in transverse section	subround (0), sub-rectangular (1), narrow-elliptic (2), fan-shaped (3)
44	Degree of endosperm dorsal compression (Width/Height)*	1.0–1.2 (0), 1.2–2 (1), > 2 (2)
45	Dorsal face of endosperm	smooth (0), undulate (1), with grooves (2)
46	Mesocarp thickness*	corky (0), thick (1), thin (2)
47	Cavity between pericarp and seed	absent (0), present (1)
48	Arrangement of vittae	almost encircling the seed (0), triple in each furrow (1), single or double in each furrow (1)
49	Vittae visibility on commissure face	invisible (0), visible (1)
50	Size of vittae*	quite small (0), medium (1), conspicuously enlarged (2)
51	Vittae number on the commissure*	numerous (0), 6–8 (1), 2–4 (2), absent (3)
52	Size of vascular bundle	reduced (0), not reduced (1)
53	Separation of mature mericarps	some adherence to each other (0), easily separated (1)

TABLE 2. Sequence characteristics of the four primary DNA datasets considered in this study (datasets I–IV), as well as partitioned data for the nrDNA ITS and ETS regions and the four cpDNA loci.

Characteristic	Data matrix									
	ITS1 + ITS2 (dataset I)	ITS	ETS	<i>rps16</i> intron	<i>rps16-trnK</i>	<i>rpl32-trnL</i>	<i>trnL-trnT</i>	nrDNA (dataset II)	cpDNA (dataset III)	combined (dataset IV)
No. of accessions	116	49	49	49	49	49	49	49	49	49
No. of total aligned positions	479	611	519	835	766	1158	919	1130	3687	4782
No. of constant positions (and %)	127 (26.5)	430 (70.4)	315 (60.7)	781 (93.5)	691 (90.2)	1002 (86.5)	859 (93.5)	744 (65.8)	3356 (91.0)	4048 (84.7)
No. of autapomorphic positions (and %)	67 (14.0)	91 (14.9)	101 (19.58)	33 (4.0)	43 (5.6)	88 (7.6)	43 (4.7)	190 (16.8)	195 (5.3)	401 (8.4)
No. of parsimony informative positions (and %)	285 (59.5)	90 (14.7)	103 (19.9)	21 (2.5)	32 (4.2)	68 (5.9)	174 (1.9)	196 (17.4)	136 (3.7)	333 (7.0)
No. of indels	—	—	2	10	62	83	57	—	—	—
Divergence range (%)	0–40.3	0–8.4	0–12.1	0–3.6	0–14.8	0.4–15.6	0–8.3	0–9.6	0.2–7.5	0.2–7.8
Average divergence (%)	12.1	3.9	4.9	1.2	3.5	5.0	2.5	4.4	3.1	3.5

ITS, ETS, and each of the four cpDNA loci examined, are presented in Table 2. These results indicate that average sequence divergence of ETS is more variable (4.9%) than ITS (3.9%) across the *Angelica* group. The ETS region is slightly smaller than ITS, but contributed more autapomorphic and parsimony informative characters than ITS. The *rpl32-trnL* region is the most variable plastid locus, with 5.0% average sequence divergence and numerous small indels (83). The next most variable plastid locus is *rps16-trnK*, with 3.5% average sequence divergence and 62 indels. Among the four plastid loci examined, the *rps16* intron has the lowest sequence divergence and the least number of indels. Overall, dataset II (nrDNA ITS & ETS) was more variable than dataset III (cpDNA), with greater average sequence divergence and more parsimony informative characters, in spite of being one-third the size.

Phylogenetic Analyses of Molecular Data—The trees generated from MP and BI analyses of dataset I were similar in topology, thus only the Bayesian tree with posterior probabilities (PP) and BS values obtained from the MP analysis is shown (Fig. 1). MP heuristic searches of dataset I resulted in 20,000 shortest trees, each with a length (L) of 1484 steps, a consistency index (CI; including uninformative characters) of 0.42, and a retention index (RI) of 0.70. Members of *Angelica* s.l. fell primarily into two major lineages: (1) the *Ostericum* clade (PP = 1.00, BS = 100), which is sister to tribe Scandiceae; and (2) the *Angelica* group (with weak branch support). The nomenclatural type of *Angelica*, *A. sylvestris*, occurs within the *Angelica* group and sequence divergence values between this species and members of the *Ostericum* clade varied from 17.6–19.7% for ITS, 20.9–24.8% for ETS, and 7.4–10.8% for the *rps16* intron. The *Angelica* group occurred in tribe Selineae and comprised six major lineages: *Angelica* s.s. clade (PP = 0.79, BS < 50), *Archangelica* clade (PP = 0.90, BS = 53), *Coelopleurum* clade (PP = 0.98, BS = 62), the Littoral *Angelica* clade (PP = 1.00, BS = 59), the North American *Angelica* clade (PP = 0.99, BS = 79), and the *Glehnia* clade (PP = 1.00, BS = 98). The *Archangelica* clade was represented by *Archangelica officinalis* (the type of the genus *Archangelica*), two additional species of *Archangelica*, and the North American species *Angelica ampla*. The *Coelopleurum* clade comprised *Coelopleurum saxatile* and *C. lucidum*, which arose from within a paraphyletic *Magadania* Pimenov & Lavrova, a small genus endemic to Northeast Asia. The Littoral *Angelica* clade comprised *A. furcijuga*, *A. japonica*, *A. morii*, and *A. shikokiana*, all of which inhabit the East Asian littoral regions or islands (i.e. southeast littorals of mainland China, Kyushu, Ryukyu, Shikoku, and Taiwan). The North American *Angelica* clade consisted of five species endemic to North America (i.e. *A. arguta*, *A. breweri*, *A. grayi*, *A. pinnata*, and *A. roseana*), and the *Glehnia* clade comprised both varieties of this monotypic genus. Four species of *Angelica* fell outside of the *Angelica* group: *A. hirsutiflora* was placed in the *Peucedanum* s.s. clade; *A. onosepala* was placed sister to *Heracleum hemsleyanum* in tribe Tordylieae; and *A. paeoniifolia* and *A. sinensis* were grouped with *Conioselinum* Fisch. ex Hoffm. and its allies in the *Sinodielsia* clade.

The *Angelica* s.s. clade comprised 43 accessions representing 37 species and four genera. Included among the species of *Angelica* within this clade were *Czernaevia laevigata*, *Heracleum xiaojinense*, *Ostericum huadongensis*, and *O. koreanum*. Although the *Angelica* s.s. clade is weakly supported, it does contain predominantly East Asian species (the two exceptions

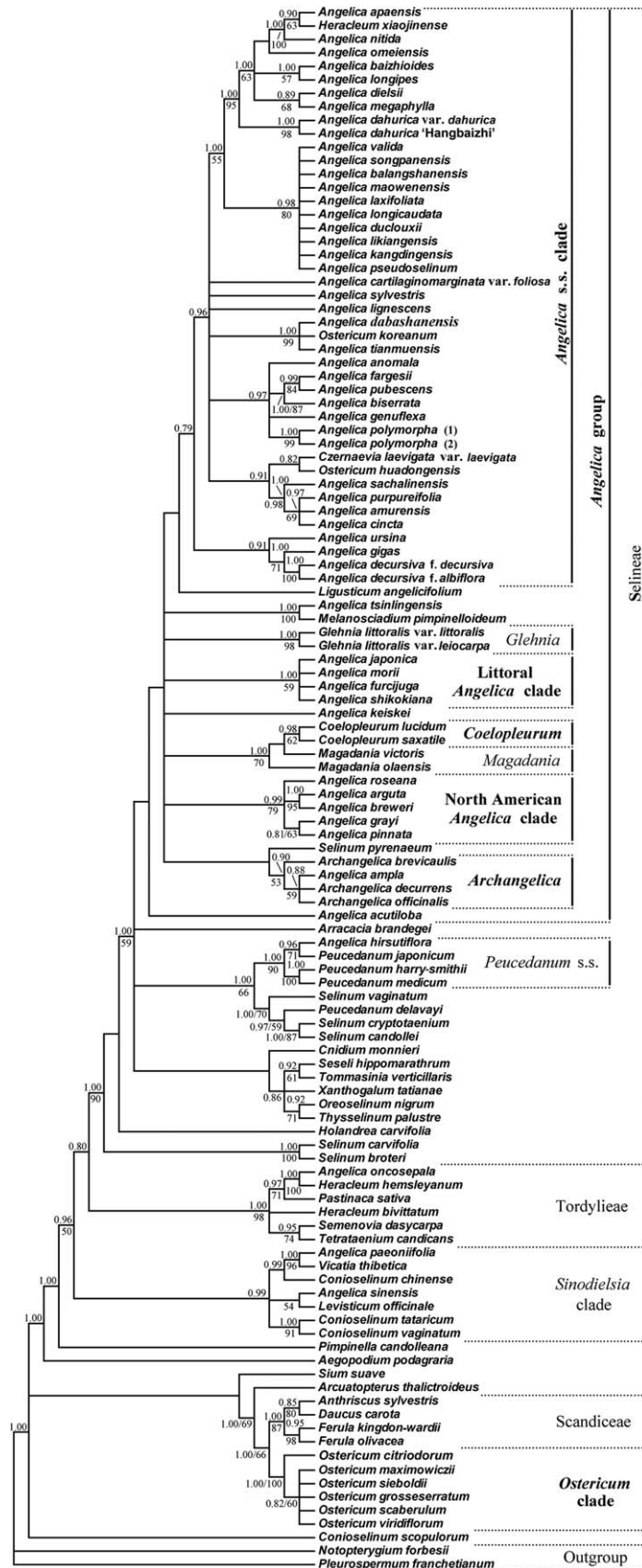


FIG. 1. Majority-rule consensus tree retrieved by Bayesian inference analysis of 116 accessions (dataset I) representing the *Angelica* group and other major lineages of Apiaceae subfamily Apioideae using a GTR+I+G nucleotide substitution model. Numbers along branches indicate Bayesian posterior probabilities (only those > 0.75 are shown). Because this tree was highly consistent to a strict consensus tree derived from maximum parsimony analysis, bootstrap support values (> 50) for similar clades are also provided. The names of the clades identified are those of Zhou et al. (2008, 2009) and Downie et al. (2010).

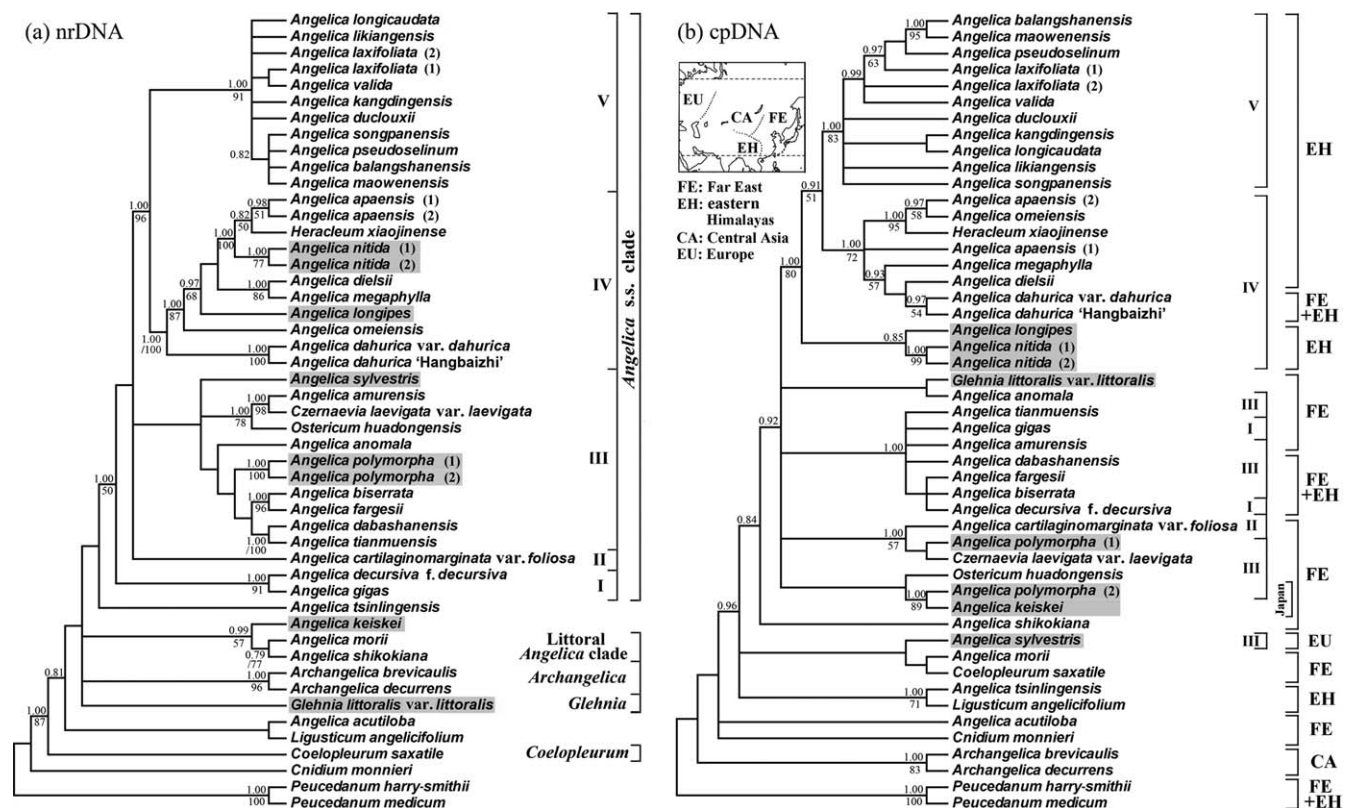
being *A. lignescens* and *A. sylvestris*, Liao et al. 2012a); relationships within this clade, however, are poorly resolved. To further explore relationships among these East Asian species, 33 of these accessions, plus additional single accessions each of *A. apaensis*, *A. laxifoliata*, and *A. nitida*, were analyzed using ETS and cpDNA evidence (datasets II and III). Ten representatives from each of the other major *Angelica* group clades (with the exception of the North American *Angelica* clade), plus three outgroups were also included. Excluded from these analyses were *A. baizhioides* (Xue et al. unpubl. data), *A. cincta*, *A. genuflexa*, *A. lignescens*, *A. pubescens*, *A. purpureifolia*, *A. sachalinensis*, *A. ursina*, and *O. koreanum* because their ITS sequences were obtained from GenBank and we did not have access to material to sequence their corresponding plastid genes. Additionally, we could not sequence the cpDNA regions in *A. decursiva* f. *albiflora* despite our best efforts.

For datasets II (nrDNA) and III (cpDNA), the topologies of the MP, BI, and ML trees were similar to each other, therefore only the ML trees of each are presented, with BS and PP values from the other analyses indicated along their branches (Fig. 2). Twenty minimal length trees (L = 655 steps, CI = 0.72, RI = 0.80) were retrieved in the MP analysis of dataset II. NrDNA evidence provides varying support for the *Angelica* s.s. clade (PP = 1.00, BS = 50). Within this clade, five major lineages were identified (I–V), with three of them well supported (PP = 1.00, BS > 90). MP analysis of the cpDNA dataset retrieved 20,000 minimal length trees (L = 485 steps, CI = 0.74, RI = 0.73). The trees resulting from the three

analyses of these plastid DNA data, however, were generally poorly resolved, with only a few strongly supported clades, such as the *Archangelica* clade (PP = 1.00, BS = 83) and a group comprised of the aforementioned lineages IV and V (PP = 1.00, BS = 80). The latter group represents plants exclusively from the eastern Himalayas and has been referred to as the eastern Himalayan lineage (Liao et al. 2012a).

Results of a partition homogeneity test for the nrDNA and cpDNA datasets indicated that these genomes provide significantly different phylogenetic estimates (ILD probability value = 0.01). Those taxa involved in this conflict are highlighted in Fig. 2, and include *A. keiskei*, *A. longipes*, *A. nitida*, *A. sylvestris*, *Glehnia littoralis* var. *littoralis*, and two populations of *A. polymorpha*. *Glehnia littoralis* var. *littoralis*, which is widely distributed on East Asian beaches, and the Japanese endemic species *A. keiskei* fell outside of the *Angelica* s.s. clade in the nrDNA trees, but were nested among the northeast Asian members of the *Angelica* s.s. clade in the cpDNA trees; in particular, *A. keiskei* is sister to Japanese *A. polymorpha* in the latter. *Angelica sylvestris* from Europe does not ally with members of the *Angelica* s.s. clade in the cpDNA trees, whereas it does in the nrDNA trees.

MP analysis of the matrix of combined nrDNA and cpDNA data (dataset IV) resulted in 11 maximally parsimonious trees (L = 652 steps, CI = 0.79, RI = 0.57). The strict consensus of these trees was topologically congruent to those inferred using BI and ML methods, therefore only the ML tree is shown with PP and BS values indicated along its



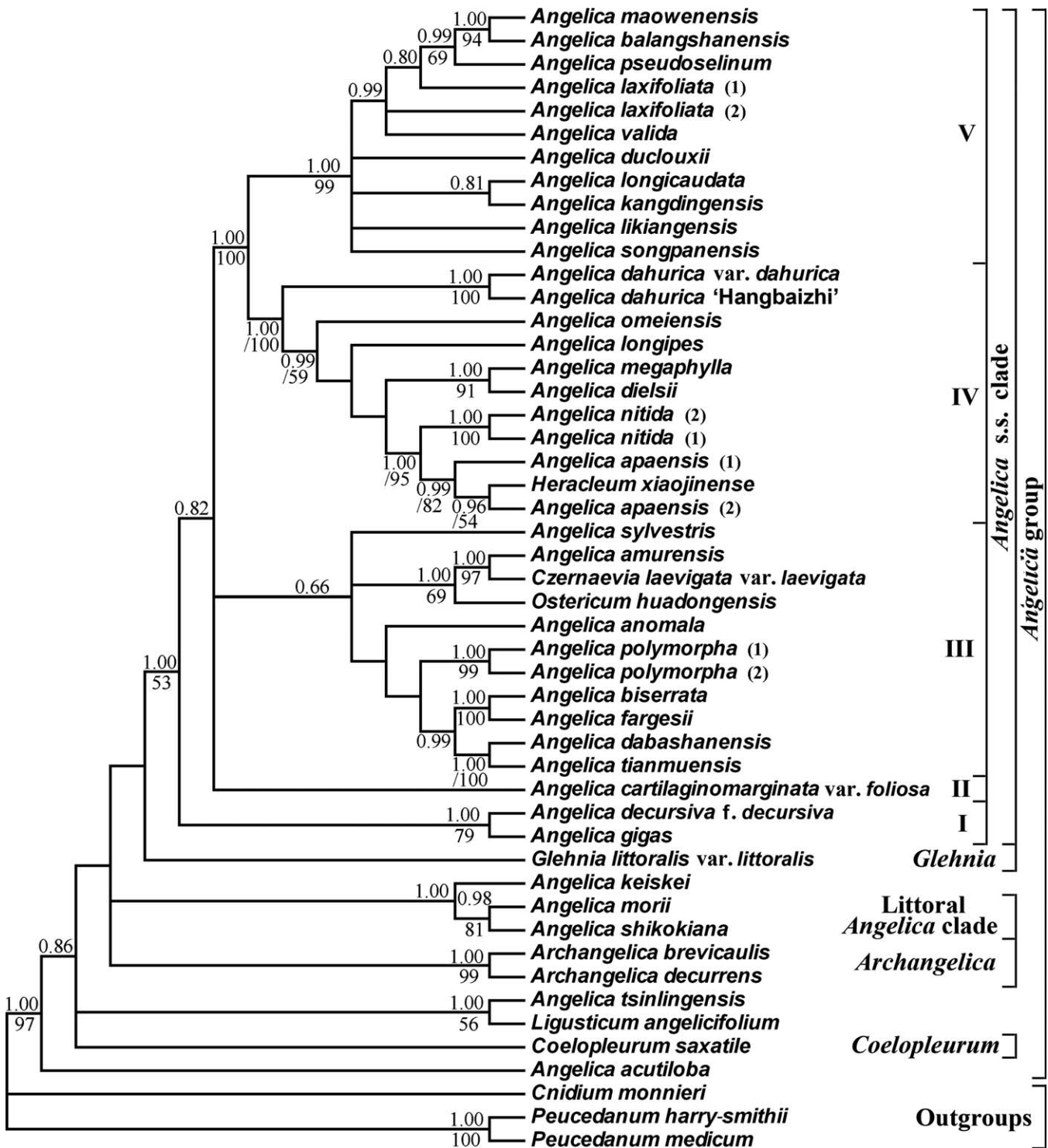


FIG. 3. Maximum likelihood tree derived from analysis of combined nrDNA and cpDNA data (dataset IV) derived from 49 accessions of the *Angelica* group and outgroups. This tree is topologically congruent with those trees inferred using Bayesian inference and maximum parsimony analyses, therefore numbers above and below branches are posterior probability (> 0.75) and bootstrap support (> 50) values, respectively. The Roman numerals indicate the same lineages as identified in Fig. 2.

branches (Fig. 3). Monophyly of the *Angelica* s.s. clade was supported in all analyses (PP = 1.00, BS = 53), as was the monophyly of the Littoral *Angelica* clade (PP = 0.98, BS = 81), with *A. keiskei* as its sister group. The same five lineages as determined previously were identified in the *Angelica* s.s. clade. *Angelica decursiva* grouped with *A. gigas* in Clade I with strong branch support (PP = 1.00, BS = 79). *Angelica cartilagi-*

nomarginata var. *foliosa* comprised an isolated lineage (Clade II). Clade III included eight species of *Angelica*, including its type *A. sylvestris*, plus *Czernaevia laevigata* and *Ostericum huadongensis*; however, this clade was weakly supported (PP = 0.66, BS < 50). Clade IV was supported strongly (PP = 1.00, BS = 100) and included *A. apaensis*, *A. dahurica*, *A. dielsii*, *A. longipes*, *A. megaphylla*, *A. nitida*, *A. omeiensis*, and

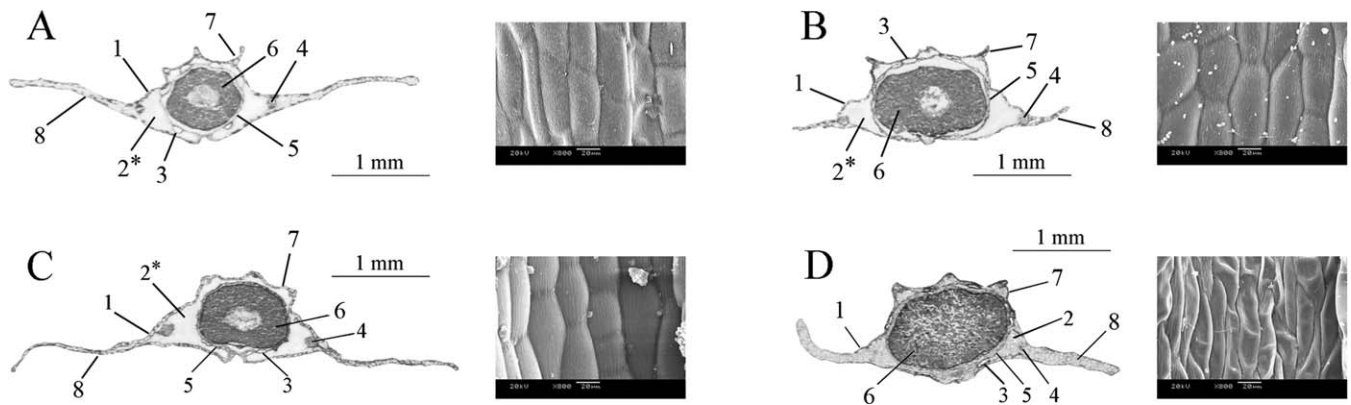


FIG. 4. Fruit structure and epidermal micromorphology (800 \times) of the genus *Ostericum*. (A) *O. maximowiczii*, (B) *O. sieboldii*, (C) *O. scaberulum*, and (D) *O. huadongensis*. 1. exocarp, 2. mesocarp, 2*. destroyed mesocarp (cavity), 3. vittae, 4. vascular bundles, 5. endocarp, 6. endosperm, 7. dorsal ribs, 8. lateral ribs.

Heracleum xiaojinense. The fifth lineage also received strong support (PP = 1.00, BS = 99) and consisted of *A. balangshanensis*, *A. duclouxii*, *A. kangdingensis*, *A. laxifoliata*, *A. likiangensis*, *A. longicaudata*, *A. maowenensis*, *A. pseudoselinum*, *A. songpanensis*, and *A. valida*. *Angelica acutiloba* and *A. tsinlingensis* were isolated from the *Angelica* s.s. clade. The latter allied with *Ligusticum angelicifolium* (PP = 1.00, BS = 56) and the former arose as an early-diverging branch within the *Angelica* group.

Fruit Anatomy and Morphology of *Angelica* s.l.—With one exception, the fruit anatomy of *Ostericum* is significantly different from that of all other members of *Angelica* s.l. For example, the exocarp consists of one layer of relatively large cells with convex and thickened outer walls (most easily seen in Fig. 4C); in *Angelica* s.s. species, these cells are much smaller and not easily distinguishable (cf. Figures 4D and 6, as examples). In addition, the mesocarp cells of *Ostericum* species are destroyed when mature, and all of its vascular bundles are reduced (Fig. 4A–C). Typically, vascular bundles are located in the mesocarp below each rib and are surrounded by parenchyma cells; a reduced vascular bundle, however, is smaller in size and always placed outside of the mesocarp or in a cavity. Under scanning electron micros-

copy, epidermal cells of the fruits of *Ostericum* are convex, rhombic-rectangular in shape and protruding with lineate ornamentation (Fig. 4A–C). In contrast, the fruit of *O. huadongensis* is quite different anatomically—its mesocarp is well-developed at maturity, as are the vascular bundles, the exocarp consists of relatively small cells, and the epidermal cells are concave in shape (Fig. 4D). These features are also found in all other members of the *Angelica* group (Figs. 5 and 6), but not in any other *Ostericum* species.

The mericarps of *Archangelica*, *Coelopleurum*, and *Glehnia* F. Schmidt each have a corky mesocarp, more or less equally developed winged ribs with inflated bases, and a slightly compressed endosperm (Fig. 5A–C); moreover, their dual mericarps always adhere to each other at maturity. Equally developed ribs refers to their general shape and length, and while the lengths of the dorsal ribs may not always be equal to each other or to those of the lateral ribs, they are similar to one another in overall shape and structure. *Coelopleurum* can be distinguished from the other two genera by its fewer and enlarged vittae (Fig. 5B, structure 3) and reduced vascular bundles that occur outside of the mesocarp, adhering to its inner face (Fig. 5B, structure 4*).

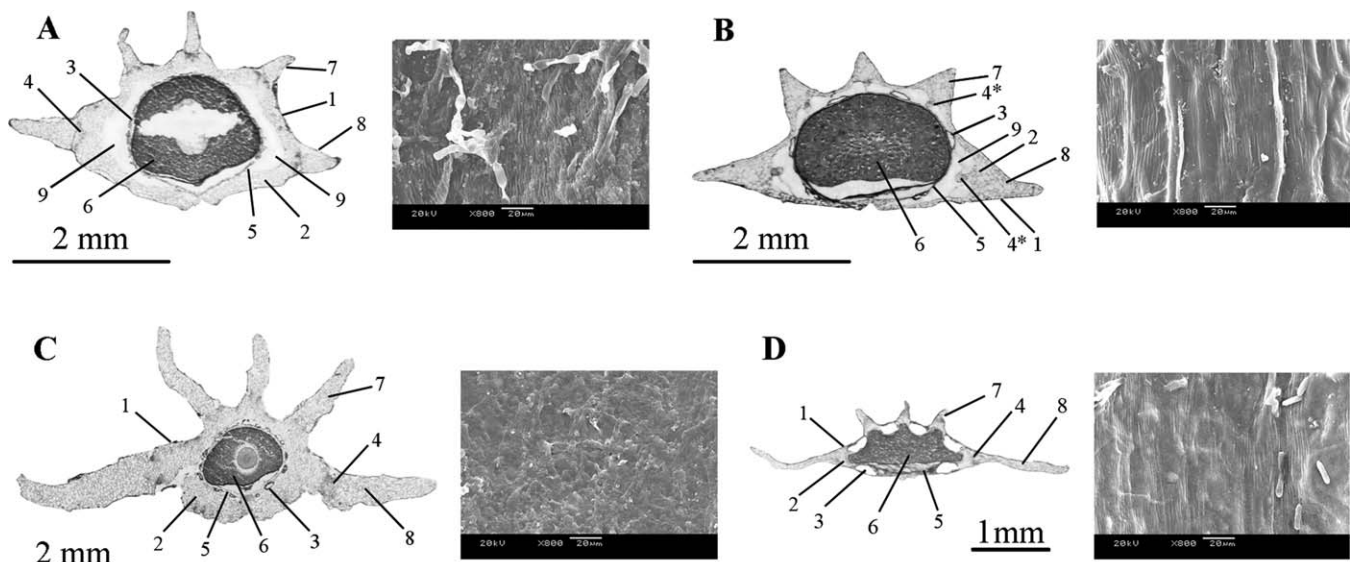


FIG. 5. Fruit structures and epidermal micromorphology (800 \times) of (A) *Archangelica decurrens*, (B) *Coelopleurum saxatile*, (C) *Glehnia littoralis* var. *littoralis*, and (D) *Angelica morii*. 1. exocarp, 2. mesocarp, 3. vittae, 4. vascular bundles, 4*. reduced vascular bundles, 5. endocarp, 6. endosperm, 7. dorsal ribs, 8. lateral ribs, 9. cavity.

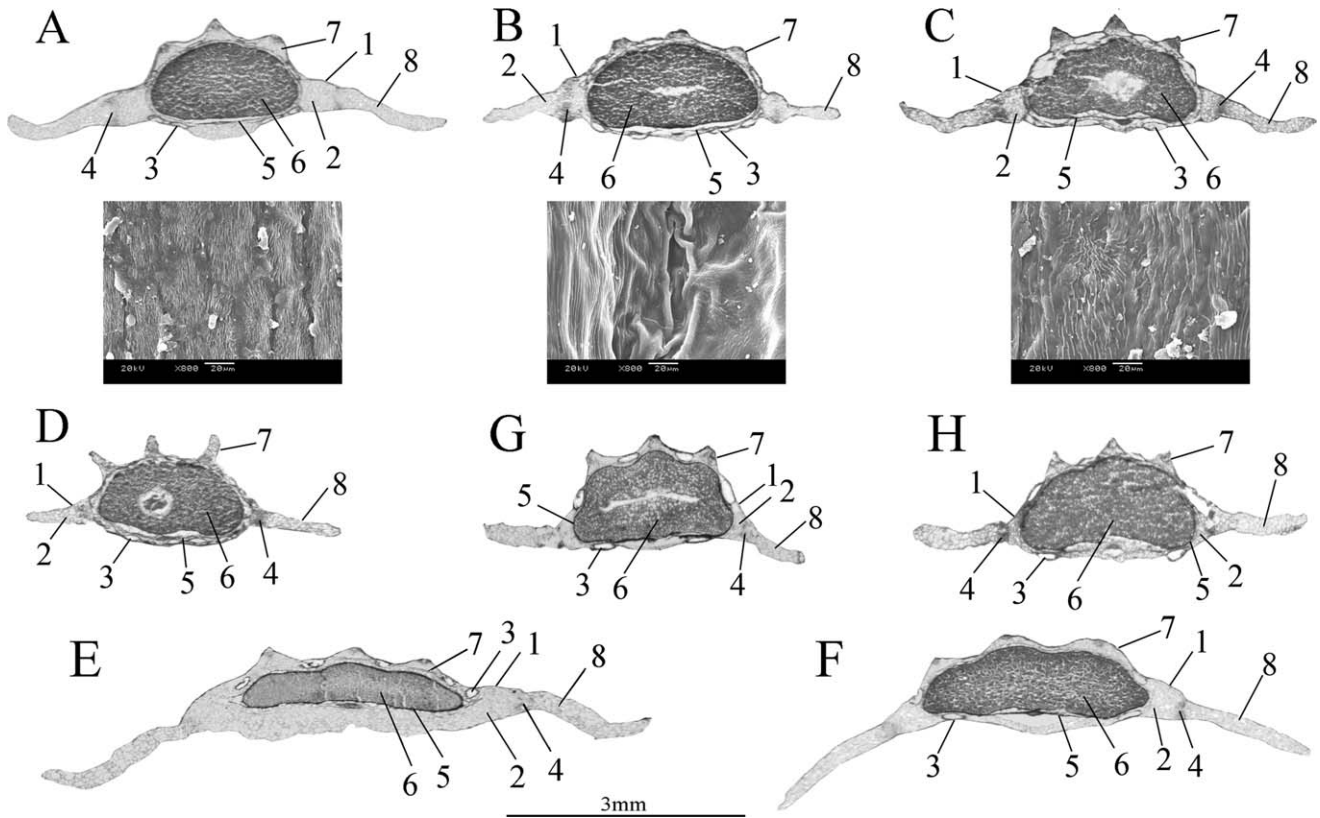


FIG. 6. Fruit structures of eight representatives of the *Angelica* s.s. clade and epidermal micromorphology (800×) of three representatives. (A) *Angelica gigas*, (B) *A. cartilaginomarginata* var. *foliosa*, (C) *A. amurensis*, (D) *Czernaevia laevigata* var. *laevigata*, (E) *A. apaensis*, (F) *A. dahurica* var. *dahurica*, (G) *A. longicaudata*, and (H) *A. pseudoselinum*. 1. exocarp, 2. mesocarp, 3. vittae, 4. vascular bundles, 5. endocarp, 6. endosperm, 7. dorsal ribs, 8. lateral ribs.

The vittae of *Archangelica* and *Glehnia* are more numerous and smaller (and are almost encircling and adhering to the seed) and their vascular bundles are well-developed. Members of the Littoral *Angelica* clade, as represented by *A. morii* (Fig. 5D), have a mericarp bearing narrowly winged dorsal ribs and a fan-shaped endosperm with four grooves on its dorsal face. In

contrast, members of *Angelica* s.s. have ribs that are unequally-developed because the dorsal ribs (low, filiform or rounded) are different from the two lateral ribs (broad winged) both in shape and structure (Fig. 6).

Fruit sections of eight representative members of the *Angelica* s.s. clade (Fig. 6A–H) show the following

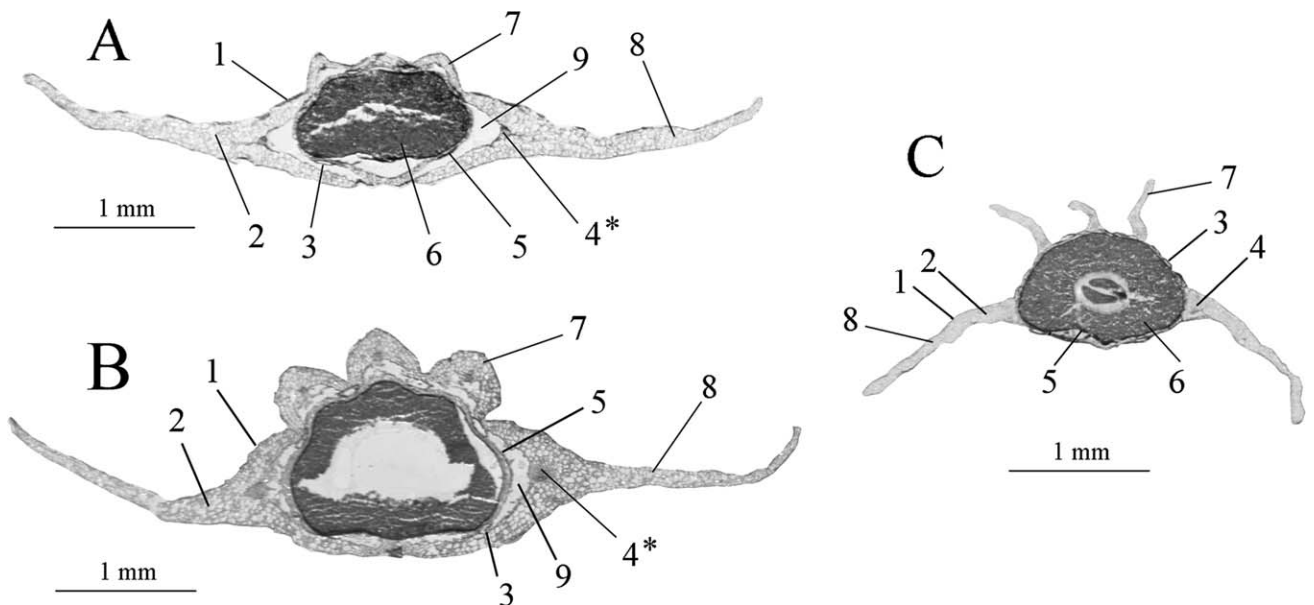


FIG. 7. Fruit structures of (A) *Angelica sinensis*, (B) *A. ternata*, and (C) *A. tsinlingensis*. 1. exocarp, 2. mesocarp, 3. vittae, 4. vascular bundles, 4*. reduced vascular bundles, 5. endocarp, 6. endosperm, 7. dorsal ribs, 8. lateral ribs, 9. cavity.

characteristics: mesocarp not corky and mostly adhering to the seed; ribs unequally developed (i.e. the dorsal ribs are low, filiform or rounded, and the lateral ribs are broad-winged and wider than half the body); endosperm conspicuously dorsally compressed; vittae relatively few and enlarged; and vascular bundles non-reduced. In addition, the mature mericarps are easily separated.

Angelica sinensis and *A. ternata* share similar fruit features. Their dorsal ribs are low-rounded, the exocarp is interrupted near the carpophore, the mesocarp is relatively thick, the vascular bundles are significantly reduced and adhere to the inside of the mesocarp, and the vittae are barely visible on the commissural face (Fig. 7A and B). The fruit of *A. tsinlingensis* is characterized by a thin pericarp, thin-winged

Characters

- 5. Stem presence
 - 10. Form of blade division
 - 18. Sheath shape
 - 20. Petiole internal structure
 - 26. Bract shape
 - 29. Flower color
 - 36. Fruit shape
 - 39. Fruit rib development
 - 40. Dorsal ribs form
 - 41. Lateral rib width
 - 44. Degree of endosperm dorsal compression
 - 46. Mesocarp thickness
 - 48. Arrangement of vittae
 - 52. Size of vascular bundle
 - 53. Separation of mature mericarps
- State: 0, □; 1, ▨; 2, ■

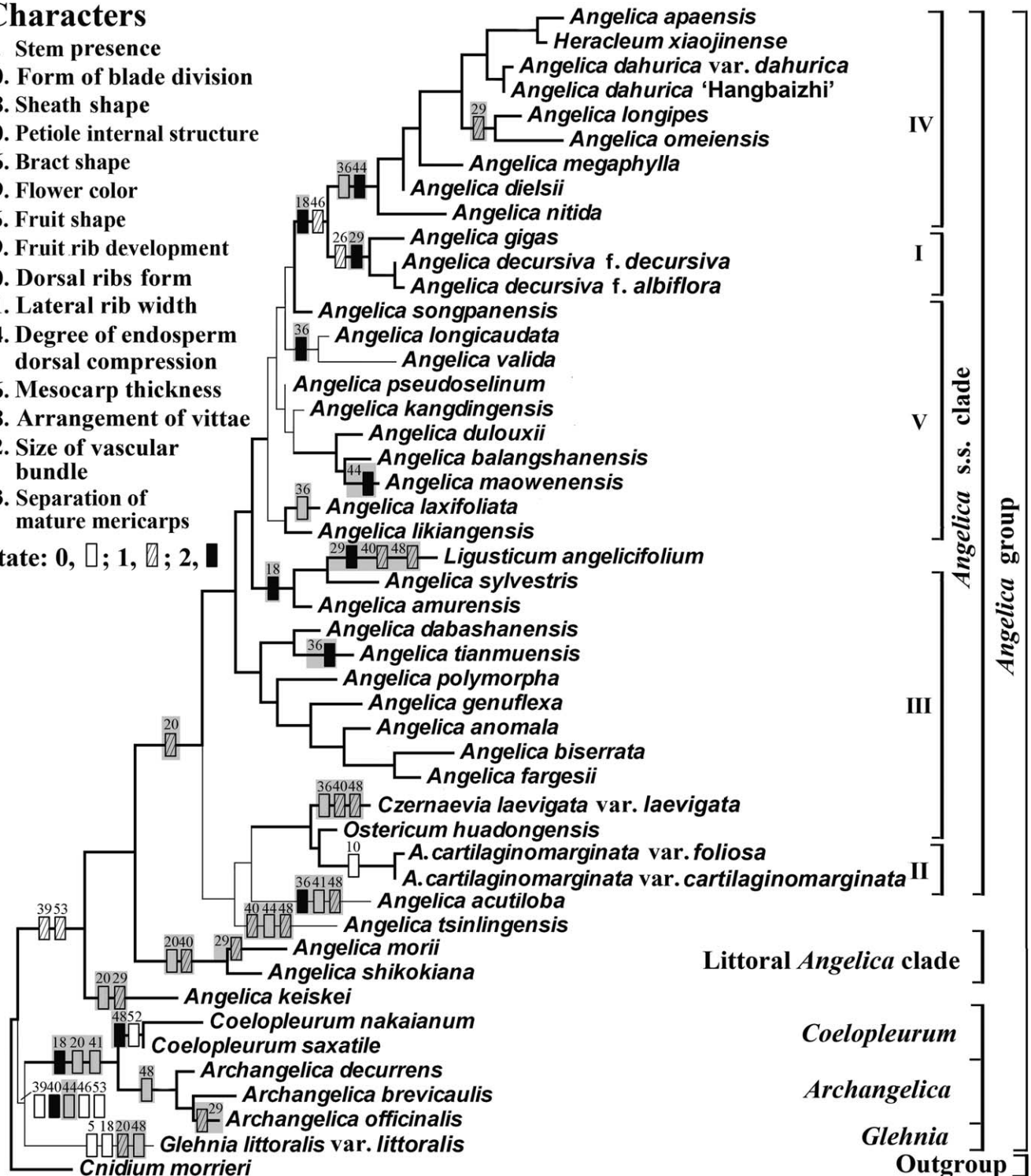


FIG. 8. One of 1097 minimal length, 367-step trees derived from maximum parsimony analysis of 53 morphological characters (CI = 0.25, RI = 0.56). Branches supported in the strict consensus tree of this analysis are marked with bold lines. The Roman numerals indicate the same lineages as identified in Fig. 2. The distribution of 15 morphological characters is mapped onto this single tree; homoplastic character states are shadowed.

dorsal ribs, and three vittae in each furrow (Fig. 7C). These characters easily distinguish *A. tsinlingensis* from all other *Angelica* species examined herein.

The fruit epidermal micromorphology of the *Angelica* group is various and complex, especially in its surface roughness, ornamentation type, and epidermal secretion, but these characters are not useful in distinguishing taxa within the *Angelica* group. However, the presence of epidermal cells that are invisible or flat/concave with vague outlines (Figs. 5 and 6) is useful to distinguish members of the *Angelica* group from those of the *Ostericum* clade.

Phylogenetic Analysis of Morphological Data—MP analysis of the morphological dataset retrieved 1097 minimal length trees, each of 367 steps (CI = 0.25, RI = 0.56). One of these trees shows the distribution of several characters and character states, with branches supported in the strict consensus tree indicated by bold lines (Fig. 8). Most characters commonly emphasized in traditional classification systems of *Angelica* were homoplastic, such as sheath shape (character 18, state 2; Table 1), flower color (character 29, states 1 and 2), fruit shape (character 36, states 0 and 2), form of dorsal ribs (character 40, state 1), lateral rib width (character 41, state 0), and degree of endosperm dorsal compression (character 44, states 0 and 2). Endosperm dorsal compression, lateral rib width, and features of the secretory system correspond approximately to those characters traditionally considered important in distinguishing among genera of *Angelica* s.l., specifically the degree of fruit compression, width of wings, and vittae number, respectively. Some morphological characters, however, were especially useful in demarcating major clades despite their homoplasy, such as those of petiole internal structure (character 20), features of the ribs (characters 39–41), and arrangement of vittae in the fruit (character 48). The *Archangelica* and *Coelopleurum* clades were each moderately to well supported (BS = 65 and 77, respectively; support values not shown in Fig. 8), and along with *Glehnia* this entire group can be distinguished from other taxa by the shared presence of more or less equally-developed (character 39, state 0) and corky-winged ribs (character 40, state 2), a slightly compressed endosperm (character 44, state 0), a corky pericarp (character 46, state 0), and adhering mericarps (character 53, state 0). The Littoral *Angelica* clade (BS = 45) is characterized by members sharing a hollow petiole (character 20, state 0) and narrow-winged dorsal ribs (character 40, state 1).

The *Angelica* s.s. clade, however, exhibited extensive variation in morphology, with only the previously designated Clades I, II, and IV retrieved as monophyletic. Clade I (BS = 84) is characterized by plants possessing persistent, saccate bracts (character 26, state 1) and dark-purple flowers (character 29, state 2). Clade II (BS = 96) is characterized by plants possessing a unique, once-pinnate blade (character 10, state 0) with 4–9 pairs of pinnae. Clade IV (BS < 50) is distinguished by plants bearing subround mericarps (character 36, state 0) with heavily-compressed endosperm (character 44, state 2).

DISCUSSION

Discordance Between ITS and cpDNA Phylogenies—The topologies of the nrDNA and cpDNA trees differed significantly, with this discordance centered around *A. keiskei*, *A. longipes*, *A. nitida*, *A. polymorpha*, *A. sylvestris*, and *Glehnia littoralis* (Fig. 2). Because the tree topology inferred from

cpDNA exhibited some geographic structure, we favor chloroplast capture as the most probable explanation for this discordance. Although incomplete lineage sorting could also cause such a conflict, the coalescence of organelle DNA is much faster than that of nuclear genes (Moore 1995). Therefore, it is unlikely that lineage sorting for nuclear genes had been completed before the divergence of chloroplast genes in *Angelica* s.s. and its closest relatives. It is not uncommon for chloroplast transfer to happen between genera; for instance, Yuan and Olmstead (2008) suggested two independent, sympatric, chloroplast transfer events for both *Verbena* L. and *Glandularia* J. F. Gmel. (Verbenaceae) occurring in North and South America, respectively. Furthermore, the ancestor of *A. sylvestris* was estimated to be isolated from other East Asian species of the *Angelica* s.s. clade for more than eight million years since a dispersal westward from East Asia to Central Asia and Europe during the middle Late Miocene (Liao et al. 2012a). Therefore, vicariance is probably responsible for its greater genetic distance from its East Asian relatives. The conflicts between nuclear and plastid DNA datasets revealed that *A. nitida*, a rare tetraploid of *Angelica* ($4n = 44$; Pan et al. 1991; Zhang et al. 2005), arose through hybridization with *A. apaensis* as one of the diploid parents because of their similar fruit characters and close position in the nrDNA tree. *Angelica longipes* is a rather poorly known species (Sheh et al. 2005) and requires further study to explain its discordance in the cladograms.

Phylogenetics of *Angelica sensu lato*—Results of the large scale ITS study (Fig. 1) show that *Angelica* s.l. is not monophyletic, a finding in accordance with several other molecular systematic studies with more limited sampling (e.g. Kondo et al. 1996, Downie et al. 1998, Xue et al. 2007, Feng et al. 2009). The majority of these species occur in what we have coined the *Angelica* group in tribe Selineae. Several other species of *Angelica* s.l., however, fell outside of Selineae, and in this study, six *Ostericum* members allied as a sister group to tribe Scandiceae in the *Ostericum* clade. Downie et al. (2010) treated the *Ostericum* clade in a broader *Acronema* clade, alongside other taxa such as *Pterygopleurum* Kitag. and *Ligusticum* L. The distant relationship between *Ostericum* and other *Angelica* s.l. members is congruent with immunological (Shneyer et al. 2003), chemical (Harborne et al. 1986), and pollen ultrastructural (Sheh et al. 1997; Shu and Sheh 2004) evidence. The most obvious feature of *Ostericum* is its unique mericarp structure. Its exocarp consists of convex, rhombic-rectangular cells with thickened outer walls, and its mesocarp cells and vascular bundles are reduced at fruit maturity (Fig. 4A–C). Clearly, members of the *Ostericum* clade are distinguishable from those of the *Angelica* group on the basis of both molecular and morphological evidence. One exception, however, is *O. huadongensis* (Fig. 4D). In trees derived from DNA sequences and morphology (Figs. 1–3, and 8), *O. huadongensis* occurs within the *Angelica* s.s. clade. This placement is supported by fruit morphology (non-reduced mesocarp and vascular bundles) and concave epidermal cells (Fig. 4D), features also found in *Angelica* s.s. but not members of the *Ostericum* clade. This suggests that *O. huadongensis* is better treated in *Angelica* s.s. than in *Ostericum*.

Six major clades were identified within the *Angelica* group. The vegetative organs of *Archangelica* and *Coelopleurum* are considered “*Angelica*-like” (Vasilèva and Pimenov 1991; Sheh et al. 2005), but their anatomical characters are remarkably different from those species of the *Angelica* s.s. clade.

For instance, the petioles of *Archangelica brevicaulis*, *A. decurrens*, and *Coelopleurum saxatile* are hollow-tubular, whereas solid petioles characterize all members of *Angelica* s.s. (Liao et al. 2012b). The mericarps of *Archangelica* and *Coelopleurum* are similar in appearance, as are their mesocarps and ribs. However, the vittae of *Archangelica* are quite small, almost encircling and adhering to the seed (Fig. 5A, structure 3), and these serve to distinguish *Archangelica* from all other members of *Angelica* s.l. According to Qin et al. (1995), the North American species *Angelica ampla* shares similar fruit features with that of *Archangelica*. In the ITS trees, *A. ampla* allies with three species of *Archangelica* and away from the other members of the North American *Angelica* clade (Fig. 1). The North American species *Angelica atropurpurea* L. and *A. laurentiana* Fernald, while not included in our study, also share these distinguishable features with *Archangelica* (Qin et al. 1995), which suggests they should also be transferred to *Archangelica* pending confirmation from further study. The mericarps of *Coelopleurum* and *Archangelica* differ in their secretory system (*Coelopleurum* has only one or two enlarged vittae in each furrow, Fig. 5B, structure 3) and vascular bundles (*Coelopleurum* has reduced vascular bundles, Fig. 5B, structure 4*). Moreover, *Coelopleurum gmelinii* (DC.) Ledeb., *C. lucidum*, and *C. saxatile* have a karyotype of $2n = 28$, which is unique in the Selineae (Vasiléva and Pimenov 1991; Pan et al. 1994; Shneyer et al. 2003). These multiple lines of evidence suggest that *Archangelica* and *Coelopleurum* should be maintained as independent genera, distinct from *Angelica* s.s.

The genus *Glehnia*, comprising two closely related taxa occurring on eastern Asian and western North American beaches, is placed in the subtribe Angelicinae (Shan 1992). Both taxa are perennial, with white-pubescent throughout, and have conspicuously shortened stems. The mericarps of *Glehnia* are among the largest in Apiaceae, approaching 1.2×1.0 cm in size with wide-winged ribs. They have a corky pericarp and small, numerous vittae (Fig. 5C, structures 2 and 3, respectively) similar to those found in *Archangelica* and *Coelopleurum*. Considering that these three genera also share pollen that is super-rectangular in shape (Sheh et al. 1997), *Glehnia* appears more closely related to *Archangelica* and *Coelopleurum* than to members of the *Angelica* s.s. clade. *Glehnia*, therefore, should also be maintained as a distinct genus.

The Littoral *Angelica* clade is represented by *A. morii* and several island species, such as *A. shikokiana*. While this clade occurs outside of *Angelica* s.s., its members are quite *Angelica*-like in appearance. However, they do differ from those of the *Angelica* s.s. clade in several anatomical features. As examples, *A. shikokiana* has a hollow, tubular petiole, and *A. morii* also has a hollow petiole, but with a vascular partition (Liao et al. 2012b). Their fruits possess highly developed vascular bundles (Fig. 5D, structure 4), which are not usually as developed in members of the *Angelica* s.s. clade. In *A. morii*, the dorsal ribs are narrow-winged and the endosperm is fan-shaped with four grooves on the dorsal face, all features that allow this species to be distinguished readily.

The *Angelica* s.s. clade is characterized by many features that distinguish it from other members of the *Angelica* group: the petioles are solid (Liao et al. 2012b), the mesocarp is not corky and mostly adheres to the seed, the ribs are unequally developed (with the dorsal ribs low or rounded and the lateral ribs broad-winged and wider than half of the body), the endosperm is conspicuously dorsally compressed, the vittae

are generally fewer but enlarged, the vascular bundles are non-reduced, and the mericarps are easily separated from one another at maturity (Fig. 6).

In the analysis of combined nrDNA and cpDNA data (Fig. 3), Clade I is an early diverging branch of the *Angelica* s.s. clade and is characterized by members possessing dark-purple flowers and persistent, saccate bracts. Clade II is characterized by plants possessing a unique, once-pinnate leaf blade with 4–9 pairs of pinnae, yet its fruits have low dorsal ribs and its mesocarp consists of 1–3 layers of cells (Fig. 6B), features similar to those found in members of Clade V. Most members of Clade IV are restricted to the eastern Tibetan plateau and adjacent regions, except for *A. dahurica*, which is widely cultivated for medicine (Kao 1993; Sheh et al. 2005). In Clade IV, diagnostic morphological characters include saccate-inflated sheaths, subround mericarps with a thickened mesocarp and a highly compressed endosperm (W/H, Width/Height, > 2), low, rounded dorsal ribs, and broad wings that are much wider than the body (Fig. 6E and F). Pimenov and Kljuykov (2003) suggested that *Heracleum xiaojinense* was conspecific with *A. apaensis* on the basis of fruit anatomy, a relationship supported by the molecular evidence presented herein. Clade V is sister to Clade IV and comprises nine species endemic to the Hengduan Mountains of southwestern China. These species are characterized by thin, leathery leaves, long-tubular sheaths, a thin mesocarp comprising 1–3 layers of cells, moderately compressed endosperm (W/H = 1.5–2), and narrow-winged lateral ribs mostly as wide as the body (Fig. 6G and H). Clade V was not retrieved as monophyletic in the morphological phylogeny, however this lineage possesses low sequence divergence values (nrDNA: 0–2.47%; cpDNA: 0.71–3.42%) such that some species showing morphological differences had identical ITS sequences (i.e. *A. kangdingensis*, *A. laxifoliata*, *A. longicaudata*, *A. pseudoselinum*, and *A. songpanensis*). This phenomenon may be attributed to the recent and rapid radiation of the group, and incomplete lineage sorting. The initial split of Clade V was estimated as occurring less than 4.0 Myr (Liao et al. 2012a), which seems too recent to accumulate enough mutations to complete lineage sorting for both nuclear and plastid genes.

The remainder of the species of the *Angelica* s.s. clade formed a weakly supported Clade III (Fig. 3). Included in this group is *Czernaevia laevigata*, a species recognized in the Flora of China because of its distinct petal morphology and absence of coumarin and flavonoid compounds (Sheh et al. 2005). Despite the triple vittae in each of its furrows, *Czernaevia laevigata* is indeed *Angelica*-like, both in its morphology and habit. Vasiléva and Pimenov (1991) suggested that *Czernaevia* should be placed into *Angelica* based on karyotype characters, and the results of this study support the transfer proposed previously by Vasiléva and Pimenov (1991) and Pimenov et al. (2003).

Several species once recognized in *Angelica* are better excluded from the *Angelica* s.s. clade. The transfer of *A. onosepala* to tribe Tordylieae is supported by both morphological and DNA evidence (Pimenov and Kljuykov 2003; Feng et al. 2009). *Angelica paeoniifolia* and *A. sinensis* ally with *Contioselinum* spp., *Levisticum officinale*, and *Vicatia tibetica* in the *Sinodielsia* clade, a relationship also in accordance with chemical data (Xue et al. 2007). The morphology and anatomy of *A. sinensis* are also quite different from members of

the *Angelica* s.s. clade; as examples, the petioles are hollow-tubular (Liao et al. 2012b), the exocarp is interrupted near the carpophore, the mesocarp is relatively thick, the vascular bundles are reduced and adhere to the inside of the mesocarp, and the vittae are hardly visible on the commissural face (Fig. 7A). Such features also occur in *A. ternata* (Fig. 7B) and other Himalayan *Angelica* species not included in this study, such as *A. glauca* Edgew., *A. indica* Pimenov & Kljuykov, *A. multicaulis* Pimenov, and *A. paeoniifolia* (Qin et al. 1995; Pimenov and Kljuykov 2003). Immunological studies of seed proteins revealed that *A. glauca*, *A. multicaulis*, and *A. ternata* showed a close affinity, although they were distant from the type species, *A. sylvestris* (Shneyer et al. 2003). Pending further study, we believe that *A. sinensis* and its allies form another major lineage of *Angelica* s.l. located in the *Sinodielsia* clade. These species are all endemic to the Himalayas, with only *A. sinensis* distributed in the eastern Himalayas, and *A. sinensis* distributed in the eastern Himalayas, with only *A. sinensis* distributed in the eastern Himalayas, and *A. sinensis* distributed in the eastern Himalayas. Further study of these Himalayan plants may eventually reveal a new genus of Apiaceae.

Angelica tsinlingensis used to be considered conspecific with *Notopterygium forbesii* (Pimenov and Kljuykov 2003) because of their winged ribs and triple vittae in the furrows (Fig. 7C). However, *A. tsinlingensis* is indeed distinguishable from *Notopterygium forbesii* with leaves that are 2-ternate (with nine leaflets total), flowers white with slightly enlarged outer petals, mature mericarps gray and about 6 × 4 mm in size, and the commissure face of the endosperm flat. In contrast, *N. forbesii* has 3–4-pinnate leaves, yellow flowers, and 4 × 4 mm, brownish-yellow fruits with a concave endosperm commissure. Inferred from the ITS results (Fig. 1), *A. tsinlingensis* is distantly related to *N. forbesii* and sister to *Melanosciadium pimpinelloideum* (which is similar to *A. tsinlingensis* in the vegetative stage, but significantly differs based on inflorescence and fruit morphologies). On the other hand, *A. tsinlingensis* is clearly different from members of the *Angelica* s.s. clade with its thin-winged dorsal ribs and triple vittae in each furrow; this discordance corresponds with the molecular results in placing *A. tsinlingensis* outside of the *Angelica* s.s. clade.

The taxonomic position of *A. acutiloba* has been disputed for many years because of its unusual fruit characteristics. All of its ribs are reduced, yellowish dots occur on the surfaces of its ribs, vallecular canals, and commissure, numerous yellow crystals occur in its exocarp and mesocarp cells, and triple vittae are distributed in each furrow (Qin et al. 1995; Chu and Liu 2007). *Angelica acutiloba* is isolated from the *Angelica* s.s. clade and occupies an early diverging branch of the *Angelica* group (Figs. 1–3). Its unusual morphology doesn't reveal any close relationship and its placement as an isolated lineage in the phylogenetic trees suggests that recognition of a new, monotypic genus might be in order. Based on fruit morphology, Chu and Liu (2007) suggested that *A. acutiloba* might represent a new genus and the results presented herein support that conclusion.

Angelica hirsutiflora, an endemic species of Taiwan, appears more similar morphologically to *Peucedanum* than it does to *Angelica*. Similar features include a woody stem base, leathery leaflets with entire, wavy margins, strong dorsally compressed fruits with low, reduced dorsal ribs, broad and lignified lateral wings, and a commissure with

7–8 vittae (Shan 1992; Kao 1993). The strong morphological similarity between these taxa is also reflected in the molecular results, as *A. hirsutiflora* occurs within the *Peucedanum* s.s. clade (Fig. 1).

According to Pimenov (1968a) and Vasiléva and Pimenov (1991), *Angelica* s.l. can be divided into three subgenera: *Angelica*, *Archangelica*, and *Ostericum*. Their subgenus *Archangelica* comprised sections *Archangelica* and *Coelopleurum*. Subgenus *Angelica* included sections *Angelica*, *Czernaevia*, *Porphyroscias*, and *Callisace*, the latter comprising four subsections. Results from the molecular phylogenetic analyses presented herein partly support these treatments, in the sense that *Angelica*, *Archangelica*, *Coelopleurum*, and *Ostericum* each formed monophyletic branches, *Czernaevia laevigata* was submerged in the *Angelica* s.s. clade, and Clade I of the *Angelica* s.s. clade corresponds to section *Porphyroscias*. However, based on an expanded sampling compared to previous phylogenetic studies of *Angelica* s.l., we suggest major rearrangements to these traditional treatments. Clade III of *Angelica* s.s. integrates *Angelica* sections *Angelica* and *Czernaevia* and three subsections of section *Callisace* (subsections *Anisopleura*, *Stenophyllum*, and *Angelophyllum*). Clade II and the two eastern Himalayan lineages (clades IV and V) are recognized herein as distinct groups for the first time (one member of clade IV, *A. dahurica*, was treated as subsection *Callisace* by Russian authors). Still, further research on *Angelica* is needed, especially for those species from Japan and Russia. The results presented herein provide new insights into the phylogeny and classification of East Asian *Angelica* s.l., a group whose relationships have heretofore been unclear. A bevy of taxonomic realignments and possible new genera are suggested from this study (including those that involve a number of North American species currently treated in *Angelica*), which will be presented in subsequent papers.

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- APPENDIX 1. List of 120 accessions examined for molecular and fruit anatomical and micromorphological studies. GenBank accession numbers are provided for six loci: ITS / ETS / *rps16* intron / *rps16-trnK* / *rpl32-trnL* / *trnL-trnT*. A dash (—) indicates that the locus was not sequenced for that taxon and an asterisk (*) indicates that the accession was newly considered in this study. Specimen voucher information is provided (locality, collector and collector number, herbarium) unless the sequence data were published by others and obtained by us from GenBank; then the place of original publication is provided. Specimen voucher information for those accessions used in the related study of the historical biogeography of the East Asian *Angelica* group (Liao et al. 2012a), but also considered herein, is also provided. Specimens examined for the fruit anatomical and SEM studies are indicated by daggers (†). Herbarium abbreviations follow Holmgren et al. (1990), and author abbreviations follow Brummitt and Powell (1992).
- Aegopodium podagraria* L., U30536 & U30537 /—/—/—/—/—, Downie & Katz-Downie (1996); *Angelica acutiloba* (Siebold & Zucc.) Kitag. f., GU395147 / HM443666 / GU395095 / JF279349 / JF279395 / JF279303, China, Sichuan, Chengdu, cultivated, Liao CY 666779 (SZ); *A. ampla* A. Nelson, U79597 & U79598 /—/—/—/—/—, Sun et al. (2004); *A. amurensis* Schischk. f., GU395148 / HM443667 / GU395096 / JF279350 / JF279396 / JF279304, China, Jilin, Changbai Mountain, Liao CY 746005 (SZ); *A. anomala* Avé-Lall. f., GU395149 / HM443668 / GU395097 / JF279351 / JF279397 / JF279305, China, Jilin, Changbai Mountain, Liao CY 744106 (SZ); *A. apaensis* R.H. Shan & C.Q. Yuan (1)†, GU395150 / HM443669 / GU395098 / JF279352 / JF279398 / JF279306, China, Sichuan, Balang Mountain, Liao CY 727237 (SZ); *A. apaensis* R.H. Shan & C.Q. Yuan (2), JN107553 / JN107556 / JN107547 / JN107544 / JN107550 / JN107541, China, Sichuan, Mengbi Mountain, Liao CY 666949 (SZ); *A. arguta* Nutt. ex Torr. & A. Gray, U79599 & U79600 /—/—/—/—/—, Sun et al. (2004); *A. baizhioides* (Mss.), DQ263588 /—/—/—/—/—, Xue et al. (2007); *A. balangshanensis* R.H. Shan & F.T. Put, HQ896671 / JF279293 / JF279301 / JF279393 / JF279439 / JF279347, China, Sichuan, Balang Mountain, Liao CY 742598 (SZ); *A. biserrata* (R.H. Shan & C.Q. Yuan) C.Q. Yuan & R.H. Shant, GU395180 / HM443670 / GU395099 / JF279353 / JF279399 / JF279307, China, Anhui, Jinzhai, Liao CY 666796 (SZ); *A. breveri* A. Gray, U78396 & U78456 /—/—/—/—/—, Sun et al. (2004); *A. cartilaginomarginata* var. *foliosa* C.Q. Yuan & R.H. Shant, GU395177 / HM443671 / GU395100 / JF279354 / JF279400 / JF279308, China, Jiangsu, Nanjing, Liao CY 667428 (SZ); *A. cincta* H. Boissieu, AF008601 & AF009080 /—/—/—/—/—, Katz-Downie et al. (1999); *A. dabashanensis* C.Y. Liao & X.J. He†, GU395179 / HM443688 / GU395124 / JF279371 / JF279417 / JF279325, China, Shaanxi, Daba Mountains, Liao CY 666810 (SZ); *A. dahurica* (Hoffm.) Benth. & Hook. f. ex Franch. & Sav.†, GU395152 / HM443673 / GU395102 / JF279356 / JF279402 / JF279310, China, Jilin, Changbai Mountain, Liao CY 703009 (SZ); *A. dahurica* ‘Hangbaizhi’, GU395151 / HM443672 / GU395101 / JF279355 / JF279401 / JF279309, China, Sichuan, Chengdu, cultivated, Liao CY 673892 (SZ); *A. decursiva* (Miq.) Franch. & Sav.†, GU395153 / HM443674 / GU395103 / JF279357 / JF279403 / JF279311, China, Sichuan, Chengdu, cultivated, Liao CY 673892 (SZ); *A. decursiva* (Miq.) Franch. & Sav. f. *albiflora* (Maxim.) Nakai, HQ256684 /—/—/—/—/—, Japan, Nikko, 0561599 (KUN); *A. dielsii* H. Boissieu†, GU395154 / HM443675 / GU395130 / JF279358 / JF279404 / JF279312, China, Shaanxi, Daba Mountains, Liao CY 666961 (SZ); *A. duclouxii* Fedde ex H. Wolff†, GU395155 / HM443676 / GU395129 / JF279359 / JF279405 / JF279313, China, Yunnan, Dongchuan, Liao CY 674189 (SZ); *A. fargesii* H. Boissieu†, GU395181 / HM443677 / GU395128 / JF279360 / F279406 / JF279314, China, Shaanxi, Daba Mountains, Liao CY 673574 (SZ); *A. furcijuga* Kitag., DQ278164 /—/—/—/—/—, Xue et al. (2007); *A. gemflexa* Nutt. ex Torr. & A. Gray, DQ263566 /—/—/—/—/—, Xue et al. (2007); *A. gigas* Nakai†, GU395156 / HM443678 / GU395104 / JF279361 / JF279407 / JF279315, China, Jilin, Changbai Mountain, Liao CY 744110 (SZ); *A. grayi* J.M. Coult. & Rose, AY146825 & AY146891 /—/—/—/—/—, Sun et al. (2004); *A. hirsutiflora* S.L. Liu, C.Y. Chao & T.I. Chuang, HQ256683 /—/—/—/—/—, China, Taiwan, Yang TY 0091455 (KUN); *A. japonica* A. Gray, AY548214 /—/—/—/—/—, Choi et al., unpublished; *A. kangdingensis* R.H. Shan & F.T. Pu, GU395157 / HM443679 / GU395131 / JF279362 / JF279408 / JF279316, China, Sichuan, Kangding, Liao CY 666988 (SZ); *A. keiskei* Koidz.†, GU395158 / HM443680 / GU395123 / JF279363 / JF279409 / JF279317, China, Yunnan, Kunming, cultivated, Liao CY 673598 (SZ); *A. laxifoliata* Diels (1)†, GU395159 / HM443681 / GU395105 / JF279364 / JF279410 / JF279318, China, Sichuan, Lixian, Liao CY 727232 (SZ); *A. laxifoliata* Diels (2), JN107554 / JN107557 / JN107548 / JN107545 / JN107551 / JN107542, China, Shaanxi, Taibai Mountain, Liao CY 673689 (SZ); *A. lignescens* Reduron & Danton, AY179030 /—/—/—/—/—, Spalik et al. (2004); *A. likiangensis* H. Wolff†, HQ267716 / HQ267717 / JF279300 / JF279392 / JF279437 / JF279346, China, Yunnan, Lijiang, Wang ZX 802456 (SZ); *A. longicaudata* C.Q. Yuan & R.H. Shant, GU395160 / HM443682 / GU395122 / JF279365 / JF279411 / JF279319, China, Sichuan, Emei Mountain, Liao CY 673421 (SZ); *A. longipes* H. Wolff, HQ256679 / HQ256694 / JF279299 / JF279391 / JF279438 / JF279345, China, Tibet, Nielamu, Yu Y 802447 (SZ); *A. maowenensis* C.Q. Yuan & R.H. Shant, GU395161 / HM443683 / GU395106 / JF279366 / JF279412 / JF279320, China, Sichuan, Balang Mountain, Liao CY 667004 (SZ); *A. megaphylla* Diels†, GU395162 / HM443684 / GU395107 / JF279367 / JF279413 / JF279321, China, Chongqing, Jinpo Mountain, Liao CY 706596 (SZ); *A. morii* Hayata†, GU395182 / HM443685 / GU395108 / JF279368 / JF279414 / JF279322, China, Jiangxi, Yingtan, Liao CY 667421 (SZ); *A. nitida* H. Wolff (1)†, GU395163 / HM443686 / GU395109 / JF279369 / JF279415 / JF279323, China, Sichuan, Hongyuan, Liao CY 667003 (SZ); *A. nitida* H. Wolff (2), JN107555 / JN107558 / JN107549 / JN107546 / JN107552 / JN107543, China, Qinghai, Ledu, Feng T 666949 (SZ); *A. omeiensis* C.Q. Yuan & R.H. Shant, GU395164 / HM443687 / GU395110 / JF279370 / JF279416 / JF279324, China, Sichuan, Emei Mountain, Liao CY 673680 (SZ); *A. onosepala* Hand.-Mazz., EU418382 /—/—/—/—/—, Feng et al. (2009); *A. paeoniifolia* R.H. Shan & C.Q. Yuan, HQ256678 /—/—/—/—/—, China, Tibet, Biru, Tao FD 0562119 (KUN); *A. pinnata* S. Watson, AF358465 & AF358532 /—/—/—/—/—, Sun et al. (2004); *A. polymorpha* Maxim. (1)†, GU395165 / HM443689 / GU395125 / JF279372 / JF279418 / JF279326, China, Heilongjiang, Harbin, Liao CY 744122 (SZ); *A. polymorpha* Maxim. (2), HQ256680 / HQ256687 / JF279294 / JF279386 / JF279432 / JF279340, Japan, Kgeji, Yusuhara Town, Taku 0867045 (KUN); *A. pseudoselinum* H. Boissieu†, GU395166 / HM443690 / GU395111 / JF279373 / JF279419 / JF279327, China, Sichuan, Zhegu Mountain, Liao CY 666969 (SZ); *A. pubescens* Maxim., DQ263567 /—/—/—/—/—, Xue et al. (2007); *A. purpureifolia* (nom. illeg.), AY548229 /—/—/—/—/—, Choi et al.,

- unpublished; *A. roseana* L.F. Hend., AF358466 & AF358533 /—/—/—/—/—, Sun et al. (2004); *A. sachalinensis* Maxim., AF007873 /—/—/—/—/—, Xue et al. (2007); *A. shikokiana* Makino ex Y. Yabe, HQ256682 / HQ256689 / JF279296 / JF279388 / JF279434 / JF279342, Japan, Shimozui, Taishou Town, Taku 0769490 (KUN); *A. sinensis* (Oliv.) Diels†, GU395144 /—/—/—/—/—, China, Gansu, Minxian, Yu Y 667002 (SZ); *A. songpanensis* R.H. Shan & F.T. Pu, GU395167 / HM443691 / GU395112 / JF279374 / JF279420 / JF279328, China, Sichuan, Songpan, Ma XG 714746 (SZ); *A. sylvestris* L., HQ256681 / HQ256688 / JF279295 / JF279387 / JF279433 / JF279341, Germany, Bavarian Swabia, German-Chinese Exped. 0562104 (KUN); *A. ternata* Regel & Schmalh.t, —/—/—/—/—, China, Xinjiang, Wuqia, Kerram 00022959 (XJBI); *A. tianmuensis* Z.H. Pan & T.D. Zhuang†, GU395178 / HM443693 / GU395113 / JF279375 / JF279421 / JF279329, China, Zhejiang, Tianmu Mountain, Liao CY 667442 (SZ); *A. tsinlingensis* K.T. Fut, GU395168 / HM443694 / GU395126 / JF279376 / JF279422 / JF279330, China, Shaanxi, Hua Mountain, Liao CY 714752 (SZ); *A. ursina* Regel, DQ263565 /—/—/—/—/—, Xue et al. (2007); *A. valida* Diels†, GU395169 / HM443695 / GU395114 / JF279377 / JF279423 / JF279331, China, Chongqing, Jinpo Mountain, Liao CY 727244 (SZ); *Anthriscus sylvestris* (L.) Hoffm.*, EU236159 /—/—/—/—/—, Zhou et al. (2008); *Archangelica brevicaulis* Rchb.f.t, GU395170 / HM443696 / GU395115 / JF279378 / JF279424 / JF279332, China, Xinjiang, Houxia, Wang CB 727248 (SZ); *Archangelica decurrens* Ledeb.t, GU395171 / HM443697 / GU395116 / JF279379 / JF279425 / JF279333, China, Xinjiang, Kanas, Wang CB 727249 (SZ); *Archangelica officinalis* Hoffm., U30576 & U30577 /—/—/—/—/—, Downie & Katz-Downie (1996); *Arcuatopterus thalictroides* M.L. Sheh & R.H. Shan, EU236160 /—/—/—/—/—, Zhou et al. (2008); *Arracacia brandegei* J.M. Coult. & Rose, U30570 & U30571 /—/—/—/—/—, Downie & Katz-Downie (1996); *Cnidium monnieri* (L.) Spreng., HQ316168 / HQ256692 / JF279297 / JF279389 / JF279435 / JF279343, China, Sichuan, Chengdu, Liao CY 802525 (SZ); *Coelopleurum lucidum* Fernald, DQ270196 /—/—/—/—/—, Xue et al. (2007); *Coelopleurum saxatile* Drud†, GU395172 / HM443698 / GU395117 / JF279381 / JF279334, China, Jilin, Changbai Mountain, Liao CY 706634 (SZ); *Conioselinum chinense* Britton, Sterns & Poggenb., GU395145 /—/—/—/—/—, China, Anhui, Jinzhai, Liao CY 666797 (SZ); *Conioselinum scopulorum* J.M. Coult. & Rose*, AF008634 & AF009113 /—/—/—/—/—, Katz-Downie et al. (1999); *Conioselinum tataricum* Hoffm., AF008623 & AF009102 /—/—/—/—/—, Katz-Downie et al. (1999); *Conioselinum vaginatum* (Spreng.) Thell., FJ385041 /—/—/—/—/—, Zhou et al. (2009); *Czernaevia laevigata* Turcz.t, GU395173 / HM443699 / GU395118 / JF279381 / JF279427 / JF279335, China, Jilin, Changbai Mountain, Liao CY 714738 (SZ); *Daucus carota* L.*, AF077779 /—/—/—/—/—, Lee & Downie (2006); *Ferula kingdon-wardii* H. Wolff *, EU236166 /—/—/—/—/—, Zhou et al. (2008); *Ferula olivacea* (Diels) H. Wolff ex Hand.-Mazz.*, FJ385043 /—/—/—/—/—, Zhou et al. (2009); *Glehnia littoralis* F. Schmidt var. *littoralis*†, GU395183 / HM443700 / GU395119 / JF279382 / JF279428 / JF279336, China, Beijing, cultivated, Liao CY 666775 (SZ); *Glehnia littoralis* F. Schmidt var. *leiocarpa* (Mathias) B. Boivin, AY146850 & AY146916 /—/—/—/—/—, Sun et al. (2004); *Heracleum bivittatum* H. Boissieu, EU236168 /—/—/—/—/—, Zhou et al. (2008); *Heracleum hemsleyanum* Diels, EU001371 /—/—/—/—/—, He et al., unpublished; *Heracleum xiaojinense* F.T. Pu & X.J. He†, FJ812132 / HM443701 / FJ986011 / JF279383 / JF279429 / JF279337, China, Sichuan, Balang Mountain, Yu Y 10012 (SZ); *Holandrea carvifolia* (Vill.) Reduron, Charpin & Pimenov, AF495828 /—/—/—/—/—, Spalik et al. (2004); *Levisticum officinale* W.D.J. Koch, GU395146 /—/—/—/—/—, China, Heilongjiang, Harbin, cultivated, Liao CY 673560 (SZ); *Ligusticum angelicifolium* Franch.t, GU395174 / HM443702 / GU395127 / JF279384 / JF279430 / JF279338, China, Yunnan, Lijiang, Liao CY 673560 (SZ); *Magadania olaensis* (Gorovoj & N.S. Pavlova) Pimenov & Lavrova, AY328946 & AY330512 /—/—/—/—/—, Valiejo-Roman et al. (2006); *Magadania victoris* (Schischk.) Pimenov & Lavrova, AY328947 & AY330513 /—/—/—/—/—, Valiejo-Roman et al. (2006); *Melanosciadium pimpinelloideum* H. Boissieu*, FJ385049 /—/—/—/—/—, Zhou et al. (2009); *Notopterygium forbesii* H. Boissieu*, GU390407 /—/—/—/—/—, China, Gansu, Lianhua Mountain, Feng T 666939 (SZ); *Oreoselinum nigrum* Delarbre, AF495836 & AF495837 /—/—/—/—/—, Spalik et al. (2004); *Ostericum citriodorum* (Hance) R.H. Shan & C.Q. Yuan*, JX312701 / JX312699 / JX312700 /—/—/—, China, Guangxi, Luzhai, Liao CY 908277 (SZ); *Ostericum grosseserratum* (Maxim.) Kitag.*†, GU390409 / HQ393883 / GU395136 /—/—/—, China, Anhui, Jinzhai, Liao CY 667435 (SZ); *Ostericum huadongensis* Z.H. Pan & X.H. Lit, GU395175 / HM443703 / GU395120 / JF279385 / JF279431 / JF279339, China, Anhui, Jinzhai, Liao CY 667429 (SZ); *Ostericum koreanum* (Maxim.) Kitag., AY534622 /—/—/—/—/—, Choi et al., unpublished; *Ostericum maximowiczii* (F. Schmidt) Kitag.*†, GU390410 / HQ393880 / GU395137 /—/—/—, China, Jilin, Changbai Mountain, Liao CY 744114 (SZ); *Ostericum scaberulum* (Franch.) R.H. Shan & C.Q. Yuan*†, GU390411 / HQ393882 / GU395140 /—/—/—, China, Yunnan, Shangri-la, Liao CY 703013 (SZ); *Ostericum sieboldii* (Miq.) Nakai*†, GU390412 / HQ393881 / GU395141 /—/—/—, China, Jilin, Changbai Mountain, Liao CY 667568 (SZ); *Ostericum viridiflorum* (Turcz.) Kitag.*†, GU390413 / HQ393879 / GU395139 /—/—/—, China, Heilongjiang, Yichun, Wang CB 674185 (SZ); *Pastinaca sativa* L., EU185668 /—/—/—/—/—, Paik & Watson, unpublished; *Peucedanum delavayi* Franch., FJ385054 /—/—/—/—/—, Zhou et al. (2009); *Peucedanum harry-smithii* Fedde ex H. Wolff, GU390408 / HM443665 / GU395121 / JF279348 / JF279394 / JF279302, China, Shaanxi, Daba Mountains, Liao CY 666964 (SZ); *Peucedanum japonicum* Thunb., DQ270201 /—/—/—/—/—, Xue et al. (2007); *Peucedanum medicum* Dunn, HQ256686 / HQ256693 / JF279298 / JF279390 / JF279436 / JF279344, China, Anhui, Yixian, Liao CY 666876 (SZ); *Pimpinella candolleana* Wight & Arn., EU236194 /—/—/—/—/—, Zhou et al. (2008); *Pleurospermum franchetianum* Hemsl.*, EU236198 /—/—/—/—/—, Zhou et al. (2008); *Selinum broteri* Hoffm. & Link*, AY179029 /—/—/—/—/—, Spalik et al. (2004); *Selinum candollei* DC., U30564 & U30565 /—/—/—/—/—, Downie & Katz-Downie (1996); *Selinum carvifolia* (L.) L.*, AY179028 /—/—/—/—/—, Spalik et al. 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