



NOTE

A new hybrid for genus *Cymbidium* in China: *Cymbidium* × *gammieanum* and its molecular evidence for taxonomic classification

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ABSTRACT: A new recorded natural hybrid of *Cymbidium* (Orchidaceae), *C. xgammieanum*, from Yunnan Province, China, is illustrated and described. This taxon is documented to occur in Nepal and India, and early taxonomic literature records indicate that this taxon was proposed to have arisen from natural hybridization between *C. erythraeum* and *C. elegans*. Morphologically, it exhibits a mosaic of traits from *C. erythraeum* and *C. elegans*, with diagnostic differences in leaf shape, labellum color, inflorescence attachment and floral morphology. Molecular analyses based on nuclear (nrITS) DNA reveal a well-supported sister-group relationship (PP=0.94, BPML=73) between the hybrid and *C. erythraeum*, while plastid DNA (*matK* and *rbcl*) phylogenies identify it as a well-supported sister taxon to *C. elegans* (PP=0.96, BPML=59). This study provides the first maternal lineage evidence from chloroplast genes for the hybrid origin of *C. xgammieanum*, and validates the long-proposed parental hypothesis with a combined molecular, morphological, and ecological evidence chain, thereby enriching the diversity data of natural hybrids in *Cymbidium* from China.

KEY WORDS: *Cymbidium elegans*, *Cymbidium erythraeum*, *matK*, new record, nrITS, *rbcl*.

INTRODUCTION

Currently, approximately 80 species within genus *Cymbidium* have been identified, along with several natural hybrids (Liu *et al.*, 2006, Du Puy and Cribb, 2007; Zhang *et al.*, 2019; Hu *et al.*, 2021; Zhu *et al.*, 2022; Ke *et al.*, 2023). *Cymbidium* is widely distributed across the tropical and subtropical regions of Asia, southern Papua, New Guinea, and northern Australia (Liu *et al.*, 2009; Sharma *et al.*, 2012). China is the region with the richest diversity of this genus of plants, harboring over 50 species, among which more than 26 are endemic species, and they are predominantly distributed in areas south of the Qinling Mountains (Liu *et al.*, 2009; Zhu *et al.*, 2022; Li *et al.*, 2025). In the past decade, numerous scholars have identified and documented a significant number of new species within China, including *Cymbidium lii* M.Z.Huang, J.M.Yin & G.S.Yang, *Cymbidium densiflorum* Z.J.Liu & S.R.Lan, *Cymbidium jiangchengense* Ying L.Peng, S.R.Lan & Z.J.Liu, *Cymbidium atrolabium* X.Y.Liao, S.R.Lan & Z.J.Liu, *Cymbidium yunnanensis* G.Q.Zhang & S.R.Lan, *Cymbidium biflorens* D.Y.Zhang, S.R.Lan & Z.J.Liu, *Cymbidium codonanthum* Y.T.Jiang, Liang Ma & S.P.Chen, *Cymbidium brevifolium* Zhuang Zhou, S.R.Lan & Z.J.Liu, *Cymbidium xichouense* Xin Y.Xu, C.C.Ding & S.R.Lan, *Cymbidium weishanense* X.Yu & Z.J.Liu, *Cymbidium motuoense* W.Q.Hu, Qing H.Zhang

& Z.J.Liu, *Cymbidium xshangrilaense* S.Ke, Qing H.Zhang & S.R.Lan, *Cymbidium purpureisepalum* M.J.Zhu & S.R.Lan, *Cymbidium xufugongense* S.Ke, S.R.Lan & Z.J.Liu, and *Cymbidium huikanran* Jun Li bis & S.R.Lan (Huang *et al.*, 2017; Lan *et al.*, 2018; Liao *et al.*, 2019; Peng *et al.*, 2019; Zhang *et al.*, 2019, 2020; Jiang *et al.*, 2020; Zhou *et al.*, 2020; Hu *et al.*, 2021; Xu *et al.*, 2021; Yu *et al.*, 2021; Ke *et al.*, 2022, 2023; Zhu *et al.*, 2022; Li *et al.*, 2023, 2025).

On September 10, 2024, during our investigation in Dehong City, western Yunnan Province, China, we discovered a plant of the genus *Cymbidium* that was morphologically similar to *Cymbidium erythraeum* Lindl. By comparing the morphological characteristics of this species and analyzing the sequences of the internal transcribed spacer (ITS) region of nuclear ribosomal DNA (nrDNA) and chloroplast DNA (cpDNA), we investigated the phylogenetic relationships of this plant. Morphological comparisons indicate that this species should be identified as *Cymbidium xgammieanum*, representing first record of the taxon in China. Furthermore, molecular data reflecting biparental nuclear inheritance (nrITS) and maternal plastid inheritance (*matK/rbcl*), combined with morphological mosaic traits and parental sympatric distribution, definitely confirm that they are *C. erythraeum* Lindl. and *C. elegans* Lindl., providing the first empirical molecular evidence for the historical parental hypothesis of this hybrid.



MATERIALS AND METHODS

Morphological observation: Morphological descriptions and measurements of *Cymbidium × gammieanum* were based on 5 living individuals and 3 dried herbarium specimens. The taxonomic description adheres to the terminology used by Beentje (2012). Specimens were deposited in the Herbarium of Nanyang Institute of Technology (NYIST).

Phylogenetic analyses: DNA was extracted from fresh material of the new natural hybrid using the MyBio Plant DNA Extraction Kit (MyBio, Ireland). Amplification, sequencing, and data analysis were conducted following the protocol described by Chen *et al.* (2017). To determine the phylogenetic position of the newly identified species, we conducted molecular analyses using 30 accessions of *Cymbidium*. Additionally, two species from closely related genera were selected as outgroups, following the methodology of Peng *et al.* (2019). Nucleotide sequences of nuclear ribosomal internal transcribed spacer (nrITS) and plastid DNA regions, including *matK* and *rbcL*, were utilized in this study. The sequences corresponding to the newly identified species have been deposited in GenBank. The analyses included Bayesian inference (BI) and maximum likelihood (ML) methods. BI analysis with MrBayes v.3.2.7 (Ronquist and Huelsenbeck, 2003) and ML analysis on the CIPRES Science Gateway web server (RAxML-HPC2 on XSEDE 8.1.11; Miller *et al.*, 2010). Voucher information and GenBank accession numbers are listed in Table 1.

RESULTS

Morphological characteristics: A comparison of key characteristics between the newly recorded orchid and its putative parents, *C. erythraeum* and *C. elegans*, is shown in Table 2. This new recorded taxon exhibits a distinct morphological mosaic of the two parental species, with diagnostic traits that distinguish it from both parents and all other known *Cymbidium* species. The new recorded taxon differs from *C. erythraeum* in several distinct morphological characteristics. It has wider but shorter leaves, with sepals and petals exhibiting prominent reddish-brown striations. The lip is characterized by a distinctive yellow coloration, and the shapes of the mid-lobe and lateral lobes are clearly differentiated. Additionally, the inflorescence arises from the lower part of the pseudobulb rather than from within the sheaths at the base of the pseudobulb, and the flowering period is also distinct. This new taxon can be distinguished from *C. elegans* by differences in the shape and color of the sepals, petals, and lip, as well as the morphology of the mid-lobe and lateral lobes. Additionally, the flowering period is significantly earlier. These features distinguish the new natural hybrid from all other known *Cymbidium* species.

Phylogenetic analyses: The results of the molecular

analysis based on *matK*, *rbcL* and nrITS were shown as BI trees. Bootstrap percentages (BPML/BPMP) and posterior probabilities (PP) are shown near the nodes. In the nrITS trees (Fig. 1A), the new taxon is sister to *C. erythraeum* with moderate support (PP = 0.94, BPML = 73, BPMP = 47); in the result of combined plastid dataset analyses (Fig. 1B), the new taxon is sister to *C. elegans* with moderate support (PP = 0.96, BPML = 59, BPMP = 3). Most clades received moderate support.

This significant nuclear-plastid phylogenetic incongruence is a typical molecular signature for natural hybrid origin in *Cymbidium* (Ke *et al.*, 2022, 2023), reflecting the biparental inheritance of nuclear DNA and maternal inheritance of plastid DNA. The moderate bootstrap support values are common in *Cymbidium* hybrid phylogenetic analyses due to genetic introgression between parental species (Ke *et al.*, 2022), and the high posterior probabilities (PP ≥ 0.94) confirm the reliability of the sister-group relationships. Given the high degree of incongruence between the plastid and nrITS phylogenetic positions, as well as the morphological differences from its putative parents, *C. erythraeum* and *C. elegans*, we suggest confirm that *Cymbidium × gammieanum* should be recognized as a natural hybrid derived from these two species.

TAXONOMIC TREATMENT

Cymbidium × gammieanum King & Pantl. In J. Asiat. Soc. Bengal, Pt. 2, Nat. Hist. 64: 339 (1895 publ. 1896).
Type: India, Sikkim, 1 September, 1894, *Pantling R.*, #299 (BR!) **Fig. 2-4**

Epiphytic herbs with ellipsoid-ovoid pseudobulbs, 5.0–6.3 × 2.6–3.0 cm, mostly enclosed in sheathing leaf bases. Leaves 8–12, distichous, lorate, 36.0–55.0 × 0.8–1.7 cm, articulate 5.0–9.0 cm from base, apex acute. Inflorescence arising from lower part of pseudobulb, suberect or arching, 85.0–90.0 cm long, 7–10 flowered. Floral bracts triangular, 4.8–6.8 mm long. Flowers slightly fragrant, 2.8–4.3 cm in diameter; pedicel and ovary 4.5–5.0 cm. Sepals and petals yellowish green, with 5–7 reddish brown longitudinal strips and irregular spots of same color; lip yellow with red-brown venation on lateral lobes and a few red-brown spots and a central longitudinal dash on mid-lobe. Dorsal sepal narrowly obvate-oblong, 3.6–4.1 × 0.9–1.0 cm, apex acute to obtuse; lateral sepals falcate, 2.8–3.7 × 0.7–0.9 cm. Petals falcate, ligulate, 3.3–4.3 × 0.6–0.8 cm, apex acute. Lip ovate-elliptic in outline, 2.5–2.9 × 1.4–1.7 cm, base fused to basal margins of column for 1.9–2.6 mm, trilobed; lateral lobes erect, narrowly lanceolate; mid-lobe slightly recurved, cordate, 9.0–10.0 × 12.0–13.0 mm, sparsely shortly hairy; disc hairy at apices of lateral lobes, papillate near base and on mid-lobe, with 2 puberulent lamellae extending to base of mid-lobe. Column arcuate, 2.5–3.0 cm long, narrowly winged; pollinia 2, yellow, ovate, 2.0–2.5 mm.

**Table 1.** Accession numbers of the specimens used in this study. A dash (-) indicates missing data.

Taxa	Voucher	ITS	<i>matK</i>	<i>rbcL</i>
<i>Cymbidium banaense</i>	Z.J.Liu 5331	MK439806	MK439759	MK439782
<i>Cymbidium eburneum</i>	Z.J.Liu 2625	MF861124	MF861040	MF861083
<i>Cymbidium maguanense</i>	Z.J.Liu 3257	MF861125	MF861084	MF861041
<i>Cymbidium wadae</i>	Peng F002	MK626688	MK644111	MK644109
<i>Cymbidium mastersii</i>	Z.J.Liu 2924	MK439816	MK439769	MK439792
<i>Cymbidium gaoligongense</i>	Z.J.Liu 6432	MF861142	MF861057	MF861101
<i>Cymbidium wilsonii</i>	Z.J.Liu 7025	MF861156	MF861072	MF861114
<i>Cymbidium iridioides</i>	Z.J.Liu 6429	MF861141	MF861056	MF861100
<i>Cymbidium tracyanum</i>	Z.J.Liu 6426	MF861123	MF861039	MF861082
<i>Cymbidium insigne</i>	Z.J.Liu 3251	MF861140	MF861055	MF861099
<i>Cymbidium whiteae</i>	-	AF470508	KX298621	-
<i>Cymbidium wenshanense</i>	Z.J.Liu 6431	MF861128	MF861043	MF861087
<i>Cymbidium hookerianum</i>	Z.J.Liu 6425	MF861143	MF861058	MF861102
<i>Cymbidium schroederi</i>	Z.J.Liu 2837	MF861155	MF861071	MF861113
<i>Cymbidium changningense</i>	Z.J.Liu 6430	MF861126	MF861042	MF861085
<i>Cymbidium elegans</i>	Z.J.Liu 6399	MF861147	MF861062	MF861106
<i>Cymbidium ensifolium</i>	Z.J.Liu 6599	MF861138	MF861053	MF861097
<i>Cymbidium sinensis</i>	Z.J.Liu 2596	MK439819	MK439771	MK439795
<i>Cymbidium faberi</i>	Z.J.Liu 7071	MF861148	MF861063	MF861107
<i>Cymbidium goeringii</i>	Z.J.Liu 2522	MK439812	MK439765	MK439788
<i>Cymbidium serratum</i>	Z.J.Liu 2575	MF861134	MF861049	MF861093
<i>Cymbidium tortisepalum</i>	Z.J.Liu 6403	MF861133	MF861048	MF861092
<i>Cymbidium lancifolium</i>	Z.J.Liu 7013	MF861137	MF861052	MF861096
<i>Cymbidium rhizomatsum</i>	Z.J.Liu 2559	MF861150	MF861066	MF861109
<i>Cymbidium aloifolium</i>	Z.J.Liu 6591	MF861139	MF861054	MF861098
<i>Cymbidium canaliculatum</i>	Z.J.Liu 6326	MF861161	MF861078	MK848061
<i>Cymbidium atropurpureum</i>	Z.J.Liu 6592	MF861153	MF861069	MF861111
<i>Cymbidium mannii</i>	Z.J.Liu 6590	MF861121	MF861037	MF861080
<i>Cymbidium paucifolium</i>	Z.J.Liu 2112	MF861151	MF861067	MF861110
<i>Cymbidium rectum</i>	MWC1460	AF470494	HM137051	-
<i>Cymbidium ×gammieanum</i>	Wu BY02	PZ316039	PZ312994	PZ312994
Outgroup				
<i>Galeandra devoniana</i>	JBL Pupulin 1133	EU877142	KF660268	AF074171
<i>Eulophia graminea</i>	Peter 7598	KF318890	MH767976	KF358040

Table 2. Morphological comparisons for *Cymbidium ×gammieanum*, *C. erythraeum* and *C. elegans*.

Character	<i>C. ×gammieanum</i>	<i>C. erythraeum</i>	<i>C. elegans</i>
Leaves	36.0–55.0 cm × 0.8–1.7 cm	45.0–92.0 cm × 0.7–1.5 cm	45.0–76.0 cm × 1.7–2.1 cm
Flower color	sepals and petals yellowish green, with reddish brown longitudinal strips and irregular spots of same color; lip white or pale yellow with red-brown venation	sepals and petals green, with dark red brown dashes and spots; lip white or pale yellow with dark red brown stripes and spots	sepals and petals cream-yellow to pale yellowish green, sometimes tinged pale green
Inflorescence	Inflorescence arising from lower part of pseudobulb, suberect or arching	Inflorescence arising from within sheaths at base of pseudobulb, suberect or arching	Inflorescence arising from lower part of pseudobulb, rachis pendulous or nodding
Sepals	Dorsal sepal narrowly obovate-oblong, 3.6–4.1 × 0.9–1.0 cm, apex acute to obtuse; lateral sepals falcate, 2.8–3.7 × 0.7–0.9 cm	narrowly oblong-oblancheolate to narrowly obovate-lanceolate, 34–43 × 7–14 mm, apex acute to obtuse	34–43 × 7–11 mm, apex acute to obtuse
Petals	falcate, ligulate, 3.3–4.3 × 0.6–0.8 cm, apex acute	falcate, ligulate, 33–53 × 4–7 mm, apex acute	broadly linear-oblancheolate, 30–40 × 5–8 mm, apex obtuse
Lip profile	ovate-elliptic, 2.5–2.9 × 1.4–1.7 mm	elliptic-ovate, 24–43 mm	oblancheolate-triangular, 30–40 × 4–8 mm
Lateral lobes	narrowly lanceolate	erect, subovate, sometimes shortly ciliate	loosely clasping column, not ciliate
Midlobe	cordate, 9.5–10.0 × 12.0–13.0 mm	slightly recurved, cordate to ensiform, 8–9 × 10–11 mm, sometimes sparsely shortly hairy	small, 6–10 × 5–8 mm, margin slightly undulate, apex slightly bilobed and incurved
Flowering period	July to October	October to January	October to December

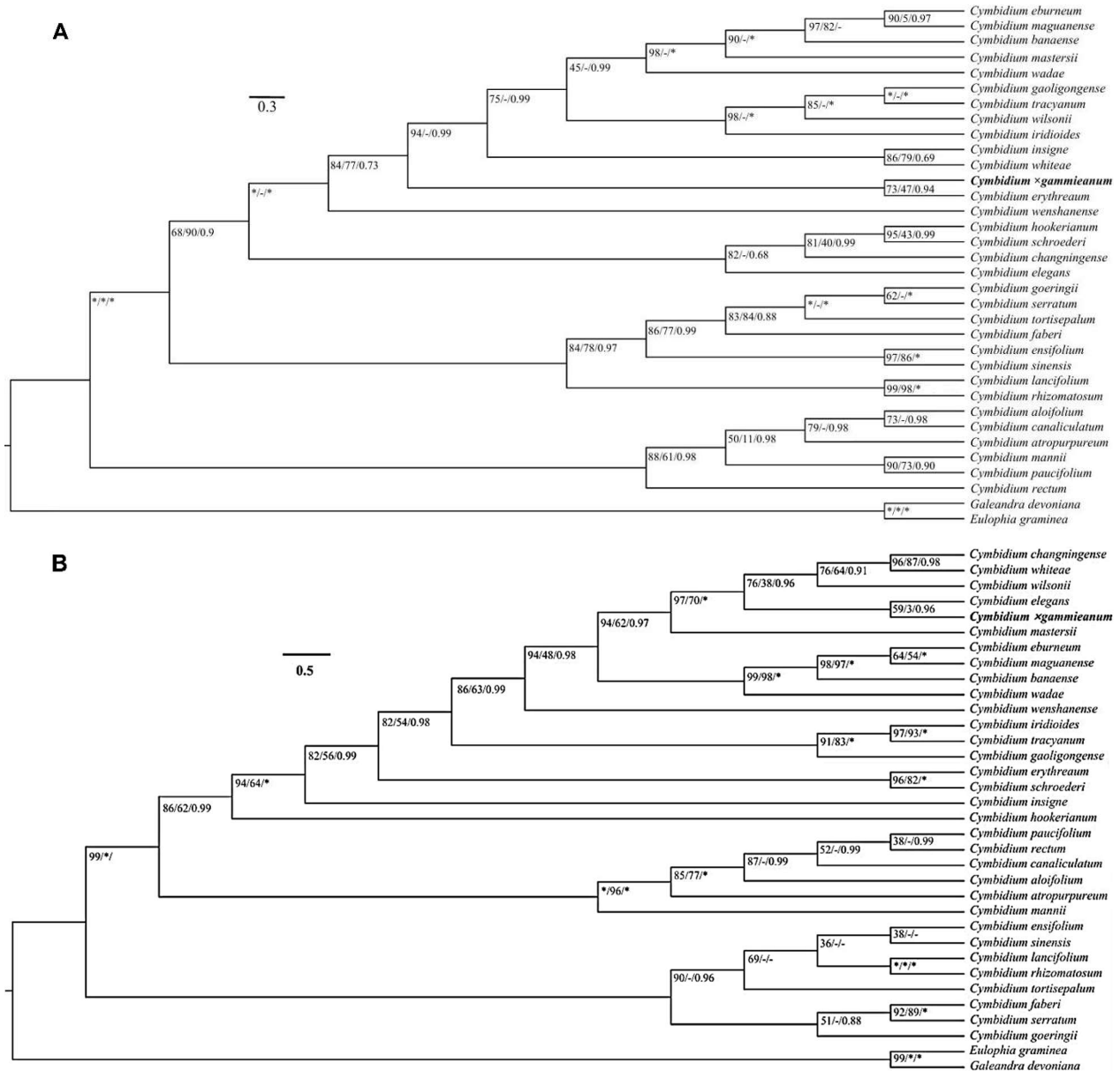


Fig. 1. Phylogenetic relationships of *C. xgammieanum* based on **A.** the nuclear DNA (ITS) **B.** the combined plastid DNA. *Galeandra devoniana* and *Eulophia graminea* were used as outgroups. The numbers near the nodes are maximum likelihood bootstrap percentages (BPML), maximum parsimony bootstrap percentages (BPMP) and Bayesian posterior probabilities (PP) respectively. “**” indicates that the node has BP 100 or PP 1.00. “-” indicates that the node is incongruent between the topology of the Bayesian and MP/ML trees.

Phenology: Flowering July to October.

Distribution: So far, this taxon has been documented in Nepal, India and Vietnam, as well as in Baoshan City and Dehong City, Yunnan Province, China.

Habitat: Epiphytic in subtropical open forest at elevations of 1700 m to 2600 m (Fig. 5).

Vernacular name: De Hong Lan 德宏兰 (Chinese name).

Additional specimens examined: Vietnam, 1935, *M. Poilane* 32079 (USNM); India, West Bengal, 1753 m, 01 January, 1984, *Bailes*, *C. 1004* (K); Nepal, Gandaki Province, 2200 m, 09 November,

2002, *A. Subedi*, *RP Chaudhary* & *LR Shakya* 1023 (N?); China, Yunnan Province, Dehong City, 2100 m, 8 August, 2025, *Wu* BY02 (NYIST).

Note: *Cymbidium xgammieanum* is a natural hybrid of *C. erythraeum* and *C. elegans*, characterized by a morphological mosaic of the two parental species. It shares floral morphology with *C. erythraeum* but differs in its yellow labellum, wider leaves, inflorescence emergence from the lower pseudobulb, and differentiated lobe morphology; it is similar to *C. elegans* in leaf texture but distinct in striated sepals/petals, floral coloration, lobe morphology.

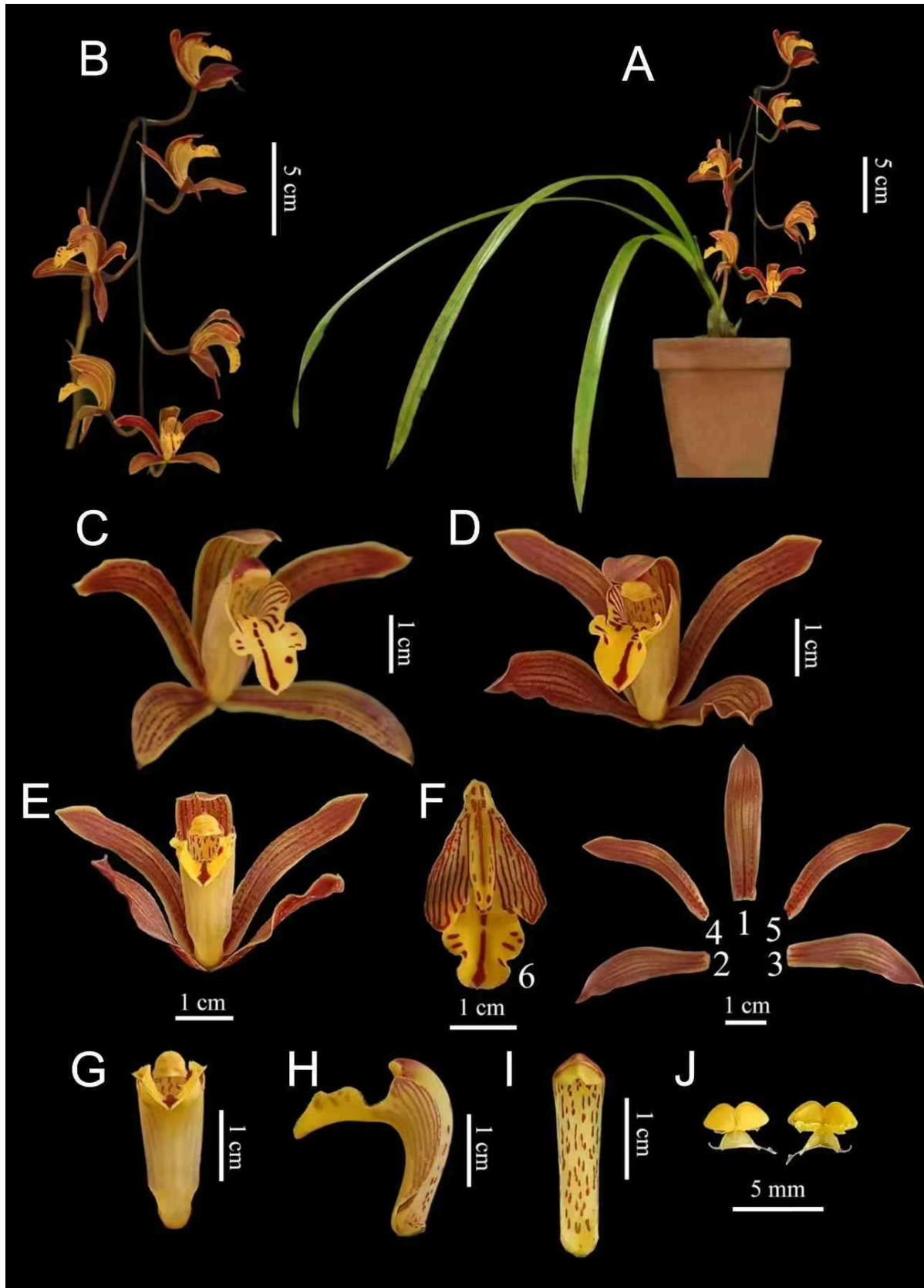


Fig. 2. *Cymbidium x gammieanum*. **A.** Flowering plant. **B.** Inflorescence. **C, D** and **E.** Flower (front view). **F.** Anatomy of flowers (F1: dorsal sepals; F2 and F3: lateral sepals; F4 and F5: petals; F6: labellum). **G** and **H.** Column with lip. **I.** Column. **J.** pollinia with stipe.

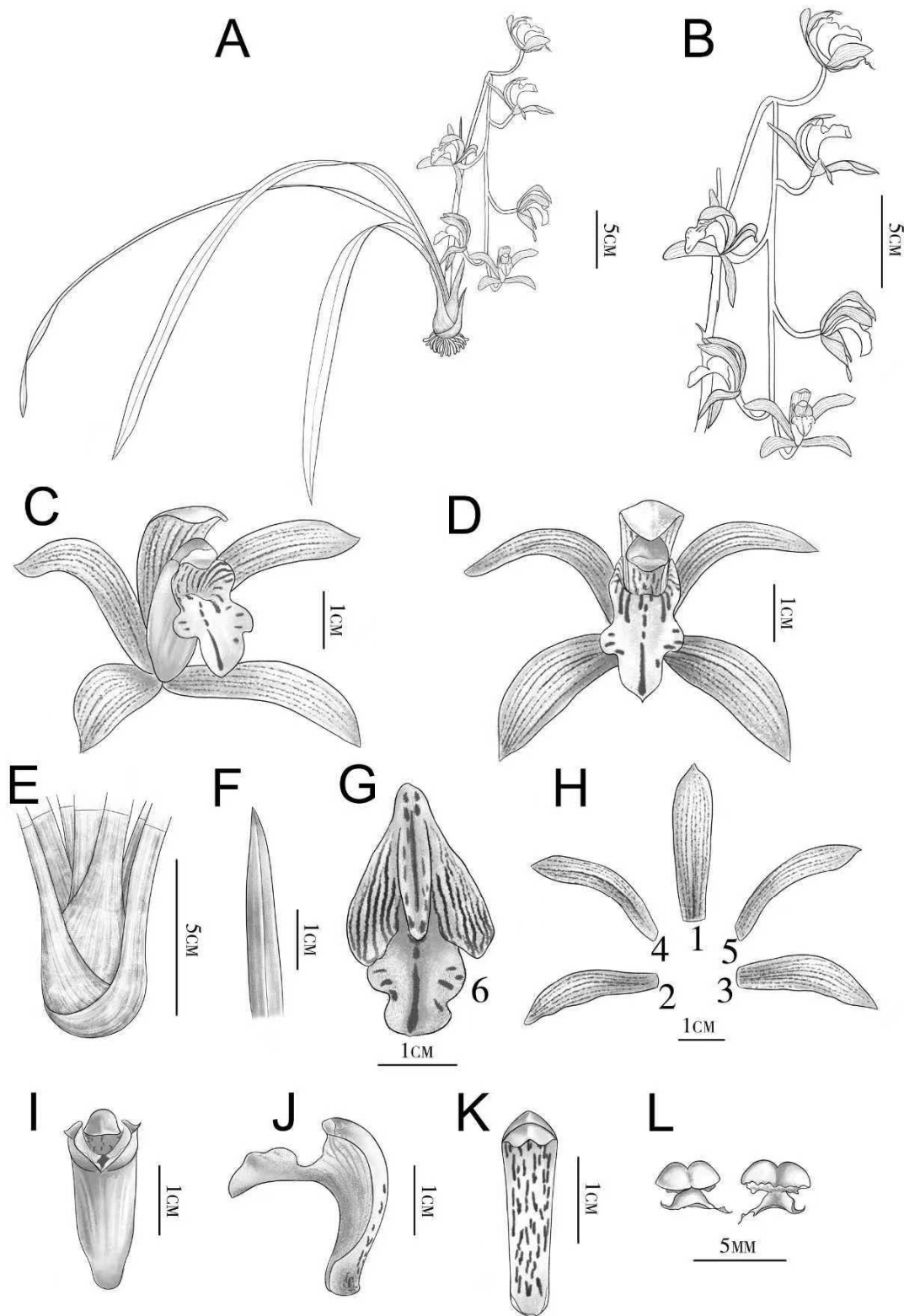


Fig. 3. *Cymbidium xgammieanum*. A. Flowering plant. B. Inflorescence. C—D. Flower. E. Pseudobulbs enveloped by leaf sheaths. F. Leaf apex. G. Labellum. H. Anatomy of flowers (H1: dorsal sepals; H2—H3: lateral sepals; H4—H5: petals). I—J. Column with lip. K. Column. L. pollinia with stipe. All figure should be labeled with the front type letter Arial, send me the file to replaced

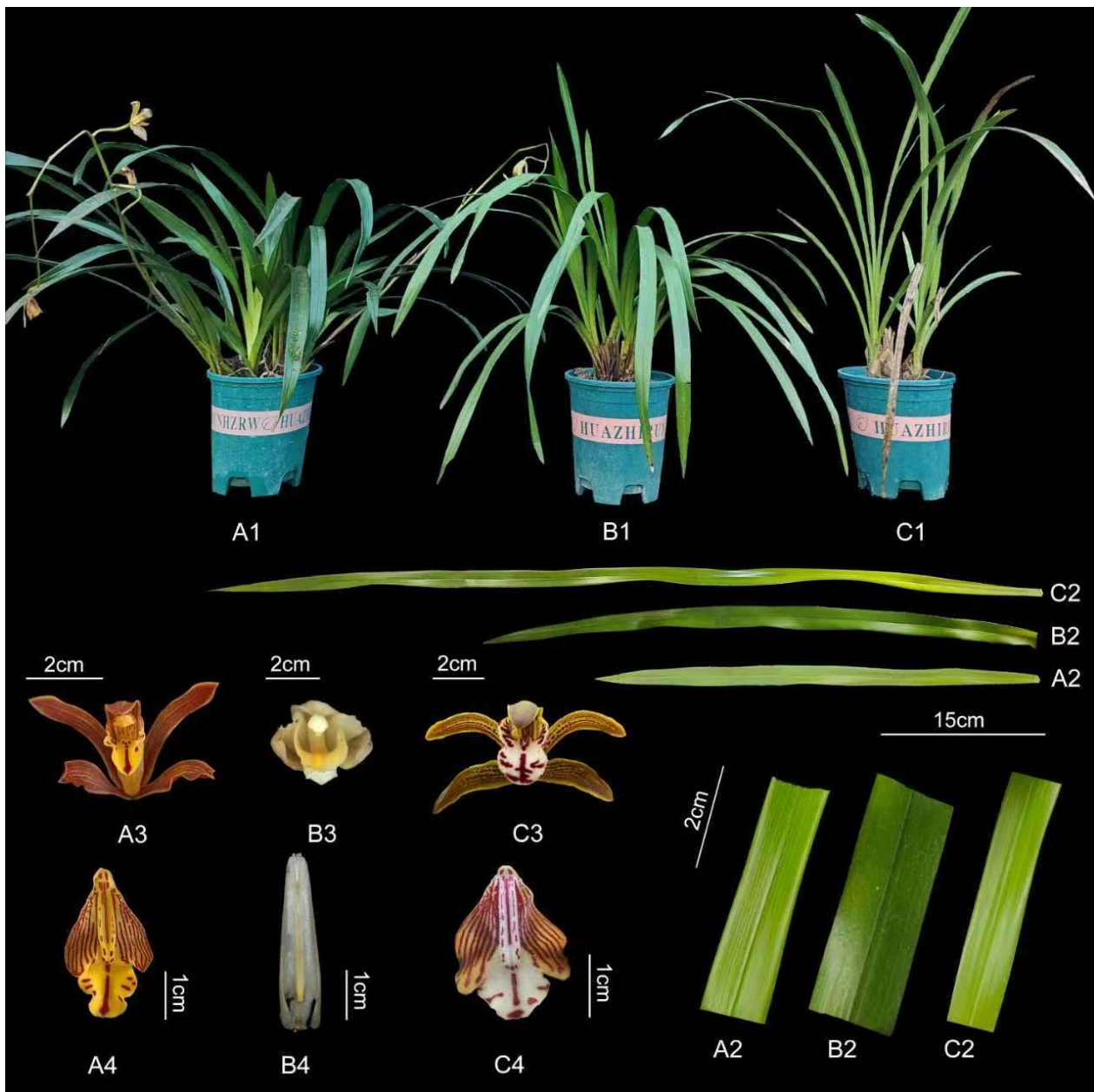


Fig. 4. . Comparisons among the three taxa of *Cymbidium* (**A.** *Cymbidium xgammieanum*. **B.** *Cymbidium elegans*. **C.** *Cymbidium erythraeum*) 1. Plant. 2. Leaf. 3. Flower (front view). 4. labellum (front view).

DISCUSSION

In China, *Cymbidium* comprises three subgenera, with *Cymbidium* subgenus *Cyperorchis* being the largest, encompassing roughly 30 species as documented by Liu *et al.*, (2006). In general, it is defined by epiphytic habits, typically large flowers, bilobed pollinia, and lateral lip lobes basally fused to the column's basal margins over 2–10 mm (Liu *et al.*, 2006). *Cymbidium xgammieanum* exhibits all these defining traits and is confirmed as a member of *Cyperorchis* via phylogenetic analyses, with a distinctive yellow labellum that differentiates it from all

other known species in this subgenus.

Phylogenetic relationships from combined plastid (*matK*, *rbcL*) and nuclear (nrITS) datasets confirm the hybrid is closely related to *C. erythraeum* and *C. elegans* within *Cyperorchis*, and the nuclear-plastid phylogenetic incongruence directly clarifies its parental lineage and inheritance pattern: the nrITS sister relationship with *C. erythraeum* identifies it as the paternal parent, while the plastid sister relationship with *C. elegans* identifies it as the maternal parent—consistent with the maternal plastid inheritance pattern of *Cymbidium* (Zhang *et al.*, 2019). This chloroplast-derived maternal lineage evidence not



Fig. 5. Habitat and flowering plant of *Cymbidium xgammieanum*. A and B. Habitat; C and D. Flowering plant in wild.



only clarifies the parental direction of the hybrid's origin, but also provides new insights into the natural hybridization mechanism of *Cymbidium* in western Yunnan, suggesting *C. elegans* may be a frequent maternal parent in interspecific hybridization of *Cyperorchis*.

Morphological traits of the hybrid align perfectly with the molecular evidence: it inherits floral structural features from the paternal parent *C. erythraeum* (e.g., falcate lateral sepals, ligulate petals) but differs in leaf morphology, labellum color and inflorescence attachment; it shares leaf texture and pseudobulb characteristics with the maternal parent *C. elegans* but differs in floral morphology and flowering period. This typical morphological mosaicism is a core diagnostic feature of natural hybrids (Soltis and Soltis, 2009), and the hybrid's unique traits (yellow labellum, inflorescence from the lower pseudobulb) further confirm it is not an intraspecific variant of either parent.

Ecologically, *C. erythraeum* and *C. elegans* are strictly sympatric with *C. xgammieanum* in western Yunnan (Baoshan and Dehong Cities) at an elevation of 1700–2600 m (Fig. 5), and their flowering periods overlap for 2–3 months—providing the necessary spatial and temporal conditions for natural cross-pollination.

The parentage of this species was initially inferred from floral color and inflorescence morphology in earlier studies, with some reports proposing *C. elegans* and *C. erythraeum* as the putative progenitors, while others suggested *C. elegans* and *C. giganteum* (Atkinson, 1895). Based on both molecular and morphological evidence, we conclude that this taxon—designated as *C. xgammieanum*, represents a natural hybrid originating from *C. erythraeum* and *C. elegans*, two species with overlapping geographic distributions and flowering periods that facilitate hybridization (Liu *et al.*, 2009; Ormerod *et al.*, 2021).

These findings provide the first empirical molecular-morphological combined evidence confirming the parental lineage of *C. xgammieanum* as *C. erythraeum* and *C. elegans*, validating the long-proposed historical hypothesis and filling a gap in the molecular characterization of this poorly documented natural hybrid.

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