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Authors: Ning, Zu-Lin, Pan, Bo, Wen, Fang, Kang, Ming, and Zhuang, Xue-Ying

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ZU-LIN NING^{1,2*}, BO PAN³, FANG WEN³, MING KANG² & XUE-YING ZHUANG^{1*}

***Primulina yingdeensis*, a new species from Guangdong, China, and *P. rosulata*, a new combination (*Gesneriaceae*), based on morphological and molecular evidence**

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Abstract: Based on morphological and molecular data, a new species of *Gesneriaceae*, *Primulina yingdeensis*, from Guangdong, China, is described and illustrated. It is morphologically most similar to *P. hochiensis* and *P. hochiensis* var. *rosulata*, but the three taxa differ from each other in discrete characters. They share yellow honey guides, but in the new species these are strongly ridged and with glands. At the same time, also based on morphological and molecular data, *P. hochiensis* var. *rosulata* is raised to the rank of species, as *P. rosulata*.

Key words: *Gesneriaceae*, *Primulina*, taxonomy, China, Guangdong, Guangxi

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Introduction

The genus *Primulina* Hance (1883) formerly composed of the single species *P. tabacum* Hance, now includes all species of *Chirita* sect. *Gibbosaccus* C. B. Clarke (1883), *Chiritopsis* W. T. Wang (1981) and two species of *Wentsaiboea* D. Fang & D. H. Qin (2004), based on taxonomic transfers supported by recent phylogenetic analyses (Möller & al. 2009, 2011; Liu & al. 2010; Wang & al. 2011; Weber & al. 2011a). The newly defined *Primulina* is one of the largest genera of the Old World *Gesneriaceae*, comprising currently around 170 species distributed in S and SW China and N Vietnam (Weber & al. 2011a; Möller & Clark 2013) with more than 40 taxa described in the last few years (Möller & al. 2016). The karst regions of S and SW China and N Vietnam show the high-

est biodiversity and differentiation of *Primulina* species, and most are narrowly endemic species with small populations distributed at only one or a few localities (Wen & Zhong 1998; Li & Wang 2004; Wei & al. 2004; Kang & al. 2014).

In 2011, during field investigations in Yingde City, N Guangdong, we collected an unknown species of *Gesneriaceae* with stolons and white flowers with a slender funnelform corolla tube. Although in corolla shape and size it is similar to *Primulina hochiensis* (C. C. Huang & X. X. Chen) Mich. Möller & A. Weber (Huang & Chen 1992; Weber & al. 2011a) and *P. hochiensis* var. *rosulata* F. Wen & Y. G. Wei (Wen & al. 2012), it differs in several other morphological characters, summarized in Table 1. These differences mean that the new collection can easily be distinguished from the described species in *Primulina*.

1 College of Forestry and Landscape Architecture, Guangdong Key Laboratory for Innovative Development and Utilization of Forest Plant Germplasm, South China Agricultural University, Guangzhou 510642, China; *e-mail: xyzhuang@scau.edu.cn (author for correspondence).

2 South China Botanical Garden, Chinese Academy of Sciences, Guangzhou 510650, China; *e-mail: ningzulin@163.com (author for correspondence).

3 Guangxi Institute of Botany, Guangxi Zhuang Autonomous Region and Chinese Academy of Sciences, Guilin 541006, China.

Table 1. Morphological comparison of *Primulina yingdeensis*, *P. hochiensis* and *P. rosulata*.

Character	<i>Primulina yingdeensis</i>	<i>Primulina hochiensis</i>	<i>Primulina rosulata</i>
Stolons	present	present	absent
Leaf blade size	6–11(–15) × 4–6.5(–10) cm	2–5.5 × 1.3–3.5 cm	3.5–6 × 2.5–4 cm
Leaf blade indumentum	abaxially densely appressed white pubescent, adaxially densely long villous and short pubescent	densely appressed puberulent on both surfaces	densely appressed puberulent on both surfaces
Lateral veins per leaf blade	5–7	3 or 4	4 or 5
Branches per cyme	1 or 2	0	0 or 1
Flowers per cyme	4–16	1–4	2–4
Peduncle length	6–15 cm	4–7 cm	3–9.5 cm
Bract shape	oblong or linear-lanceolate	linear-lanceolate	linear
Bract size	15–23 × 2–7 mm	3–4 × c. 1.2 mm	4–5 × c. 1 mm
Pedicel length	1–3.5 cm	0.7–2.3 cm	≤ 0.7 cm
Calyx lobe colour	green with purple apex or rarely all green	green	purple
Corolla colour	white	purple	white or pale pink
Corolla inside indumentum	densely glandular pubescent below stamen insertion	glabrous	glabrous
Corolla tube length	1.8–2 cm	1.5–1.8 cm	c. 1 cm
Corolla honey guides	strong ridged, covered with glands	gently ridged, without glands	gently ridged, without glands
Anther colour	light yellow	purple	dark purple or purple
Anther indumentum	abaxially densely white villous	abaxially white villous	glabrous
Pistil length	2.2–2.6 cm	1.5–2 cm	c. 1 cm

To evaluate distinctness and phylogenetic affinities, we carried out a phylogenetic analysis on a set of *Primulina* taxa including the three above-mentioned, morphologically similar taxa.

Material and methods

Material — Material of the undescribed species was collected on a limestone hill in Yingde City, Guangdong Province, China. Voucher specimens were deposited in IBSC (herbarium code according to Thiers [continuously updated]). For molecular work, leaf material from three different localities was collected. In addition, leaf material of the two morphologically similar taxa, *Primulina hochiensis* and *P. hochiensis* var. *rosulata*, was collected from two localities and one locality, respectively (see Appendix 1). All leaves were dried in silica gel. Relevant literature was consulted to compare the new collection to other species in *Primulina* (Wang 1981; Liu & Guo 1989; Wang & al. 1990, 1998; Huang & Chen 1992; Wei & al. 2000; Fang & Qin 2004; Li & Wang 2004; Shen & al. 2010; Wei & al. 2010; Wen & al. 2012; Wu & al. 2012; Chung & al. 2013; Ning & al. 2013; Zhao & al. 2013; Zheng & Deng 2014; Zhou & al. 2014; Guo & al. 2015; Ning & al. 2015).

The ingroup comprised 95 *Primulina* samples, comprising 91 taxa and 87 species including the presumed new species. Two additional species, i.e. *Petrocodon dealbatus* Hance (1883) and *Petrocodon hancei* (Hemsl.)

Mich. Möller & A. Weber (Weber & al. 2011b), were selected as outgroups based on previous phylogenetic analyses (Möller & al. 2009, 2011).

DNA sequencing and phylogenetic analyses — Sequence data for all but five samples used in this study were obtained from GenBank (Appendix 2). For the extraction of genomic DNA from the five samples, PCR procedure followed Kang & al. (2014). DNA sequences generated in this study were deposited in NCBI GenBank with accession numbers KU528868 to KU528887 (Appendix 2). A total of 95 samples were used to reconstruct the molecular phylogeny using the nuclear internal transcribed spacer (ITS) and three plastid markers (*trnL-trnF*, *rpl32-trnL* and *atpB-rbcL*). The reconstruction of the phylogenetic tree by Bayesian inference (BI) was carried out in MrBayes v.3.2.2 (Huelsenbeck & Ronquist 2001; Ronquist & Huelsenbeck 2003; Ronquist & al. 2012). The GTR + I + Γ model was obtained for the combined dataset with the Akaike Information Criterion (AIC) in MrModeltest v.2.3 (Nylander 2004). Eight million generations were run in two independent analyses each with four chains (one cold and three heated). One tree was saved every 1000 generations (= 8000 trees), and the first 800 trees (10%) were discarded as burn-in, determined after plotting the log likelihoods in the “.stat” output file. The remaining trees were used to estimate a majority rule consensus tree with the “sumt” command and the Bayesian posterior probabilities (PP) obtained in MrBayes.

Results and Discussion

Phylogenetic analysis

The concatenated and aligned DNA sequences were 4034 base pairs long. A total of 825 polymorphic sites were found, of which 456 were parsimony informative. Bayesian analysis of the molecular data revealed that the three accessions of the newly collected taxon clustered into a clade with maximum support (Fig. 1). The BI analysis also showed that the new species was sister to *Primulina hochiensis* var. *rosulata* with maximum support. Additionally, the clade comprising these two taxa nested on a polytomy including *P. hochiensis* var. *hochiensis* and *P. glandulosa* var. *yangshuoensis* (F. Wen & al.) Mich. Möller & A. Weber (Fig. 1). This configuration rendered *P. hochiensis* paraphyletic. The genetic distance is 0.015 between *P. hochiensis* var. *hochiensis* and *P. hochiensis* var. *rosulata*; it is 0.011 between *P. hochiensis* var. *hochiensis* and *P. yingdeensis*; 0.015 between *P. glandulosa* (D. Fang & al.) Yin Z. Wang and *P. fimbrisepala* (Hand.-Mazz.) Yin Z. Wang; 0.014 between *P. verecunda* (Chun) Mich. Möller & A. Weber and *P. villosissima* (W. T. Wang) Mich. Möller & A. Weber; and 0.013 between *P. fimbrisepala* and *P. villosissima*. Thus the genetic distance between *P. hochiensis* var. *hochiensis* and *P. hochiensis* var. *rosulata* is the same or larger than that between the other taxa. Because the sister relationship between *P. hochiensis* var. *rosulata* and the new taxon is strongly supported, but the monophyly of *P. hochiensis* is not supported, we raise *P. hochiensis* var. *rosulata* to the rank of species.

Taxonomic treatment

After consulting the relevant literature and performing molecular phylogenetic analyses, we conclude that our collection from Yingde City represents a new species of *Primulina*, which is described and illustrated here. In addition, some key characters of *P. hochiensis* var. *rosulata* are obviously different from *P. yingdeensis* and *P. hochiensis* var. *hochiensis*, such as plants without stolons, leaf blades elliptical to slightly ovate, and anthers glabrous; hence *P. hochiensis* var. *rosulata* is raised to the rank of species.

Primulina rosulata (F. Wen & Y. G. Wei) Z. L. Ning & X. Y. Zhuang, **comb. & stat. nov.** ≡ *Primulina hochiensis* var. *rosulata* F. Wen & Y. G. Wei in Phytotaxa 54: 37. 2012. — Holotype: China, Guangxi Zhuang Autonomous Region, Pingle county, Tong'an town, 24°34'47"N, 110°55'34"E, altitude 150 m, 17 Aug 2008, B. Gao 08171 (IBK!; isotype: BJFC!).

Distribution — Pingle County, NE Guangxi Autonomous Region, S China.

Primulina yingdeensis Z. L. Ning, M. Kang & X. Y. Zhuang, **sp. nov.** — Fig. 2 & 3.

Holotype: China, Guangdong Province, Yingde City,

Huanghua Town, 24°09'36"N, 112°54'00"E, altitude 185 m, on limestone rock surface in a karst cave, 27 Aug 2011, M. Kang & al. YD03-3 (IBSC!; isotypes: IBSC! and others to be distributed).

Diagnosis — The new species is most similar to *Primulina rosulata*, but differs by having stolons present (vs absent), peduncle longer, 6–15 cm (vs 3–9.5 cm), bracts larger, 15–23 × 2–7 mm (vs 4–5 × c. 1 mm), corolla inside densely glandular pubescent below stamen insertion (vs glabrous), honey guides strongly ridged, covered with glands (vs gently ridged, without glands), and anthers light yellow (vs dark purple or purple). The new species differs from *P. hochiensis* by having cymes 1- or 2-branched with 4–16 flowers (vs unbranched with 1–4 flowers), peduncle longer, 6–15 cm (vs 4–7 cm), bracts larger, 15–23 × 2–7 mm (vs 3–4 × c. 1.2 mm), corolla white (vs purple), inside densely glandular pubescent below stamen insertion (vs glabrous), honey guides strongly ridged, covered with glands (vs gently ridged, without glands), and anthers light yellow (vs purple).

Description — *Herbs* perennial, with stolons. *Stem* rhizomatous, subterete, 1.5–2.5 cm long, c. 1 cm in diam.; internodes inconspicuous. *Leaves* 15–18, crowded at stem and stolon apex. *Petiole* 3.5–5 cm long, 0.8–1 cm in diam., densely white appressed pubescent. *Leaf blade* ovate or ovate-elliptic, slightly asymmetric, 6–11(–15) × 4–6.5(–10) cm, herbaceous, abaxially densely appressed white pubescent, adaxially densely long villous and shortly pubescent, base cuneate, margin serrate or rarely entire, apex acute or rounded; lateral veins 5–7 on each side of midrib. *Cymes* 3–8, 1- or 2-branched, 4–16-flowered; *peduncle* 6–15 cm long, c. 0.3 cm in diam., densely puberulent; *bracts* 2, opposite, oblong or linear-lanceolate, 15–23 × 2–7 mm, pubescent, margin entire. *Pedicel* 1–3.5 cm long, pubescent. *Calyx* green with purple apex, rarely all green, 5-lobed nearly to base; *lobes* linear-lanceolate, 4–8 × 1–1.5 mm, pubescent. *Corolla* white, 2.4–3 cm long, outside glandular puberulent, inside densely glandular pubescent below stamen insertion, throat with two strongly ridged yellow longitudinal honey guides covered with glands; *tube* narrowly funnel-form, 1.8–2 cm long, orifice 6–8 mm in diam.; *upper lip* 2-lobed, lobes suborbicular, c. 5 × 6 mm, apex rounded; *lower lip* 3-lobed, lateral lobes oblong, 4–6 × 4–5 mm, apex rounded, central lobe subrounded, 4–5 × 4–5 mm, apex rounded. *Stamens* 2, adnate to c. 1.2 cm above base of corolla tube; *filaments* geniculate near middle, c. 7 mm long, glabrous; *anthers* light yellow, reniform, c. 5 × 4 mm, abaxially densely white villous. *Staminodes* 3, c. 2 mm long, glabrous, adnate to 6–8 mm above base of corolla tube. *Disc* annular, 0.5–1 mm high, glabrous, margin repand. *Pistil* 2.2–2.6 cm long; *ovary* linear, 8–10 mm long, puberulent; *style* 1.3–1.5 cm long, glandular puberulent; *stigma* obtrapeziform, c. 1 mm long, 2-lobed. *Capsule* linear, 1.5–1.8 cm long.

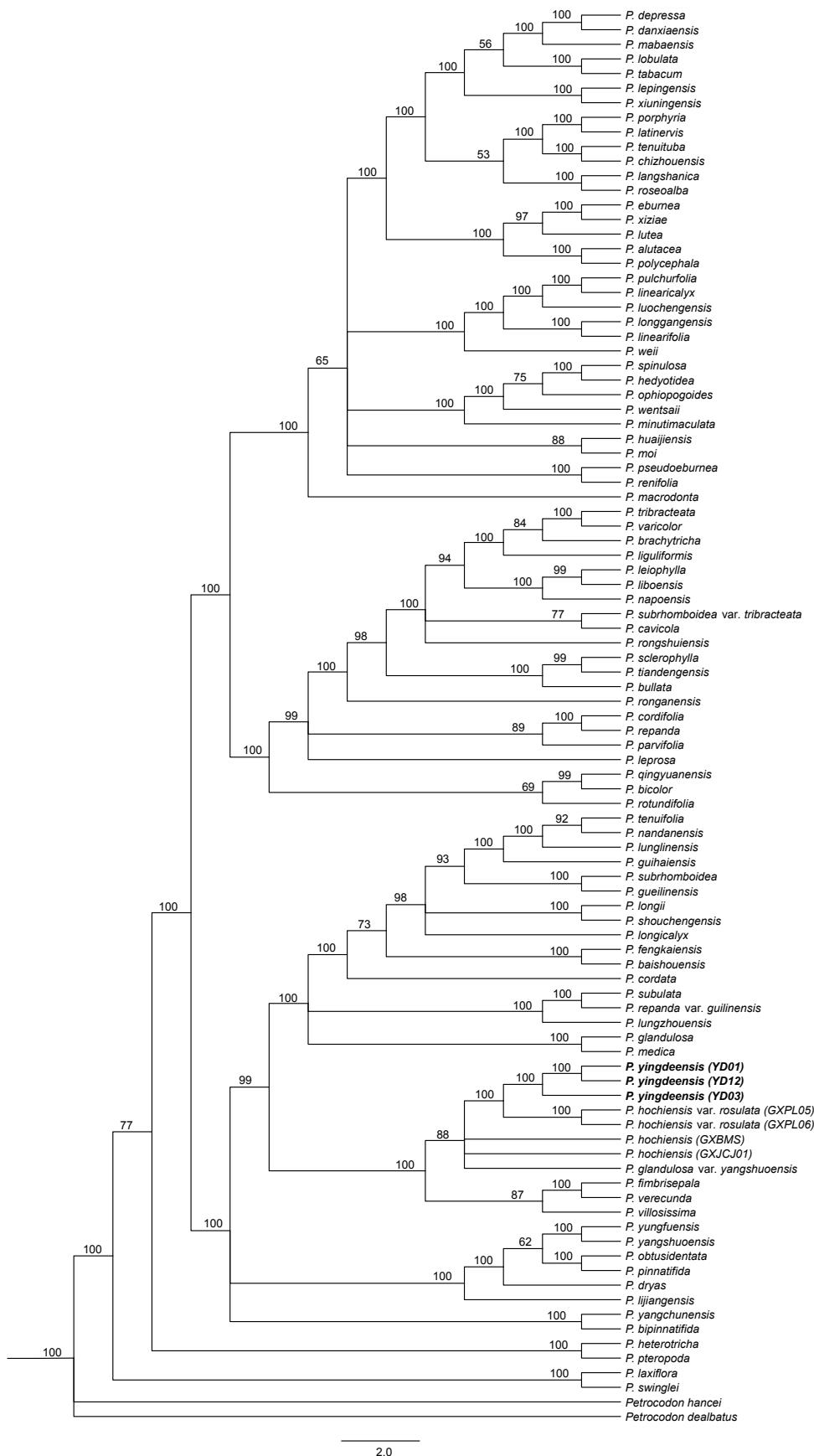


Fig. 1. Majority rule consensus Bayesian tree based on combined sequence data from the nuclear internal transcribed spacer (ITS) and three plastid markers (*trnL-trnF*, *rpl32-trnL* and *atpB-rbcL*). Numbers above branches are posterior probability values.



Fig. 2. *Primulina yingdeensis* – A: habit; B: front view of flower; C: side view of flower; D: opened corolla showing stamens and staminodes; E: adaxial view of stamens; F: abaxial view of stamens; G & H: pistil with calyx; I: stigma; J: young fruit with persistent calyx. – All drawn from the holotype by Yun-Xiao Liu.



Fig. 3. *Primulina yingdeensis* – A & B: plants in natural habitat at type locality, 4 Aug 2013; C & D: plants cultivated in South China Botanical Garden; E: pistil, calyx removed; F: inflorescence; G: front view of flower; H: corolla opened showing stamens and staminodes; I: style and stigma; J: side view of flower; K: side view of stamens; L: calyx lobes. – Scale bars: H, J & L = 1 cm. – All photographs by Zu-Lin Ning.

Phenology — Flowering from August to October, and fruiting from September to November.

Distribution and ecology — *Primulina yingdeensis* is currently known from five populations in Yingde City, NW Guangdong, S China (24.13–24.38°N, 112.90–113.09°E). During field surveys in Yingde City, we found that it is locally abundant and grows mainly on moist rock surfaces at altitudes of 125–225 m.

Conservation status — The population occurs close to the edge of a provincial road and near a village without any protective measures. Its habitat is vulnerable to destruction from human activities. The plant is easy to propagate from stolon cuttings. At present, we have introduced some individuals from the wild population into cultivation in the South China Botanical Garden and Guilin Botanical Garden, China.

Etymology — The specific epithet *yingdeensis* is derived from the name of the type locality, Yingde City.

Additional specimens seen (paratypes) — CHINA: GUANGDONG: Yingde City, Xiniu Town, Xiazai Village, 24.13°N, 113.00°E, altitude 185 m, 27 Aug 2011, Kang Ming YD01 (IBSC); Huanghua Town, Mingjing Village, 24.17°N, 112.93°E, altitude 205 m, 27 Aug 2011, Kang Ming YD110802 (IBSC); Huanghua Town, Guanyinyan, 24.16°N, 112.90°E, altitude 225 m, 27 Jun 2012, Kang Ming & Ning Zu-Lin YD03 (IBSC); Dawan Town, Changshan Village, 24.38°N, 112.97°E, altitude 200 m, 6 Aug 2012, Kang Ming & Ning Zu-Lin YD120806 (IBSC); Shigutang Town, 24.20°N, 113.09°E, altitude 125 m, 4 Aug 2015, Kang Ming & Yang Li-Hua YD12 (IBSC).

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References

Chung K. F., Huang H. Y., Peng C. I. & Xu W. B. 2013: *Primulina mabaensis* (*Gesneriaceae*), a new species

- from a limestone cave of northern Guangdong, China. — *Phytotaxa* **92**: 40–48.
- Clarke C. B. 1883: *Cyrtandreae*. — In: Candolle A. & Candolle C. (ed.), *Monographiae phanerogamarum* **5(1)**. — Parisiis: G. Masson.
- Don D. 1822: Descriptions of two new genera of Nepal plants. — *Edinburgh Philos. J.* **7**: 82–86.
- Fang D. & Qin D. H. 2004: *Wentsaiboea* D. Fang & D. H. Qin, a new genus of *Gesneriaceae* from Guangxi, China. — *Acta Phytotax. Sin.* **42**: 533–536.
- Guo J., Pan B., Liu J., Xu W. B. & Chung K. F. 2015: Three new species of *Primulina* (*Gesneriaceae*) from limestone karsts of China based on morphological and molecular evidence. — *Bot. Stud. (Taipei)* **56**: 34–46.
- Hance H. F. 1883: New Chinese *Cyrtandreae*. — *J. Bot.* **21**: 165–170.
- Huang C. C. & Chen X. X. 1992: A new medicinal species of *Chirita* (*Gesneriaceae*) from Guangxi. — *Bot. J. S. China* **1**: 14–16.
- Huelsenbeck J. P. & Ronquist F. 2001: MRBAYES: Bayesian inference of phylogeny. — *Bioinformatics* **17**: 754–755.
- Kang M., Tao J. J., Wang J., Ren C., Qi Q., Xiang Q. Y. & Huang H. W. 2014: Adaptive and nonadaptive genome size evolution in karst endemic flora of China. — *New Phytol.* **202**: 1371–1381.
- Li Z. Y. & Wang Y. Z. 2004: Plants of *Gesneriaceae* in China. — Zhengzhou: Henan Science Technology Publication House.
- Liu X. L. & Guo X. H. 1989: A new species of *Chiritopsis* from Anhui. — *Bull. Bot. Res. (Harbin)* **9(3)**: 51–54.
- Liu Y., Xu W. B. & Pan B. 2010: *Wentsaiboea tian-dengensis* sp. nov. and *W. luochengensis* sp. nov. (*Gesneriaceae*) from karst caves in Guangxi, southern China. — *Nordic J. Bot.* **28**: 739–745.
- Möller M. & Clark J. L. 2013: The state of molecular studies in the family *Gesneriaceae*: a review. — *Selbyana* **31**: 95–125.
- Möller M., Forrest A., Wei Y. G. & Weber A. 2011: A molecular phylogenetic assessment of the advanced Asiatic and Malesian didymocarpoid *Gesneriaceae* with focus on non-monophyletic and monotypic genera. — *Pl. Syst. Evol.* **292**: 223–248.
- Möller M., Pfosser M., Jang C. G., Mayer V., Clark A., Hollingsworth M. L., Barfuss M. H. J., Wang Y. Z., Kiehn M. & Weber A. 2009: A preliminary phylogeny of the didymocarpoid *Gesneriaceae* based on three molecular data sets: incongruence with available tribal classifications. — *Amer. J. Bot.* **96**: 989–1010.
- Möller M., Wei Y. G., Wen F., Clark J. L. & Weber A. 2016: You win some you lose some: updated generic delineations and classification of *Gesneriaceae* — implications for the family in China. — *Guizhaia* **36**: 44–60.

- Ning Z. L., Pan B. & Kang M. 2015: *Primulina fengkaiensis* (*Gesneriaceae*), a new species from limestone areas in western Guangdong, China. – *Phytotaxa* **197**: 296–300.
- Ning Z. L., Wang J., Smith J. F. & Kang M. 2013: *Primulina qingyuanensis* (*Gesneriaceae*), a new species from limestone areas in Guangdong, China. – *Phytotaxa* **137**: 48–52.
- Nylander J. A. A. 2004: MrModeltest v2. – Uppsala: Evolutionary Biology Centre, Uppsala University.
- Ronquist F. & Huelsenbeck J. P. 2003: MrBayes 3: Bayesian phylogenetic inference under mixed models. – *Bioinformatics* **19**: 1572–1574.
- Ronquist F., Teslenko M., van der Mark P., Ayres D. L., Darling A., Höhna S., Larget B., Liu L., Suchard M. A., Huelsenbeck J. P. 2012: MrBayes 3.2: efficient Bayesian phylogenetic inference and model choice across a large model space. – *Syst. Biol.* **61**: 539–542.
- Shen R. J., Lin S. S., Yu Y., Cui D. F. & Liao W. B. 2010: *Chiritopsis danxiaensis* sp. nov. (*Gesneriaceae*) from Mount Danxiashan, south China. – *Nordic J. Bot.* **28**: 728–732.
- Thiers B. [continuously updated]: Index Herbariorum: A global directory of public herbaria and associated staff. New York Botanical Garden's Virtual Herbarium. – Published at <http://sweetgum.nybg.org/science/ih/> [last accessed 24 Oct 2016].
- Wang W. T. 1981: Quinque genera nova Gesneriacearum e Sina. – *Bull. Bot. Res.* **1(3)**: 21–51.
- Wang W. T., Pan K. Y. & Li Z. Y. 1990: *Gesneriaceae*. – In: Wang W. T., Pan K. Y. & Li Z. Y. (ed.), *Flora Reipublicae Popularis Sinicae* **69**. – Beijing: Science Press.
- Wang W. T., Pan K. Y., Li Z. Y., Weitzman A. L. & Skog L. E. 1998: *Gesneriaceae*. – In: Wu Z. H. & Raven P. H. (ed.), *Flora of China* 18. – Beijing: Science Press; St Louis: Missouri Botanical Garden Press.
- Wang Y. Z., Mao R. B., Liu Y., Li J. M., Dong Y., Li Z. Y. & Smith J. F. 2011: Phylogenetic reconstruction of *Chirita* and allies (*Gesneriaceae*) with taxonomic treatments. – *J. Syst. Evol.* **49**: 50–64.
- Weber A., Middleton D. J., Forrest A., Kiew R., Lim C. L., Rafidah A. R., Sontag S., Triboun P., Wei Y. G., Yao T. L. & Möller M. 2011a: Molecular systematics and remodelling of *Chirita* and associated genera (*Gesneriaceae*). – *Taxon* **60**: 767–790.
- Weber A., Wei Y. G., Puglisi C., Wen F., Mayer V. & Möller M. 2011b: A new definition of the genus *Petrocodon* (*Gesneriaceae*). – *Phytotaxa* **23**: 49–67.
- Wei Y. G., Wen F., Möller M., Monro A., Zhang Q., Gao Q., Mou H. F., Zhong S. H. & Cui C. 2010: *Gesneriaceae* of south China. – Nanning: Guangxi Science Technology Publishing House.
- Wei Y. G., Wen H. Q. & Zhong S. H. 2000: New materials of *Gesneriaceae* from Guangxi, China. – *Acta Phytotax. Sin.* **38**: 297–301.
- Wei Y. G., Zhong S. H. & Wen H. Q. 2004: Studies of the flora and ecology *Gesneriaceae* in Guangxi Province. – *Acta Bot. Yunnan.* **26**: 173–182.
- Wen F., Qin G. L., Wei Y. G., Liang G. Y. & Gao B. 2012: *Primulina hochiensis* var. *rosulata* (*Gesneriaceae*) a new variety at an entrance of a limestone cave from Guangxi, China. – *Phytotaxa* **54**: 37–42.
- Wen H. Q. & Zhong S. H. 1998: The resources of ornamental plants in *Gesneriaceae* in Guangxi. – *Guishaia* **18**: 209–212.
- Wu W. H., Meng T. & Xu W. B. 2012: *Primulina sinovietnamica* (*Gesneriaceae*), a new species identified by both morphological and molecular characters from the limestone area in Guangxi, China. – *Phytotaxa* **60**: 32–40.
- Zhao B., Pan B. & Zhang Y. 2013: *Primulina guizhongensis* (*Gesneriaceae*), a new species from Guangxi, China. – *Phytotaxa* **109**: 27–35.
- Zheng Y. L. & Deng Y. F. 2014: A new species of *Primulina* (*Gesneriaceae*) from Guangdong, China. – *Phytotaxa* **163**: 48–53.
- Zhou S. B., Hong X., Ma W. & Wen F. 2014: *Primulina lechangensis* (*Gesneriaceae*), a new species from a limestone cave of northern Guangdong, China. – *Bangladesh J. Pl. Taxon.* **21**: 187–191.
- Zhou Y. L. 2004: Plant biology. – Beijing: Higher Education Press.

Appendix 1. Voucher information of samples sequenced for this study.

Taxon	Locality	Voucher
<i>Primulina hochiensis</i>	China, Guangxi, Gongcheng County, Baimian Mountain	GXBMS (IBSC)
<i>P. hochiensis</i>	China, Guangxi, Hechi City, Jinchengjiang	GXJCJ01 (IBSC)
<i>P. rosulata</i>	China, Guangxi, Pingle County, Tongan Town	GXPL05 (IBSC)
<i>P. rosulata</i>	China, Guangxi, Pingle County, Tongan Town	GXPL06 (IBSC)
<i>P. yingdeensis</i>	China, Guangdong, Yingde City, Xiuniu Town	YD01 (IBSC)
<i>P. yingdeensis</i>	China, Guangdong, Yingde City, Huanghua Town	YD03 (IBSC)
<i>P. yingdeensis</i>	China, Guangdong, Yingde City, Shigutang Town	YD12 (IBSC)

Appendix 2. GenBank accession numbers for phylogenetic reconstruction of *Primulina*. Sequences generated in this study are indicated in bold; otherwise they were obtained from Kang & al. (2014).

Taxon	nuclear ITS	<i>trnL-trnF</i>	<i>rpl32-trnL</i>	<i>atpB-rbcL</i>
<i>Primulina alutacea</i> F. Wen & al.	KF498125	KF498152	KF498313	KF497940
<i>P. baishouensis</i> (Y. G. Wei & al.) Yin Z. Wang	KF498103	KF498223	KF498261	KF497989
<i>P. bicolor</i> (W. T. Wang) Mich. Möller & A. Weber	KF498116	KF498242	KF498269	KF498002
<i>P. bipinnatifida</i> (W. T. Wang) Yin Z. Wang & J. M. Li	KF498113	KF498235	KF498331	KF497937
<i>P. brachytricha</i> (W. T. Wang & D. Y. Chen) R. B. Mao & Yin Z. Wang	KF498048	KF498155	KF498360	KF497943
<i>P. bullata</i> S. N. Lu & F. Wen	KF498071	KF498184	KF498333	KF497964
<i>P. cavicola</i> ined.	KF498075	KF498188	KF498266	KF498021
<i>P. chizhouensis</i> Xin Hong & al.	KF498108	KF498230	KF498321	KF497995
<i>P. cordata</i> Mich. Möller & A. Weber	KF498137	KF498209	KF498301	KF497977
<i>P. cordifolia</i> (D. Fang & W. T. Wang) Yin Z. Wang	KF498134	KF498193	KF498344	KF497970
<i>P. danxiaensis</i> (W. B. Liao & al.) W. B. Liao & K. F. Chung	KF498050	KF498157	KF498332	KF497945
<i>P. depressa</i> (Hook. f.) Mich. Möller & A. Weber	KF498149	KF498256	KF498358	KF498042
<i>P. dryas</i> (Dunn) Mich. Möller & A. Weber	KF498055	KF498164	KF498264	KF497951
<i>P. eburnea</i> (Hance) Yin Z. Wang	KF498126	KF498158	KF498339	KF498011
<i>P. fengkaiensis</i> Z. L. Ning & M. Kang	KF498098	KF498217	KF498362	KF498034
<i>P. fimbrisepala</i> (Hand.-Mazz.) Yin Z. Wang	KF498046	KF498153	KF498285	KF497941
<i>P. glandulosa</i> (D. Fang & al.) Yin Z. Wang var. <i>glandulosa</i>	KF498087	KF498201	KF498357	KF498029
<i>P. glandulosa</i> var. <i>yangshuoensis</i> (F. Wen & al.) Mich. Möller & A. Weber	KF498138	KF498210	KF498355	KF497978
<i>P. gueilinensis</i> (W. T. Wang) Yin Z. Wang & Yan Liu	KF498045	KF498238	KF498272	KF498036
<i>P. guihaiensis</i> (Y. G. Wei & al.) Mich. Möller & A. Weber	KF498078	KF498191	KF498271	KF497969
<i>P. hedyotidea</i> (Chun) Yin Z. Wang	KF498084	KF498198	KF498270	KF498026
<i>P. heterotricha</i> (Merr.) Y. Dong & Yin Z. Wang	KF498099	KF498218	KF498302	KF497984
<i>P. hochiensis</i> (C. C. Huang & X. X. Chen) Mich. Möller & A. Weber (GXBMS)	KU528873	KU528883	KU528878	KU528868
<i>P. hochiensis</i> (GXJCJ01)	KF498068	KF498180	KF498274	KF498019
<i>P. hochiensis</i> var. <i>rosulata</i> : see <i>P. rosulata</i>				
<i>P. huaijensis</i> Z. L. Ning & Jing Wang	KF498127	KF498159	KF498315	KF497946
<i>P. langshanica</i> (W. T. Wang) Yin Z. Wang	KF498109	KF498232	KF498277	KF497997
<i>P. latinervis</i> (W. T. Wang) Mich. Möller & A. Weber	KF498148	KF498255	KF498258	KF498041
<i>P. laxiflora</i> (W. T. Wang) Yin Z. Wang	KF498079	KF498192	KF498327	KF498023
<i>P. leiophylla</i> (W. T. Wang) Yin Z. Wang	KF498072	KF498185	KF498329	KF497965
<i>P. lepingensis</i> Z. L. Ning & Ming Kang	KF498142	KF498229	KF498322	KF497994
<i>P. leprosa</i> (Yan Liu & W. B. Xu) W. B. Xu & K. F. Chung	KF498081	KF498195	KF498276	KF498024
<i>P. liboensis</i> (W. T. Wang & D. Y. Chen) Mich. Möller & A. Weber	KF498073	KF498186	KF498280	KF497966
<i>P. liguliformis</i> (W. T. Wang) Mich. Möller & A. Weber	KF498132	KF498177	KF498293	KF498018
<i>P. lijiangensis</i> (B. Pan & W. B. Xu) W. B. Xu & K. F. Chung	KF498112	KF498151	KF498278	KF498009
<i>P. linearicalyx</i> F. Wen, B. D. Lai & Y. G. Wei	KF498091	KF498207	KF498297	KF497975
<i>P. linearifolia</i> (W. T. Wang) Yin Z. Wang	KF498085	KF498199	KF498298	KF498027

<i>Primulina lobulata</i> (W. T. Wang) Mich. Möller & A. Weber	KF498054	KF498163	KF498288	KF497950
<i>P. longgangensis</i> (W. T. Wang) Yan Liu & Yin Z. Wang	KF498150	KF498257	KF498259	KF498043
<i>P. longicalyx</i> (J. M. Li & Yin Z. Wang) Mich. Möller & A. Weber	KF498131	KF498175	KF498305	KF497958
<i>P. longii</i> (Z. Yu Li) Z. Yu Li	KF498092	KF498208	KF498282	KF497976
<i>P. lunglinensis</i> (W. T. Wang) Mich. Möller & A. Weber	KF498097	KF498216	KF498284	KF497983
<i>P. lungzhouensis</i> (W. T. Wang) Mich. Möller & A. Weber	KF498074	KF498187	KF498283	KF497967
<i>P. luochengensis</i> (Yan Liu & W. B. Xu) Mich. Möller & A. Weber	KF498077	KF498190	KF498345	KF497968
<i>P. lutea</i> (Yan Liu & Y. G. Wei) Mich. Möller & A. Weber	KF498067	KF498179	KF498275	KF497961
<i>P. mabaensis</i> K. F. Chung & W. B. Xu	KF498118	KF498244	KF498312	KF498037
<i>P. macrodonta</i> (D. Fang & D. H. Qin) Mich. Möller & A. Weber	KF498065	KF498176	KF498354	KF497959
<i>P. medica</i> (D. Fang ex W. T. Wang) Yin Z. Wang	KF498094	KF498212	KF498353	KF497980
<i>P. minutimaculata</i> (D. Fang & W. T. Wang) Yin Z. Wang	KP954419	KP954427	KP954491	KP954423
<i>P. moi</i> F. Wen & Y. G. Wei	KF498115	KF498241	KF498342	KF498001
<i>P. nandanensis</i> (S. X. Huang & al.) Mich. Möller & A. Weber	KF498069	KF498182	KF498287	KF497963
<i>P. napoensis</i> (Z. Yu Li) Mich. Möller & A. Weber	KF498070	KF498183	KF498286	KF498020
<i>P. obtusidentata</i> (W. T. Wang) Mich. Möller & A. Weber	KF498096	KF498214	KF498323	KF498033
<i>P. ophiopogoides</i> (D. Fang & W. T. Wang) Yin Z. Wang	KF498062	KF498172	KF498260	KF497956
<i>P. parvifolia</i> (W. T. Wang) Yin Z. Wang & J. M. Li	KF498057	KF498166	KF498325	KF497952
<i>P. pinnatifida</i> (Hand.-Mazz.) Yin Z. Wang	KF498111	KF498234	KF498328	KF497999
<i>P. polycephala</i> (Chun) Mich. Möller & A. Weber	KF498147	KF498249	KF498299	KF498007
<i>P. porphyria</i> X. L. Yu & Ming Li	KU173793	KU173799	KU173802	KU173796
<i>P. pseudoeburnea</i> (D. Fang & W. T. Wang) Mich. Möller & A. Weber	KF498060	KF498169	KF498338	KF497954
<i>P. pteropoda</i> (W. T. Wang) Yan Liu	KF498100	KF498219	KF498349	KF497985
<i>P. pulchurfolia</i> ined.	KF498082	KF498196	KF498279	KF498025
<i>P. qingyuanensis</i> Z. L. Ning & Ming Kang	KF498129	KF498162	KF498350	KF497949
<i>P. renifolia</i> (D. Fang & D. H. Qin) J. M. Li & Yin Z. Wang	KF498061	KF498171	KF498296	KF497955
<i>P. repanda</i> var. <i>guilinensis</i> (W. T. Wang) Mich. Möller & A. Weber	KF498066	KF498178	KF498273	KF497960
<i>P. repanda</i> (W. T. Wang) Yin Z. Wang var. <i>repanda</i>	KF498089	KF498204	KF498300	KF497974
<i>P. ronganensis</i> (D. Fang & Y. G. Wei) Mich. Möller & A. Weber	KF498135	KF498202	KF498289	KF497973
<i>P. rongshuiensis</i> (Yan Liu & Y. S. Huang) W. B. Xu & K. F. Chung	KF498088	KF498203	KF498290	KF498030
<i>P. rosealba</i> (W. T. Wang) Mich. Möller & A. Weber	KF498123	KF498251	KF498335	KF498039
<i>P. rosulata</i> (F. Wen & Y. G. Wei) Z. L. Ning & X. Y. Zhuang (GXPL05)	KU528874	KU528884	KU528879	KU528869
<i>P. rosulata</i> (GXPL06)	KU528875	KU528885	KU528880	KU528870
<i>P. rotundifolia</i> (Hemsl.) Mich. Möller & A. Weber	KP954420	KP954428	KP954492	KP954424
<i>P. sclerophylla</i> (W. T. Wang) Yan Liu	KF498130	KF498170	KF498330	KF498016
<i>P. shouchengensis</i> (Z. Yu Li) Z. Yu Li	KF498114	KF498239	KF498294	KF498010
<i>P. spinulosa</i> (D. Fang & W. T. Wang) Yin Z. Wang	KF498063	KF498173	KF498265	KF497957

<i>Primulina subrhomboidea</i> (W. T. Wang) Yin Z. Wang var. <i>subrhomboidea</i>	KF498044	KF498237	KF498281	KF497939
<i>P. subrhomboidea</i> var. <i>tribracteata</i> ined.	KF498133	KF498181	KF498292	KF497962
<i>P. subulata</i> (W. T. Wang) Mich. Möller & A. Weber	KF498056	KF498165	KF498307	KF498014
<i>P. swinglei</i> (Merr.) Mich. Möller & A. Weber	KF498090	KF498205	KF498356	KF498031
<i>P. tabacum</i> Hance	KF498110	KF498233	KF498348	KF497998
<i>P. tenuifolia</i> (W. T. Wang) Yin Z. Wang	KF498058	KF498167	KF498262	KF497953
<i>P. tenuituba</i> (W. T. Wang) Yin Z. Wang	KF498095	KF498213	KF498308	KF497981
<i>P. tiandengensis</i> (F. Wen & H. Tang) F. Wen & K. F. Chung	KF498136	KF498206	KF498295	KF498032
<i>P. tribracteata</i> (W. T. Wang) Mich. Möller & A. Weber	KF498064	KF498174	KF498352	KF498017
<i>P. varicolor</i> (D. Fang & D. H. Qin) Yin Z. Wang	KF498086	KF498200	KF498263	KF498028
<i>P. verecunda</i> (Chun) Mich. Möller & A. Weber	KF498143	KF498231	KF498324	KF497996
<i>P. villosissima</i> (W. T. Wang) Mich. Möller & A. Weber	KF498145	KF498240	KF498306	KF498000
<i>P. weii</i> Mich. Möller & A. Weber	KF498076	KF498189	KF498291	KF498022
<i>P. wentsaii</i> (D. Fang & L. Zeng) Yin Z. Wang	KF498083	KF498197	KF498336	KF497972
<i>P. xiuningensis</i> (X. L. Liu & X. H. Guo) Mich. Möller & A. Weber	KF498124	KF498252	KF498320	KF498040
<i>P. xiziae</i> F. Wen & al.	KF498122	KF498250	KF498337	KF498008
<i>P. yangchunensis</i> Y. L. Zheng & Y. F. Deng	KF498146	KF498245	KF498303	KF498004
<i>P. yangshuoensis</i> Y. G. Wei & F. Wen	KF498093	KF498211	KF498334	KF497979
<i>P. yingdeensis</i> Z. L. Ning & al. (YD01)	KF498120	KF498247	KF498310	KF498006
<i>P. yingdeensis</i> (YD03)	KU528876	KU528886	KU528881	KU528871
<i>P. yingdeensis</i> (YD12)	KU528877	KU528887	KU528882	KU528872
<i>P. yungfuensis</i> (W. T. Wang) Mich. Möller & A. Weber	KF498144	KF498236	KF498304	KF497938
<i>Petrocodon dealbatus</i> Hance	KF498053	KF498254	KF498364	KF498013
<i>Petrocodon hancei</i> (Hemsl.) Mich. Möller & A. Weber	KF498051	KF498253	KF498363	KF498012

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