



Taxonomic revision of the genus *Pseudocodon* (Campanulaceae) based on character analysis and molecular phylogeny

QIANG WANG & DE-YUAN HONG*

State Key Laboratory of Systematic & Evolutionary Botany, Institute of Botany, Chinese Academy of Sciences, Xiangshan, Beijing 100093, China.

*Author for correspondence: hongdy@ibcas.ac.cn

Abstract

All taxa of the newly established genus *Pseudocodon* (= *Codonopsis* subg. *Pseudocodonopsis*), except for *Pseudocodon rosulatus*, form a complicated group extremely controversial on species delimitation. In the present study, numerous field trips were made, 29 populations were observed, and over 800 specimens of the genus preserved in 18 herbaria were critically examined. Statistical analysis of the characters was carried out based on 148 samples from specimens and 16 field populations. Four fragments of chloroplast DNA (*atpB*, *matK*, *rbcL*, *petD* with *petB-petD* spacer) and *ITS* of nuclear DNA were used in the molecular phylogenetic analysis of 40 samples from specimens and 22 field populations. A taxonomic revision of the genus *Pseudocodon* is presented on the basis of these studies. We recognize eight species: *Pseudocodon convolvulaceus*, *Pse. graminifolius*, *Pse. grey-wilsonii*, *Pse. hirsutus*, *Pse. petiolatus*, *Pse. retroserratus*, *Pse. rosulatus*, and *Pse. vinciflorus*. In the present study, one new species and one new subspecies are described, and one new combination is made. In addition, nine taxa are treated as synonyms, and six lectotypes are designated.

Key words: morphology, multivariate statistical analyses, phylogenetics, population sampling, taxonomy

Introduction

Pseudocodon Hong & Sun (in Wang *et al.* 2014: 546) is a newly established genus in the Campanulaceae. This new genus is distributed in the Himalaya and SW China, from central Nepal eastward to S Yunnan and W Guizhou. *Pseudocodon* was treated as a subgenus (Shen & Hong 1983; Hong *et al.* 2011) of *Codonopsis* Wallich (1824: 103), i.e. *Cod.* subg. *Pseudocodonopsis* Komarov (1908: 102). However, *Pseudocodonopsis* is distinct from *Codonopsis* in a number of morphological characters. For example, it possesses a rotate corolla, globose (rarely elongated) roots, flowers without nectary disc, and a unique type of filaments. Wang *et al.* (2014) raised *Cod.* subg. *Pseudocodonopsis* to the generic rank based on molecular phylogenetics, palynology, cytology, and analysis of morphological characters.

The delimitation of species in *Pseudocodon* has been problematic and highly controversial. Within *Pseudocodon*, only *Pseudocodon rosulatus* (W. W. Smith) Hong in Wang *et al.* (2014: 548) (= *Cod. rosulata* Smith [1921: 157–158]) is clearly distinct from all the other species, and has been continuously recognized as such. In contrast, taxonomic treatments of the other 14 described taxa have long been controversial (Table 1). For the sake of convenience of discussion, we here use the term ‘*Cod. convolvulacea* complex’ to cover all 14 of these taxa. This extremely complicated ‘*Cod. convolvulacea* complex’ comprises *Cod. convolvulacea* Kurz (1873: 195), *Cod. efilamentosa* Smith (1913: 107–108), *Cod. forrestii* Diels (1912: 171), *Cod. forrestii* var. *heterophylla* Wu in Wu & Li (1965: 80), *Cod. forrestii* var. *hirsuta* Tsoong & Shen in Shen *et al.* (1975: 55), *Cod. graminifolia* Lévillé (1916: 24), *Cod. grey-wilsonii* Shaw (1996: 93), *Cod. limprichtii* Lingelsheim & Borza (1914: 391–392), *Cod. limprichtii* var. *hirsuta* Handel-Mazzetti (1924: 169), *Cod. limprichtii* var. *pinifolia* Handel-Mazzetti (1924: 170), *Cod. macrophylla* Lammers & Klein (2010: 557–558), *Cod. mairei* Lévillé (1916: 24), *Cod. retroserrata* Wang & Xu (1993: 186), and *Cod. vinciflora* Komarov (1908: 103–104).

to rounded, apex obtuse to acute, 2–3.5 cm long, 1.5–2.5 cm wide, margin entire to shallowly crenulate. Flowers solitary, terminal on main stem. Hypanthium obtriangular; calyx lobes 5, strongly reflexed even at anthesis, linear-lanceolate, entire, 7–9 mm long, 3–3.5 mm wide. Corolla blue, 5-lobed to near base, rotate; corolla lobes elliptic, ca 2 cm long, 1 cm wide. Stamens free; filaments 4 mm long, basal part strongly dilated into triangular, ciliate; anthers 4 mm long. Ovary inferior; style glabrous, ca 2 mm long; stigma 3-fid, stigma lobes elliptic-oblong, ca 4 mm long, 2.5 mm wide. Fruit unknown.

Pseudocodon petiolatus differs distinctly from all other species of the genus in having elongate roots, long petioles, strongly reflexed calyx lobes, and large stigma lobes.

Distribution:—Endemic to China: SW Sichuan.

8. *Pseudocodon rosulatus* (W. W. Smith) Hong in Wang *et al.* (2014: 548). *Codonopsis rosulata* Smith (1921: 157). Type:—CHINA. Sichuan, Muli, 28°12'N, 3350 m, August 1918, *G. Forrest 16856* (holotype E).

Distribution:—Endemic to China: SW Sichuan.

Acknowledgements

We are grateful to the curators of the herbaria B, BM, CAL, CAS, CDBI, E, G, GH, HNWP, K, KATH, KUN, LD, PE, SM, SZ, TI, W, and WU for permission to examine specimens. Thanks are due to Dr. Shi-Liang Zhou and Mr. Tao Chen for their advice and considerable help in methodology, and to Miss Ai-Li LI for drawing the figure 5. This research was financially supported by the National Natural Science Foundation of China (Grant Nos. 31170175, 31110103911), and the National Science Infrastructure Platform Foundation of China (Grant No. 2013FY112100).

References

- Anthony, J. (1926) A key to the genus *Codonopsis* Wall. with an account of two undescribed species. *Notes from the Royal Botanic Garden, Edinburgh* 15: 173–190.
- Ballard, F. (1939) *Codonopsis convolvulacea* var. *forrestii*. *Curtis's Botanical Magazine* 162: 499, tab. 9581.
- Besant, J.W. (1925) Notes from Glasnevin. *Codonopsis forrestii*. *Gardeners Chronicle, Series 3* 78: 125, 128.
- Diels, L. (1912) *Plantae Chinenses Forrestianae*: new and imperfectly known species. *Notes from the Royal Botanic Garden, Edinburgh* 5: 161–304.
- Finlay, M.K. (1972) On *Codonopsis* which grow at Keillour. *Journal of the Royal Horticultural Society* 97: 82–87, figs. 33–37.
- Grey-Wilson, C. (1990) A survey of *Codonopsis* in cultivation. *The Plantsman* 12: 64–99.
- Grey-Wilson, C. (1995) *Codonopsis convolvulacea* and its allies. *The New Plantsman* 2: 213–225.
- Handel-Mazzetti, H.R.E. (1924) *Plantae novae Sinenses*. *Anzeiger der Akademie der Wissenschaften in Wien. Mathematisch-naturwissenschaftliche Klasse, Vienna* 61: 162–170.
- Hara, H. (1978) New or noteworthy flowering plants from eastern Himalaya (21). *Journal of Japanese Botany* 53: 134–140.
- Hong, D.Y. (1985) Campanulaceae: *Codonopsis*. In: Wu, C.Y. (Ed.) *Flora Xizangica. Vol. 4*. Science Press, Beijing, pp. 568–583.
- Hong, D.Y. (2010) Taxonomic notes on Chinese Campanulaceae. *Novon* 20: 420–425.
<http://dx.doi.org/10.3417/2010001>
- Hong, D.Y. & Ma, L.M. (1992) Campanulaceae. In: Li, H.W. & Zhu, Z.Y. (Eds.) *Flora of Sichuan. Vol. 10*. Sichuan National Press, Chengdu, pp. 515–586.
- Hong, D. & Ma, L. (1994) *Codonopsis*. In: Wang, W. & Wu, S. (Eds.) *Vascular Plant of the Hengduan Mountains. Vol. 2*. Science Press, Beijing, pp. 1954–1961.
- Hong, D.Y., Lammers, T.G. & Klein, L.L. (2011) *Codonopsis*. In: Wu, Z.Y., Raven, P.H. & Hong, D.Y. (Eds.) *Flora of China. Vol. 19*. Science Press & Missouri Botanical Garden Press, Beijing & St. Louis, pp. 513–526.
- Komarov, V.L. (1908) Prolegomena ad Floras Chinae nec non Mongoliae. *Trudy Imperatorskago S.-Peterburgskago Botaničeskago Sada* 29 (1): 1–179.
- Kurz, S. (1873) On a few new plants from Yunan. *Journal of Botany, British and Foreign* 11: 193–196.
- Larkin, M.A., Blackshields, G., Brown, N.P., Chenna, R., McGettigan, P.A., McWilliam, H., Valentin, F., Wallace, I.M., Wilm, A., Lopez,

- R., Thompson, J.D., Gibson, T.J. & Higgins, D.G. (2007) Clustal W and Clustal X version 2.0. *Bioinformatics* 23: 2947–2948. <http://dx.doi.org/10.1093/bioinformatics/btm404>
- Lammers, T.G. & Klein, L.L. (2010) A synopsis of *Codonopsis* subg. *Pseudocodonopsis* (Campanulaceae: Campanuloideae), with description of a new species of uncertain provenance. *Botanical Studies* 51: 553–561.
- Léveillé, H. (1916) *Catalogue des Plantes de Yun-Nan [2]*. Imprimerie Monnoyer, Le Mans, pp. 16–32.
- Li, J.L., Wang, S., Yu, J., Wang, L. & Zhou, S.L. (2013) A Modified CTAB Protocol for Plant DNA Extraction. *Chinese Bulletin of Botany* 48: 72–78. <http://dx.doi.org/10.3724/SP.J.1259.2013.00072>
- Lingelsheim, A. & Borza, A. (1914) Plantae novae Limprichtianae in Yunnan collectae. *Repertorium Specierum Novarum Regni Vegetabilis* 13: 385–392. <http://dx.doi.org/10.1002/fedr.19140132403>
- Miller, M.A., Pfeiffer, W. & Schwartz, T. (2010) Creating the CIPRES Science Gateway for inference of large phylogenetic trees. In: *Proceedings of the Gateway Computing Environments Workshop (GCE), 14 Nov. 2010*. New Orleans, LA., pp. 1–8. <http://dx.doi.org/10.1109/GCE.2010.5676129>
- Nannfeldt, J.A.F. (1936) Campanulaceae. In: Handel-Mazzetti, H.R.E. (Ed.) *Symbolae sinicae. Vol. 7*. Springer, Vienna, pp. 1067–1081.
- Nylander, J.A.A., Wilgenbusch, J.C., Warren, D.L. & Swofford, D.L. (2008) AWTY (are we there yet?): a system for graphical exploration of MCMC convergence in Bayesian phylogenetics. *Bioinformatics* 24: 581–583. <http://dx.doi.org/10.1093/bioinformatics/btm388>
- Rambaut, A. (2002) *Se-Al Sequence Alignment Editor v. 2.0a11*. Department of Zoology, University of Oxford, Oxford.
- Ronquist, F. & Huelsenbeck, J.P. (2003) MrBayes version 3.0: Bayesian phylogenetic inference under mixed models. *Bioinformatics* 19: 1572–1574. <http://dx.doi.org/10.1093/bioinformatics/btg180>
- Shaw, J.M.H. (1996) A new name in *Codonopsis*. *New Plantsman* 3: 93.
- Shen, L.D. & Hong, D.Y. (1983) *Codonopsis*. In: Hong, D.Y. (Ed.) *Flora Reipublicae Popularis Sinicae. Vol. 73 (2)*. Science Press, Beijing, pp. 32–69.
- Shen, L.D., Tang, X.Y., Yue, S.J. & Xie, C.K. (1975) The genus *Codonopsis* Wall. in Szechuan. *Acta Phytotaxonomica Sinica* 13 (3): 55–68.
- Smith, W.W. (1913) Diagnoses specierum novarum chinensium in herbario Horti Regii Botanici Edinburgensis cognitarum I–L. *Notes from the Royal Botanic Garden, Edinburgh* 13: 105–136.
- Smith, W.W. (1921) Diagnoses specierum novarum in herbario Horti Regii Botanici Edinburgensis cognitarum DI–DL. *Notes from the Royal Botanic Garden, Edinburgh* 13: 149–187.
- Swofford, D.L. (2002) *PAUP*: phylogenetic analysis using parsimony (*and other methods)*, version 4.0b10. Sinauer, Sunderland, Massachusetts.
- Wallich, N. (1824) *Codonopsis* Wall. In: Roxburgh, W. (Ed.) *Flora Indica. Vol. 2*. The Mission Press, Serampore, pp. 103–107.
- Wang, Q., Zhou, S.L. & Hong, D.Y. (2013) Molecular phylogeny of the Platycodonoid group (Campanulaceae s. str.) with special reference to circumscription of *Codonopsis*. *Taxon* 62: 498–504. <http://dx.doi.org/10.12705/623.2>
- Wang, Q., Ma, X.T. & Hong, D.Y. (2014) Phylogenetic analyses reveal three new genera of the Campanulaceae. *Journal of Systematics and Evolution* 52: 541–550. <http://dx.doi.org/10.1111/jse.12096>
- Wang, Z.T. & Xu, G.J. (1993) Two new species of *Codonopsis* from China. *Acta Phytotaxonomica Sinica* 31: 184–187.
- Wu, C.Y. (1965) Campanulaceae. In: Wu, C.Y. & Li, H.W. (Eds.) *Flora report on tropic & subtropic Yunnan. Vol. 1*. Science Press, Beijing, pp. 57–97.