



## Two new species of *Dendrobium* (Orchidaceae: Epidendroideae) from China: evidence from morphology and DNA

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### Abstract

Two new orchid species, *Dendrobium wenshanense* and *D. longlingense*, from Yunnan, China, are described and illustrated in this study based on morphological and molecular evidence. Morphological analysis indicates that *D. wenshanense* is similar to *D. wilsonii* and *D. fanjingshanense*, but they differ in floral color, length of column foot and shapes/sizes of sepals, petals and lip; *D. longlingense* is similar to *D. longicornu*, but differs in presence of black hairs on adaxial surface of leaf, position of inflorescences, floral color and shapes of lip and spur. Molecular analysis of combined nuclear and plastid datasets (ITS, *matK*, *rbcL*, *trnH-psbA*, *trnL* intron) reveals that *D. wenshanense* is sister to *D. wilsonii* and *D. fanjingshanense* and a member of section *Dendrobium*; *D. longlingense* is sister to *D. longicornu* and a member of section *Formosae*.

**Key words:** Chinese orchid flora, *Dendrobium* sect. *Dendrobium*, *Dendrobium* sect. *Formosae*, molecular phylogenetics of *Dendrobium*

### Introduction

The genus *Dendrobium* Swartz (1799: 82; Orchidaceae: Epidendroideae), with more than 1100 species, is one of the largest genera in Orchidaceae. It is widely distributed in India, Japan, Malaysia, Indonesia, Australia and New Guinea, with some species extending to the Pacific islands (Wood, 2006). Some 80 species are found in China, of which 14 are endemic (Zhu *et al.*, 2009). The genus is characterized by lateral inflorescences, lateral sepals adnate to elongate column foot and part of lip to form a mentum and four naked pollinia. Lindley (1844) proposed four sections initially, later expanded to ten (1851). Schlechter (1912) recognised four subgenera and 41 sections. Recently, Clements *et al.* (2002, 2003, 2006) divided the broad genus *Dendrobium* into three subtribes, but Schuiteman (2011) proposed a broad circumscription of *Dendrobium* including the other genera of the subtribe Dendrobiinae.

Given the widely varying habitats, large number of species and diverse morphologies, the taxonomy of this genus is complicated, and many species have not been found since they were originally described. Recently, during our fieldwork in Yunnan, two new species were discovered. After careful observation of transplanted individuals in The National Orchid Conservation Center of China (NOCC), morphological studies and literature comparison of similar *Dendrobium* species, molecular phylogenetic analysis was conducted to determine their phylogenetic positions. The morphological analyses indicated that they are new species of *Dendrobium*, and the molecular analyses placed one in section *Dendrobium* and the other in section *Formosae*.

### Materials and methods

**Macromorphology and micromorphology observation:**—Gross morphological data were obtained during fieldwork, and specimens were deposited in the herbarium of NOCC. Measurements, shapes, colours and other details given in the description are based on living material. Pollinia were taken from fully opened flowers, and the micromorphological observations were conducted under a Guiguang XTL-500 microscope.

**Conservation status:**—Using the World Conservation Union Red List Categories and Criteria (IUCN, 2001), *D. longlingense* should be treated as critically endangered.

**Species recognition:**—*Dendrobium longlingense* is distinct among species in the genus because it has dense black hairs on leaf abxial surface and leaf sheaths, subterminal inflorescences arising from leafless stems, and a golden yellow lip slightly curved to form spur. The lip is subflabellate with a sharp tooth at the apex of midlobe and a disk with one thick, fleshy longitudinal ridge. It is morphologically similar to *D. longicornu*, but can be easily distinguished from the latter, which has blackish brown rigid hairs on both leaf surfaces and leaf sheaths and subterminal inflorescences on leafy stems. The lip is subobovate or rhombic. The central part of the disk is orange with 3 or 4 keeled longitudinal ridges. The mentum is straight and forms a spur.

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## References

- Clements, M.A. & Jones, D.L. (2002) Nomenclatural changes in the Dendrobieae (Orchidaceae) 1: the Australasian region. *Orchadian* 13: 485–497.
- Clements, M.A. (2003) Molecular phylogenetic systematics in the Dendrobiinae (Orchidaceae), with emphasis on *Dendrobium* section *Pedilonum*. *Telopea* 10: 247–298.
- Clements, M.A. (2006) Molecular phylogenetic systematics in Dendrobieae (Orchidaceae). *Aliso* 22: 465–480.
- Doyle, J.J. & Doyle, J.L. (1987) A rapid isolation procedure from small quantities of fresh leaf tissue. *Phytochem Bull* 19: 11–15.
- IUCN (2001) IUCN red list categories and criteria, version 3.1. Gland, IUCN Species Survival Commission.
- Lindley, J. & Paxton, J. (1851) The transparent dendrobe. *Paxton's Flower Garden* 1: 133–136.  
<http://dx.doi.org/10.1017/cbo9781139095648.028>
- Lindley, J. (1844) *Dendrobium*. *Botanical Register* 30: 46–64.
- Liu, Z.J., Chen, L.J., Chen, S.C., Cai, J., Tsai, W.C., Hsiao, Y.Y., Ma, X.Y. & Zhang, G.Q. (2011) *Paraholcoglossum* and *Tsiorchis*, two new orchid genera established by molecular and morphological analyses of the *Holcoglossum* alliance. *PLoS ONE* 6: e24864.  
<http://dx.doi.org/10.1371/journal.pone.0024864>
- Mike, T., Lena, S. & Joachim, W.K. (1999) The phylogenetic relationships and evolution of the Canarian laurel forest endemic *Ixanthus viscosus* (Aiton) Griseb. (Gentianaceae): evidence from *matK* and ITS sequences, and floral morphology and anatomy. *Plant Systematics Evolution* 218: 299–317.  
<http://dx.doi.org/10.1007/bf01089233>
- Reeves, G., Chase, M.W., Goldblatt, P., Rudall, P., Fay, M.F., Cox, A.V., Lejeune, B. & Souza-Chies, T. (2001) Molecular systematics of Iridaceae: evidence from four plastid DNA regions. *American Journal of Botany* 88: 2074–2087.  
<http://dx.doi.org/10.2307/3558433>
- Sang, T., Crawford, D.J., Stuessy, T.F. (1997) Chloroplast DNA phylogeny, reticulate evolution, and biogeography of *Paeonia* (Paeoniaceae). *American Journal of Botany* 84: 1120–1136.  
<http://dx.doi.org/10.2307/2446155>
- Schlechter, R. (1912) Die Orchidaceen von Deutsch-Neu-Guinea. In: Fedde, F. (Eds.) *Repertorium Specierum Novarum Regni Vegetabilis*. Beihefte Band I. Koeltz, Berlin, Germany.
- Schuiteman, A. (2011) *Dendrobium* (Orchidaceae): to split or not to split? *Gardens' Bulletin Singapore* 63: 245–257.
- Sulaiman, S.F., Culham, A. & Harborne J.B. (2003) Molecular phylogeny of Fabaceae based on *rbcL* sequence data: with special emphasis on the tribe Mimoseae (Mimosoideae). *Asia Pacific Journal of Molecular Biology and Biotechnology* 11: 9–35.
- Swartz, O. (1799) *Dendrobium*. *Nova Acta Regiae Societatis Scientiarum Upsaliensis* 6: 82–85.
- Taberlet, P., Gielly, L., Pautou, G. & Bouvet, J. (1991) Universal primers for amplification of three non-coding regions of chloroplast DNA. *Plant Molecular Biology* 17: 1105–1109.  
<http://dx.doi.org/10.1007/bf00037152>
- Wood, H.P. (2006) *The dendrobiums*. Gantner, Ruggell, pp. 3–10.

- Xiang, X.G., Schuiteman, A., Li, D.Z., Huang, W.C., Chung, S.W., Li, J.W., Zhou, H.L., Jin, W.T., Lai, Y.J., Li, Z.Y. & Jin, X.H. (2013) Molecular systematics of *Dendrobium* (Orchidaceae, Dendrobieae) from mainland Asia based on plastid and nuclear sequences. *Molecular Phylogenetics and Evolution* 65: 950–960.  
<http://dx.doi.org/10.1016/j.ympev.2013.06.009>
- Zhang, G.Q., Liu, K.W., Chen, L.J., Xiao, X.J., Zhai, J.W., Li, L.Q., Cai, J., Hsiao, Y.Y., Rao, W.H. Huang, J., Ma, X.Y., Chung, S.W., Huang, L.Q., Tsai, W.C. & Liu, Z.J. (2013) A new molecular phylogeny and a new genus, *Pendulorchis*, of the *Aerides–Vanda* alliance (Orchidaceae: Epidendroideae). *PloS One* 8: e60097.  
<http://dx.doi.org/10.1371/journal.pone.0060097>
- Zhu, G.H., Ji, Z.H., Wood, J.J. & Wood, H.P. (2009) *Dendrobium*. In: Wu, C.Y., Raven, P.H. & Hong, D.Y. (Eds.) *Flora of China*. Scientific Press, Beijing, pp. 367–397.