

## Two new species of *Mycena* in Taiwan

Chiung-Chih Chang<sup>1</sup>, Wen-Neng Chou<sup>2\*</sup>

<sup>1</sup>*Biodiversity Research Center, Academia Sinica, Nankang, Taipei 115, Taiwan*

<sup>2</sup>*Department of Biology, National Museum of Natural Science, Taichung 404, Taiwan*

(Accepted: July 3, 2019)

### ABSTRACT

*Mycena diplazii* and *M. subcyanocephala* are proposed as new species, supported by morphological and phylogenetic analyses. The phylogenetic tree inferred from ITS sequences suggests the placement of the new taxa within the genus *Mycena*. These two new species were found in subtropical Taiwan. *Mycena diplazii*, a species of section *Basipedes* inhabiting living stem of *Diplazium esculentum*, is characterized mainly by a distinct basal disc and forming a pseudocollarium lamellae. *Mycena subcyanocephala*, a lignicolous species of section *Spinosaes*, is characterized mainly by tomentulose fruitbodies, a white cap with pale blue tone, thin-walled pileiocystidia and caulocystidia, smooth round-headed cheilocystidia, inamyloid basidiospores, and the 2-spored basidia.

**Key words:** section *Spinosaes*, section *Basipedes*, ITS, taxonomy

### Introduction

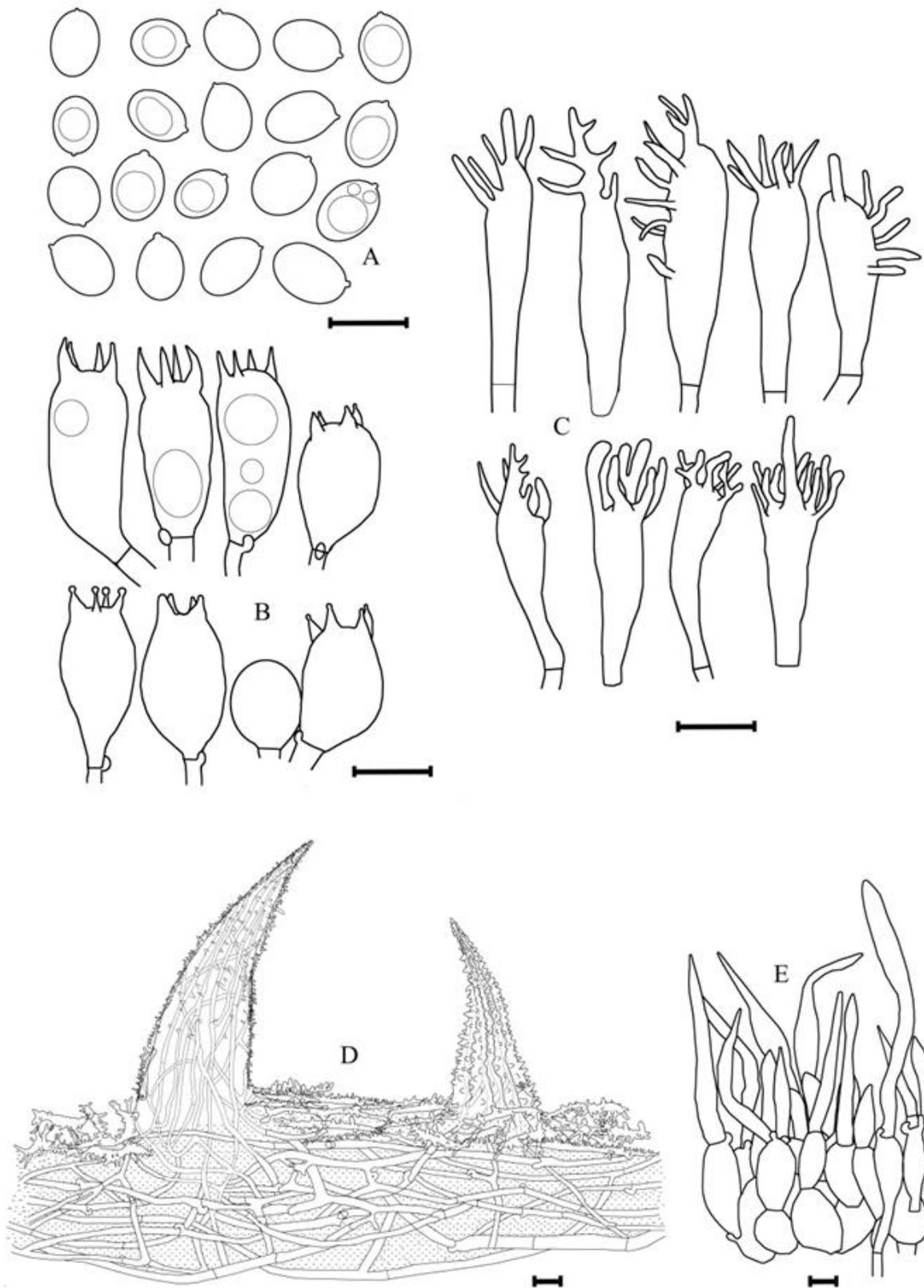
The genus *Mycena* (family Mycenaceae Overeem) is generally characterized by having tiny fruiting bodies, white spore prints, and amyloid basidiospores. The genus so far accommodates over 500 species worldwide, and classified in at least 60 sections (Desjardin et al. 2008). Twenty-one species of *Mycena* have been reported in Taiwan (Chang and Ju 2017, Wei and Kirschner 2018). Field surveys of *Mycena* in Taiwan conducted in 2013 and 2015 have revealed two undescribed species based on morphological and phylogenetic evidence. They are described as new herein.

### Materials and Methods

The studied specimens were deposited in the

herbarium of National Museum of Natural Science, Taichung, Taiwan (TNM). For microscopic observation, thin sections of specimens were mounted in 3% KOH and stained with 1% phloxine. Melzer's reagent (IKI) was used to detect the amyloid reactions of basidiospores. DNA extraction and PCR amplification of the internal transcribed spacers (ITS) were described in Huang and Wang (2011). The obtained ITS sequences were assembled by using BioEdit (Hall 1999), and subsequently submitted to GenBank. For phylogenetic analyses, the ITS dataset included the sequences of the two *Mycena* species described herein and their related *Mycena* species retrieved from GenBank. The alignment was conducted by using MAFFT 7 with Q-INS-I strategy (Katoh and Standley 2013). The aligned dataset was adjusted manually using BioEdit when necessary. The phylogenetic trees were in-

\*Corresponding author, e-mail: [cwn@mail.nmns.edu.tw](mailto:cwn@mail.nmns.edu.tw)



**Fig. 1.** *Mycena diplazii*. A. Basidiospores. B. Basidia. C. Cheilocystidia. D. Pileipellis. E. Disc hairs. Bars = 10  $\mu$ m.

ferred from Maximum Likelihood (ML) and Bayesian Inference (BI) methods using RAxML (Stamatakis 2014) and MrBayes v. 3.2.6. (Ronquist et al. 2012) at the CIPRES Science Gateway (<http://www.phylo.org/>), respectively. The best-fit model was selected by jModeltest v. 0.1 (Posada 2008) based on the Bayesian information criterion (BIC). Bootstrap (BS) values were calculated after 1000 replications in ML analysis. For BI analysis, 10 million generations were initiated from random starting trees and sampled every 1000 generations. The first 2500 trees were discarded as burn-in, and the Posterior Probability (PP) values were calculated from the remaining trees. Since both BI and ML analyses yielded similar topologies, only the phylogram inferred from ML analysis was shown. Only supporting values of  $BS \geq 70$  and those of  $PP \geq 0.7$  were shown on nodes of the ML tree.

## Results

The ITS dataset consisted of 34 taxa of 945 sites including gaps after trimming. The best-fit model GTR + I + G was selected for both ML and BI analyses. The BI analysis was concluded while the average standard deviation of split frequencies reduced to 0.009888. In the phylogenetic tree (Fig. 4), *M. diplazii* clustered together with *M. substylobates* with strong statistical supports (BP:100%, PP:1). *Mycena subcyanocephala* clustered with *M. acicula* with a high PP value in BI analysis but without a significant support in ML analysis.

## Taxonomy

*Mycena diplazii* W. N. Chou, *sp. nov.*

Figs. 1, 3A–E

Mycobank No: MB 830232.

**Holotype.** TAIWAN. Taichung: National Museum of Natural Science, on stem of living *Diplazium esculentum*, W. N. Chou, Oct. 9. 2013, CWN 10507 (TNM F0032107), sequences of ITS deposited at GenBank as MK400425.

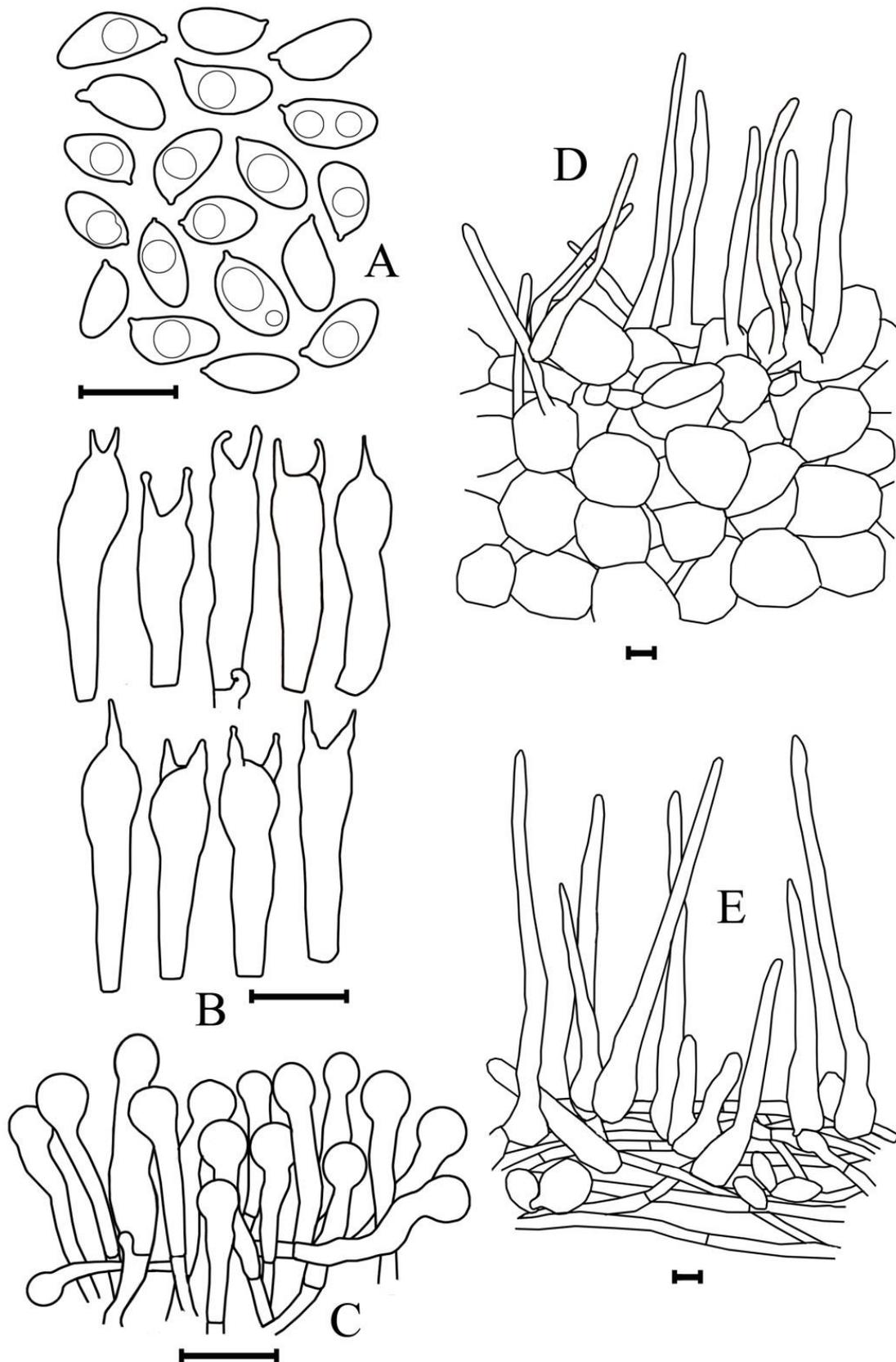
**Etymology.** *diplazii*, referring to the habitat on stem of *Diplazium esculentum*.

Pileus hemispherical to convex, 3–8 mm diam; surface with small and sparse white hirtose, gelatinous when moist, translucent-striate, gray-brownish to white. Lamellae white, broad, 14–21, adnate to almost free, forming a pseudocollarium, generally with two, occasionally with one series of lamellulae, smooth at edges. Stipe cylindrical, 10–15 × 0.6–0.8 mm, whitish, translucent, smooth or finely pubescent, with downy radially striate whitish basal disc, 1.5–2 mm wide. Flesh thin, without odor; taste not determinable.

Basidiospores broadly elliptic, 8–10 × 6–7 μm, hyaline, smooth, with or without droplets, amyloid. Basidia clavate, 20–30 × 8–10 μm, with 4 sterigmata. Cheilocystidia cylindrical-clavate, 25–40 × 6–9 μm, with digitiform outgrowths at apex. Pileipellis of branched, gelatinized hyphae, with brush-like outgrowths at apex, with occasional exserted, thorn-like elements, 60–100 × 25–35 μm, hyaline. Disc hairs cylindrical to fusiform, 35–95 × 5–10 μm. Septa with clamps.

**Habitat.** On stem of living *Diplazium esculentum* (Vegetable Fern), solitary or scattered.

**Additional specimen examined (paratype).** TAIWAN. Taichung, National Museum of Natural Science, W. N. Chou, CWN 11039, Dec. 1. 2015 (TNM).



**Fig. 2.** *Mycena subcyanocephala*. A. Basidiospores. B. Basidia. C. Cheilocystidia. D. Pileipellis. E. Cortical layer of the stipe. **Bars** = 10  $\mu$ m.

**Notes.** *Mycena diplazii* has a well-defined basal disc and distinct pseudocollarium, thus belonging to the section *Basipedes*. *Mycena stylobates* resembles *M. diplazii* in cheilocystidia and pileipellis but differs from the latter by having cylindrical-elliptic basidiospores and growing on needle litter, leaves, and branchlets. *Mycena bulbosa* has a similar basal disc but differs from *M. diplazii* by having inamyloid basidiospores and growing on dead stems of *Juncus* and *Carex* (Smith 1947, Amy 1981, Moser 1983, Singer 1986, Imazeki and Hongo 1987, Breitenbach and Kränzlin 1991). *Mycena diplazii* is easy to recognize by its habitat and basal discs.

The phylogenetic analyses suggested that *M. substylobates* is the closest species to *M. diplazii* (Fig. 4). These two species are similar in appearance with white fruiting bodies and a hirsute basal disc. However, *M. substylobates* has a smaller pileus (1.5–4.5 mm diam) and a bulbous base in stipe, smaller basidia (15–24 × 6–8 µm), obpyriform to spherical cheilocystidia with short excrescences on the entire surface, and non-gelatinous pileipellis with various shaped acanthocysts. Moreover, *M. substylobates* grew on dead Lauraceae twigs or leaves, while *M. diplazii* was collected from stems of living *Diplazium esculentum*.

***Mycena subcyanocephala*** W. N. Chou, *sp. nov.*  
Figs. 2, 3F–I  
MycoBank No: MB 830233.

**Holotype.** TAIWAN. Taichung: National Museum of Natural Science, on rotten wood, W. N. Chou, Nov. 4. 2013, CWN 10510 (TNM F0032108). GenBank: ITS = MK400426.

**Etymology.** *subcyanocephala*, referring to the

light blue pileus.

Pileus hemispherical to convex, 5–10 mm diam; surface white, densely tomentulose, hygrophanous when moist, obliterate striate, blue when young, fading to light blue or whitish at maturity. Lamellae white, broad, 16–19, adnexed, generally with two or occasionally with three series of lamellulae, white-granulose at edges. Stipe cylindrical, 10–25 × 0.7–1 mm, whitish-yellow, tomentulose, densely covered with whitish mycelium at base. Flesh thin, odor and taste not determinable.

Basidiospores elliptic, 8–11 × 4–6 µm, hyaline, smooth, with or without droplets, inamyloid. Basidia cylindrical-clavate, 20–30 × 8–10 µm, with 2 sterigmata. Cheilocystidia cylindrical-fusiform, 15–24 × 4–6 µm, with a smooth rounded apex. Pileipellis of spherical to clavate cells, with pileocystidia at apex, cylindrical, 50–100 µm long. Caulocystidia of cortical layer at the stipe apex cylindrical to subulate, up to 150 µm long. Septa with clamps.

**Habitat.** On rotted wood, scattered.

**Notes.** *Mycena subcyanocephala* has a pubescent pileus, pubescent stipe with fibrils at base, and smooth, clavate cheilocystidia, thus belonging to the section *Spinosaes*. *Mycena rasada* also has round-headed cheilocystidia but differs in having narrower basidiospores. *Mycena setulipes* also has a rooting stipe base but its cheilocystidia are narrower, up to 1.5 µm diam. *Mycena variicystis* differs from *M. subcyanocephala* by having a discoid stipe base and cylindrical cheilocystidia with numerous excrescences at apex; *M. setigera* differs by lacking caulocystidia; *M. paediscula* differs by having a



**Fig. 3.** A–E. *Mycena diplazii*. A. Fruiting bodies. B. Pileus. C. Lamellae and disc. D. Lamellae and lamellulae. E. Discs. F–I. *Mycena subcyanocephala*. F–G. Fruiting bodies. H. Pileus. I. Lamellae and lamellulae.

pale brown stipe base and lageniform cheilocystidia; *M. dermatogloea* differs by having an orange stipe base and shorter basidiospores, 7–9 µm in length; and *M. mridula* differs by having cylindrical cheilocystidia with short excrescences at apex (1–5 × 0.5–1 µm) (Métrod 1949, Desjardin et al. 2003, Boonpratuang 2009, Aravindakshan and Manimohan 2013). *Mycena subcyanocephala* is easy to recognize by having cheilocystidia with a smooth rounded head, inamyloid basidiospores, 2-sterigmate basidia, and a tomentulose, blue to light blue pileus.

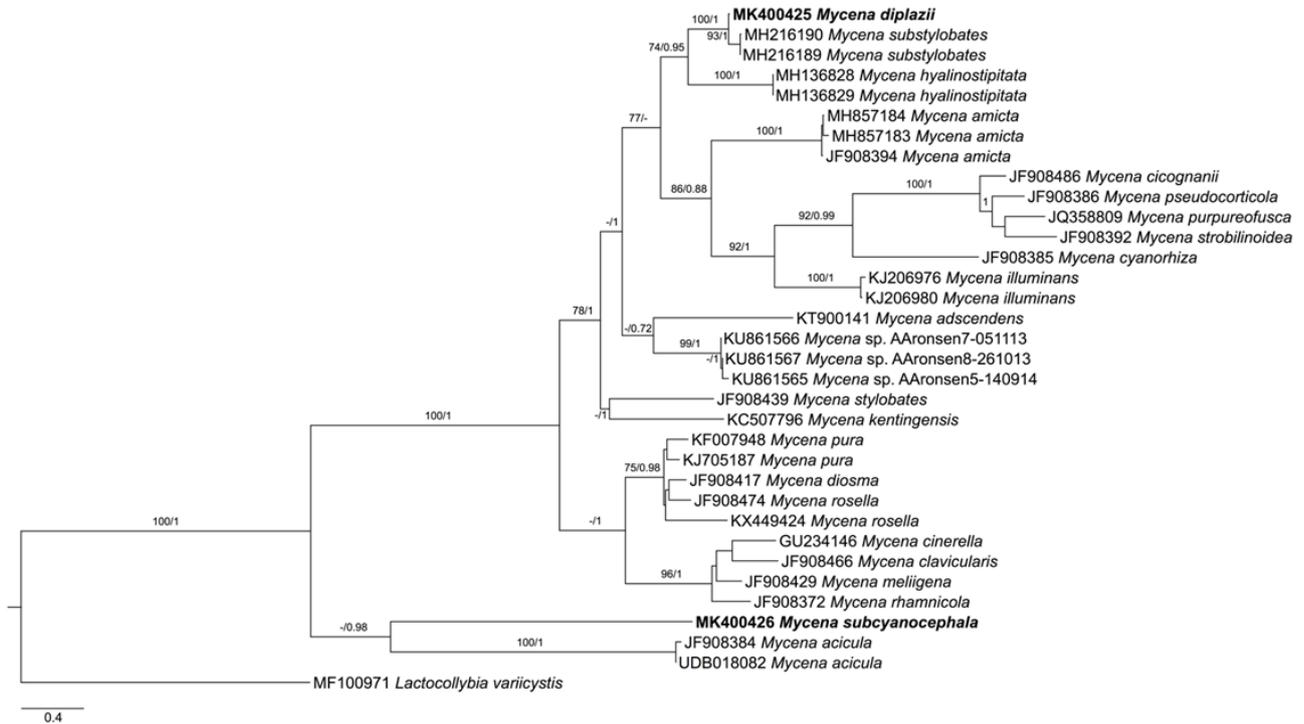
Species of *Mycena* in section *Spinosa* were mostly found in Southeast and South Asia. The discovery of *M. subcyanocephala* in Taiwan indicates that members of this section can also be found in subtropical East Asia.

### Acknowledgments

We are grateful to Ms. Y.P. Chen for laboratory works, and Dr. Y.Z. Wang, National Museum of Natural Science, Taiwan, for reading the manuscript.

### References

- Amy M. 1981. Trial key to the mycenoid species in the Pacific Northwest. Pacific Northwest Key Council.
- Aravindakshan DM, Manimohan P. 2013. A new section and two new species of *Mycena*. *Mycosphere* 4:930–935.
- Boonpratuang T. 2009. *Mycena variicystis*, a new spinose species from Phru Toh Daeng Peat Swamp in Thailand. *Mycotaxon* 109: 185–188.
- Breitenbach J, Kränzlin F. 1991. *Fungi of Switzerland* vol. 3 Boletes and Boletaceae. *Mycologia Luzern*, 361 pp.
- Chang YY, Ju YM. 2017. Small agarics in Taiwan: *Mycena albopilosa* sp. nov. and *Gloiocephala epiphylla*. *Botanical studies* 58:19, 1-11.
- Desjardin DE, Oliveira AG, Stevani CV. 2008. Fungi bioluminescence revisited. *Photochemical & Photobiological Sciences* 7: 170–182.
- Desjardin DE, Boonpratuang T, Hywel-Jones N. 2003. New spinose species of *Mycena* in sections *Basipedes* and *Polyadelphia* from Thailand. *Fungal Divers* 12:7–17.
- Hall TA. 1999 BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. *Nucl Acids Symp Ser* 41:95–98.
- Huang CL, Wang YZ. 2011. New records of endophytic fungi associated with the Araucariaceae in Taiwan. *Collection and Research* 24:87–95.
- Imazeki R, Hongo T. 1987. *Colored illustrations of mushrooms of Japan* vol. I. Hoikusha Publ. Co., Ltd. 325 pp.
- Katoh K, Standley DM. 2013. MAFFT multiple sequence alignment software version 7: improvements in performance and usability. *Molecular biology and evolution* 30:772–780.
- Kuo M. 2010. Mycenoid mushrooms retrieved from the MushroomExpert.Com. Web site: <http://www.mushroomexpert.com/myceno id.html>
- Singer F. 1986. *The agaricales in modern taxonomy*. Koeltz scientific books, 981 pp.
- Métrod G. 1949. *Les Mycènes de Madagascar*:



**Fig. 4.** Maximum likelihood phylogeny inferred from ITS dataset. Branches are labeled with bootstrap support values (BS  $\geq$  70) and posterior probabilities (PP  $\geq$  0.7). New species are indicated in boldface.

(*Mycena*, *Corrugaria*, *Pterospora*). Laboratoire de cryptogamie du Museum national d'histoire naturelle, ? pp.

Moser M. 1983. Key to Agarics and Boleti (Polyporaceae, Boletales, Agaricales, Russulales). Roger Phillips Pub., 535 pp.

Posada D. 2008. jModelTest: phylogenetic model averaging. *Molecular Biology and Evolution* 25:1253–1256.

Ronquist F, Teslenko M, van der Mark P, Ayres D L, Darling A, Höhna S, Larget B, Liu L, Suchard MA, Huelsenbeck JP. 2012. MrBayes 3.2: efficient Bayesian phylogenetic inference and model choice across a

large model space. *Systematic biology* 61: 539–542.

Smith AH. 1947. North American species of *Mycena*. Univ. of Michigan press, 521 pp.

Stamatakis A. 2014. RAxML version 8: a tool for phylogenetic analysis and post-analysis of large phylogenies. *Bioinformatics* 30: 1312–1313.

Wei CL, Kirschner R. 2019. A new *Mycena* species with blue basidiomata and poroid hymenophore from Taiwan. *Mycoscience* 60:10–13.

## 兩種在臺灣發現的小菇屬新種

張瓊之<sup>1</sup>、周文能<sup>2\*</sup>

<sup>1</sup>中央研究院生物多樣性研究中心，臺北市南港區研究院路二段 128 號，臺灣

<sup>2</sup>國立自然科學博物館，臺中市館前路一號 404，臺灣

### 摘 要

本文報導在台灣發現的兩個小菇屬新種：雙蓋蕨小菇 (*Mycena diplazii*) 與近藍蓋小菇 (*Mycena subcyanocephala*)，兩新種皆分佈於台灣亞熱帶地區。本研究使用內轉錄間隔區 (internal transcribed spacers, ITS) 序列建構親緣關係樹，探討兩新種與相近種的關係。雙蓋蕨小菇隸屬於吸盤小菇節 (section *Basipedes*)，生長在過溝菜蕨 (*Diplazium esculentum*) 的活體上，鑑定特徵為菌柄基部明顯的吸盤狀構造及菌褶中心的假項圈狀 (pseudocollarium) 構造。近藍蓋小菇隸屬於針毛小菇節 (section *Spinosa*)，生長於腐木，鑑定特徵為子實體表面具有細絨毛、淡藍色菌蓋、薄壁的菌蓋囊狀體與菌柄囊狀體、光滑頭狀緣囊狀體、無類澱粉反應的擔孢子及雙孢的擔子。包含本研究的兩個新種，臺灣小菇屬種數增加到 23 種。

**關鍵詞：**針毛小菇節、柄吸盤小菇節、內轉錄間隔區、分類學