



## *Phallus haitangensis*, a new species of stinkhorn from Yunnan Province, China

HUILI LI<sup>1,2,3,6</sup>, XUELAN MA<sup>1,3,5</sup>, PETER E. MORTIMER<sup>1,3\*</sup>, SAMANTHA C. KARUNARATHNA<sup>1,3</sup>, JIANCHU XU<sup>1,3</sup> & KEVIN D. HYDE<sup>1,2,3,4</sup>

<sup>1</sup>Key Laboratory of Economic Plants and Biotechnology, Kunming Institute of Botany, Chinese Academy of Sciences, 132 Lanhei Road, Kunming 650201, China

<sup>2</sup>Center of Excellence in Fungal Research, Mae Fah Luang University, 57100 Chiang Rai, Thailand

<sup>3</sup>World Agroforestry Centre, East and Central Asia, 132 Lanhei Road, Kunming 650201, China

<sup>4</sup>Mushroom Research Foundation, 128 M.3 Ban Pa Deng T. Pa Pae, A. Mae Taeng, Chiang Mai 50150, Thailand

<sup>5</sup>University of Chinese Academy of Science, Beijing 100049, China

<sup>6</sup>School of Science, Mae Fah Luang University, Chiang Rai 57100, Thailand

\*Corresponding author: [peter@mail.kib.ac.cn](mailto:peter@mail.kib.ac.cn)

### Abstract

Four specimens of *Phallus* were collected during surveys in a *Pinus armandii* forest in Baoshan, Yunnan Province, China. Macro- and micro-characteristics, together with Internal Transcribed Spacer (ITS) sequence data, showed that the four specimens belong to a new species, here named *Phallus haitangensis*. The ITS phylogenetic analyses, morphological descriptions, color photographs, and line drawings are provided, and compared with closely related species in the genus.

**Keywords:** Phallaceae, phylogeny, taxonomy

### Introduction

The genus *Phallus* Junius ex L. is classified under the family Phallaceae, order Phallales (Index Fungorum 2016). Species of the genus *Phallus* are typically characterized by a foetid odour and a bell-shaped or nearly half-rounded receptacle, an erect and sponge-like pseudostipe, a saccate volva with rhizomorphs, and sometimes the presence of a skirt-like structure named an indusium (Arora 1986, Liu 2005). *Phallus* species are used in China for food (e.g., *Dictyophora duplicata* (Bosc) E. Fish. (1888: 6), *Phallus echinovolvens* (M. Zang, D.R. Zheng & Z.X. Hu) Kreisel (1996: 277), *P. indusiatus* Vent. (1798: 520), *D. multicolor* Berk. & Broome (1883: 65), *P. rubrovolvens* (M. Zang, D.G. Ji & X.X. Liu) Kreisel (1996: 280) and *P. flavocostatus* Kreisel (1996: 278), *P. impudicus* L. (1753: 1178)) (Dai *et al.* 2010) and medicine (e.g., *D. duplicata*, *P. indusiatus*, *D. multicolor*, *P. impudicus* and *P. rubicundus* (Bosc) Fr. (1823: 284)) (Dai & Yang 2008). *Phallus indusiatus* is an example of a medicinal species from this genus, with antioxidant and antimicrobial properties (Oyetayo *et al.* 2009).

To date, the genus *Phallus* contains 31 species, excluding synonyms, on the basis of 107 records in Species Fungorum (Index Fungorum 2016). Known *Phallus* species of the world are widely distributed in different vegetation and climate types, for examples, grassland, bamboo forest or broad-leaved forest from tropical, subtropical or temperate areas (Calonge *et al.* 2002, Kreisel & Hausknecht 2002, Baseia *et al.* 2003, Liu 2005, Calonge *et al.* 2008, Kasuya 2008, Desjardin *et al.* 2009, Li *et al.* 2014). Twenty six species of *Phallus* have been recorded in China as *Phallus* and *Dictyophora* including varieties and orphan species (Liu 2005). Li *et al.* (2008) indicated that abundant resources of Phallaceae were presented in Yunnan Province, where thirteen species have been recorded. In this study, all specimens were collected in conifer forests in Yunnan Province.

Molecular phylogenetic methods have been widely applied in basidiomycete studies, and have elucidated several taxonomic issues (Drehmel *et al.* 1999, Zhang *et al.* 2004, Dentinger *et al.* 2010, Zhao *et al.* 2011, Karunarathna *et al.* 2014; Thongklang *et al.* 2014; Gui *et al.* 2015; Liu *et al.* 2015; Chen *et al.* 2016; Zhao *et al.* 2016). However, only a few studies (Li *et al.* 2014, Adamčík *et al.* 2015) have incorporated phylogenetic analyses in their taxonomic descriptions of the genus *Phallus*. Although the taxonomic position of *Phallus* and *Dictyophora* is still controversial, this study follows the taxonomic system of Dring (1964), who advocated that the genus *Dictyophora* should be merged

with *Phallus*. Li *et al.* (2014) also provided phylogenetic data to show that the two genera should be treated as *Phallus*. In the present paper, we aim to introduce a new species from montane, subtropical, pine forests in Yunnan Province, China, with support from morphological and phylogenetic analyses, and to provide evidence for the taxonomic position of *Phallus* and *Dictyophora*.

## Materials and Methods

### Collecting site and morphological study

Four specimens of *Phallus* (Table 1) were collected in the forests surrounding Haitang Village, Baoshan County, Yunnan Province, China (25.234°N, 99.290°E), whose locations are KU705381 (25.275°N, 99.299°E), KU5382 (25.266°N, 99.300°E), KU705383 & KU705384 (25.267°N, 99.305°E). These forests are dominated by *Pinus armandii* Franch. (1884: 285). Photographs of the fresh basidiocarps were taken *in situ* and the number of basidiocarps, odor, forest type, substrate type, location and date were recorded. The fresh basidiocarps were wrapped in aluminum foil and taken to the laboratory where they were described, placed in a food dryer (Shanghao FD-61WHC) at 40°C for 15 h until they were completely dehydrated and then stored in sealed, labeled plastic bags. Color terms used in the description were according to Kornerup and Wanscher (1978). All herbarium specimens were deposited in the Herbarium of the Kunming Institute of Botany (HKAS), Chinese Academy of Science, China. Facesoffungi numbers and Index Fungorum numbers were obtained as detailed in Jayasiri *et al.* (2015) and Index Fungorum (2016).

**TABLE 1.** Taxa information and GenBank accession numbers of *Phallus* and *Dictyophora* specimens used in the molecular phylogenetic analyses.

Species name	GenBank no.	Source	Sequence	Length
<i>D. phalloidea</i> Lév. (1809: 88)	AF324162	GenBank	ITS	619bp
<i>P. atrovolutus</i> Kreisel & Calonge (2005: 5–8)	KP012745	GenBank	ITS	617bp
<i>P. cinnabarinus</i> (W.S. Lee) Kreisel (1996: 278) ( <i>D. cinnabarina</i> W.S. Lee (1957: 156))	KJ764821	Cabral <i>et al.</i> (2015)	ITS	620bp
<i>P. echinovolutus</i> ( <i>D. echinovoluta</i> M. Zang, D.R. Zheng & Z.X. Hu (1988: 146))	AF324168	GenBank	ITS	618bp
<i>P. haitangensis</i>	KU705381	This study	ITS	422bp
<i>P. haitangensis</i>	KU705382	This study	ITS	638bp
<i>P. haitangensis</i>	KU705383	This study	ITS	639bp
<i>P. haitangensis</i>	KU705384	This study	ITS	640bp
<i>P. hadriani</i> Vent. (1798: 517)	KF481956	Moreno <i>et al.</i> (2013)	ITS	584bp
<i>P. impudicus</i> ( <i>D. duplicata</i> var. <i>obliterata</i> Malençon (1957: 66))	KR673719	Kim <i>et al.</i> (2015)	ITS	587bp
<i>P. indusiatus</i> ( <i>D. indusiata</i> (Vent.) Desv. (1809: 92))	JN182874	GenBank	ITS	616bp
<i>P. mengsongensis</i> H.L. Li, L. Ye, Mortimer, J.C. Xu & K.D. Hyde (2014: 94)	KF052624	Li <i>et al.</i> (2014)	ITS	614bp
<i>P. multicolor</i> (Berk. & Broome) Cooke (1882: 57)	KP012762	GenBank	ITS	632bp
<i>P. rubrovolutus</i> ( <i>D. rubrovoluta</i> M. Zang, D.G. Ji & X.X. Liu (1976: 11))	KF939505	Lu <i>et al.</i> (2014)	ITS	613bp
<i>P. rugulosus</i> Lloyd (1908: 402)	AF324170	GenBank	ITS	570bp
<i>P. serrata</i> H.L. Li, L. Ye, P.E. Mortimer, J.C. Xu & K.D. Hyde (2014: 98)	KF052623	Li <i>et al.</i> (2014)	ITS	587bp
<i>P. ultraduplicatus</i> X.D. Yu (2015: 147)	KJ591584	Adamčík <i>et al.</i> (2015)	ITS	615bp
<i>Mutinus caninus</i> (Huds.) Fr. (1849: 434)	GQ981513	GenBank	ITS	661bp

Hand sections of dried specimens were made under a dissecting microscope (Motic SMZ-140 Series), and mounted in 3%–5% KOH for examination. All micro-morphological characteristics were observed under the objective lenses of 200×, 400× and 1000× of a Nikon compound microscope (Nikon Model Eclipse Ni-U). Basidiospore size, colour, shape, and the cells or hyphae of the pseudostipe, indusium and volva were recorded and photographed. In addition, line drawings of the volva hyphae, and mature fruiting bodies were made. The range of values of basidiospore sizes in side view was calculated from at least 25 basidiospores. The value range of the cells of the pseudostipe, indusium and hyphae of the volva were calculated from at least 20 hyphae or cells. The photographs and drawing plates were edited in Adobe Photoshop CS3.

#### *DNA extraction, PCR amplification and sequencing*

Total deoxyribonucleic acid (DNA) of the four specimens was extracted using the Biospin Fungus Genomic DNA Extraction Kit (Bioer Technology Co., Ltd., Hangzhou, P.R. China). The primers ITS4 and ITS5 were used to amplify the complete ITS region (Gardes & Bruns 1993). For PCR amplification, 25 µl of the PCR reaction mixture contained 10 mM 2Mix 10 µl, 1 µM ITS5 0.35 µl, 1 µM ITS4 0.35 µl, 50 ng/µl DNA 0.6 µl, ddH<sub>2</sub>O 13.7 µl. The PCR thermal profile was set according to Douanla-Meli *et al.* (2005) and Li *et al.* (2014) with the following modifications: an initial denaturation at 94°C for 3 min, followed by 35 cycles consisting of denaturation at 94°C for 40s, annealing at 55°C for 45s, and extension at 72°C for 1 min, and a final elongation step of 7 min at 72°C. To check the PCR products, 1% agarose gel electrophoresis for 30 minutes at 220V was used. All PCR products were sent to Shanghai Majorbio Bio-Pharm Technology Co., Ltd. for purification and sequencing.

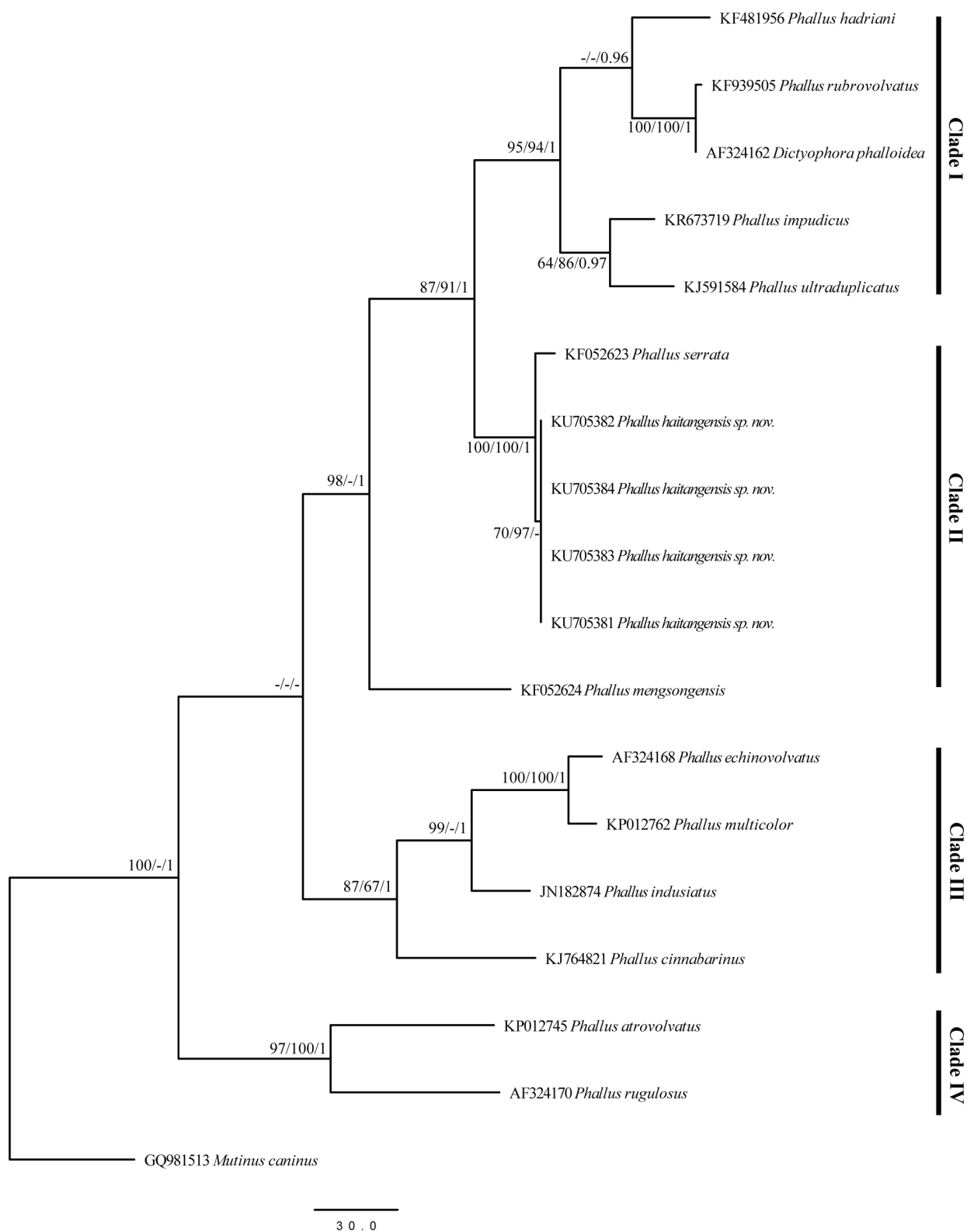
#### *Sequence alignment and phylogenetic analyses*

Sequences of the four *Phallus* specimens were assembled using SeqMan (DNA Star) (DNASTRAT Lasergene 13) (Burland 1999) and Bioedit (Hall 1999). The ITS dataset includes 17 taxa from the genus *Phallus* as the ingroup, four new sequences from our samples, and 13 obtained from GenBank (Table 1). *Mutinus caninus* was chosen as the outgroup taxon (Fig. 1). These sequences were aligned in ClustalX Version 2.1 (Thompson *et al.* 1997), and manual adjustments were made to improve the alignments. The phylogenetic analysis was carried out using PAUP 4.0 software (Swofford 2001). Data were analyzed with random addition sequence, unweighted parsimony and gaps were treated as missing data. The maximum parsimony trees with bootstrap values were obtained by using the heuristic search option with 1,000 random taxa addition and tree bisection and reconnection (TBR) as the branch-swapping algorithm. The ML analysis was performed using raxmlGUI v.0.9b2 with the model “GTRGAMMA” (Stamatakis 2006; Silvestro & Michalak 2010), while Bayesian analysis was conducted with MrBayes v. 3.1.2 (GTR+I+Gmodel) (Huelsenbeck & Ronquist 2001) to evaluate Posterior probabilities (PP) (Rannala & Yang 1996; Zhaxybayeva & Gogarten 2002) by Markov Chain Monte Carlo sampling (BMCMC). Phylogenetic trees were sampled every 100<sup>th</sup> generation in 455,000 generations from the running of six simultaneous Markov chains. The first 980 trees which contained the burn-in phase of the analyses were discarded. The remaining 2,560 trees were used to calculate the posterior probabilities (PP) in the majority rule consensus tree (Liu *et al.* 2011). The best tree was viewed in FigTree and edited in Adobe Illustrator CS5.

## Results

#### *Phylogenetic analyses*

The equally weighted maximum parsimony tree had a consistency index (CI) of 0.683, a retention index (RI) of 0.681, a rescaled consistency index (RC) of 0.465, and a homoplasy index (HI) of 0.317. There were 615 characters in the present analysis, of which 253 were constant, 98 were parsimony-uninformative and 264 were parsimony-informative (Fig. 1). The phylogenetic tree revealed four clades (Fig. 1): Clade II, with relatively high bootstrap support (70 MP and 97 ML), comprised four *Phallus* species, derived from the four field collections, belonging to *P. haitangensis* and closely related to *Phallus serrata* with very high support (100 MP, 100 ML and 1.00 PP).



**FIGURE 1.** Maximum parsimony phylogram based on ITS rDNA sequence data showing the phylogenetic position of *Phallus haitangensis* with selected *Phallus* species. Bootstrap support values for maximum parsimony (MP, left), maximum likelihood (ML, middle) higher than 50% and Bayesian posterior probabilities (PP, right) greater than 0.95 are provided above the nodes. The tree is rooted with *Mutinus caninus*. The bar represents the distance between species.

## Taxonomy

*Phallus haitangensis* H.L. Li, P.E. Mortimer, J.C. Xu & K.D. Hyde, *sp. nov.* Fig. 2

*Index Fungorum number*: IF552109; *Facesoffungi number*: FoF: 02184

**Etymology**:—The species epithet “haitangensis” refers to the place where the type species was collected.

**Holotype**:—CHINA. Yunnan Province: Baoshan, Haitangwa Village, 25.266°N, 99.300°E, alt. 2473m, 8 April 2013, Huili Li (HKAS 88193).

**Diagnosis**:—Receptacle strongly reticulate, golden orange, with a prominent opening at the apex. Pseudostipe creamy white, with a well-developed and light orange indusium, and a sac-like volva with rhizomorphs.

**Description**:—Immature fruiting body not seen. Expanded basidioma up to 130–165 mm high, unbranched, with an indusium flaring out from beneath the receptacle, a stipe-like pillar and a volva with thin rhizomorphs. Receptacle 30–35 mm high × 25–50 mm wide, bell shape to umbrella shape, dark orange (5B7) to golden yellow (5B7), strongly reticulated, gold yellow (5B7) ridges, yellowish brown (5E5–7) sticky gleba in the pits, with a prominent hole at the apex with 1–8 mm diameter. Receptacle margin is lightly upturned. Pseudostipe creamy white (4A1), 100–132 mm high × 8–11/15–20/18–22 mm wide (apex-middle-base), nearly equal or lightly tapering upwards, fragile or soft, spongiform, holes in pseudostipe is 1–10 mm diameter. Indusium 85–95 mm long, 80–155 mm broad, pale orange (5A3) to light orange (5A4–5), well-developed, at first contracted under the edge of the receptacle, later expanding almost touching the ground. The holes of the indusium are nearly round to polygonal, 4–35 mm diameter and 1–4 mm thick, smooth edges. Volva present at base of pseudostipe, 30–140 mm high and 20–45 mm thick, yellowish white (4A2), smooth surface, attached to the substrate by a pinkish white (13A2) rhizomorph. Odor foetid.

**Basidiospores** [100/4/4] 2.8–4.2 (3.4±0.3) µm length, 1.1–2.6 (1.7±0.3) µm width, L=3.4 µm, W=1.7 µm, Q=2, elongate or cylindrical, hyaline, smooth surface. Cells of pseudostipe 15–38.8 (28±6.2) µm diameter, bubble-like or foam-like, hyaline, smooth surface. Cells of indusium 21.2–41.6 (30.1±5.7) µm diameter, bubble-like or foam-like, hyaline, smooth surface. Hyphae of volva 1.8–3.8 (2.6±0.5) µm width, tubular and branched, septate with clamp-connection, hyaline, smooth surface (Fig.3).

**Habitat and Distribution**:—solitary or scattered on soil with decaying litter under *P. armandii* trees, Yunnan Province, China.

**Material examined**:—CHINA. Yunnan Province: Baoshan, Haitangwa Village, 25.266°N, 99.300°E, alt. 2473m, 8 April 2013, *Huili Li* (HKAS 88193, *holotype*); *Ibid.*, Haitang Village, 4 August 2013, *Huili Li* (HKAS 88191, *paratype*); *Ibid.* 26 August 2014, *Huili Li* (HKAS 88197, *paratype*); *Ibid.* 2 September 2014, *Huili Li* (HKAS 88199, *paratype*).

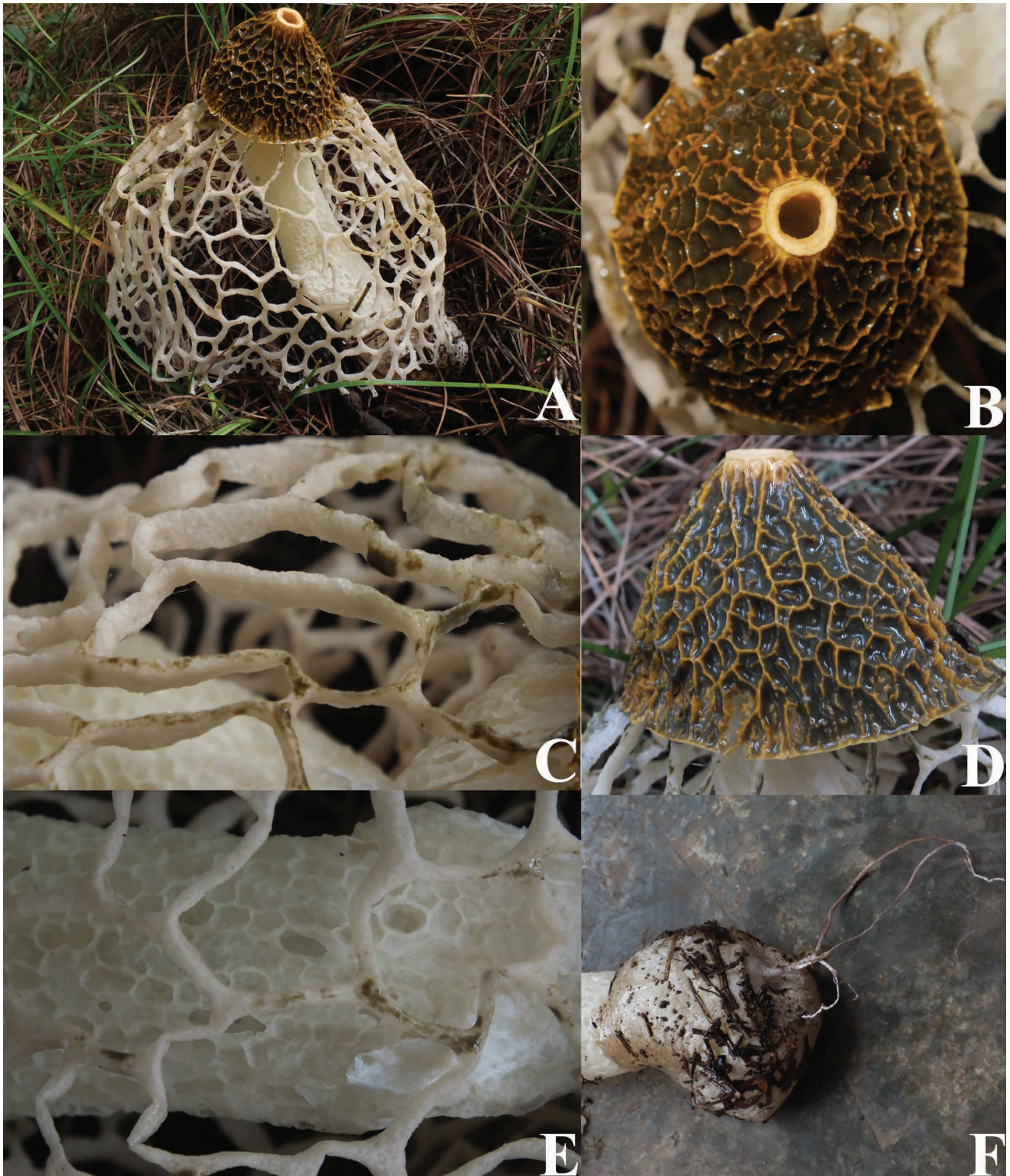
**Notes**:—The main distinguishing characteristics of *Phallus haitangensis* are the golden orange receptacle that is strongly reticulate, with a somewhat prominent apex, which has a light orange opening, and a light orange indusium with rounded to polygonal meshes without serrated margins (Fig. 2), and a creamy white spongy pseudostipe which tapers upwards. The results of the phylogenetic tree showed that *P. haitangensis* grouped with *P. serrata*, *P. mengsongensis* and *P. cinnabarinus*, however *P. serrata* has a white and half-egg receptacle, a white indusium with serrated margins of the holes, and larger basidiospores (Li *et al.* 2014); *P. mengsongensis* has small fruiting bodies without an indusium (Li *et al.* 2014); the receptacle of *P. cinnabarinus* is cinnabar-red and the pseudostipe is pale cinnabar-red and shorter than that in *P. haitangensis*, and the indusium of *P. cinnabarinus* is also cinnabar-red above to paler below, and the basidiospores of *P. cinnabarinus* are brownish green (Lee 1957, Hemmes & Desjardin 2009) (Table 2).

Some indusiate species are morphologically similar to *P. haitangensis*, however, the indusium of *P. impudicus* (= *P. impudicus* var. *pseudoduplicatus* O. Andersson (1989: 233)) is pure white and up to 1/3 to 2/3 of the pseudostipe length; the indusium of *P. indusiatus* is longer than that of *P. haitangensis* with a white receptacle (Kreisel & Hausknecht 2002, Das *et al.* 2007, Li *et al.* 2008, Dash *et al.* 2010) (Table 3); *P. indusiatus* var. *roseus* Lloyd (1909) (Kreisel & Hausknecht 2002) has a white or cream or yellowish receptacle with a light pinkish indusium; *P. haitangensis* has a taller fruiting body (130mm–165mm high) than *P. indusiatus* (= *P. indusiatus* f. *citrinus* K. Das, S. K. Singh & Calonge (2007: 136)) (75mm–110mm high) and *P. flavidus* Kreisel & Hauskn (2009: 152) (50mm–80mm) (Table 3); the pseudostipe of *P. multicolor* is yellowish white (Kreisel & Hausknecht 2002, Li *et al.* 2008, Dutta *et al.* 2012) (Table 3); the orange or chrome yellow indusium of *P. multicolor* var. *laeticolor* D. A. Reid (1976) (Kreisel & Hausknecht 2002) up to 1/2 to 3/4 of pseudostipe length (Kreisel & Hausknecht 2002); *P. luteus* (Liou & L. Hwang 1936) T. Kasuya (2008: 9) has an orange to yellowish white receptacle and a chrome yellow or orange-yellow indusium (Kreisel & Hausknecht 2002).

*Phallus haitangensis* should also be compared with *P. echinovolvatus*, however, the receptacle of *P. echinovolvatus*



is whitish yellow under a dark brown gleba, the indusium is white, and the volva is echinulated (Calonge 2005, Li *et al.* 2008, Li *et al.* 2014). *Phallus haitangensis* closely resembles *P. duplicatus* Bosc (1811: 86), but this species has a white receptacle and indusium, an annulus under the indusium, and the basidiospores are light greenish. (Li *et al.* 2008, Cortez *et al.* 2011, Kibby & McNeil 2012) (Table 2).

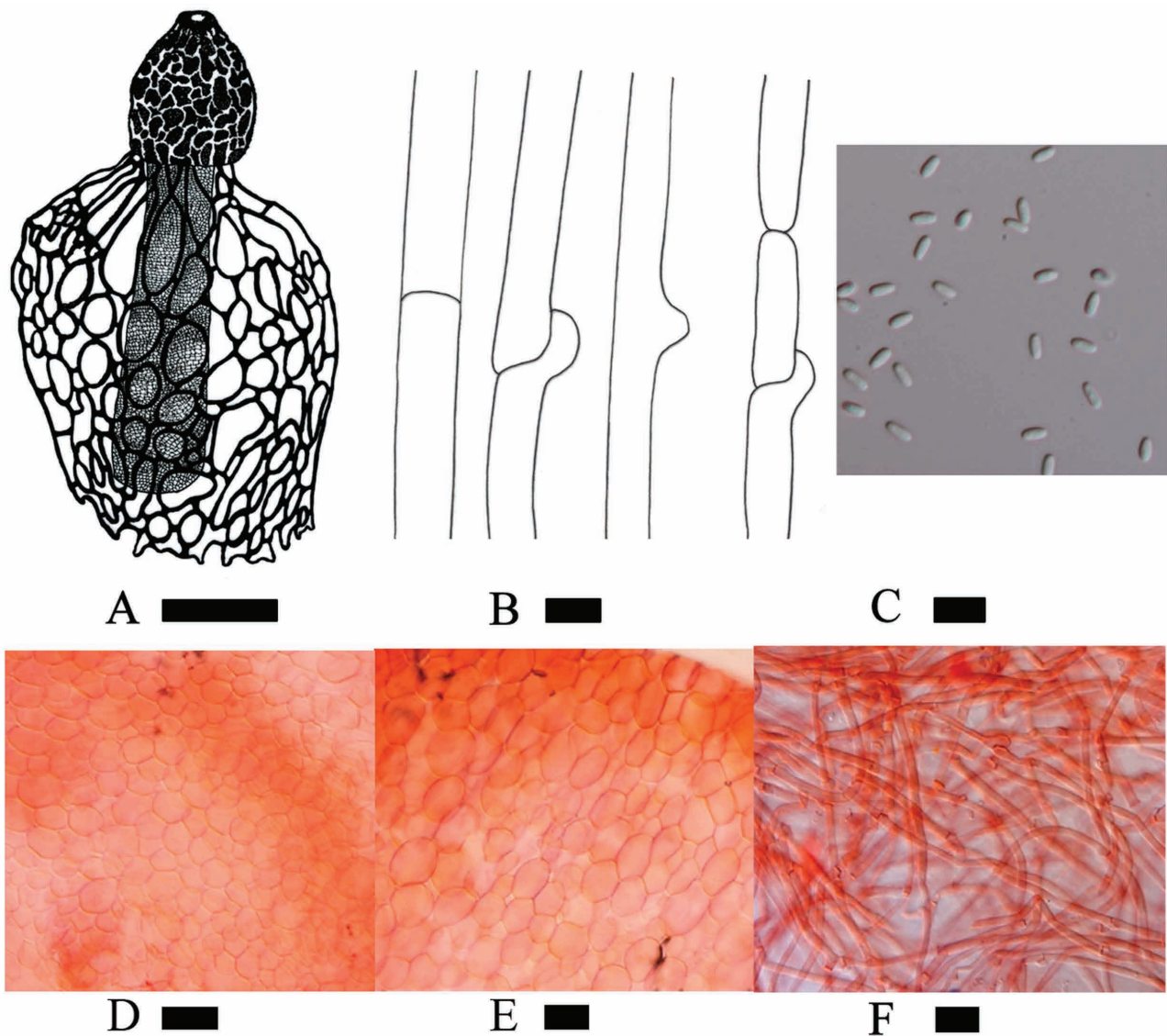


**FIGURE 2.** Plate of the *Phallus haitangensis* (HKAS 88193). A: Mature fruiting body of *P. haitangensis*, B: Hole at the apex of the receptacle; C: Indusium; D: Receptacle; E: Pseudostipe; F: Volva and rhizomorph. Photos by Huili Li.

TABLE 2. Synopsis of macro- and micro-characteristics of *Phallus haitangensis*, *P. serrata*, *P. mengsongensis*, *P. cinnabarinus*, *P. echinovolvatus*, and *P. duplicatus*. Abbreviations: H=Height, W=Width, Chars.=Characteristics.

Species name	<i>Phallus haitangensis</i> <i>sp. nov.</i>	<i>P. serrata</i>	<i>P. mengsongensis</i>	<i>P. cinnabarinus</i>	<i>P. echinovolvatus</i>	<i>P. duplicatus</i>
Receptacle shape	Bell-shape or umbrella	Half-egg	Egg-shape to elliptical or ovoid	Conical	Campanulated	Conical or campanulated
Receptacle size (H × W, mm)	30–35 × 25–50	40–42 × 34–35	17–20 × 9–11	25 × 30	25–30 × 25–30	35–40 × 40–45
Receptacle color	Gold orange	White	Greyish white or light grey	Cinnabar-red	Nearly white to yellow	White under dark brown gleba
Receptacle chars.	Strongly reticulated	Reticulated	Reticulated	Reticulated	Reticulated	Strongly rugose
Gleba color	Olive or greenish brown	Olive	Yellowish-brown	Dark brown	Olive to dark brown	Dark greenish brown
Indusium length (mm)	85–95	135–140	Absence	20–100	70–100	60–70
Indusium color	Golden orange or light orange	White	Absence	Brilliant cinnabar-red, paler	White	White to creamy white
Indusium chars.	Nearly round or polygonal holes	Round, nearly round or polygon hole with the serrated margin in hole	Absence	Netted, smaller nearer the margin	Polygonal holes	Nearly round to polygonal holes
Annulus	Absent	Absent	Absent	Absent	Absent	Present
Pseudostipe shape	Nearly equal or tapering upwards	Cylindrical	Nearly cylindrical	Tapering upward or tapering upwards	Cylindrical to fusiform	Cylindrical, tapering downwards
Pseudostipe size (H × W, mm)	100–132 × 15–20	150–155 × 150–200	94–142 × 3–5	70–130 × 8–20	90–150 × 20–30	150–200 × 35–40
Pseudostipe color	Creamy white	White	Pure white to pale orange	Cinnabar-red above, fading to paler below	Nearly white	White to creamy white
Pseudostipe chars.	Fragile or soft, spongiform	Hollow	Hollow	Hollow	Hollow	Sponge, hollow
Volva chars.	Non-echinulate	Non-echinulate	Non-echinulate	Non-echinulate	Echinulate	Non-echinulate
Spore shape	Elongate or cylindrical	Ellipsoid	Elongate or cylindrical	Obovate-ellipsoid or subclavate	Oval to ellipse	Ellipse to oblong
Spore size (µm)	2.8–4.2 × 1.1–2.6	4–5 × 2–3	3.5–5 × 1.5–2	3–4 × 1.6–2.2	3–4 × 1.3–2.0	3.5–4.5 × 1.3–1.7
Spore color	Hyaline	Hyaline	Hyaline	Brownish green	Light brownish green	Hyaline to light greenish to olive





**FIGURE 3.** Morphological characteristics of *Phallus haitangensis*. A. Line drawing of basidiocarp B. Line drawing of hyphae of volva C. Basidiospores D. Cells of pseudostipe E. Cells of indusium F. Hyphae of volva. Scale bars: A=20 mm, B=20  $\mu$ m, C=5  $\mu$ m, D=40  $\mu$ m, E=40  $\mu$ m, F=10  $\mu$ m. Tissues of the structures D, E and F were mounted in Congo Red for microscopic observation. Drawings by Samantha C. Karunaratna and photos by Huili Li.

## Discussion

In this study, we introduce *Phallus haitangensis* based on its unique macro- and micro- morphological characteristics together with the support of phylogenetic analyses. This species has a prominent opening at the apex, a strongly reticulate and golden orange receptacle, a creamy white pseudostipe, a well-developed net-like and light orange indusium without a serrated margin, and a sac-like volva with rhizomorphs. In the ITS phylogeny, this species was well separated from other *Phallus* species with high bootstrap support values (Fig.1), and the comparison with the morphologically similar taxa in *Phallus* showed *P. haitangensis* has unique characteristics (Table2), indicating that *P. haitangensis* is a distinct species within *Phallus*.

The phylogenetic tree (Fig.1) shows that *Phallus* species with and without indusium are mixed together. In Clade I, the species with indusium—*P. ultraduplicatus*, *P. rubrovolvatus*, *Dictyophora phalloidea* and the species without indusium—*P. impudicus* and *P. hadriani* were also grouped in a single clade. In Clade II, the species with indusium—*P. serrata* and *P. haitangensis* and the species without indusium—*P. mengsongensis* grouped in a single clade. In Clade IV, the species with indusium—*P. atrovolvatus* and the species without indusium—*P. rugulosus* grouped in a single clade. This supports the argument that *Dictyophora* should be merged with *Phallus* (Dring 1964). Similarly, Cabral



*et al.* (2012) also mentioned that the indusium should not be used as a characteristic to distinguish between genera of Phallaceae. Thus, we consider that *Dictyophora* should be a synonym of *Phallus* as we showed in our previous study (Li *et al.* 2014).

To date, most taxonomic studies of *Phallus* have been based on morphological observation and very few studies have used molecular analyses for their studies. This study used ITS data of new collections and all available ITS data of *Phallus* in the GenBank for phylogenetic analyses of *P. haitangensis*, so the molecular analyses should be used in *Phallus* taxonomy in future studies.

**TABLE 3.** Synopsis of macro- and micro-characteristics of *Phallus haitangensis*, *P. indusiatus*, *P. flavidus* and *P. multicolor*. Abbreviations: H=Height, W=Width, Chars.=Characteristics.

Species name	<i>Phallus haitangensis</i> <i>sp. nov.</i>	<i>P. indusiatus</i>	<i>P. flavidus</i>	<i>P. multicolor</i>
Receptacle shape	Bell-shape or umbrella	Conical or campanulate	Conical-truncate	Campanulate
Receptacle size (H × W, mm)	30–35 × 25–50	18–32 × 16–27	18–25 × 15–20	25–30 × 20
Receptacle color	Gold orange	Lemon-yellow, luteus, orange	Pallid orange	Lemon-yellow
Receptacle chars.	Strongly reticulated	Coarsely reticulated	Reticulated	Reticulated
Gleba color	Olive or greenish brown	Olive	Olivaceous brown	Olive-brown
Indusium length (mm)	85–95	100–120	50 (1/2 to 2/3 of pseudostipe length)	78
Indusium color	Golden orange or light orange	Yellowish white, pale luteus, salmon, light orange to orange	Cream to yellow	Lemon yellow to yellowish-orange
Pseudostipe shape	Nearly equal or tapering upwards	Cylindrical	Cylindrical	Narrow upward
Pseudostipe size (H × W, mm)	100–132 × 15–20	75–110 × 11–22	--	60–85 × 25–30
Pseudostipe color	Creamy white	White	Cream to yellowish	Yellowish-white
Pseudostipe chars.	Fragile or soft, spongiform	Hollow, spongy	Hollow, spongy	Hollow
Spore shape	Elongate or cylindrical	Ovoid to cylindrical	Cylindrical	Long-elliptical to nearly cylindrical
Spore size (µm)	2.8–4.2 × 1.1–2.6	3–4.4 × 1.2–2	2.8–3.6 × 1.5–1.8	3.94–4.33 × 1.77–1.97
Spore color	Hyaline	Hyaline	Yellowish	Hyaline

## Acknowledgements

We would like to thank CGIAR Research Program 6: Forest, Trees and Agroforestry, the Kunming Institute of Botany, Chinese Academy of Science (CAS) and the Chinese Ministry of Science and Technology, under the 12th 5-year National Key Technology Support Program (NKTSP) 2013BAB07B06 integration and comprehensive demonstration of key technologies on Green Phosphate-mountain Construction for providing the financial support for this study. Thailand Research Fund grant-Taxonomy, Phylogeny and biochemistry of Thai Basidiomycetes (BRG 5580009); the National Research Council of Thailand (NRCT), projects-Taxonomy, Phylogeny and cultivation of *Lentinus* species in northern Thailand (NRCT/55201020007), Mae Fah Luang University project-Taxonomy, Phylogeny and cultivation of *Lentinus* species in northern Thailand (MFU/54 1 01 02 00 48), and Thailand Research Fund grant- Taxonomy,

Phylogeny and biochemistry of Thai Basidiomycetes (BRG 5580009) are also thanked for supporting this study. Kevin D. Hyde thanks the Chinese Academy of Sciences, project number 2013T2S0030, for the award of Visiting Professorship for Senior International Scientists at Kunming Institute of Botany. The authors also would like to thank Jiayu Guo, Lei Ye, Xia Luo, Aseni Navoda Ediriweera, Youji Yang and Qiuxiang Yang for the contribution of fieldwork.

## References

- Adamčík, S., Cai, L., Chakraborty, D., Chen, X.H., Cotter, H.V.T., Dai, D.Q., Dai, Y.-C., Das, K., Deng, C., Ghobad-Nejhad, M., Hyde, K.D., Langer, E., Latha, K.P.D., Liu, F., Liu, S.-L., Liu, T., Lv, W., LV, S.-X., Machado, A.R., Pinho, D.B., Pereira, O.L., Prasher, I.B., Rosado, A.W.C., Qin, J., Qin, W.-M., Verma, R.K. Wang, Q., Yang, Z.-L., Yu, X.-D., Zhou, L.-W. & Buyck, B. (2015) Fungal Biodiversity Profiles 1–10. *Cryptogamie Mycologie* 36: 121–166.  
<http://dx.doi.org/10.7872/crym/v36.iss2.2015.121>
- Andersson, O. (1989) Stinksvampen, *Phallus impudicus*, i Norden. *Svensk bot. Tidskr.* 83: 219–241.
- Arora, D. (1986) *Mushrooms demystified*. Ten Speed Press, pp 1056.
- Baseia, I.G., Gibertoni & T.B., Maia, L.C. (2003) *Phallus pygmaeus*, a new minute species from a Brazilian tropical rain forest. *Mycotaxon* 85: 77–79.
- Burland, T.G. (1999) DNASTAR's Lasergene sequence analysis software. *Bioinformatics Methods and Protocols* 132: 71–91.  
<http://dx.doi.org/10.1385/1-59259-192-2:71>
- Cabral, T.S., Clement, C.R. & Baseia, I.G. (2015) Amazonian phalloids: new records for Brazil and South America. *Mycotaxon* 130: 315–320.  
<http://dx.doi.org/10.5248/130.315>
- Cabral, T.S., Marinho, P., Goto, B.T. & Baseia, I.G. (2012) *Abrachium*, a new genus in the Clathraceae, and Itajahya reassessed. *Mycotaxon* 119: 419–429.  
<http://dx.doi.org/10.5248/119.419>
- Calonge, F.D., Menezes de Sequeira, M., Freitas, T., Rocha, E. & Franquinho, L. (2008) *Phallus maderensis* sp. nov., found in Madeira, Portugal. *Boletín de la Sociedad Micológica de Madrid* 32: 101–104.
- Calonge, F.D. (2005) A tentative key to identify the species of *Phallus*. *Boletín de la Sociedad Micológica de Madrid* 29: 9–18.
- Calonge, F.D. & Kreisel, H. (2002) *Phallus minusculus* sp. nova from Tropical Africa. *Feddes Repertorium* 113: 600–602.  
<http://dx.doi.org/10.1002/fedr.200290007>
- Calonge, F.D., Kreisel, H. & Mata, M. (2005) *Phallus atrovolvatus*, a new species from Costa Rica. *Boletín de la Sociedad Micológica de Madrid* 29: 5–8.
- Chen, J., Parra, L.A., De Kesel, A.N.D.R.É., Khalid, A.N., Qasim, T., Ashraf, A., Bahkali, A., Hyde, K.D., Zhao, R. & Callac, P. (2016) Inter- and intra-specific diversity in *Agaricus endoxanthus* and allied species reveals a new taxon, *A. punjabensis*. *Phytotaxa* 252: 1–16.  
<http://dx.doi.org/10.11646/phytotaxa.252.1.1>
- Commonwealth Mycological Institute (various authors) (1951–1960) *Index of Fungi* 2: 342. Published quarterly by the Institute.
- Cooke, M.C. (1882) Australian fungi. *Grevillea*. 11: 57–65.
- Cortez, V.G., Baseia, I.G. & da Silveira, R.M.B. (2011) Two noteworthy *Phallus* from southern Brazil. *Mycoscience* 52: 436–438.  
<http://dx.doi.org/10.1007/s10267-011-0124-5>
- Dai, Y. & Yang, Z. (2008) A revised checklist of medicinal fungi in China. *Mycosystema* 27: 801–824.
- Dai, Y., Zhou, L., Yang, Z., Wen, H., Bau, T. & Li, T. (2010) A revised checklist of edible fungi in China. *Mycosystema* 29: 1–21.
- Das, K., Singh, S. & Calonge, F. (2007) Gasteromycetes of Western Ghats, India: I. A new form of *Phallus indusiatus*. *Boletín de la Sociedad Micológica de Madrid* 31: 135–138.
- Dash, P., Sahu, D., Sahoo, S. & Das, R. (2010) *Phallus indusiatus* Vent. & Pers. (Basidiomycetes)-a new generic record for Eastern Ghats of India. *Journal of Threatened Taxa* 2: 1096–1098.  
<http://dx.doi.org/10.11609/JoTT.o2305.1096-8>
- Dentinger, B.T., Ammirati, J.F., Both, E.E., Desjardin, D.E., Halling, R.E., Henkel, T.W., Moreau, P., Nagasawa, E., Soyong, K., Taylor, A.F., Watling, R., Moncalvo, J. & McLaughlin, D.J. (2010) Molecular phylogenetics of porcini mushrooms (*Boletus* section *Boletus*). *Molecular Phylogenetics and Evolution* 57: 1276–1292.  
<http://dx.doi.org/10.1016/j.ympev.2010.10.004>
- Desjardin, D.E. & Perry, B.A. (2009) A new species of *Phallus* from Sao Tome, Africa. *Mycologia* 101: 545–547.  
<http://dx.doi.org/10.3852/08-166>

- Douanla-Meli, C., Langer, E. & Calonge, F.D. (2005) *Geastrum pleosporus* sp. nov., a new species of Geastraceae identified by morphological and molecular phylogenetic data. *Mycological Progress* 4: 239–250.  
<http://dx.doi.org/10.1007/s11557-006-0127-3>
- Drehmel, D., Moncalvo, J.M. & Vilgalys, R. (1999) Molecular phylogeny of *Amanita* based on large-subunit ribosomal DNA sequences: implications for taxonomy and character evolution. *Mycologia* 1999: 610–618.  
<http://dx.doi.org/10.2307/3761246>
- Dring, D.M. (1964) *Gasteromycetes of west tropical Africa*. Vols. 98–109. Commonwealth Mycological Institute.
- Dutta, A.K., Chakraborty, N., Pradhan, P. & Acharya, K. (2012) Phallales of West Bengal, India. II. Phallaceae: *Phallus* and *Mutinus*. *Researcher* 4: 21–25.
- Franchet, A. & David, A. (1884) *Plantae Davidianae ex Sinarum imperio: pte. Plantes de Mongolie, du nord et du contre de la Chine* (Vol. 1). G. Masson, 285.
- Fries, E.M. (1849) *Summa Vegetabilium Scandinaviae Sectio posterior* 259–572: 381.
- Gardes, M. & Bruns, T.D. (1993) ITS primers with enhanced specificity for basidiomycetes-application to the identification of mycorrhizae and rusts. *Molecular ecology* 2: 113–118.  
<http://dx.doi.org/10.1111/j.1365-294X.1993.tb00005.x>
- Gui, Y., Zhu, G.S., Callac, P., Hyde, K.D., Parra, L.A., Chen, J., Yang, T.J., Huang, W.B., Gong, G.L. & Liu, Z.Y. (2015) *Agaricus* section *Arvenses*: three new species in highland subtropical Southwest China. *Fungal Biology* 119: 79–94.  
<http://dx.doi.org/10.1111/j.1365-294x.1993.tb00005.x>
- Hall, T.A. (1999) BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. *Nucl. Acids. Symp. Ser.* 41: 95–98.
- Hara, C., Kiho, T., Tanaka, Y. & Ukai, S. (1982) Anti-inflammatory activity and conformational behavior of a branched (1→3)-β-D-glucan from an alkaline extract of *Dictyophora indusiata* Fisch. *Carbohydrate Research* 110: 77–87.  
[http://dx.doi.org/10.1016/0008-6215\(82\)85027-1](http://dx.doi.org/10.1016/0008-6215(82)85027-1)
- Hemmes, D.E. & Desjardin, D.E. (2009) Stinkhorns of the ns of the Hawaiian Isl aiiian Isl aiiian Islands. *Fungi* 2: 8–10.
- Huelsensbeck, J.P. & Ronquist, F. (2001) MRBAYES: Bayesian inference of phylogenetic trees. *Bioinformatics* 17: 754–755.  
<http://dx.doi.org/10.1093/bioinformatics/17.8.754>
- Index Fungorum (2016) Available from: <http://www.indexfungorum.org/names> (accessed 16 February 2016)
- Jayasiri, S.C., Hyde, K.D., Ariyawansa, H.A., Bhat, J., Buyck, B., Cai, L., Dai, Y.C., Abd-Elsalam, K.A., Ertz, D., Hidayat, I., Jeewon, R., Jones, E.B.G., Bahkali, A.H., Karunarathna, S.C., Liu, J.K., Luangsa-ard, J.J., Lumbsch, H.T., Maharachchikumbura, S.S.N., McKenzie, E.H.C., Moncalvo, J.M., Ghobad-Nejhad, M., Nilsson, H., Pang, K., Pereira, O.L., Phillips, A.J.L., Raspé, O., Rollins, A.W., Romero, A.I., Etayo, J., Selçuk, F., Stephenson, S.L., Suetrong, S., Taylor, J.E., Tsui, C.K.M., Vizzini, A., Abdel-Wahab, M.A., Wen, T.C., Boonmee, S., Dai, D.Q., Daranagama, D.A., Dissanayake, A.J., Ekanayaka, A.H., Fryar, S.C., Hongsanan, S., Jayawardena, R.S., Li, W.J., Perera, R.H., Phookamsak, R., De Silva, N.I., Thambugala, K.M., Tian, Q., Wijayawardene, N.N., Zhao, R.L., Zhao, Q., Kang, J.C. & Promputtha, I. (2015) The Faces of Fungi database: fungal names linked with morphology, phylogeny and human impacts. *Fungal Diversity* 74: 3–18.  
<http://dx.doi.org/10.1007/s13225-015-0351-8>
- Karunarathna, S.C., Guinberteau, J., Chen, J., Vellinga, E.C., Zhao, R., Chukeatirote, E., Yan, J., Hyde, K.D. & Callac, P. (2014) Two new species in *Agaricus* tropical clade I. *Chiang Mai Journal of Science* 41: 771–780.
- Kasuya, T. (2008) *Phallus luteus* comb. nov., a new taxonomic treatment of a tropical phalloid fungus. *Mycotaxons* 106: 7–18.
- Kibby, G. & McNeil, D. (2012) *Phallus duplicatus* new to Britain. *Field Mycology* 13: 86–89.  
<http://dx.doi.org/10.1016/j.fldmyc.2012.06.009>
- Kim, C.S., Jo, J.W., Kwag, Y.-N., Sung, G.-H., Lee, S.-G., Kim, S.-Y., Shin, C.-H. & Han, S.-K. (2015) Mushroom flora of ulleung-gun and a newly recorded *Bovistas* species in the Republic of Korea. *Mycobiology* 43: 239–257.  
<http://dx.doi.org/10.5941/MYCO.2015.43.3.239>
- Kirk, P., Cannon, P., Minter, D. & Stalpers, J. (2008) *Ainsworth & Bisby's dictionary of the fungi*. 10<sup>th</sup> Edition, CABI, Europe, UK.
- Kornerup, A. & Wanscher, J.H. (1978) *Methuen handbook of colour*. Eyre Methuen, London, pp 252.
- Kreisel, H. (1996) A preliminary survey of the genus *Phallus* sensu lato. *Czech Mycol* 48: 273–281.
- Kreisel, H. & Hausknecht, A. (2002) The gasteral basidiomycetes of Mascarenes and Seychelles 3. Some recent records. *Österr. Z. Pilzk* 18: 149–159.
- Lee, W.S. (1957) Two new phalloids from Taiwan. *Mycologia* 49: 156–158.  
<http://dx.doi.org/10.2307/3755742>
- Li, H.L., Mortimer, P.E., Karunarathna, S.C., Xu, J.C. & Hyde, K.D. (2014) New species of *Phallus* from a subtropical forest in Xishuangbanna, China. *Phytotaxa* 163 (2): 91–103.  
<http://dx.doi.org/10.11646/phytotaxa.163.2.3>



- Li, T., Song, B., Wu, X., Liu, B. & Deng, W. (2008) A study on phallaceae from Yunnan, Guizhou and Guangxi, China. *Guizhou Science* 22: 80–89.
- Linnaei, C.V. & Salvius, L. (1753) *Species plantarum. Holmiae, Impensis Laurentii Salvii* 2: 1178.
- Linnean Society of London (1881–1887) *Transaction of the Linnean Society of London, 2nd series, Botany* 2: 65.
- Liu, B., Fan, L., Li, J.Z., Li, T.H., Song, B. & Li, J.H. (2005) *Flora Fungorum Sinicorum Vol. 23*. Science Press, Beijing: Science Press, pp 137–171.
- Liu, J.K., Jones, E.B.G., Ariyawansa, H.A., Bhat, D.J., Boonmee, S., Maharachchikumbura, S., McKenzie, E.H.C., Phookamsak, R., Phukhamsakda, C., Shenoy, B.D., Abdel-Wahab, M.A., Buyck, B., Chen, J., Chethana, K.W.T., Singtripop, C., Dai, D.Q., Dai, Y.C., Daranagama, D.A., Dissanayake, A.J., Doliom, M., D'souza, M.J., Fan, X.L., Goonasekara, I.D., Hirayama, K., Hongsanan, S., Jayasiri, S.C., Jayawardena, R.S., Karunarathna, S.C., Li, W.J., Mapook, A., Norphanphoun, C., Pang, K.L., Perera, R.H., Peršoh, D., Pinruan, U., Senanayake, I.C., Somrithipol, S., Suetrong, S., Tanaka, K., Thambugala, K.M., Tian, Q., Tibpromma, S., Udayanga, D., Wijayawardene, N.N., Wanasinghe, D.N., Wisitrasameewong, K., Abdel-Aziz, F.A., Adamčík, S., Bahkali, A.H., Boonyuen, N., Bulgakov, T., Callac, P., Chomnunti, P., Greiner, K., Hashimoto, A., Hofstetter, V., Kang, J.C., Lewis, D.A., Li, X.H., Liu, X.X., Liu, Z.Y., Matumura, M., Mortimer, P.E., Rambold, G., Randrianjohany, E., Sato, G., Indrasudhi, V.S., Tian, C.M., Verbeken, A., Wolfgang, von. Brackel., Wang, Y., Wen, T.C., Xu, J.C., Yan, J.Y., Zhao, R.L., Camporesi, E. & Hyde, K.D. (2015). Fungal Diversity Notes 1–110: Taxonomic and phylogenetic contributions to fungal species. *Fungal Diversity* 72: 1–197.  
<http://dx.doi.org/10.1007/s13225-015-0324-y>
- Liu, J.K., Phookamsak, R., Jones, E.B.G., Zhang, Y., Ko-Ko, T.W., Hu, H.L., Boonmee, S., Doilom, M., Chukeatirote, E., Bahkali, A.H., Wang, Y. & Hyde, K.D. (2011) *Astrosphaeriella* is polyphyletic, with species in *Fissuroma* gen. nov., and *Neoastrosphaeriella* gen. nov. *Fungal Diversity* 51: 135–154.  
<http://dx.doi.org/10.1007/s13225-011-0142-9>
- Lloyd, C.G. (1909) Synopsis of the known Phalloids. *Bulletin Lloyd Library* 7: 10.
- Lu, Y., Gui, Y., Gong, G., Wei, S. & Zhu, G. (2014) Genetic diversity of 18 *Dictyophora rubrovolvata* germplasm resources from Guizhou. *Guizhou Agricultural Sciences* 42: 17–20.
- Moreno, G., Khalid, A., Alvarado, P. & Kreisel, H. (2013) *Phallus hadriani* and *P. roseus* from Pakistan. *Mycotaxon* 125: 45–51.  
<http://dx.doi.org/10.5248/125.45>
- Oyetayo, V., Dong, C. & Yao, Y. (2009) Antioxidant and antimicrobial properties of aqueous extract from *Dictyophora indusiata*. *The Open Mycology Journal* 3: 20–26.  
<http://dx.doi.org/10.2174/1874437000903010020>
- Persoon, C.H. (1801) *Synopsis Methodica Fungorum* 2: 244.
- Peterak, F. (1922–1928) *Petrak's lists* 3: 91.
- Peterak, F. (1922–1928) *Petrak's lists* 3: 258.
- Rannala, B. & Yang, Z. (1996) Probability distribution of molecular evolutionary trees: a new method of phylogenetic inference. *Journal of Molecular Evolution* 43: 304–311.  
<http://dx.doi.org/10.1007/BF02338839>
- Reid, D.A. (1976) Some gasteromycetes from Trinidad and Tobago. *Kew Bulletin* 31: 657–690.  
<http://dx.doi.org/10.2307/4119418>
- Saccardo, P.A. (1888) *Sylloge Fungorum VII*. pp. 3–11.
- Silvestro, D. & Michalak, I. (2010) raxmlGUI: a graphical front-end for RAxML Program and documentation. Available from: <http://www.sourceforge.com> (accessed 22 July 2016)
- Stamatakis, A. (2006) RAxML-VI-HPC: Maximum likelihood-based phylogenetic analyses with thousands of taxa and mixed models. *Bioinformatics* 22: 2688–2690.  
<http://dx.doi.org/10.1093/bioinformatics/btl446>
- Swofford, D.L. (2001) *PAUP\*. Phylogenetic Analysis Using Parsimony (\*and Other Methods). Version 4*. Sinauer Associates, Sunderland, Massachusetts.
- Thompson, J.D., Gibson, T.J., Plewniak, F., Jeanmougin, F. & Higgins, D.G. (1997) The CLUSTAL\_X windows interface: flexible strategies for multiple sequence alignment aided by quality analysis tools. *Nucleic acids research* 25: 4876–4882.  
<http://dx.doi.org/10.1093/nar/25.24.4876>
- Thongklang, N., Nawaz, R., Khalid, A.N., Chen, J., Hyde, K.D., Zhao, R., Parra, L., Hanif, M., Moinard, M. & Callac, P. (2014) Morphological and molecular characterization of three species from tropical Asia (Pakistan, Thailand) reveals a new group in *Agaricus* section *Xanthodermatei*. *Mycologia* 106: 1220–1232.  
<http://dx.doi.org/10.3852/14-076>
- Ventenat, E.P. (1798) Dissertation sur le genre *Phallus*. *Mém. Inst. Nat. Sci. Arts Mat. Phys.* 1: 503–523.
- Zang, M., Ji, D.G. & Liou, S.C. (1976) *Materiae ad diagnosis fungorum oeconomiarum Yunnanicum (II)*. *Yunnan Zhiwu Yanjiu* 2: 1–12.

- Zang, M., Zheng, D.R. & Hu, Z.X. (1988) A new species of the genus *Dictyophora* from China. *Mycotaxon* 33: 146.
- Zhang, L.F., Yang, J.B. & Yang, Z.L. (2004) Molecular phylogeny of eastern Asian species of *Amanita* (Agaricales, Basidiomycota): taxonomic and biogeographic implications. *Fungal Diversity* 17: 219–238.
- Zhao, R., Karunarathna, S., Raspé, O., Parra, L.A., Guinberteau, J., Moinard, M., Kesel, A.D., Barroso, G., Courtecuisse, R., Hyde, K.D., Gueilly, A.K., Desjardin, D.E. & Callac, P. (2011) Major clades in tropical *Agaricus*. *Fungal Diversity* 51: 279–296.  
<http://dx.doi.org/10.1007/s13225-011-0136-7>
- Zhao, R., Zhou, J.L., Chen, J., Margaritescu, S., Sánchez-Ramírez, S., Hyde, K.D., Callac, P., Parra, L.A., Li, G.J. & Moncalvo, J.M. (2016) Towards standardizing taxonomic ranks using divergence times—a case study for reconstruction of the *Agaricus* taxonomic system. *Fungal Diversity* 78: 239–292.  
<http://dx.doi.org/10.1007/s13225-016-0357-x>
- Zhaxybayeva, O. & Gogarten, J.P. (2002) Bootstrap, Bayesian probability and maximum likelihood mapping: exploring new tools for comparative genome analyses. *BMC Genomics* 3: 4.  
<http://dx.doi.org/10.1186/1471-2164-3-4>