# **Fungal Biodiversity Profiles 11-20**

Sinang HONGSANAN<sup>a,b,c</sup>, Kevin D. HYDE<sup>a,b,c,d</sup>, Ali H. BAHKALI<sup>d</sup>, Erio CAMPORESI<sup>j</sup>, Putaruk CHOMNUNTI<sup>c</sup>, Hasini EKANAYAKA<sup>a,b,c</sup>, André A.M. GOMES<sup>f</sup>, Valérie HOFSTETTER<sup>h</sup>, E.B.Gareth JONES<sup>e</sup>, Danilo B. PINHO<sup>g</sup>, Olinto L. PEREIRA<sup>g</sup>, Qing TIAN<sup>a,b,c</sup>, Dhanushka N. WANASINGHE<sup>a,b,c</sup>, Jian-Chu XU<sup>a,b</sup> & Bart BUYCK<sup>i\*</sup>

> <sup>a</sup>World Agroforestry Centre, East and Central Asia, Kunming 650201, Yunnan, China

<sup>b</sup>Key Laboratory of Economic Plants and Biotechnology, Kunming Institute of Botany, Chinese Academy of Sciences, Lanhei Road No 132, Panlong District, Kunming, Yunnan Province, 650201, PR China

<sup>c</sup>Center of Excellence in Fungal Research, Mae Fah Luang University, Chiang Rai, 57100, Thailand, email address: kdhyde3@gmail.com

<sup>d</sup>Botany and Microbiology Department, College of Science, King Saud University, Riyadh, KSA 11442, Saudi Arabia

<sup>e</sup>Department of Botany and Microbiology, College of Science, King Saud University, P.O. Box 2455 Riyadh 11451, Kingdom of Saudi Arabia

<sup>f</sup>Departamento de Microbiologia, Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil

<sup>g</sup>Departamento de Fitopatologia, Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil; e-mail: oliparini@ufv.br

<sup>h</sup>Department of plant protection, Agroscope Changins-Wadenswil Research Station, ACW, Rte de Duiller, 1260, Nyon, Switzerland

<sup>i</sup>Museum National d'Histoire Naturelle, Dept. Systematique et Evolution CP 39, ISYEB, UMR 7205 CNRS MNHN UPMC EPHE, 12 Rue Buffon, F-75005 Paris, France; email: buyck@mnhn.fr

> <sup>j</sup>A.M.B. Gruppo Micologico Forlivese "Antonio Cicognani", Via Roma 18, Forlì, Italy

Abstract – The authors describe ten new taxa for science using mostly both morphological and molecular data. In Ascomycota, descriptions are provided for *Ceramothyrium ficus* (Chaetothyriales, Eurotiomycetes), *Lachnum fusiforme* (Leotiomycetes, Helotiales), four new

<sup>\*</sup> Correspondence: Bart Buyck, buyck@mnhn.fr

species in Dothideomycetes, namely *Micropeltis dendrophthoes* (Microthyriales), *Montagnula bellevaliae* and *M. scabiosae* (Pleosporales), *Scorias mangiferae* (Capnodiales), as well as for *Mucodor coffeanum* (Xylariales, Sordariomycetes). In Basidiomycota, three new subgenera are introduced in *Russula* (Russulales).

# Dothideomycetes / Eurotiomycetes / Leotiomycetes / new species / new subgenera / Phylogeny / Russula / Sordariomycetes / Taxonomy

# 11. Ceramothyrium ficus Hongsanan, Q. Tian, A.H. Bahkali & K.D. Hyde, sp. nov. Figs 1-2

*Index Fungorum number*: IF 551525, *Facesoffungi number*: FoF: 01059. *Etymology: ficus* referring to the host on which the taxon was collected.

*Systematic placement*: Ascomycota, Pezizomycotina, Eurotiomycetes, Chaetothyriomycetidae, Chaetothyriales, Chaetothyriaceae.

Holotype: MFU 15-2252.

GenBank: KT588601 (ITS), KT588599 (LSU).

*Epiphytic* on the upper surface of living leaves, appearing as small black dots, leaves remaining healthy. Superficial hyphae branched, septate, slightly constricted at the septa, brown to dark brown, radiating outwardly, and covering the ascomata as a subiculum, without penetrating host tissues. Sexual morph: Ascomata 475-550 high  $\times$  120-130 µm diam. ( $\bar{x} = 530 \times 124$  µm, n = 5), solitary or scattered, superficial on the surface of leaves, globose to subglobose, flattened when dry, brown to dark brown, easily removed, covered by a subiculum or hyphal layer. *Peridium* 5-10 µm wide, comprising two strata, the outer stratum comprising brown to dark brown cells of textura angularis, the inner stratum comprising pale brown to hyaline flattened cells. Hamathecium comprising pseudoparaphyses embedded in mucilage. Asci 95-110  $\times$  30-39 µm ( $\bar{x} = 104 \times 33$  µm, n = 10), 8-spored, bitunicate, clavate, with long pedicellate, ocular chamber not seen, evanescent. Ascospores  $36-39 \times 7-8 \ \mu\text{m}$  ( $\bar{x} = 38 \times 7.5 \ \mu\text{m}$ , n = 10), 3-5-seriate, hyaline, subcylindrical, muriform, with 7-8 transverse septa and 1 longitudinal septa, sometimes with 2 longitudinal septa in the end cells, slightly constricted at the septa, narrowly rounded at both ends, verrucose or smooth-walled, surrounded by a mucilaginous sheath. Asexual morph: undetermined.

*Culture on PDA*: Ascospores germinating on PDA at 25-28°C in 12 h of light/12 h of dark, germ tubes appearing from most cells of the ascospores, hyaline to brown, and becoming brown to grayish. Colonies reaching 1 cm diam. after 7 days on PDA at 25-28°C, colonies erumpent on the surface of media, surface smooth, velvety, slightly wavy at the margin, no asexual morph was produced in PDA after 60 days incubation.

*Material examined*: THAILAND, Chiang Rai Province, Mae Fah Luang University, on living leaves of *Ficus* sp. (Moraceae), 23 January 2015, S. Hongsanan AD01 (MFU 15-2252, **holotype**), (**isotype** MFU 15-2253; in KUN), ex-type living culture, **MFLUCC 15-0228**, MFLUCC 15-0229, and in BCC.

*Notes*: The genus *Ceramothyrium* was introduced by Batista and Maia (1957), with the type species *Ceramothyrium paiveae* Bat. & H. Maia in the family Phaeosaccardinulaceae. Species of *Ceramothyrium* are referred to as sooty molds because of their morphological and ecological similarity with other sooty mold species in Capnodiaceae (Chomnunti *et al.*, 2012a, 2014). Phylogenetic analyses place *Ceramothyrium* in the family Chaetothyriaceae (Chaetothyriales,

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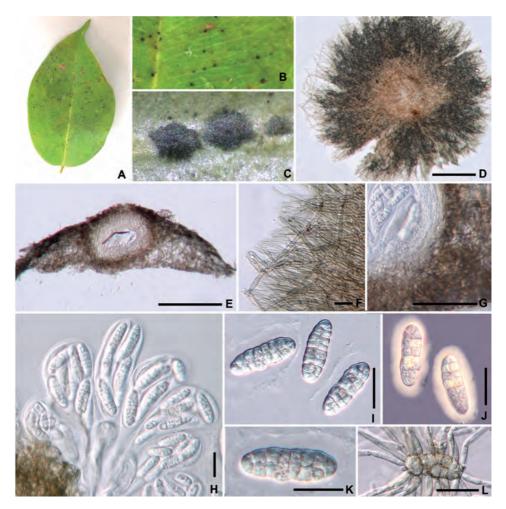


Fig. 1. *Ceramothyrium ficus* (holotype). A. Specimen. B, C. Ascomata developing on surface of leaves. D. Squash mount of ascomata covered by a subiculum. E. Section through ascoma. F. Margin of ascoma. G. Peridium of ascoma. H. Bitunicate asci. I. Ascospores with mucilaginous sheath. J, K. Ascospores with mucilaginous sheath in India Ik. L. Germinating ascospore. Scale bars: D,  $E = 100 \mu m$ ,  $G = 20 \mu m$ , F, H-L = 20  $\mu m$ .

Eurotiomycetes) with strong support (Winka *et al.*, 1998; Lutzoni *et al.*, 2004; Miadlikowska & Lutzoni, 2004; Reeb *et al.*, 2004; Chomnunti *et al.*, 2012a, 2012b). Thirty epithets are listed in *Ceramothyrium* in Index Fungorum (2015).

*Ceramothyrium ficus* and *C. longisolcano* differ, as *C. ficus* was found on living leaves, and has small ascomata, which are pale brown to yellowish at the center when viewed in squash mounts, and smaller ascospores with 7-8 transverse septa and 1-2 longitudinal septa, while *C. longisolcano* was found on dead leaves, produces larger ascomata which are dark brown to reddish in the center when viewed in squash mounts, and has larger ascospores with 7 transverse septa and 1 longitudinal septum.

*Ceramothyrium ficus* is also distinct from *C. thailandicum* Chomnunti & K.D. Hyde which has ascomata darkened at the center when viewed in squash mounts, and clavate ascospores with 7-9 transverse septa and lacking longitudinal septa. Molecular analyses using ITS and LSU sequence data (Fig. 2) demonstrate a close relationship between *C. ficus* and *C. longisolcano* (90% ML and 1.0 PP support), but support *C. ficus* as a distinct species with high bootstrap support (100% ML and 1.0 PP).

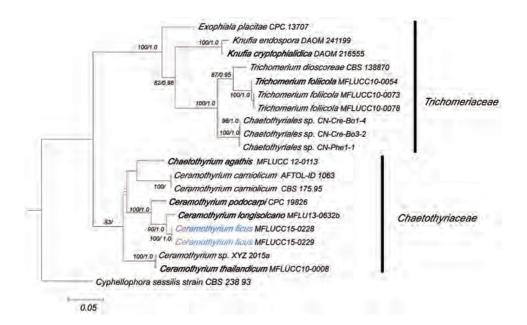


Fig. 2. The phylogenetic tree was obtained by RAxML maximum likelihood methods using ITS and LSU regions. The first set of numbers above the nodes are RAxML bootstrap value expressed values above 70% shown. The second set of numbers above the nodes are Bayesian posterior probabilities, with values above 0.95 shown. Strain numbers are indicated after species names. New sequence data are in blue bold, and other types are in black bold. The analysis included 10 strains from Trichomeriaceae, 8 strains from Chaetothyriaceae, and is rooted with Cyphellophora sessilis (Cyphellophoraceae) for the out-group; the alignment comprises 1,435 characters. The newly generated nucleotide sequences were compared against the GenBank database using the Mega BLAST program. Sequences that relate to Ceramothyrium were obtained from GenBank and were aligned using the multiple sequence alignment program, MAFFT (Katoh & Standley, 2013), checked manually using BioEdit software (Hall, 2014). Maximum-likelihood (ML) analysis was performed using raxmlGUIv.0.9b2 (Silvestro & Michalak, 2012), with 1.000 replicates. The model of evolution was estimated by using MrModeltest 2.2 (Nylander et al., 2008). Posterior probabilities were determined by Markov Chain Monte Carlo sampling (MCMC) in MrBayes v3.1.2 (Huelsenbeck & Ronquist, 2001). Six simultaneous Markov chains were run for 1,000,000 generations and trees were sampled every 100th generation, and 10,000 trees were obtained. The first 2,000 trees, representing the burn-in phase were discarded (Cai et al., 2006, 2008). Phylogenetic trees were drawn by using Treeview v. 1.6.6 (Page, 2001).

Index Fungorum number: IF551388, Facesoffungi number: FoF 00970. Etymology: The specific epithet fusiforme is named according to its ascospores.

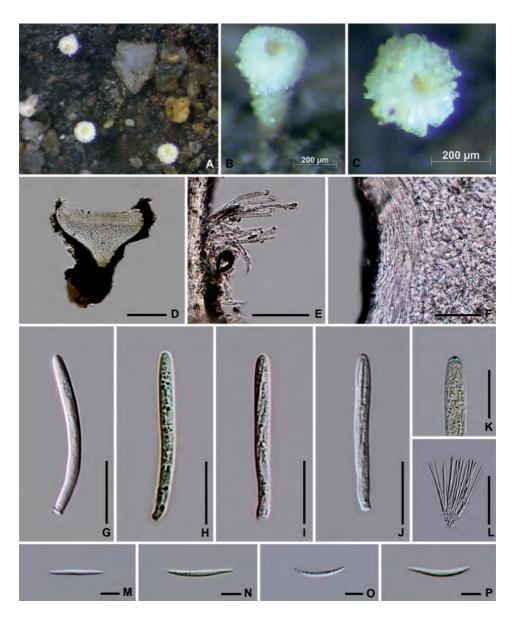


Fig. 3. Lachnum fusiforme (holotype). A. Discomata on wood. B, C. Discomata on wood. D. Cross section of an discoma. E. Cylindrical hairs. F. Close up of the excipulum. H-J. Short pedicellate asci. K Apex of the asci with the plug bluing in Melzer's reagent. L. Septate paraphyses. M-P. Filiform ascospores. Scale bars: B-D = 200  $\mu$ m, E = 50  $\mu$ m, F = 30  $\mu$ m, G-J= 15  $\mu$ m, K = 30  $\mu$ m, L-P = 5  $\mu$ m.

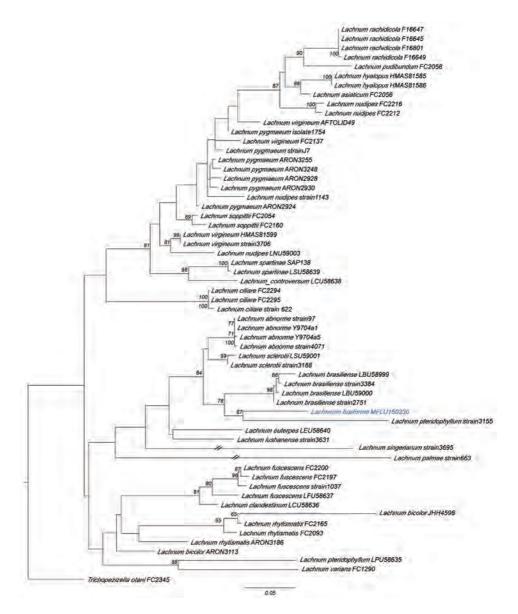


Fig 4. Phylogenetic tree obtained by Maximum likelyhood analysis using ITS sequence data. Maximum likelihood bootstrap support values (significant support threshold value > 70%) are indicated at the nodes. Newly generated sequences from this study are in bold. The analyses included 58 *Lachnum* strains and is rooted with *Trichopezizella otanii* (*Lachnaceae*) for the out-group; the alignment comprises 486 characters. The newly generated nucleotide sequences were edited with BioEdit software (Hall, 2014) and checked manually. The other sequences were retrieved from GenBank. The consensus sequences were aligned using the multiple sequence alignment program, MAFFT v. 6.864b (http://mafft.cbrc.jp/alignment/server/index.html). The alignments were checked, and manual adjustments were performed when necessary. The phylogenetic analyses were conducted as described by Machado *et al.*, (2014).

*Systematic placement*: Lachnaceae, Helotiales, Leotiomycetidae, Leotiomycetes, Pezizomycotina, Ascomycota.

Holotype: MFLU 15-0230.

GenBank: KT384413 (ITS).

Saprobic on dead stems. Sexual morph: Apothecia  $435-440 \times 378-382 \ \mu m$  $(\bar{x} = 437 \times 380 \ \mu\text{m}, n = 10)$  arising singly or in small groups, superficial, stipitate, white when fresh, covered with long, bright white hairs. Receptacle cupulate, disc concave and light yellow, margins white. Stipe hairy, medium to long. Hairs  $70-90 \times 3-4 \ \mu m \ (79.4 \times 3.5 \ \mu m, n = 30)$  on flanks and stipe, cylindrical or tapered to a blunt hemispherical tip, straight, thin-walled, septate, white, covered with dense colorless granules, occasionally tips are covered with a vellow to white resinous-like substance. Ectal excipulum 32-37 µm wide ( $\bar{x} = 35.5$  µm, n = 10) composed of hyaline cells textura prismatica. Medullary excipulum 48-52 µm wide ( $\bar{x} = 51$  µm, n = 10), comprising hyaline cells of *textura intricata*. Hymenium hyaline. Paraphyses 1.5-2 µm wide ( $\bar{x} = 1.7$  µm, n = 20), numerous, filiform, septate. Asci 47-52 × 4-4.5  $\mu$ m ( $\bar{x} = 49.4 \times 4 \mu$ m, n = 30), 8-spored, unitunicate, cylindrical, short pedicellate, with inoperculate apical plug bluing in Melzers reagent. Ascospores 18- $25 \times 1.5$ -2 µm ( $\bar{x} = 21.9 \times 1.6$  µm, n = 40), biseriate, fusiform with pointed tips, hyaline, aseptate, smooth-walled, lacking sheath or appendages, with refractive inclusions. Asexual morph: undetermined.

*Material examined*: THAILAND, Chiang Rai Province, Kun Korn waterfall, on dead stems, 20 January 2015, A.H. Ekanayaka, HD0012 (MFLU 15-0230, **holotype**).

*Notes*: This is a very large genus with a diverse distribution not only in temperate, but also in tropical zones. They inhabit dead stems and leaves of herbaceous plants (Spooner, 1987). *Lachnum* is characterized by discoid apothecia covered by finely granulate, subcylindrical and septate hairs on the receptacle surface, subcylindrical to clavate asci with an amyloid apical ring, ascospores with different shapes, lanceolate to subcylindrical paraphyses, and an ectal excipulum often composed of prismatic cells (Spooner, 1987; Kirk *et al.*, 2008). Currently 522 epithets are listed in Index Fungorum and 250 species are estimated in this genus (Kirk *et al.*, 2008; Index Fungorum 2015). Although there are large number of described species, there are limited phylogenetic studies. Hence, species identifications have mainly been based on morphological features (Zhao & Zhuang, 2011).

Single spore isolation was not successful for *Lachnum fusiforme*. Therefore fungal DNA was isolated directly from the discomata and no living cultures are available. The helotialean genus *Lachnum* is widely distributed in the tropics and characterized by discoid apothecia, which are small in size with numerous hairs on the receptacle surface and inhabit a wide range of plant tissues (Zhao & Zhuang, 2011). Phylogenetic analyses and morphological comparison indicate that our specimen (MFLU 15-0230) belongs in *Lachnum*. *Lachnum fusiforme* is morphologically similar to *L. albidulum* (Penz. & Sacc.) M.P. Sharma, but differs in having wider paraphyses and much longer ascospores. Paraphyses of *L. albidulum* are 2-3 µm at the widest point and ascospores 8-9 µm long (Haines, 1992), but in *L. fusiforme* paraphyses are 1.5-2 µm wide and ascospores 18-25 µm long.

# 13. Micropeltis dendrophthoes Hongsanan & K.D. Hyde, sp. nov. Figs 5-6

Index Fungorum number: IF 551523, Facesoffungi number: FoF: 01058. Etymology: dendrophthoes, referring to the host on which the taxon was collected. S. Hongsanan et al.

*Systematic placement*: Ascomycota, Pezizomycotina, Dothideomycetes, Incertae sedis, Microthyriales, Micropeltidaceae.

Holotype: MFLU 15-2255.

GenBank: KT588595 (LSU), KT588597 (SSU).

*Epiphytic* on the upper surface of leaves, appearing as small black dots. Superficial hyphae absent. Sexual morph: Thyriothecia 85-93 high  $\times$  346-538 µm diam. ( $\bar{x} = 91 \times 495 \mu$ m, n = 5), solitary to clustered, superficial on the surface of hosts, circular, membranous, bluish to grey black, easy removed, base poorly developed, with a central, irregular ostiole. Upper wall comprising an irregular, meandering, arrangement of hyphae, from the central ostiole to the outside. Peridium

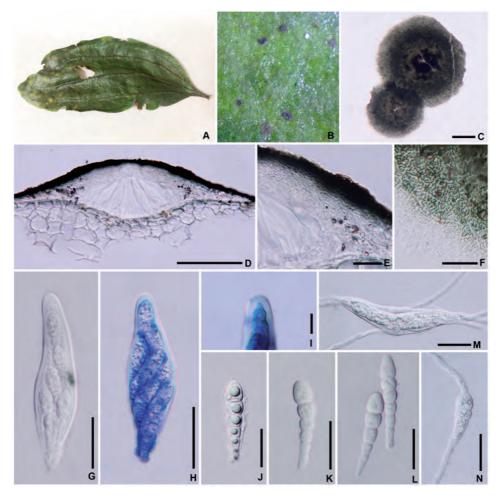


Fig. 5. *Micropeltis dendrophthoes* (holotype). A. Leaf. B. Thyriothecia developing on surface of leaves. C. Thyriothecia viewed in squash mount. D. Section through thyriothecium. E. Peridium of thyriothicium. F. Upper wall of thyriothecium in squash mount. G. Ascus. H. Ascus in cotton blue reagent. I. Ocular chamber of ascus in cotton blue reagent. J. Ascospore with gelatinous sheath. K, L. Ascospores in 70% lactic acid. M, N. Germinating ascus and ascospore. Scale bars: C,  $D = 100 \mu m$ , E-H, J-N = 20  $\mu m$ , I = 5  $\mu m$ .

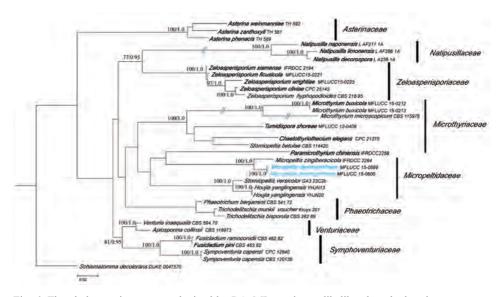


Fig. 6. The phylogenetic tree was obtained by RAxML maximum likelihood methods using sequences of LSU and SSU regions. The first set of numbers above the nodes are RAxML bootstrap value expressed values above 70% shown. The second set of numbers above the nodes are Bayesian posterior probabilities, with values above 0.95 shown. Strain numbers are indicated after species names. New sequence data are in blue bold, and other types are in black bold. The analyses included 13 strains from Microthyriales, 20 strains from closely related orders, and is rooted with Schismatomma decolorans (Roccellaceae) for the out-group; the alignment comprises 1,151 characters. The newly generated nucleotide sequences were compared against the GenBank database using the Mega BLAST program. Sequences that relate to Micropeltis were obtained from GenBank and were aligned using the multiple sequence alignment program, MAFFT (Katoh & Standley, 2013), checked manually using BioEdit software (Hall, 2014). Maximum-likelihood (ML) analysis was performed using raxmlGUIv.0.9b2 (Silvestro & Michalak, 2012), with 1,000 replicates. The model of evolution was estimated by using MrModeltest 2.2 (Nylander et al., 2008). Posterior probabilities were determined by Markov Chain Monte Carlo sampling (MCMC) in MrBayes v3.1.2 (Huelsenbeck & Ronquist, 2001). Six simultaneous Markov chains were run for 1,000,000 generations and trees were sampled every 100th generation, and 10,000 trees were obtained. The first 2,000 trees, representing the burn-in phase were discarded (Cai et al., 2006, 2008). Phylogenetic trees were drawn by using Treeview v. 1.6.6 (Page, 2001).

5-10 µm wide, comprising two strata, the outer stratum having bluish to black, occluded walls, inner stratum of greenish to hyaline, flattened cells. *Hamathecium* with evanescent pseudoparaphyses, asci embedded in mucilage, inclined towards the central ostiole,. *Asci* 60-62 × 16-18 µm ( $\bar{x} = 61 \times 17$  µm, n = 10), 6-8-spored, bitunicate, fissitunicate, broadly cylindrical to fusiform, with a short pedicel, apically rounded with ocular chamber. *Ascospores* 36-39 × 7-8 µm ( $\bar{x} = 38 \times 7.5$  µm, n = 10), 2-3-seriate, hyaline, clavate, 4-5-septate, constricted at the septa, narrowly rounded at both ends, lower end cell long, smooth-walled, surrounded by a mucilaginous sheath. **Asexual morph:** undetermined.

*Culture on PDA*: Ascospores germinating on PDA at 25-28°C for 12 h of light/12 h of dark, germ tubes appearing from each end of the ascospores, hyaline to brown, but becoming brown to reddish later. Colonies reaching 1 cm diam. after 7 days on PDA at 25-28°C, colonies superficial to erumpent, surface smooth, pale brown at the center, darkened at the margin, producing brown pigment on PDA.

*Material examined*: THAILAND, Chiang Rai Province, Tasud, on living leaves of *Dendrophthoe* sp. (Loranthaceae), 29 January 2015, S. Hongsanan PM02 (MFLU 15-2255, **holotype**), (**isotype** in KUN), ex-type living culture, MFLUCC 15-0599, MFLUCC 15-0600.

*Notes: Micropeltis* is the type genus of Micropeltidaceae (Wu *et al.*, 2011, 2014). The genus was formally established by Montagne (1842), with the type species *M. applanata* Mont. *Micropeltis* is characterized by dark brown to black or bluish to greenish thyriothecia, with the peridium comprising an irregular, meandering, arrangement of compact hyphae, and multi-septate ascospores (Wu *et al.*, 2011, 2014; Hyde *et al.*, 2013). Free-hand sections did not reveal any penetration of the host, and it is not clear how these taxa obtain their nutrients. Several researchers used the variation of colour and size of thyriothecia, the variation of size and shape of ascospores, and septation to distinguish species; however, there are a very little sequence data in GenBank (Batista *et al.*, 1959; Reynolds & Gilbert., 2005, 2006; Wu *et al.*, 2011).

*Micropeltis dendrophthoes* is most similar to *M. rhopaloides* Syd. & P. Syd. in having superficial thyriothecia, producing bitunicate, 4-8-spored asci and 4-5-septate, hyaline, clavate ascospores, which are constricted at the septa. However, *M. dendrophthoes* differs in the size and shape of its ascospores, surrounded by a thin sheath. *Micropeltis dendrophthoes* is also similar to *M. borneensis* Syd. & P. Syd. but the latter has 8-spored asci, with fusiform ascospores lacking a sheath, and having a short basal cell at the base of the ascospores. Molecular analyses based on a combined data set of LSU and SSU sequence data indicate that *M. dendrophthoes* is a distinct species (100% ML and 1.0 PP), closely related to *M. zingiberacicola* H.X. Wu & K.D. Hyde (100% ML and 1.0 PP).

# 14. Montagnula bellevaliae Wanasinghe, Camporesi, E.B.G. Jones & K.D. Hyde, sp. nov. Figs 7-8

Index Fungorum number: IF551512, Facesoffunginumber: FoF01051.

*Systematic placement*: Ascomycota, Pezizomycotina, Dothideomycetes, Pleosporomycetidae, Pleosporales, Didymosphaeriaceae.

*Etymology*: Name reflects the host genus *Bellevalia*, from which the species was collected.

Holotype: MFLU 15-1881.

GenBank: KT443906 (ITS), KT443902 (LSU), KT443904 (SSU).

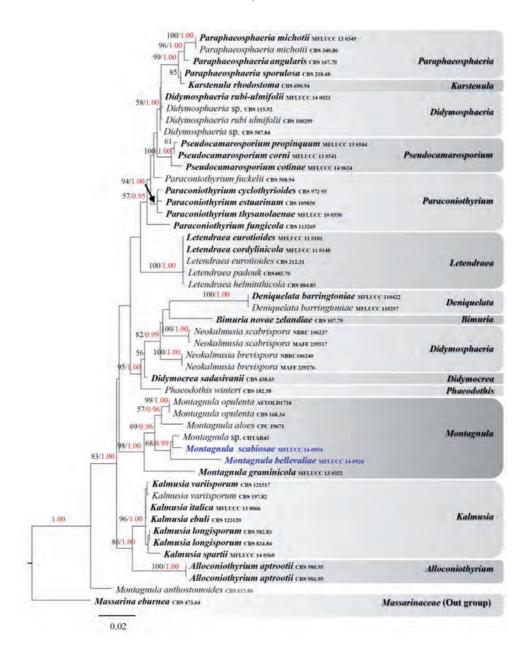
Saprobic on dead herbaceous branches. Sexual morph: Ascomata 100-120 µm high, 150-175 µm diam. ( $\bar{x} = 117 \times 166 µm$ , n = 10), solitary, scattered, immersed to erumpent, globose to subglobose or obpyriform, dark brown to black, coriaceous, ostiolate. Ostiole 60-90 µm high, 30-50 µm diam. ( $\bar{x} = 73 \times 39.5 µm$ , n = 5), eccentric, papillate, black, smooth, filled with hyaline cells when mature. Peridium 5-10 µm wide at the base, 10-20 µm wide at the sides, comprising 2-3 layers of blackish to dark brown, thin-walled cells of *textura angularis*. Hamathecium comprising numerous, 2-3.5 µm (n = 30) wide, filamentous, branched, septate, pseudoparaphyses. Asci 70-100 × 9-12 µm ( $\bar{x} = 86 \times 10.5 µm$ , n = 40), 8-spored, bitunicate, fissitunicate, clavate, pedicel furcate and up to 20-30 µm long, rounded and thick-walled at the apex, with a ocular chamber. Ascospores 15-18 × 5-6 µm ( $\bar{x} = 16 \times 5.5 µm$ , n = 50), overlapping 1-2-seriate, narrowly ovoid to clavate, 2-septate, constricted at the septa, initially hyaline, becoming reddish brown at maturity, with conical and narrowly rounded ends, guttulate, lacking a mucilaginous sheath. Asexual morph: Undetermined.



Fig. 7 *Montagnula bellevaliae* (holotype). **A.** Appearance of ascomata on host substrate. **B.** Section of the ascoma. **C.** Section of the peridium cells. **D.** Pseudoparaphyses. **E-G.** Asci. **H-J.** Ascospores. **K.** Germinating spore. **L, M.** Colonies on PDA (m from below). Scale bars: **B** = 50  $\mu$ m, **B** = 10  $\mu$ m, **D** = 5  $\mu$ m, **E-G** = 20  $\mu$ m, **H-K** = 10  $\mu$ m.

*Culture characteristics*: Colonies on PDA reaching 15-20 mm diam. in 21 days at 16°C, white grey in center, cream to orangish-white at the outer region, spreading with moderate aerial mycelium, and smooth, even margins; white to cream or yellowish white from below.

*Material examined:* ITALY, Forlì-Cesena Province, Predappio, Fiumana, dead stems of *Bellevalia romana*, 04 Feb 2013, E. Camporesi (MFLU 15-1881, **holotype**, **isotype** in BBH 39898), ex-type living culture, **MFLUCC 14-0924**, in BCC.



Notes: Recent studies (Leuchtmann, 1984; Aptroot, 1995; Barr, 2001; Zhang *et al.*, 2012a, b; Hyde *et al.*, 2013; Ariyawansa *et al.*, 2013, 2014), currently place *Montagnula* in the family Didymosphaeriaceae including some phragmosporous and didymosporous species, making it a heterogenic genus. Phylogenetic analyses have shown a robust clustering at family level of *M. opulenta* (De Not.) Aptroot with *Alloconiothyrium* Verkley, Göker & Stielow, *Bimuria* D. Hawksw., Chea & Sheridan,

Fig. 8. RAxML tree based on a combined dataset of LSU, SSU, and ITS partial sequences. Bootstrap support values for maximum likelihood (ML, black) higher than 70% and Bayesian posterior probabilities greater than 0.95 (red) are given above the nodes. The ex-types (reference strains) are in bold; the new isolates are in blue. The tree is rooted to Massarina eburnea (CBS 473.64). The analyses included 49 strains and is rooted with Massarina eburnea (Massarinaceae) for the out-group: the alignment comprises 2346 characters. The newly generated nucleotide sequences were edited with BioEdit software (Hall, 2014) and checked manually. The other sequences were retrieved from GenBank. The consensus sequences were compared against the GenBank database using the Mega BLAST program. The multiple alignments were automatically done by MAFFT v. 7.036 (Katoh & Standley, 2013), but manual adjustments for improvement were made by eye where necessary using BioEdit v. 7.2 (Hall, 2014) and ClustalX (Kohli & Bachhawat, 2003). MODELTEST v. 3.7 (Posada & Crandall 1998) following Akaike Information Criterion was used to determine the best-fit model of evolution for each data set for Bayesian and Maximum Likelihood analyses. Maximum-likelihood (ML) analysis was performed in RAxML (Stamatakis, 2008) implemented in raxmlGUI v.0.9b2 (Silvestro & Michalak, 2010), employing mixed models of evolution settings of the program and Bootstrap support obtained by running 1000 pseudo replicates. The online tool Findmodel was used to determine the best nucleotide substitution (http://www.hiv.lanl.gov/content/sequence/findmodel/findmodel.html) model for each partition. A Bayesian analysis was conducted with MrBayes v. 3.1.2 (Huelsenbeck & Rongvist, 2001) to valuate Posterior probabilities (PP) (Rannala & Yang 1996; Zhaxybayeva & Gogarten, 2002) by Markov Chain Monte Carlo sampling (BMCMC). Two parallel runs were conducted, using the default settings, but with the following adjustments: Six simultaneous Markov chains were run for 1,000,000 generations and trees were sampled every 100th generation and 10,000 trees were obtained. The first 2,000 trees, representing the burn-in phase of the analyses and discarded. The remaining 8000 trees were used for calculating PP in the majority rule consensus tree (Cai et al., 2006, 2008; Ariyawansa et al., 2015). Maximum trees were visualized with Tree View (Page, 2001).

Deniquelata Ariyawansa & K.D. Hyde, Didymocrea Kowalski, Didymosphaeria Fuckel, Kalmusia Niessl, Letendraea Sacc., Neokalmusia Ariyawansa & K.D. Hyde, Paracamarosporium Wijayaw. & K.D. Hyde, Paraconiothyrium Verkley, Paraphaeosphaeria O.E. Erikss., Phaeodothis Syd. & P. Syd., Pseudocamarosporium Wijayaw. & K.D. Hyde and Tremateia Kohlm., Volkm.-Kohlm. & O.E. Erikss. (Zhang et al., 2009; Hyde et al., 2013; Ariyawansa et al., 2014; Wijayawardene et al., 2014; Liu et al., 2015; Ariyawansa et al., 2015). Ariyawansa et al. (2014) synonymized Montagnulaceae under the oldest family name Didymosphaeriaceae.

The phylogenetic analyses of combined LSU, SSU and ITS sequence data (Fig. 8) show that *M. bellevaliae* and *M. scabiosae* belong in *Montagnula* and form a sister clade to *M. opulenta* (De Not.) Aptroot (CBS 168.34, AFTOL-ID 1734), *M.* sp. CHTAR43 (= *Letendraea helminthicola* (Berk. & Broome) Weese), *M. aloes* Crous (CPC 19671) and *M. graminicola* Chethana, Thambugala, Camporesi & K.D. Hyde (MFLUCC 13-0352) with high bootstrap support and confirm their generic placement in Didymosphaeriaceae. Molecular sequence data are not available for the type species (*M. infernalis*), hence it was not included in our analysis.

*Montagnula bellevaliae* is distinct in the genus in having an eccentric papilla, a 3-layered peridium of brown cells and producing 3-septate curved ascospores (Fig. 7).

15. Montagnula scabiosae Wanasinghe, Camporesi, E.B.G. Jones & K.D. Hyde, sp. nov. Fig. 9

Index Fungorum number: IF551513, Facesoffungi number: FoF01052.

*Systematic placement*: Ascomycota, Pezizomycotina, Dothideomycetes, Pleosporomycetidae, Pleosporales, Didymosphaeriaceae.

*Etymology*: Name reflects the host genus *Scabiosa*, from which the species was collected.

Holotype: MFLU 15-1882.

GenBank: KT443907 (ITS), KT443903 (LSU), KT443905 (SSU).

Saprobic on dead herbaceous branches. Sexual morph: Ascomata 300-320 µm high, 300-360 µm diam. ( $\bar{x} = 308 \times 330$  µm, n = 10), solitary, scattered, immersed, globose or subglobose, dark brown to black, coriaceous, ostiolate. Ostiole 90-110 µm high, 50-60 µm diam. ( $\bar{x} = 99 \times 56.5$  µm, n = 5), papillate, black, smooth, filled with brown cells when mature. Peridium 9-12 µm wide at the base, 12-18 µm wide in sides, comprising 4-5 layers, having a pigmented outer part of reddish brown to dark brown, thin-walled cells of *textura angularis*. Hamathecium comprising numerous, 1.5-3 µm (n = 30) wide, filamentous, branched, septate, pseudoparaphyses. Asci 110-130 × 14-20 µm ( $\bar{x} = 116 \times 15.6$  µm, n = 40), 8-spored, bitunicate, fissitunicate, clavate, pedicel furcate and up to 25-35 µm long, rounded and thick-walled at the apex, with an ocular chamber. Ascospores 20-23 × 7-9 µm ( $\bar{x} = 22 \times 7.7$  µm, n = 50), overlapping 1-2-seriate, ellipsoidal to broadly fusiform, flattened on one side, 3-septate, slightly constricted at the septa, initially hyaline, becoming reddish brown at maturity, with conical and narrowly rounded ends, guttulate, lacking a mucilaginous sheath. Asexual morph: Undetermined.

*Culture characteristics*: Colonies on PDA reaching 15-25 mm diam. in 21 days at 16°C, olivaceous-grey in center, creamy to creamy-white at the outer region, spreading with moderate aerial mycelium, and smooth, even margins; white to cream or yellowish white from below.

*Material examined:* ITALY, Forlì-Cesena Province, Predappio, Rocca delle Caminate, dead stems of *Scabiosa* sp. (Caprifoliaceae), 20 April 2014, E. Camporesi (MFLU 15-1882, **holotype**, **isotype** in BBH 39899), ex-type living culture, **MFLUCC 14-0954**, in BCC.

*Notes: Montagnula scabiosae* is distinct from other species of the genus in its thin peridium and inequilaterally fusiform ascospores that are flattened on one side.

## 16. Muscodor coffeanum A. A. M. Gomes, D. B. Pinho and O. L. Pereira, sp. nov. Figs 10-11

*MycoBank*: MB810171.

GenBank: KM514680 (ITS), KP862880 (RPB2).

Systematic position: Ascomycota, Ascomycetes, Xylariales, Xylariaceae.

Diagnosis: Similar to other Muscodor species, Muscodor coffeanum presents a sterile mycelium; it differs from other Muscodor species by producing hyphae and derived structures of different sizes (hyphae 1.5-5.5  $\mu$ m in diam.; coils 12-17  $\mu$ m in diam.). Additionally, M. coffeanum is phylogenetically distinct from other Muscodor spp.

Etymology: Named after its host genus, Coffea.

*Holotype*: BRAZIL, Viçosa, in the region of Zona da Mata in the state of Minas Gerais, on the stems of *Coffea arabica*, H. Z. Motter, November, 2011, (VIC42835 holotype, culture ex-type COAD 1842).

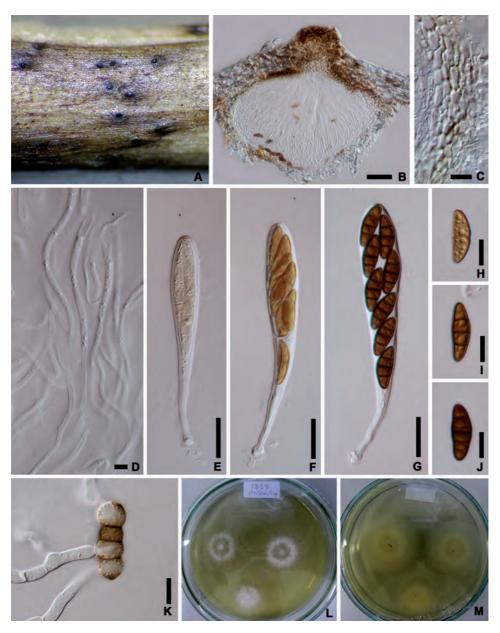


Fig. 9 *Montagnula scabiosae* (holotype). **A.** Appearance of ascomata on host substrate. **B.** Section of the ascoma. **C.** Section of the peridium cells. **D.** Pseudoparaphyses. **E-G.** Asci. **H-J.** Ascospores. **K.** Germinating spore. **L, M.** Colonies on PDA (M from below). Scale bars: **B** = 50  $\mu$ m, **C** = 10  $\mu$ m, **D** = 5  $\mu$ m, **E-G** = 20  $\mu$ m, **H-K** = 10  $\mu$ m.

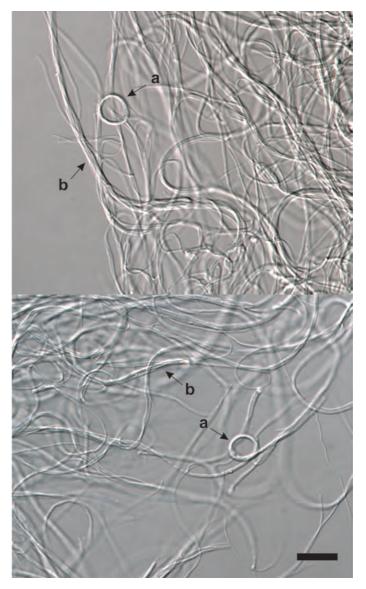


Fig. 10. *Muscodor coffeanum*. **a.** coils appear as fused rope-like hyphal strands. **b.** the hyphae intertwining and forming rope-like strands. Scale bars =  $20 \ \mu m$ .

Additional specimens examined. BRAZIL, Viçosa, in the region of Zona da Mata in the state of Minas Gerais, on the stems of *Coffea arabica*, H. Z. Motter, November 2011: *Muscodor coffeanum* (culture COAD 1899; ITS sequence GenBank KM514681) and *Muscodor coffeanum* (culture COAD 1900; ITS sequence GenBank KP862879 and RPB2 sequence GenBank KP862881).

**Hyaline hyphae** (1.5-5.5  $\mu$ m in diam.), thin-walled, septate, branched, frequently intertwining and forming rope-like strands 3-15  $\mu$ m wide; coils measuring 12-17  $\mu$ m in diam. appear as fused, rope-like, hyphal strands that are branching at

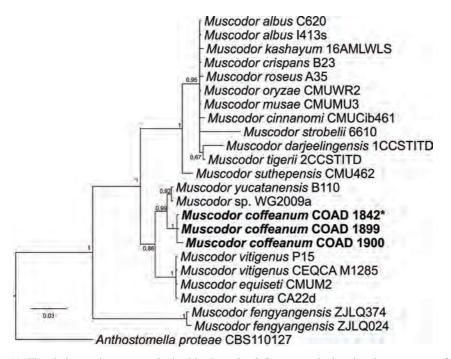


Fig. 11. The phylogenetic tree was obtained by Bayesian inference methods using the sequences of the ITS region. The posterior probability values (significant support threshold value = 0.95) are indicated at the nodes. The species from this study is highlighted in bold and type reference strains indicated. The analyses included 23 *Muscodor* specimens and was rooted with *Anthostomella proteae* (Xylariaceae) for out-group; the alignment contained 563 characters. The newly generated nucleotide sequences were edited with BioEdit software (Hall, 2014) and checked manually. The other sequences were retrieved from GenBank. The consensus sequences were aligned using the multiple sequence alignment program, MUSCLE<sup>®</sup> (Edgar, 2004), built in MEGA v. 5 software (Tamura *et al.*, 2011). The alignments were checked, and manual adjustments were performed when necessary. The alignment and tree were deposited into TreeBASE under accession number 17181. The phylogenetic analyses were conducted as described by Machado *et al.* (2014).

right angles. Mycelium sterile. **No conidia** or other sporulation structures were observed under laboratory conditions.

*Culture characteristics*: The colony grows slowly on potato dextrose agar (PDA), at a daily average of 2.3 mm, reaching 21-34 mm in diam. in 15 days at 25°C with a photoperiod of 12 hours. Older colonies (40 days) feature a soft beige colouration, flocculose surface, have an intense beige colouration on the reverse and produce a weak mouldy odour.

*Notes: Muscodor* is a genus of endophytic fungi characterized by the production of volatile organic compounds (VOCs) that inhibit the growth of other microorganisms (Strobel, 2011). Because of their potential for biofumigation, *Muscodor* species have been tested against a wide variety of plant and human pathogenic fungi (Goates & Mercier, 2009). A preliminary evaluation of the VOCs produced by *Muscodor coffeanum* showed that it completely inhibited growth of *Aspergillus ochraceous*, *A. niger*, *A. flavus* and *Fusarium semitectum* on PDA, but also of *A. ochraceous* when coffee beans were inoculated with a suspension of 10<sup>7</sup> conidia/ml

of the latter species. The newly proposed species is morphologically similar to M. *yucatanensis* (Gonzalez *et al.*, 2009); however, it produces smaller coils (the typical structure of the genus), thicker hyphae (Fig. 10) and a beige pigment on PDA. A megablast search of the NCBI GenBank nucleotide sequence database using the ITS sequence shows that M. *coffeanum* differs from M. *yucatanensis* (Accession No. FJ917287) by 7 bp over 563 nucleotides. Additionally, Bayesian inference analyses show that M. *coffeanum* is phylogenetically close to – although clearly distinct from – M. *yucatanensis* (Fig. 11). To our knowledge, this report is the first study concerning *Muscodor* on Rubiaceae and the first study of this genus in Brazil.

#### Genus Russula Pers., Observ. mycol. 1: 100. 1796.

In view of a forthcoming multigene phylogeny of the genus (Buyck *et al.*, in prep.), several new subgenera are here introduced for species having unequal gills:

# 17. Russula subg. Archaea Buyck & V. Hofstetter subg. nov.

*MycoBank*: MB 814390.

*Systematic position*: Basidiomycota, Agaricomycetes, Russulales, Russulaceae. *Etymology*: named after the type species.

Type species: R. archaea Heim, R. Heim, Candollea 7: 382. 1938.

Moderately large to small species, compact to very thin-fleshed. Cap dull coloured, yellowish, brownish or gray. Annulus never present. Gills irregularly unequal, with lamellulae either more or less abundant than normal gills. Context yellowing, browning, greying or reddening; mild to acrid. Spore print white. Secotioid and gasteroid representatives unknown.

Spores very small, with inamyloid suprahilar spot. Primordial hyphae absent. Gloeocystidia in all parts of the fruiting body, mucronate to obtuse. Hyphal extremities of cap surface variably inflated or not.

Ectomycorrhizal mantle with a plectenchymatic outer layer, producing abundant, emergent, hyphal extremities. Gloeocystidia inconspicuous, terminal, onecelled, minutely capitate with mostly one terminal knob Rhizomorphs common. Associations with mycoheterotrophic Orchidaceae and Ericaceae unknown.

*Notes*: This subgenus harbors species that belong in sections *Archaeinae* Buyck & Sarnari and *Gossypinae* Buyck.

## 18. Russula subg. Brevipes Buyck & V. Hofstetter subg. nov.

#### MycoBank: MB 814591.

Systematic position: Basidiomycota, Agaricomycetes, Russulales, Russulaceae.

*Etymology*: the name refers to the typically very short stipe that is characteristic for most species in this subgenus.

*Type species: Russula brevipes* Peck, Rep. (Annual) New York State Mus. Nat. Hist. 43: 20. 1890.

Mostly medium to very large species that are very thick-fleshed, exceptionally also small and thin-fleshed. Cap whitish, often rapidly developing yellowish brown to reddish brown stains. Well-developed annulus never present. Gills regularly unequal. Context turning yellowish to rusty brown, mostly with distinct smell, acrid to strongly acrid, (rarely mild ?). Spore print whitish to yellow. Secotioid and gasteroid representatives known.

Spores with inamyloid or amyloid suprahilar spot. Primordial hyphae absent. Gloeocystidia mucronate to obtuse-rounded, in all parts of the fruiting body. Hyphal extremities of cap surface inflated or not.

Ectomycorrhizal mantle with a plectenchymatic outer layer, covered with emergent, one-celled to secondarily septate, short, flask-shaped, mostly thick-walled gloeocystidia that are generally minutely capitate with one or rarely two, central knobs Rhizomorphs common. Associations with mycoheterotrophic Orchidaceae and Ericaceae known from subsection *Lactarioideae* Maire (only the *R.delica-brevipes* group).

*Notes*: This subgenus contains essentially species that belong to widespread subsections *Lactarioideae* Maire (sensu Bon) and *Pallidosporinae* Bon, but it is very likely that also the type species of sections *Delicoarchaeae* Singer and *Metachromaticae* Singer, as well as some other tropical whitish species with unequal gills will belong here. We have excluded the presence of a well-developed annulus in our definition of this subgenus although the Mexican *R. herrerae* was described as having an annulus. We consider, however, the arachnoid-cortinarioid tissue of the 'ring'-structure emanating from the stipe tissue in this species quite different from the development of the annulus in all other Russulas as a result of the proliferation of pileipellis tissue.

#### 19. Russula subg. Malodora Buyck & V. Hofstetter subg. nov.

#### *MycoBank*: MB 814391.

*Systematic position*: Basidiomycota, Agaricomycetes, Russulales, Russulaceae.

*Etymology*: refers to the frequently disagreeable smell that characterizes most species at maturity.

*Type species: R. compacta* Frost in Peck, New York St. Mus. Ann. Rept. 32: 32. 1879.

Medium to large species, often firm and compact, never extremely thinfleshed. Cap dull coloured, yellow brown, grey to almost black or whitish. Annulus never present. Gills regularly unequal to frequently and almost regularly forking. Greying, browning context, mostly developing rapidly a strongly disagreeable smell and taste. White spore print. Secotioid and gasteroid representatives unknown.

Spores with inamyloid suprahilar spot. Gloeocystidia moderately numerous to numerous on gill surface, mucronate and inconspicuous to absent elsewhere from the fruit body. Hyphal extremities of cap surface typically with inflated, often voluminous cells, and strongly septate.

Ectomycorrhizal mantle with a plectenchymatic outer layer, with dispersed to rare gloeocystidia that are emergent, one-celled, flask-shaped, minutely capitate with one or rarely two knobs, sometimes accompanied by emergent, apically tapering to cylindrical, thick-walled hyphal extremities Rhizomorphs common. Associations with mycoheterotrophic Orchidaceae and Ericaceae rare.

*Notes*: Apart from the type species, this subgenus contains some of the species that were previously placed in subsections *Meleagrinae* Buyck and *Brunneodermatinae* Buyck of section *Fistulosae* (Singer) Buyck.

As a result of the above new subgenera, subgen. Compactae is therefore now restricted to only some of the *Russula* species having unequal gills, viz. species of (sub)sections *Polyphyllinae* Singer, *Nigricantinae* Bataille and *Fistulosinae* Heim:

*Russula* subg. *Compactae* (Fr.) Bon, Doc. Mycol. 17 (65): 53. 1986, emend. Buyck & V. Hofstetter

Fruiting bodies very large to very small, thick-fleshed. Cap dull-coloured, white, brown, grey to black. Annulus never present. Gills regularly unequal. Context reddening, greying, blackening, rarely browning, with or without distinct, mostly

disagreeable smell, mild to very acrid. Spore print white. Secotioid and gasteroid representatives unknown.

Spores with inamyloid suprahilar spot. Gloeocystidia presence varying from present in all tissues to restricted to the hymenium only, mostly minutely capitate with one central knob, elsewhere often with two excentrical knobs, more rarely obtuse rounded. Hyphal extremities of cap surface inflated or not.

Ectomycorrhizal mantle with a plectenchymatic outer layer, covered with emergent, one-celled, flask-shaped gloeocystidia that are mostly mucronate with one central knob or, more frequently, two excentrical knobs Rhizomorphs common. Associations with mycoheterotrophic Orchidaceae and Ericaceae known from sect. *Nigricantinae* Bataille.

Type species: Russula nigricans Fr., Epicr. syst. mycol.: 350. 1838.

20. Scorias mangiferae Hongsanan, P. Chomnunti, J.C. XU & K.D. Hyde, sp. nov. Figs 12-13

> *Index Fungorum number*: IF 551526, *Facesoffungi number*: FoF 01060. *Etymology: mangiferae* referring to the host on which the taxon was found.

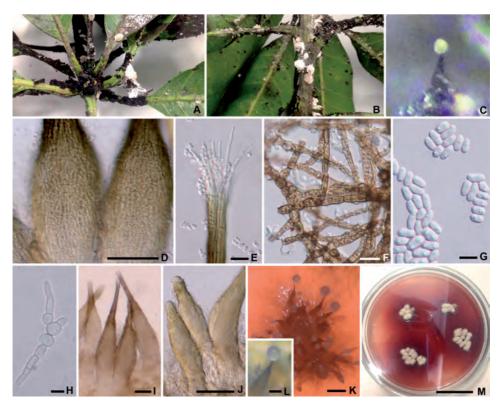


Fig. 12. *Scorias mangiferae* (holotype). **A.** Appearance of sooty mould on host. **B.** Insects (Mealy bugs) associated with *Scorias mangiferae*. **C.** Pycnidium. **D.** Pycnidia wall. **E.** Ostiole. **F.** Hyphal network. **G.** Conidia. **H.** Germinating conidium. **I-L.** Pycnidia developing on media. **M.** Colonies producing red pigmentation on PDA. Scale bars: **D. I**, **J**, **L** = 50  $\mu$ m, **K** = 100  $\mu$ m, **E-H** = 5  $\mu$ m, **M** = 3 cm.

*Systematic placement*: Ascomycota, Pezizomycotina, Dothideomycetes, Dothideomycetidae, Capnodiales, Capnodiaceae.

Holotype: MFLU 15-2254.

GenBank: KT588604 (ITS), KT588603 (LSU).

*Epiphytic, saprobic* on sugary exudates from insects growing on the surface of branches and living leaves. *Superficial hyphae* 4-6  $\mu$ m wide ( $\bar{x} = 5 \mu$ m, n = 10), branched, septate, slightly constricted at the septa, brown to dark brown dark brown

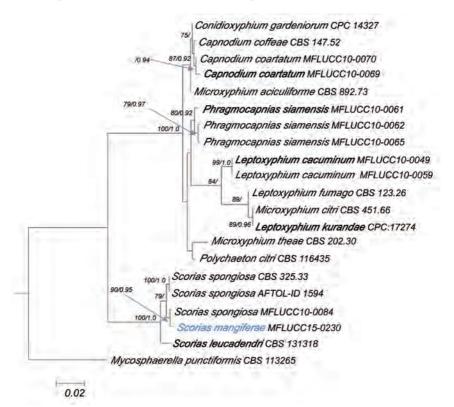


Fig. 13. The phylogenetic tree was obtained by RAxML maximum likelihood methods using sequences of ITS and LSU regions. The first set of numbers above the nodes are RAxML bootstrap value expressed expressed with values above 70% shown. The second set of numbers above the nodes are Bayesian posterior probabilities, with values above 0.95 shown. Strain numbers are indicated after species names. New sequence data are in blue bold, and other types are in black bold. The analyses included 5 strains from Scorias, 15 strains from others species in Capnodiaceae, and is rooted with Mycosphaerella punctiformis (Mycosphaerellaceae) for the out-group; the alignment comprises 1,325 characters. The newly generated nucleotide sequences were compared against the GenBank database using the Mega BLAST program. Sequences that relate to the Scorias were obtained from GenBank and were aligned using the multiple sequence alignment program, MAFFT (Katoh & Standley, 2013), checked manually using BioEdit software (Hall, 2014). Maximum-likelihood (ML) analysis was performed using raxmlGUIv.0.9b2 (Silvestro & Michalak, 2012), with 1,000 replicates. The model of evolution was estimated by using MrModeltest 2.2 (Nylander et al., 2008). Posterior probabilities were determined by Markov Chain Monte Carlo sampling (MCMC) in MrBayes v3.1.2 (Huelsenbeck & Ronquist, 2001). Six simultaneous Markov chains were run for 1,000,000 generations and trees were sampled every 100th generation, and 10,000 trees were obtained. The first 2,000 trees, representing the burn-in phase were discarded (Cai et al., 2006, 2008). Phylogenetic trees were drawn by using Treeview v. 1.6.6 (Page, 2001).

towards the edge, forming as thallus cover the surface of plants. Asexual morph: *Pycnidia* 295-385 high × 46-55 µm diam. ( $\bar{x} = 370 \times 53$  µm, n = 5), gregarious, arising from the hyphae, long stalked, flask-shaped, brown to pale brown at the base, brown to dark brown towards the tapering apex. *Ostiole* surrounded by septate, hyaline hyphae, with rounded tips. *Pycnidial wall* helical twisting, synnemata-like. *Conidiogenous cells* formed in the inner cells of the oval part. *Conidia* 6-7 × 2-3 µm ( $\bar{x} = 6.7 \times 2.5 \mu$ m, n = 10), ellipsoidal, unicellular, hyaline to greenish, with rounded ends. Sexual morph: Undetermined.

*Culture on PDA*: Conidia germinating on PDA at 27°C for 12 h of light/12 h of dark, germ tubes appearing from conidia, hyaline, and becoming darkened after 10 days. Colonies reaching 1 cm diam. after 5 days on PDA at 25-28°C, colonies superficial to erumpent, velvety, thin at the margin, water droplets forming on the surface of colonies, white to ivory, darkened and greenish at the margin after 14 days incubation, produce abundant pycnidia and red pigment on PDA after 5 days incubation.

*Material examined*: THAILAND, Chiang Rai Province, Bandu, on branch of *Mangifera* sp. (Anacardiaceae), 4 February 2015, S. Hongsanan BJ01 (MFLU 15-2254, holotype), (isotype in KUN), ex-type living culture, MFLUCC 15-0230, in BCC.

*Notes*: The genus *Scorias* was established by Fries (1832), with the type species *Scorias spongiosa* (Schwein.) Fr. Species of *Scorias* grow on insect honeydew or sugary plant exudates (Hughes, 1976; Reynolds, 1978; Schoch *et al.*, 2006; Chomnunti *et al.*, 2011, 2014), and is characterized by subglobose to broadly ellipsoidal ascomata, bitunicate asci, and 3-4-trans-septate, hyaline to pale brown ascospores. The asexual morph *Scorias* is characterized by superficial hyphae covering the surface of hosts, long or short-stalked, flask-shaped pycnidia, arising from hyphae, and hyaline, unicellular conidia tapering towards the apex (Chomnunti *et al.*, 2011, 2014). Phylogenetic analyses indicated that the genus *Scorias* belongs in Capnodiaceae (Schoch *et al.*, 2006, 2009; Crous *et al.*, 2009; Chomnunti *et al.*, 2011, 2014; Hyde *et al.*, 2013). There are ten known species in this genus, but only two species have molecular data.

*Scorias mangiferae* is most typical of *S. spongiosa* (Schwein.) Fr. in having hyphal networks covering the surface of hosts, and flask-shaped pycnidia arising from the hyphae, producing unicellular, hyaline conidia. However, our species differs from all other known species in the genus in its conidiomata having short stalks, which are brown to yellowish when immature, becoming brown at the base and darkened in the upper part when mature, and in producing larger, hyaline conidia. Molecular analyses using combined LSU and ITS sequence data indicate *S. mangiferae* is a distinct species (Fig. 13), which is related to *S. spongiosa* (90% ML and 0.95 PP support).

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