ANDRÉ LUIZ FIRMINO

# NEW TAXA AND PHYLOGENETIC PLACEMENT OF ASTERINALES FROM BRAZIL

Dissertação apresentada à Universidade Federal de Viçosa, como parte das exigências do Programa de Pós-Graduação em Fitopatologia, para obtenção do título de *Magister Scientiae*.

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APROVADA: 27 de fevereiro de 2013.

Harold Charles Evans

Robert Weingart Barreto

Olinto Liparini Pereira
(Orientador)

“Escolhe um trabalho de que gostes, e não terás
que trabalhar nem um dia na tua vida”.
(Confúcio)

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## BIOGRAFIA

ANDRÉ LUIZ FIRMINO, filho de Selma Regina Pacheco e Helson José Firmino (*in memoriam*), nasceu na cidade de São Paulo-SP no dia 6 de novembro de 1986.

Cursou o ensino fundamental em São Paulo-SP e Santo Antônio do Grama-MG e o ensino médio na cidade de Rio Casca-MG.

No ano de 2006, ingressou no curso de Engenharia Florestal na Universidade Federal de Viçosa, graduando-se em janeiro de 2011.

Em fevereiro de 2011, iniciou o Mestrado no Programa de Pós-graduação em Fitopatologia, na Universidade Federal de Viçosa, submetendo-se à defesa de dissertação em 27 de fevereiro de 2013.

No mesmo ano, ingressou no Doutorado em Fitopatologia, na mesma instituição.

**SUMÁRIO**

# RESUMO

FIRMINO, André Luiz, M.Sc., Universidade Federal de Viçosa, Fevereiro de 2013.
**Novos taxa e posicionamento filogenético da ordem *Asterinales* do Brasil.**
Orientador: Olinto Liparini Pereira. Coorientador: José Luiz Bezerra.

Folhas vivas de 18 espécies de plantas de 10 famílias botânicas apresentando colônias escuras suspeitas de pertencerem à ordem *Asterinales* foram coletadas em áreas de Mata Atlântica do Brasil em várias viagens de campo durante 2009–2012. As viagens de campo foram conduzidas em quatro estados brasileiros e em seis áreas: Minas Gerais (Mata do Paraíso, Mata do Seu Nico, Mata da Biologia e próximo a Nova Lima), Espírito Santo (Reserva Natural da Vale do Rio Doce), Bahia (Reserva Ecológica da Michelin e Serra da Jibóia) e Rio Grande do Sul (próximo a Bento Gonçalves). O exame da morfologia dos fungos nas amostras indicam que havia, 13 prováveis novas espécies, 4 primeiros relatos para o Brasil e 5 novos hospedeiros para espécies conhecidas de Asterinales. Os fungos são descritos e ilustrados. Uma análise filogenética combinada de sequências ribossomais (18S e 28S rDNA) e codificadores de proteínas (TEF e RPB2) confirmaram que *Asterinales* formam um clado monofilético bem suportado dentro de *Dothideomycetes*. A relação entre *Schizothyriaceae* e *Mycosphaerellaceae* dentro de *Capnodiales* e a não relação entre *Asterinales* e *Capnodiales* foram corroboradas.

# ABSTRACT

FIRMINO, André Luiz, M.Sc., Universidade Federal de Viçosa, February 2013. **New taxa and phylogenetic placement of *Asterinales* from Brazil.** Adviser: Olinto Liparini Pereira. Co-adviser: José Luiz Bezerra.

Infected living leaves of different host plants showing black colonies belonging to *Asterinales* were collected in areas of Atlantic forest of Brazil in field trips during 2009–2012. The field trips were conducted in four Brazilian states and six areas: Minas Gerais (Mata do Paraíso, Mata do Seu Nico, Mata da Biologia and near Nova Lima), Espírito Santo (Reserva Natural da Vale do Rio Doce), Bahia (Reserva Ecológica da Michelin and Serra da Jibóia) and Rio Grande do Sul (near Bento Gonçalves). Through morphological and biometric data, 13 probably new species, 4 first reported to Brazil and 5 new host records are described and illustrated. A combined phylogenetic analysis of the ribosomal sequences (18S and 28S rDNA) and protein-coding (TEF and RPB2) confirmed that *Asterinales* form a monophyletic clade well supported within the *Dothideomycetes*. The sister relationship of *Schizothyriaceae* and *Mycosphaerellaceae* within the *Capnodiales* and the non-relationship between *Asterinales* and *Capnodiales* were corroborated. A revision of the genera of the families *Asterinaceae* and *Englerulaceae* shows a small divergence in the number of genera when compared with others authors. An index of *Asterinales* from Brazil containing 151 species distributed in 47 botanical families, including two families: *Asterinaceae* and *Englerulaceae* is also provided.

# 1. INTRODUÇÃO GERAL

A classificação dos fungos pertencentes à ordem *Asterinales*, assim como ordens de fungos que apresentam como principal característica, o ascoma dimidiato, sempre se baseou exclusivamente em características morfológicas, levando a interpretações divergentes por parte de diferentes especialistas.

A primeira descrição conhecida de fungo pertencente à atual ordem *Asterinales* é a do gênero *Triposporium* Corda, em 1837, um anamorfo do gênero *Batistinula* Arx, em 1960. Léveillé (1845) descreveu os dois gêneros que possuem o maior número de espécies dentro da ordem *Asterinales*: *Asterina* (*A. melastomatis*, *A. compacta*, *A. pulla* e *A. azarrae*) e *Lembosia* (*L. tenella*, *L. macula*, *L. dendrochili*, e *L. drymidis*). Em 1899, *Asterina* foi incluída em *Microthyriaceae* e a família foi divida em duas subfamílias, *Asterineae* e *Microthyrieae* devido à presença ou ausência de micélio superficial (Theiss. 1913 a, b). Hansford (1946) descreveu a família *Asterinaceae* incluindo 13 gêneros em sua publicação.

A primeira descrição da ordem *Asterinales*, abrangia os fungos que possuiam ascocarpos cleistotecioides ou achatados, superficiais, epifíticos ou hiperparasitas; com peridio delgado (frequentemente compostos por uma camada simples), sem poro apical, com ascocarpo abrindo-se por divisão entre as células paralelas, desintegração ou gelatinização das células apicais. Nesta classificação incluíam-se as famílias: *Saccardiaceae, Vizellaceae, Stephanothecaceae, Schizothyriaceae, Brefeldiellaceae, Capnodiaceae, Asterinaceae, Englerulaceae e Parodiopsidaceae.* Note-se que a família *Parmulariaceae* estava ausente do grupo nesta classificação, tendo sido alocada na ordem *Hysteriales*, onde se encontravam os fungos que possuem ascocarpos apotecioides ou histerotecioides, arredondados ou elongados, sapróbios,

parasitas ou liquenizados; com peridio pseudoparenquimatoso ou prosenquimatoso; pseudoparáfises frequentemente largas ou ramificadas e/ou pigmentadas e ascosporos geralmente assimétricos (Barr 1979).

Atualmente os fungos que compreendem a ordem *Asterinales* são caracterizados por serem epifíticos ou hiperparasitas, biotróficos, com ascas bitunicadas e geralmente globosas, podendo ser ovais ou cilíndricas, ascomas escutelares-dimidiatos que crescem superficialmente sobre as folhas ou caules das plantas, com ou sem micélio superficial, sendo que estes podem conter ramificações opostas, alternas ou irregulares; hifopódios unicelulares ou bicelulares, podendo ser opostos, alternados ou alternados e opostos, sendo ovais, conóides, sublobados, lobados ou variáveis em forma; Haustórios presentes em vários gêneros, hamatécio ausente, ascomas não ostiolados, abrindo-se irregularmente à maturidade por meio de fendas estelares, longitudinais, irregulares ou por deliquescência parcial da parede superior e apresentando hipostroma intramatricial presente somente em alguns gêneros da família *Parmulariaceae* (Arx & Müller 1975; Eriksson 1981; Bezerra 2004; Hosagoudar 2012).

*Asterinales* são frequentemente confundidos em observações superficiais com os fungos da Ordem *Meliolales* pela presença do micélio epifítico negro e hifopodiado. Porém os *Meliolales* possuem células conidiogênicas (fiálides), ascas unitunicadas e ascosporos geralmente com 4 septos (Hansford 1961). Além disso, são filogeneticamente distantes dos *Asterinales* por estarem dentro da ordem *Sordariales* (Bezerra 2004).

A ordem *Microthyriales* possui representantes com e sem micélio hifopodiado assim como os *Asterinales*, mas possuem abertura ostiolar assim com os *Meliolales* (Bezerra 2004), além de possuírem ascas inclinadas. Entretanto, diversos gêneros se encontram em posição duvidosa. Após re-exame de materiais depositados em herbários, Wu *et al.* (2011), transferiram

11 gêneros da família *Microthyriaceae* para *Asterinaceae*, a saber: *Asterinema*, *Asteritea*, *Govindua*, *Maublancia*, *Polycyclinopsis*, *Polystomellina*, *Resendea*, *Trichopeltospora* e *Xenostomella* (*Asterinaceae*); *Phaeothyriolum* e *Platypeltella* (*Asterinaceae* incertae sedis). Adicionalmente transferiram 2 gêneros para *Aulographaceae*, a saber: *Cirsosina* e *Lembosiella*. Estes autores também supõem que as famílias *Aulographaceae, Asterinaceae* e *Parmulariaceae* possuem poucas diferenças morfológicas e podem ser consideradas sinônimas. Entretanto essa opinião não é amplamente aceita.

Alguns representantes da ordem *Asterinales* aqui tratados, tiveram quatro loci gênicos sequenciados, sendo: nuc SSU rDNA, nuc LSU rDNA, *TEF1* e *RPB2*, que são usados para o posicionamento de Ordens dentro da Classe *Dothideomycetes* (Schoch *et al.* 2006).

Até o presente momento, não há um posicionamento filogenético da ordem *Microthyriales* dentro da classe *Dothideomycetes*.

Batista e colaboradores foram os micologistas que mais estudaram os *Asterinales* no Brasil, coletando na Mata Atlântica do nordeste brasileiro e recebendo materiais provenientes de todas as regiões do Brasil, principalmente da região amazônica (Bezerra, 2004). Nos últimos dez anos, os trabalhos publicados envolvendo "*Asterinales* brasileiros" têm se concentrado na família *Parmulariaceae* (Inácio C & Inácio 2003; Inácio & Cannon 2008; Inácio *et al*. 2011, 2012) e havendo apenas descrições eventuais e isoladas de *Asterinaceae* (Inácio & Cannon 2003; Silva & Pereira 2008; Bezerra *et al*. 2010) e nenhum trabalho de *Englerulaceae*. Entretanto, os estudos realizados por Batista e colaboradores e Inácio ainda são insuficientes para amostrar os gêneros e as espécies que ocorrem no país. Atualmente pouquíssimos pesquisadores se dedicam a essa extensa ordem.

O estudo de grande diversidade de "*Asterinales* brasileiros" pode contribuir significativamente para um melhor entendimento da filogenia do grupo, especialmente trabalhos envolvendo novas regiões gênicas, incluindo codificadores de proteínas.

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# ARTIGO

## New taxa and phylogenetic placement of Asterinales from Brazil

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**Key words:** *Asterinaceae*, *Englerulaceae*, phylogeny, taxonomy, tropical fungi

### INTRODUCTION

*Asterinales* is one of the largest orders within the class *Dothideomycetes*, represented by 3 families: *Asterinaceae* with 46 genera and 653 species, *Englerulaceae* with 11 genera and 106 species and *Parmulariaceae* with 34 genera and 119 species (Erikson 1981; Kirk *et al.* 2008; Hofmann *et al.* 2010). However there is a disagreement between authors about these numbers and regarding the number of species in the genus *Asterina*. Hosagoudar & Abraham (2000) report 578 species and Hofmann & Piepenbring (2011) estimated a number between 578 to 708 species. Although there are more than 1000 species listed in Mycobank (http://www.mycobank.org) a number that probably includes synonyms and it must be reviewed with more caution.

The history of the current Order *Asterinales* began with the description of the genus *Triposporium* Corda, in 1837 (Corda 1837), where is the anamorph of *Batistinula* Arx (Arx 1960). Léveillé (1845) described the two genera having the greatest number of species within the 0*Asterinales*: *Asterina* (*A. melastomatis*, *A. compacta*, *A. pulla* and *A. azarrae*) and *Lembosia* (*L.*

*tenella*, *L. macula*, *L. dendrochili*, and *L. drymidis*). In 1899, *Asterina* was included in *Microthyriaceae* and the family has been divided into two subfamilies: *Asterineae* and *Microthyrieae* due to the presence or absence of superficial mycelium (Theissen 1913 a, b). Later Hansford (1946) described the family *Asterinaceae* and included 13 genera. In 1979 the order *Asterinales* was proposed by Barr to accommodate members of *Dothideomycetes* having clestothecioid or flattened ascocarps that are superficial, epiphytic or hyperparasitic; a thin peridium (often only a single layer), without apical pore, opening by crumbling or gelatinization of apical cells or splitting between rows of cells. In this classification included the families: *Saccardiaceae*, *Vizellaceae*, *Stephanothecaceae*, *Schizothyriaceae*, *Brefeldiellaceae*, *Capnodiaceae*, *Asterinaceae*, *Englerulaceae* and *Parodiopsidaceae*. Note that the family *Parmulariaceae* was not included, being allocated in the order *Hysteriales*, where were fungi that having apothecioid or hysterothecioid ascocarps, discoid or cupulate in section, rounded or elongate, saprobic, parasitic or lichenized; peridium pseudoparenchymatous or prosenchymatous; pseudoparaphyses above asci often enlarged or branched and/or pigmented to form an epithecium or pseudoepithecium; ascospores mostly asymmetric (Barr 1979).

Currently the order *Asterinales* includes species that are epiphytic or hyperparasitic, obligately biotrophic, with bitunicate asci that generally are globose, and may be oval or cylindrical, ascomata dimidiate that grow on surface of leaves or stems of plants, with or without superficial mycelium, that may have opposite, alternate or irregular branches; Apressoria that are uni or bicellular, and may be opposed, alternate or alternate and opposite, having shapes varying between oval, ampulliform, lobate or variable; haustoria are present in various genera; hamatecium is absent; dehiscence is not ostiolate, but opening irregular at maturity by means of stellar slits, longitudinal, irregular or partial deliquescence of the upper wall and only presenting

hypostroma (internal stroma or internal hyphae) in some members of the *Parmulariaceae*, (von Arx & Müller 1975; Eriksson 1981; Bezerra 2004; Hofmann *et al.* 2010; Hofmann & Piepenbring 2011; Hosagoudar 2012).

Wu *et al.* (2011) examined and transfered nine genera of the family *Microthyriaceae* to *Asterinaceae*: *Asterinema*, *Asteritea*, *Govindua*, *Maublancia*, *Polycyclinopsis*, *Polystomellina*, *Resendea*, *Trichopeltospora* and *Xenostomella*.

The classification within the order *Asterinales* is still based exclusively on morphological characteristics (Figure 1), but phylogenetic studies of *Asterinales* are also needed to establish the value of such morphological features in the delimitation of genera and species

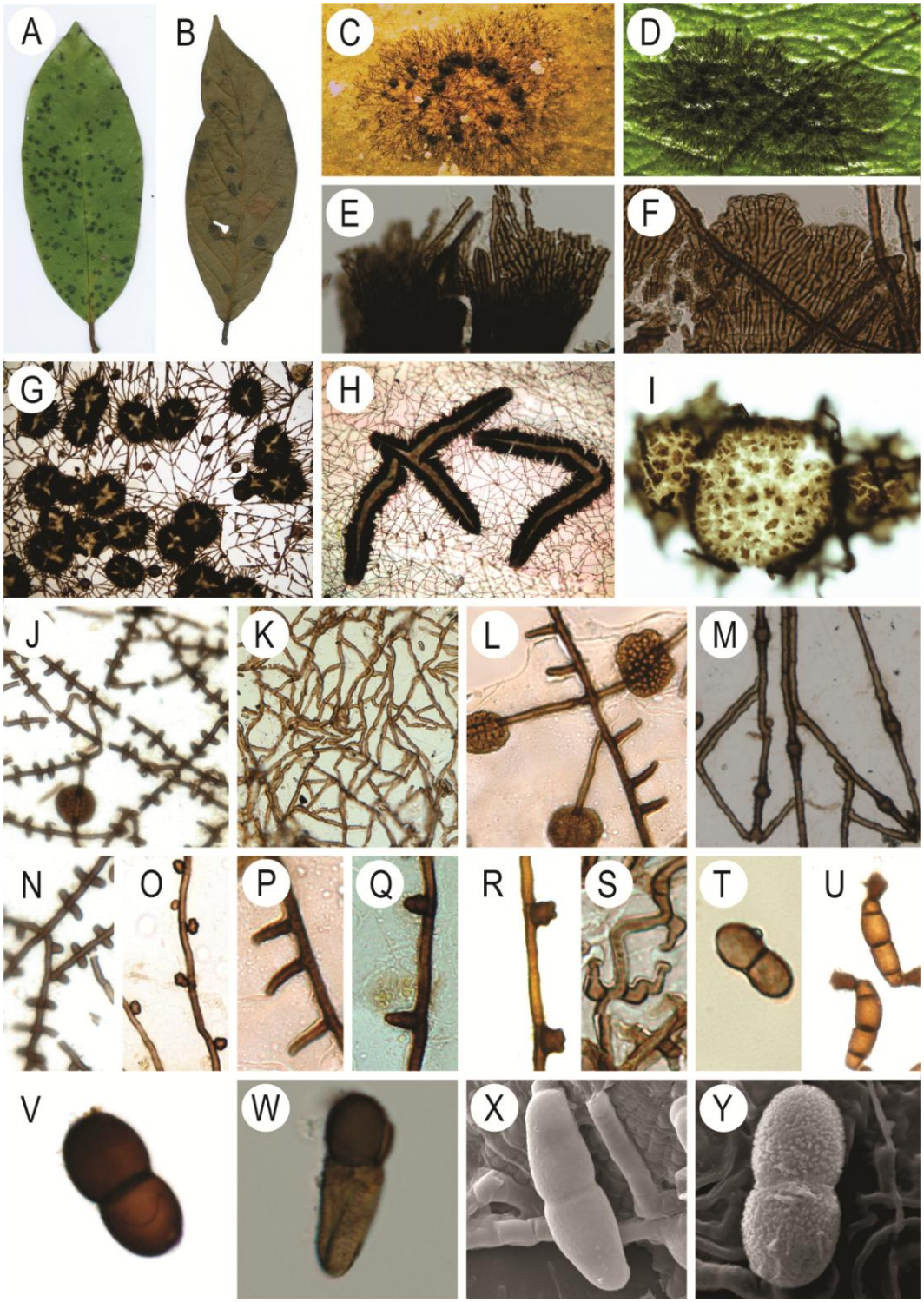


**Fig. 1.** Main morphological characteristics used in the taxonomy of the *Asterinales*. A. Epiphyllous colonies. B. Hypophyllous colonies. C. Single colony. D. Confluent colony. E. Radiate scutellum. F. Reticulate scutellum. G. Stellar slits. H. longitudinal slits. I. Partial deliquescence of the upper wall. J. Mycelium with apressoria. K. Mycelium without apressoria. L. Lateral apressoria. M. Intercalary appressoria. N-S Apressoria: N. Opposites. O. Alternate. P. Unilateral. Q. Rounded. R. Lobate. S. Pyriform. Ascospore: T. Uniseptate. U. Multiseptate. V. Constricted at the middle. W. Non central constriction. X. Smooth. Y. Verruculous.

## MATERIAL & METHODS

### Morphology

Infected living leaves of different host plants showing black colonies possibly belonging to *Asterinales* were collected in areas of Atlantic forest of Brazil in field trips during 2009–2012 in three Brazilian states: Minas Gerais (Mata do Paraíso, Mata do Seu Nico, Mata da Biologia and near to Nova Lima), Espírito Santo (Reserva Natural da Vale do Rio Doce) and Bahia (Reserva Ecológica da Michelin).

Fresh leaves were photographed, dried, and examined under an Olympus SZ 40 stereomicroscope. The nail polish technique was used to observe the colonies (Hosagoudar & Kapoor 1984). Individual fungal structures from the plant surface were removed with a scalpel and mounted in lactophenol. Observations and measurements were made with a Carl Zeiss Standard W light microscope (Göttingen, Germany) and photographs were taker on an Olympus BX 51 microscope equipped with a digital camera E-volt 330. For scanning electron microscopy (SEM), air-dried material was mounted directly and coated with a thin layer of gold using a sputter coater (Balzers® model FDU 010) for 2 min. Photographs were obtained using a Carl-Zeiss Model LEO VP 1430 scanning electron microscope. Biometric data were based on 30 measurements of structures for mycelial hyphae, apressoria, ascomata and ascospores.

### DNA extraction

DNA of the fungi was extracted from selected ascomata of *Asterinales* on infected leaves (free from mycoparasites) following the methods described by Pinho *et al*. (2012) using Wizard® Genomic DNA Purification Kit (Promega Corporation, WI, U.S.A.).

## PCR and Sequencing

The four genes used for nucleotide sequence comparisons are partial regions of nuclear large subunit (nuc LSU rDNA), nuclear small subunit (nuc SSU rDNA), elongation factor gene (*TEF1*) and the second largest subunits of RNA polymerase II gene (*RPB2*) (see Table 1). PCRs reactions were run under conditions described previously (Pinho *et al.* 2012). PCR products were purified and sequenced by Macrogen Inc., Korea (http://www.macrogen.com).

**Table 1.** Primers used in this work.

| Gene | Primers | Primer Sequences (5'– 3') | Authors |
|---|---|---|---|
| LSU | LR0R | ACCCGCTGAACTTAAGC | (Vilgalys & Hester 1990) |
| | LR5 | TCCTGAGGGAAACTTCG | (Kodsueb *et al*. 2006) |
| SSU | NS1 | GTAGTCATATCG TTGTCTC | (White *et al*. 1990) |
| | NS4 | CTTCCGTCAATTCCTTTAAG | (Tsui *et al*. 2006) |
| *TEF1* | 983F | GCYCCYGGHCAYCGTGAYTTYAT | (Rehner 2001) |
| | 2218R | ATGACACCRACRGCRACRGTYTG | (Rehner 2001) |
| *RPB2* | 5F2 | GGGGWGAYCAGAAGAAGGC | (Liu *et al*. 1999) |
| | 7cR | CCCATRGCTTGYTTRCCCAT | (Liu *et al*. 1999) |

## Phylogenetic analysis

For phylogenetic analysis, sequences of the nuc LSU rDNA, nuc SSU rDNA, *TEF1* and *RPB2* obtained in this work and retrieved from Schoch *et al.* (2009) in TreeBase (http://www.treebase.org/). The nucleotide sequences were edited with the DNA Baser v.3 (Heracle BioSoft S.R.L., Pitesti, Romania). The sequences were checked manually and aligned using the multiple sequence alignment program MUSCLE® (Edgar 2004) built in MEGA v. 5

software (Tamura *et al*. 2011). For the Bayesian inference (BI) the nucleotide substitution models through the MrMODELTEST 2.3 (Posada & Buckley 2004) was used. As soon as the likelihood scores were calculated, the models were selected in Akaike Information Criterion (AIC). Four Analysis of the Markov Chain Monte Carlo method (MCMC) chains were run simultaneously, starting from random trees for 10 000 000 generations. Trees were sampled every 1000$^{th}$ generation for a total of 10 000 trees. The first 2 500 trees were discarded as the burn-in phase of each analysis. Posterior probabilities (Rannala & Yang, 1996) were determined from a majority-rule consensus tree generated with the remaining 7 500 trees.

A phylogenetic analysis of the alignment was performed by CIPRES webportal (Miller *et al.* 2010) using MrBayes v.3.1.1 (Ronquist & Huelsenbeck 2003). Trees were visualized in FigTree (Rambaut 2009). *Laurela megasperma* and *Trypethelium nitidiusculum*, members of *Trypetheliales* as outgroup were used as outgroup in these analysis.

## RESULTS

### Phylogenetic analysis of Asterinales

Representative sequences from orders related to *Asterinales* were utilized for the combined phylogenetic analysis (Fig. 2). The first phylogenetic study based on DNA sequences of 18S and 28S rDNA showed that *Asterinales* form a well-supported monophyletic clade within the *Dothideomycetes* (Hofmann *et al*. 2010). In the same study, the authors addressed the need for sequences of protein-coding for true positioning of the order.

With the use of ribosomal sequences (18S and 28S rDNA) and protein-coding (TEF and RPB2) we confirmed that *Asterinales* form a monophyletic clade well supported within the *Dothideomycetes*. The present study corroborates the sister relationship of *Schizothyriaceae* and

*Mycosphaerellaceae* within the *Capnodiales* and a distant relationship between *Asterinales* and *Capnodiales*. Future studies are also needed to establish the value of specific morphological features in the delimitation of genera and species within the *Asterinales*.

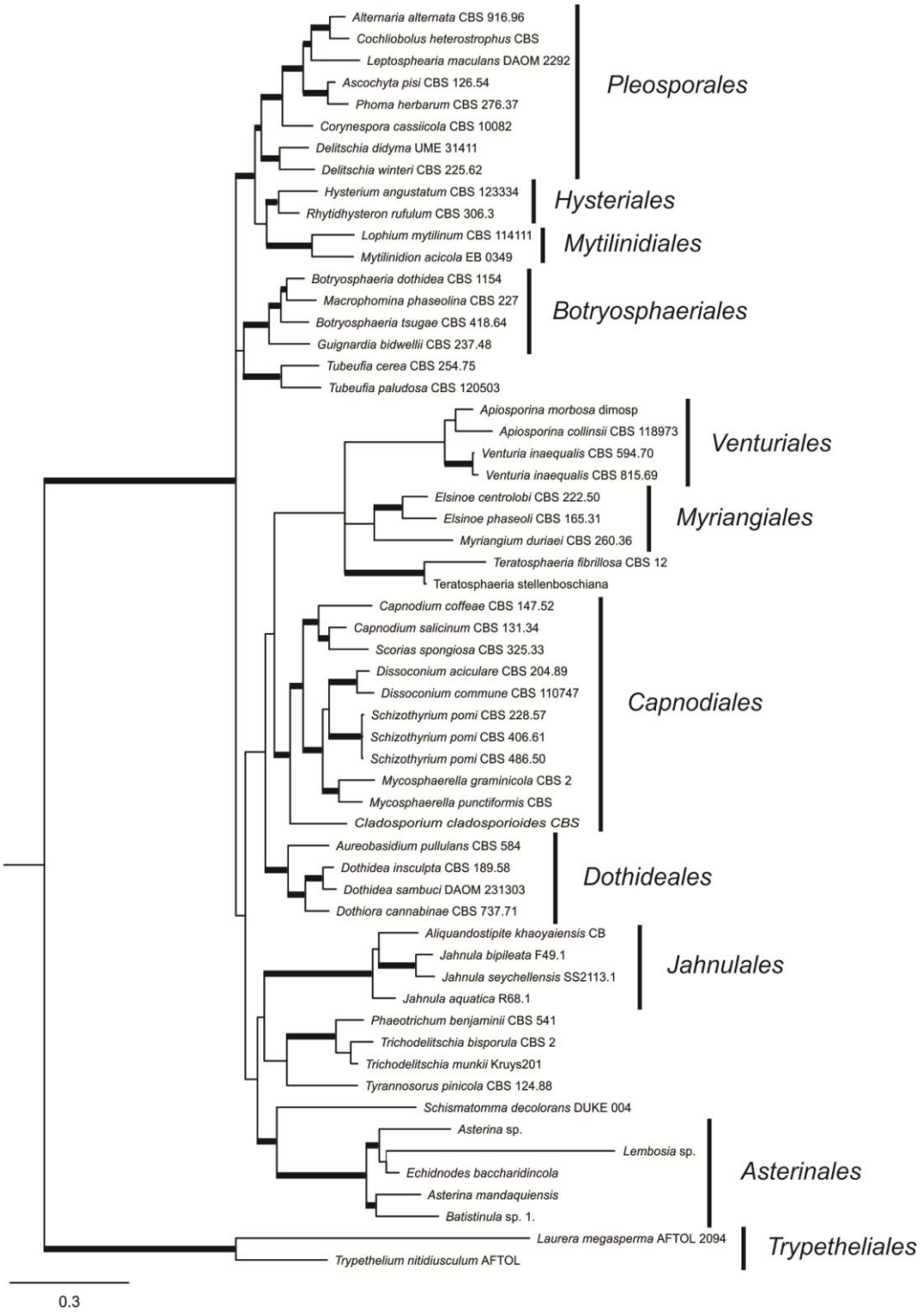


**Fig. 2.** Bayesian inference tree from obtained from a data set of 58 taxa including representatives of all known orders in *Dothideomycetes* and potential species included in *Asterinales* comparing four genes (SSU, LSU, TEF and RPB2) with *Laurela megasperma* and *Trypethelium nitidiusculum*, members of *Trypetheliales* as outgroup. Only values above 96% of posterior probability on Bayesian tree are thickened. Culture and voucher numbers are indicated after species names.

## Taxonomy

### Members of the family *Asterinaceae*

***Asterina claviflori*** Kar & Maity. Transactions of the British Mycological Society. 54(3):435–444. 1970. Fig. 3.

*Colonies* epiphyllous, circular, single, becoming confluent, black. *Hyphae* straight to slightly flexuous, branching unilateral or alternate, pale brown to brown, septate, hyphal cells cylindrical, 4.5–6 µm, smooth. *Apressoria* numerous, entire, sessile, straight to crooked, elliptic to angular, unicellular, alternate or unilateral, never opposed, 10–17.5×5–7.5 µm, brown, penetration peg in central part of apressorial cell. *Thyriothecia* superficial, develop below surface mycelium, circular, single to confluent, fringed at margins, 152–304 µm diam., dark brown to blackish, open with central star-shaped fissures. *Scutellum* radiate, composed of cells isodiametric to cylindrical, straight. *Asci* ovoid to globose, bitunicate, 8 ascospores, hyaline. *Ascospores* 2–celled, cylindrical to ellipsoid, straight, constricted at central septum, 30–37.5×12.5–15 µm, pale brown to brown, smooth. *Anamorph* absent.

***Notes*:** Three specimens collected were identified as being *Asterina clavifolia*. All of them were collected in Reserva Natural Vale do Rio Doce from leaves of *Eugenia punicifolia*, *Eugenia santensis* and *Eugenia* sp. *Asterina clavifolia* was described by Kar & Maity in 1970 in India on leaves of *Syzygium claviflorum*. This is the first record of *A. clavifolia* in Brazil and *E. punicifolia* and *E. santensis* are new records of host plant species. This species is illustrated here for the first time.

*Specimens examined*: Brazil, Espírito Santo, Reserva Natural Vale do Rio Doce from leaves of *Eugenia punicifolia*, *Eugenia santensis* and *Eugenia* sp. (*Myrtaceae*), 2012. A.L. Firmino.

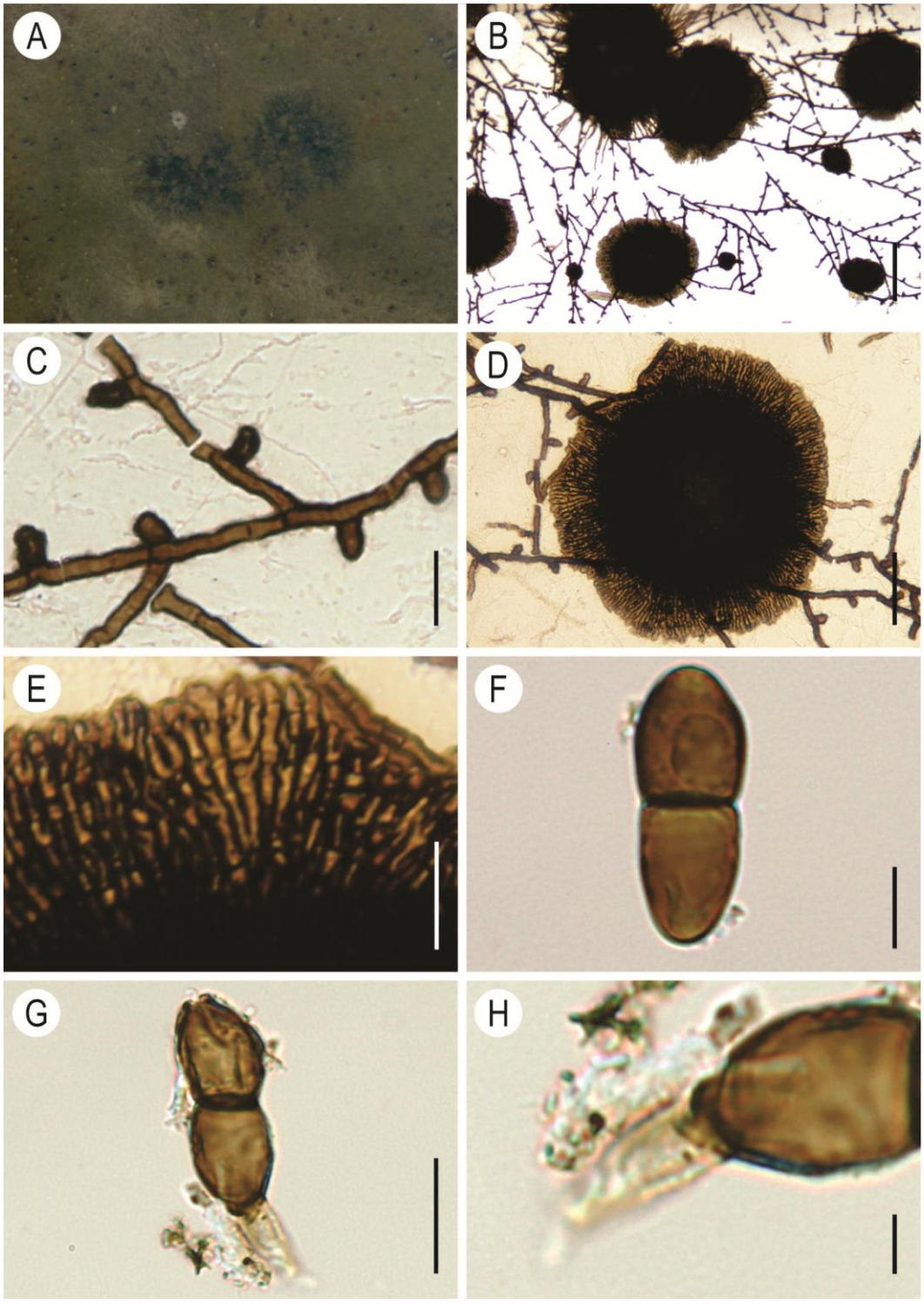


**Fig. 3.** *Asterina claviflori*. A. Colonies epiphyllous. B. Part of a colony with open thyriothecia and surface mycelium. C. Hyphae, branching and apressoria. D. Thyriothecia. E. Fringed at margins. F. Ascospore. G. Ascospore germination. H. Detail of the germination with hyaline hyphae. Scale bars: B=200 µm; C–E–G= 20 µm; D= 100 µm; F= 10 µm; H= 5 µm.

***Asterina colliculosa*** Speg. Boletín de la Academia Nacional de Ciencias en Córdoba. 11(4):381–622. 1889. Fig. 4.

*Colonies* epiphyllous, circular, single, becoming confluent, black. *Hyphae* straight to slightly ondulate, branching unilateral or alternate, brown to dark brown, septate, hyphal cells cylindrical, 5–7 µm, smooth. *Apressoria* numerous, entire, sessile, straight, facing forward, oblong to angular, unicellular, alternate or unilateral, rarely opposed, 9–10×6–7.5 µm, brown, penetration peg in central part of apressorial cell. *Thyriothecia* superficial, develop below surface mycelium, circular, single to confluent, fringed at margins, 147–218 µm diam., dark brown to blackish, open with central star-shaped fissures. *Scutellum* radiate, composed of cells isodiametric to cylindrical, straight. *Asci* ovoid, bitunicate, 8 ascospores, hyaline. *Ascospores* 2–celled, oblong to ellipsoid, straight, constricted at central septum, 27.5–32.5×12–15 µm, brown to dark brown, smooth. *Anamorph* absent.

***Notes*:** One specimen collected was identified as being *Asterina colliculosa*. It was collected in Reserva Natural Vale do Rio Doce from leaves of *Eugenia sulcata*. *Asterina colliculosa* was described by Spegazzini in 1889 on leaves of *Eugenia* sp. This is the first record of *A. colliculosa* in Brazil and *E. sulcata* is new record of host plant species. This species is illustrated here for the first time.

*Specimens examined*: Brazil, Espírito Santo, Reserva Natural Vale do Rio Doce from leaves of *Eugenia sulcata* (*Myrtaceae*), 2012. A.L. Firmino.

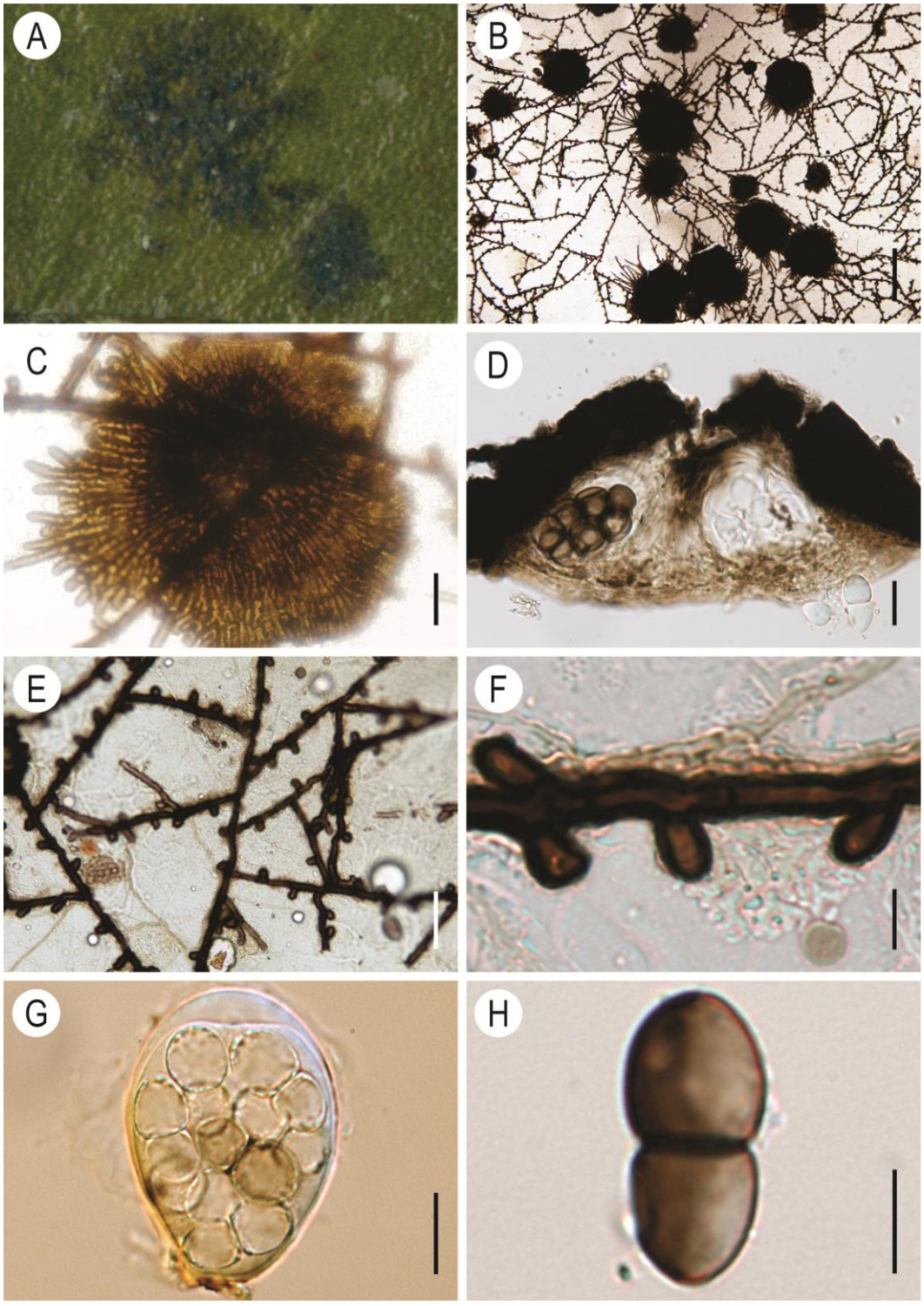


**Fig. 4.** *Asterina colliculosa*. A. Colonies epiphyllous. B. Part of a colony with open thyriothecia and surface mycelium.. C. Initial young thyriothecia. D. Cross section of the thyriothecia. E. Mycelium branching. F. Apressoria. G. Bitunicate ascus, ovoid with immature ascospore. H. Brown and smooth ascospore. Scale bars: B=200 µm; C–D–G= 20 µm; E= 50 µm; F–H= 10 µm.

***Asterina mandaquiensis*** Henn. Hedwigia 48:12. 1908. Fig. 5.

*Colonies* epiphyllous, circular, single, becoming confluent, black. *Hyphae* straight to slightly ondulate, branching unilateral or alternate, ferruginous to brown, septate, hyphal cells cylindrical, 4.5–5 µm, smooth. *Apressoria* numerous, entire, sessile, facing forward, ovate, unicellular, alternate or unilateral, never opposed, 6–10×5–7 µm, brown, penetration peg in central part of apressorial cell. *Thyriothecia* superficial, develop below surface mycelium, circular, single to confluent, fringed at margins, 140–218 µm diam., dark brown to blackish, open with central star-shaped fissures. *Scutellum* radiate, composed of cells isodiametric to cylindrical, straight. *Asci* ovoid, bitunicate, 8 ascospores, hyaline. *Ascospores* 2–celled, cylindrical to ellipsoid, straight to curved, constricted at central septum, 22.5–27.5×7.5–10.5 µm, brown, smooth. *Anamorph* absent.

***Notes*:** Three specimens collected were identified as being *Asterina mandaquiensis*. Two were collected from leaves of *Eugenia uniflora* in Reserva Natural Vale do Rio Doce (Espírito Santo). And the third specimen was collected from leaves of *Eugenia* sp. in Mata da Biologia (Minas Gerais). *Asterina mandaquiensis* was described by Hennings in 1908 in Brazil on leaves of *Eugenia uniflora*. This species is illustrated here for the first time.

*Specimens examined*: Brazil, Espírito Santo and Minas Gerais, Reserva Natural Vale do Rio Doce and Viçosa from leaves of *Eugenia uniflora* and *Eugenia* sp. (*Myrtaceae*), 2012. A.L. Firmino, D.B. Pinho and O.L. Pereira.

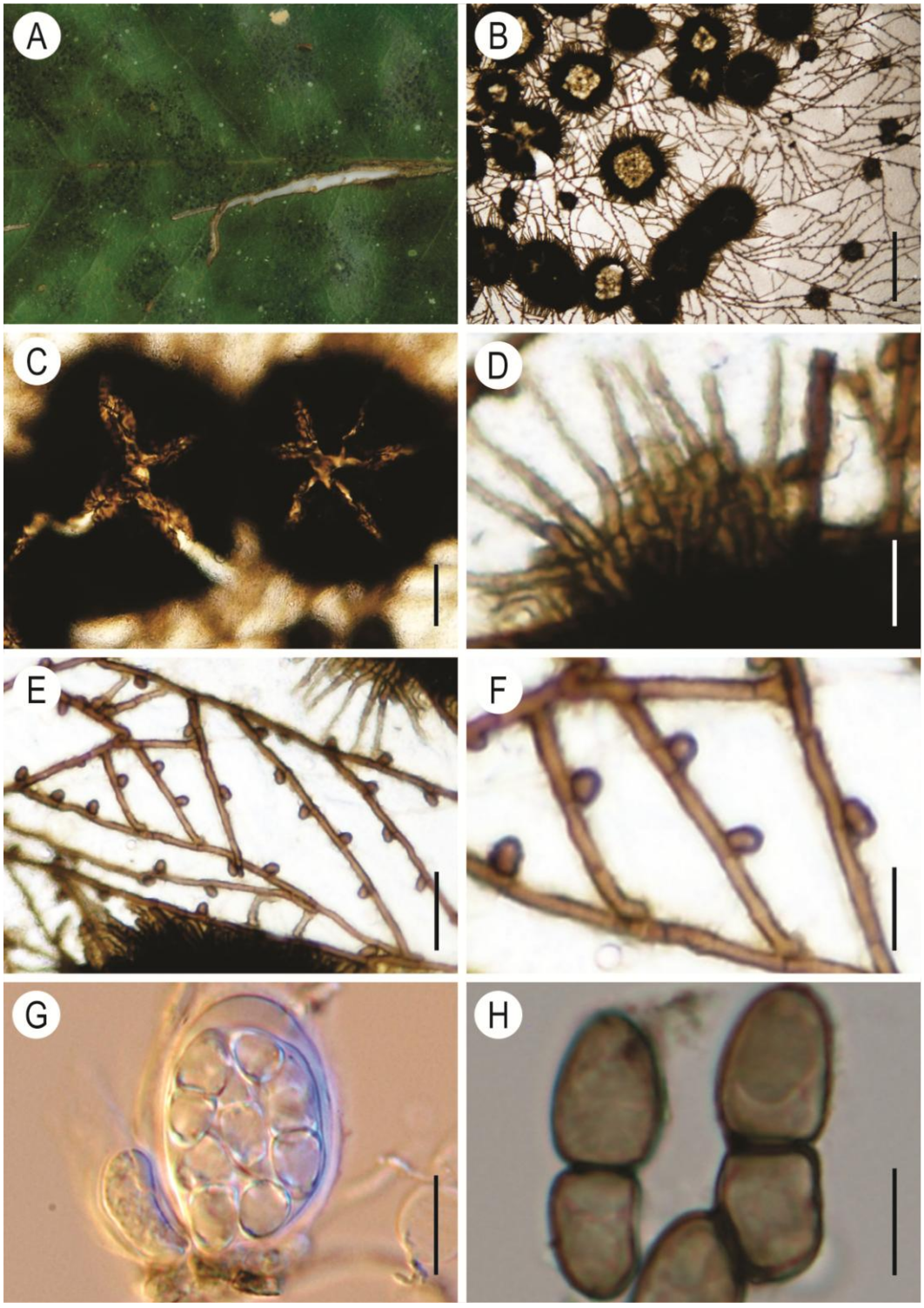


**Fig. 5.** *Asterina mandaquiensis*. A. Colonies epiphyllous. B. Part of a colony with open thyriothecia and surface mycelium. C. Thyriothecium opened by a central star-shaped fissure. D. Thyriothecia fringed at margins. E. Mycelium branching. F. Apressoria. G. Bitunicate asci ovoid. H. Brown and smooth ascospore. Scale bars: B=200 µm; C–E= 50 µm; D–F–G= 20 µm; H= 10 µm.

***Asterina zeyheri*** Doidge. Bothalia. 4(2):273–420. 1942. Fig. 6.

*Colonies* epiphyllous, circular, single, becoming confluent, dark brown to black. *Hyphae* straight to flexuous, branching unilateral or alternate, brown to dark brown, septate, hyphal cells cylindrical, 4.5–5.5 µm, smooth. *Apressoria* numerous, entire, sessile, straight to facing forward, cylindrical to elliptic, unicellular, alternate or unilateral, never opposed, 10–15×6–8 µm, brown, penetration peg in central part of apressorial cell. *Thyriothecia* superficial, develop below surface mycelium, circular, single, slightly fringed at margins, 162–304 µm diam., dark brown to blackish, open with central star-shaped fissures. *Scutellum* radiate, composed of cells isodiametric to cylindrical, straight. *Asci* ovoid to subovoid, bitunicate, 8 ascospores, hyaline. *Ascospores* 2–celled, cylindrical to elliptic, straight, constricted at central septum, 35–40×14–15.5 µm, pale brown to brown, smooth. *Anamorph* absent.

***Notes*:** Two specimens collected were identified as being *Asterina zeyheri*. Both were collected in Reserva Natural Vale do Rio Doce from leaves of *Eugenia rotundifolia* and *Eugenia* sp. *Asterina zeyheri* was described by Doidge in 1942 in South Africa on leaves of *Eugenia zeyheri*. This is the first record of *A. zeyheri* in Brazil and *Eugenia rotundifolia* is new record of host plant species. This species is illustrated here for the first time.

*Specimens examined*: Brazil, Espírito Santo and Minas Gerais, Reserva Natural Vale do Rio Doce and Viçosa from leaves of *Eugenia uniflora* and *Eugenia* sp. (*Myrtaceae*), 2012. A.L. Firmino, D. B. Pinho and O.L. Pereira.

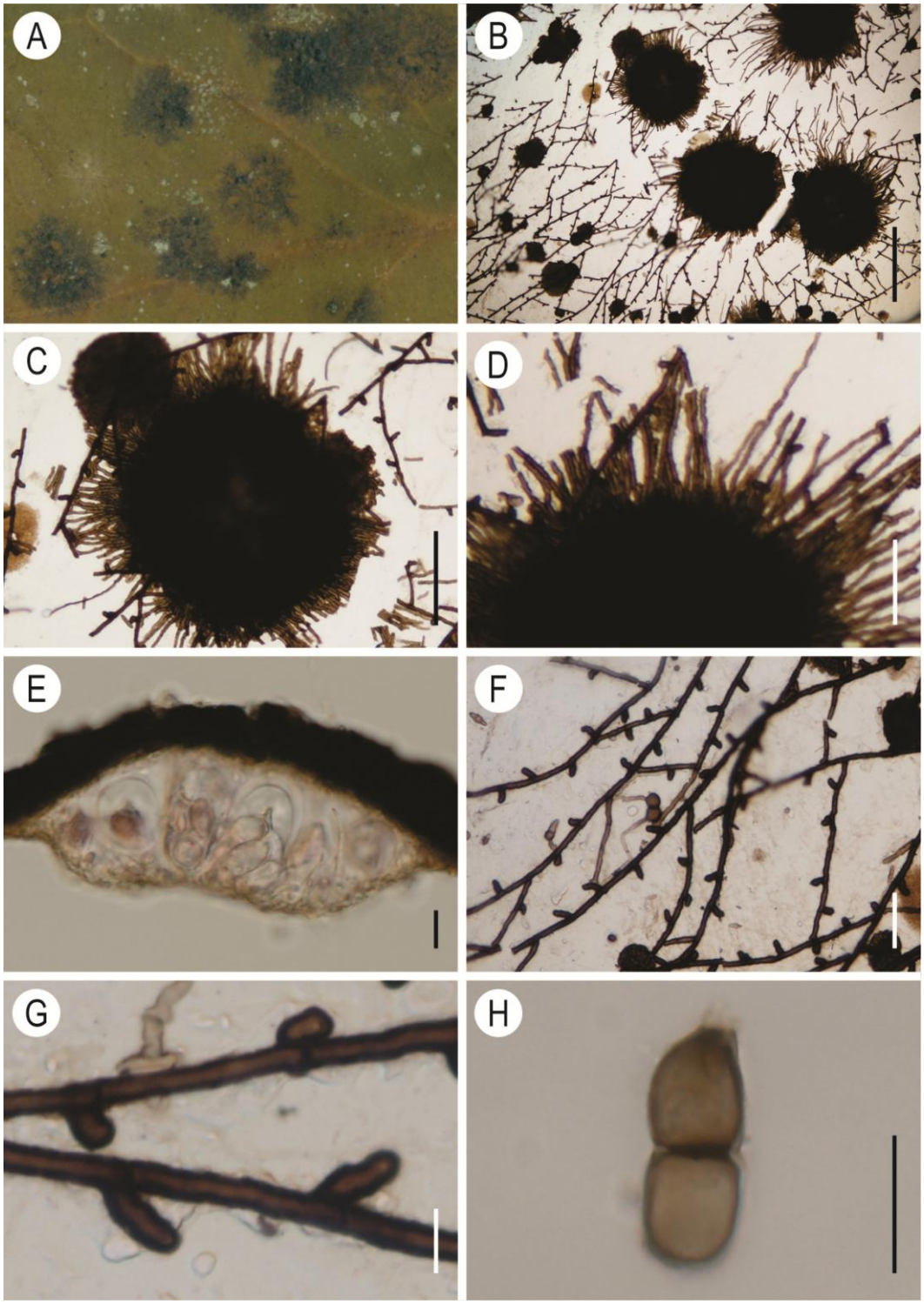


**Fig. 6.** *Asterina zeyheri*. A. Colonies epiphyllous. B. Part of a colony with open thyriothecia and surface mycelium. C. Thyriothecium opened by a central star-shaped fissure. D. Thyriothecia fringed at margins. E. Cross section of the thyriothecia. F. Mycelium branching. G. Long and unicelular apressoria. H. Pale brown and smooth ascospore. Scale bars: B=200 µm; C= 100 µm; D–F= 50 µm E–H= 20 µm; G= 10 µm.

***Asterina* sp. 1** (To be proposed as new) Fig. 7.

*Colonies* epiphyllous, irregular, single, becoming confluent, black. *Hyphae* straight, branching opposite or rarely unilateral, brown, septate, hyphal cells cylindrical, 4–5 µm, smooth. *Apressoria* numerous, entire to irregularly sublobate, (0–3) lobes, sessile, straight to crooked, ovate, long-ovate, unicellular, unilateral or rarely alternate, never opposed, 7.5–12.5×4.5–7 µm, brown, penetration peg in top part of apressorial cell. *Thyriothecia* superficial, develop below surface mycelium, circular, single, fringed at margins, 97.5–157.5 µm diam., dark brown to blackish, open with central star-shaped fissures. *Scutellum* radiate, composed of cells isodiametric to cylindrical, straight. *Asci* globose, bitunicate, 8 ascospores, hyaline. *Ascospores* 2–celled, oblong, straight, constricted at central septum, 20–22.5×9.–12 µm, brown, smooth. *Anamorph* absent.

***Notes*:** Twenty species of Asterina are known in association with members of the family *Rutaceae* (Hosagoudar 2000). The only one reported from Brazil is *Asterina rhabdodendri* Syd. on leaves of *Rhabdodendron crassipes* (Spruce ex Benth.) Huber (Sydow & P. Sydow 1916). *Asterina* sp. 1 on *Dictyoloma vandellianum* differs from *Asterina acronychiae* Hosagoudar & Goos in having apressoria mainly unilateral, larger in length and smaller in wide, ascospores smaller in length and width, smooth (Hosagoudar & Goos 1996). Differs from *Asterina atalantiae* Hosagoudar & Agarwal in having colonies epiphyllous, smaller thyriothecia and ascospore (Hosagoudar & Agarwal 2003). Differs from *Asterina banguinensis* H.S. Yates in having smaller hyphae in wide, lobate apressoria and dispersed thyriothecia (Yates 1918). Differs from *Asterina clausenicola* Doidge in having straight hyphae, unilateral appresoria and larger thyriothecia (Doidge 1920). Differs from *Asterina correicola* Cooke & Massee in having larger thyriothecia and smaller ascospores (Cooke 1887). Differs from *Asterina decora* Syd. in having epiphyllous colonies,

smaller apressoria, ascospore in lenght and larger ascospore in wide (Sydow 1937). Differs from *Asterina delicatula* Syd. & Bal in having flexuous hyphae, smaller apressoria and larger ascospores (Sydow 1921). Differs from *Asterina dictylomae* Henn. in having larger hyphae, unilateral apressoria and larger ascospores (Hennings 1904). Differs from *Asterina fagarae* H.S. Yates in having unilateral apressoria, smaller thyriothecia and ascospore (H.S Yates 1918). Differs from *Asterina glycosmidigena* Hosagoudar & Thomas in having larger thyriothecia and ascospore (Hosagoudar & Thomas 2010). Differs from *Asterina glycosmidis* Hosagoudar & Biju in having unilateral apressoria and smaller ascospore (Hosagoudar 2005). Differs from *Asterina melicopicola* Hosagoudar & Abraham in having unilateral apressoria and smaller ascospores (Hosagoudar & Abraham 1997). Differs from *Asterina murrayae* Hansf. in having epiphyllous colonies, straight hyphae, unilateral apressoria and smaller ascospores (Hansford 1947). Differs from *Asterina murrayicola* Hosagoudar & Sabeena in having epiphyllous colonies, smaller thyriothecia and ascospore, ascospore smooth (Hosagoudar 2012). *Asterina rhabdodendri* Syd. & P. Syd. in having epiphyllous colonies, unilateral apressoria, smaller thyriothecia and ascospore, ascospore smooth (Sydow & P. Sydow 1916). Differs from *Asterina rhabdodendri* var. *lavibus* Patil & Pawar in having epiphyllous colonies, unicellular, unilateral and smaller apressoria, smaller ascospore (Patil & Pawar 1989). Differs from *Asterina toddaliae* Kar & Ghosh in having smaller apressoria and ascospore (Kar & Ghosh 1986). Differs from *Asterina toddaliicola* Hosagoudar Agarwal, Biju & Archana in having unilateral and unicellular apressoria, larges thyriothecia, smaller ascospore in wide (Hosagoudar 2006). Differs from *Asterina vepridis* Doidge in having epiphyllous colonies, unilateral apressoria, smaller thyriothecia and ascospore (Doidge 1942). And differs from *Asterina zanthoxyli* Yamamoto in having unilateral apressoria and smaller ascospore in wide (Yamamoto 1957).

*Specimens examined*: Brazil, Espírito Santo, Reserva Natural Vale do Rio Doce from leaves of *Dictyoloma vandellianum* (*Rutaceae*), 2012. A.L. Firmino.

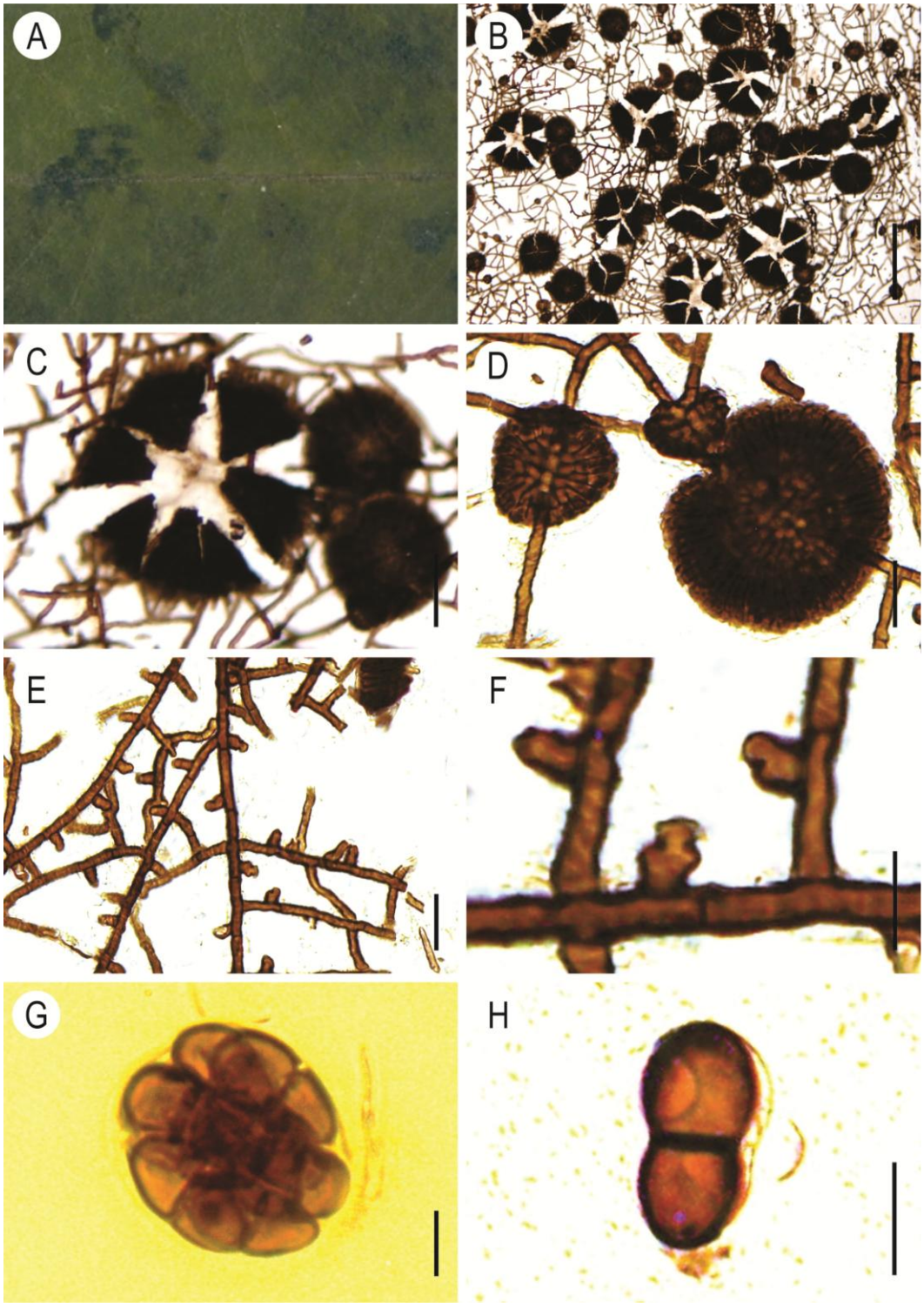


**Fig. 7.** *Asterina* sp. 1. A. Colonies epiphyllous. B. Part of a colony with open thyriothecia and surface mycelium. C. Open thyriothecia. D. Initials young thyriothecia. E. Mycelium branching. F. Unicellular and sublobate apressoria. G. Asci with ascospores. H. Mature ascospore. Scale bars: B=200 µm; C= 50 µm; D–E= 20 µm; F–G–H= 10 µm.

***Asterina* sp. 2** (to be proposed as new) Fig. 8.

*Colonies* epiphyllous, irregular to circular, single, becoming confluent, black. *Hyphae* straight to slightly flexuous, branching irregular, pale brown to brown, septate, hyphal cells cylindrical, 5–6 µm, smooth. *Apressoria* numerous, entire, sessile, straight to crooked, long-ovate, unicellular, alternate to unilateral, never opposed, 9–12.5×6–7.5 µm, brown, penetration peg in top part of apressorial cell. *Thyriothecia* superficial, developing below surface mycelium, circular, single to small groups confluent, fringed at margins, 142–253 µm diam., dark brown to blackish, open with central star-shaped fissures. *Scutellum* radiate, composed of isodiametric to cylindrical cells, straight. *Asci* globose, bitunicate, 8 ascospores, hyaline. *Ascospores* 2–celled, oblong, straight, constricted at central septum, 27.5–34.5×14–15 µm, brown, smooth. *Anamorph* absent.

***Notes*:** Twelve species of *Asterina* are known in association with members of the family *Sapotaceae* (Hosagoudar 2000). The only one species reported from Brazil is *Asterina chrysophylli* Henn. on leaves of *Chrysophyllum* sp. (Hennings 1908). *Asterina* sp. 2 on *Sapotaceae* member differs from *Asterina bosmanae* Doidge in having smaller apressoria and larger ascospore (Doidge 1942). Differs from *Asterina chrysophylli* Henn. in having smaller thyriothecia and ascospores in wide (Hennings 1908). Differs from *Asterina diaphorella* Syd. & P. Syd. in having smaller ascospore and apressoria in wide (Sydow & Sydow P. 1919). Differs from *Asterina dipholidis* Petr. & Cif. in having smaller thyriothecia and ascospore in wide (Petrak & Ciferri 1930). Differs from *Asterina laxiuscula* Syd. & P. Syd. in having smaller ascospore (Sydow & P. Sydow 1917). Differs *Asterina mimusopsidicola* Hosagoudar, Sabeena & Agarwal in having epiphyllous colonies, unicellular apressoria, smaller thyriothecia and larger ascospores (Hosagoudar *et al* 2009). Differs from *Asterina opaca* Syd. & P. Syd. in having alternate or

unilateral apressoria and larger ascospores (Sydow & P. Sydow 1912). Differs from *Asterina palaquii* Hosagoudar & Goos in having alternate to unilateral apressoria, smaller apressoria and ascospores (Hosagoudar & Goos 1996). Differs from *Asterina paraguayensis* Speg. in having epiphyllous colonies, larger hyphae, thyriothecia and ascospores (Spegazzini 1885). Differs from *Asterina robusta* Doidge in having smaller thyriothecia and ascospores (Doidge 1920). Differs from *Asterina saccardoana* Theiss. in having thyriothecia fringed at margins, smaller hyphae and ascospores (Theissen 1913). Differs from *Asterina sydowiana* R.W. Ryan in having smaller apressoria and ascospores, ascospore smooth (Ryan 1924).

*Specimens examined*: Brazil, Espírito Santo, Reserva Natural Vale do Rio Doce from leaves of *Sapotaceae* member (*Sapotaceae*), 2012. A.L. Firmino.

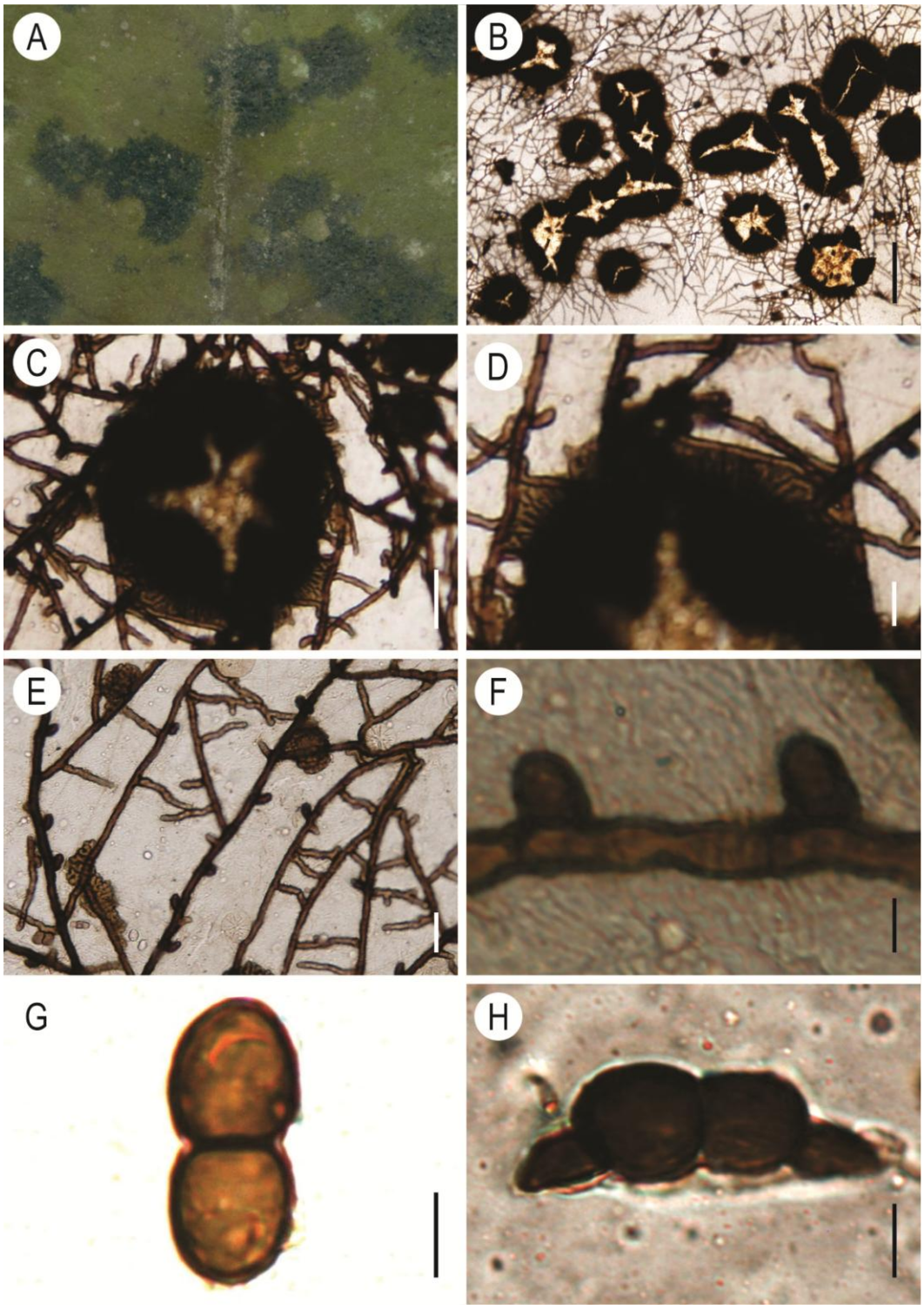


**Fig. 8.** *Asterina* sp. 2. A. Colonies epiphyllous. B. Part of a colony with open thyriothecia and surface mycelium. C. Thyriothecium opened by a central star-shaped fissure. D. Thyriothecium fringed at margins. E. Mycelium branching. F. Apressoria. G. Smooth, brown, constricted at central septum and mature ascospore. H. Germinating ascospore. Scale bars: B=200 µm; C= 50 µm; D–E= 20 µm; F= 5 µm; G–H= 10 µm.

***Asterina* sp. 3** (to be proposed as new) Fig. 9.

*Colonies* epiphyllous, irregular to circular, single, rarely confluent, black, not causing leaf spot, but causing a purple discoloration of the leaf tissues under the colony. *Hyphae* straight to slightly flexuous, branching alternate, pale brown to brown, septate, hyphal cells cylindrical, 4–5 µm, smooth. *Apressoria* numerous, entire, sessile, straight to crooked, cylindrical to long-ovate, unicellular, alternate to unilateral, never opposed, 8–10×6–7.5 µm, brown, penetration peg in top part of apressorial cell. *Thyriothecia* superficial, developing below surface mycelium, circular, single to small group confluent, fringed at margins, 152–253 µm diam., dark brown to blackish, open with central star-shaped fissures. *Scutellum* radiate, composed of cells isodiametric to cylindrical, straight. *Asci* clavate, bitunicate, 8 ascospores, hyaline. *Ascospores* 2–celled, oblong, straight, constricted at central septum, 22.5–25×10.5–13 µm, brown, smooth. *Anamorph* absent.

***Notes*:** Twelve species of Asterina are known in association with members of the family *Sapotaceae* (Hosagoudar 2000). The only one species reported from Brazil is *Asterina chrysophylli* Henn. on leaves of *Chrysophyllum* sp. (Hennings 1908). *Asterina* sp. 3 on *Sapotaceae* member differs from *Asterina bosmanae* Doidge in having larger apressoria, thyriothecia fringed at margins, asci clavate and ascospores with uniform coloring (Doidge 1942). Differs from *Asterina chrysophylli* Henn. in having clavate asci and smaller ascospore (Hennings 1908). Differs from *Asterina diaphorella* Syd. & P. Syd. in having clavate asci and smaller ascospores in length and width (Sydow & Sydow P. 1919). Differs from *Asterina dipholidis* Petr. & Cif. in having smaller apressoria, thyriothecia and ascospores (Petrak & Ciferri 1930). Differs from *Asterina laxiuscula* Syd. & P. Syd. in having clavate asci and smaller ascospores (Sydow & P. Sydow 1917). Differs *Asterina mimusopsidicola* Hosagoudar, Sabeena & Agarwal in having

epiphyllous colonies, smaller apressoria and thyriothecia, unicellular apressoria and larger ascospores (Hosagoudar *et al*. 2009). Differs from *Asterina opaca* Syd. & P. Syd. in having never opposed apressoria, clavate asci and smaller ascospores (Sydow & P. Sydow 1912). Differs from *Asterina palaquii* Hosagoudar & Goos in having unilateral apressoria, larger apressoria and smaller ascospores (Hosagoudar & Goos 1996). Differs from *Asterina paraguayensis* Speg. in having epiphyllous colonies, larger thyriothecia and clavate asci (Spegazzini 1885). Differs from *Asterina robusta* Doidge in having smaller thyriothecia, fringed at margins, clavate asci and smaller ascospores (Doidge 1920). Differs from *Asterina saccardoana* Theiss. in having smaller apressoria and ascospores, clavate asci (Theissen 1913). Differs from *Asterina sydowiana* R.W. Ryan in having larger and smooth ascospores (Ryan 1924).

*Specimens examined*: Brazil, Espírito Santo, Reserva Natural Vale do Rio Doce from leaves of *Pradosia lactescens* (*Sapotaceae*), 2012. A.L.Firmino, D.B. Pinho & O.L. Pereira.

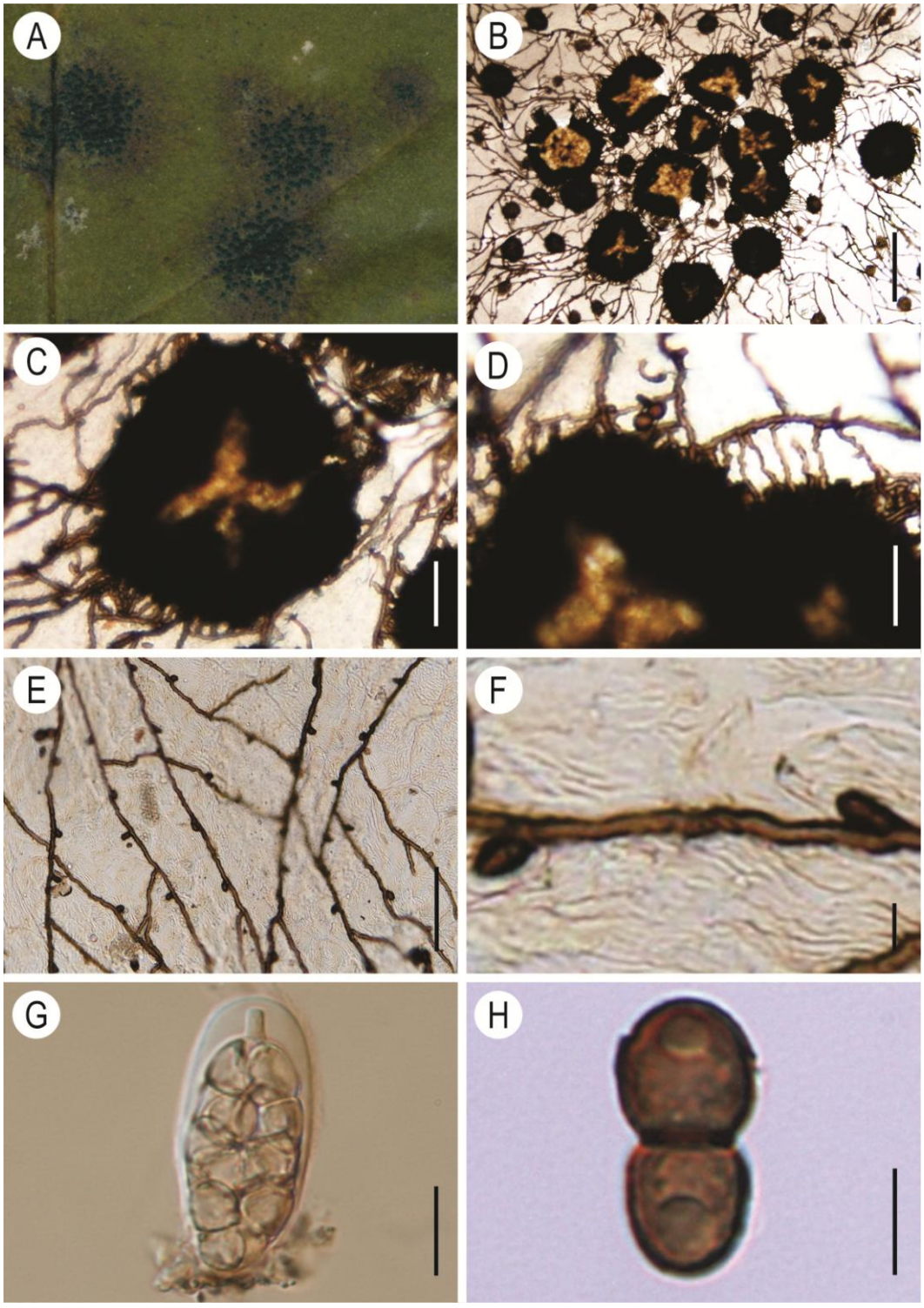


**Fig. 9.** *Asterina* sp. 3. A. Colonies epiphyllous. B. Part of a colony with open thyriothecia and surface mycelium. C. Thyriothecium opened by a central star-shaped fissure. D. Thyriothecia fringed at margins. E. Mycelium branching. F. Alternate, sessile apressoria. G. Bitunicate ascus with hyaline and immature ascospores. H. Smooth, brown, constricted at central septum and mature ascospore. Scale bars: B=200 µm; C–D= 50 µm; E= 100 µm; F–H= 10 µm; G = 20 µm.

***Asterina* sp. 4** (to be proposed as new) Fig. 10.

*Colonies* epiphyllous, irregular to circular, single, rarely confluent, black. *Hyphae* straight, branching opposed, alternate or unilateral, dark brown, septate, hyphal cells cylindrical, 5–6 µm, smooth. *Apressoria* numerous, entire, sessile, straight to crooked, angular, unicellular, alternate to unilateral, never opposed, 15–19×7.5–10 µm, brown, penetration peg in middle part of apressorial cell. *Thyriothecia* superficial, develop below surface mycelium, circular, single, fringed at margins, 182–263 µm diam., dark brown to blackish, open with central star-shaped fissures. *Scutellum* radiate, composed of cells isodiametric to cylindrical, straight. *Asci* globose, bitunicate, 4–8 ascospores, hyaline. *Ascospores* 2–celled, conoide, straight to curved, constricted at central septum, 32.5–40×14–17.5 µm, ferruginous to brown, smooth. *Anamorph* absent.

***Notes*:** Fourteen species of *Asterina* are known in association with members of the family *Fabaceae* (Hosagoudar 2000; Farr & Rossman 2013). There are not species reported from Brazil. *Asterina* sp. 4 on *Fabaceae* member differs from *Asterina ciferriana* Petr. in having straight hyphae, larger and angular apressoria and larger ascospores (Petrak & Ciferri 1932). Differs from *Asterina contigua* Syd. in having epiphyllous colonies, larger and straight hyphae, with apressoria, larger thyriothecia and ascospores (Sydow 1938). *Asterina contigua* must belong to the genus *Prilleuxina* by absence of apressoria. Differs from *Asterina derridis* Henn. in having epiphyllous colonies, larger thyriothecia and ascospores (Hennings 1908). Differs from *Asterina elaeocarpi* Syd. & P. Syd. in having larger apressoria, thyriothecia and ascospores (Sydow & P. Sydow 1911). Differs from *Asterina holocalycis* Speg, in having straight hyphae, smooth apressoria, larger thyriothecia and ascospores (Spegazzini 1912). Differs from *Asterina hoveafolia* Cooke & Massee in having dispersal thyriothecia and larger ascospores (Cooke 1893). Differs from

*Asterina manihotis* Syd. in having larger hyphae, larger and unicellular apressoria, larger thyriothecia and ascospores (Sydow 1939). Differs from *Asterina platystoma* Cooke & Massee in having larger ascospores (Cooke 1889). Differs from *Asterina prosopidis* (Ellis & Everh) Farl. in having smaller thyriothecia, globose asci and larger ascospores (Farlow 1905). Differs from *Asterina saracae* Hosagoudar, T.K. Abraham & J.L. Crane in having larger, straight to crooked, angular and never opposed apressoria, larger ascospores (Hosagoudar *et al.* 1998). Differs from *Asterina scitula* Syd. in having larger and straight hyphae, larger and unicellular apressoria, larger thyriothecia and ascospores (Sydow 1938). Differs from *Asterina singaporensis* Syd. & P. Syd. in having larger hyphae, apressoria, thyriothecia and ascospores (Sydow & P. Sydow 1920). Differs from *Asterina trachycarpa* Syd. & P. Syd. in having epiphyllous colonies, larger thyriothecia, larger and smooth ascospores (Sydow & P. Sydow 1912). Differs from *Asterina yucatanensis* Ellis & Everh. in having epiphyllous colonies, larger thyriothecia and ascospores (Ellis & Everh 1896).

*Specimens examined*: Brazil, Bahia, Reserva Reserva Ecológica da Michelin from leaves of *Inga* sp. (*Fabaceae*), 2010. A.L. Firmino & D.B. Pinho.

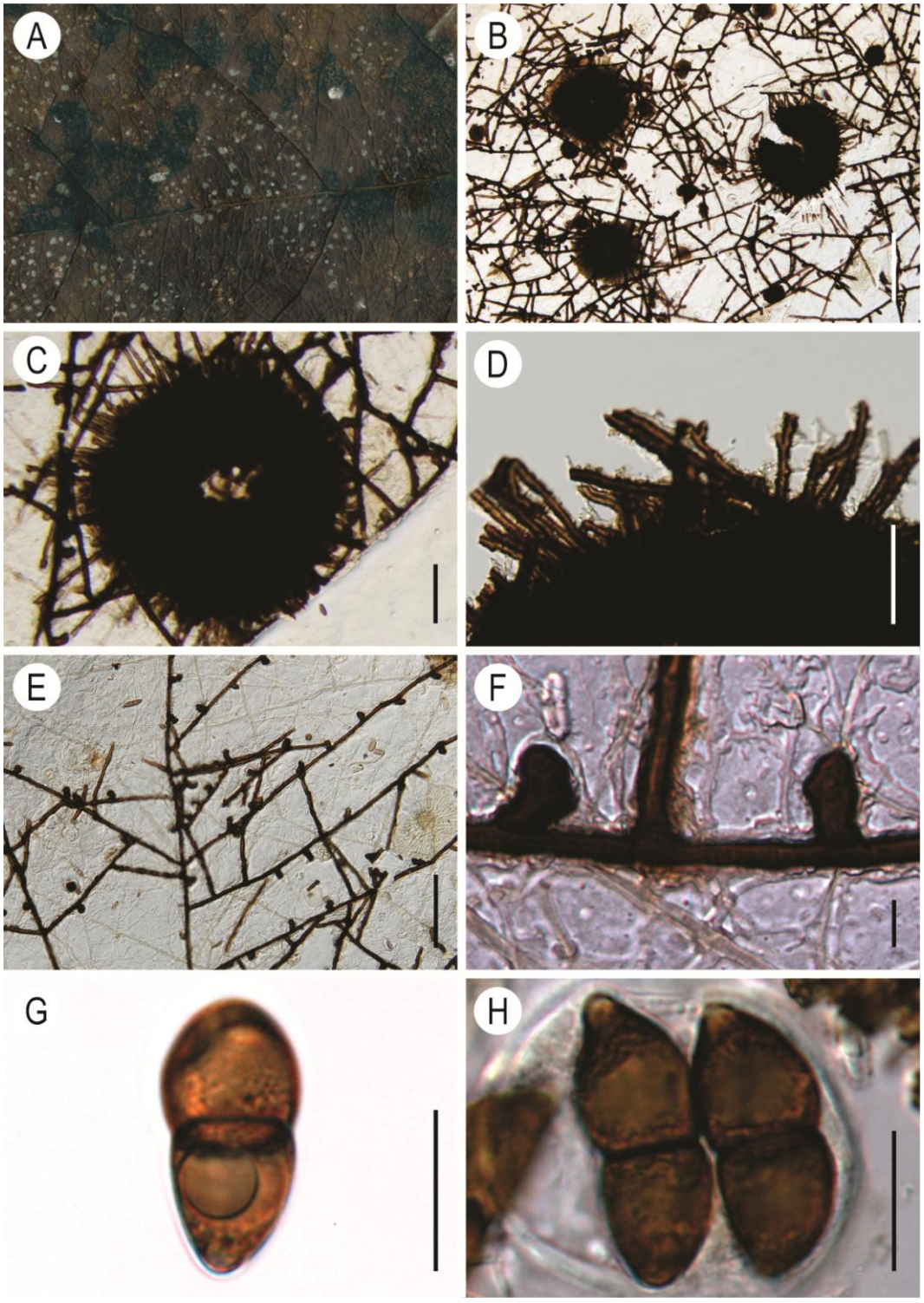


**Fig. 10.** *Asterina* sp. 4. A. Colonies epiphyllous. B. Part of a colony with open thyriothecia and surface mycelium. C. Thyriothecium opened by a central star-shaped fissure. D. Thyriothecia fringed at margins. E. Mycelium branching. F. Straight to crooked, angular and unicellular apressoria. G. Smooth, brown, constricted at central septum and mature ascospore. H. Bitunicate ascus with mature ascospores. Scale bars: B=200 µm; C–D= 50 µm; E= 100 µm; F= 10 µm; G–H = 20 µm.

***Asterolibertia* sp. 1** Fig. 11.

*Colonies* epiphyllous, irregular, single, becoming confluent, black. *Hyphae* straight to ondulate, branching opposite or rarely alternate, brown, septate, hyphal cells cylindrical, 4.5–5 µm, smooth. *Apressoria* numerous, intercalary, sessile, elliptic or elongated, unicellular, 10–14×6–8 µm, brown, penetration peg in central part of apressorial cell. *Thyriothecia* superficial, develop below surface mycelium, circular, single, fringed at margins, 142–202.5 µm diam., dark brown to blackish, open with central star-shaped fissures. *Scutellum* radiate, composed of cells isodiametric to cylindrical, straight. *Asci* globose, bitunicate, 4–ascospores, hyaline. *Ascospores* 2–celled, oblong, ends broadly rounded, straight to slightly arched, constricted at extreme end septum, 36–40×12–14.5 µm, pale brown to brown, very verruculous. *Anamorph* absent.

***Notes*:** There are not species of *Asterolibertia* described from the *Euphorbiaceae*. Among the families belonging to order *Malpighiales*, *Malpighiaceae* and *Chrysobalanaceae* were the only that had representatives of *Asterolibertia* to make the comparison. Three *Asterolibertia* are founded on members of *Chrysobalanaceae* and one on *Malpighiaceae* (Hosagoudar 2010). *Asterolibertia* sp. 1 on leaves of *Senefeldera multiflora* differs from *Asterolibertia schroeteri* (Rehm) Arx in having dispersal thyriothecia, ascospore oblong with ends broadly rounded, Asci with 4 ascospores, verruculose (Müller & Arx 1962). Differs from *Asterolibertia licaniae* (Cooke) Hansf. in having smaller hyphae, thyriothecia, and apressoria in wide, larger and verruculose ascospore, ascospores with unequal cells (Hansford 1949). Differs from *Asterolibertia licaniicola* Hansf. in having larger and verruculose ascospore, ascospores with unequal cells (Hansford 1949). Differs from *Asterolibertia ulei* Hansf. in having smaller apressoria in wide and elliptic or elongated, larger ascospore (Hansford 1949). *Asterolibertia* sp. 1 is the first *Asterolibertia* described from the family *Euphorbiaceae*.

*Specimens examined*: Brazil, Espírito Santo, Reserva Natural Vale do Rio Doce from leaves of *Senefeldera multiflora* (*Euphorbiaceae*), 2012. A.L. Firmino, D.B. Pinho & O.L. Pereira.

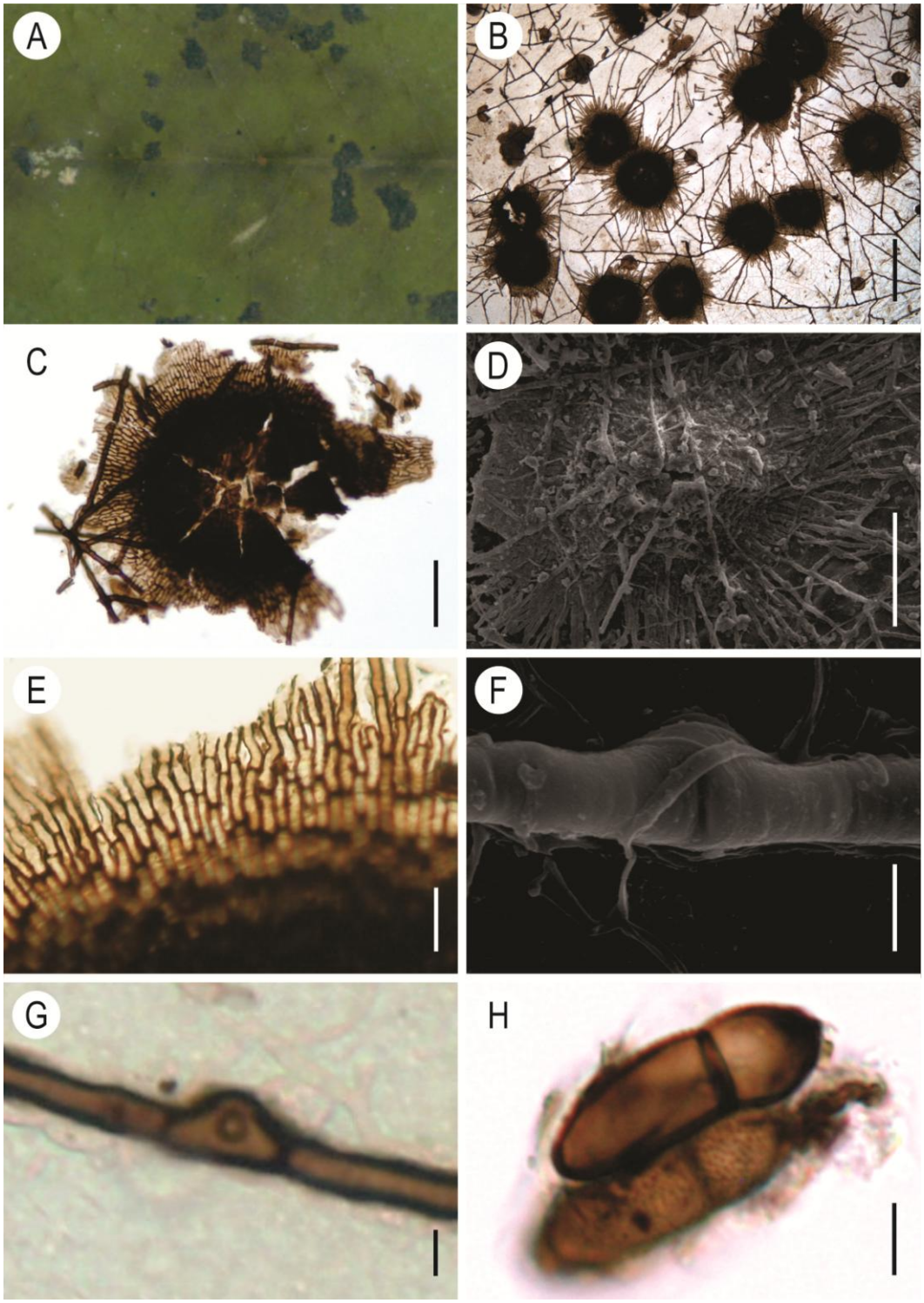


**Fig. 11.** *Asterolibertia* sp. 1. A. Colonies epiphyllous. B. Part of a colony with open thyriothecia and surface mycelium. C. Thyriothecium opened by a central star-shaped fissure. D. Thyriothecia on SEM. E. Thyriothecia fringed at margins. F. Intercalary apressoria on SEM. G. Intercalary, orbicular to elliptic or elongated and unicellular apressoria. H. Verruculous, pale brown, constricted at extreme end septum and mature ascospore. Scale bars: B=200 µm; C–F–G= 5 µm; D= 100 µm; E= 50 µm; H = 10 µm.

***Asterolibertia* sp. 2** Fig. 12.

*Colonies* epiphyllous, circular to irregular, single, becoming confluent, black. *Hyphae* straight to slightly ondulate, branching opposite or rarely alternate, pale brown to brown, septate, hyphal cells cylindrical, 5–6 µm, smooth. *Apressoria* numerous, intercalary, sessile, orbicular, unicellular, 10–15×7.5–11 µm, pale brown to brown, penetration peg in central part of apressorial cell. *Thyriothecia* superficial, develop below surface mycelium, circular, single, fringed at margins, 142–202.5 µm diam., dark brown to blackish, open with central star-shaped fissures. *Scutellum* radiate, composed of cells isodiametric to cylindrical, straight. *Asci* globose to ovoid, bitunicate, 4 ascospores, hyaline. *Ascospores* 2–celled, oblong, ends broadly rounded, straight, slightly constricted at central septum, 29.5–37.5×15.5–18 µm, pale brown to brown, very verruculose. *Anamorph* absent.

***Notes*:** Two species of *Asterolibertia* are known in association with members of the family *Malpighiaceae* (Hosagoudar 2010). However *Asterolibertia couepiae* (Henn.) G. Arnaud probably belongs to the genus *Bheemamyces* Hosagoudar in having lateral and intercalary apressoria (Arnaud 1918). *Asterolibertia* sp. 2 on leaves of *Pera* sp. differs from *Asterolibertia ulei* Hansf. in having asci with 4 ascospores, larger ascospore and equal cells (Hansford 1949).

*Specimens examined*: Brazil, Espírito Santo, Reserva Natural Vale do Rio Doce from leaves of *Pera* sp. (*Malpighiaceae*), 2012. A.L. Firmino, D.B. Pinho & O.L. Pereira.

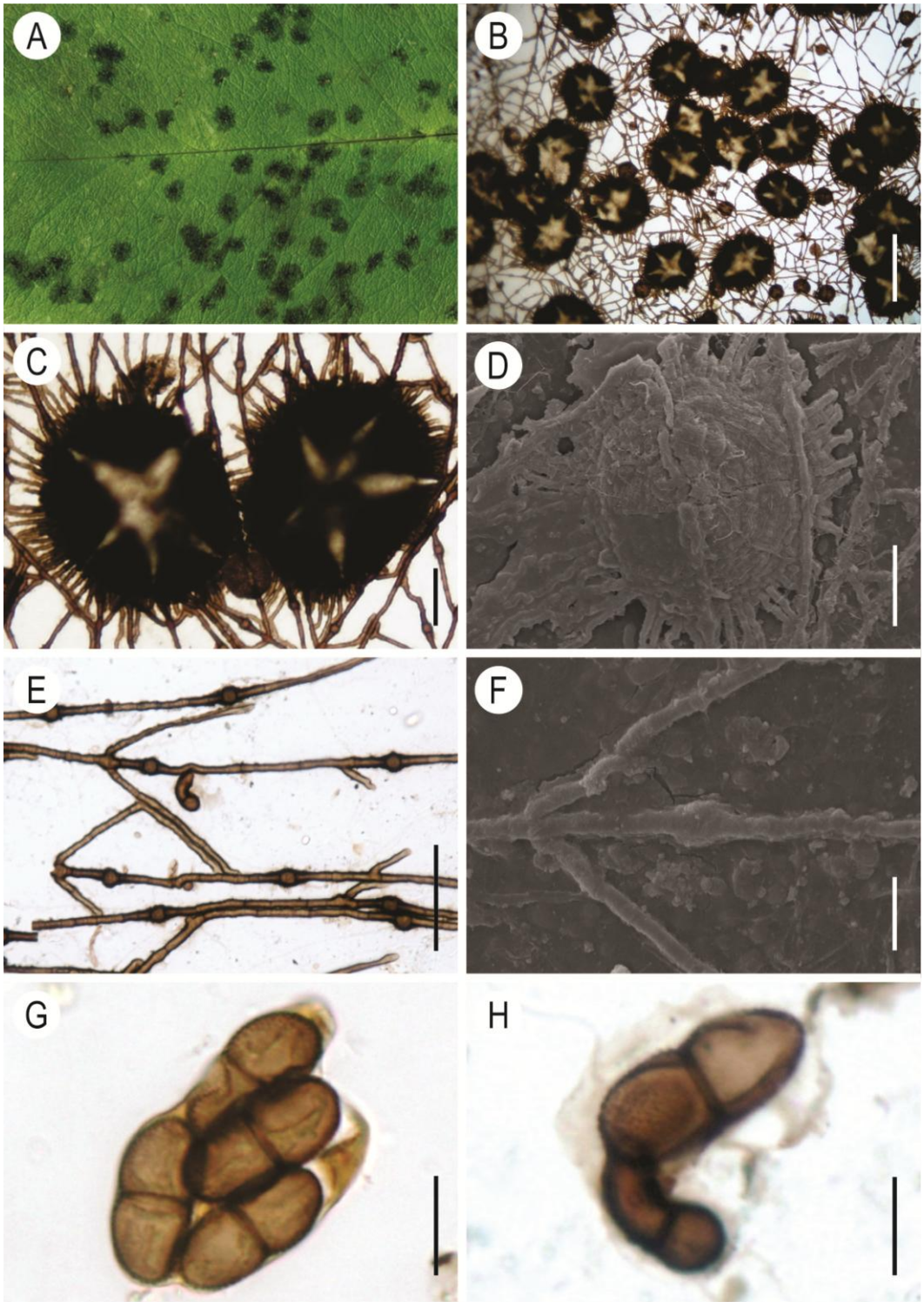


**Fig. 12.** *Asterolibertia* sp. 2. A. Colonies epiphyllous. B. Part of a colony with open thyriothecia and surface mycelium. C. Thyriothecium opened by a central star-shaped fissure. D. Thyriothecia fringed at margins on SEM. E. Mycelium branching with intercalary apressoria. F. Intercalary, orbicular and unicellular apressoria on SEM. G. Bitunicate ascus with verruculous, pale brown, constricted at central septum and mature ascospore. H. Germinating ascospore. Scale bars: B=200 µm; C–D–E= 50 µm; F= 20 µm; G–H= 10

***Batistinula* sp. 1** (to be proposed as new) Fig. 13-14.

*Colonies* amphigenous, irregular to circular, single, becoming confluent, black. *Hyphae* straight, branching alternate, unilateral or opposite, pale brown to brown, septate, hyphal cells cylindrical, 4.5–5 µm, smooth. *Apressoria* numerous, (2–3) lobes, sessile, straight, cylindrical, unicellular, alternate or unilateral, never opposed, 9.5–15×9.5–14 µm, brown, penetration peg in central part of apressorial cell. *Thyriothecia* superficial, develop below surface mycelium, circular, single, fringed at margins, 152–213 µm diam., dark brown to blackish, open with central star-shaped fissures. *Scutellum* radiate, composed of cells isodiametric to cylindrical, straight. *Asci* globose, bitunicate, (4–8) ascospores, hyaline. *Ascospores* 4–celled, oblong, ends broadly rounded, straight to slightly arched, constricted at central septum, 40–48×11–15 µm, pale to pale brown, smooth. *Anamorph* present, in few amounts, superficial, developing above the surface mycelium, brown to dark brown. *Conidiophores* in erectile hyphae, monoblastic, cylindrical, simple, septate, brown. *Conidia* single, staurospore, with 3 branches, germinating at the 3 ends, brown, smooth.

***Notes*:** *Batistinula* sp. 1 on *Caesalpinia echinata* is characterized by lobate apressoria with a sessile base (Fig. 13 g-h) and fringed at margins of Thyriothecia. *Batistinula* sp. 2 on *Chamaecrista ensiformis* differs from *Batistinula* sp. 1 in having obpyriform apressoria (Fig. 15 d), smaller ascospore, larger hyphae and without fringed margin of Thyriothecia. *B. gallesiae* from Brazil is morphologically similar to *Batistinula* sp. 1 (Table 2). However, *Batistinula* sp. 1 differs in having lobate apressoria, ascospore most light and smaller hyphae.

*Specimens examined*: Brazil, Espírito Santo, Reserva Natural Vale do Rio Doce from leaves of *Caesalpinia echinata* (Pau-brasil) (*Leguminosae*), 2012. A.L. Firmino, D.B. Pinho & O.L. Pereira.

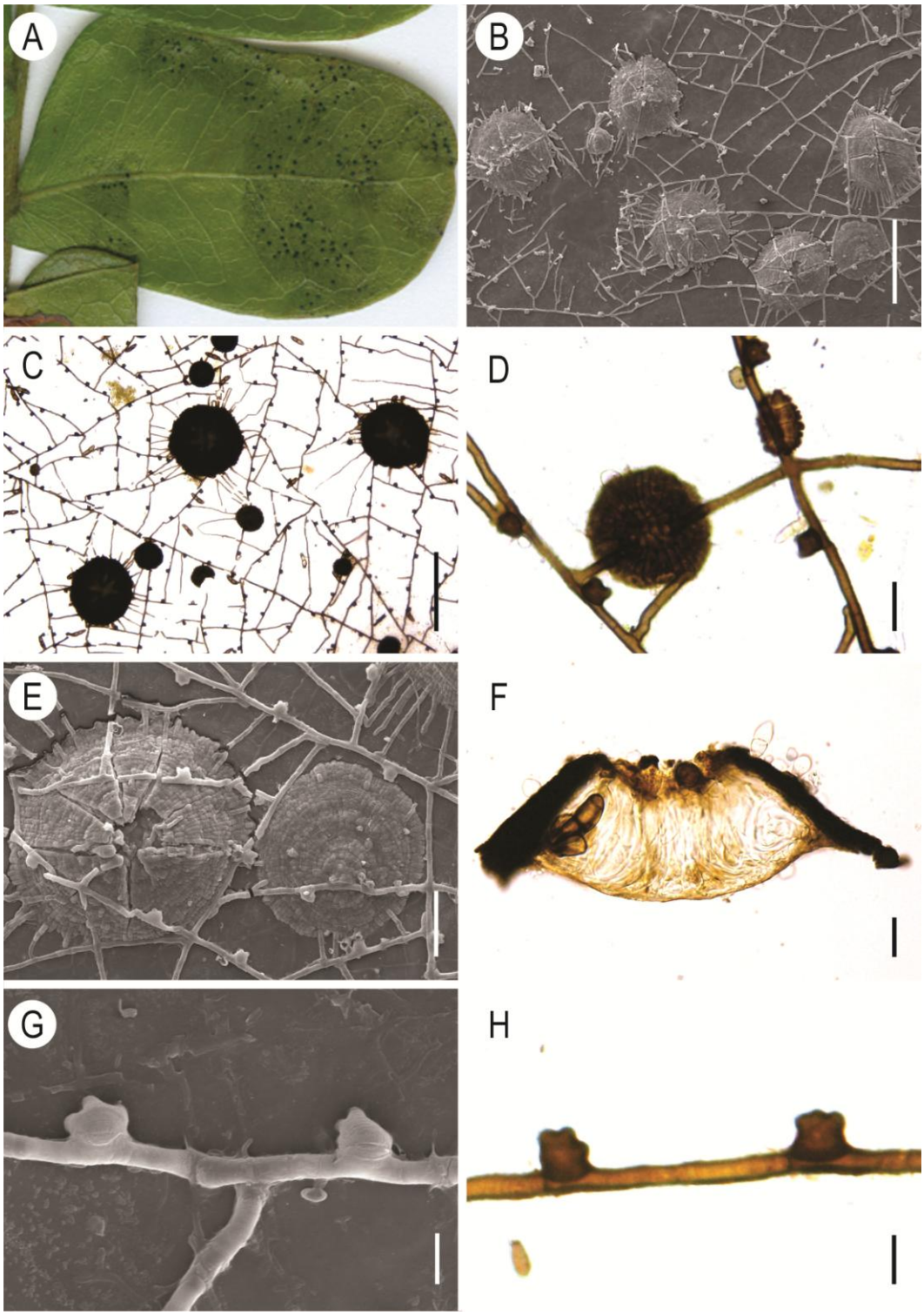


**Fig. 13.** *Batistinula* sp. 1. A. Colonies epiphyllous. B. Colonies on scanning electronic microscope (SEM). C. Part of a colony with open thyriothecia and surface mycelium. D. Initial young thyriothecia. E. Thyriothecium opened by a central star-shaped fissure on SEM. F. Cross section of the thyriothecia. G. Apressoria on SEM. H. lobate, sessile, straight, cylindrical, unicellular and brown apressoria. Scale bars: B–C=200 µm; D–F= 20 µm; E= 50 µm; G–H = 10 µm.

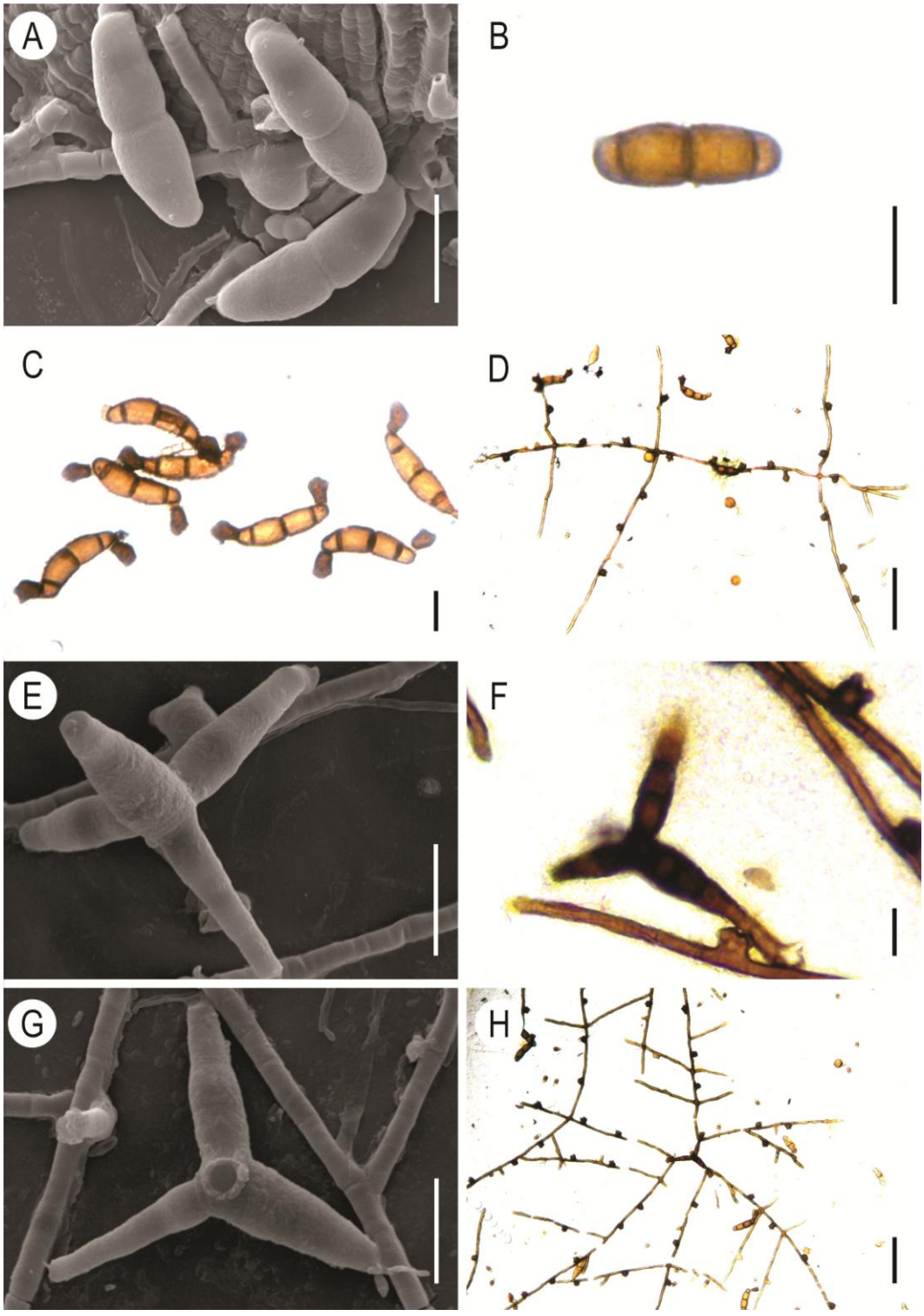


**Fig. 14.** *Batistinula* sp. 1. A. Smooth, oblong, ends broadly rounded, straight to slightly arched, constricted at central septum ascosporo on SEM. B. pale to pale brown and 4–celled ascospore. C. Ascospore with apressoria. D. Germinating ascospore. E. *Triposporium* and erect conidiophore on SEM. F. *Triposporium* on microscope. G. *Triposporium* fallen on the leaf surface showing the conidiogenous locis. H. Germinating *triposporium*. Scale bars: A–B–C–E–F–G=20 µm; D = 100 µm; E–H= 50 µm.

***Batistinula* sp. 2** (to be proposed as new) Fig. 15.

*Colonies* epiphyllous, irregular to circular, single, black. *Hyphae* straight, branching alternate, unilateral or opposite, pale brown to brown, septate, hyphal cells cylindrical, 5–5.5 µm, smooth. *Apressoria* numerous, obpiriform, sessile, unicellular, alternate or unilateral, never opposed, 9.5−12 × 9−12.5 µm, brown, penetration peg in central part of apressorial cell. *Thyriothecia* superficial, develop below surface mycelium, circular, single, without fringed at margins, 122.5−180 µm diam., dark brown to blackish, open with central star-shaped fissures. *Scutellum* radiate, composed of cells isodiametric to cylindrical, straight. *Asci* globose, bitunicate, (4–8) ascospores, hyaline. *Ascospores* 4–celled, oblong, ends broadly rounded, straight to slightly arched, constricted at central septum, 32.5−40 × 12−14.5 µm, brown, smooth. *Anamorph* present, in few amounts, superficial, developing above the surface mycelium, brown to dark brown. *Conidiophores* in erect hyphae, monoblastic, cylindrical, simple, septate, brown. *Conidia* single, staurospore, with 3 branches, germinating at 3 ends, brown, smooth.

***Notes*:** *Batistinula gallesiae* from Brazil is morphologically similar to *Batistinula* sp. 2 (Table 2, Arx 1960). However, *Batistinula* sp. 2 differs in having obpyriform apressoria and smaller thyriothecia, ascospore and hyphae.

*Specimens examined*: Brazil, Espírito Santo, Reserva Natural Vale do Rio Doce from leaves of *Chamaecrista ensiformis* (*Leguminosae*), 2012. A.L. Firmino.

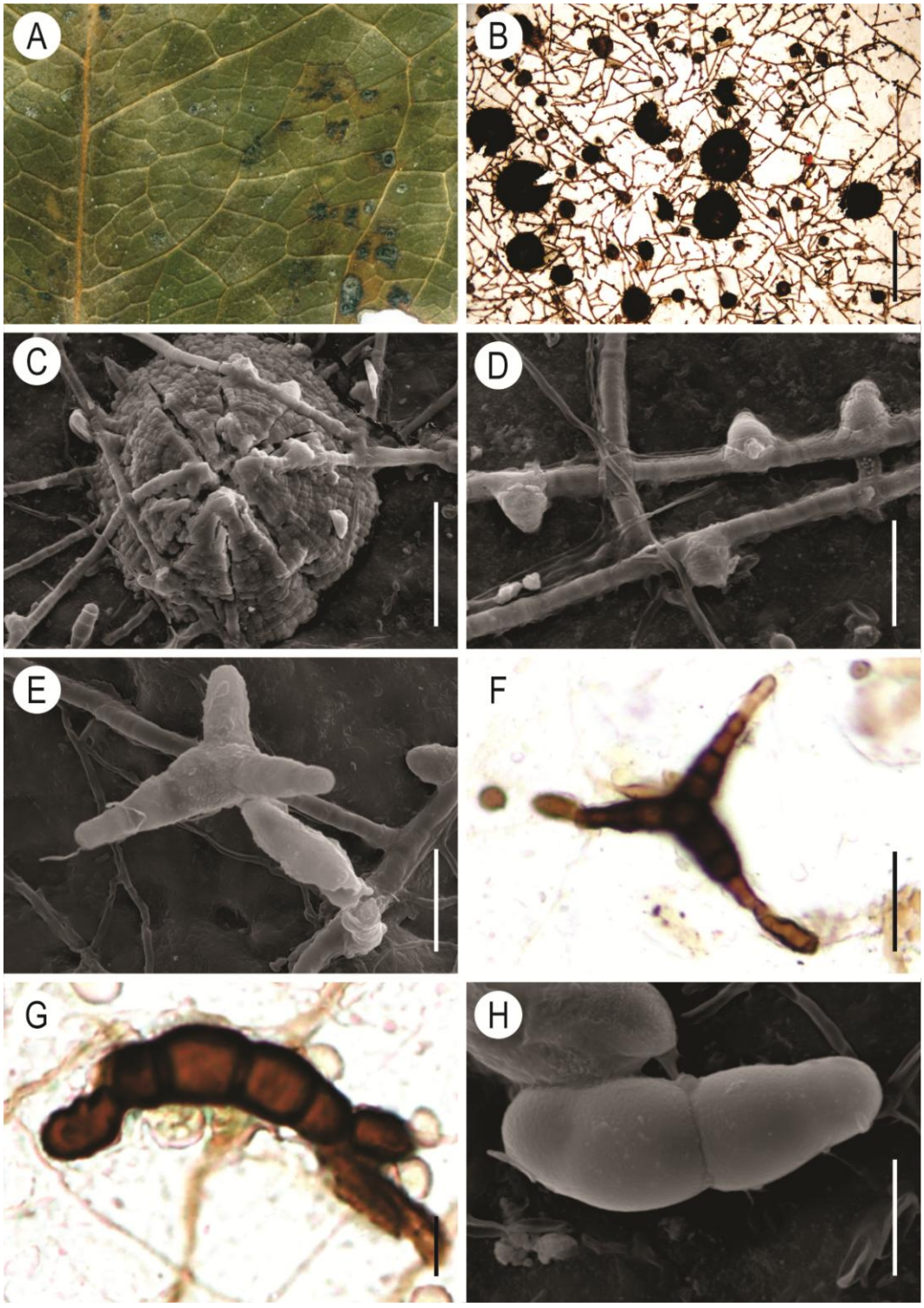


**Fig. 15.** *Batistinula* sp. 2. A. Colonies epiphylous. B. Part of a colony with open thyriothecia and surface mycelium. C. Thyriothecium opened by a central star-shaped fissure on SEM. D. Obpiriform, sessile and unicellular apressoria. E. *Triposporium* and erect conidiophore on SEM. F. Germinating *Triposporium*. G. Germinating ascospore. H. Smooth, 4–celled and constricted at central septum ascosporo on SEM. Scale bars: B= 200 µm; C–F=50 µm; D–E= 20 µm; G–H= 10 µm.

**Table 2.** Biometric data (µm) of the species of *Batistinula* spp. according to literature.

| Species | Thyriothecia | Ascospores | Apressoria | Hyphae |
|---|---|---|---|---|
| *Batistinula gallesiae* | 180−270 | 38−52 × 14−18 | 9−12 wide | 5−7 |
| *Batistinula* sp. 1 | 152−213 | 40−48 × 11−15 | 9.5−15 × 9.5−14 | 4.5−5 |
| *Batistinula* sp. 2 | 122.5−180 | 32.5−40 × 12−14.5 | 9.5−12 × 9−12.5 | 5−5.5 |

***Echidnodes baccharidincola*** (Rehm) Theiss. & Syd., Annales Mycologici. 15(6):422. 1917. Fig. 16.

*Colonies* epiphyllous, irregular to circular, single, black. *Hyphae* straight to flexuous, branching irregular, pale brown, septate, hyphal cells cylindrical, 3–4 µm, smooth. *Apressoria* absent. *Hysterothecia* superficial, develop below surface mycelium, circular to ellipsoid, single to confluent, 102−160 µm diam., dark brown to blackish, open with longitudinal fissures. *Asci* ovoid to subclavate, bitunicate, 8 ascospores, hyaline. *Ascospores* 2–celled, cylindrical to oblong, ends broadly rounded, straight, constricted at central septum, 15−22 × 9−11.5 µm, brown, smooth. *Anamorph* absent.

***Notes*:** One specimen collected was identified as being *Echidnodes baccharidincola*. It was collected near Nova Lima city from leaves of *Baccharis dracunculifolia*. *Echidnodes baccharidincola* was reported in Minas Gerais, Brazil on leaves of *Baccharis* sp. *Baccharis dracunculifolia* is new record of host plant species.

*Specimens examined*: Brazil, Minas Gerais, Nova Lima from leaves of *Baccharis dracunculifolia* (*Asteraceae*), 2012. O.L. Pereira.

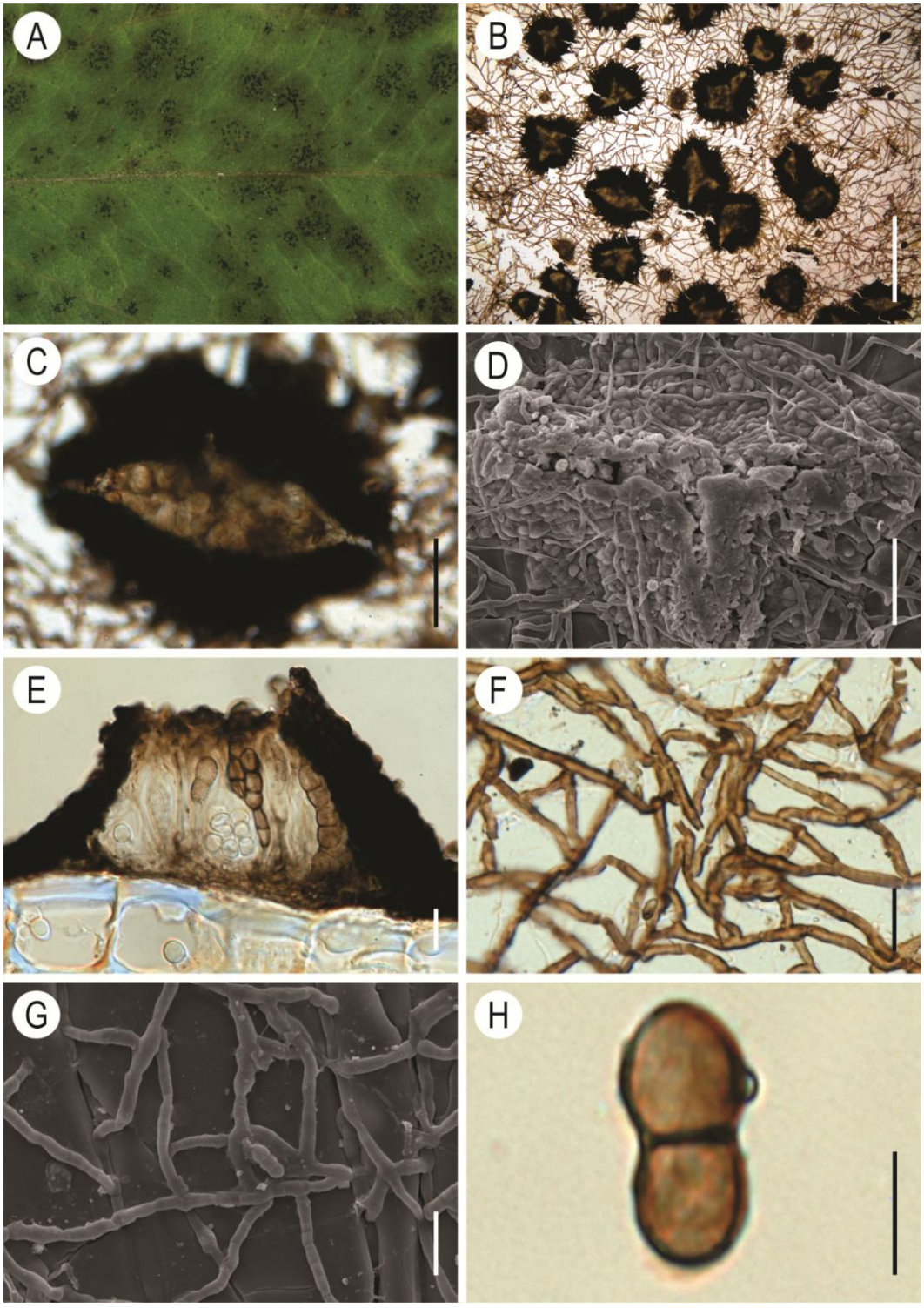


**Fig. 16.** *Echidnodes baccharidincola*. A. Colonies epiphyllous. B. Part of a colony with open hysterothecia and surface mycelium. C. Hysterothecia opened by a longitudinal fissure. D. Hysterothecia on SEM. E. Cross section of the hysterothecia. F. Mycelium branching. G. Mycelium without apressoria. H. Brown and smooth scospore. Scale bars: B= 200 µm; C–D=50 µm; E–F–G= 20 µm; H= 10 µm.

***Lembosia* sp. 1.** (to be proposed as new) Fig. 17.

*Colonies* epiphyllous, irregular to circular, single to confluent, black. *Hyphae* straight to flexuous, branching unilateral and alternate, brown, septate, hyphal cells cylindrical, 5–6 µm, smooth. *Apressoria* unilateral to alternate, entire, sessile, globose, straight to angular, unicellular, 8–10×7.5–9.5 µm, brown, penetration peg in middle part of apressorial cell. *Hysterothecia* superficial, develop below surface mycelium, mostly linear, rarely Y-shaped, single, fringed at margins, 294–729 × 192–233 µm diam., dark brown to blackish, open with longitudinal fissures. *Scutellum* radiate, composed of cells isodiametric to cylindrical, straight. *Asci* ovoid, bitunicate, 8 ascospores, hyaline. *Paraphyses* hyaline, filiform and unbranched. *Ascospores* oblong to cylindrical, 1-septate in the middle, constricted, hyaline, becoming brown at maturity, smooth, 25–30 × 14–15 µm. *Anamorph* absent.

**Notes:** Two species of *Lembosia* are known in association with members of the family *Malpighiaceae* (Hosagoudar 2000). *Lembosia* sp. 1 on *Malpighiaceae* member differs from *Lembosia byrsonima* Henn. in having larger hyphae and hysterothecia in length and wide, asci with 8 ascospores and ascospore oblong to cylindrical (Hennings 1904). Differs from *Lembosia stevensii* Hansf. in having smaller hysterothecia and wide, smooth ascospore (Hansford 1955).

*Specimens examined*: Brazil, Bahia, Reserva Ecológica da Michelin from leaves of *Byrsonima sericea* (*Malpighiaceae*), 2010. A.L. Firmino & D.B. Pinho.

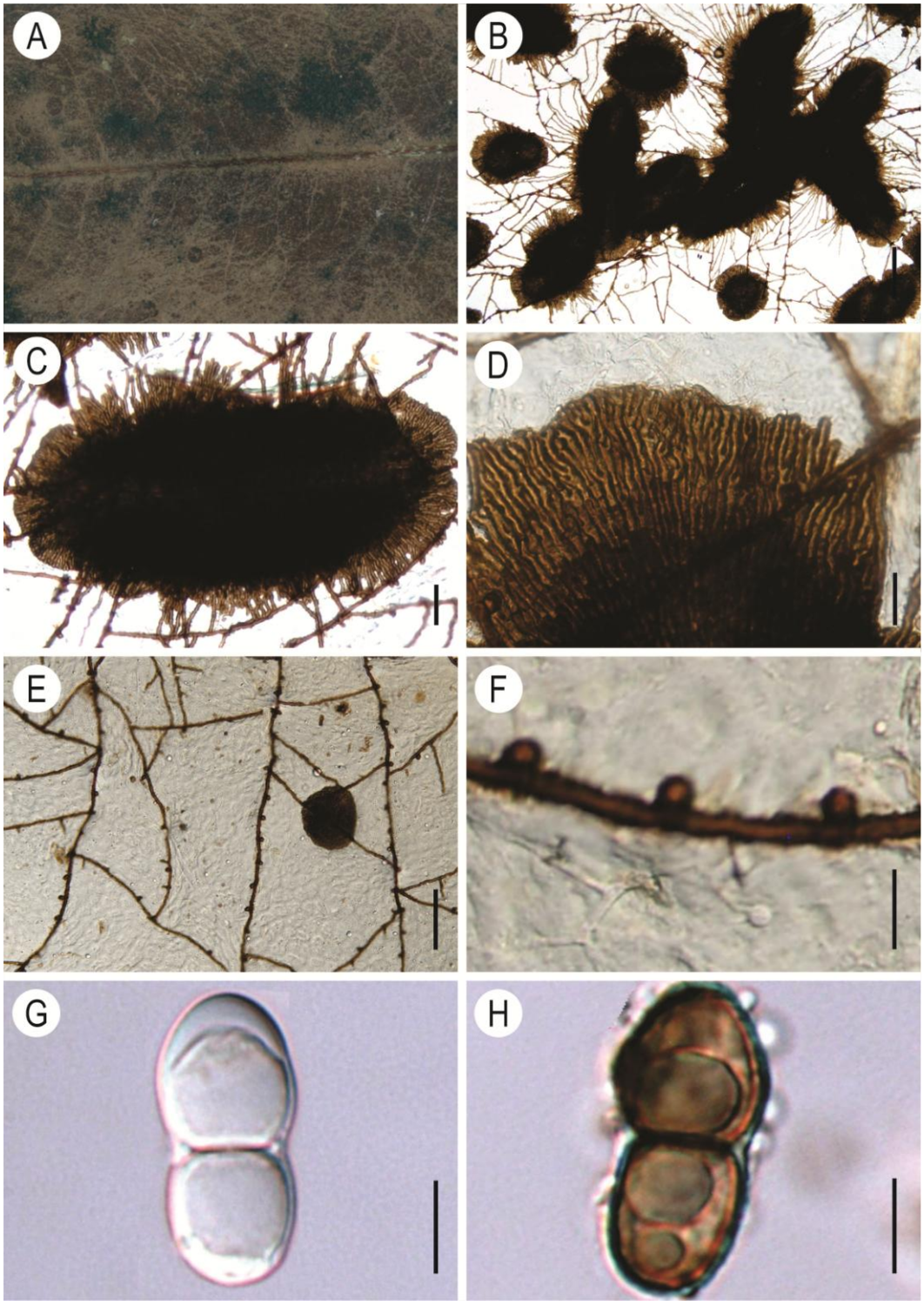


**Fig. 17.** *Lembosia* sp. 1. A. Colonies epiphyllous. B. Part of a colony with open hysterothecia and surface mycelium. C. Hysterothecia open by a longitudinal fissure. D Hysterothecia fringed at margins. E. Mycelium branching. F. Sessile, globose, straight to angular and unicellular apressoria. G. Hyaline and immature ascospore. H. Brown, smooth, constricted at the central septum ascospore. Scale bars: B= 200 µm; C=50 µm; D–F= 20 µm; E= 100 µm; G= 10 µm; H= 10 µm.

***Lembosia* sp. 2** (to be proposed as new) Fig. 18.

*Colonies* epiphyllous, irregular to circular, single to confluent, black. *Hyphae* straight to flexuous, branching irregular, brown, septate, hyphal cells cylindrical, 4–5 µm, smooth. *Apressoria* few, entire, sessile, globose, straight to angular, unicellular, 4.5–5×4.5–5 µm, brown, penetration peg in middle part of apressorial cell. *Hysterothecia* superficial, develop below surface mycelium, mostly linear, rarely Y-shaped, single, fringed at margins, 521.5–1853 × 85–109 µm diam., dark brown to blackish, open with longitudinal fissures. *Scutellum* radiate, composed of cells isodiametric to cylindrical, straight. *Asci* saccate to ovoid, bitunicate, 8 ascospores, 27.5–34 × 12–17.5 µm, hyaline. *Paraphyses* hyaline, filiform and unbranched. *Ascospores* fusiform, 1-septate in the middle, constricted, hyaline, becoming brown at maturity, smooth, 14–16 × 4–5 µm. *Anamorph* absent.

**Notes:** Four species of *Lembosia* are known in association with members of the family *Orchidaceae* (Hosagoudar 2000; Silva & Pereira 2008). Only *Lembosia epidendri* is reported from Brazil (Silva & Pereira 2008). *Lembosia* sp. 1 on *Orchidaceae* member differs from *Lembosia rolfsii* Horne in having larger ascospore, laerger hysterothecia in length and smaller in wide. *L. rolfsii* probably does not belong to the genus *Lembosia*, because it presents conidia on mycelium, being better allocated on genus *Maheshwaramyces* Hosagoudar, Archana & Mathew Dan. (Horne 1905). Differs from *Lembosia sertiferae* Syd. in having straight hyphae, smaller and rounded apressoria, larger hysterothecia and smaller ascospore (Sydow 1939). Differs from *Lembosia dendrochili* Lév in having smaller apressoria and ascospore, larger hysterothecia (Léveillé 1845). Differs from *Lembosia epidendri* Silva & Pereira in having smaller apressoria and ascospore, larger hysterothecia (Silva & Pereira 2008).

*Specimens examined*: Brazil, Bahia, Santa Terezinha, Serra da Jibóia from leaves of *Sobralia liliastrum* (*Orchidaceae*), 2012. O.L. Pereira.

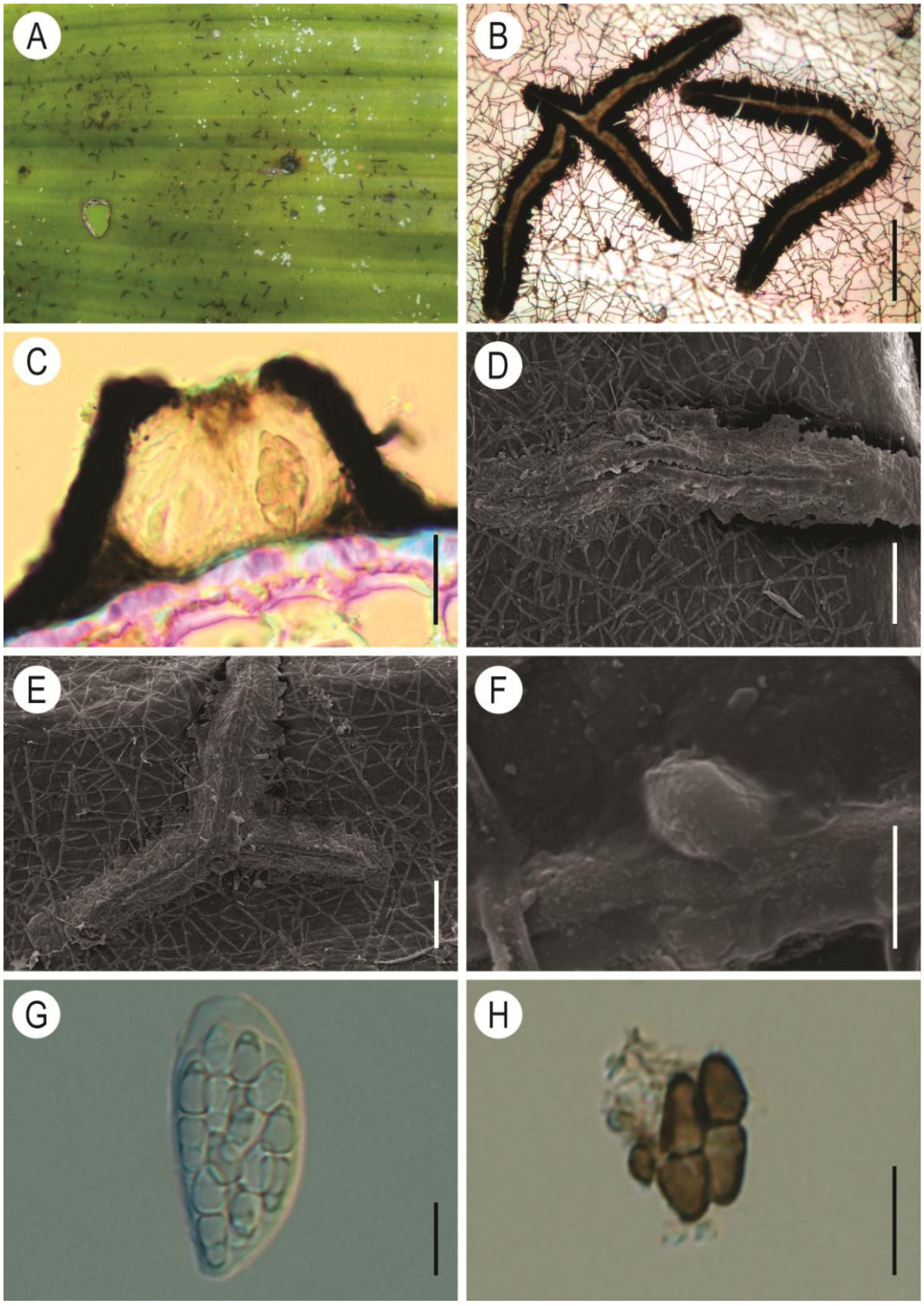


**Fig. 18.** *Lembosia* sp. 2. A. Colonies epiphyllous. B. Part of a colony with open hysterothecia and surface mycelium. C. Cross section of the hysterothecia. D Hysterothecia linear. E. Hysterothecia in Y-shaped. F. Sessile, globose, straight to angular and unicellular apressoria. G. Bitunicate ascus with hyaline ascospore. H. Brown, smooth and fusiform ascospores. Scale bars: B= 200 µm; C=20 µm; D–E= 100 µm; F= 5 µm; G–H= 10 µm.

***Lembosia* sp. 3** (to be proposed as new) Fig. 19.

*Colonies* epiphyllous, irregular, single to confluent, black. *Hyphae* flexuous, branching irregular, pale brown, septate, hyphal cells cylindrical, 2–2.5 µm, smooth. *Apressoria* few, entire, sessile, globose, angular, unicellular, 4–5×4–5 µm, brown, penetration peg in middle part of apressorial cell. *Hysterothecia* superficial, develop below surface mycelium, linear, never in Y-shaped, single, fringed at margins, 521.5−1853 × 85−109 µm diam., dark brown to blackish, open with longitudinal fissures. *Scutellum* radiate, composed of cells isodiametric to cylindrical, straight. *Asci* clavate to ovoid, bitunicate, 8 ascospores, hyaline. *Paraphyses* hyaline, filiform and unbranched. *Ascospores* fusiform, 1-septate in the middle, constricted, hyaline, becoming brown at maturity, smooth, 14−16 × 4−5 µm. *Anamorph* absent.

**Notes:** There are no species of *Lembosia* have been found on *Bignoniaceae* family members. Among the families belonging to order *Lamiales*, *Oleaceae* was the only family that had representatives of *Lembosia* to make the comparison. Three *Lembosia* are found on *Oleaceae* family (Song & Hosagoudar 2003). *Lembosia* sp. 2 on leaves of *Tabebuia elliptica* differs from *Lembosia rizalensis* Petr. in having larger hysterothecia and that are smooth and bigger ascospores (Petrak 1955). Differs from *Lembosia micrasca* Syd. in having smaller and angulate apressoria, larger and rarely Y-shaped hysterothecia (Sydow 1937). Differs from *Lembosia notelaeae* Hansf. in having smaller hyphae, apressoria, larger hysterothecia and rarely in Y-shaped (Hansford 1957). *Lembosia* sp. 2 is the first *Lembosia* described in the family *Bignoniaceae*.

*Specimens examined*: Brazil, Bahia, Reserva Ecológica da Michelin from leaves of *Tabebuia elliptica* (*Bignoniaceae*), 2011. A.L. Firmino & D.B. Pinho.

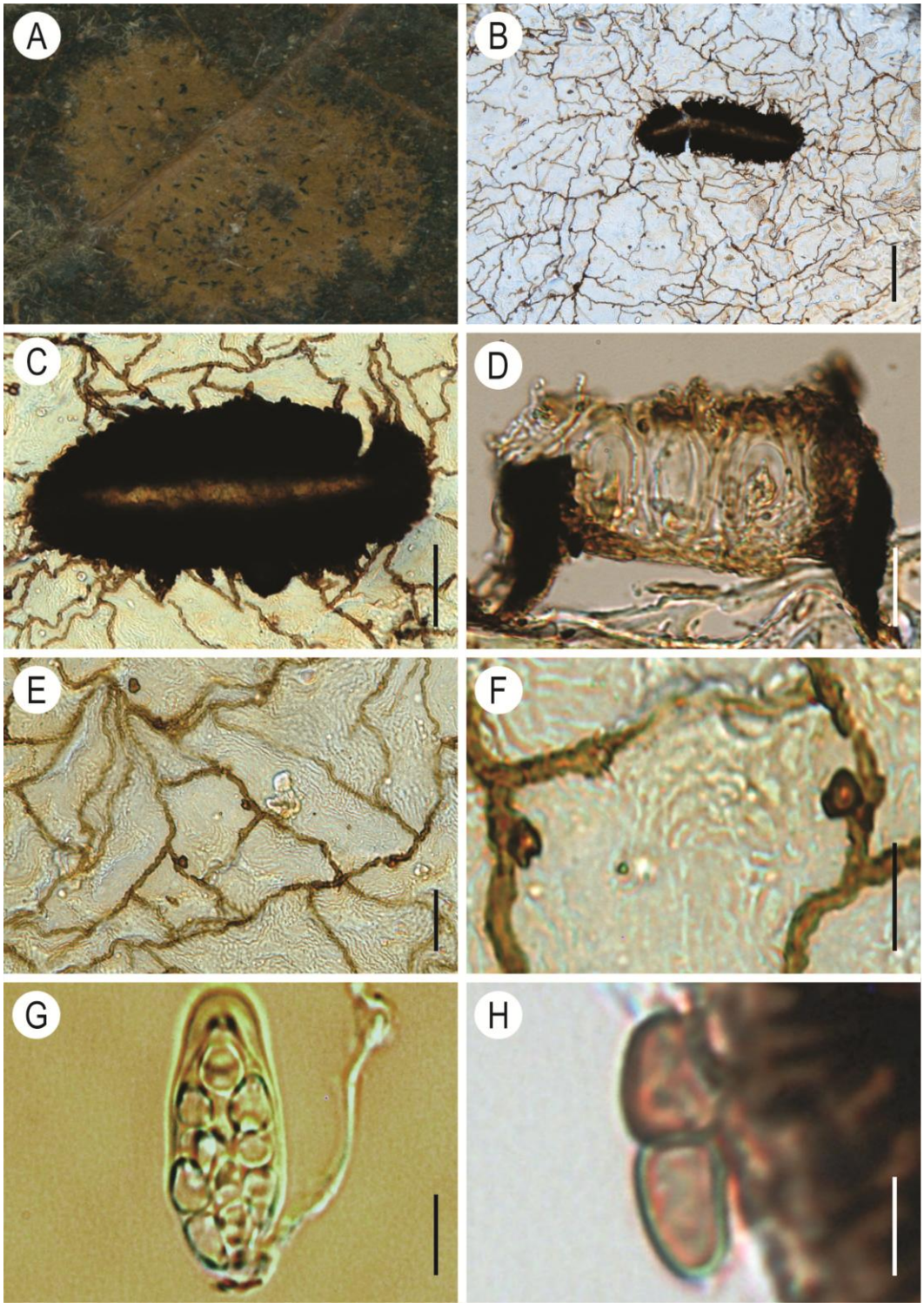


**Fig. 19.** *Lembosia* sp. 3. A. Colonies epiphyllous. B. Part of a colony with open hysterothecia and surface mycelium. C. Hysterothecia opened by a longitudinal fissure. D. Cross section of the hysterothecia. E. Mycelium branching. F. Sessile, globose, angular and unicellular apressoria. G. Bitunicate ascus with hyaline ascospore. H. Brown, smooth and fusiform ascospore. Scale bars: B= 100 µm; C=50 µm; D–E= 20 µm; F–G= 10 µm; H= 5 µm.

**Members of the family *Englerulaceae***

***Digitosarcinella* sp. 1** (to be proposed as new) Fig. 20-21.

*Colonies* epiphyllous, irregular to circular, single to confluent, brown to dark brown. *Hyphae* straight to slightly flexuous, branching opposed, alternate or unilateral, brown, septate, hyphal cells cylindrical, 4.5–5.5 µm, smooth. *Apressoria* numerous, entire, sessile, pyriform, unicellular, alternate to unilateral, never opposed, 7.5–12×7.5–10 µm, brown. Apressoria produce a hypha which arches around and is closely appressed to the apressoria; the curved hyphae may bear a lateral hypha or conidiophore. *Ascomata* superficial, develop below surface mycelium, circular, single, 253–607.5 µm diam., pale to pale brown, open with irregular fissures. *Scutellum* composed of cells irregular. *Asci* clavate, bitunicate, 4–8 ascospores, hyaline. *Ascospores* 2–celled, conoide, straight, slightly constricted at central septum, 24–30×12–15 µm, yellowish to ferruginous, smooth. *Anamorph* present,

(***Digitosarcinella***)

Anamorph superficial, developing above the hypha arched, brown to dark brown. *Conidiophores* in erectile hyphae, monoblastic, cylindrical, simple, septate, brown. *Conidiogenous cells* scattered or in groups, sessile, subcylindrical to ellipsoidal, monoblastic, brown. *Conidia* produce a cheiroid conidium with parallel, straight or curved, up to 5-septate and constricted at the septa, and gradually takes a conoid form, 37.5–50 µm in lengh, 25–37.5 in larger wide and 17.5–27.5 in smaller wide, dark brown with the lower end hyaline, smooth.

(***Questieriella***)

Anamorph superficial. *Conidiogenous cells* shorter, hyaline to subhyaline, cylindrical, sessile. *Conidia* blastic, single, terminal, curved to arched, 3-septate, not constricted at the septa, 70–87.5×12.5–15 µm, yellowish to pale, verruculous.

***Notes*:** Only one species of *Digitosarcinella* is known (Hugues 1984). *Digitosarcinella casearіae* Hugues was found on leaves of *Casearіae sylvestri* in Brazil, São Paulo, Campinas. *Digitosarcinella* sp. 1 differs from *Digitosarcinella casearіae* in having a large arched hypha around the appressorium, smaller *Digitosarcinella* and larger and verrucose *Questieriella*. Hugues improperly allocates this genus as anamorph of *Shiffnerula*. Being that its teleomorph is clearly distinct from that genus (Hugues 1984; Hosagoudar 2003).

*Specimens examined*: Brazil, Espírito Santo, Reserva Natural Vale do Rio Doce, from leaves of *Terminalia kuhlmannii* (*Combretaceae*), 2012. A.L. Firmino, D.B. Pinho & O.L. Pereira.

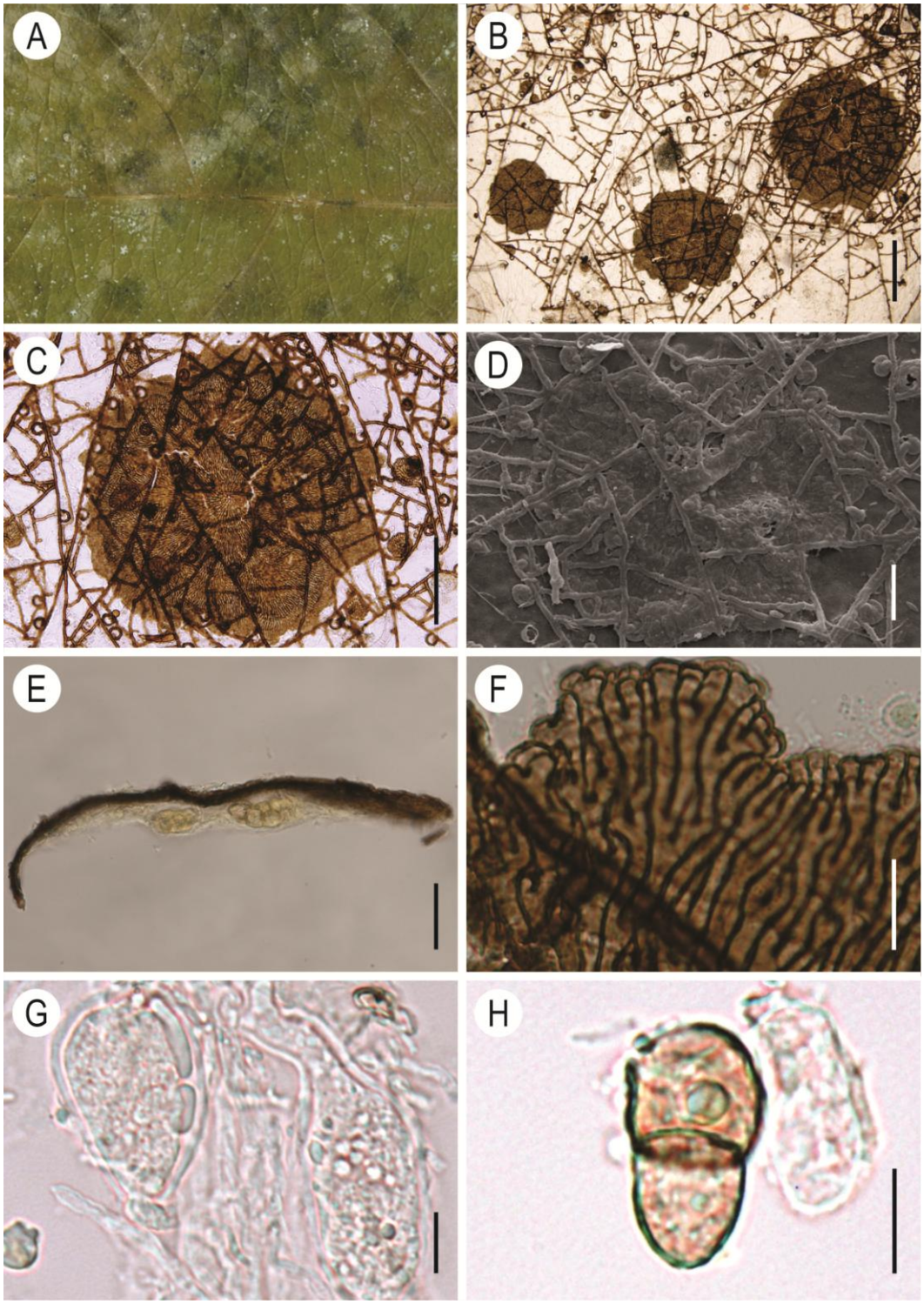


**Fig. 20.** *Digitosarcinella* sp. 1. A. Colonies epiphyllous. B. Part of a colony with open ascomata and surface mycelium. C. Ascomata composed by reticulate cells. D Ascomata on SEM. E. Cross section of the ascomata. F. Ascomata fringed at the margin. G. Bitunicate ascus. H. Brown, smooth, conoid ascospore. Scale bars: B= 200 µm; C= 100 µm; D–E= 50 µm; F= 20 µm; G–H= 10 µm.

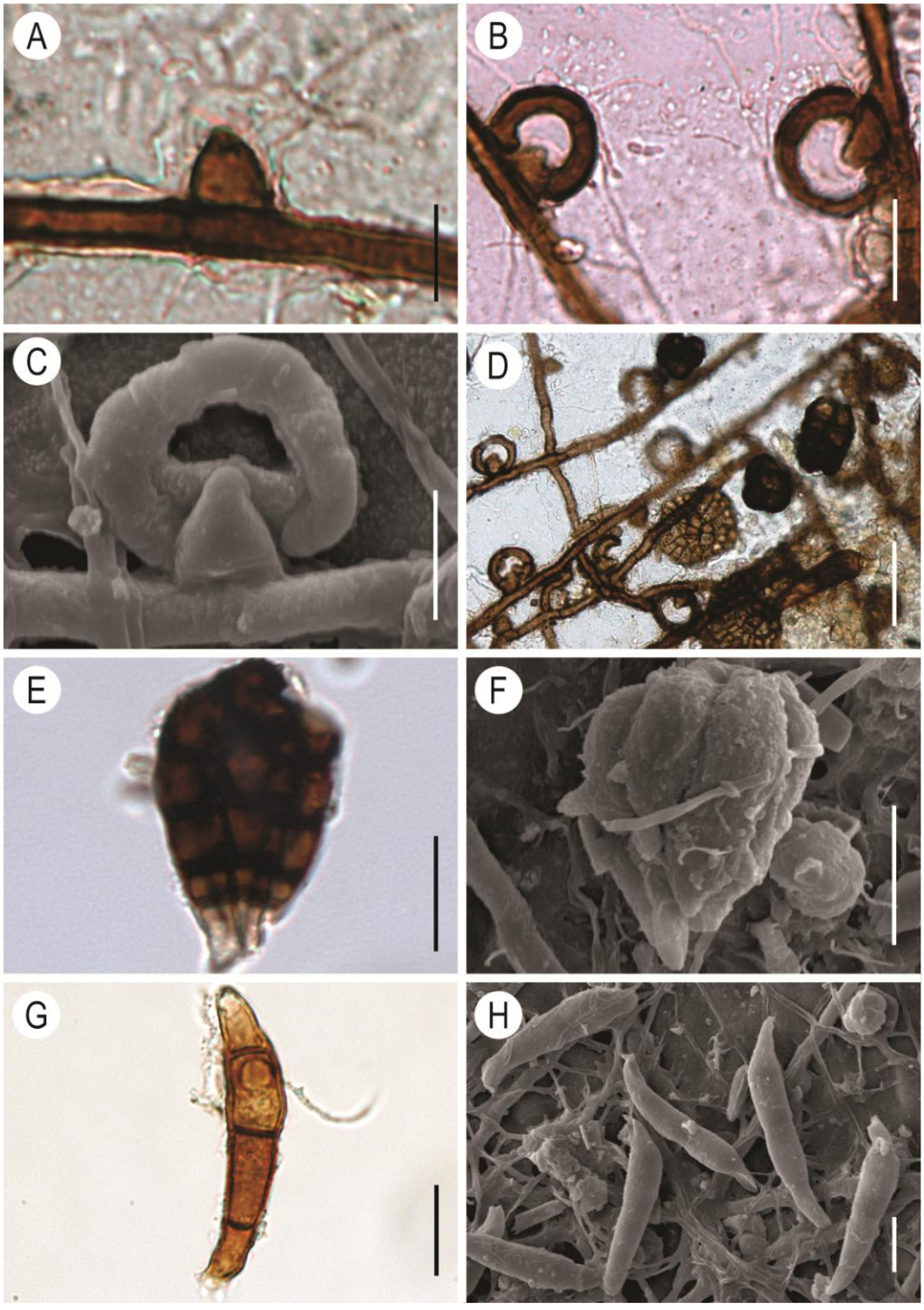


**Fig. 21.** *Digitosarcinella* sp. 1. A. Sessile, pyriform and unicellular apressoria. B. Apressoria with arched hyphae around. C. Apressoria with arched hyphae on SEM. D Ascomata on SEM. E. Dark brown with the lower end hyaline and smooth cheiroid conidia. F. Cheiroid conidia on SEM. G. Blastic, single, terminal, curver to arched and with 3-septate *Questieriella*. H. *Questieriella* on SEM. Scale bars: A–G–H= 10 µm; B= 200 µm; C= 100 µm; D–E= 50 µm; F= 20 µm.

***Rhytidenglerula trematis*** (Syd.) Arx. Beiträge zur Kryptogamenflora der Schweiz 11 (2): 1–922. 1962 Fig. 22.

*Colonies* hypophyllous, irregular to circular, single, becoming confluent, dark brown. *Hyphae* straight to flexuous, branching irregular, pale brown to brown, septate, hyphal cells cylindrical, 4–5 µm, smooth. *Apressoria* numerous, entire, sessile, lobed to denticulate, irregular in shape, unicellular, alternate to unilateral, never opposed, 6–9×6–9.5 µm, brown, penetration peg in central part of apressorial cell. *Ascomata* superficial, develop below surface mycelium, circular to irregular, single to clustered, partial deliquescence of the upper wall, 65.5–112.5 µm diam., dark brown to blackish at the margin, pale brown at central part. *Asci* globose, bitunicate, 8 ascospores, hyaline. *Ascospores* 2–celled, oblong-ovoid to ellipsoid, straight, with a slight constriction in the septum, 24–29×12.5–15 µm, brown with a dark band in each cell, smooth. *Anamorph* absent.

***Notes*:** *Rhytidenglerula trematis* (Syd.) Arx and *Rhytidenglerula trematicola* Castlebury are described on *Trema micrantha*. Both species are similar in ascomata, ascospore and asci measurements, but *R. trematis* has a dark band in each cell and a slight constriction in the septum. This is the third species of the genus *Rhytidenglerula* known from Brazil (Arx 1962) and this species was known from Venezuela and USA (Sydow 1930; Castlebury *et al.* 1995) and is reported here for the first time from Brazil.

*Specimens examined*: Brazil, Espírito Santo, Reserva Natural Vale do Rio Doce from leaves of *Trema micrantha* (*Cannabaceae*), 2012. A.L. Firmino, D.B. Pinho & O.L. Pereira.

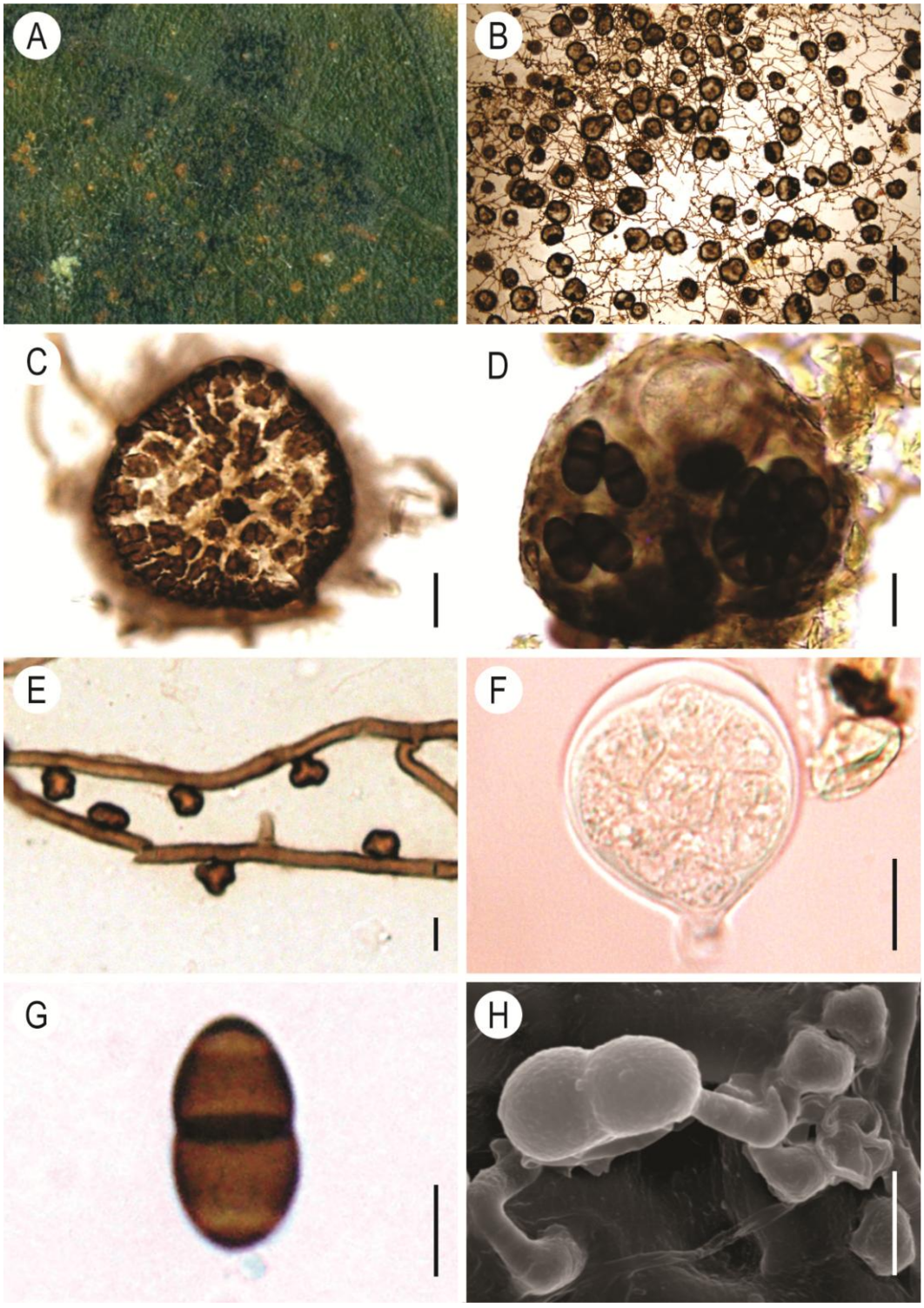


**Fig. 22.** *Rhytidenglerula trematis*. A. Colonies epiphylous. B. Part of a colony with open ascomata and surface mycelium. C. Ascomata with partial deliquescence of the upper wall. D Ascomata laterally. E. Mycelium and apressoria. F. Bitunicate ascus globose. G. Ascospore with dark bands. H. Smooth ascospore on SEM. Scale bars: B= 200 µm; C–D–F= 20 µm; E–G–H= 10 µm.

***Rhytidenglerula* sp. 1** (to be proposed as new) Fig. 23.

*Colonies* amphigenous, irregular to circular, single, becoming confluent, dark brown. *Hyphae* straight, branching opposed or alternate, brown, septate, hyphal cells cylindrical, 4.5–5.5 µm, smooth. *Apressoria* numerous, entire, sessile, rounded, cylindrical, unicellular, opposed, 7.5–12×5–7.5 µm, brown, penetration peg in central part of apressorial cell. *Ascomata* superficial, develop below surface mycelium, circular to irregular, single to clustered, partial deliquescence of the upper wall, 101–213 µm diam., dark brown to blackish at the margin, pale brown at central part. *Asci* ovoid, bitunicate, 8 ascospores, hyaline. *Ascospores* 2–celled, cylindrical, straight, 32.5–42.5×15–20 µm, brown verrucose. *Anamorph* absent.

***Notes*:** Eleven species of *Rhytidenglerula* have been described (Müller & Arx 1962). *Rhytidenglerula* sp. 1 differs from all other species in having opposed apressoria, larger and verruculose ascospore (Table 3).

*Specimens examined*: Brazil, Rio Grande do Sul, Bento Gonçalves. From leaves of *the Ilex paraguariensis* (Erva-mate) (*Aquifoliaceae*), 2011. A.L. Firmino.

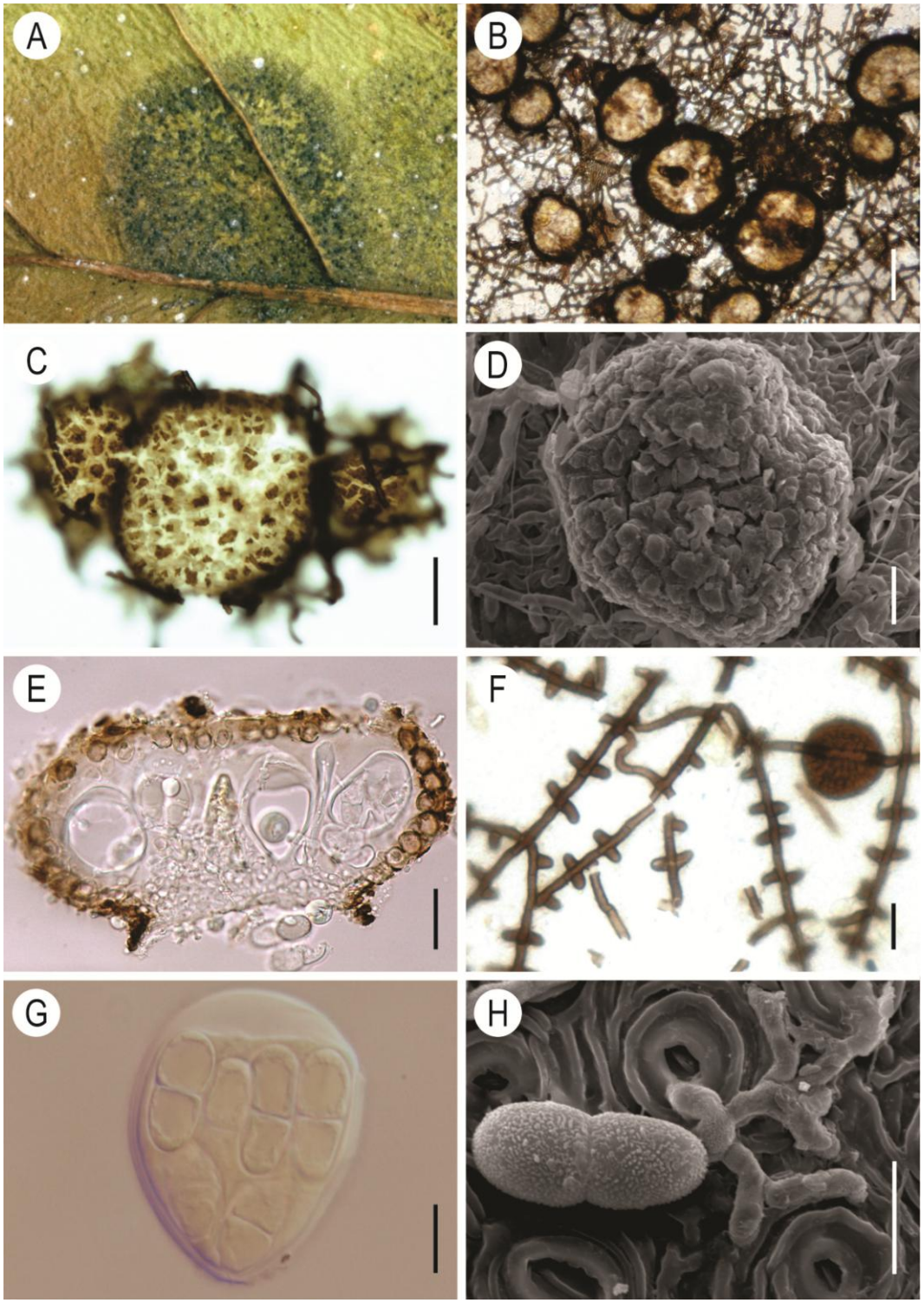


**Fig. 23.** *Rhytidenglerula* sp. 1. A. Colonies amphigenous B. Part of a colony with open ascomata and surface mycelium. C. Ascomata with partial deliquescence of the upper wall on microscope. D Ascomata with partial deliquescence of the upper wall on SEM. E. Cross section of the ascomata. F. Branching mycelium and composition of apressoria. G. Bitunicate ascus ovoid. H. Verruculose ascospore on SEM. Scale bars: B= 100 µm; C–E= 50 µm; D–F–G–H= 20 µm.

**Table 3.** Biometric data (µm) of the species of *Rhytidenglerula* spp. according to the literature.

| Species | Ascoma | Ascospore | Apressoria | Hyphae |
|---|---|---|---|---|
| *Rhytidenglerula* sp 1. | 101−213 | 32.5−42.5 × 15−20 | 7.5−12 × 5−7.5 | 4.5−5.5 |
| *Rhytidenglerula trematis*(Syd.) Arx This work | 62.5−112.5 | 24−29 × 12.5−15 | 6−9 × 6−9.5 | 4−5 |
| *Rhytidenglerula trematis* (Syd.) Arx | 50−110 | 24−28 × 13−15 | 10−12.5 × 7.5−10 | 4−6 |
| *Rhytidenglerula carnea* (Ellis & Mart.) Höhn. | 100−150 | 18−21 × 8−10 | 5−8 wide | 3−4 |
| *Rhytidenglerula cissi* (Syd.) Arx | 40−120 | 18−22 × 10−12.5 | 8−13 wide | 3.5−8 |
| *Rhytidenglerula davillae* (Hansf.) Arx | 100 | 18 × 8 | 8−15 × 8−10 | 5 |
| *Rhytidenglerula hendrickxii* (Hansf.) Arx | 75 | 22−25 × 10−12 | 8−12 × 11−14 | 6−8 |
| *Rhytidenglerula homalanthi* (Hansf.) Arx | 80−120 | 13−14 × 6−7 | 10−14 wide | − |
| *Rhytidenglerula landolphiae* (Hansf.) Arx | 80−130 | 21−24 × 8−9 | 18−30 × 3−4 | 3−4 |
| *Rhytidenglerula minima* (Hansf.) Arx | 40−50 | 15 × 6 | 8 × 3−4 | 3 |
| *Rhytidenglerula solani* Bat.,Bezerra & Matta | 57−107 | 22−25 × 11−13 | 9−13 × 8−11 | 5.5−7.5 |
| *Rhytidenglerula trematicola* Castlebury | 46−80 | 18−26 × 10−14 | 8−10 wide | 3−5 |
| *Rhytidenglerula vilis* (Petr.) Arx | 50−100 | 18−21 × 9−11 | 7−9 × 5−6.5 | 2.5−5 |

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