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Novel and uncommon bryophilous fungi from Brazil

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Abstract. Bryophilous fungi are an understudied group with unknown biodiversity in tropical regions, and are often underestimated worldwide. In Brazil, research on this ecological group has primarily focused on the southern and southeastern regions. During a survey of bryophilous fungi in the northeastern region, specifically in Bahia State, two novel species, *Brachydesmiella bahiensis* and *Dendryphiosphaera longispora*, were discovered. *Brachydesmiella bahiensis* resembles *B. biseptata* and *B. caudata*, but exhibits conidial distal cell morphology between these two species. *Dendryphiosphaera longispora* has longer conidia than other species within the genus. Also, the uncommon microfungus *Dwibeeja sundara* was reported for the second time globally. Detailed descriptions and illustrations of each species are provided.

Key words: asexual ascomycetes, fungal diversity, liverworts, taxonomy, tropical fungi

Introduction

The term bryophilous fungi is used to describe fungi that grow on or near bryophytes, particularly mosses and liverworts, as originally described by Racovitza (1959). According to Döbbeler (1997), mosses and liverworts are excellent fungal hosts. In forest environments, these fungi typically prefer areas covered by mosses (Grzesiak & Wolski 2015).

Pressel et al. (2010) proposed that investigating the interactions between bryophytes and fungi could provide new perspectives on the colonization of terrestrial environments. They presented a comprehensive review of the enduring partnerships between fungi and bryophyte rhizoids, illustrating the diversity and historical significance of these relationships.

Davey & Currah (2006) noted that bryophilous pathogens exhibit structural and functional adaptations to their habits. Fungi explore different nutritional microniches within the gametophyte and possess the capacity to degrade the cell wall, which contains abundant polyphenolics compounds (lignin-like). This degradation may represent a product of the co-evolution of plants and parasites or a recent change in these fungi to this habit (Davey & Currah 2006). Felix (1988) stated that fungus parasitism is not always evident and depends on the presence of intracellular hyphae. Furthermore, this author considers that many associations may be coincidental, because fungi and bryophytes frequently occur on the same substrates, mainly decaying wood and litter.

According to Racovitza (1959), most bryophyte-associated fungi are saprobes. Greiff (2019) discussed the different modes of nutrition of bryophilous fungi and highlighted the lack of knowledge regarding saprobes. Analysis of the morphological and ecological characteristics of these fungi, their relationships with non-bryophilous fungi, interactions with hosts, specificity, geographic distribution, and decomposition can expand our thinking regarding the systematics and biology of fungi (Döbbeler 1997).

Ptaszyńska et al. (2009) emphasized the significance of bryophytes in understanding global mycodiversity. They argued that 90% of known fungi are associated with plant substrates or their derivatives. A significant portion of mycological inventories are related to vascular plants, especially those of economic interest. However, bryophytes are distributed worldwide, even in polar regions, presenting the potential for discovery of a substantial associated fungal diversity.

In Brazil, more than 1,600 species of bryophytes have been recorded, of which 18% are endemic (Costa & Luizi-Ponzo 2010; Koga et al. 2021). Approximately 280 species (including mosses and liverworts) have been recorded in the State of Bahia, with 130 species identified in a single study conducted at Serra da Jibóia (Valente et al. 2009), underscoring the remarkable diversity of this

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group. Studies on bryophilous fungi in Brazil are incipient and disperse, and are marked by scattered observations and significant knowledge gaps. Racovitza (1959), Döbbeler (1978; 2003), Felix (1988) and Döbbeler & Hertel (2013) documented eight fungal species from Brazil, and, recently, 50 bryophilous fungi have been reported from the southern and southeastern regions (Grandi et al. 2008; Furlan-Lopes et al. 2022). This study aimed to investigate microfungi associated with liverworts, contributing to the understanding of bryophilous fungal diversity and geographic distribution.

Material and methods

The Atlantic Forest, reduced to only 8% of its original area in Brazil, is widely recognized as a crucial priority area for the conservation of global biodiversity (Mittermeier et al. 2004). Serra da Jiboia is one of the last remaining fragments of this forest in northeastern Brazil, covering a region of 440 km² (Ramos et al. 2020). The area has a subhumid to dry climate, altitudes reaching 820 m above sea level and an average annual precipitation of 1,200 mm (Blengini et al. 2015).

Expeditions were carried out in Serra da Jibóia to gather specimens of epixilious liverworts in search of microfungi. Samples were placed in paper bags and sent to the Mycology Laboratory of the State University of Feira de Santana (UEFS). They were regularly examined for 60 days under a Leica S8APO stereomicroscope to collect reproductive structures. Permanent slides were mounted using PVL resin (polyvinyl alcohol + lactic acid + phenol), and deposited in the Herbarium of the State University of Feira de Santana (HUEFS). Microphotographs were obtained using an Olympus BX51 microscope equipped with a DP25 digital camera. Unfortunately, despite multiple efforts, pure cultures of the specimens under investigation could not be obtained.

Results

Two new species, *Brachydesmiella bahiensis* and *Dendryphiosphaera longispora*, are introduced, and the second global occurrence of *Dwibeeja sundara* Subram. is detailed with accompanying illustrations. Due to the unavailability of molecular data for all genera treated here, their precise phylogenetic placement remains undetermined, and they are considered "*Incertae sedis*", belonging to the subphylum Pezizomycotina, Ascomycota (IndexFungorum 2024).

Taxonomy

Brachydesmiella bahiensis S.M. Leão & Gusmão, sp. nov. (Fig. 1A–C)

MycoBank MB 852552

Diagnosis: Morphologically, it closely resembles *B. biseptata* and *B. caudata*, but differs by having intermediate measurements of the apical cells of conidia.

Type: Brazil, Bahia State: Santa Terezinha, Serra da Jibóia (12°51′13.9″S, 39°28′32.5″W, 800 m), 11 June 2022, on liverworts *Riccardia* sp., leg. S.M. Leão-Ferreira (HUEFS 266443 – holotype).

Etymology. The name is in reference to Bahia state, where the specimen was collected.

Ecology. On epixilous liverworts.

Distribution. Only known from the type.

Description. Colonies on the natural substrate caespitose, effuse, brown. Mycelium superficial and immersed. Conidiophores differentiate, simple or rarely branched, erect, sinuate to slightly geniculate, somewhat nodose, smooth, 1–2-septate, pale brown, 20–50 × 5–7.5 µm (n = 50). Conidiogenous cells polytretic, terminal and intercalary, integrated, indeterminate, sympodially, smooth-walled, pale brown. Conidiogenous loci slightly melanized around

Table 1. Synopsis of the *Brachydesmiella* species.

Species	Conidiophores (µm)	Conidia				References
		Form	Size (µm)	Apical cell (µm)	Septa	
B. anthostomelloidea	40–90 × 3.5–4.5 × 5–7	Limoniform to ampulliform	35–47 × 14–18	ausent	1	Goh & Hyde (1996), Sivichai et al. (1998), Castañeda-Ruiz et al. (2006), Barbosa & Gusmão (2011)
B. biseptata	28–70 × 4–6 × 5–9	Limoniform	36–51 × 15–22	3.9–7.5 × 3.9–7.5	1–2	Hughes (1971), Goh & Hyde (1996), Sivichai et al. (1998), Castañeda-Ruiz et al. (2006), Jiang et al. (2008)
B. brasiliensis	20–110 × 6–7	Navicular to narrow fusiform	30–36 × 6–7	–	3	Castañeda-Ruiz et al. (2006)
B. caudata	19–46 × 3.8–8	Limoniform-caudate	52–77 × 13–33	17–25 × 2–3.8	2	Hao & de Hoog (1986), Sivichai et al. (1998), Castañeda et al. (2006)
B. eugecapiellana	Up 70 × 6–8	Navicular to narrow fusiform	32–40 × 4–6.5	–	2–3	Castañeda et al. (2006)
B. bahiensis	20–50 × 5–7.5	Limoniform	38–53 × 18–20	8–11 × 5–6	2	This paper
B. obclavata	40–75	Obclavate	18–23(–26) × 3	–	1	Castañeda-Ruiz et al. (2006)
B. orientalis	20–60 × 5–8	Pyriform	30–37.5 × 17–22.5	–	1(2)	Hao & de Hoog (1986), Sivichai (1998), Castañeda-Ruiz et al. (2006)
B. verrucosa	36–76 × 5–7 × 5–8	Ampulliform	56–92 × 12–17	22–46 × 4–6	2	Castañeda-Ruiz et al. (2006)

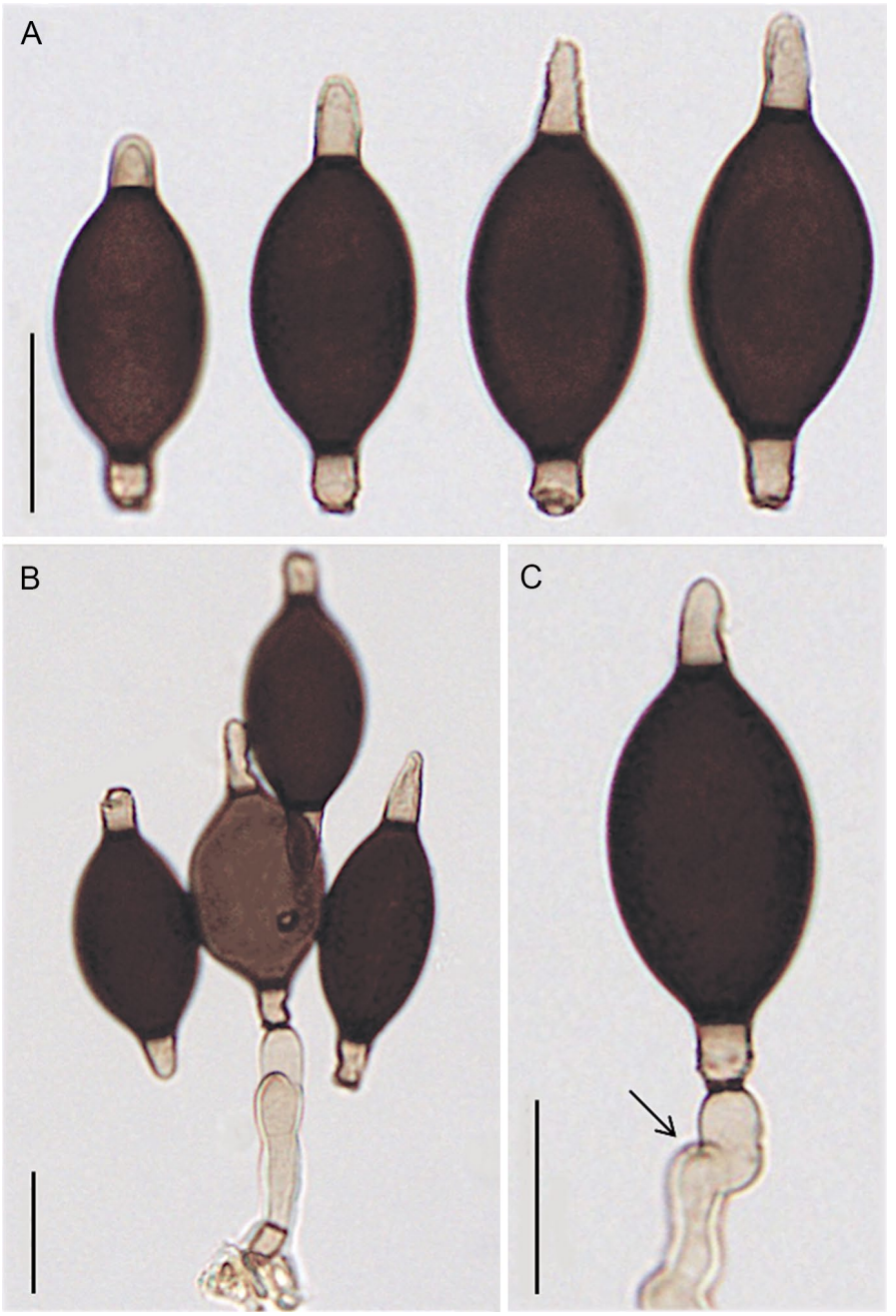

Figure 1. *Brachydesmiella bahiensis* (HUEFS 266443 – holotype). A – conidia; B – general aspect; C – detailed conidiogenous cell. Scale bars = 20 µm.

the pores. Conidial secession schizolytic. Conidia acrogenous, solitary, dry, limoniform, unequal colored, 2-septate, 41–55.5 × 14.5–35 µm (n = 50), apical cell pale brown, cylindrical to acicular, slightly verrucose, 3.7–13.7 × 3.5–6 µm (n = 50), wider central cell dark brown, limoniform, smooth, 27.5–37.5 × 14.5–35 µm (n = 50), basal cells pale brown, cylindrical, truncated at the base, slightly verrucose, 6–8 × 5–6 µm (n = 50).

Notes. *Brachydesmiella* G. Arnaud ex S. Hughes is characterized by erect, nodose conidiophores with polytretic, indeterminate conidiogenous cells and solitary, septate conidia (Hughes 1961). Sivichai et al. (1998) redescribed and illustrated all known species at the time, including a new combination, *B. orientalis* (V. Rao & de Hoog) Goh and a new species *B. verrucosa* Goh, Sivichai, KD Hyde & Hywel-Jones. However, these authors described all species as having polyblastic conidiogenesis, which differs from the circumscription of the genus given by Hughes (1961), and followed by several authors (Hughes 1971; Rao & de Hoog 1986; Goh & Hyde 1996; Castañeda-Ruiz et al. 2003, 2006) that characterized the conidiogenesis as polytretic. Currently, eight species are known, four of which occur in Brazil: *B. anthostomelloidea* Goh & K.D. Hyde, *B. brasiliensis* R.F Castañeda, Gusmão & Heredia, *B. caudata* V. Rao & de Hoog, and *B. obclavata* R.F. Castañeda, Gusmão & Saikawa (Castañeda-Ruiz et al. 2006; Barbosa & Gusmão 2011). Countries such as India, Korea, Mexico, New Zealand, the United Kingdom and Venezuela also have records of this genus (GBIF 2024).

Among the *Brachydesmiella* species (Table 1), *B. anthostomelloidea*, *B. obclavata*, *B. orientalis* (V. Rao

& de Hoog) Goh have 1-septate conidia and *B. eugecapiellana* R.F Castañeda, Iturr. & Saikawa and *B. brasiliensis* have 2 or 3 septate conidia (Goh & Hyde 1996; Sivichai et al. 1998; Castañeda-Ruiz et al. 2003) (Fig. 2). Based on the number of septa, *B. bahiensis* is more closely related to *B. biseptata* G. Arnaud ex S. Hughes, *B. caudata*, and *B. verrucosa* Goh, Sivichai, K.D Hyde & Hywel-Jones. *Brachydesmiella verrucosa* differs in shape, pigmentation, ornamentation, and conidial dimensions and is much larger than other species in the genus (Sivichai et al. 1998; Castañeda-Ruiz et al. 2006). In contrast, *B. biseptata* and *B. caudata* have similar morphology, pigmentation, and ornamentation of their apical and basal cells, which are slightly rough. The conidia of *B. biseptata* have smaller basal and apical cells, while *B. caudata* has larger conidia basal and apical cells compared with *B. bahiensis*.

Additional examined material. *Brachydesmiella anthostomelloidea* Goh & K.D. Hyde (HUEFS 155249; HUEFS 155250). *Brachydesmiella brasiliensis* R.F Castañeda, Gusmão & Heredia (HUEFS 97984 – holotype!). *Brachydesmiella caudata* V. Rao & de Hoog (HUEFS 165766). *Brachydesmiella obclavata* R.F. Castañeda, Gusmão & Saikawa (HUEFS 97983 – holotype!).

Dendryphiosphaera longispora S.M. Leão & Gusmão, sp. nov. (Fig. 3A–D)

MycoBank MB 852553

Diagnosis: Differs from all described species by the length of its conidia.

Type: Brazil, Bahia State: Santa Terezinha, Serra da Jibóia (12°51′13.9″S, 39°28′32.5″W, 800 m), 26 Apr. 2022, on liverworts, leg. S.M. Leão-Ferreira (HUEFS 266444 – holotype).

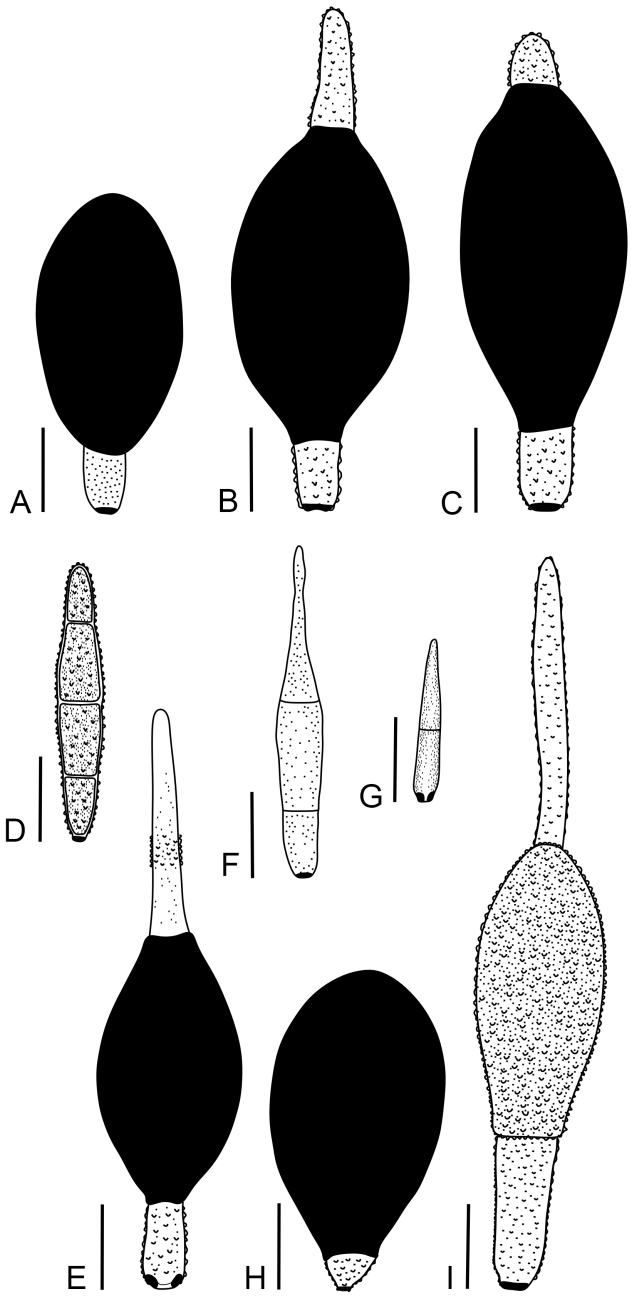

Figure 2. *Brachydesmiella* conidia. A – *B. anthostomelloidea*; B – *B. bahiensis*; C – *B. biseptata*; D – *B. brasiliensis*; E – *B. caudata*; F – *B. eugecapiellana*; G – *B. obclavata*; H – *B. orientalis*; I – *B. verrucosa*. Scale bars = 10 μm.

Table 2. Synopsis of the *Dendryphiosphaera* species.

Species	Conidiophores (µm)	Conidiogenous cells			Conidia			References
		Form	Size (µm)	Nº chains	Form	Size (µm)	Septa	
D. minuta	200–300 × 7	Suglobose to obovoid	–	4	Obovoid	11–14 × 5–6.5	2	Rao & de Hoog (1986)
D. longispora	100–400 × 7.5–13	Lageniform	5–15 × 3–6	Up 6	Clavate to cylindrical	12.5–23 × 4–9	2–3 (+2)	This paper
D. parvula	60–185 × 5–8	Subglobose to ovoid	5–11 × 3–4	2	Clavate obovoide	8–12 × 3.5–4.5	1 1 (pseudo)	Nawawi & Kuthubutheen (1988)
D. taiensis	140–368 × 4.5–8.9	Globose	3.5–4.5 × 2.3–4.8	2	Elliptic to cylindrical	10.5–16 × 4–6.9	2	Lunghini & Rambelli (1978), Leão-Ferreira et al. (2013)
D. uniseptata	140–200 × 5–8	Cylindrical to lageniform	5–8 × 2.5–3	0	Clavate to cuneate	10–13 × 2–3	1	Castañeda-Ruiz et al. (1998)

Description. Colonies scattered, black. Mycelium superficial, immersed. Conidiophores macronematous, mononematous, unbranched, erect, straight or slightly flexuous, 7–12 septate, smooth, brown to dark brown, 100–400 × 7.5–12.5 µm (n = 50). Conidiogenous cells monoblastic, lateral and terminal, discrete, percurrent, sometimes whorled, up 6 extensions, lageniform, smooth, pale brown to brown, 3.5–11 × 2.5–5 µm (n = 50). Conidial secession schizolytic. Conidia acrogenous, solitary, dry, clavate to cylindrical, pale brown, equally pigmented, 2–3-septate, 12.5–23 × 5–8.5 µm (n = 50).

Etymology. The name is in reference for the long conidia of the fungus.

Ecology. On epixilous liverworts.

Distribution. Only known from the type.

Notes. *Dendryphiosphaera* was established by Lunghini & Rambelli (1978) with the type species *D. taiensis* Lunghini & Rambelli. This genus has setiform brown conidiophores, with conidiogenous cells forming whorls in the apical region and solitary, septate, or pseudoseptate conidia with schizolytic secession (Fig. 4). Subsequently, three species were proposed: *D. minuta* V. Rao & de Hoog, *D. parvula* Nawawi & Kuthub., and *D. uniseptata* R.F. Castañeda & Guarro (Rao & de Hoog 1986; Nawawi & Kuthubutheen 1988; Castañeda-Ruiz et al. 1998). *Dendryphiosphaera parvula* and *D. taiensis* have been recorded in Brazil by Almeida et al. (2011) and Leão-Ferreira et al. (2013). The geographic distribution of this genus is restricted to intertropical countries such as Brazil, Ivory Coast, Cuba, India, Malaysia, and Mexico (GBIF 2024).

The genera *Dendryphiosphaera* is closest with *Brachysporiella* Bat. and its synonyms: *Edmundmasonia* Subram., *Monosporella* S. Hughes, *Monotosporella* S. Hughes (Indexfungorum 2024). Rao & de Hoog (1986) suggested that *Dendryphiosphaera* and *Brachysporiella* Bat. be separated based on the latter having larger, darker conidia with thick walls.

Upon closer examination of the descriptions and illustrations of the type material of *B. gayana* Bat., it becomes evident that numerous species classified under *Brachysporiella* do not align well with the original description. Examples include *B. biseptata* Backer & Partr., *B. dennisii* J.L. Crane & Dumont, *B. indica* Munjal & H.S. Gill, *B. navarica* Hern.-Restr., R.F. Castañeda & Gené, and *B. sinensis* W.P. Wu. These discrepancies are particularly notable in the conidial morphology, especially the short distance present in the basal septa and the pronounced presence of the swollen apical cell. On the other hand, phylogenetic analysis of *B. navarica* and *B. setosa* (Berk. & M.A. Curtis) M.B. Ellis (formerly *Monotosporella setosa*) provided evidence that the former belongs to the order Kirschsteiniotheliales and the latter to Pleurotheciales (Hernández-Restrepo et al. 2017), highlighting the necessity for an extensive morphological and phylogenetic study to accurately delineate the species within this genus. Despite the similarities between *Dendriosphaeria* and *Brachysporiella* "s.s.", we consider them to be distinct genera.

The size and septation of conidia (Table 2) are the most relevant characteristics for differentiating *Dendryphiosphaera* species (Rao & de Hoog 1986; Nawawi & Kuthubutheen 1988; Castañeda-Ruiz et al. 1998). Other species in the genus have smaller conidiophores and conidia than *D. longispora*, as well as fewer extensions of conidiogenous cells and septa in the conidia. *Dendryphiosphaera minuta* has up to four extensions of conidiogenic cells. However, these are subglobose to ovoid and their conidia are smaller than those of the proposed species. *D. parvula* and *D. taiensis*, in addition to differing in the morphology of the conidiogenic cells, have only two extensions and conidia are smaller.

Additional examined material. *Dendryphiosphaera parvula* Nawawi & Kuthub. (HUEFS 154969). *Dendryphiosphaera taiensis* Lunghini & Rambelli (HUEFS 136879).

Dwibeeja sundara Subram, Kavaka 20/21(1–2): 57. 1995. (Fig. 5A–C)

Description. Colonies scattered, black. Mycelium superficial, and immersed. Conidiomata indeterminate, synnematous, intricate stipe with terminal conidiogenous cells, fertile along the stipe and at the ápice, black, 70–182 × 30–75 µm (n = 30). Conidiophores macronematosous, sinematous, erect, straight or slightly flexuous, smooth, dark. Conidiogenous cells monoblastic, determinate, terminal, discrete, cylindrical to pyriforme, smooth, brown to dark brown, 13–22 × 6–8 × 2–3 µm (n = 30). Conidial

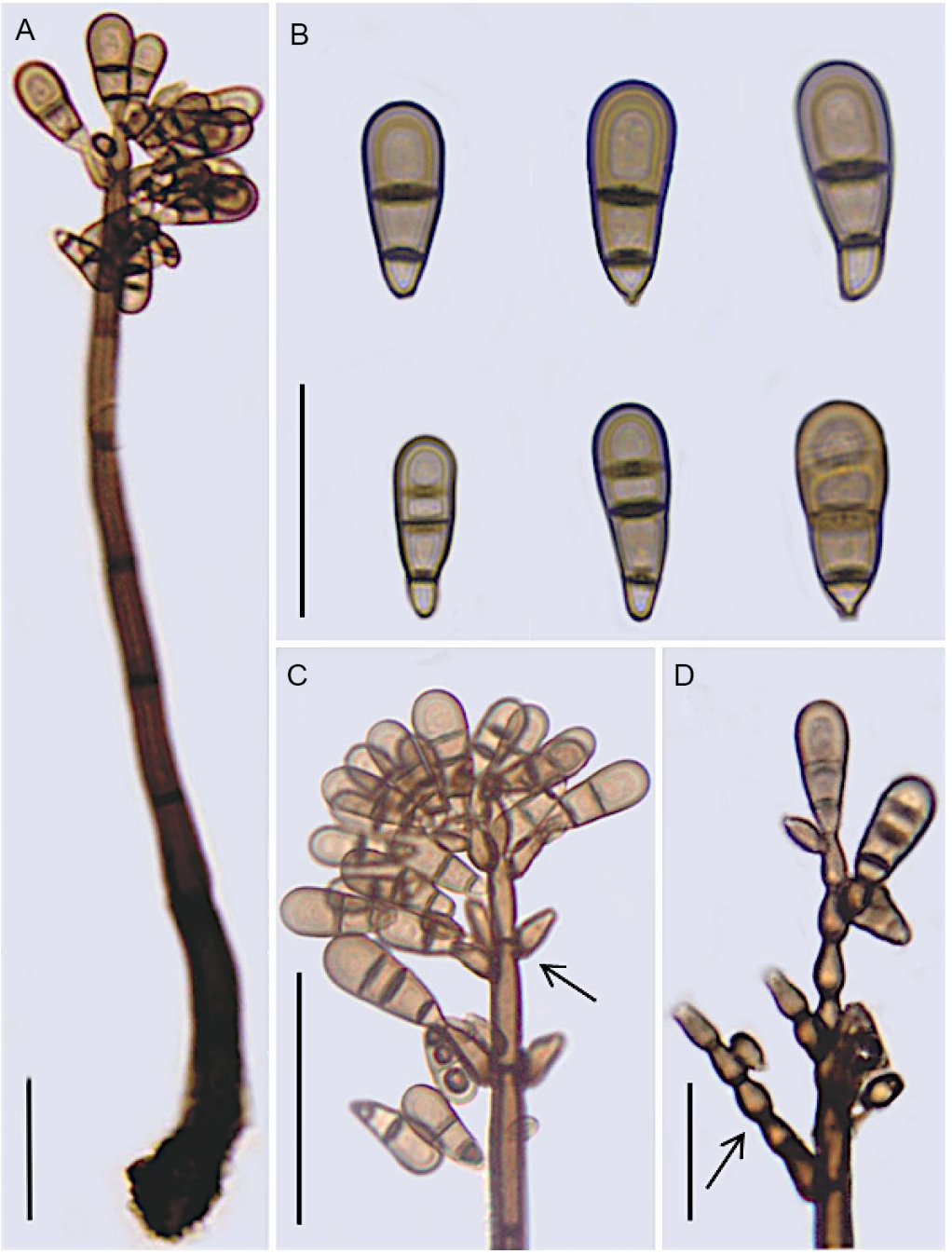

Figure 3. *Dendryphiosphaera longispora* (HUEFS 266444 – holotype). A – general aspect; B – conidia; C – arrangement conidiogenous cell; D – detailed catenate conidiogenous cell. Scale bars = 20 µm.

secession schizolytic. Conidia acrogenous, amero, dry, in acropetal chains of two, globose to limoniform, smooth, dark brown to black, apical conidia with light or dark brown mamillate at each end, 15–20 × 11–13 µm (n = 30), basal conidia truncate at the base, 17–21 × 10–13 µm (n = 30).

Ecology. On epixilous liverworts.

Distribution. Brazil (this paper) and Singapore (Subramanian 1992).

Notes. *Dwibeeja* was proposed by Subramanian as the type species, *D. sundara*, which is associated with the decomposition of *Calophyllum inophyllum* L. branches in Singapore (Subramanian 1992). To date, no other records of this species have been published. It is a monotypic genus characterized by the formation of a synnema with simple conidiogenous cells that project in the upper portion, where the acropetal chains of two black conidia are formed. *Yinmingella* Goh, C.K.M. Tsui & K.D. Hyde is considered a genus similar to *Dwibeeja* in terms of ontogeny and conidial morphology, however, this genus presents sporodochia conidioma with stroma (Seifert et al. 2011).

Until now, this species has been found as a saprobic fungus in terrestrial environments, and the sexual phase and phylogenetic position remain unknown (Wijayawardene et al. 2017). The conidioma of the specimen studied is shorter and wider when compared to the original description [up 250 × 20–40(–60) µm], which may suggest

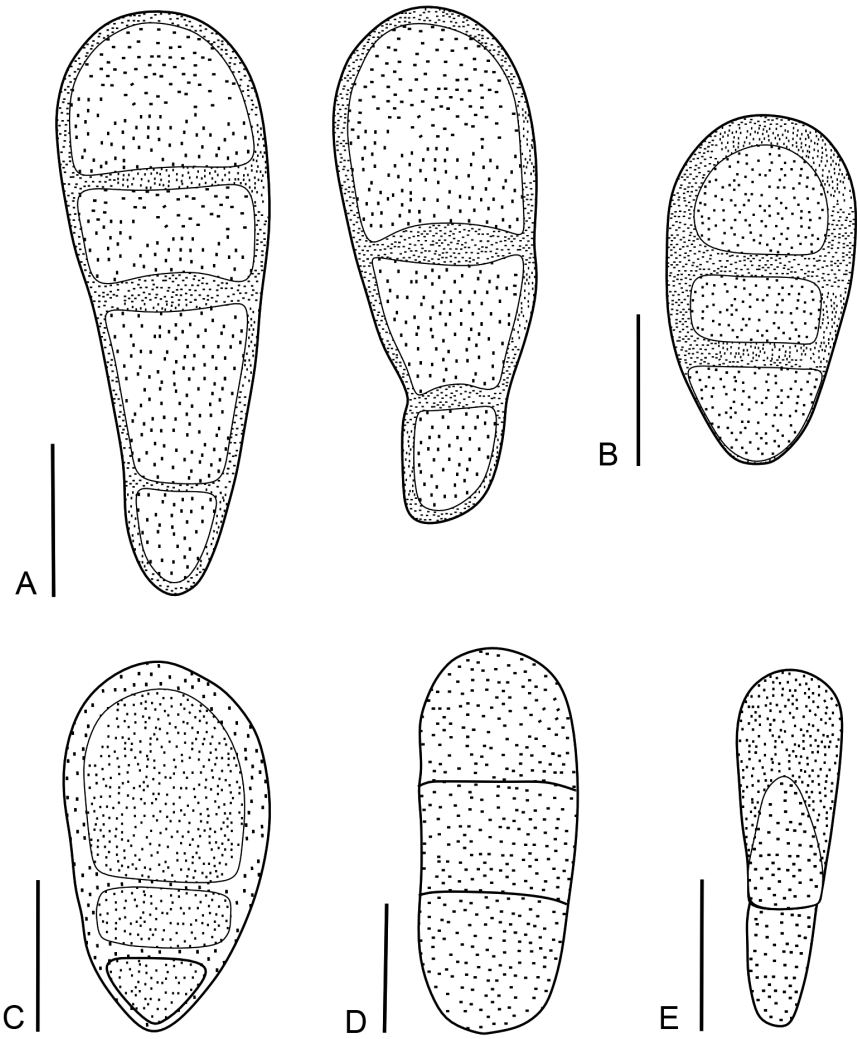

Figure 4. *Dendryphiosphaera* conidia. A – *D. longispora*; B – *D. minuta*; C – *D. parvula*; D – *D. taiensis*; E – *D. uniseptata*. Scale bars = 10 µm.

an adaptation to the substrate whose dimensions are considerably smaller than those of a vascular plant. However, the conidiogenous cells, have slightly larger dimensions compared to the type (up 20 × 3–7 × 2–5 µm; 13–22 × 6–8 × 2–3 µm), except for the apex of the conidiogenous cell. Therefore, there is variation in the shape of the conidiogenous cells in both materials, from cylindrical/subcylindrical to cylindrical/piriform. The young conidia are darker than those observed by Subramanian (1992), and the apical conidia are generally globose. The mode of secession of the conidia is schizolitic as reported by Seifert et al. (2011). This species has been collected only in Singapore in the Asian continent.

Specimen examined. BRAZIL. Bahia State: Santa Terezinha, Serra da Jibóia (12°51′13.9″S, 39°28′32.5″W, 800 m), 04 Dec. 2021, on decaying *Frullania* sp., leg. S.M. Leão-Ferreira (HUEFS 266445).

Discussion

Brachydesmiella species decay submerged or terrestrial wood in temperate and tropical regions (Sivichai et al. 1998; Castañeda-Ruiz et al. 2006). *Dendryphiosphaera* is recorded on almost all continents associated with decaying leaves and wood (Rao & de Hoog 1986; Nawawi & Kuthubutheen 1988; GBIF 2024). The species described here were also observed growing on bryophyte substrates. Since other records for the species of *Brachydesmiella* and *Dendryphiosphaera*, as well as for the type *Dwibeeja*, are related to vascular plants, we consider the association of these species optional. According to Felix (1988), the association between fungi and bryophytes can be obligatory or optional because both occur on the same substrate. The discovery of new species in this study reinforces the argument for the great diversity of bryophilous fungi still to be discovered and the need for more taxonomic surveys to better understand the biology of this group. None of these genera have sequences available in GenBank, and attempts to cultivate the specimens presented here failed, therefore, morphological characteristics were predominant in the definition of taxa.

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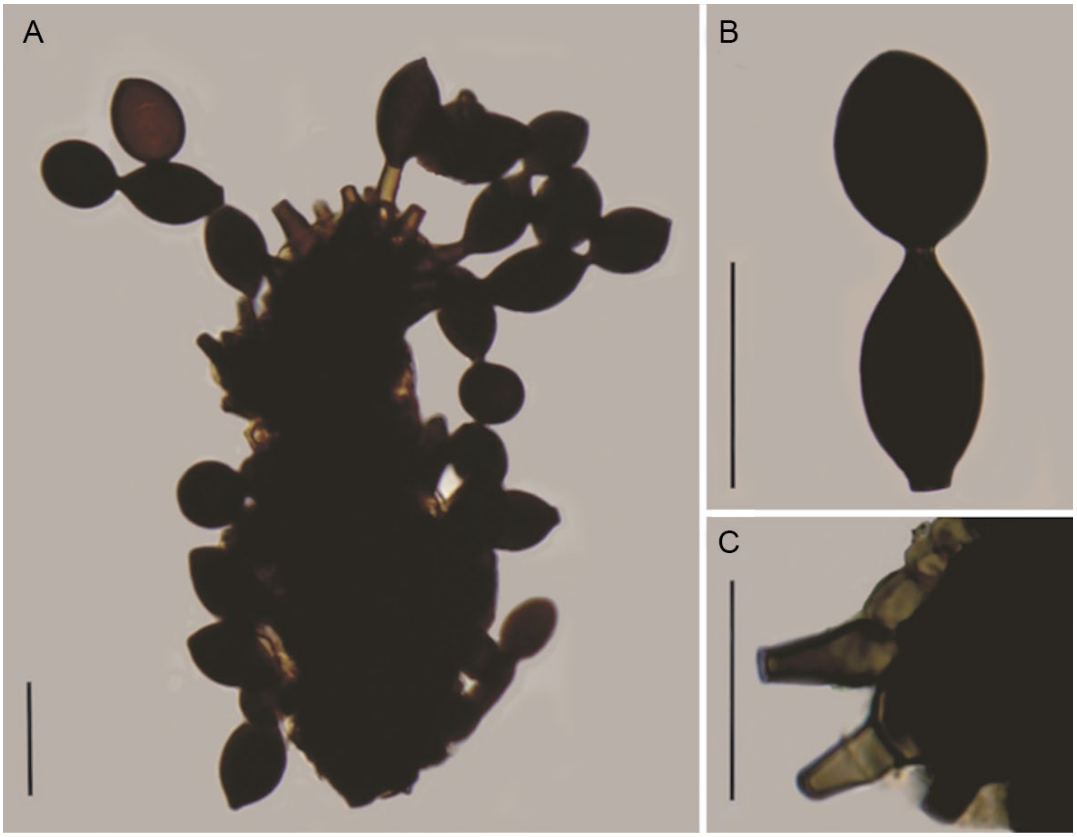

Figure 5. *Dwibeeja sundara* (HUEFS 266445). A – general aspect; B – catenate conidia; C – detailed conidiogenous cell. Scale bars = 20 µm.

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