

# The genus *Brachydesmiella* from submerged wood in the tropics, including a new species and a new combination

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During studies of fungi occurring on wood submerged in freshwater streams in the tropics, five species of *Brachydesmiella*, namely, *B. anthostomelloidea*, *B. biseptata*, *B. caudata*, *B. orientalis* comb. nov., and *B. verrucosa* sp. nov., were collected. These are described and illustrated from natural substrata, wooden test blocks, and from pure culture, based mainly on collections made in Thailand. *Brachydesmiella verrucosa* differs from the other four species in having 3-celled, coarsely verrucose, ampoule-shaped conidia, with a long, thin, cylindrical, verruculose apical cell. A key to *Brachydesmiella* species is provided.

Key Words—aquatic fungi; Hyphomycetes; mitosporic fungi; systematics.

Species of *Brachydesmiella* G. Arnaud ex S. Hughes (Hughes, 1961) are known from decaying wood around the world. *Brachydesmiella biseptata* G. Arnaud ex S. Hughes, the type species, has been previously reported from countries in the temperate regions, e.g., Canada, France, Japan, New Zealand, Uganda, and United Kingdom (Arnaud, 1953; Hughes, 1961, 1971; Ellis, 1971; Nicot, 1971; Tubaki, 1975; Matsushima, 1983). Three other members of the genus, however, are known from wood submerged in freshwater in the tropics: *B. anthostomelloidea* Goh & K. D. Hyde, *B. biseptata* var. *orientalis* V. G. Rao & de Hoog, and *B. caudata* V. G. Rao & de Hoog (Rao and de Hoog, 1986; Goh and Hyde, 1996).

During surveys of lignicolous freshwater fungi in the tropics, we have collected all four *Brachydesmiella* taxa. In this paper, these taxa are redescribed and illustrated. A further species, *B. verrucosa* sp. nov., isolated from decaying wood submerged in freshwater streams in Australia and Malaysia and from submerged test blocks in streams in Thailand, is also described. This new taxon differs from all previously described species of *Brachydesmiella* in having 3-celled, coarsely verrucose, ampoule-shaped conidia, with a long, thin, cylindrical, verruculose apical cell. Collections and cultural details of other *Brachydesmiella* taxa collected in Thailand are also given. Based on observations from natural substrata and from pure culture, we consider *B. biseptata* var. *orientalis* to be a distinct species, rather than a variety. A new combination is therefore proposed. An amended key to *Brachydesmiella* species, based on Goh and Hyde (1996), is provided.

## Materials and Methods

Specimens of naturally submerged wood were collected from streams in Australia, Hong Kong, Malaysia, and Thailand. In Thailand, an experiment to look at natural colonization of soft and hard woods (*Alstonia scholaris* and *Anisoptera oblonga*, respectively) in small streams in Khao Yai National Park, Nakorn Ratchassima Province has been in progress since September, 1996. Test blocks (15 × 2.5 × 2.5 cm), free of wood preservative, threaded on nylon rope, with five blocks (of the same species) per set were submerged in two streams. Each month, test blocks were recovered from the two sites.

All wood samples (both natural and test blocks) were placed in separate polyethylene bags and taken to the laboratory. Material was incubated in plastic boxes on sterilized damp tissue paper at 20–22°C. Material was examined periodically under a stereo-microscope and the fungi were identified. Preparations were mounted in lactophenol-cotton blue and sealed with polyvinyl alcohol. Single-conidium isolations were made and grown on corn-meal agar (CMA) or potato-dextrose agar (PDA). The conidia were spread over the agar surface with a flame-sterilised inoculation loop dipped in 0.05% (w/v) Triton X-100. Plates were incubated at 20°C in a cabinet with cool white fluorescent light and examined with a microscope each day for signs of germination. Six to eight of the germinated conidia were transferred to new plates and incubated in the same cabinet. Slides from cultures were made in water or in lactophenol-cotton blue. Dried specimens collected in Thailand in this study are kept in the BIOTEC herbarium collection, whereas those collected in Australia, Hong Kong, and Malaysia are deposited in HKU(M).

## Taxonomy

*Brachydesmiella anthostomelloidea* Goh & K. D. Hyde, Mycol. Res. **100**: 1364–1366. 1996. Figs. 1, 2

Colonies on PDA reaching a diam of 20 mm in 60 d at 25°C with normal daylight illumination. The colonies composed of three growing zones: the outer growing zone effuse, very pale milky brown, ca. 1.5–2.5 mm wide; the middle zone slightly raised, fluffy, white, composed of aerial hyphae 2–3 µm in diam; the central region dome-shaped, sporulating, grey, with aerial mycelium composed of numerous conidiophores and conidia at different stages of development. Conidiogenous cells integrated, polyblastic, sympodially proliferating, with conspicuously thickened conidial scars. Conidiophores clavate, 40–90 µm long, 3.5–4.5 µm wide at the base, 5–7 µm wide at the apex. Conidia pyriform, 2-celled, 35–47 × 14–18 µm, basal cell cylindrical, central cell darkly olivaceous brown.

Specimen examined: Australia, North Queensland, Mount Lewis, on unidentified wood submerged in a small rain forest stream, Apr. 1995, T. M. and K. D. Hyde, BRIP 23200.

Known distribution: Australia.

*Brachydesmiella biseptata* G. Arnaud ex S. Hughes, Can. J. Bot. **39**: 1095. 1961. Figs. 3–6

Colonies on natural substrate effuse, farinose, black, shiny. Conidiophores scattered or in small groups, smooth, thick-walled, pale brown, cylindrical-clavate, narrower at the basal part, flexuous, rarely branched, with 0–2 septa, 28–54 µm long, 5–8 µm wide, base 4–6 µm wide. Conidiogenous cells integrated, polyblastic, sympodially proliferating, with 1–4 conspicuously thickened conidial scars which are 2–3 µm in diam. Conidia solitary, limoniform, 3-celled, 37.5–51.3 × 17.5–20 µm, unequally coloured, the terminal cell short, triangular, thin-walled, subhyaline, verrucose, 5–7.5 × 5–7.5 µm, the central cell 27.5–34 × 17.5–20 µm, black, thick-walled, smooth, the basal cell 5–7.5 × 5–7.5 µm, trapezoid, thin-walled, subhyaline, verrucose, base obconically truncate, with a hilum 2–3 µm in diam.

Colonies on CMA reaching a diam of 10–15 mm in

28 d at 20°C under cool white fluorescent light, effuse, with immersed and superficial mycelium; hyphae up to 4 µm wide, subhyaline to pale brown. Conidia produced in the centre of the colony, mainly 3-celled, occasionally 2-celled.

Specimen examined: Thailand, Khao Yai National Park, Tad Ta Phu, on submerged wood (*Anisoptera oblonga*, test block), 3 Feb. 1997, S. Sivichai, BIOTEC SS285.

Known distribution: Canada, France, Japan, New Zealand, Thailand, Uganda, United Kingdom.

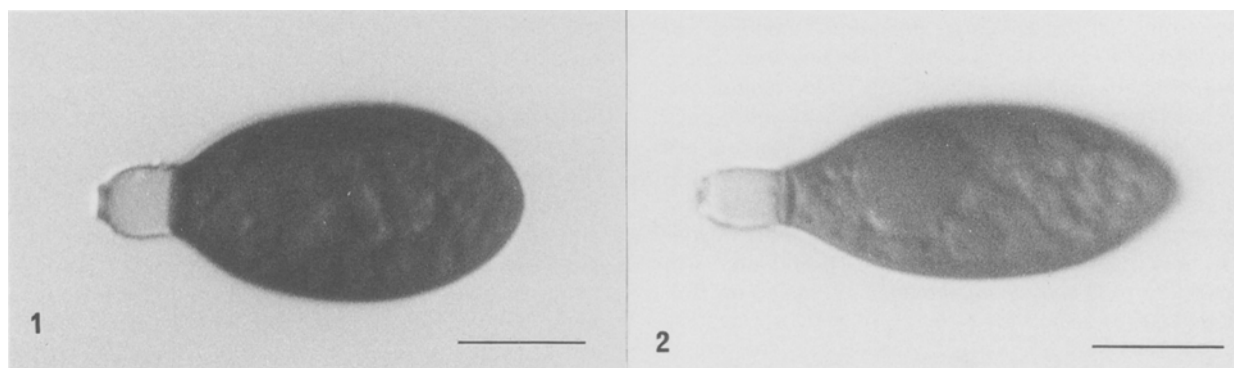
Notes: *Brachydesmiella biseptata* has been previously reported from North America and several European countries, and it appeared that this species has a frequent occurrence in the temperate zones. The present collection of *B. biseptata* in Thailand may be an indication of its occurrence in the tropics.

*Brachydesmiella caudata* V. G. Rao & de Hoog, Stud. Mycol. **28**: 5. 1986. Figs. 7–10

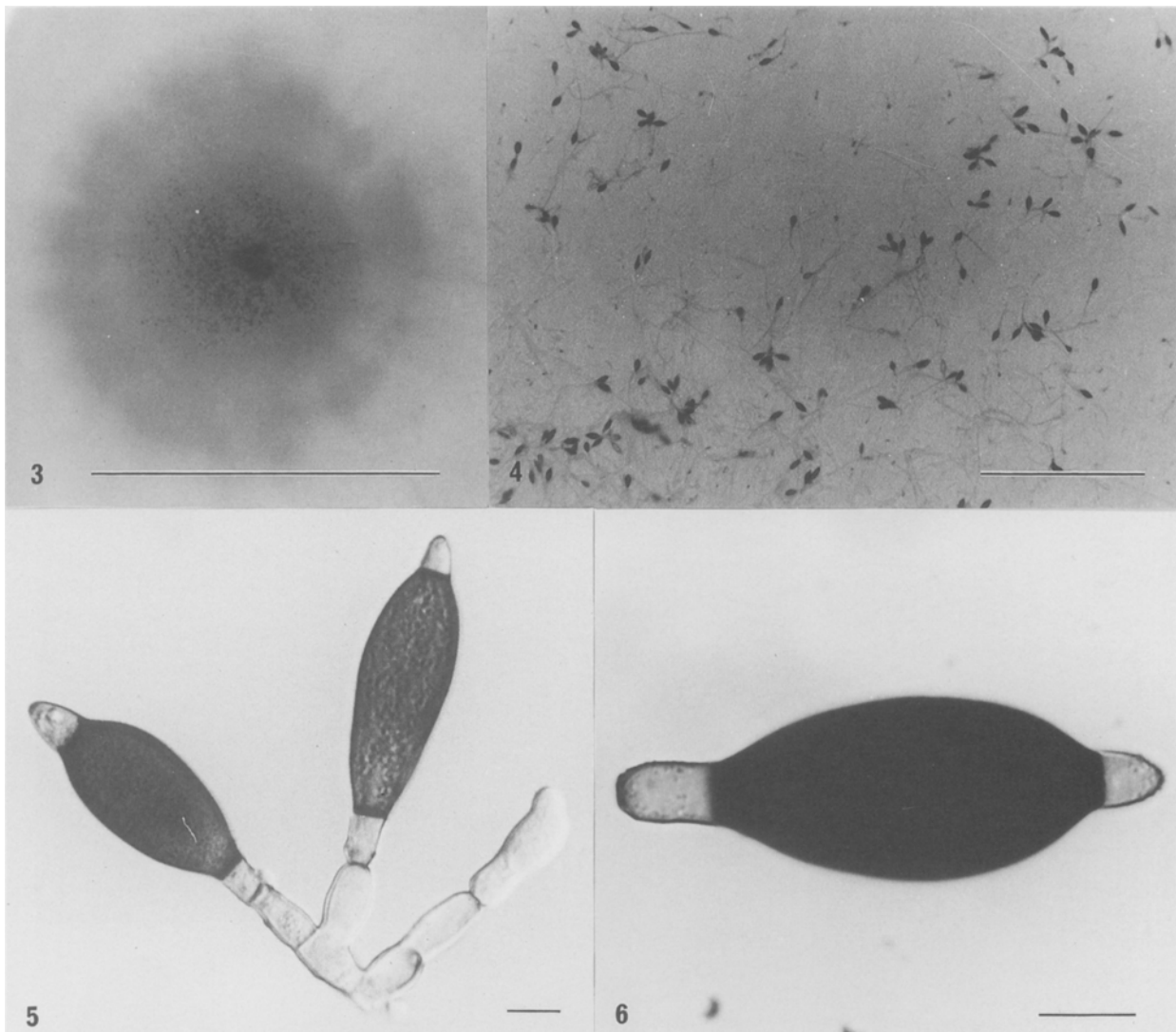
Colonies on natural substrate effuse, farinose, black, shiny. Conidiophores solitary or in small groups, cylindrical-clavate, flexuous, unbranched, stout, thick-walled, smooth, 0–3-septate, pale-brown, 28–46 × 4.5–8 µm. Conidiogenous cells integrated, polyblastic, sympodially proliferating, with 1–4 conspicuously thickened conidial scars which are 2–3 µm in diam. Conidia solitary, limoniform, 3-celled, 52.5–72.5 × 13–33 µm, unequally coloured, the terminal cell subcylindrical or acicular, subacute at the apex, thin-walled, subhyaline, verrucose in the middle part, 17–25 × 2–3.5 µm, the central cell 10–13 × 7–8 µm, dark reddish brown, ellipsoidal or truncate-limoniform, thick-walled, smooth, the basal cell 4–5 × 2–2.5 µm, cylindrical, thin-walled, subhyaline, base obconically truncate, with a hilum 2–3 µm in diam.

Colonies on CMA reaching a diam of 10–15 mm in 28 d at 20°C under cool white fluorescent light, effuse; mycelium immersed and superficial; hyphae up to 4 mm wide, hyaline to subhyaline; conidia mainly 3-celled, occasionally 2-celled, with the terminal cells relatively shorter than those from natural substrata.

Specimens examined: Hong Kong, New Territories, Tai Po, Tai Po Kau Forest Stream, on unidentified



Figs. 1, 2. Conidia of *Brachydesmiella anthostomelloidea* from PDA culture. Scale bar = 10 µm.



Figs. 3–6. *Brachydesmiella biseptata*, growth and sporulation on CMA.

3. Colony with sporulation. 4. Higher magnification of a sporulating portion. 5. Conidiophores and developing conidia. 6. Conidium. Scale bars: 3=1 cm; 4=100  $\mu\text{m}$ ; 5, 6=10  $\mu\text{m}$ .

submerged wood, 21 Sep. 1996, W. H. Ho and K. M. Tsui, HKU(M) 4599; *ibid.*, on submerged wood (*Machilus velutina*, test block), HKU(M) 4854; *ibid.*, HKU(M) 4857. Thailand, Khao Yai National Park, Km 29.2, on unidentified hardwood submerged in a small stream, 10 Jan. 1997, S. Sivichai, BIOTEC SS128.

Known distribution: Hong Kong, India, Thailand.

*Brachydesmiella orientalis* (V. G. Rao & de Hoog) Goh, *comb. nov.* Figs. 11–15

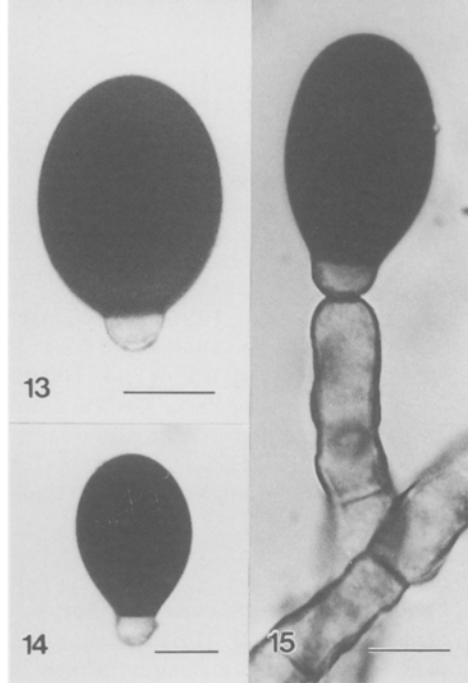
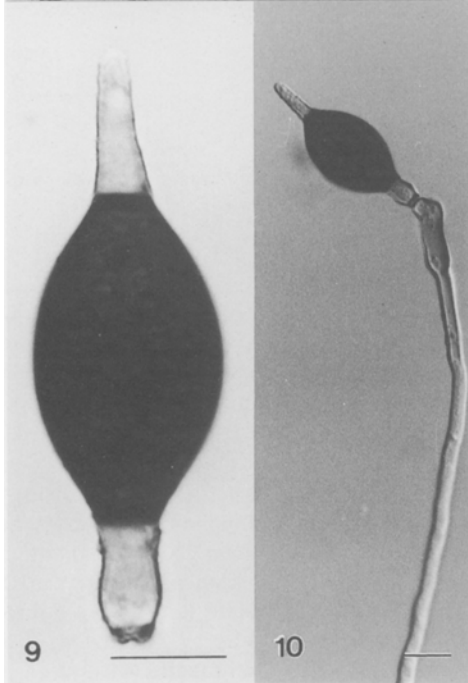
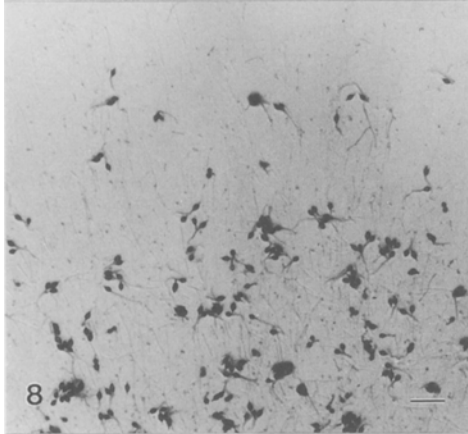
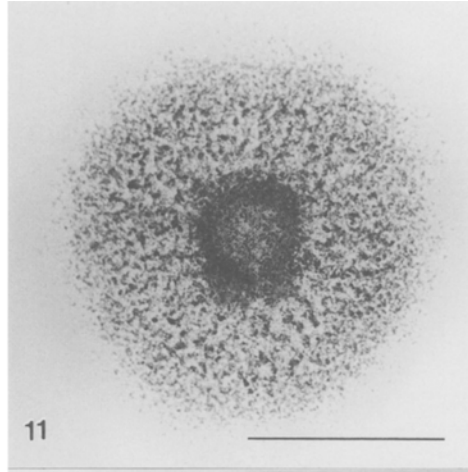
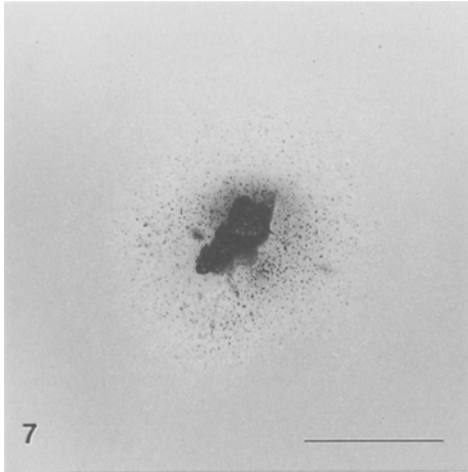
$\equiv$  *Brachydesmiella biseptata* var. *orientalis* V. G. Rao & de Hoog, *Stud. Mycol.* **28**: 3. 1986.

Colonies on natural substratum effuse, farinose, black, shiny. Conidiophores solitary or in small groups, flexuous, unbranched, smooth, thick-walled, subhyaline to pale brown, with 1–3 septa, 30–60  $\mu\text{m}$  long, 5–8  $\mu\text{m}$

wide, base 4–6  $\mu\text{m}$  wide. Conidiogenous cells integrated, polyblastic, sympodially proliferating, with 1–6 conspicuously thickened conidial scars which are 2–3  $\mu\text{m}$  in diam. Conidia solitary, pyriform, 2-celled, 30–37.5  $\times$  17.5–22.5  $\mu\text{m}$ , unequally coloured, the terminal cell 25–32.5  $\times$  17.5–22.5  $\mu\text{m}$ , blackish brown, smooth, thick-walled, the basal cell 4.5–5.5  $\times$  5–7.5  $\mu\text{m}$ , broadly trapezoid, thin-walled, subhyaline, verrucose, base obconically truncate, with a hilum 2–3  $\mu\text{m}$  in diam.

Colonies on CMA reaching a diam of 5–10 mm in 28 d at 20°C under cool white fluorescent light, effuse, no aerial mycelium; hyphae up to 4  $\mu\text{m}$  wide, subhyaline to pale-brown, partly immersed in agar. Conidia produced in the centre of the colony, predominantly 2-celled, occasionally 3-celled and limoniform.

Specimen examined: Thailand, Khao Yai National



Park, Tad Ta Phu, on submerged wood (*Anisoptera oblonga*, test block) 25 Mar. 1997, S. Sivichai, BIOTEC SS150.

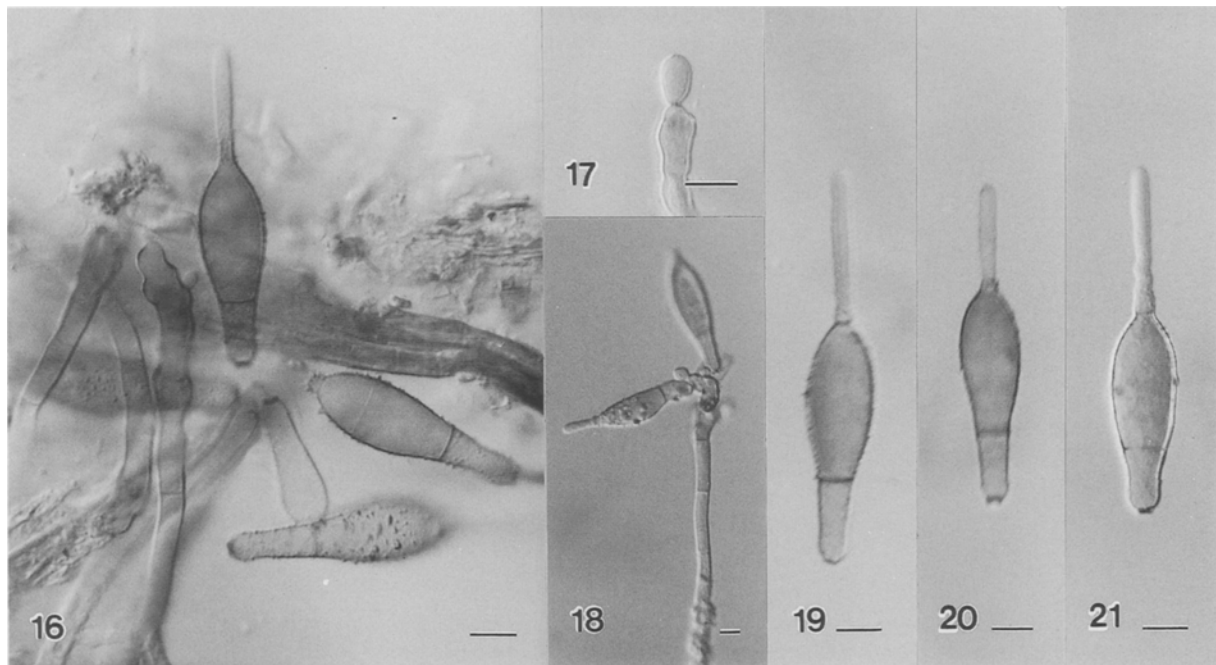
Known distribution: India, Thailand.

Notes: This is the first occasion where *B. biseptata* and *B. orientalis* were recorded in the same geographical region. Both species were collected from the same site in Thailand and found to occur on the same type of wood. *Brachydesmiella biseptata* was found on wood that had been immersed for 5 mo, while *B. orientalis* was found on wood that had been immersed for 6 mo. The simultaneous occurrence of these two species on wooden test blocks and their growth in pure culture has allowed us to compare these two taxa. *Brachydesmiella orientalis* differs from *B. biseptata* in producing conidia which are predominantly 2-celled on natural substratum and also in vitro, as compared to the mostly 3-celled conidia of *B. biseptata*, although both species may occasionally produce the other form of conidia (3-celled and 2-celled, respectively) in culture. Moreover, the conidia of *B. biseptata* are larger, with a broader (19–22  $\mu\text{m}$  wide), lighter (brown to dark brown) central cell, and a longer,

slender, obtriangular basal cell (4–5.5  $\times$  4–5  $\mu\text{m}$ ). The conidia of *B. orientalis* are comparable to those of *B. anthostomelloidea*, both being 2-celled. The latter species, however, differs in having longer and narrower conidia, with a short-cylindrical basal cell (Goh and Hyde, 1996).

***Brachydesmiella verrucosa*** Goh, Sivichai, K. D. Hyde & Hywel-Jones, sp. nov. Figs. 16–33

Coloniae in substrato naturali farinosae, brunneae. Mycelium partim superficiale et partim immersum, ex hyphis pallide brunneis compositum. Conidiophora solitaria vel numero paucio fasciculata, cylindro-clavata, ad basim angustata, flexuosa, usque 7-geniculata, crassitunicata, laevia vel leniter verruculosa ad basim, 0–4-septata, ad septa non constricta, pallide brunnea, ad basim hyalina vel subhyalina, 36–76  $\mu\text{m}$  longa, ad apicem 6–8  $\mu\text{m}$  lata, ad basim 5–7  $\mu\text{m}$  lata. Cellulae conidiogonae in conidiophoris integratae, terminales, polyblasticae, sympodiales, cylindricae; cicatrices conidiales 2–3  $\mu\text{m}$  diam. Conidia solitaria, acropleurogena, ampulliformia, tricellularia, 56–92  $\times$  12–17  $\mu\text{m}$ , versicoloria, illa



Figs. 16–21. *Brachydesmiella verrucosa*, on submerged wood (from holotype, HKU(M) 5855).

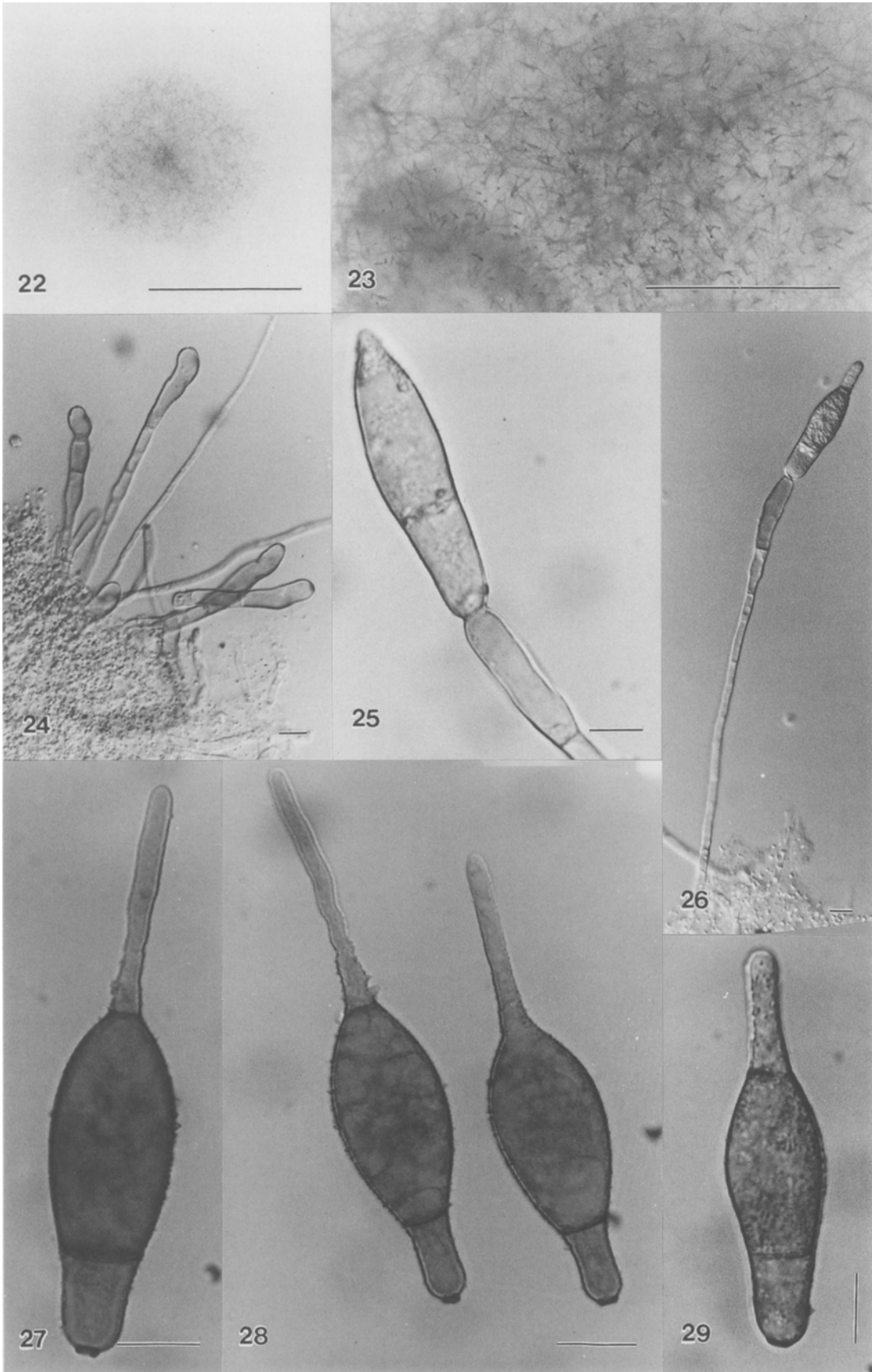
16. Squash mount in lactophenol illustrating conidiophores, young and mature conidia. Note verrucose wall ornamentation of the conidia. 17. Apex of a conidiophore bearing an incipient conidium. 18. Conidiophore bearing developing conidia at the apex. 19–21. Mature conidia. Scale bar = 10  $\mu\text{m}$ .

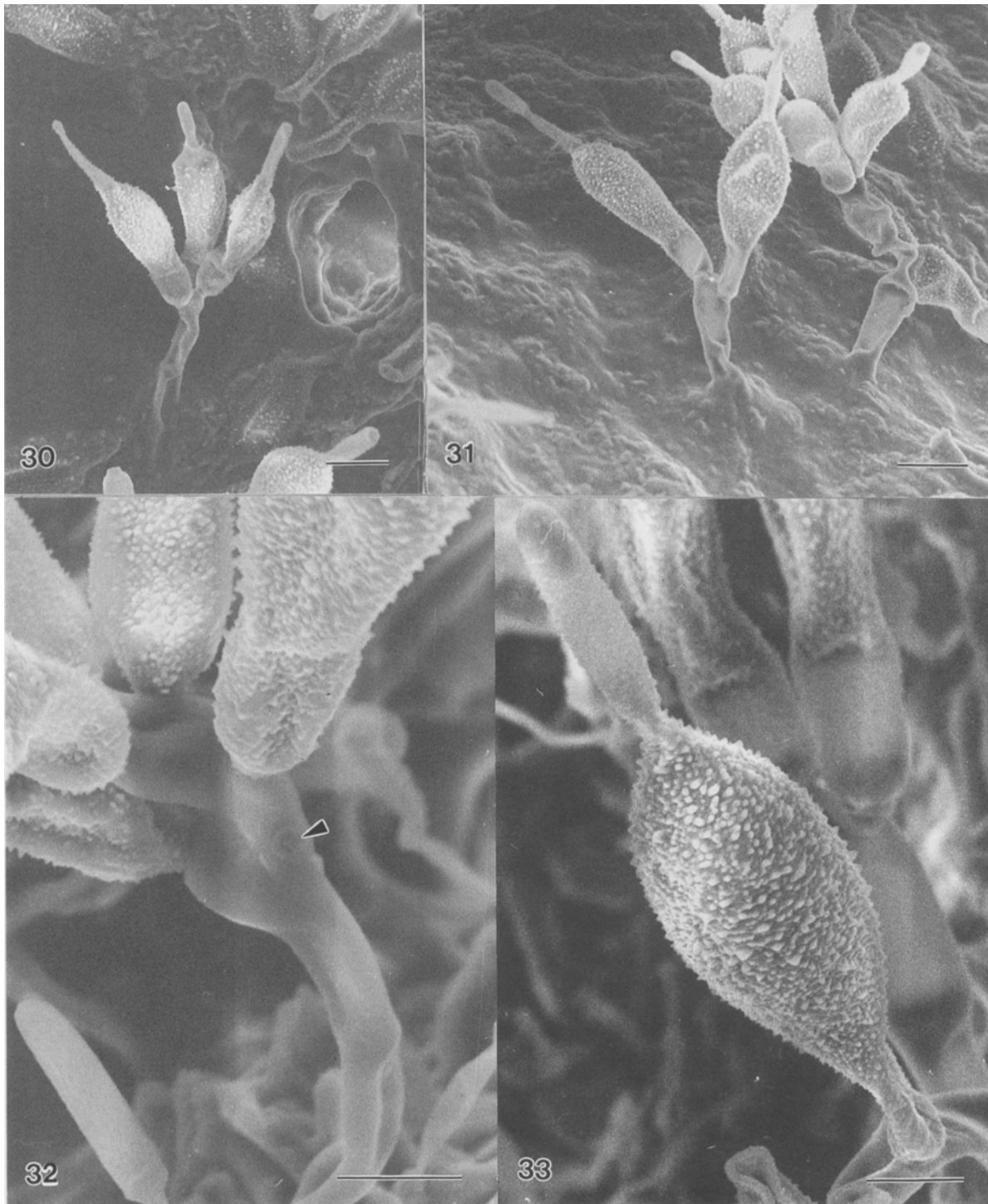
Figs. 7–10. *Brachydesmiella caudata*, growth and sporulation on CMA.

7. Colony with sporulation. 8. Higher magnification of a sporulating portion. 9. A mature conidium. 10. Conidiophore bearing a conidium at the apex. Scale bars: 7 = 1 cm; 8 = 100  $\mu\text{m}$ ; 9, 10 = 10  $\mu\text{m}$ .

Figs. 11–15. *Brachydesmiella orientalis*, growth and sporulation on CMA.

11. Colony with sporulation. 12. Higher magnification of a sporulating portion. 13, 14. Conidia. 15. Conidiophore bearing a conidium at the apex. Scale bars: 11 = 1 cm; 12 = 100  $\mu\text{m}$ ; 13–15 = 10  $\mu\text{m}$ .





Figs. 30–33. *Brachydesmiella verrucosa*, cryo-SEM micrographs of material on submerged wood (from holotype, HKU(M) 5855). 30, 31. Conidiophores bearing conidia. 32. Close-up of a conidiophore with sympodially produced conidia. A conidial scar on the conidiophore is arrowed. 33. A conidium. Note verrucose wall ornamentation. Scale bars: 30, 31 = 10  $\mu\text{m}$ ; 32, 33 = 5  $\mu\text{m}$ .

Figs. 22–29. *Brachydesmiella verrucosa*, growth and sporulation on CMA. 22. Colony with sporulation. 23. Higher magnification of a sporulating portion. 24. Conidiophores. Note clavate shape, septation, and conidial scars. 25. Apex of a conidiophore bearing a developing conidium. 26. Conidiophore bearing a young conidium at the apex. 27–29. Conidia. Note verrucose wall ornamentation. Scale bars: 22 = 1 cm; 23 = 100  $\mu\text{m}$ ; 24, 26 = 20  $\mu\text{m}$ ; 25, 27–29 = 5  $\mu\text{m}$ .

terminalia longe cylindrica, apice rotundata, verruculosa, tenuitunicata, subhyalina vel pallide brunnea,  $22\text{--}46 \times 4\text{--}6 \mu\text{m}$ , illa centralia late ellipsoidea,  $22\text{--}34 \times 12\text{--}17 \mu\text{m}$ , verrucosa, pallide olivaceobrunnea, crassitunicata, illa basalia parviora,  $8\text{--}16 \times 6\text{--}8 \mu\text{m}$ , cylindrica, crassitunicata, hyalina vel subhyalina, leniter verruculosa vel laevia, ad basim obconico-truncata, hilo  $2\text{--}3 \mu\text{m}$  lato conspicue incrassato praedita.

Holotypus: Malaysia, Johore, Keluang, Gunung Lambak, on submerged wood, 15 Feb. 1997, T. K. Goh and C. S. Lim, HKU(M) 5855.

Etym.: *verrucosa* – referring to the coarse ornamentation of the conidial walls.

Colonies on natural substratum farinose, brown. Mycelium partly superficial, partly immersed, consisting of light brown hyphae. Stroma none. Setae and hyphopodia absent. Conidiophores macronematous, solitary or in small groups, scattered, cylindro-clavate, narrower at the basal part, flexuous, up to 7-geniculate, stout, thick-walled, smooth or slightly verruculose at the basal portion, 0–4-septate, not constricted at the septa, pale brown, hyaline to subhyaline at the base,  $36\text{--}76 \mu\text{m}$  long, apex  $6\text{--}8 \mu\text{m}$  wide, base  $5\text{--}7 \mu\text{m}$  wide. Conidigenous cells integrated, terminal, polyblastic, sympodial, cylindrical, cicatrized with conidial scars  $2\text{--}3 \mu\text{m}$  in diam. Conidia solitary, acropleurogenous, ampulliform, 3-celled,  $56\text{--}92 \times 12\text{--}17 \mu\text{m}$ , unequally coloured; the terminal cell long cylindrical, ends rounded, verruculose, thin-walled, subhyaline to pale brown,  $22\text{--}46 \times 4\text{--}6 \mu\text{m}$ ; the central cell broadly ellipsoidal, coarsely verrucose,  $22\text{--}34 \times 12\text{--}17 \mu\text{m}$ , pale olivaceous brown, darker than the apical and basal cells, thick-walled; basal cell  $8\text{--}16 \times 6\text{--}8 \mu\text{m}$ , cylindrical, thin-walled, hyaline to subhyaline, slightly verruculose or otherwise smooth, base obconically truncate, with a conspicuously thickened hilum of  $2\text{--}3 \mu\text{m}$  in diam.

Conidia germinating on CMA within 24–36 h in cabinet under cool white fluorescent light at  $20^\circ\text{C}$ , producing a germ tube from either end, or from the apical end only.

Colonies on CMA reaching a diam of 10–15 mm in 14 d at  $20^\circ\text{C}$  under cool white fluorescent light, effuse in the middle zone, with aerial mycelium which is up to  $5 \mu\text{m}$  wide and subhyaline to pale brown, darker in the centre where mycelium immersed into the agar. Conidia forming in the centre only after 8–10 d.

Other specimens examined: Australia, North Queensland, Atherton Tablelands, near Ravenshoe, ca 20 km along Tully Falls Rd. to Koombooboo Dam, Creek, 1745 S, 14535 E, on submerged wood, K. D. Hyde and C. Pearce, HKU(M) 5128; *ibid.*, HKU(M) 5135; Thailand, Nakorn Ratchassima, Khao Yai National Park, stream at Tad Ta Phu, on submerged wood (*Anisoptera oblonga*, test block), 10 April 1997, S. Sivichai, BIOTEC SS341.

Known distribution: Australia, Malaysia, Thailand.

Notes: In culture the conidia of *B. verrucosa* are most similar to those of *B. caudata* (Rao and de Hoog, 1986). In both species conidia are 3-celled, with a long cylindrical terminal cell, a large central cell, and a shorter, more-or-less cylindrical basal cell. In *B. caudata*, however, the conidia lack the distinct verrucose ornamentation found in *B. verrucosa*. Conidia of *B. caudata* also differ from those of *B. verrucosa* in that the central cell is smooth and darker, and the apical cell is shorter ( $17\text{--}25 \mu\text{m}$  long), narrower ( $2\text{--}4 \mu\text{m}$  wide), and verruculose only at the middle portion. Other differences between these two species are compared in Table 1. In Thailand, *B. caudata* was discovered from an unidentified hardwood which had fallen naturally into a seasonally flowing stream. It was sought in an area where test blocks had been placed but was not found on any of these. In contrast, *B. verrucosa* was found in a stream that flows all through the year and was discovered from test blocks of a hardwood which had been immersed in the river for 7 mo. *Brachydesmiella verrucosa* is known from Australasia and SE Asia, whereas *B. caudata* is known from S and SE Asia. Thailand is an area of overlap for the known ranges of these two species.

Table 1. Comparison of *Brachydesmiella caudata* and *B. verrucosa*.

	<i>B. caudata</i>	<i>B. verrucosa</i>
Conidiophores	$28\text{--}46 \times 4.5\text{--}8 \mu\text{m}$	$37\text{--}76 \times 6\text{--}8 \mu\text{m}$
Conidia		
Shape	limoniform	ampulliform
Overall size	$52.5\text{--}72 \times 13\text{--}33 \mu\text{m}$	$56\text{--}92 \times 12\text{--}17 \mu\text{m}$
Number of cells	3	3
Terminal cell	$17\text{--}25 \times 2\text{--}3.5 \mu\text{m}$ , acicular, with subacute end, verruculose at the middle portion	$22\text{--}46 \times 4\text{--}6 \mu\text{m}$ , long-cylindrical, with bluntly rounded end, verruculose throughout
Central cell	$25\text{--}35 \times 15\text{--}22 \mu\text{m}$ , dark reddish brown, ellipsoidal or truncate-limoniform, smooth	$22\text{--}34 \times 12\text{--}17 \mu\text{m}$ , pale olivaceous brown, broadly ellipsoidal, coarsely verrucose
Basal cell	$10\text{--}19 \times 3\text{--}8 \mu\text{m}$ , cylindrical, verruculose	$8\text{--}16 \times 6\text{--}8 \mu\text{m}$ , cylindrical, verruculose
Distribution	Hong Kong, India, Thailand	Australia, Malaysia, Thailand



Key to *Brachydesmiella* speices

1. Conidia exclusively 2-celled, pyriform ..... 2
1. Conidia exclusively or predominantly 3-celled, limoniform or ampulliform ..... 3
  2. Conidia 35–47 × 14–18 μm; basal cell 6–9 μm long, cylindrical ..... *B. anthostomelloidea*
  2. Conidia 30–35 × 17–20 μm; basal cell 3–4 μm long, broadly trapezoid ..... *B. orientalis*
3. Conidia 50–92 μm long; apical cell acicular or elongated cylindrical; basal cell cylindrical to subcylindrical ..... 4
3. Conidia 37–51 × 17–22 μm; apical and basal cells triangular or trapezoid ..... *B. biseptata*
  4. Conidia 52–72 × 13–33 μm, smooth or verruculose, more or less limoniform; apical cell 19–23 × 2–3.5 μm, subcylindrical or acicular; central cell dark reddish brown ..... *B. caudata*
  4. Conidia 56–92 × 12–17 μm, coarsely verrucose, ampulliform; apical cell 22–46 × 4–6 μm, elongate-cylindrical; central cell pale olivaceous brown ..... *B. verrucosa*

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