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Annellosympodia orbiculata gen. et sp. nov. and *Scolecostigmina flagellariae* sp. nov. from Australia

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Abstract. Two new fungi, *Annellosympodia orbiculata* and *Scolecostigmina flagellariae*, on *Acacia ligulata* and *Flagellaria indica*, respectively, from Australia are described and illustrated. The former species is placed in the new genus *Annellosympodia*, which is characterised by an unusual combination of features, *viz*. fasciculate conidiogenous cells (conidiophores reduced to conidiogenous cells), holoblastic conidiogenesis with sympodial, but rectilinear proliferation leaving annular structures and lateral conspicuous conidiogenous loci, and rhexolytic conidial secession. The generic placement of these two fungi is discussed.

Introduction

Ellis (1959, 1971, 1976) assigned numerous dematiaceous hyphomycetes with sporodochial conidiomata and percurrent, annellate conidiogenous cells to *Stigmina* Sacc., which widened the generic concept and circumscription of this genus considerably. Sutton and Pascoe (1989) reduced the heterogeneity of *Stigmina* by confining it to foliicolous species with sporodochia associated with stomata and holoblastic conidia formed on percurrently proliferating, verruculose, ragged conidiogenous cells. Further narrowing of the generic limits of *Stigmina* by Sutton and Crous (1997), Crous (1998) and Braun *et al*. (1999) resulted in the segregation of four other *Stigmina*-like genera, namely *Cercostigmina* U. Braun, *Lecanostictopsis* B. Sutton & Crous, *Scolecostigmina* U. Braun and *Xenostigmina* Crous. Braun *et al*. (1999) provided a key that demonstrated the differences between these four genera as well as *Stigmina s. str*. and *Thyrostroma* Höhn. Later, *Cercostigmina* was shown to be an anamorphic genus of *Mycosphaerella* indistinguishable from *Pseudocercospora* (Crous *et al*. 2001). The formation of percurrent conidiogenous cells is not tenable within *Mycosphaerella* anamorphs to distinguish anamorphic genera (Crous *et al*. 2006).

Using sequence analyses of the small ribosomal subunit (SSU) region of the nrDNA, Crous *et al*. (2006) demonstrated that *Pseudocercospora vitis* (Lév.) Speg., the type species of *Pseudocercospora*, *Phaeoisariopsis griseola* (Sacc.) Ferraris, the type species of *Phaeoisariopsis*, and *Stigmina platani* (Fuckel) Sacc., the type species of *Stigmina*, cluster together in a mixed clade with other species of *Pseudocercospora*. The three genera are proven *Mycosphaerella* anamorphs with a uniform scar type (conidiogenous loci truncate, unthickened). The shape and septation of the conidia has undoubtedly no value at generic rank in *Mycosphaerella* anamorphs. Thus, it was inevitable that *Stigmina*, *Phaeoisariopsis* and *Pseudocercospora* should merge. Although *Stigmina* is the earliest legitimate name, Braun and Crous (2006) proposed to conserve *Pseudocercospora* against *Stigmina* and *Phaeoisariopsis*. For most *Stigmina*-like species, excluding the type species *S. platani*, it is unproven whether they represent *Mycosphaerella* anamorphs because there are no cultures available and molecular sequence analyses have not been conducted. At present, the traditional application of the generic names concerned has to be maintained.

A foliar pathogen on *Acacia ligulata* and *Acacia* sp. from arid regions in central and north-western Australia, was first thought to be close to *Stigmina s. str*., due to young conidiogenous cells with single terminal conidiogenous loci, but older developmental stages showed proliferating conidiogenous cells, elongated up to 50 µm, with distinct annular structures and conspicuous lateral scars. This unusual mode of conidiogenesis and proliferation is quite distinct from *Stigmina* and allied genera, but resembles that of *Annellophragmia* Subran. (Ellis 1971) and *Spiropes dictyosporus* Seifert & S. Hughes (Seifert and Hughes 2000). A second species of *Stigmina*-like fungus on the leaves of *Flagellaria indica* (Flagellariaceae), which is native to Australian rainforests and extends from Africa to Polynesia, was identified as belonging to *Scolecostigmina*, using the key of Braun *et al*. (1999).

Methods

For light microscopy the specimens were mounted in lactoglycerol, gently heated and examined with a compound microscope (Leica DMLB, Hamburg, Germany). Specimens are deposited in the herbaria BRIP, HAL and PERTH (Holmgren *et al*. 1990).

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Taxonomy

Annellosympodia orbiculata McTaggart, R.G. Shivas & U. Braun, gen. et sp. nov. (Figs 1 and 2)
MycoBank # MB510980 (genus) et MycoBank # MB510981 (species).

Holotypus in matrice *Acacia* sp., south side of Walcott Inlet, Western Australia, Australia, on 24 Mar. 1994, A.A. Mitchell; holo: PERTH 03270173; iso: BRIP 49805, HAL 2031 F; para: in matrice *Acacia ligulata* A. Cunn. ex Benth., Corroboree Rock, East MacDonald Ranges, Northern Territory, Australia, 28 Aug. 1991, D.R. Spowart (BRIP 19908); isopara: HAL 2050 F.

Descriptio generico-specifica: *Fungi imperfecti*, *Hyphomycetes. Coloniae* gregariae in areis decoloratis in phyllodiis. *Conidiomata* punctiformia, amphigena, fusca, pulverulenta, diametro 40–60 µm, coniuncta super stomata. *Mycelium* immersum. *Stromata* nulla. *Conidiophora* nulla. *Cellulae conidiogenae* ampulliformes, doliiformes vel obovoideae, fuscae, aseptatae, verrucosae, parietibus incrassatis, 15–20 µm longae, 9–13 µm latae in parte latissima, dense fasciculatae, fere 12–27 in sporodochiis minutis, pulvinatis. Conidiogenesis holoblastica, primo monoblastica, loco apiculi singulari cum annulo unico, denso, circulari circa porum centralem, deinde polyblastica, cum cellulis conidiogenis rectis, successive sympodialiter sed rectilineare proliferantibus, ad 50 µm longis, cicatricibus conspicuis, primo apicalibus, deinde lateralibus, applanatis, non-protuberantibus, leviter incrassatis et fuscatis, ~2 µm latis. *Conidia* solitaria, brunnea, grosse verrucosa, cylindracea ad ellipsoidea, 18–28 × 9–12 (–14) µm, aseptata vel equatoriale 1-septata, interdum constricta ad septum, apex rotundatus, basis truncata cum segmentis marginalibus et hilo atro, conspicuo; secessio rhexolytice.

Fig. 1. *Annellosympodia orbiculata* on phyllodes of *Acacia* sp. (BRIP 49805).

Etymology: referring to the sympodial, but rectilinear proliferation of the conidiogenous cells, leaving annular structures and conspicuous lateral scars (genus name); from the Latin word *orbiculatus* meaning circular referring to the annellation on the conidiogenous cells (epithet).

Fungi imperfecti, *Hyphomycetes. Colonies* gregarious on discoloured areas on phyllodes. *Conidiomata* punctiform, amphigenous, dark brown, pulverulent, 40–60 µm diam., centred over stomata. *Mycelium* immersed. *Stromata* absent. *Conidiophores* absent. *Conidiogenous cells* ampulliform, doliiform or obovoid, macronematous, dark brown, aseptate, verrucose, thick-walled, 15–20 µm long, 9–13 µm wide at widest part, in groups of ~12–27 on minute pulvinate sporodochia. Conidiogenesis holoblastic, at first monoblastic, occurring apically to form a single conidiogenous locus with a thick, ring-like annellation around a central pore, later polyblastic, conidiogenous cells with successive sympodial but rectilinear proliferation, up to 50 µm long, rupturing the outer wall layer around each scar, resulting in a lateral displacement of scars, leaving conspicuous circumferential annular fringes of the torn wall, lateral scars in face view conspicuous, flat, non-protuberant, slightly thickened and darkened, orbiculate, ~2 µm diam. *Conidia* solitary, brown, coarsely verrucose, cylindrical to ellipsoidal, 18–28 × 9–12(–14) µm, apex rounded, base truncate with a marginal frill and a dark conspicuous hilum, 0–1-septate with an equatorial septum, sometimes constricted at the septum; secession rhexolytic.

Conidiogenous loci of early monoblastic developmental stages of *A. orbiculata* are reminiscent of loci in *Stigmina* (*s. str*.), but the new species differs from all *Stigmina*-like species described on *Acacia* (Bessey 1953; Sutton and Pascoe 1989). *Stigmina inconspicua* B. Sutton & Pascoe has stromata composed of pseudoparenchyma and mycelium, discrete conidiophores and conidia that are 1–3-septate and smaller than *A. orbiculata. Camptomeris verruculosa* (Syd.) E.A. Bessey (≡ *Stigmina verruculosa* Syd.), unlike *A. orbiculata*, has megalophyses, which protrude through the stomata and give rise to the conidiophores.

The conidiogenous cells of *A. orbiculata* may proliferate and elongate up to ~50 µm. The mode of proliferation, the conidiogenesis and structure of the conidiogenous loci are rather unusual and quite distinct from *Stigmina* and allied genera. The unusual combination of annular structures and conspicuous lateral conidiogenous loci is similar to *Annellophragmia* (Ellis 1971) and *S. dictyosporus* (Seifert and Hughes 2000). But *Annellophragmia* differs from *Annellosympodia* in having long, filiform conidiophores arranged in synnemata and distoseptate

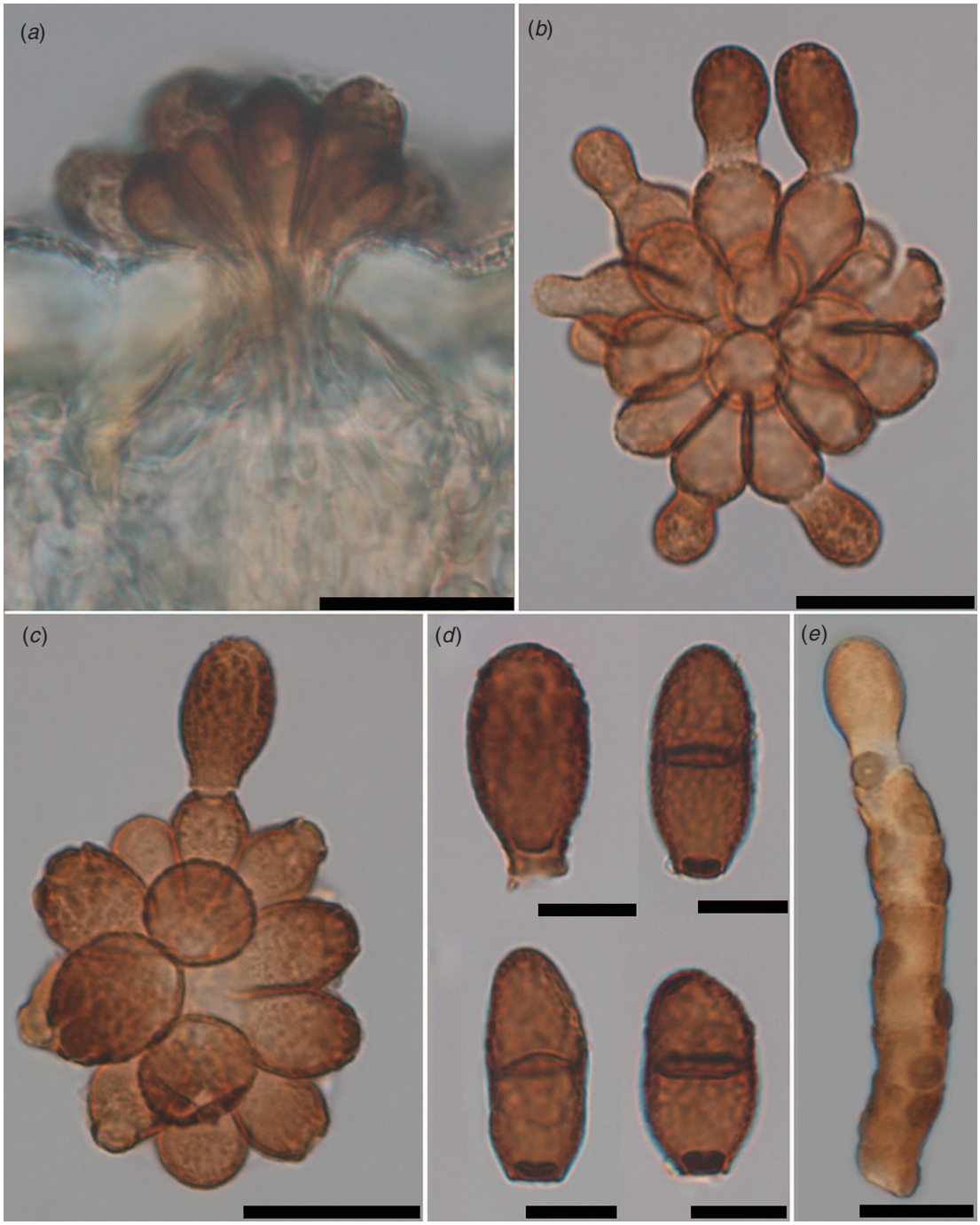

Fig. 2. *Annellosympodia orbiculata* (BRIP 19908): (*a*) sporodochia arising through stomata; (*b*, *c*) conidiogenous cells with annellations showing conidiogenesis; (*d*) conidia; (*e*) mature conidiogenous cell with visible scars and annellations (BRIP 49805). Bars = 20 µm (*a*–*c*), 10 µm (*d*, *e*).

phragmosporous conidia. *S. dictyosporus* is also synnematous and has muriform conidia. The conidiogenesis and proliferation of the conidiogenous cells, described and illustrated in detail by Seifert and Hughes (2000), is close to *Annellosympodia*. It is questionable if *S. dictyosporus* is congeneric with *Spiropes*. *Spiropes* has been confined to hyphomycetes hyperparasitic on sooty moulds with a wide range of conidiophore types, conidiogenous cells and conidia. The type species has distinctly sympodial-geniculate (zigzag) conidiogenous cells and distoseptate conidia. Some other species referred to as *Spiropes*, e.g. *S. dialii* (Bat.) M.B. Ellis and *S. shoreae* M.B. Ellis (Ellis 1971), show similar conidiogenesis and proliferation as in *S. dictyosporus*. Similar structures are also evident in some species of *Pseudospiropes* M.B. Ellis, e.g. *P. simplex*

(Kunze) M.B. Ellis (Ellis 1971). The type species of *Pseudospiropes* as well as *P. simplex* are characterised by having distoseptate (pluriseptate) conidia. Castañeda *et al*. (2001) confined *Pseudospiropes* to species with distoseptate conidia. Furthermore, they introduced *Minimelanolocus* R.F. Castañeda & Heredia for former *Pseudospiropes* species having sympodial and few enteroblastic regenerative percurrent proliferations (not connected with conidiogenesis), euseptate (pluriseptate) conidia, and inconspicuous or slightly prominent conidiogenous loci. *Nigrolenticulus* R.F. Castañeda & Heredia was proposed for similar taxa with euseptate conidia differing from *Minimelanolocus* by its enlarged, thickened, protuberant, strongly melanised, lenticular scars. The characters of *Pseudospiropes s. str*., *Minimelanolocus* and *Nigrolenticulus* do not allow an inclusion of the new species on *Acacia* in Australia.

Finally, *Matsushimiella* R.F. Castañeda & Heredia (Castañeda *et al*. 2001) was established to accommodate the saprobic *Pseudospiropes queenslandicus* Matsush. (Matsushima 1989). It is interesting and noteworthy that *Matsushimiella* is characterised by having rhexolytic conidial secession. This genus is easily distinguishable from *Annellosympodia* by having enlarged, protuberant conidiogenous loci and distoseptate conidia. Annellations are lacking.

Scolecostigmina flagellariae R.G. Shivas & U. Braun, sp. nov. (Figs 3–5)

MycoBank # MB510982.

Holotypus in matrice *Flagellaria indica* L., Mossman Gorge National Park, Queensland, Australia, 28 July 1992, R.G. Shivas; holo: BRIP 20539; iso: HAL 1622 F, PERTH 3801179; para: Cape Tribulation National Park, Queensland, on *Flagellaria indica*, 10 Aug. 1992, R.G. Shivas, BRIP 20531, PERTH 3203468; Mossman Gorge National Park, 27 July 1993, R.G. Shivas, BRIP 21842; Mossman Gorge National Park, 1 Aug. 1993, R.G. Shivas, BRIP 21843.

Fig. 3. *Scolecostigmina flagellariae* on leaves of *Flagellaria indica* (BRIP 20531).

Scolecostigminae fici-elasticae et *S. pilostigmatis* similis, sed conidiomatibus, conidiophoris et conidiis saepe rubro-brunneis et conidiis laevibus.

Etymology: after the host plant *Flagellaria*.

Lesions on living leaves, amphigenous, forming numerous speckles, scattered to dense, subcircular to angular-irregular, 0.5–2 mm wide, dark brown, colour mainly caused by the large fungal colonies, but host tissue also becoming brown, necrotic, often with a diffuse yellowish to ochraceous to light reddish brown halo, confluent lesions covering much of the leaf surface. *Mycelium* internal, at first aggregated in the substomatal cavities, composed of hyaline intercellular hyphae and dark brown or reddish brown extracellular hyphae, which collectively form a compact stromatic column up to 80 µm wide and often emerging from both leaf surfaces. *Conidiomata* amphigenous, punctiform to pustulate, sporodochial, dry, centred over stromatic column, pulvinate, appressed to the leaf surface, dark brown, dark reddish brown, up to 250 µm high and 500 µm wide. *Stromatic cells* subglobose to angular-irregular in outline, 2–10 µm diam. *Conidiophores* very numerous, dense, arising from the stromatic hyphal aggregations, macronematous, erect, subcylindrical, somewhat sinuous, stiff to flexuous, 15–70 × 3–6 µm, pale to medium brown or reddish brown, irregularly 0–2-septate, occasionally branched at the base, wall thin or somewhat thickened, smooth. *Conidiogenous cells* integrated, terminal, subcylindrical, 4–20 × 3–4 µm, producing holoblastic conidia each separated by an enteroblastic, percurrent proliferation to form 1–15 conspicuous, transverse, ragged annellations. *Conidia* solitary, cylindrical, subcylindrical-fusiform, subclavate, 40–85 × 3–8 µm, (0–)3(–4)-euseptate, without constrictions, wall somewhat thickened, (0.5–)0.75–1.25(–2) µm wide, smooth, pale to medium brown, reddish brown, apex obtuse, broadly rounded, gradually tapered to the base, which is usually somewhat bulging, convex, 3–4 µm wide, with a ragged marginal frill, secession schizolytic.

Braun (1999) transferred numerous *Stigmina* species to a new genus *Scolecostigmina* characterised by having pulvinate sporodochial conidiomata, conidiogenous cells with conspicuous, coarse annellations and scolecosporous, euseptate, more or less thick-walled conidia. *S. flagellariae* fits well into the concept of *Scolecostigmina* as it is foliicolous, biotrophic; its conidiomata are sporodochial, pulvinate; the conidiogenous cells have conspicuous annellations; the conidia are solitary, scolecosporous, with a slightly thickened wall. The annellations are coarse, ragged and the conidia are provided with conspicuous, ragged, marginal frills at the base, which are characteristic for *Scolecostigmina*. Teleomorphs of *Scolecostigmina* species and molecular data are not yet known, so that a possible affinity to the *Mycosphaerellaceae* and relations to *Pseudocercospora* (incl. *Cercostigmina*) remain unresolved.

Species of *Scolecostigmina* appear to be host specific. *S. flagellariae* is the first species of any *Stigmina*-like fungus on a host in the Flagellariaceae. *S. flagellariae* differs from all other species of *Scolecostigmina* in having conidiomata,

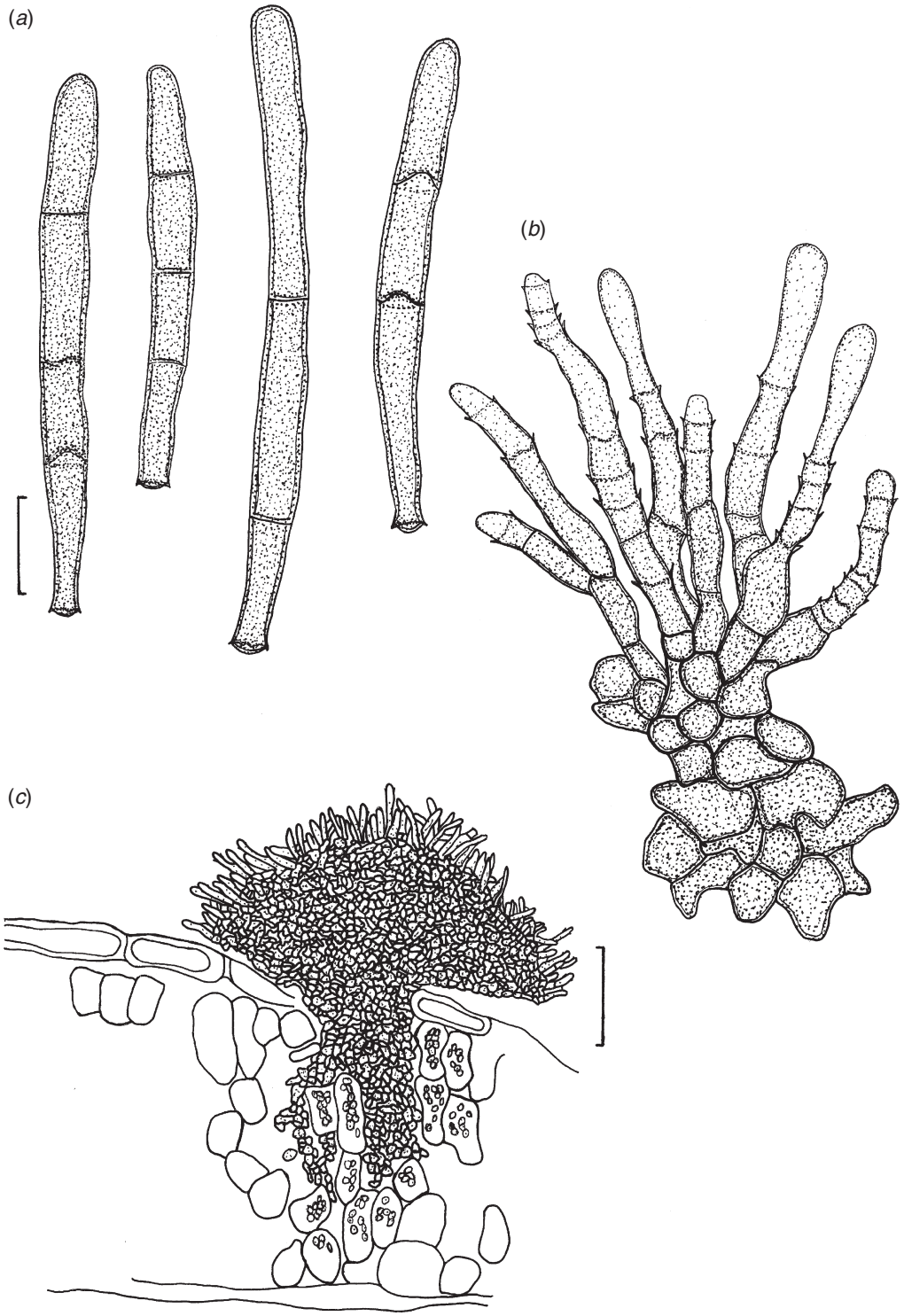

Fig. 4. *Scolecostigmina flagellariae*: (*a*) conidia; (*b*) conidiophores; (*c*) conidioma. Bars = 10 µm (*a*, *b*), 100 µm (*c*) (drawings by R.G. Shivas).

conidiophores and conidia with a distinctive reddish tinge and smooth conidia, which are almost consistently 3-septate. All other species of *Scolecostigmina* are either pluriseptate, or if they have relatively few septa, then the conidia are verruculose, examples are *Scolecostigmina fici-elasticae* (Kapoor) U. Braun and *S. pilostigmatis* (M.B. Ellis) U. Braun.

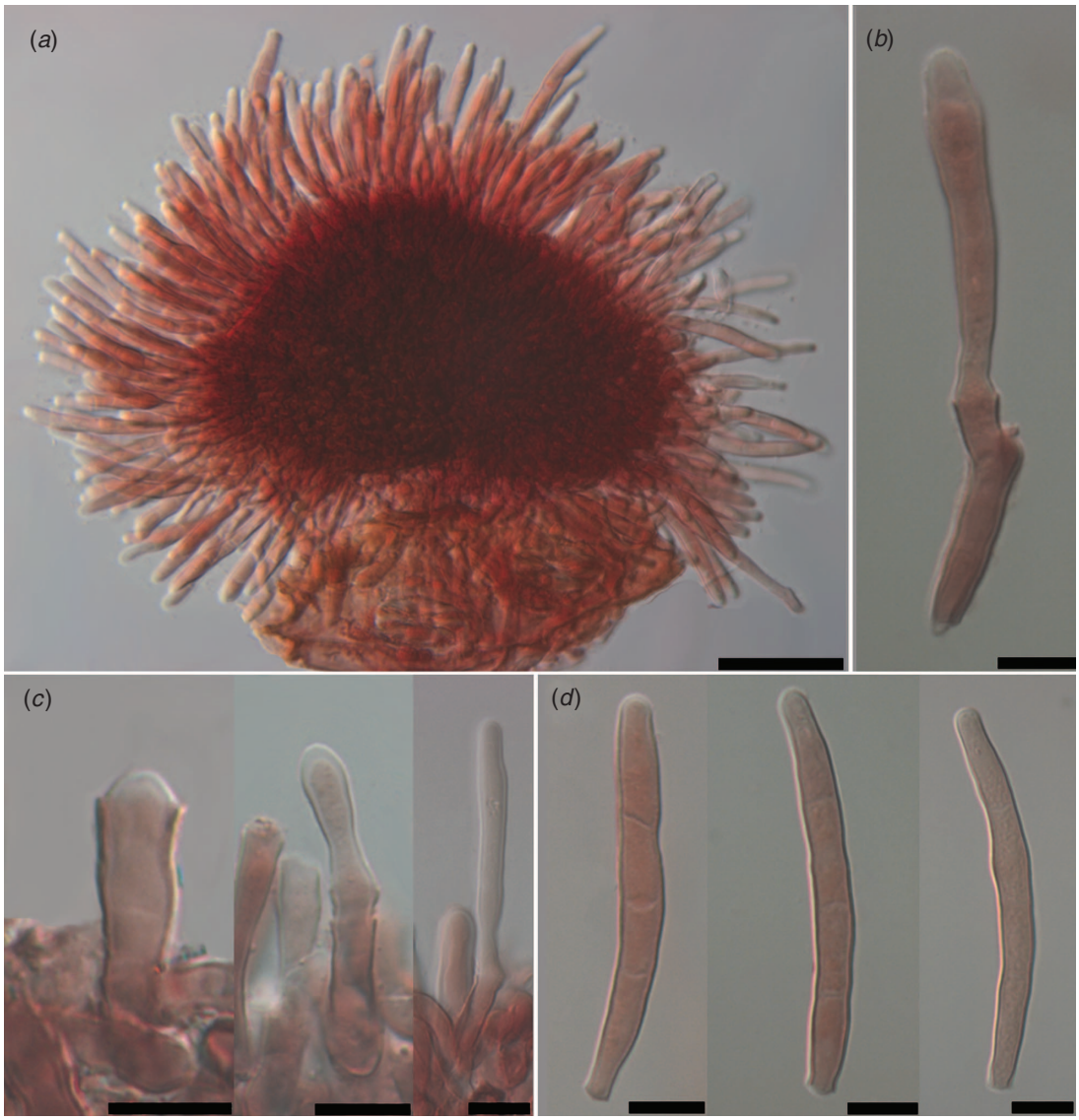

Fig. 5. *Scolecostigmina flagellariae* (BRIP 21843): (*a*) conidioma; (*b*) conidiogenesis; (*b*, *c*) annellations on conidiophores; (*d*) conidia. Bars = 50 µm (*a*), 10 µm (*b–d*).

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