New species of *Anthostomella* on fynbos, with a key to the genus in South Africa

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A study of saprobic fungi occurring on the fynbos of the Western Cape Province of South Africa yielded four unknown *Anthostomella* species. *A. proteae* from *Protea nitida*, *A. cynaroides* from *P. cynaroides*, *A. leucospermi* from *Leucospermum oleifolium*, and *A. brabeji* from *Brabejum stellatifolium* are described as new. New records for South Africa include *A. conorum* from *Leucadendron* sp., *Protea magnifica* and *P. neriifolia*, and *A. clypeata* from *Ischyrolepis subverticellata*, *Cannomois virgata*, *Restio egregius*, and *R. cfr confusus*. A dichotomous key to the *Anthostomella* species in South Africa is also provided.

INTRODUCTION

Species of Anthostomella occur on leaves and stems of numerous hosts worldwide and are generally accepted to be saprobes. More than 300 described species were monographed by Lu & Hyde (2000), who accepted 86 species; 95 species were reduced to synonymy, and 105 taxa were allocated to other genera. Eleven species were acknowledged from South Africa: A. vestita (syn. A. capensis; Doidge 1948) on Phoenix dactylifera, A. acuminata, A. applanata, A. caffrariae, A. leptospora, A. meerensis, A. palmae and A. spiralis on Phoenix reclinata, A. colligata and A. raphiae on Raphia australis, and A. umbrinella (syn. A. cassinopsidis; Rehm 1907) on Cassinopsis capensis (Lu, Hyde & Liew 2000). A. alpigena and A. xylosteri that were originally reported from South Africa by Martin (1969) were placed in Amphisphaerella, while A. melanotes (syn. Barrmaelia oxyacanthae; Rappaz 1995) was excluded from Anthostomella. Francis (1975) also reported A. rubicola on Rubus fruticosus from South Africa, but this material was not examined by Lu & Hyde (2000).

Fynbos is a vegetation type dominated by shrubs and comprises species specific to South Africa's south-western and southern Cape (Cowling & Richardson 1995). Over 7700 plant species are found in the fynbos. Of these about 70% are endemic to the area. The most

striking features of the fynbos are the conspicuous members of Proteaceae (shrubs with large leaves), Ericaceae (health-like shrubs), Restionaceae (reed-like plants) and geophytes (bulbous herbs) (Cowling & Richardson 1995). Because the fynbos is so floristically diverse and unique, a similar degree of diversity in fungi was expected. Previous studies have shown the pathogens of Proteaceae to be unique, and in most cases host specific (Taylor 2001). The present study was therefore initiated to determine if the same would also be the case for the saprobic fungi. To test this hypothesis, dicotyledonous Proteaceae and monocotyledonous Restionaceae in the Western Cape Province as host plants and the saprobic genus Anthostomella was chosen. During the course of this study four new species and two new records of Anthostomella were collected from South African fynbos. These fungi are illustrated and described, and a dichotomous key to the South African species of Anthostomella is provided.

MATERIALS AND METHODS

Eleven sites in the Western Cape Province were chosen depending on the host, the accessibility, and habitat characteristics. Dead leaves, twigs, and old flowerheads of *Proteaceae* and culms of *Restionaceae* were collected over a 2 yr period in (2000–01). The samples were inspected immediately for fungal structures, or air-dried for further study. Air-dried samples were incubated in moisture chambers for 2–3 d before examination. Single spore colonies were established on 5% malt

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extract agar (MEA; Biolab, Midrand, Johannesburg), supplemented with $0.04 \text{ g} \text{ l}^{-1}$ streptomycin sulphate. Cultural characteristics were rated for each isolate in triplicate from MEA plates after 10–30 d of incubation at 25 °C in the dark, and colours determined according to Rayner (1970). Observations, measurements and photography of characteristic structures were made from mounts in lactophenol. The 95% confidence intervals were derived from 30 observations to determine the range of variation in size of structures, with the extremes given in parentheses. Ascal subapical structures were stained using Melzer's solution. Sections of ascomata were made on a Leica CM1100 Cryostat microtome and mounted with Jung tissue freezing mediumTM (Leica Instruments, Germany). Photographic images were taken with Nikon Digital Camera DXM 1200 on a Nikon Eclipse E600 light microscope or a Nikon SMZ800 dissecting microscope. Herbarium specimens are lodged at PREM (National Collection of Fungi, Pretoria) and reference cultures are maintained in the culture collection of the Department of Plant Pathology, University of Stellenbosch (STE-U), and the Centraalbureau voor Schimmelcultures (CBS), Utrecht, in the Netherlands.

TAXONOMY

Anthostomella brabeji S. Lee & Crous, sp. nov.

(Figs 1-10)

Etym.: in reference to its host genus, *Brabejum*.

Ascomata subglobosa, ostiolata, clypeata, usque ad 500 µm alta et 530 µm lata. Asci unitunicati, octospori, cylindrici, pedicellati, apice rotundato, $(102-)105-122.5(-134) \times (5-)$ 6–8(–9) µm, apparatu subapicali, amyloideo, cuneato, 1.5– 2 × 2–2.5 µm. Ascosporae ellipsoideae vel fusiformes, pallide brunneae, glabro-tunicatae, $(12-)14-17.5(-19) \times (4.5-)5-$ 6 µm, unicellulares; nulla fissura germinalis observata, sine tunica gelatinosa.

Typus: **South Africa**: *Western Cape Province*: Kogelberg Nature Reserve, on dead leaves of *Brabejum stellatifolium*, 3 Nov. 2000, *S. Lee* SL348 (PREM 57445 – holotypus; ex-type cultures STE-U 4684, CBS 110128).

Ascomata perithecioid, immersed in host tissue, scattered, separate beneath a clypeus or infrequently gregarious beneath fused clypei, visible as black, shiny and slightly raised areas with a central papillate pore (Fig. 1); in vertical section subglobose, to 500 µm high and 530 µm wide (Fig. 2), with a single central periphysate ostiolar canal, up to 30 µm wide (Fig. 3). Clypeus black, shiny, globose to subglobose, margin distinct, 0.5-1 mm diam, consisting of dark, thick-walled hyphae in subepidermal cells. Peridium coriaceous, pseudoparenchymatous, comprising 2 layers of compressed cells, (25-)30-37.5(-42.5) µm thick, cells of outer layer angular, thick-walled, brown, cells of inner layer thin-walled, hyaline, flattened (Fig. 4), sometimes cells of transient layer between outer and inner layer present, moderately thick-walled, light brown. Paraphyses hyaline, filamentous, infrequently branched, flexuose, septate, numerous, $1.5-2.5(-3) \mu m$ thick (Fig. 5). Asci unitunicate, octosporous, cylindrical, pedicellate, apically rounded, $(102-)105-122.5(-134) \times (5-)6-8(-9) \mu m$ (Fig. 6); a subapical apparatus amyloid, wedge-shaped, $1.5-2 \times 2-2.5 \mu m$ (Fig. 7). Ascospores uniseriate, overlapping, ellipsoidal to fusiform, ends rounded, pale brown, smooth walled, $(12-)14.5-16(-19) \times (4.5-)5-6 \mu m$ (av. $15.3 \times 5.3 \mu m$), unicellular; no germ-slit observed, lacking a gelatinous sheath (Figs 8–10).

Cultural characteristics: Colonies circular with entire margin, white with a ochreous (15'b) central part, reverse buff (19"d) and dark brick (7"k) in the middle, with moderately dense, fluffy aerial mycelium. Colonies reaching 66 mm diam on MEA after 10 d at 25 $^{\circ}$ in the dark.

Geographic distribution: South Africa.

Host: Brabejum stellatifolium (Proteaceae).

Notes: Morphologically Anthostomella brabeji is similar to A. leucospermi by not having a dwarf cell, germ-slit, or gelatinous sheath, but differs in having ellipsoidal to fusiform ascospores. Additionally, the presence of a clypeus, the paler pigmentation of ascospores, and the moderately fast growth in culture further distinguishes A. brabeji from other taxa. Anthostomella bromeliaceae and A. lugubris are similar to A. brabeji. However, A. bromeliaceae has narrower ascospores $(14-18 \times 4-5 \mu m)$ with germ-slits, while A. lugubris has a gelatinous sheath and larger ascospores $(15-24 \times 9-11.5 \mu m)$.

Anthostomella clypeata (De Not.) Sacc., Syll. Fung. 1: 283 (1882). (Figs 11–21)

Ascomata perithecioid, immersed in host tissue, scattered, separate beneath a clypeus or sometimes gregarious beneath fused clypei, visible as black areas with a halo around the central pore or as black and conical areas with a central papillate pore (Figs 11-12); in vertical section subglobose, to 300 µm high and 320 µm wide, with a single central periphysate ostiolar canal, up to 20 µm wide (Fig. 13). Clypeus black or sometimes with a halo around the central pore, subglobose, margin indistinct, 0.2-0.3 mm diam, consisting of dark, thickwalled hyphae in both epidermal and subepidermal cells. Peridium coriaceous, pesudoparenchymatous, comprising 2 layers of compressed cells, $(10-)15-20(-22.5) \mu m$ thick, cells of outer layer angular, thick-walled, brown, cells of inner layer thin-walled, hyaline, flattened (Fig. 14). Paraphyses hyaline, filamentous, branched, flexuose, septate, numerous, (1.5-)2-2.5 µm thick. Asci unitunicate, octosporous, cylindrical, pedicellate, apically rounded, $(92.5-)95-105(-125) \times (7-)7.5-10(-12.5) \,\mu\text{m}$ (Figs 15-16); subapical apparatus amyloid, wedgeshaped, $2.5-3 \times 2-3 \mu m$ (Figs 17–18). Ascospores uniseriate, overlapping, ellipsoidal, ends rounded, brown, smooth walled, $(12-)12.5-14.5(-15) \times 5-7(-7.5) \,\mu\text{m}$ (av. $13.4 \times 6.1 \,\mu\text{m}$), bicellular; a dwarf basal cell hyaline, $2-3 \times 2-2.5 \,\mu\text{m}$; no germ-slit or gelatinous sheath observed (Figs 19-21).



Figs 1–10. *Anthostomella brabeji* (holotype, PREM 57445). **Fig. 1.** Ascomata on the host surface. **Fig. 2.** Section of an ascoma showing a clypeus. **Fig. 3.** Ostiole with periphyses. **Fig. 4.** Peridium. **Fig. 5.** Paraphyses. **Fig. 6.** Ascus. **Fig. 7.** Subapical apparatus. **Figs 8–10.** Ascospores. Bars: Fig. $2 = 100 \mu m$; Figs $3-6=10 \mu m$; Figs $7-10=5 \mu m$.



Figs 11–21. Anthostomella clypeata (PREM 57447). **Fig. 11.** Ascoma on the host surface. **Figs 12–13.** Sections through ascomata. **Fig. 14.** Peridium. **Figs 15–16.** Asci. **Figs 17–18.** Subapical apparatus. **Figs 19–21.** Ascospores. Bars: Figs $12-13 = 100 \,\mu\text{m}$; Figs $14-18 = 10 \,\mu\text{m}$; Figs $19-21 = 5 \,\mu\text{m}$.

Cultural characteristics: Colonies circular with entire margin or irregular, buff (19"d), reverse ochraceous (15'b), with dense, chiefly immersed, felty mycelium, slightly raised. Colonies reaching 2.5 mm diam on MEA after 10 d at 25 $^{\circ}$ in the dark.

Geographic distribution: Australia, Chile, Czech Republic, France, Italy, Portugal, South Africa, Turkey, USA (Florida, Hawaii).

Hosts: Araucaria brasiliana (Araucariaceae), Archontophoenix alexandrae (Palmae), Bambusa sp. (Gramineae), Ischyrolepis subverticellata (Restionaceae), Fraxinus excelsior (Oleaceae), Cannomois virgata (Restionaceae), Lonicera sp. (Caprifoliaceae), Oraniopsis appendiculata (Palmae), Restio egregius, R. cfr confusus (Restionaceae), Rubus ulmifolius, R. fruticosus, Rubus sp. (Rosaceae), Sabal palmetto (Palmae), Serenoa serrulata (Palmae), Smilax sp. (Smilacaceae), Uncinia erinacea (Cyperaceae).

Notes: The description above is based on isolate PREM 57447. This fungus is cosmopolitan and known from various substrates. This is the first report from South Africa, where it occurs on *Restionaceae*. Clypei of some isolates show a halo structure around the ostiolar pore which was not mentioned by Lu & Hyde (2000). South African collections also had asci with slightly longer subapical structures and wider ascospores than given by Lu & Hyde (2000) (subapical structures $1.3-1.5 \times 1.5-2 \,\mu$ m, ascospores $12-15 \times 5-5.5 \,\mu$ m).

Specimens examined: South Africa: Western Cape Province: Kogelberg Nature Reserve, on dead culms of Restio egregius, 3 Nov. 2000, S. Lee SL557 (PREM 57446); Jonkershoek Nature Reserve, on culms of Ischyrolepis subverticellata, 5 Apr. 2001, S. Lee SL678 (PREM 57447, cultures STE-U 4686); on dead culms of Cannomois virgata, 15 Jun. 2001, S. Lee SL764 (PREM 57448, cultures STE-U 4685), on dead culms of Restio cfr confusus, S. Lee SL796 (PREM 57449, cultures STE-U 4692).

Anthostomella conorum (Fuckel) Sacc., Syll. Fung. 1: 283 (1882). (Figs 22–31)

Ascomata perithecioid, immersed in host tissue, scattered, separate beneath a clypeus or infrequently gregarious beneath fused clypei, visible as black and conical areas with a central papillate pore (Fig. 22); in vertical section obpyriform, to 380 µm high and 320 µm wide (Fig. 23), with a single central periphysate ostiolar canal, to 32.5 µm wide (Fig. 24). Clypeus black, globose, margins indistinct, 0.2-0.3 mm diam, consisting of dark, thick-walled hyphae in subepidermal cells. Peridium coriaceous, pseudoparenchymatous, comprising 2 layers of compressed cells, 17.5-32.5 µm thick, cells of outer layer angular, moderately thick-walled, light brown becoming paler towards the outside, cells of inner layer thin-walled, hyaline, flattened (Fig. 25). Paraphyses hyaline, filamentous, unbranched, flexuose, septate, numerous, 2.5-4(-4.5) µm wide. Asci unitunicate, octosporous, cylindrical, pedicellate, apically rounded, $(10-)105-135(-147.5) \times (8-)10-12.5 \ \mu\text{m}$ (Fig. 27); subapical apparatus amyloid, wedge-shaped to discoid, $1.5-2 \times 4-5 \ \mu\text{m}$ (Fig. 26). *Ascospores* uniseriate, overlapping, ellipsoidal, ends rounded, brown, smooth walled, $(12.5-)13-15 \times 7.5-8(-9) \ \mu\text{m}$ (av. $14.1 \times 7.7 \ \mu\text{m}$), unicellular; germ-slit straight, extending over the full length; gelatinous sheath $1-2 \ \mu\text{m}$ thick, slightly thinner at each end (Figs 28–31).

Cultural characteristics: Colonies circular with entire margin, surface white and reverse ochreous (15'b), with dense, cottony to felty superficial mycelium, raised. Colonies reaching 11 mm diam. on MEA after 30 d at 25 $^{\circ}$ in the dark.

Geographic distribution: Belgium, France, Germany, Greece, Italy, Portugal, South Africa, Spain, UK.

Hosts: Artemisia campestris (Compositae), Cytisus fontanesius (Leguminosae), Coronilla glauca (Leguminosae), Galium fruticosum (Rubiaceae), Helleborus foetidus (Ranunculaceae), Laurus nobilis (Lauraceae), Leucadendron sp. (Proteaceae), Mesembryanthemum edule (Aizoaceae), Pinus sylvestris (Pinaceae), Protea magnifica, P. neriifolia (Proteaceae), Salix sp. (Salicaceae), Urginea maritime (Hyacinthaceae).

Notes: The above description is based on the isolate PREM 57452. This is the first record of *Anthostomella conorum* from South Africa, where it occurs on *Proteaceae*. The ascospores of these isolates are slightly smaller than those cited by Lu & Hyde (2000) $(14-19 \times 7.5-10 \ \mu\text{m})$.

Specimens examined: South Africa: Western Cape Province: J. S. Marais Nature Reserve, on dead leaves of Protea magnifica, 6 Jun. 2000, S. Lee SL152 (PREM 57450, cultures STE-U 4688); Gordon's Bay, on dead leaves of Protea neriifolia, 26 Jun. 2000, S. Lee SL187 (PREM 57451, culture STE-U 4689); Kleinmond, on dead leaves of Leucadendron sp., 11 Jul. 2000, S. Lee SL629 (PREM 57452, cultures STE-U 4687).

Anthostomella leucospermi S. Lee & Crous, sp. nov. (Figs 32–40)

Etym.: in reference to the host genus, *Leucospermum*.

Ascomata globosa, ostiolata, clypeata, usque ad 480 µm alta et 420 µm lata. Asci unitunicati, octospori, cylindrici, pedicellati, apice rotundato, $(100-)110-120(-137.5) \times 10-11.5$ (-12.5) µm, apparatu subapicali, amyloideo, cuneato, $2-3 \times 3-4$ µm. Ascosporae inaequilateraliter ellipsoideae, brunneae, glabro-tunicatae, $(10-)12.5-15(-17) \times (5.5-)7-8(-9)$ µm, unicellulares; nulla fissura germinalis observata, sine tunica gelatinosa.

Typus: **South Africa**: *Western Cape Province*: Kleinmond Costal Nature Reserve, on dead leaves of *Leucospermum oleifolium*, 11 Jul. 2000, *S. Lee* SL632 (PREM 57453 – holo-typus; ex-type cultures STE-U 4690, CBS 110126).

Ascomata perithecioid, immersed in host tissue, scattered, separate beneath a clypeus, visible as black and conical areas with a central papillate pore (Fig. 32); in vertical section globose, to 480 μ m high and 420 μ m wide, with a single central periphysate ostiolar canal, to 20 μ m wide (Fig. 33). *Clypeus* black, globose to subglobose, margin indistinct, 0.2–0.5 mm diam, consisting



Figs 22–31. Anthostomella conorum (PREM 57452). **Fig. 22.** Ascomata on the host surface. **Fig. 23.** Section through an ascoma. **Fig. 24.** Ostiole. **Fig. 25.** Peridium. **Fig. 26.** Subapical apparatus. **Fig. 27.** Ascus. **Figs 28–31.** Ascospores. Bars: Fig. 23 = 100 μ m; Fig. 24 = 50 μ m; Figs 25–27 = 10 μ m; Figs 28–31 = 5 μ m.

of dark, thick-walled hyphae in subepidermal cells. *Peridium* coriaceous, pseudoparenchymatous, comprising 2 layers of compressed cells, $20-35 \,\mu\text{m}$ thick, cells of outer layer angular, thick-walled, brown, cells of inner layer thin-walled, hyaline, flattened (Fig. 34). *Paraphyses* hyaline, filamentous, unbranched, flexuose, septate, numerous $1-2.5(-3) \,\mu\text{m}$ thick (Fig. 36). *Asci* unitunicate, octosporous, cylindrical, pedicellate, apically rounded, $(100-)110-120(-137.5) \times 10-11.5$ (-12.5) μm (Fig. 35), subapical apparatus amyloid, wedge-shaped, $2-3 \times 3-4 \,\mu\text{m}$ (Fig. 37). *Ascospores*

uniseriate, overlapping, inequilaterally ellipsoidal, ends rounded or infrequently apiculate, brown, smooth walled, $(10-)12.5-15(-17) \times (5.5-)7-8(-9) \mu m$ (av. $14.2 \times 7.0 \mu m$), unicellular; no germ-slit or gelatinous sheath observed (Figs 38–40).

Cultural characteristics: Colonies circular with entire margin, surface white and reverse luteous (17b), with dense, cottony aerial mycelium. Colonies reaching 9 mm diam on MEA after 10 d at 25 $^{\circ}$ in the dark.

Geographic distribution: South Africa. Host: Leucospermum oleifolium (Proteaceae).



Figs 32–40. Anthostomella leucospermi (holotype, PREM 57453). Fig. 32. Ascomata on the host surface. Fig. 33. Section through an ascoma. Fig. 34. Peridium. Fig. 35. Asci. Fig. 36. Paraphyses. Fig. 37. Subapical apparatus. Figs 38–40. Ascospores. Bars: Fig. $33 = 100 \mu m$; Figs $34-36 = 10 \mu m$; Figs $37-40 = 5 \mu m$.

Notes: Only a few species of Anthostomella lack ascospore germ-slits, dwarf basal cells and gelatinous sheaths. Anthostomella leucospermi lacks these structures, and also has inequilaterally ellipsoidal ascospores. Anthostomella scirpi is one of the species most similar to A. leucospermi, but has a non-amyloid subapical ascal structure. A. tumulosa differs from A. leucospermi by having larger ascospores (17.5–22.5 × $6.5-9.5 \mu m$), and a gelatinous sheath; and A. auatica has ascospores of similar dimensions to those of A. leucospermi, but with short germ-slits.

Anthostomella proteae S. Lee & Crous, sp. nov. (Figs 41–49)

Etym.: in reference to its host genus, Protea.

Ascomata globosa, ostiolata, clypeata, usque ad 370 µm alta et 320 µm lata. Asci unitunicati, octospori, cylindrici, pedicellati, apice rotundato, $(95-)115-137.5(-142.5) \times (6.5-)7.5-9$ µm, apparatu subapicali, amyloideo, cuneato. Ascosporae inaequilateraliter ellipsoideae, brunneae, glabro-tunicatae, $(12.5-)13.5-15(-17.5) \times (5-)5.5-7.5(-8)$ µm, bicellulares, cella nana basilari hyalina, $1.5-2 \times 2-3$ µm, fissura germinalis



Figs 41–49. *Anthostomella proteae* (holotype, PREM 57454). **Fig. 41.** Ascomata on the host surface. **Fig. 42.** Section through an ascoma. **Fig. 43.** Ostiole. **Fig. 44.** Peridium. **Figs 45–46.** Asci with subapical apparatus. **Figs 47–49.** Ascospores. Bars: Fig. $42 = 100 \mu m$; Fig. $43 = 50 \mu m$; Figs $44-48 = 10 \mu m$; Fig. $49 = 5 \mu m$.

recta, indistincta, extensa per totam longitudinem, tunica gelatinosa 1.5–2 μm crassa.

Typus: **South Africa**: *Western Cape Province*: Jonkershoek Nature Reserve, on dead leaves of *Protea nitida*, 6 Jun. 2000, *S. Lee* SL59 (PREM 57454 – holotype; ex-type cultures STE-U 4691, CBS 110127).

Ascomata perithecioid, immersed in host tissue, scattered, separate, visible as faintly darkened and raised areas with a central papillate pore (Fig. 41); in vertical section globose, to 370 μ m high and 320 μ m wide (Fig. 42), with a single central periphysate ostiolar canal, up to 37.5 μ m wide (Fig. 43). *Clypeus* absent. *Peridium* coriaceous, pseudoparenchymatous, comprising 2 layers of compressed cells, 15–17.5(–20) μ m thick, cells of outer layer angular, thick-walled, brown, cells of inner layer thin-walled, hyaline, flattened (Fig. 44). *Paraphyses* hyaline, filamentous, unbranched, flexuose, septate, sparse (2–)3–4 μ m thick. *Asci* unitunicate,

octosporous, cylindrical, pedicellate, apically rounded $(95-)115-137.5(-142.5) \times (6.5-)7.5-9 \ \mu m$ (Fig. 45); a subapical apparatus amyloid, wedge-shaped, $4-5 \times (2-)3 \ \mu m$ (Fig. 46). *Ascospores* uniseriate, overlapping, inequilaterally ellipsoidal, ends rounded, brown, smooth walled, $(12.5-)13.5-15(-17.5) \times (5-)5.5-7.5$ (-8) μm (av. $15.0 \times 6.2 \ \mu m$), bicellular; a basal dwarf cell hyaline, $1.5-2 \times 2-3 \ \mu m$; germ-slit straight, indistinct, extending over the full length; gelatinous sheath $1.5-2 \ \mu m$ thick (Figs 47-49).

Cultural characteristics: Colonies irregular with erose edges, surface white and reverse ochreous (15'b), with a medium density, raised, fluffy aerial mycelium. Colonies reaching 14 mm after 10 d at 25 $^{\circ}$ in the dark.

Geographic distribution: South Africa.

Host: Protea nitida (Proteaceae).

Notes: Perithecia of Anthostomella proteae lack a clypeus. A slight darkened area is visible around the ostiole, however, which is the consequence of some intracellular hyphal growth in the subepidermal cells. Vertical sections through the perithecium revealed a heavily pigmented area around the ostiolar canal, which is the thickened peridium usually extending up into the ostiole. A. proteae has asci with amyloid subapical apparati and ascospores with dwarf cells, germ slits and gelatinous sheaths. There are a few species morphologically similar to A. proteae. A. proteae is similar to A. tomicoides in ascospore dimensions and ascus morphology, to which Bingsheng Lu (pers. comm.) considers it to be a conspecific. We believe it to be distinct, however, based on the following differences: the as cospores of A. tomicoides $(14-20 \times 5-8 \mu m)$ are slightly longer than those of A. proteae $(12.5-17.5 \times 5-8 \,\mu\text{m})$; in all collections of A. proteae studied, the germ-slit consistently extended over the full spore length; and the absence of a clypeus. A. proteae is thus different from A. tomicoides. Ascospores of A. proteae $(15-17.5 \times 6.5-$ 9 µm) are also similar to those of A. foveolaris (12.5- $14.5 \times 5.5 - 7 \,\mu\text{m}$), but differ in being somewhat larger. Among South African species, A. meerensis is the most similar to A. proteae. The two differ, however, in ascospore size, and the nature of the ascal subapical structure. Ascospores of A. proteae are smaller than those of A. meerensis $(17.5-19.5 \times 7.5-9 \,\mu\text{m})$, and the subapical structure of A. meerensis does not stain blue in Melzer's solution.

Anthostomella cynaroides S. Lee & Crous, sp. nov. (Figs 50–58)

Etym.: in reference to its host species, *Protea* cynaroides.

Ascomata subglobosa, ostiolata, clypeata, usque ad 500 µm alta, usque ad 530 µm lata. Asci unitunicati, fere octospori, cylindrici, pedicellati, apice rotundato, (137.5-)142.5-155 (-164) × (11-)12.5-15 µm, apparatu subapicali, amyloideo, cuneato, 4.5-5 × 3.5-4 µm. Ascosporae ellipsoideae, brunneae, glabro-tunicatae, $(17.5-)20-22.5(-25) \times (7-)7.5-8(-10)$ µm, bicellulares, cella nana basilari hyalina $1.5-2 \times 3$ µm, nulla fissura germinalis observata, sine tunica gelatinosa.

Typus: **South Africa**: *Western Cape Province*: Helderberg Nature Reserve, on a decorticated stem of *Protea cynaroides*, 14 Aug. 2000, *S. Lee* SL208 (PREM 57445 – holotypus).

Ascomata perithecoid, immersed in host tissue, scattered, separate beneath a clypeus or sometimes gregarious beneath fused clypei; visible as black and conical areas with a central papillate pore (Fig. 50); in vertical section obpyriform, to 250 µm high and 190 µm wide (Fig. 51), with a central single ostiolar canal, to 30 µm wide (Fig. 52). Clypeus black, ellipsoidal, margin indistinct, 0.2–1 mm diam, consisting of dark, thick-walled hyphae in both epidermal and subepidermal cells. Peridium coriaceous, pseudoparenchymatous, comprising 2 layers of compressed cells, (12.5-)15-20 µm thick, cells of outer layer angular, thick-walled, brown, cells of inner layer thin-walled, hyaline, flattened (Fig. 53), boundaries between host cell and peridium indistinct. Paraphyses hyaline, filamentous, unbranched, flexuose, septate, numerous 1.5-2(-3) µm thick. Asci unitunicate, mostly octosporous, cylindrical, pedicellate, apically roundend, $(137.5-)142.5-155(-164) \times (11-)12.5-15 \,\mu m$ (Figs 54-55); subapical apparatus amyloid, wedgeshaped, $4.5-5 \times 3.5-4 \,\mu m$ (Fig. 56). Ascospores uniseriate, overlapping, ellipsoidal, laterally flattened, ends rounded or apex slightly apiculate, brown, smooth walled, $(17.5-)20-22.5(-25) \times (7-)7.5-8(-10) \mu m$ (av. $21.7 \times 7.7 \,\mu\text{m}$), bicellular; a basal dwarf cell hyaline, $1.5-2 \times 3 \,\mu\text{m}$; no germ-slit or gelatinous sheath observed (Figs 57-58).

Geographic distribution: South Africa.

Host: Protea cynaroides (Proteaceae).

Notes: Anthostomella cynaroides is a typical species of Anthostomella possessing a basal dwarf cell. Ascospores of A. cynaroides are devoid of gelatinous sheaths and germ slits. Even in the young stage of ascospore development when these structures are easily recognizable, no germ slit or gelatinous sheath was found. Lu & Hyde (2000) considered the presence or absence of a gelatinous sheath not to be a decisive character, but only considered it when it was associated with other characters. Among the species of Anthostomella having ascospores that have dwarf cells but neither germ-slits nor gelatinous sheaths, A. rubicola $(20-27.5 \times 5-8 \,\mu\text{m})$ is the closest to A. cynaroides $(17.5-25 \times 7-10 \,\mu\text{m})$ in ascospore dimensions, and B. Lu (pers. comm.) considers them to be conspecific. We choose to separate these two taxa, however, based on the following differences: the ascospores of A. rubicola have rostrate dwarf cells and sharply tapering apices, while dwarf cells of A. cynaroides are semi-globose, and ascospores have obtuse apices: further, ascospores of A. rubicola remain hyaline in their asci, while those of A. cynaroides become brown in asci.

DISCUSSION

In this study six species of *Anthostomella* were isolated from eleven different host plants in either the *Proteaceae* or *Restionaceae*. Of these, four taxa were recorded



Figs 50–58. Anthostomella cynaroides (holotype, PREM 57445). **Fig. 50.** Ascomata on the host surface. **Fig. 51.** Section through an ascoma. **Fig. 52.** Ostiole. **Fig. 53.** Peridium. **Figs 54–55.** Asci. **Fig. 56.** Subapical apparatus. **Figs 57–58.** Ascospores. Bars: Fig. 51 = 100 μ m; Figs 52 = 25 μ m; Figs 53–55 = 10 μ m; Figs 56–58 = 5 μ m.

as new to science and proved to be unique to the fynbos. A further two species were newly reported from these hosts. These data are consistent with that of a previous study, which highlighted the richness of new fungal species occurring in the fynbos (Taylor, Lee & Crous 2001). It is estimated that fynbos harbours 75% of South Africa's rare and threatened plants

(Hall & Veldhuis 1985). Moreover, urbanization, agriculture, invasion of alien plants, and frequent fires continuously accelerate the obliteration of the flora and fauna component of the fynbos (Cowling & Richardson 1995). This phenomenon in turn threatens its mycota. In the present study, only a few resios and protea species were examined for species of *Anthostomella*,

Key to Anthostomella species from South Africa

As As	scospores with a spiral germ-slit	-slit			•	•	•	•	•	•	•		•	•		2
As	scospores 16.5–20.5 μm in length	h														umbrinella
As	scospores 20–34 µm in length	•	•	•	•	•	•		•	•	•			•	•	. spiralis
As As	scospore wall verrucose .				•											vestitata
As	scospores with a dwarf cell			•												5
Ba	asal dwarf cell rostrate .															. rubicola
Ba	asal dwarf cell not rostrate	•	•	•	•	•	•	•	•	•	•	•		•		6
Br Br	cown ascospore cell without a ge cown ascospore cell with germ-sl	erm-sli lit	t		•			•								7
As As	scospores without a gelatinous s scospores with a thick gelatinous	heath s sheat	th			•	•		•	•						. clypeata . colligata
As As	scospores without a gelatinous s scospores with a gelatinous shea	heath th				•	•								•	cynaroides
As As	scal subapical structure I – scal subapical structure I +	•	•	•	•	•	•		•	•					•	. meerensis
Asc As	cal subapical structure I –						•								•	. palmae
As	scospores with a germ-slit															12
As	scospores without a gelatinous s	heath	•	•	•	•	•	•	•	•	•	•	•	•	•	leucosnermi
As	scospores with a gelatinous shea	th														. brabeji
As As	scospores with hyaline polar gela scospores without polar pads	atinou	s pads		•			•								. applanata
As As	sci with an apical structure															. caffrariae
As	scal subapical structure discoid														•	. raphiae
As	scal subapical wedge-shaped	•	•	•	•	•	•		•	•	•		•	•	•	16
As	scospores inequilateral .				•			•	•							
As As	scospores 12–14.5 × 3.5–5 μm scospores 12.5–15 × 7.5–9 μm															. acuminata . conorum

which in turn revealed several taxa new to science. This study therefore underlines the great potential of finding unusual and diverse mycoflora associated with members of the fynbos, as well as other habitats and substrates in the fynbos yet to be studied. Further studies on the biodiversity of saprobic fungi in the fynbos are presently being undertaken, and will contribute to the better understanding and support for conservation of the fynbos biome.

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