Sclerococcum phaeophysciae and *S. toensbergii*, two new lichenicolous asexual Ascomycetes, with a revised key to the species of *Sclerococcum*

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Abstract. The new species *Sclerococcum phaeophysciae* Diederich & van den Boom, collected on *Phaeophyscia orbicularis* in Belgium, Germany, Luxembourg and the Netherlands, is characterized by smooth, 1-septate conidia, and is distinguished from *S. montagnei* by larger conidiomata and conidia. The new *S. toensbergii* Diederich, collected on *Megalaria pulverea* and *Pertusaria carneopallida* in the USA, Washington, is characterized by multicellular conidia with a mostly smooth and regularly thickened cell wall. A revised key to the lichenicolous species of *Sclerococcum* is given.

1. Introduction

The genus *Sclerococcum* Fr. was described for the asexual lichenicolous hyphomycete *S. sphaerale* (Ach.) Fr., forming blackish sporodochia with dark brown muriform conidia on the thallus of *Pertusaria corallina* (Hawksworth 1975). More and more species were described within the genus, until Diederich et al. (2013) transferred five species with 0–1-septate, catenate conidia to the genus *Cladophialophora* Borelli (Chaetothyriales), based on molecular phylogenetic results, whilst the type species proved to be closely related to *Dactylospora* Körb. (Eurotiomycetes). The genus *Sclerococcum* is possibly still heterogeneous, as exemplified by the rather different morphological and chemical (pigmentation) characters observed in two recently described species collected on Graphidaceae hosts (Diederich 2015). A key to all known lichenicolous species of *Sclerococcum* was published by Diederich (2015).

In this paper, two new species of sporodochioid hyphomycetes, one apparently confined to *Phaeophyscia orbicularis*, the other collected on *Megalaria pulverea* and *Pertusaria carneopallida* in the USA, are described. Unfortunately attempts to culture fresh material of the former species failed, and herbarium specimens of the latter were too old for obtaining a culture, hence the inclusion within *Sclerococcum* is merely based on morphological similarities with the other species of the genus.

2. Material and Methods

The studied specimens are kept in BG, BR and in the private collection of the authors. Dry herbarium specimens were examined and measured under a binocular microscope Leica MZ 7.5 (magnification up to ×50), and photographed using a Canon 40D camera with a Nikon BD Plan 10 microscope objective, StackShot (Cognisys) and Helicon Focus (HeliconSoft) for increasing the depth of field. Handmade sections of conidiomata were studied in water, 10% KOH, Phloxine B and Congo red. Microscopic photographs were prepared using a Leica DMLB microscope, a Leica EC3 camera and Helicon Focus. Conidial measurements are indicated as (min.–)$\overline{X}$-σ_x–$\overline{X}$+σ_x (–max.), followed by the number of measurements (n); the length/breadth ratio of conidia is indicated as Q and given in the same way. Chemical reactions were tested using 10% KOH (K).

3. Results

Sclerococcum phaeophysciae Diederich & van den Boom sp. nov. (Figs 1–2)

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Distinguished from the only other known *Sclerococcum* species with smooth, 1-septate conidia (*Sclerococcum montagnei*) by larger conidiomata, 200–600 µm diam., larger conidia, (9.7–)11.1–14.6(–17) × (8.0–)9.1–10.9(–12.0) µm, and a different host selection, *Phaeophyscia orbicularis*.

Typus: Germany, Brandenburg, SW of Berlin, NE of Potsdam, old churchyard in mixed forest, 52°22' N, 13°11' E, alt. 30 m, top of concrete pole, on *Phaeophyscia orbicularis*, 22 Febr. 2004, *P. & B. van den Boom* 32074 & *H. Sipman* (BR–holotype; herb. van den Boom, herb. Diederich–isotypes).

Colonies lichenicolous on *Phaeophyscia orbicularis*, forming superficial, flattened or rarely convex sporodochia, dark brown to blackish, rounded, elongate or irregular in form, 200–600 µm diam., not or occasionally confluent. Vegetative hyphae hyaline, immersed in the host thallus, indistinct. Conidiophores aggregated into dense sporodochia, not or sparsely branched, hyaline or pale brown. Conidiogenous cells monoblastic or rarely polyblastic, terminal, integrated, hyaline or pale brown, subspherical to ellipsoid, mainly 5–10 µm diam., hardly distinguishable from other conidiophore cells. Conidia produced singly, separating easily, dry, acrogenous, subspherical, ellipsoid or angular, medium to dark brown, smooth-walled, usually 1-septate, (9.7–)11.1–14.6(–17) × (8.0–)9.1–10.9(–12.0) µm, Q = (1.0–)1.1–1.5(–1.8) (n = 40), exceptionally 2-septate; septum 1.5–3 µm thick, dark brown, often with a distinct blackish lamella; wall mostly medium brown, 0.7–1.2 µm thick, in some portions much thicker and darker, up to 2.2 µm thick. All parts K– (becoming slightly darker).

Distribution and hosts. Known from Belgium, Germany, Luxembourg and the Neth-

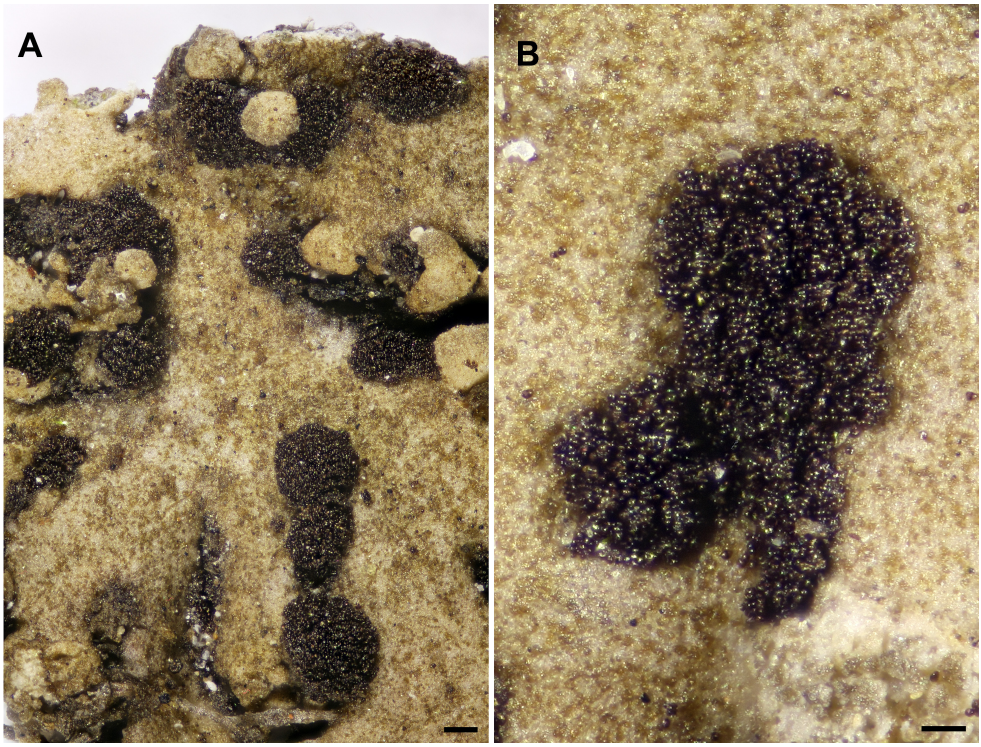

Fig. 1. *Sclerococcum phaeophysciae* (holotype). A, Sporodochia developing on the thallus of *Phaeophyscia orbicularis*. B, Sporodochium at a higher magnification. Scale bars: A = 100 µm; B = 50 µm.

erlands, on thalli of *Phaeophyscia orbicularis* that are not visibly damaged. It has been found mostly on saxicolous *P. orbicularis* and more rarely on epiphytic specimens.

Observations. The only other known *Sclerococcum* species with mainly 1-septate conidia is *S. montagnei* Hafellner, which differs in having smaller conidiomata, 200–300 µm

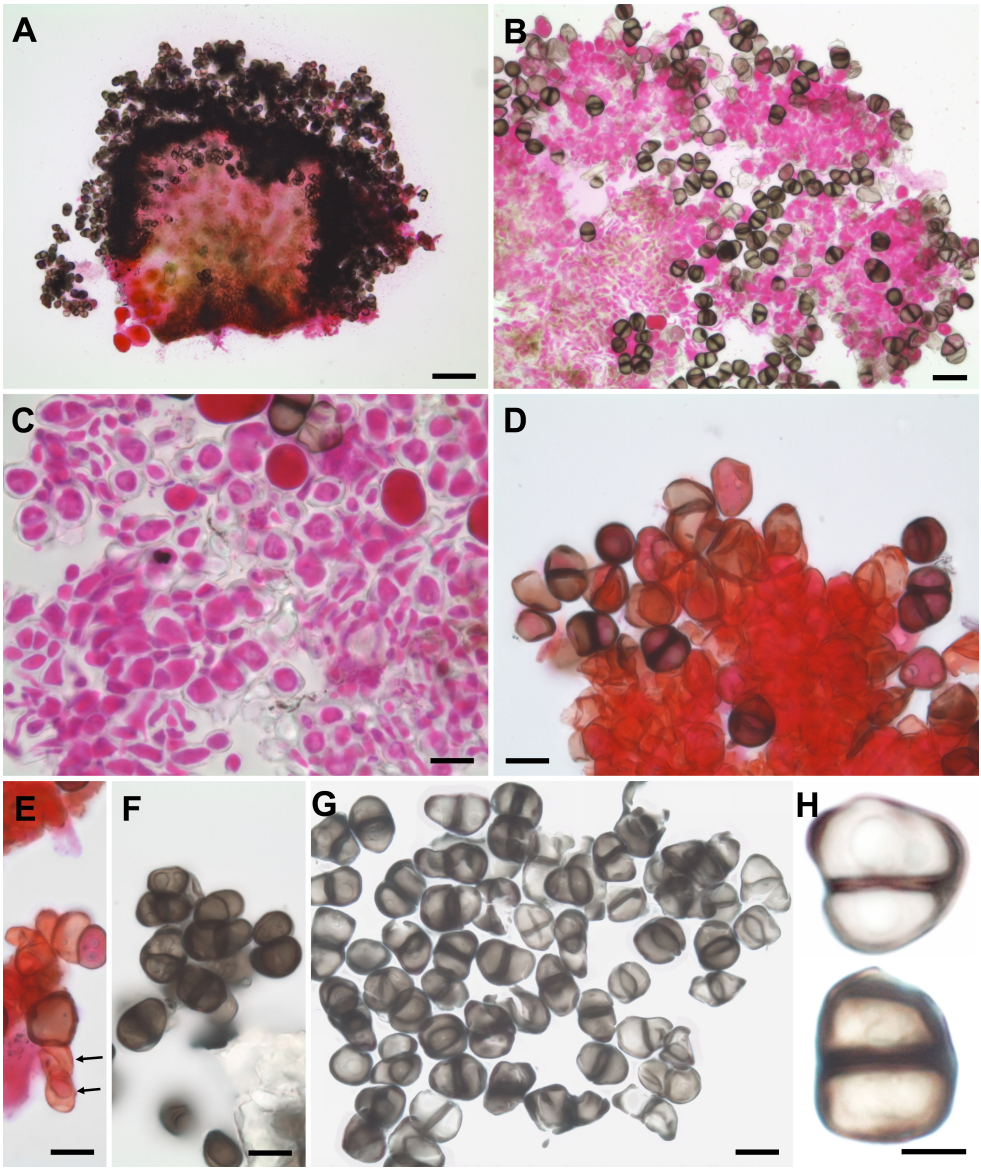

Fig. 2. *Sclerococcum phaeophysciae* (holotype). A, Section through conidioma. B, Section through conidioma, with conidiophores separated through pressure on cover glass. C, Basal part of conidioma in contact with host thallus (A–C, in Phloxine B after K pre-treatment). D, Exposed part of conidioma, indistinctly showing (partly collapsed) conidiophores and conidia. E, Young, aseptate conidium attached to two cells of conidiophore (arrows) (D–E, in a mixture of Phloxine B and Congo red). F, Conidia in water. G, Conidia in K. H, Conidia in K, showing irregularly thickened wall, and septum with darker lamella. Scale bars: A = 50 µm; B = 20 µm; C–G = 10 µm; H = 5 µm.

diam., distinctly smaller and often irregularly catenate conidia, 10–13 × 6–9 µm when 1-septate, and a different host selection, *Lecanora rupicola* (Hafellner 1996).

Additional specimens examined (all on *Phaeophyscia orbicularis*; all in the private herbaria of the collectors): **Belgium:** Prov. Antwerpen: Baarle-Hertog, enclave-area, churchyard (IFBL B5.18), on gravestone, 2014, *van den Boom* 50258. Prov. Limburg: S of Molenbeersel, nature reserve 'Zig', W edge, bridge over stream Abeek (C7.35), on horizontal surface of concrete, 2004, *van den Boom* 32657; NE of Bree, E of Bocholt, S side along nature reserve 'Luysen', between Urlobroek and Mariahof (C7.34), on roadside concrete fence posts along meadow, 2004, *van den Boom* 33766; E of Maaseik, S of Aldeneik, churchyard (C7.57), on gravestone, 2012, van den Boom 47412; Hamont, centre, churchyard (C7.12), on gravestone, 2012, van den Boom 48137. Prov. Liège: N of Verviers, Aubel centre, churchyard (F7.18), on gravestone, 2012, *van den Boom* 48201. Prov. Luxembourg: Bomal, W side of city, path to top of hill Le Calvaire, 2005, *van den Boom* 34678. **Luxembourg:** SSW of Bascharage, near Moulin de Bascharage (M8.31), on *Malus*, 1987, *Diederich* 8979; NE of Bergem, Schéierbësch, on *Populus*, 1987, *Diederich* 8524. **Netherlands:** Prov. Noord-Brabant: St. Oedenrode centre of village, churchyard, on concrete of gravestone, 2012, *van den Boom* 47339; Son centre, churchyard, on horizontal concrete surface of tombstone, 2005, *van den Boom* 34129; Bergeyk centre, old churchyard, on tombstones, 2012, *van den Boom* 48112, 48118; Nuenen centre, churchyard, on tombstones, 2006, *van den Boom* 37378; SW of Valkenswaard, Westerhoven, old churchyard, on gravestones, 2010, *van den Boom* 45001.

Sclerococcum toensbergii Diederich sp. nov. (Figs 3–4)

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Distinguished from the other known *Sclerococcum* species with muriform, smooth-walled conidia by the number of conidial cells (8–12 cells visible in optical section vs 2–6-celled in *Sclerococcum epiphytorum* and *S. sphaerale*, and up to 60-celled in *S. phyllobaeis*), by conidia with evenly thickened walls (unevenly thickened in *S. serusiauxii*), and by conidiomata 150–250 µm diam. (43–145 µm diam. in *S. epicladonia*); conidia 13.5–18.8 × 10–14.6 µm, K–; conidial cells 3.5–6 µm diam.

Typus: USA, Washington, Cowlitz Co., 7–8 km SW of summit of Mount St. Helens, E of Goat Mtn, NE of Goat Marsh Lake, N of Coldspring Creek, W of gravel Rd FR 8123, 46°10' N, c. 122°16.5' W, alt. 900–1000 m, corticolous on trunk of *Alnus rubra*, on *Pertusaria carneopallida* (accompanied by *Phlyctis speirea*), 8 August 1996, *T. Tønsberg* 24085 (BG L-71917–holotype; WTU, herb. Diederich–isotypes).

Colonies lichenicolous on corticolous lichens, forming superficial, slightly convex sporodochia, blackish, rounded, elongate or irregular in form, (100–)150–250(–300) µm diam., occasionally confluent, often poorly delimited because of mature conidia spreading around the conidiogenous area. Vegetative hyphae pale to medium brown, immersed in the host thallus, (2–)3–4(–5) µm diam. Conidiophores aggregated into dense sporodochia, not or sparsely branched, pale to medium brown, 2–3.5 µm thick. Conidiogenous cells monoblastic or polyblastic, terminal, integrated, brown, subspherical to ellipsoid or elongate, hardly distinguishable from other conidiophore cells. Conidia produced singly, rarely catenate, separating easily, dry, acrogenous, subspherical or ellipsoid, medium to dark brown, muriform, with less than 20 cells [(6–)8–12(–15) cells visible in optical section], (12.5–)13.5–18.8(–23.0) × (7.7–)10.0–14.6(–17.5) µm, Q = (1.0–)1.1–1.5(–1.9) (n = 40), often spreading around sporodochia on host thallus; conidial cells subspherical to ellipsoid, 3.5–6 µm diam.; septa 0.5–1 µm thick, dark brown; wall dark brown, 0.5–1 µm thick, smooth or rarely becoming rough. All parts K– (becoming slightly darker).

Distribution and hosts. Known from two localities in the USA, Washington, on corticolous thalli of *Megalaria pulverea* and *Pertusaria carneopallida* that are not visibly damaged. It should be noted, however, that the thallus of *P. carneopallida* had only a few apothecia, which are usually more numerous in this species (Dibben 1980).

Observations. The other known *Sclerococcum* species with muriform, mainly smooth-walled conidia are *S. epicladonia* Zhurb., described from *Cladonia* (Zhurbenko & Pino-Bodas 2017), *S. epiphytorum* Diederich, described from *Varicellaria hemisphaerica* (Diederich 1990), *S. phyllobaeis* Etayo, described from *Phyllobaeis* (Etayo 2017), *S. serusiauxii* Boqueras & Diederich, known from *Parmelina* species (Boqueras & Diederich 1993), and *S. sphaerale* (Ach.) Fr., growing on *Pertusaria corallina* or rarely on other saxicolous *Pertusaria* species (Hawksworth 1975). *Sclerococ-*

cum epiphytorum and *S. sphaerale* are readily distinguishable from the new species by its 2–6-celled conidia, whilst *S. serusiauxii* differs by conidia with an unevenly thickened cell wall and distinctly smaller conidiomata, 60–180 µm diam; *S. epicladonia* differs in having conidia up to 50-celled and much smaller conidiomata, 43–145 µm diam.; *S. phyllobaeis* has larger conidia, 11–30 × 10–20 µm, becoming up to 60-celled when mature.

All other lichenicolous *Sclerococcum* species are known from a single host genus, or from closely related genera. It is therefore surprising to have two morphologically very similar specimens on two non-related host genera, *Megalaria* (Lecanorales) and *Pertusaria* (Pertusariales). Either this is the first known non-specialized species of *Sclerococcum*, or the two specimens belong to two morphologically similar, but genetically distinct species. To avoid any confusion, the description above is based solely on the type specimen. Freshly collected specimens from both hosts are needed to obtain a molecular answer.

Etymology. The new species is named after Tor Tønsberg, collector of both specimens of the new species and expert of corticolous, crustose lichens from the Northern Hemisphere.

Additional specimen examined: **USA**: Washington: Clallam Co., [Olympic Peninsula,] between Forks and La Push, E of Quillayute, S of Soleduck River Bridge at junction Quillayute Rd/La Push Rd, 0.4 km along gravel road E of Soleduck River, Norm Cowan Memorial Fishing and Picnic Area, at the parking lot, 47°57' N, 124°28' W, alt. 50 m, corticolous on trunk of *Alnus rubra*, on *Megalaria pulverea* (accompanied by *Lecanora cinereofusca*), 1 Dec. 1994, *Tønsberg* 21577 (BG, WTU, herb. Diederich).

Revised key to the lichenicolous species of *Sclerococcum*

A key to all known lichenicolous *Sclerococcum* species was provided by Diederich (2015). Two species were excluded from that key: *Sclerococcum griseosporodochium* Etayo, nowadays considered as a lichenized, non lichenicolous species, and *Sclerococcum acarosporae* S. Y. Kondr., described from *Acarospora* cf. *laqueata* in Israel, a species with deviating morphological characters, resembling some species of *Lichenostigma* subgen. *Lichenogramma*. Three additional species have recently been described by Etayo (2017) and Zhurbenko & Pino-Bodas (2017). We therefore present a revised key to the lichenicolous species of *Sclerococcum*.

1. Conidia 1(–2)-celled **2**
1. Conidia 2- to multi-celled **6**

2. Conidia smooth-walled **3**
2. Conidial wall ornamented **5**

3. Conidia 7.5–9.5 × 6.5–8 µm, with a thick wall up to 2 µm wide; on *Cladonia* ..... ***S. crassitunicatum* Zhurb., Diederich & U. Braun**
3. Conidia less than 7 µm long, with a thinner wall **4**

4. Conidiomata 50–100 µm diam., not confluent; conidia medium to dark brown, mostly 5–6.5 × 4–5 µm when 1-septate, cells easily separating in part-conidia; on *Fissurina* (Diederich 2015) ..... ***S. aptrootii* Diederich**
4. Conidiomata (50–)100–300 µm diam., often confluent; conidia paler, 4–7 µm diam. when 1-septate, not separating in part-conidia; on corticolous *Pertusaria* (Hawksworth 1979) ... . ***S. simplex* D. Hawksw.**

5. Conidiomata 100–200(–250) µm diam.; conidia aseptate, mostly 7–9 × 5–6 µm, or 1-septate, 11–13.5 × 5–6.5 µm, outer wall splitting at maturity, giving a mosaic-like ornamentation; on *Anomomorpha* (Diederich 2015) ..... ***S. sipmanii* Diederich**
5. Conidiomata up to 600 µm diam.; conidia aseptate, 5–8 × 4.5–6 µm, with a verrucose wall; on *Bellemerea diamarta* (Alstrup 1993) ..... ***S. verrucisporum* Alstrup**

6 Conidia smooth-walled **7**
6 Conidial wall ornamented **14**

7 Conidia 2-celled **8**
7 Conidia mostly more than 2-celled **9**

8 Conidiomata 200–300 µm diam.; conidia mainly 2-celled, occasionally aseptate or submuriform, wall occasionally fissured, 10–13 × 6–9 µm; on *Lecanora rupicola* (Hafellner 1996) ..... ***S. montagnei* Hafellner**
8 Conidiomata larger at maturity, 200–600 µm diam.; conidia consistently 2-celled, smooth-walled, 11–14.5 × 9–11 µm; on *Phaeophyscia orbicularis* ....***S. phaeophysciae* Diederich & van den Boom**

9 Conidiomata 45–180 µm diam. **10**
9 Conidiomata 150–500 µm diam. **11**

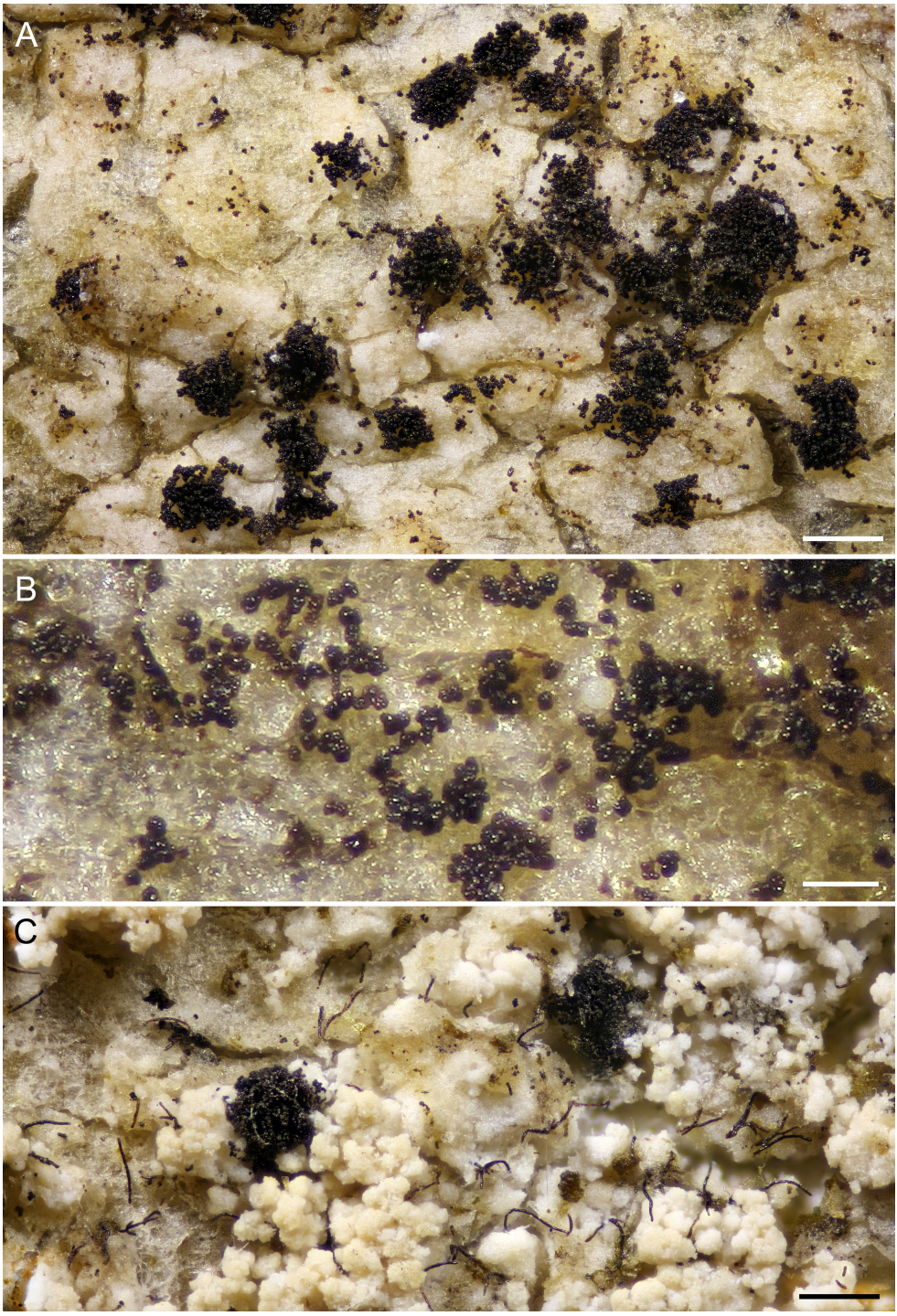

Fig. 3. *Sclerococcum toensbergii* (A–B, holotype; C, *Tønsberg* 21577). A, Sporodochia developing on the thallus of *Pertusaria carneopallida*. B, Conidia spreading around sporodochia. C, Sporodochia developing on the thallus of *Megalaria pulverea*. Scale bars: A, C = 200 µm; B = 50 µm.

10 Conidia 2–14(–20)-celled, with irregular, well-delimited darker regions due to an unevenly thickened cell wall; conidiomata 60–180 µm diam.; on *Parmelina* (Boqueras & Diederich 1993).....***S. serusiauxii* Boqueras & Diederich**

10 Conidia 2–50-celled, with an evenly thickened cell wall; conidiomata 43–145 µm diam.; on *Cladonia* (Zhurbenko & Pino-Bodas 2017)..... .....***S. epicladonia* Zhurb.**

11 Conidia 2–6-celled; conidiomata 170–500 µm diam.....**12**

11 Conidia more than 6-celled when mature; conidiomata 150–250 µm diam. **13**

12 Conidiomata 170–350 µm diam., dark brown to black; conidia in surface view (×40) hardly visible, slightly shiny, often arranged in groups of 30–70 µm diam.; conidial cells (4–)6–10 µm diam.; on *Pertusaria corallina*, rarely

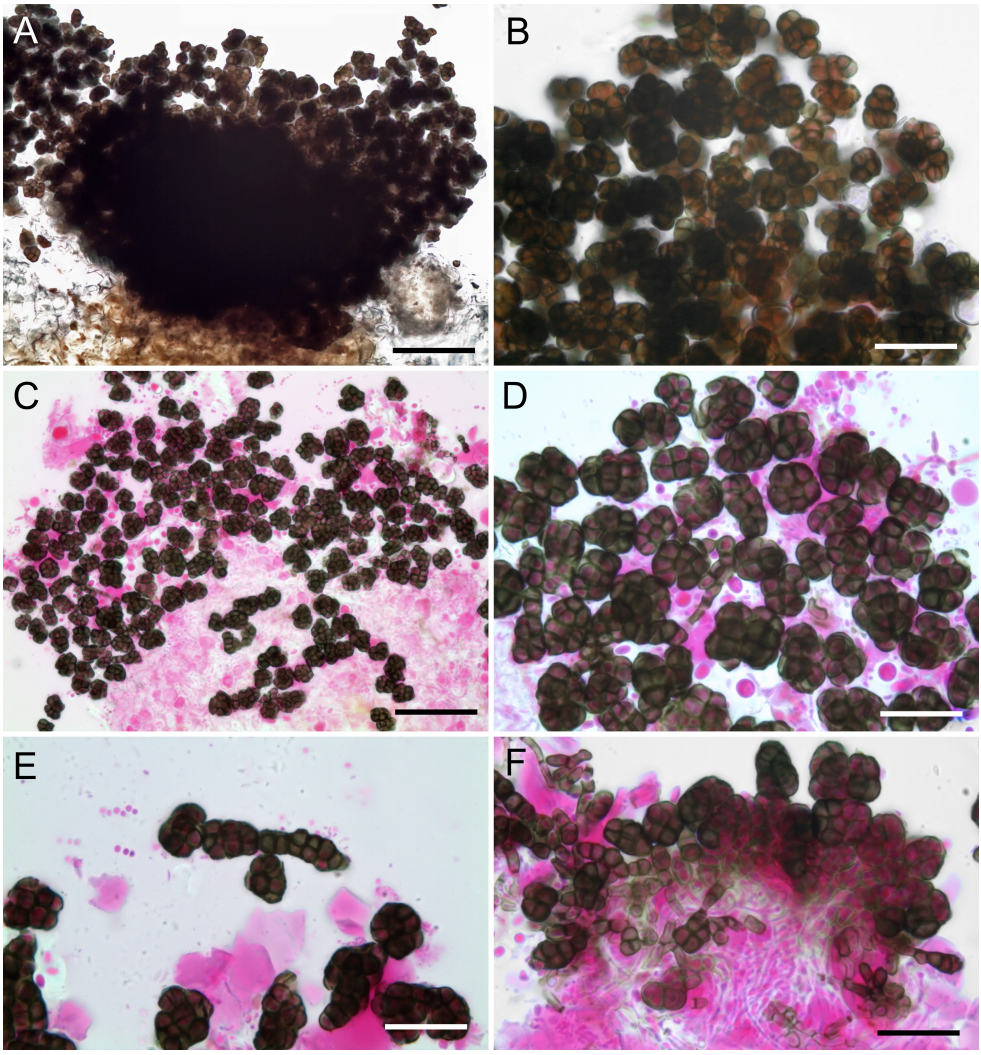

Fig. 4. *Sclerococcum toensbergii* (A–B, *Tønsberg* 21577; C–F, holotype). A, Section through conidioma. B–D, Sections through conidiomata, with conidiophores separated through pressure on cover glass. E, Catenate conidia. F, Conidiophores and conidia. (A–B, in water; C–F, in Phloxine B after K pre-treatment). Scale bars: A, C = 50 µm; B, D–F = 20 µm.

other saxicolous *Pertusaria* spp. (Hawksworth 1975) ..... ***S. sphaerale* (Ach.) Fr.**

12 Conidiomata 225–500 µm diam., dark reddish brown; conidia in surface view granular, easily visible, matt, not arranged in groups; conidial cells 3–6 µm diam.; on *Varicellaria hemisphaerica* (Diederich 1990) ..... ***S. epiphytorum* Diederich**

13 Conidia 13.5–18.8 × 10–14.5 µm, up to 20-celled when mature; on *Megalaria pulverea* and *Pertusaria carneopallida* .... ***S. toensbergii* Diederich**

13 Conidia 11–30 × 10–20 µm, up to 60-celled when mature; on *Phyllobaeis* (Etayo 2017)..... ..... ***S. phyllobaeis* Etayo**

14 Conidia 10–15 × 7–13 µm, greenish brown, wall verruculose; conidiomata 100–400 µm diam.; on *Buellia aethalea* (Diederich & Scholz 1995)..... ***S. leuckertii* Diederich & Scholz**

14 Conidia not greenish..... **15**

15 Conidiomata 50–150 µm diam., finally concave; conidia dark brown, 2–4-celled, 11–21 × 8–15 µm, wall verrucose-fissured; on *Tephromela atra* (Etayo & Calatayud 1998) ... . ***S. tephromelarum* Etayo & Calatayud**

15 Conidiomata at least 200 µm diam.; conidia with an ornamentation of polygonal areoles..**16**

16 Conidia blue-grey when young, later dark brown, 2–5-celled, 8–12.5 × 6.5–8 µm, irregularly covered by dispersed, thick, almost squamulose areoles; conidiomata 250–300 µm diam.; on *Placopsis gelida* (Berger 2000) ..... ***S. gelidarium* Etayo & F. Berger**

16 Conidia brown to black, 2–3-celled, 10–12 × 5.5–8 µm, covered by thinner areoles; conidiomata 200–400 µm diam.; on cf. *Xylographa* (Etayo & Sancho 2008).....***S. areolatum* Etayo**

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