Aabaarnia and Normanogalla, two new lichenicolous genera of Ostropales, Ascomycota

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Abstract. The new genus and species Aabaarnia siphulicola is described for a lichenicolous, gall-inducing ascomycete growing on Siphula decumbens in Australia (Tasmania) and New Zealand (Auckland Islands). The species is characterized by pale, immersed, cleistohymenial ascomata, an ascomatal wall without crystals, missing periphysoids, a K/I+ blue hymenium, subcylindrical, 4–6-spored, K/I– asci with a massive apical cap, and oval to short-cylindrical, 3-septate ascospores. The new genus Normanogalla is described for the lichenicolous, gall-inducing N. cribriformis (previously known as Unguiculariopsis cribriformis), known on terricolous Pertusaria in Scandinavia and Greenland. It is characterized by pale, immersed, cleistohymenial ascomata, an ascomatal wall without crystals, abundant periphysoids, a K/I– hymenium, subcylindrical, 8-spored, K/I– asci with an apically not or slightly thicker wall, and subspherical, aseptate ascospores.

Introduction

The ostropalean fungi, as circumscribed by Sherwood (1977), included ascomycetes with apothecioid (rarely perithecioid) ascomata that are usually immersed and closed when young, opening by an irregular pore or a slit (cleistohymenial ascomatal development), a hamathecium of true paraphyses, periphysoids often present, subcubylindrical asci that are thick-walled when young, but functionally not bitunicate, with a distinct I– apical thickening, long-cylindrical to filiform ascospores and an ascomatal wall frequently with crystalline inclusions (Ostropales s.str.), or ovoid ascospores or wall without crystalline inclusions (Ostropales s. lat.). In addition to the Stictidaceae, Hawksworth & Sherwood (1982) described a second family of Ostropales, the Odontotremaeae, that were eventually revised by Sherwood-Pike (1987). One genus of Stictidaceae, Nanostictis, and three genera of Odontotremaeae, Odontotrema, Paralethariicola and Spirographa, included obligate lichenicolous species.

Recent molecular studies allowed enlarging the concept of Ostropales, which now also include the Coenogoniaceae, Gomphillaceae, Graphidaceae, Gyalectaceae, Myelotremidaeae, Phaneromycetaceae, Phlyctidiaceae, Porinaceae and Thelotremataeae (Lumbsch & Huhndorf 2007). On the other hand, Baloch et al. (2013) showed that Odontotrema (Odontotremaeae) is polyphyletic, and most taxa, including all lichenicolous species, were transferred to Sphaeropezia (Stictidaceae).

In this paper we describe two new genera of lichenicolous ascomycetes that we include in the Ostropales. As the type species of both have rarely been collected, and as no recent collections are available, no molecular data could be obtained, hence they cannot be attributed to a family at this moment.

Material and Methods

Material from BR and UPS has been examined. Dry herbarium specimens were examined and measured under a binocular microscope Leica MZ 7.5 (magnification up to 50×). Macroscopic photographs were done using a Canon 40D camera with a Canon MP-E 65 mm macro lens, Stack
Shot (Cognisys) and Helicon Focus (HeliconSoft) for increasing the depth of field. Hand-made sections of ascomata were studied in water, 10% KOH, Congo Red and Lugol’s iodine, without (I) or with (K/I) pretreatment with KOH. Microscopic photographs were prepared using a Leica DMLB microscope with DIC optics and a Leica EC3 camera. Measurements based on statistical data are indicated as (min.−)X−σ−X+σX−max., followed by the number of measurements (N).

Fig. 1. Aabaarnia siphulicola (holotype). Ascomata immersed in pinkish galls of Siphula decumbens. Scale bar: 1 mm.
Fig. 2. Aabaarna siphulicola (holotype). A, Section through closed, immature ascoma, in water. B, Section through immature, opened ascoma in lactophenol-cotton blue. C, The same, in polarized light, showing crystals of host thallus present in the ascomatal wall. D, Section through mature ascoma, in 10% KOH. E, The same, in polarized light, with crystals absent in ascomatal wall. F, Magnification of B, showing hamathecium near ostiole in immature ascoma. G, Section through mature ascoma, filled with asci containing ascospores (DIC). H, The same, showing reaction of Lugol's reagent entering hamathecium, after pre-treatment with 10% KOH. I, Mature ascospores in 10% KOH, showing perispore. J, Mature ascospores in water (DIC), showing perispore. K, Asci (DIC). L, Ascus and paraphyses (DIC). M, Asci in Lugol's reagent after pre-treatment with 10% KOH. Scale bars: A–E, G–H = 50 µm; F, I–M = 10 µm.
Results

*Aabaarnia siphulicola* Diederich gen. et sp. nov. (Figs 1–2)

Characterized by pale, immersed, gall-inducing, cleistohymenial ascomata that are hyaline, K– in section, c. 100–200 µm diam., an ascomatal wall without crystals, missing periphysoids, a K/I+ blue hymenium, subcylindrical, 4–6-spored, K/I– asci, 70–110 (–140) × 11–14 µm, with a massive apical cap, and oval to short-cylindrical, 3-septate ascospores, (23.0–)24.4–28.0 (–29.0) × (7.0–) 7.4–8.3 (–8.5) µm, ratio length/width (3.0–) 3.1–3.6 (–4.0) [N = 20], wall 0.5–1 µm thick, perispore present, hyaline, appearing verrucose with DIC optics, c. 1 µm thick, in KOH up to 2–3 µm thick. Pycnidia unknown.

**Distribution and host.** The species is known from Australia (Tasmania) and New Zealand (Auckland Islands: Auckland Island and Rose Island), on the thallus of terricolous *Siphula decumbens*, gall-inducing.

**Observations.** The new genus is distinguished from most ostropalean genera treated by Sherwood (1977) by the ellipsoid, not filiform ascospores, and by the absence of crystals in the ascomatal wall. The most similar genera keyed out by this author are *Absconditella* and *Cryptodiscus*. *Absconditella Vězda* is distinguished by being always lichenized, never lichenicolous, a hymenium that is always K/I–, and ascospores lacking a distinct perispore (Coppins 2009). *Cryptodiscus Corda* is distinguished by a different ecology (saprotrophic and immersed in dead wood, or lichenized), deep urceolate ascomata, the margin being always strongly elevated compared to the ascomatal disc (in *Aabaarnia*, the hymenium is almost as tall as the ascomatal margin), paraphyses that often present an abruptly enlarged tip, and thin-walled ascospores lacking a distinct perispore.

The lichenicolous genera *Sphaeropezia* Sacc. and *Paralethariicola* Calatayud, Etayo & Diederich, two genera with similar oval to short-cylindrical, transversely septate ascospores, mainly differ by dark, often superficial ascomata and a distinct periphysoid layer (Baloch et al. 2013, Diederich & Etayo 2000, Calatayud et al. 2001). *Spirographa Zahlbr.* has dark brown ascomata, indistinct periphysoids, 16–32-spored asci and acicular, helicoidal, 1-septate ascospores (Diederich 2004).

The type of *Nanostictis* M. S. Christ., *N. peltigerae* M. S. Christ., and several subsequently described species differ by ascomata with an exposed disc when mature, a much thicker lateral ascomatal wall (= part of the

Lichenicolous, inducing gall formation on host thallus. **Galls** 2–4 mm diam., basally strongly constricted, slightly more pinkish than the host thallus, containing (10–)40–100 ascomata that are initially completely immersed and thus only visible in section, later become exposed through breakup of covering host thallus layer, resulting in roundish or irregular ‘pores’ of the host thallus of 20–80 µm diam., through which part of the ascomatal disc and exceptionally part of the margin can be seen. **Ascomata** initially entirely closed (cleistohymenial development) and subspherical, c. 100 µm diam., later opening by a breakup of the upper ascomatal wall, c. 100–200 µm diam. (difficult to measure without destroying the gall, as they remain immersed and almost entirely covered by host tissues at maturity); disc and margin pale brown, margin 20–40 µm thick, not breaking away from the hymenium when dry; hairs absent. **Ascomatal wall** hyaline, K–, basally and laterally 55–65 µm thick, of cells 3–6 µm diam., without crystals (but sometimes filled with crystals from the host thallus). **Periphysoids** absent. **Hymenium** hyaline, filling the entire ascomatal cavity, 130–170 µm tall, I–, K/I+ blue. **Epiphyllum** hyaline, K–. **Subhymenium** hyaline, 5–10 µm thick. **Paraphyses** septate, simple or occasionally branched, 1.7–2.5 µm thick, apically not or slightly swollen. **Asci** subcylindrical, uniloculate, wall laterally thin, with a massive apical cap, entirely I–, K/I–, 4–6-spored, 70–110 (–140) × 11–14 µm. **Ascospores** 1–2-seriate, oval to short-cylindrical, 3–septate (first septum median), slightly constricted near septa, hyaline, multiguttulate, (23.0–)24.4–28.0 (–29.0) × (7.0–) 7.4–8.3 (–8.5) µm, ratio length/width (3.0–) 3.1–3.6 (–4.0) [N = 20], wall 0.5–1 µm thick, perispore present, hyaline, appearing verrucose with DIC optics, c. 1 µm thick, in KOH up to 2–3 µm thick. Pycnidia unknown.
ascomatal wall initially covering the hymenium in young, closed ascomata), a hymenium and asci that are K/I–, unbranched paraphyses with an enlarged tip and filiform or acicular ascospores (Christiansen 1954, Etayo & Sancho 2008). Etayo & Sancho (2008) keyed out all known Nanostictis species and described a new species with ellipsoid, 3-septate ascospores, *N. nephromatis* Etayo. That species differs from *Aabaarnaia* by a different ascus type (wall apically I+ and K/I+ blue, with a different structure, as illustrated by Etayo & Sancho, 2008 in fig. 66C) and thin-walled ascospores without a perispore.

*Etymology.* The new genus is named after my friend André Aptroot, an extremely productive and tireless explorer of lichens, who collected the new species *Aabaarnaia siphulicola* in Tasmania. The generic name is formed by the initials of his name (A. A.), followed by the name of the city Baarn in the Netherlands, where André was professionally active over many years.


**Normanogalla** Diederich gen. nov.

Characterized by pale, immersed, gall-inducing, cleistohymenial ascomata that are hyaline, K– in section, an ascomatal wall without crystals, abundant periphysoids, a K/I– hymenium, subcylindrical, 8-spored, K/I– asci with an apically not or slightly thicker wall, and subspherical, asceptate ascospores.

**Type:** *Normanogalla cribriformis* (Norman) Diederich. Mycobank MB 810085

**Observations.** By the cleistohymenial ascomatal development and the abundant periphysoids, the new genus clearly belongs to the Ostropales. An assignment to a family based on morphological characters only is hardly possible for these fungi (Baloch et al. 2013), and molecular data are therefore needed.

Amongst the lichenicolous genera currently attributed to the Ostropales, families Odontotremaeaceae and Stictidaceae, *Sphaeropoezia* is distinguished by blackish ascomata, a much thicker and darker ascomatal margin and septate ascospores (Baloch et al. 2013, Diederich & Etayo 2000). *Para lethariicola* is similar but differs furthermore by asci with a hemiamyloid apical ring, the presence of excipular hairs, and curved or slightly helicoid ascospores (Calatayud et al. 2001). *Spirographa* has dark brown ascomata, a yellowish brown ascomatal wall, indistinct periphysoids, 16–32-spored asci and acicular, helicoid, 1-septate ascospores (Diederich 2004). *Nanostictis* differs by having septate, often filiform ascospores and a more massive ascomatal margin (Christiansen 1954). The poorly known *Stictis cladonias* (Rehm) Sacc., with simple, filiform ascospores appears to have bitunicate asci, which excludes it from the Ostropales (Sherwood 1977).

We also compared the new genus with the genera of Ostropales treated and keyed out by Sherwood (1977). The most similar genera, following that key, are *Cryptodiscus* (syn. *Gloeolecta*), *Melittosporiella* and *Ramonia*. *Cryptodiscus* differs by being saprotrophic on dead wood or lichenized, a K/I+ blue hymenium, and a K/I+ blue ascus wall (Baloch et al. 2009). *Melittosporiella* Höhn. differs by much larger ascomata (1–2 mm diam.) with a flattened base (ascomata not subspherical), with a margin breaking away from the hymenium when dry, and by transversely septate ascospores (Sherwood 1977). *Ramonia* Stizenb. is rather variable and comprises numerous species, keyed out by Lendemer & Knudson (2008). All known species are distinctly or weakly lichenized with *Trentepohlia*, and no lichenicolous species are known. The hymenium is always I+ bluish at least in uppermost part (Sanderson & Purvis 2009). The generic type, *R. valenzueliana* (Mont.) Stizenb. differs by polysporous asci and 1-septate ascospores. There is no species known with 8-spored asci and simple ascospores. The main difference of both *Ramonia* and *Cryptodiscus* seems to be the periphysoidal layer that is ‘highly modified, consisting, not of distinct filaments, but of
a compact, fleshy, pseudoparenchymatous matrix in which the cells are oriented perpendicularly to the surface of the ascocarp (Sherwood 1977: 17), whereas in *Normanogalla*, this layer consists of distinct periphysoids.

Fig. 3. *Normanogalla cribriformis* (Vrang, UPS). Ascomata immersed in galls of *Pertusaria dactylina*. Scale bars: A = 1 mm, b = 0.5 mm.
Fig. 4. Normanogalla cribriformis (Vrang, UPS). A, Section through closed ascoma, showing hymenium (left, bottom and right) and periphysoid layer (top). B, Periphysoids (centre) and paraphyses (right) in squash preparation. C, Section through opened ascoma, with periphysoid layer along the ascending margins. D, Hymenium and subhymenium, with periphysoid layer visible on the top left. E, Asci. F, Ascospores. [C, D: in water. A, B, F: in a mixture of Congo Red and 10% KOH. E: in Lugol after KOH-pretreatment. A, B, D, F: DIC optics.]. Scale bars: A, C = 50 µm; B, D = 20 µm; E–F = 5 µm.
Normanogalla cribriformis (Norman) Diederich comb. nov. (Figs 3–4)


Pertusaria paradoxa Linds., Trans. Linn. Soc. Lond. 27: 344 (1871). Type: Greenland, on Pertusaria oculata (BM?). [Alstrup & Hawksworth (1990) stated that this name is a synonym of Unguiculariopsis cribriformis, without reporting if they examined the type; on page 4, they explained that they have examined types of two of four species described by Lindsay from Greenland, without saying if P. paradoxa was amongst them.]

Mycobank MB 810087

Lichenicolous, inducing gall formation on host thallus. Galls 1–3 mm diam., basally not constricted, containing 10–80 ascomata that are initially completely immersed and thus only visible in section, later become exposed through breakup of covering host thallus layer, resulting in roundish or irregular ‘pores’ of the host thallus of 40–100 µm diam., through which part of the ascomatal disc and exceptionally part of the margin can be seen. Ascomata initially entirely closed (cleistohymenial development) and subspherical, c. 100 µm diam., later opening by a breakup of the upper ascomatal wall, probably opening at the same time as the breakup of the covering host thallus, c. 100–150 µm diam. (difficult to measure without destroying the gall, as they remain immersed and partially covered by host tissues at maturity); disc and margin pale brown, margin 20–25 µm thick, not breaking away from the hymenium when dry, without crystals; hairs absent. Ascomatal wall hyaline, K–, basally and laterally 13–25 µm thick, in the upper part of young closed ascomata 20–25 µm thick, composed of elongate cells 5–12 × 4–4.5 µm (in optical section). Periphysoids abundantly covering the inner cavity of the upper ascomatal wall when young, after breaking of this wall present on the inner layer of the ascomatal margin, 25–35 µm long, often with one ramification, with several septa, apical cells 3–4 µm thick, basal cells up to 5 µm thick, periphysoid layer hyaline or with a pale olivaceous pigment, K–. Hymenium hyaline, 52–70 µm tall, I–, K/I–. Epiphyllum hyaline or with a pale olivaceous tinge, K–. Subhymenium hyaline, c. 5 µm thick. Paraphyses septate, simple, 2.5–3.2 µm thick, apically not or slightly swollen. Asci subcylindrical, unitunicate, wall laterally thin, apically slightly thickened, I–, K/I–, 8-spored, 40–58 × 5–7.5 µm. Ascospores uniseriate, subhedral, with a pale olivaceous tinge, not guttulate (only old, dead spores examined), (4.0–)4.5–5.7(–6.0) × (3.5–)4.1–5.0(–5.5) µm, ratio length/width (1.0–)1.0–1.2(–1.3) [N = 25], wall 0.5–0.8 µm thick, perispore absent. Pycnidia unknown.

Distribution and host. The species is known from Greenland (Alstrup & Hawksworth 1990), Norway and Sweden, on the thallus of terricolous Pertusaria dactyлина, more rarely on P. oculata, gall-inducing.

Observations. This species was poorly known until Alstrup & Hawksworth (1990) studied type material and combined it in Unguiculariopsis. No description was provided, however, by these authors. The new combination was not validly published as the basionym was omitted. The decision to combine the species in Unguiculariopsis was based on the supposed presence of excipular hairs, illustrated in fig. 44 C–D, which represent in reality periphysoids. Our discovery of the cleistohymenial ontogeny of the ascomata allowed excluding the species from Unguiculariopsis and the Helotiales. The species has very rarely been collected, and the most recent specimen dates from 1951 (Alstrup & Hawksworth 1990). It should be carefully searched for in arctic environments on the known hosts, and the discovery of fresh material should allow DNA sequencing.

Specimens examined (all on Pertusaria dactyлина):

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References


