

Standard Paper

Clypeococcum wedinii (Dothideomycetes), a new lichenicolous fungus on *Bunodophoron*, with an updated key to species of *Clypeococcum*

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Abstract

Clypeococcum wedinii, a pyrenocarpous ascomycete growing on the lichen genus *Bunodophoron* in Chile and New Zealand, is described as new to science. It differs from other *Clypeococcum* species in the combination of the following characteristics: a conspicuous gall formation, scattered, loose to dense stromatic growths sometimes looking like a clypeus, an ascomatal wall composed of both *textura intricata* and *angularis* in surface view, a non-amyloid hymenium, an absence of ostiolar filaments, 8-spored asci, and 1-septate ascospores arranged biserially in the ascus. An updated key to the species of the lichenicolous genus *Clypeococcum* is provided.

Key words: lichen-dwelling fungi, *Polycoccaceae*, Southern Hemisphere, taxonomy

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Introduction

While inspecting the collection of unidentified lichenicolous fungi of the late Rolf Santesson, preserved in the herbarium of the Museum of Evolution at Uppsala University (UPS), I came across an interesting specimen of a pyrenocarpous ascomycete from Chile annotated as '*Polycoccum* sp.' that causes conspicuous galls on the thallus of the lichen *Bunodophoron ramuliferum* (I.M. Lamb) Wedin. A search for similar material among the *Bunodophoron* A. Massal. specimens stored in UPS uncovered another specimen of this fungus from New Zealand. Subsequent examination of these specimens showed that they represent a species from the lichenicolous genus *Clypeococcum* D. Hawksw. that is unknown to science. This paper aims to describe this new species and provide an updated key to the 15 accepted species of *Clypeococcum*.

Materials and Methods

Microscopy was carried out and images were captured using a Zeiss Axio Zoom V16 microscope and a Zeiss Axio Imager A1 microscope equipped with Nomarski differential interference contrast optics and fitted with a Zeiss AxioCam MRc5 digital camera. Microscopic characters were studied using sections hand-cut with a razor blade and mounted in water, 10% potassium hydroxide (K), Lugol's iodine directly (I) or after a K pretreatment (K/I), or concentrated nitric acid (N). Measurements were taken

from water mounts and rounded to the nearest 0.5 μm . The length, width, length/width ratio (l/w) and upper cell length/lower cell length ratio of the ascospores, as well as the diameter of the ascomata are given as (min–) (\bar{x} – SD) – (\bar{x} + SD) (–max), where 'min' and 'max' are the extreme values observed, \bar{x} the arithmetic mean and SD the corresponding standard deviation. Specimens examined are housed in LE and UPS.

Specimens examined for comparison. *Clypeococcum cladonema* (Wedd.) D. Hawksw. (type of *Clypeococcum*). **Russia:** Krasnodar Territory: on *Flavoparmelia caperata* (L.) Hale, 2014, M. P. Zhurbenko 14326a (LE 309410a).

Didymocyrtis consimilis Vain. (type of *Didymocyrtis* Vain.). **Russia:** Republic of Adygeya: on *Parvoplaca tiroliensis* (Zahlbr.) Arup *et al.*, 2014, M. P. Zhurbenko 14431 (LE 309489).

Polycoccum tryptethelioides (Th. Fr.) R. Sant. (type of *Polycoccum* Körb.). **Norway:** Troms County: on *Stereocaulon glareosum* (Savicz) H. Magn., 2003, M. P. Zhurbenko 0383 (LE 260505).

Verrucococcum spribillei V. Atienza *et al.* **USA:** Alaska: on *Lobaria pulmonaria* (L.) Hoffm., 2001, M. P. Zhurbenko 01197 (LE 233929).

The new species

Clypeococcum wedinii Zhurb. sp. nov.

Mycobank No.: MB 844870

Differs from *Clypeococcum galloides* Etayo mainly by its non-amyloid hymenium, 8-spored asci, larger ascospores (14–)18–22 (–25) \times (5–)6.5–8(–9) μm , and host preference (*Bunodophoron*).

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Type: New Zealand, Southland, Fiordland National Park, Lake Haurako, Oblong Hill, along Look Out Track, open *Nothofagus solandri*-*Weinmannia* dominated forest, on a dead trunk of *Podocarpus* sp., 45°59'S, 167°22'E, elev. 360 m, on thallus of *Bunodophoron notatum*, 21 January 1990, M. Wedin 2947 (UPS 56614—holotype).

(Fig. 1)

Lichenicolous ascomycete growing on species of *Bunodophoron* and inducing conspicuous galls. *Vegetative hyphae* light to medium brown, 2–4.5 µm diam. *Galls* subglobose, 250–2600 µm diam., initially the same colour as the host thallus, later getting darker, becoming constricted at the base and strongly tuberculate, mottled with protruding black tops of the parasitic ascomata/clypei. The interior of the galls is filled with parasitic ascomata (up to 100 or possibly more) interspersed with loose to dense stromatic growths in the form of strands or more massive aggregations between/around the ascomata mixed with host tissues.

Ascomata perithecioid, black in the exposed parts, subglobose, not beaked, (40–)90–180(–210) µm diam. ($n = 69$), completely immersed in the interior of the galls or slightly protruding above their surface in the ostiolar area, with ostiole *c.* 20 µm diam., dispersed to aggregated, sometimes contiguous with a small number merged together, often linked by brown plurihyphal stromatic strands, 10–55 µm wide, or united in groups of up to 10 or more by more extensive and dense stromatic growths that sometimes form clypeus-like structures (Fig. 1F). *Ascomatal wall* in surface view composed of interwoven hyphae (*textura intricata*) or pseudoparenchymatous cells (*textura angularis*), in cross-section light brown (inside) to medium or dark brown (outside), with the pigment deposited in the hyphal cell walls, K+ partly greyish, N–, without warts on the outside, mainly 10–30 µm wide; composed of *c.* 8–12 layers of roughly isodiametric or slightly tangentially elongated cells 1.5–5.5 µm long with walls 0.5–3 µm wide, in places passing into stromatic growths and widening to 80 µm, especially between and above adjacent ascomata, forming in the latter case clypeus-like structures. *Hymenium* not interspersed, I–, K/I–. *Hamathecium* composed of distinct, persistent, hyaline, filiform, branched, occasionally anastomosing, septate, short-celled interascal filaments that are not swollen at the apices, 1.5–2.5 µm wide; ostiolar filaments absent. *Asci* bitunicate in structure, roughly subcylindrical, slightly widened in the middle or lower half (obclavate), rounded at the apex, sometimes with an apical beak, short-stalked, 65–80 × 15–18 µm ($n = 8$), 8-spored, I–, K/I– except for ascoplast turning orange. *Ascospores* light to occasionally medium brown when mature, K+ brownish grey, very narrowly obovate, rounded at the ends, 1-septate; upper cell wider, often longer and rarely slightly darker than the lower one, upper cell length/lower cell length = 1–1.4(–1.6) ($n = 23$), not or only slightly constricted at the septum, (14–)18–22(–25) × (5–)6.5–8(–9) µm, l/w = (2.1–)2.4–3.2(–4.0) ($n = 144$), occasionally with 1–2 conspicuous guttules in each cell, finely verruculose with verrucae *c.* 0.5 µm diam.; when immature rarely with a gelatinous sheath to 1.5 µm wide observed in K, biseriolate and mostly overlapping in the ascus.

Conidiomata not observed.

Etymology. The new species is named after the eminent Swedish lichenologist Mats Wedin, monographer of the family *Sphaerophoraceae* in the Southern Hemisphere (Wedin 1995), who collected the holotype of the new species.

Distribution and hosts. The new species is known from two specimens collected in Chile and New Zealand, therefore belonging to the Neotropical and Australasian biogeographical realms. It was collected on thalli of *Bunodophoron notatum* (Tibell) Wedin and *B. ramuliferum* (Lecanorales, *Sphaerophoraceae*). The former species occurs in Australia, Tasmania and New Zealand, and the latter in Australia, Tasmania, New Zealand, southernmost South America, Juan Fernandez and the Falkland Islands (Wedin 1995). Apart from gall-formation, the harmful effects on the hosts are not noticeable, with their surrounding parts retaining their normal coloration.

Notes. With its lichenicolous lifestyle, black, subglobose, ostiolate perithecioid ascomata, often united by stromatic growths, ascomatal wall of both *textura intricata* and *angularis* in surface view, I–, K/I– hymenium, hamathecium composed of persistent, filiform, branched and anastomosing interascal filaments, bitunicate, roughly subcylindrical, 8-spored asci, and 1-septate, brown, verruculose ascospores, biseriolate in the ascus, the examined fungus is most consistent with the protologue of the genus *Clypeococcum* (*Polycoccaceae*, *Dothideomycetes*) typified on *C. cladonema* (Hawksworth 1977). The only difference from this protologue, although significant, is the absence of a persistent distinct clypeus, but the fungus occasionally develops clypeus-like structures. Morphologically, the new species also strongly resembles pyrenocarpous lichenicolous taxa belonging to *Didymocyrtis*, *Polycoccum* and *Verrucococcum* V. Atienza *et al.* However, *Didymocyrtis* and *Polycoccum* differ in the absence of stromatic ascomata and the presence of exclusively *textura angularis* tissue in the ascomatal wall; the former is additionally distinct in its narrowly cylindrical asci and uniseriate ascospores (Hawksworth & Diederich 1988; Ertz *et al.* 2015). *Verrucococcum hymeniicola* (Berk. & Broome) D. Hawksw. *et al.* and *V. spribillei* also form tightly packed ascomata surrounded by stromatic tissue. However, their ascomata are exposed in the upper two-thirds with age, and the ascomatal wall is made exclusively of *textura angularis* with a warted surface (Atienza *et al.* 2021).

Considering other genera of ascomycetes characterized by a lichenicolous lifestyle, closed ascomata and stromatic growths, it is appropriate to compare *Clypeococcum wedinii* with *Homostegia* Fuckel, *Lasiosphaeriopsis* D. Hawksw. & Sivan, *Lichenostigma* Hafellner, *Macrographa* Etayo, *Perigrapta* Hafellner, *Plectocarpon* Fée, *Plowrightia* Sacc. and *Saania* Zhurb. They are all characterized by their well-developed, usually superficial stromata. *Lichenostigma* is also distinguished by its asci arising directly within the stromatic pseudoparenchyma (Calatayud *et al.* 2004). Other genera have perithecial locules that are completely enclosed by stromatic tissue, whereas in *Clypeococcum wedinii* only some of such locules are surrounded by a stroma. Additionally, *Homostegia* differs in the wall structure of the perithecial locules (*textura angularis*) and its 3-septate ascospores (Hawksworth *et al.* 2004); *Lasiosphaeriopsis* is distinguished by the presence of periphyses, the absence of interascal filaments, unitunicate asci and perithecial locule walls with Munk pores (Hawksworth 1980b); *Macrographa* is characterized by abundant periphysoids and hyaline ascospores (Etayo & Sancho 2008); *Perigrapta* and *Plectocarpon* are well distinguished by their K/I+ blue hymenial gel and asci with a K/I+ blue apical ring (Hafellner 1996; Ertz *et al.* 2005; Zhurbenko & Ohmura 2018); *Plowrightia* differs in the absence of interascal filaments and in its hyaline ascospores (Clauzade *et al.* 1989; Thambugala *et al.* 2014); and *Saania* is distinguished by its N+ red ascomatal

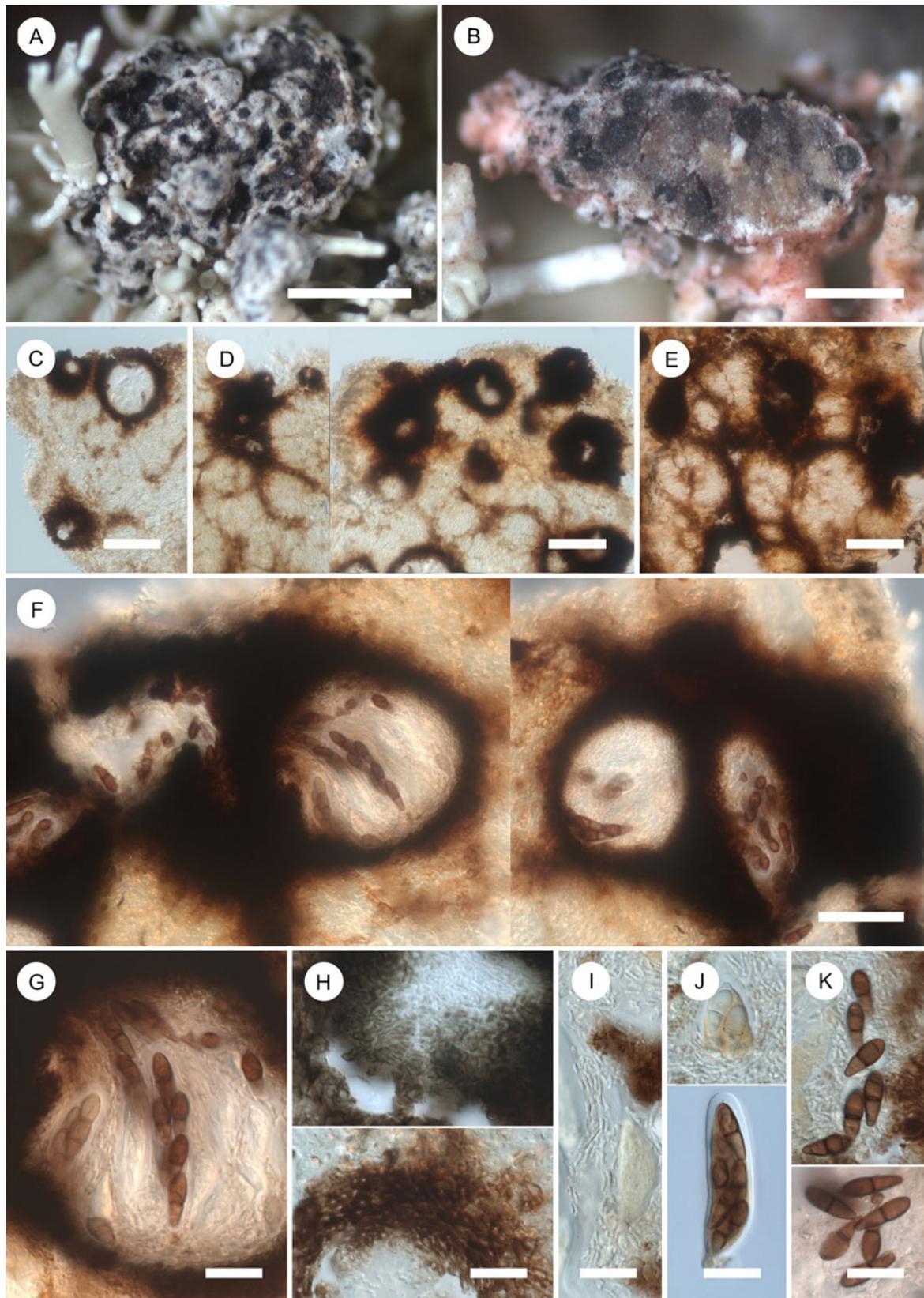


Fig. 1. *Clypeococcum wedinii* (A–C & K (below), holotype; D–J & K (above), UPS F-8925990). A, final stage of gall development. B, section through a gall showing immersed ascomata. C–E, galls with immersed ascomata and varying degrees of stromatic development in cross-section, in water. F, ascomata and clypeus-like stromatic growths in cross-section, in water. G, ascoma with asci, ascospores and interascal filaments in cross-section, in water. H, ascomatal wall in surface view, in K (above) and in cross-section, in water (below). I, interascal filaments, in I. J, asci, in I. K, ascospores in I (above) and water (below). Scales: A = 1 mm; B = 500 μ m; C–E = 100 μ m; F = 50 μ m; G–K = 20 μ m. In colour online.

wall and hamathecium, consisting only of periphysoids (Motiejūnaitė *et al.* 2019).

In the genus *Clypeococcum*, 15 species are accepted to date; all are lichen-inhabiting and rather highly selective concerning their hosts, 12 of them being confined to one lichen genus, two confined to two genera in the same family, and one to three genera in the same family (see key below). Eight of the 15 known species of *Clypeococcum*, including *C. wedinii*, are

restricted to lichens of the order *Lecanorales*, while *C. wedinii* is the first species of this genus found on members of the family *Sphaerophoraceae*, to which the genus *Bunodophoron* belongs.

Additional specimen examined. **Chile:** on *Bunodophoron ramuliferum* (thallus), leg. *Poeppig*, det. Kunze 1842, hb. Tuck, sheet no. 3746 (UPS F-8925990).

An updated key to the species of *Clypeococcum*

The previous key for *Clypeococcum* included nine species (Pirogov 2015). Subsequently, six further species have been described, making it appropriate to present an updated key for the 15 currently known in this genus. This key is based on the literature cited below under 'Lit.'

- | | | |
|--------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|---------------------------------------------------------------------|
| 1 | Ascospores muriform; on <i>Solorina</i> Ach. Lit.: Zhurbenko (2020) | C. lenae Zhurb. |
| | Ascospores 1-septate | 2 |
| 2(1) | Galls present | 3 |
| | Galls absent | 5 |
| 3(2) | Galls up to 2.6 mm diam., hymenium I–, asci 8-spored, ascospores (14–)18–22(–25) × (5–)6.5–8(–9) μm; on <i>Bunodophoron</i> .
Lit.: present paper | C. wedinii |
| | Galls up to 1 mm diam., hymenium I+ violet, ascospores shorter | 4 |
| 4(3) | Asci (2–)4-spored, ascospores 11.5–14(–16) × 5.5–7(–8) μm; on saxicolous <i>Lecidea</i> Ach. s. lat. Lit.: Etayo (2010) | C. galloides |
| | Asci 4–8-spored, ascospores 16.5–20 × 6.5–7.5 μm; on <i>Umbilicaria</i> Hoffm. Lit.: Körber (1865); Hawksworth (1982); Alstrup & Hawksworth (1990); Etayo (2010) | C. grossum (Körb.) D. Hawksw. |
| 5(2) | Hymenium predominantly I+ red; on <i>Verrucaria latebrosa</i> Körb. Lit.: Shivarov (2019) | C. hemiamyloideum Shivarov |
| | Hymenium not I+ red | 6 |
| 6(5) | Hymenium I+ violet; on <i>Parmotrema</i> A. Massal. Lit.: Etayo (2017) | C. amylaceum Etayo |
| | Hymenium I– | 7 |
| 7(6) | Ascospores longitudinally striate by wrinkles, (16–)17–20(–21) × (7–)8–9(–10) μm; on <i>Parmotrema</i> . Lit.: Etayo (2017) | C. rugosisorum Etayo & Zhurb. |
| | Ascospores not longitudinally striate | 8 |
| 8(7) | Asci 2-spored, ascospores (17–)20–27(–35) × (4–)5–5.5(–6) μm; on <i>Cetraria</i> Ach. and <i>Nephromopsis</i> Müll. Arg. Lit.: Zhurbenko (2009) | C. bisporum Zhurb. |
| | Asci with more than 2 spores, ascospores of a different size | 9 |
| 9(8) | Asci 4-spored | 10 |
| | Asci 4–8-spored | 11 |
| 10(9) | Ascospores 8–10 × 5–7 μm; on <i>Buellia melanostola</i> (Hue) Darb. Lit.: Øvstedal & Hawksworth (1986); Grube & Hafellner (1990) | C. epimelanostolum (Øvstedal & D. Hawksw.) Grube & Hafellner |
| | Ascospores (12–)14.5–17.5(–19.5) × (4.5–)5–6.5(–7.5) μm; on <i>Cetraria</i> , <i>Nephromopsis</i> and <i>Vulpicida</i> J.-E. Mattsson & M.J. Lai. Lit.: Hafellner (1994); Zhurbenko & Zhdanov (2013); Zhurbenko & Kobzeva (2014) | C. cetrariae Hafellner |
| 11(9) | Ascospores (24–)29–32(–34) × 5.5–6.5 μm; on <i>Hypotrachyna</i> (Vain.) Hale. Lit.: Etayo (2017) | C. cajasense Etayo |
| | Ascospores shorter | 12 |
| 12(11) | Ascomata 80–200 μm diam., ascospores (15–)16–21(–22.5) × 6–8.5(–9.5) μm; on <i>Squamarina</i> Poelt. Lit.: Navarro-Rosinés <i>et al.</i> (1994) | C. psoromatis (A. Massal.) Etayo |
| | Ascomata smaller, ascospores shorter | 13 |
| 13(12) | Ascomata 60–150 μm diam., ascospores (13.5–)14–16(–18) × (5.5–)6–7(–7.5) μm; on <i>Cetrelia</i> W.L. Culb. & C.F. Culb. and <i>Xanthoparmelia</i> (Vain.) Hale. Lit.: Hawksworth (1977) | C. cladonema |
| | Ascomata smaller | 14 |

- 14(13) Ascomata c. 100 µm diam., ascospores 15.5–18 × 8–10 µm; on *Placopsis* (Nyl.) Linds. Lit.: Øvstedal & Hawksworth (1986)
 **C. placopsiophilum** Øvstedal & D. Hawksw.
 Ascomata 50–100 µm diam., ascospores (9–)10–12(–13) × (4–)5–6(–6.5) µm; on *Hypocenomyce* M. Choisy. Lit.: Hawksworth
 (1980a) **C. hypocenomycis** D. Hawksw.

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