

A NEW SPECIES OF BOBOVIA (PEZIZALES)  
FROM THE HVALER ARCHIPELAGO IN NORWAY

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*Boubovia vermiphila* spec. nov. (Pezizales) is described from the mycologically interesting Hvaler archipelago in southern Norway, occurring on casts of earthworms. The relationship with *Pulvinula* and *Boubovia luteola* is discussed. Some distinctive characters between *Pulvinula* and *Boubovia* are considered.

The Hvaler archipelago is a small group of islands situated in the temperate zone of Norway, close to the Swedish border, about 100 km south-east of Oslo, up to 75 m above sea-level. The geographical position and their ecological and phenological conditions make them favourable for the growth of a wide variety of operculate discomycetes (Pezizales, *Ascomycota*). The geology is Precambrian, and consists of granites and gneisses. The landscape is covered with marine sediments, sand- and shell-beds, sand-dunes, fields of heather, shallow marshes, calcareous clay and earth which often fill the cavities and open spaces between rocks. The forests consist of coniferous and deciduous trees. The soil is rich in nutrients and lime, giving rise to a great diversity of phanerogams, including numerous rare calcicolous species.

The second author has studied the fungi of this region, with special emphasis on the Pezizales, for more than fifteen years. Thus far, over 130 species among 46 genera of Pezizales and Thelebolaceae have been recorded, the most noticeable of which are: *Aleuria bicucullata* (Boud.) Gill., *Boudiera acanthospora* Dissing & T. Schumacher, *Rhodoscypa ovilla* (Peck) Dissing & Sivertsen (Kristiansen, 1985); *Marcelleina georgii* (Svrček) J. Moravec (Moravec, 1987); *Scutellinia paludicola* (Boud.) Le Gal (Schumacher, 1990); *Lamprospora maireana* Seaver, *Boubovia luteola* (Velen.) Svrček, *Neottiella ricciae* (Crouan) Le Gal, *N. hetieri* Boud., *Peziza lividula* W. Phillips in Cooke, *Pseudascozonus racemosporus* Brumm. (Kristiansen & Schumacher, 1993); *Lamprospora kristiansenii* Benkert (Benkert, 1991); *Chalazion helveticum* Dissing, *C. sociabile* Dissing & Sivertsen (Kristiansen, 1990); *Ascodesmis nana* Brumm., *A. nigricans* Tiegh., *A. sphaerospora* Obrist (Kristiansen, 1994); *Octospora leucoloma* (Hedw.) Korf var. *tetraspora* (Fuckel) Benkert, *O. coccinea* (Crouan) Brumm. var. *tetraspora* Benkert (Benkert, 1998); *Thecotheus holmskjoldii* (E. C. Hansen) Eckblad, *Caccobius minusculus* Brumm., *Ascozonus woolhopensis* (Renny) J. Schröt. (Landvik et al., 1998a; van Brummelen & Kristiansen, 1998); and *Moravecchia hvaleri* Benkert & Kristiansen (Benkert & Kristiansen, 1999).

In 1983 an enigmatic minute discomycete was discovered, which did not seem to fit any known species at that time. Additional material was found at the same locality in 1989, 1990, and 1998, but was not found in the intervening years. Study of the latest rich collection confirmed earlier suspicions that it was a new species of *Boubovia* Svrček.

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**Boubovia vermiphila** Brumm. & R. Kristiansen, *spec. nov.* — Figs. 1, 2

Apothecia eugymnohymeniale, sparsa vel dense gregaria, sessilia, (0.1–)0.2–0.5(–0.1) mm diam., (0.1–)0.3–0.5(–1.0) mm alta. Receptaculum initio subglobulare, deinde cylindricum vel doliiforme, denique pulverulentum vel subscutellatum, hyalino-albidum, lutescens, fragile, laeve. Excipulum medullare ex textura intricata. Excipulum corticale ex textura globulosa. Asci late clavati, apice rotundati, maturitate protrusi, operculo lato,  $130\text{--}160 \times 20\text{--}27 \mu\text{m}$ , 8-spori, pariete iodo non caerulescente. Ascospores univel bi-serratae, ellipsoideae,  $17.8\text{--}20.2(–23.2) \times 9.1\text{--}10.6 \mu\text{m}$  (excl. ornamenti), verrucis admodem grossis granulisque irregularibus instructae, plerumque pileis apicalibus grandibus instructae. Paraphyses simplices vel raro ramosae, filiformae,  $2.0\text{--}2.5 \mu\text{m}$  crassae, apice non dilatatae, sed fortiter vel spiraliter contortae.

Ad terram humidam mixtus ad excrementa vermium.

Typus: *R. Kristiansen RK 89.18*, prope Putten, Kirkøy, Hvaler comm., Østfold comitatus, Norvegia, 12–17.VI.1989 (holotypus L; isotypus O, TRH).

Etymology: From Latin, vermis, a worm, and Greek, φιλέω (phileo), to love: with preference for (substrata with) worms.

Apothecia eugymnohymenial, solitary or in small groups or caespitose, sometimes aggregating, superficial, sessile on a narrow base, (0.1–)0.2–0.5(–1.0) mm diam., (0.1–)0.3–0.5 mm high. Receptacle at first subglobular, then cylindrical or doliiform, finally becoming pulvinate to subscutellate, whitish hyaline becoming yellowish on drying; consistency rather loose, fragile; surface smooth; margin not differentiated. Disc flat to convex, roughened by protruding ends of mature asci, hyaline. Hymenium  $145\text{--}160 \mu\text{m}$  thick. Hypothecium not clearly differentiated as a distinct layer, of groups of isodiametric and elongated thin-walled cells  $2\text{--}5 \times 2\text{--}3 \mu\text{m}$ . Medullary excipulum up to  $70 \mu\text{m}$  thick, of intermingled hyphae  $4.5\text{--}9 \mu\text{m}$  wide (textura intricata). Cortical excipulum clearly differentiated, near the base up to  $50 \mu\text{m}$  thick, at the margin  $15\text{--}25 \mu\text{m}$  wide, hyaline, consisting of globular and subglobular thin-walled cells  $10\text{--}25 \times 6\text{--}20 \mu\text{m}$  (textura globulosa), without hairs or hyphoids.

Asci rather broadly clavate, attenuated at the base, rounded above, with a large operculum about  $8 \mu\text{m}$  across,  $130\text{--}160 \times 20\text{--}27 \mu\text{m}$ , 8-spored, but often with only some of the spores properly developed; the wall not blue with iodine. Ascospores uni- or bi-seriate, ellipsoid (length/width ratio  $1.9\text{--}2.1(–2.3)$ , average 2.04),  $17.8\text{--}20.2(–23.2) \times 9.1\text{--}10.6 \mu\text{m}$  (without ornamentation), with 1 or 2 oil globules when young and occasional air bubbles, ornamented with a pattern of large and small irregularly shaped warts staining with methyl blue, usually with large apical caps. Paraphyses rather frequent, septate, filiform, scarcely branched, hyaline,  $2.0\text{--}2.5 \mu\text{m}$  thick, with strongly or spirally curved, not enlarged ends, containing small colourless vacuoles and granules staining red with iodine.

Habitat — On damp soil mixed with excrements of earthworms (Lumbricidae).

*Specimens examined.* NORWAY: Østfold county, Hvaler comm., Kirkøy, near Putten, on damp soil mixed with excrements of earthworms (Lumbricidae), 12–17.VI.1989, *R. Kristiansen RK 89.18* (holotype of *Boubovia vermiphila*, L; isotypes O, TRH); same locality, 18–22.VI.1983, *R. Kristiansen RK 83.145* (L); same locality, 19–24.VI.1990, *R. Kristiansen RK 90.15* (L); same locality, 13.VI.1998, *R. Kristiansen RK 98.30* (L).

The fruitbodies of *B. vermiphila* were collected four times from the same locality in the Hvaler archipelago in June. These were found growing on the rounded surface and in the crevices of wormcasts, surrounded by black calcareous soil with minute fragments of sea shells, and often covered with fallen leaves of *Populus tremula* L., one of the species of trees shading the locality. More details on the vegetation of the locality were described by Kristiansen (1990: 93).

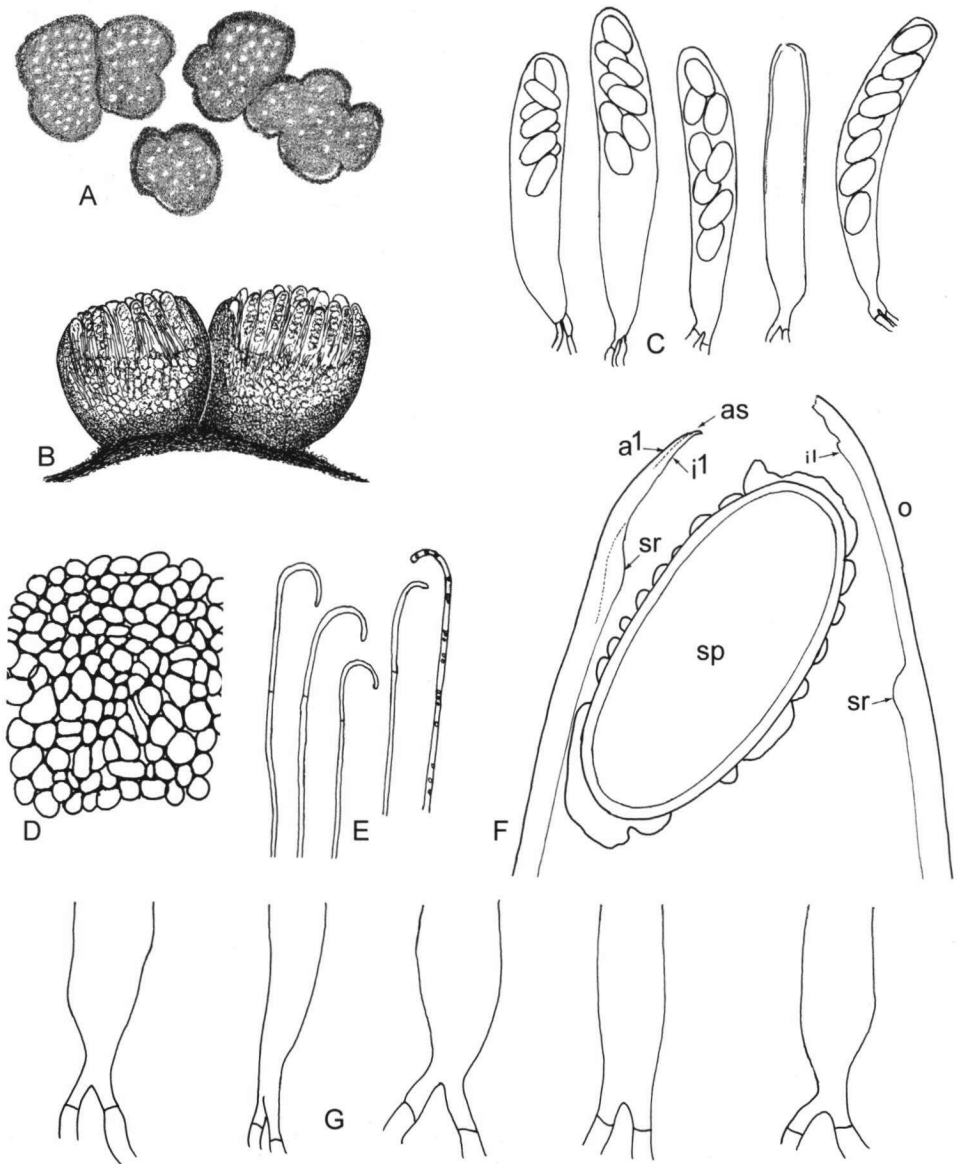


Fig. 1. *Boubovia vermiphila*. A. Habit of fruit-bodies from above,  $\times 12.5$ ; B. id., more detailed in lateral view,  $\times 100$ ; C. asci,  $\times 300$ ; D. detail of excipulum seen from outside,  $\times 300$ ; E. paraphyses,  $\times 500$ ; F. detail of top of dehisced ascus,  $\times 2700$  (as = ascostome; il = inner layer; o = operculum; ol = outer layer; sp = ascospore; sr = subapical ring); G. bases of asci,  $\times 750$  (A & E from RK 83.145; B from RK 98.30; C, D & G, from RK 90.15; F from RK 89.18, holotype).

*Boubovia vermiphila* is often found growing together with *Chalazion sociabile* Dissing & Sivertsen and *Ascobolus denudatus* Fr. Macroscopically, its minute apothecia have the appearance of a *Chalazion* and are, in the field, difficult to distinguish from those of *Chalazion sociabile*. The apothecia of *C. sociabile* are, however, white to greyish, without yellow-

ish tints, showing only a weakly developed excipulum, broadly clavate asci with huge opercula, as well as different, larger ascospores, and rather thick, straight paraphyses with thickened ends.

*Boubovia vermiphila* differs also from the recently described coprophilous *Chalazion erinaceus* Doveri et al. (Doveri et al., 1998), as we could confirm from a study of the type specimen. *Chalazion erinaceus* has much smaller (75–100 µm diam.) orbiculate to pulvinate white to dirty white apothecia; the excipulum is very restricted, showing cells that easily stain with methyl blue; while the asci are broadly clavate and smaller (62–72 × 23–34 µm) with an operculum about 24 µm across; the paraphyses are not curved and slightly swollen near the ends.

It shows great similarity in growth and structure with species of *Pulvinula* Boud. and especially with *Boubovia luteola* (Velen.) Svrček (Velenovský, 1934, as *Humaria luteola*; Svrček, 1977, 1978; Kristiansen & Schumacher, 1993).

*Chalazion* was tentatively placed in the Thelebolaceae (Hawksworth et al., 1983; Eriksson & Hawksworth, 1998), but results of DNA-sequencing (Landvik et al., 1998b) indicate that *Chalazion* groups most closely with *Glaziella* Berk. and with *Pulvinula*, both belonging to the Pyronemataceae.

*Boubovia vermiphila* differs from *Boubovia luteola* in the smaller pulvinate apothecia, the larger and more abundantly ornamented ascospores with large polar caps. It may also be related to *Octospora spaniosa* K.B. Khare (Khare, 1975), but that has much smaller very finely reticulate ascospores.

The similarity to *Pulvinula*, as defined by Rifai (1968), Pfister (1976), Korf & Zhuang (1984, 1991), and Hohmeyer (1988), is in the eugymnohymenial development, pulvinate apothecia, yellow carotenoid pigment, identical textures of the cortex and the medulla, asci with a pleurorhynchous bifurcate base, and very thin, filiform paraphyses with strongly curved ends. It differs, however, in its clearly ornamented, ellipsoid ascospores.

Species with ellipsoid ascospores were accepted in *Pulvinula* by Pfister (1976) and by Korf & Zhuang (1984, 1991), but Yao & Spooner (1996a, 1996b) transferred such species to *Boubovia* Svrček. Besides the spore shape, they considered the presence of thick walls at a certain stage in young asci to be of sufficient importance for these transfers.

The value of the last mentioned character must be considered doubtful, since it concerns a cytological character based on post-mortem observations. It is well known that the inner layer of the ascus wall may swell strongly by imbibition when the ascus turgor disappears (Boedijn, 1933; van Brummelen, 1986, 1998; Baral, 1992), especially when placed in certain media for microscopic observation.

A reliable character of species of *Pulvinula* may be found in the development of the ascospores. Wu & Kimbrough (1993) demonstrated in ascospores of *Pulvinula convexella* (P. Karst.) Pfister a strongly zonated epispore and an outermost thin layer of the secondary spore wall which is finely echinulate or granular, which is not visible with light microscopy. In the view of these authors the outer layer of the epispore is directly deposited from the epiplasm, which would mean that the epispore becomes the outer spore layer. The early wall development in *Pulvinula* differs from that found in all other representatives of the large family of the Pyronemataceae, like *Aleuria*, *Octospora*, *Humaria*, *Trichophaea*, *Pseudombrophila*, and *Scutellinia*. Unpublished studies by the first author confirm this special mode of ascospore development in *Pulvinula convexella* and *P. ovalispora* Boud. In *Boubovia luteola* and *B. vermiphila*, with complex and extensive ascospore ornamentations,

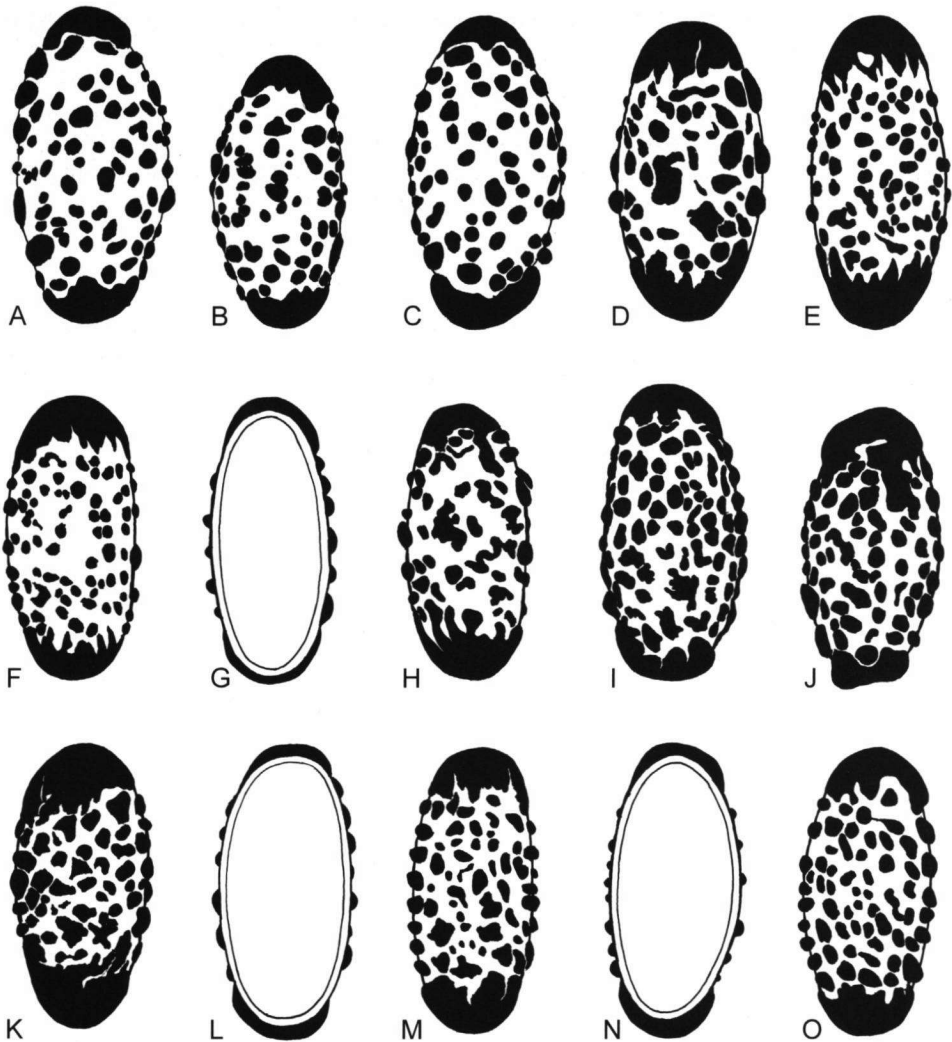


Fig. 2. *Boubovia vermiphila*. A–O. Ascospores,  $\times 1600$  (A–C from RK 83.145; D–H from RK 89.18, holotype; I–L from RK 90.15; M–O from RK 98.30).

the development of the secondary wall will be a more complex process, but ultrastructural confirmation will be needed. In *Pulvinula* the smooth ascospores are naked (i.e. devoid of a secondary wall). This would mean that *Pulvinula* should be restricted to species with smooth spores, whether globose or ellipsoid, while *Boubovia* should accommodate species with ornamented spores, so far all showing ellipsoid spores.

Air or gas inclusions are almost constantly reported as 'de Bary bubbles' for ascospores of *Pulvinula*, but in *Boubovia* these inclusions are produced only occasionally. This difference can be explained by the very dense, stratified, impermeable episporium in *Pulvinula* (Wu & Kimbrough, 1993) and a supposed more simple one in *Boubovia*.

The taxonomic position of *Boubovia* is within the Pyronemataceae, since the ascus apical apparatus is of the *Octospora* type (van Brummelen, 1978, 1986), also found in *Pulvinula convexella* and *P. ovalispora*. A subapical swelling in the shape of a ring can be observed at the inner side of the ascus wall at some distance behind the ascus tip, while the operculum and the ascostome are rather roughly delimited during ascus dehiscence (Fig. 1F). In all genera of the Pyronemataceae studied thus far this type of ascus apical apparatus could be established.

Preliminary results of DNA-sequencing by Dr. S. Landvik (pers. comm.) indicate that both *Boubovia luteola* and *B. vermiphila* are grouping close to *Pulvinula* and *Chalazion*, other representatives of the Pyronemataceae.

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