Mycoscience (2004) 45:112–115
DOI 10.1007/s10267-003-0166-4

FULL PAPER

Tsuyoshi Hosoya · Koukichi Maruyama

Pachyella globispora sp. nov. (Pezizaceae) from Japan

Received: July 24, 2003 / Accepted: November 18, 2003

Abstract Psilopezioid fungi form a group of operculate discomycetes characterized by sessile, generally pulvinate to shallow cupulate, and broadly attached apothecia occurring on wet or submerged wood and plant debris. A new member, *Pachyella globispora* sp. nov. (Pezizaceae), is described that is distinguished from other species of *Pachyella* in having markedly warted, globose ascospores.

Key words New species · *Pachyella globispora* · Pezizales · Psilopezioid · Taxonomy

Introduction

The psilopezioid fungi form a group of discomycetes characterized by sessile, generally pulvinate to shallow cupulate, and broadly attached apothecia occurring on wet or submerged wood and plant debris (Pfister 1973a). The majority of the psilopezioid fungi belong in the genera *Psilopezia* Berk., in the Pyronemataceae, or *Pachyella* Boud., in the Pezizaceae.

The genus *Pachyella* is characterized by asci diffusely amyloid without prominent crosiers; ascospores usually less than 25 μm long that lack a loosening outer spore wall; gel present in the medullary excipulum as well as surrounding the external, hyphoid hairs; medullary and excipular zones well defined; and paraphyses neither branching nor anastomosing (Pfister 1973a).

A monographic study by Pfister (1973b) was updated by Pfister and Candoussau (1981), who included nine species. Several species have been added since 1981. Four species are known in Japan: *P. adnata* (Berk. & M.A. Curtis) Pfister, *P. babingtonii* (Berk.) Boud., *P. clypeata* (Schwein.) Le Gal, (specimens cited and examined by Pfister 1973b for these three species), and *P. violaceonigra* (Rehm) Pfister (Hosoya et al. 1991).

A specimen of *Pachyella* collected by K.M. showed a unique combination of characters. This specimen has warted, globose ascospores unlike any previously described *Pachyella*. It is described here as a new species.

T. Hosoya (✉)
2-19-12-607 Toride, Toride-shi, Ibaraki 302-0004, Japan
Tel. +81-297-74-1004; Fax +81-297-74-1004
e-mail: kfe00271@nifty.ne.jp

K. Maruyama
Hachioji, Tokyo, Japan

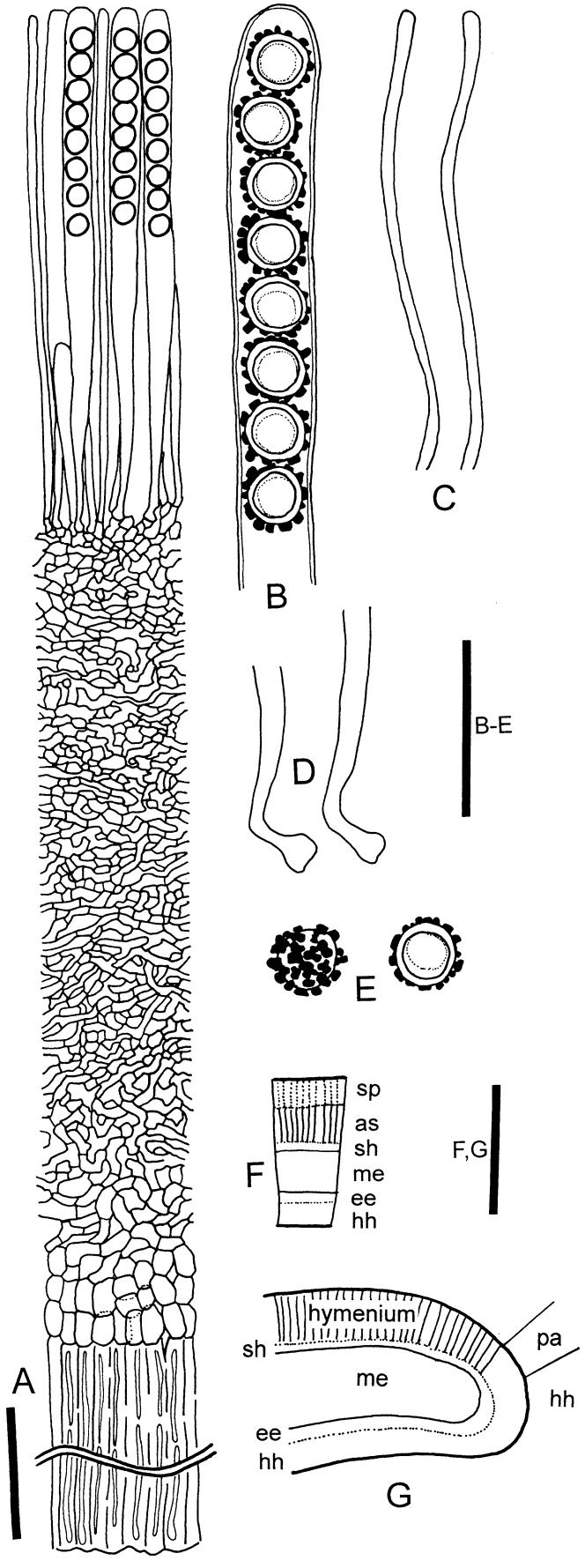
Materials and methods

The collected material was air-dried. Cultivation was not attempted. Sections were obtained using a freeze microtome as well as by hand sectioning. Color codes followed CMYK system referring to a publication (Anonymous 2002). For scanning electron microscope (SEM) observation of the ascospore ornamentation, a small portion of dried specimen was rehydrated, and crushed in a drop of water on aluminum foil to release ascospores. The aluminum foil was affixed on the stub, and the ascospores were platinum-palladium coated in an ion sputter (Hitachi E-102; Hitachi, Tokyo Japan), and observed under an SEM (Hitachi S-4000) operating at 5.0 kV.

Description

Pachyella globispora K. Maruyama & Hosoya, sp. nov.
Figs. 1–3

Apothecia 2–4 cm diametro, sessilia, substrato late affixa, vadose cupulata, cum margine irregulariter sinuato, radiatim undulata vel plicata, ab superficie summa atrobrunnea, infla alba. Hymenium 480–500 μm latum. Excipulum medullare 440–480 μm latum, "textura intricata," ex hyphis 4–6 μm latis arcte implexis, tenuitunicatis, frequenter septatis compositum. Excipulum ectale 80 μm latum, ex cellulis angularis pluristratis compositum. Pili hyphoidei ex cellulis extimis excipuli ectali emittenes, translucentes, 200–300 μm longi, 2.5–3.5 μm crassi, ab



superficie excipuli rectangulati, in matrice gelatinosa infossi. Asci 400–460 × 16–22 μm, octospori, cylindracei, iodo excluso KOH tarde coerulescenti, sine hamuli conspicuis. Ascosporae uniseriatae, globosae, pallide-brunneae, 20–24 μm diametro (verrucis. ca. 2 μm diametro inclusis), crassitunicati (1.5–2 μm), cum verrucis oblongis, uniglobulatis paraeditae. Paraphyses filiformes, simplices, ad medietem 2.5–3 μm crassae, versus apices leviter crassiores ad 6 μm crassuae.

Holotypus. TNS-F-6522, Japan-Honshu: Mt. Tsurune, Kosuge-mura, Kita-tsuru-gun, Yamanashi Pref. (alt ca. 1350 m), on *Fagus* stump, July 29, 2001, coll. K. Maruyama.

Isotypus. FH (Farlow Reference Library and Herbarium of Cryptogamic Botany, Harvard University).

Etymology. Latin, *globi-* (globose) + *spora* (spore), referring to the globose ascospores, a unique characteristic among the species of *Pachyella*.

Japanese name. "Marumi-no-kabairo-chawantake."

Apothecia sessile, broadly (at least 1/3 of the base) attached to the substrate, flat to shallowly cupulate with irregularly undulating margin, radially undulate to plicate, 2–4 cm across, dark brown (C80M80Y80K30), darker toward the center, white below, firm when fresh; becoming thin, leathery, black from both sides when dry. *Hymenium* 480–500 μm thick, young and old asci mixed, staining blue in Melzer's reagent (MLZ). *Subhymenium* 80 μm thick, almost pseudoparenchymatous, of compact cells, 4–6 μm thick. *Medullary excipulum* 440–480 μm thick, textura intricata, of tightly entangled, thin-walled, closely septate hyphae 4–6 μm thick. *Ectal excipulum* 80 μm thick, several cell layers of thicker-walled, hyaline angular cells. *Hyphoid hairs* arising from the outermost layer of ectal excipulum, 200–300 μm long, 2.5–3.5 μm thick, with blunt end, arranged rectangularly to the surface, embedded in gelatinous matrix, translucent, lumen visible up to 2 μm wide. *Asci* 400–460 × 16–22 μm, 8-spored, cylindrical in the upper one third where occupied by the ascospores, gradually tapered to the base, thin walled (up to 1.5 μm), diffusely MLZ+ without KOH pretreatment; apex rounded to flat, thin walled; base enlarged but prominent crosiers lacking. *Ascospores* globose, 20–24 μm (22.0 ± 1.1 μm on average ± SD, $n = 20$) in diameter including the warts, 15–20 μm (17.1 ± 1.2 μm on average ± SD, $n = 20$) in diameter excluding the warts, thick walled (1.5–2 μm), ornamented with warts 2 μm in

Fig. 1. *Pachyella globispora.* **A** Vertical section at the middle of the apothecium. Hyphoid hairs in the gelatinous matrix. Lower areas partially omitted for convenience. **B** Ascus, upper portion. Note uniseriate ascospores. **C** Paraphyses. **D** Asci, lower portion. Note tapering below and the somewhat enlarged ends. **E** Ascospore drawn at two different optical levels. The *left* view shows the surface structure, whereas the *right* shows the optical section with its thick wall and one oil globule. **F** Schematic drawing of a vertical median section of an apothecium to show the structure. *sp*, ascal portion where occupied by the ascospores; *as*, ascal portion where not occupied by the ascospores; *sh*, subhymenium; *me*, medullary excipulum; *ee*, ectal excipulum; *hh*, hyphoid hairs. **G** Schematic drawing of a vertical section of the apothecium near the marginal area to show the structure. *pa*, transitional region where the paraphyses dominates over asci. *Bars* **A** 100 μm; **B–E** 50 μm; **F,G** 1 mm

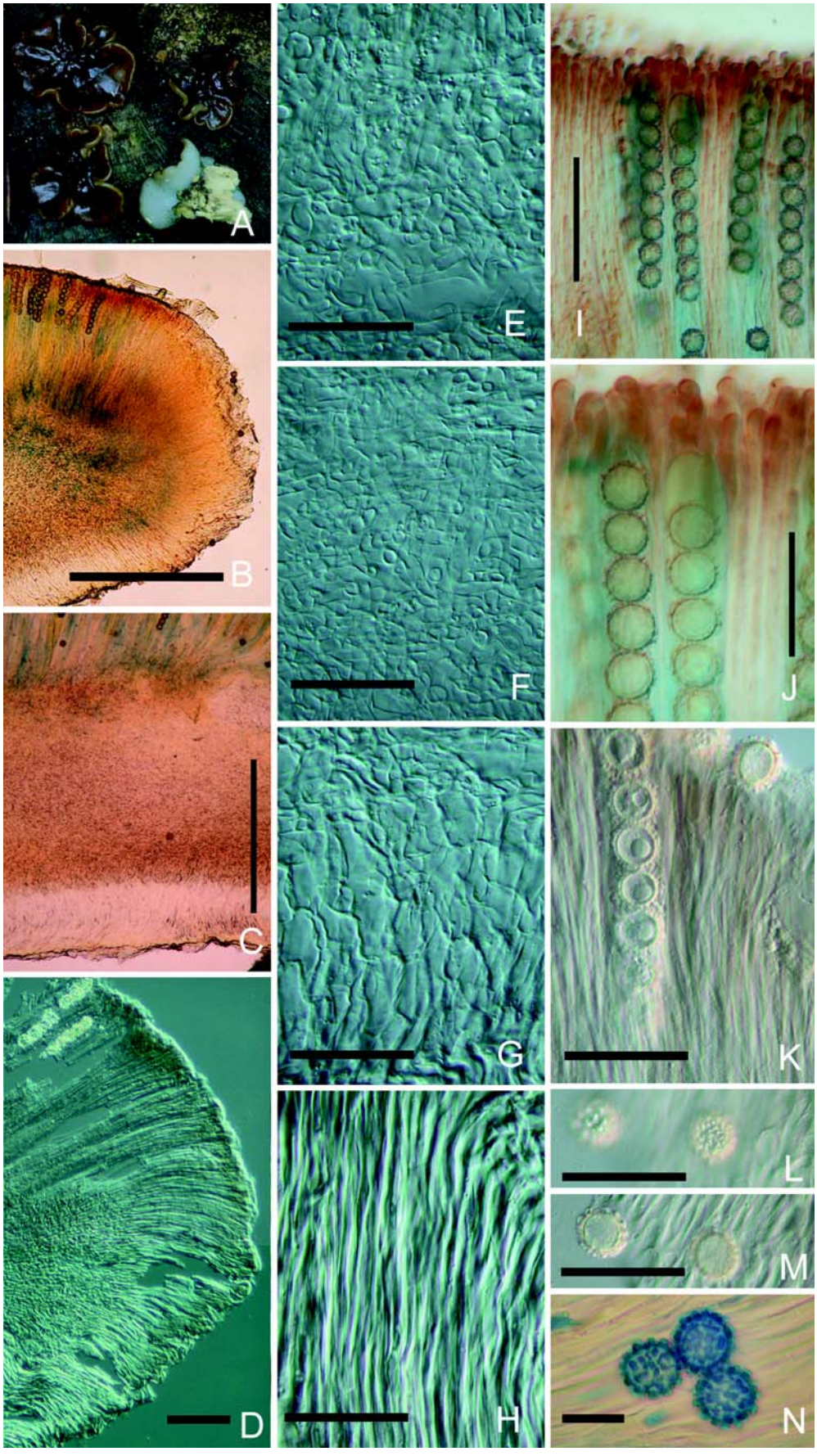


Fig. 2. *Pachyella globispora.* **A** Fresh apothecia. **B** Vertical section of the apothecium at the margin, prepared by hand section and mounted in Melzer's reagent (MLZ). Note stained portion. **C** Vertical section of the apothecium at the middle of the apothecium, prepared and mounted as above. Basal portion of the asci stained blue at the top. Note tissue organization. **D** Vertical section of the apothecium at the margin, prepared by freeze microtome, mounted in cotton blue/lactic acid (CB/LA). **E** Close-up of subhymenium/medullary excipulum. **F** Close-up of medullary excipulum. **G** Close-up of ectal excipulum. **H** Close-up of hyphoid hairs. **I** Upper portion of the hymenium, mounted in MLZ. Note paraphyses with contents stained brown, and asci diffusely stained blue. **J** Close-up of **I**. Note asci diffusely stained blue. **K** Ascospores, each containing one large oil globule. **L, M** The same ascospores at two different optical sections. Note the warts on the surface and thick walls. **N** Ascospores mounted in CB/LA. Note warts stained by cotton blue. *Bars* **B, C** 500 μm; **D** 100 μm; **E–H** 50 μm; **I** 100 μm; **J–M** 50 μm; **N** 20 μm

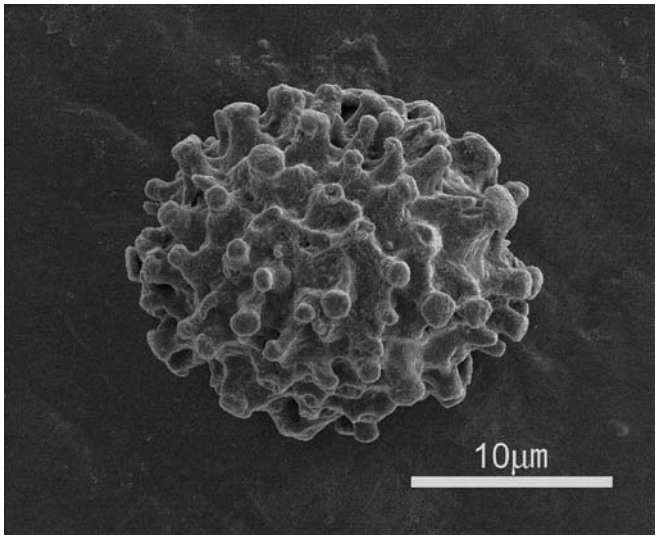


Fig. 3. SEM photograph of ascospores in *P. globispora*. Note warts with blunt apex sometimes enlarged distally. *Bar* 10 μm

diameter, uniform in thickness, sometimes enlarged distally, containing one oil globule 9.5–16 μm (11.6 ± 1.6 μm on average ± SD, $n = 20$) in diameter, pale brown, uniseriate, occupying the upper one third of the asci. *Paraphyses* filiform, simple, containing brownish pigment, 2.5–3 μm wide at the middle, slightly thickened toward the apex, up to 6 μm wide, cell content brown in MLZ.

Discussion

The present fungus is characterized by diffusely MLZ+ asci and presence of excipular hyphoid hairs embedded in the gelatinous matrix. These features match the diagnostic features of *Pachyella*. The present fugus is clearly distinguished from other known members of *Pachyella* in having strongly warted, globose ascospores.

Among the members of *Pachyella*, anatomical structures of *P. globispora* are similar to those of *P. adnata*, *P. clypeata*, *P. megalosperma* (Le Gal) Pfister, *P. habrospora* Pfister, and *P. violaceonigra.* The apothecial anatomy of these species is almost identical (Pfister 1973b, 1995). The external morphology of *P. globispora* also shows similarity to the above members to some extent. In addition, similarity to *P. peltata* Pfister & Cand. is noted because its habitat and morphology are shown to be similar to *P. clypeata* (Pfister and Candoussau 1981).

Diversity is known in the external ornamentation of the ascospores in *Pachyella*. Four have smooth ascospores, and the rest have ascospores with verrucose ornamentations of various degree. The warts are anastomosed and becoming crestlike in *P. megalosperma* (Pfister 1973b), while most prominent in *P. adnata* (Pfister 1973b, 1975). Ascospores in other members are granular to strongly verrucose (Pfister 1975). The warts in *P. globispora* most closely resemble those of *P. adnata*. However, the position of *P. globisopora* is unique in the genus *Pachyella*, because no other members of the genus *Pachyella* have globose ascospores; the anatomy is close to the majority of *Pachyella* species.

Several apothecial genera of Pezizaceae include species with globose ascospores, such as *Boudiera* Cooke, *Hapsidomyces* J.C. Krug & S.C. Jeng, *Plicaria* Fuckel, and *Scabropeziza* Dissing & Pfister. *Boudiera* and *Scabropeziza* were grouped with *Pachyella* based on large subunit rDNA sequence analysis, although the grouping lacked strong support. A phylogenetic relationship between *Boudiera* and *Pachyella* was not strongly supported (Hansen et al. 2001). The branches occurred in this group were too long for any decisive phylogenetic interpretation, but the position of these taxa might be expected to be clarified by inclusion of more members. Hansen et al. (2001) also suggested some similarity in ecology, ascal characteristics, and overall anatomy between *Boudiera* and *Pachyella*. Both occur in wet habitats, both have diffusely amyloid asci, and both have a well-developed outer excipulum of angular to globose cells. Thus, the unique combination of the apothecial anatomy and ascospore characteristics of *P. globispora* may help support the somewhat tenuous molecular data indicating a relationship between the two.

Acknowledgments The authors express their deepest thanks to Drs. M. Kakishima, and J. Abe, Tsukuba University, for kindly allowing us to use the laboratory facilities, including the light microscope. Dr. Y. Yaguchi, Tokyo University of Agriculture, is greatly appreciated due to his courtesy in SEM observation. We also wish to express our gratitude to Drs. R.P. Korf, Cornell University, and D.H. Pfister, Harvard University, for their useful advice, guidance, facilitation with the literature, and for kindly reviewing the manuscript . We also thank Mr. S. Kudo, Koujin-juku for kindly allow the first author to use his equipment that made the primary examination of the material possible.

References

Anonymous (2002) Color chart. Dainippon Ink & Chemicals, Tokyo, Japan

Hansen K, Læssøe T, Pfister DH (2001) Phylogenetics of the Pezizaceae, with an emphasis on *Peziza*. Mycologia 93:958–990

Hosoya T, Furuya K, Otani Y (1991) Materials for the fungus flora of Japan (44). Trans Mycol Soc Jpn 32:351–353

Pfister DH (1973a) The psilopezioid fungi. I. History, nomenclature, and delimitation of the psilopezioid genera. Mycologia 65:321–328

Pfister DH (1973b) The psilopezioid fungi. IV. The genus *Pachyella* (Pezizales). Can J Bot 51:2009–2023

Pfister DH (1975) Scanning electron micrographs of ascospores of *Pachyella* (Discomycetes). Mycotaxon 3:105–108

Pfister DH (1995) The psilopezioid fungi. IX. *Pachyella habrospora*, a new species from Brazil. Mycotaxon 54:393–396

Pfister DH, Candoussau F (1981) The psilopezioid fungi. VIII. Additions to the genus *Pachyella*. Mycotaxon 13:457–464