FULL PAPER

Yasunori Ono · Takao Kobayashi

Notes on new and noteworthy plant-inhabiting fungi in Japan (3)

Received: May 11, 2005 / Accepted: September 5, 2005

Abstract In the third report of the present series, four new and noteworthy plant-inhabiting fungi are described and illustrated. *Pseudodidymaria symplocarpi* on *Symplocarpus nipponicus* is reported as a new species. *Cheirospora botryospora* and *Exosporium mexicanum* are new to Japan. *Pittosporum tobira* is a new host plant for *Flosculomyces floridaensis*.

Key words Cheirospora botryospora · Exosporium mexicanum · Flosculomyces floridaensis · New species · Pseudodidymaria symplocarpi

Introduction

In the course of exploratory surveys of plant-inhabiting fungi as producers of secondary metabolites useful to the pharmaceutical industry, we encountered a number of new and noteworthy plant-inhabiting fungi (Ono and Kobayashi 2001, 2003). In this article, we describe and illustrate a new species of the genus *Pseudodidymaria*, two species newly recorded in Japan, and one species on a new host.

Material and methods

The collection, isolation and optical microscope observation of the specimens were carried out following the meth-

Y. Ono (⊠)

Core Technology Research Laboratories, Sankyo Co., Ltd., 33 Miyukigaoka, Tsukuba, Ibaraki 305-0841, Japan Tel. +81-29-856-3987; Fax +81-29-856-0572 e-mail: onoyas@sankyo.co.jp

T. Kobayashi

Department of International Agricultural Development, Tokyo University of Agriculture, Tokyo, Japan

ods of Ono and Kobayashi (2001, 2003). The specimens cited in this paper were deposited in the Herbarium of the Forestry and Forest Products Research Institute (TFM).

Description

Cheirospora botryospora (Mont.) Berk. & Broome, Ann. Mag. Nat. Hist., Ser. 2, 5:455 (1850); B. Sutton, The Coelomycetes p. 202 (1980)

Colonies punctiform, scattered, black. Mycelium immersed, branched, septate, hyaline. Conidiomata acervular, subepidermal or subperidermal, separate, hyaline to pale brown, and composed of hyphae of textura intricata. Dehiscence irregular, with strongly erumpent conidial masses adhering and forming a black pulvinate mass over the acervulus. Conidiphores $100-185 \times 2-3 \mu m$, hyaline, branched only at the base, septate, straight or flexuous, cylindrical, expanding toward the apex, smooth, often enclosed in a gelatinous sheath, and formed from the upper cells of the acervuli. Conidiogenous cells holoblastic, determinate, integrated, short cylindrical, hyaline, smooth, forming a single apical complex conidium. Conidia composed of central cells and short lateral acropetal branched cells, $18-33 \times 13-33.5 \,\mu\text{m}$ (avg. = $26.7 \times 21.1 \,\mu\text{m}$, n = 29); central cells cylindrical, obtuse at the top, septate, brown, each cell having several short branches; branch cells spherical, aseptate, smooth, brown, $3-5 \mu m$ (avg. = $3.9 \mu m$, n = 50) in diameter.

Specimen examined: On the twigs of *Fagus crenata* Blume (Fagaceae), Sanada-machi, Nagano Prefecture, May 20, 2002, Y. Ono, TFM: FPH-7764.

Note: Cheirospora botryospora has been recorded on Carpinus caroliniana Walter, Cornus alba L., Hedera helix L., Fagus grandifolia Ehrh., and Quercus robur L. in Austria, Canada, UK, and USA (Dennis 1986; Ginns 1986; Reid 1985; Sutton 1980). This is the first record of C. botryospora in Japan, and F. crenata has been newly added to the list of its host plants.

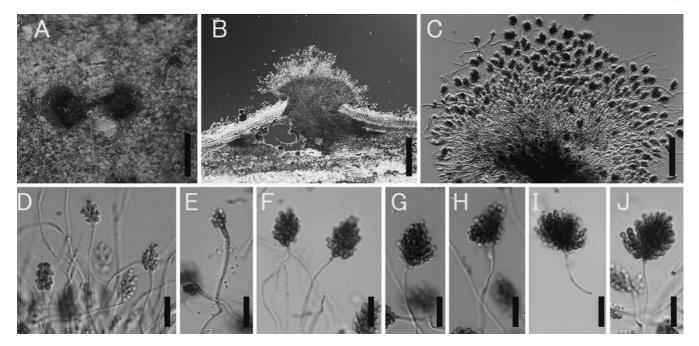


Fig. 1. Cheirospora botryospora (Mont.) Berk. & Broome. A Conidiomata on the twigs of Fagus crenata. B Cross section of a conidioma on a twig of F. crenata. C–J Conidia and conidiophores. Bars A 1 mm; B 200 μm; C 100 μm; D–J 20 μm

Exosporium mexicanum (Ellis & Everh.) M.B. Ellis, Mycol. Pap. 87:25 (1963)

- *≡Isariopsis mexicana* Ellis & Everh., Bull. Torrey Bot. Club 24:475 (1897)
- *■Phaeoisariopsis mexicana* (Ellis & Everh.) Ferraris, Ann. Mycol. 7:280 (1909)
- =Helminthosporium inversum Sacc., Ann. Mycol. 11: 557 (1913)
- *≡Corynespora inversa* (Sacc.) S. Hughes, Can. J. Bot. 36:757 (1958)
- *≡Exosporium inversum* (Sacc.) M.B. Ellis, Mycol. Pap. 82:30 (1961) Fig. 2

Stroma mostly immersed, pseudoparenchymatous, the upper cell layer brown, cells hyaline to subhyaline elsewhere. Conidiophores fasciculate, arising from the upper cells of the stroma, simple, straight or flexuous, brown, often paler near the apex, smooth-walled, sepate, 200– $295\,\mu m$ long, 6– $7.5\,\mu m$ thick at the base, 6– $8\,\mu m$ thick at the apex, with numerous, well-defined, brown to dark brown scars. Conidiogenous cells polytretic, integrated, terminal, becoming intercalary, sympodial, cylindrical or clavate, cicatrized, with the scars often dark and prominent. Conidia straight or curved, cylindrical to obclavate, smooth, pale brown to brown, with 3–11 pseudosepta, tapering toward the apex, having a thick, black, truncate scar at the base, 31– 134×11.5 – $14.5\,\mu m$ (avg. = $55.7 \times 12.6\,\mu m$, n = 33), 3– $5\,\mu m$ wide at the scar.

Specimen examined: On the dead twigs of *Erythrina* variegata var. orientalis Merr. (Leguminosae), China-cho (Okinoerabu Island), Kagoshima Prefecture, Nov 20, 2001, Y. Ono, TFM: FPH-7765 (culture MAFF 239183).

Note: The genus Exosporium Link was studied by Ellis (1961) and is characterized by its conidiophore development, conidiogenesis, and conidia. Ellis (1963) transferred Isariopsis mexicana to the genus Exosporium in his type study of Isariopsis mexicana. After his studies, Deighton (1990) transferred E. mexicanum to Phaeoisariopsis Ferraris, subsequent to his examination of the genus Phaeoisariopsis. Deighton (1990) pointed out that the main characteristics of the genus Phaeoisariopsis are conidiophores in dense subsynnematal fascicles emerging through the stroma; sympodial, polyblastic conidiogenous cells; conidial scars slightly but distinctly darkened and only slightly thickened, the old scars lying more or less flat against the sides of the conidiogenous cells; conidia more or less obclavata and with three or more transverse septa. Ellis (1961, 1963) described "conidium developed through a pore" and "conidia with 5–10 pseudosepta." Therefore, it is better placed as Exosporium mexicana.

Exosporium mexicanum has been recorded on the branches of Erythrina indica Lam., Erythrina sp., Smilax sp., Uvaria chamae P. Beauv.; and on the dead stems of Mascagnia macroptera var. jussieuana Nied. in India, Mexico, New Guinea, Philippines, Sierra Leone, and Sri Lanka (Ellis, 1961, 1963, 1971). This is the first record of E. mexicanum in Japan, and Erythrina variegata var. orientalis has been newly added to the list of host plants.

Flosculomyces floridaensis B. Sutton, Mycologia 70:789 (1978) Fig. 3

Leaf spots amphigenous, distinct, circular to irregular, grayish brown. Stroma absent. Conidiophores macronematous, mononematous, arising singly, straight or flexuous,

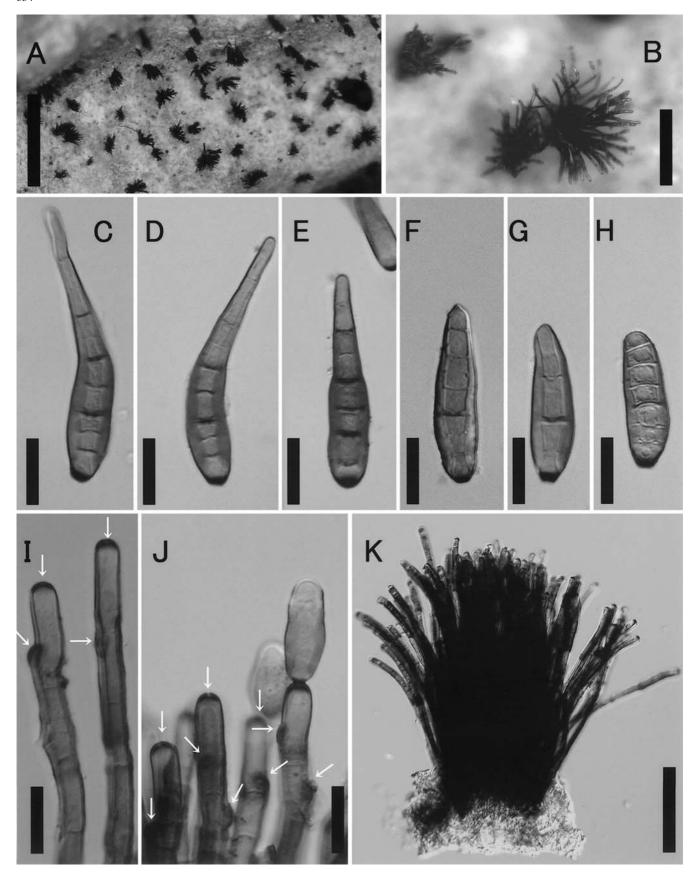


Fig. 2. Exosporium mexicanum (Ellis & Everh.) M.B. Ellis. **A,B** Conidiophores on the dead twigs of Erythrina variegata var. orientalis. **C–H** Conidia. **I,J** Conidiophores showing conidial scars (arrows). **K** Conidiophores arising from a stroma. Bars **A** 1 mm; **B** 200 μm; **C–J** 20 μm; **K** 100 μm

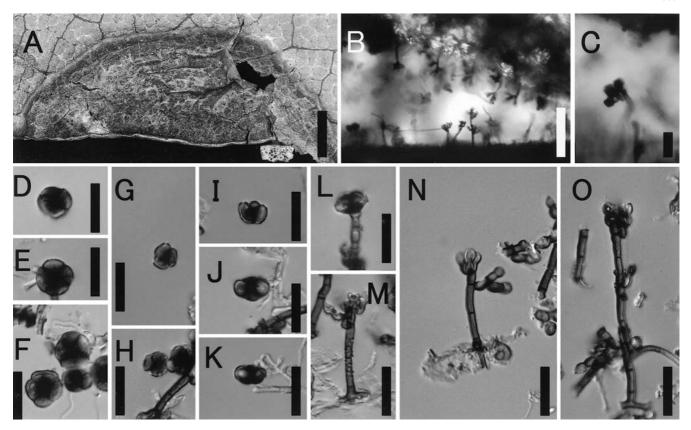


Fig. 3. Flosculomyces floridaensis B. Sutton. **A** Symptom on a leaf of *Pittosporum tobira*. **B,C** Conidiophores on *P. tobira*. **D-F** Horizontal view of four-celled conidia. **G,H** Horizontal view of three-celled

conidia. **I–K** Vertical view of conidia. **L** Conidia and conidiophores. **M–O** Conidiophores. *Bars* **A** 2 mm; **B** 40 µm; **C–O** 20 µm

branched to one or two orders in the conidiogenous region, with the distal ends being conidiogenous cells, lateral branches arising just below the septa, often in whorls of up to three, 4- to 8-septate, smooth, dark brown at the base, paler or subhyaline toward the apex, $32-84 \times 2-3.5 \,\mu m$. Conidiogenous cells holoblastic, determinate, apical on the conidiophores or lateral branches, or lateral immediately below septa, smooth, pale brown and thick-walled at the base, paler and thinner-walled toward the apex, 9-12 µm long, 3–4.5μm wide at the base, constricted to 2–3μm wide in the middle, inflating to 4-5 µm wide at the apex, and becoming cupulate after dispersing the conidia. Conidia round, moderately lobed, horizontally flattened, usually consisting of four or rarely of three cells, which adhere horizontally, brown, with the conidial base being very strongly dark brown, 12.5–16.5 μ m (avg. = 14.2 μ m, n = 42) diameter, 7–9.5 μ m (avg. = 7.8 μ m, n = 9) height.

Specimen examined: Leaf of *Pittosporum tobira* Ait. (Pittosporaceae), Naze-shi (Amami Island), Kagoshima Prefecture, Oct 21, 2003, Y. Ono, TFM: FPH-7763.

Note: The genus *Flosculomyces* B. Sutton includes two species (Sutton 1978). *Flosculomyces floridaensis* has larger 4-celled conidia (13–16.5 × 7–8 μ m) and *F. trilobatus* has smaller 3-celled conidia (11.5–13.5 × 7–9 μ m) (Onofri 1984; Sutton 1978). The present fungus has apparently been identified as *F. floridaensis* from the numbers of septation

and the dimension of the conidia. *Flosculomyces floridaensis* has previously been recorded on *Calophyllum inophyllum* L., *Lithocarpus edulis* Nakai, *Pandanus boniensis* Warb., *Pandanus furcatus* Roxb., *Pinus luchuensis* Mayr, and *Podocarpus* sp. in Hong Kong, Japan, and USA (Matsushima 1989; Sutton 1978; Yokoyama and Ito 1989). *Pittosporum tobira* Ait. has been newly added to the list of host plants.

Pseudodidymaria symplocarpi Yas. Ono & Tak. Kobay., sp. nov. Figs. 4, 5

Maculae amphigenae, distinctae, circulares vel irregulares, saepe per venas limitatae, 3–20 mm diametro, fuscae vel nigrae. Stromata substomatalia 20–70 µm diametro, ex hyphis inflatis hyalinis vel pallide flavido-viridibus, 2.5–7 µm latis composita. Conidiophora fasciculata numerosa, per stoma emergentia, macronematica, mononematica, subcylindrica vel subclavata, flexuosa vel sinuosa, continua vel raro septata, pallide brunnea vel brunnea, laevia, 42–83.5 \times 5.5–7.5 µm. Cicatrices conidiales conspicuae, refractae, interdum protuberantes. Conidia solitaria, ellipsoideo-ovoidea, subcylindrica, continua vel 1-septata, hyalina vel pallide pigmentia, laevia, apice rotundata, basi rotundata vel subtruncata et hilo paulo vel non incrassato vix refracto praedita, 17.5–32 \times 8.5–12.5 µm.

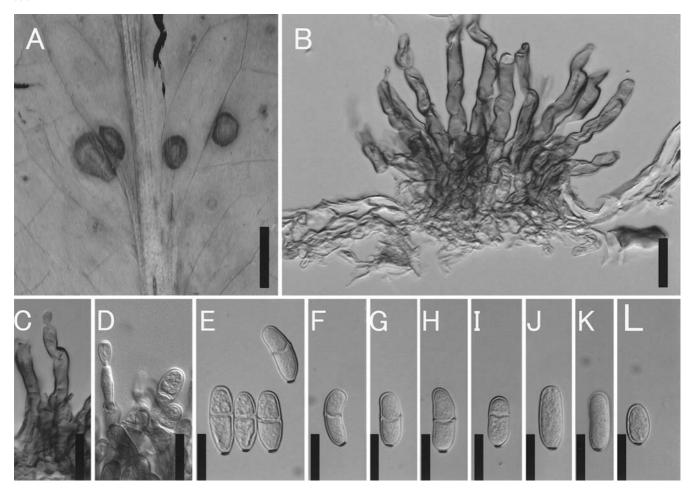


Fig. 4. Pseudodidymaria symplocarpi Yas. Ono & Tak. Kobay. **A** Symptom on a leaf of Symplocarpus nipponicus. **B** Cross section of a conidioma on S. nipponicus. **C,D** Conidia and conidiophores. **E–L** Conidia. Bars **A** 1 cm; **B–L** 20 μm

Holotypus: In foliis vivis *Symplocarpi nipponici* Makino, Sugadaira Lake, Sanada-machi, Nagano Prefecture, Japonia, May 21, 2002, Y. Ono, TFM: FPH-7766 (ex-type culture MAFF 239811).

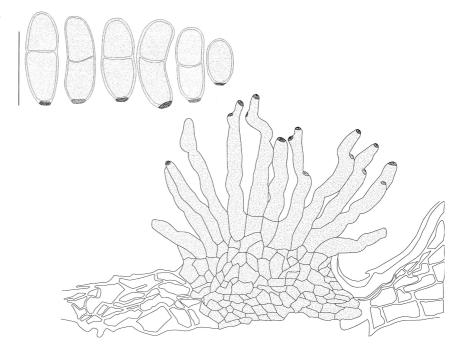
Etymology: Refers to the generic name of the host plant. Leaf spots amphigenous, distinct, circular to irregular, 3–20 mm in diameter, dark brown to black. Stroma amphigenous, brown, 22– $50\,\mu m$ in height, 53– $72\,\mu m$ in diameter. Conidiophores in fascicles, arising from the upper part of the stromata, macronematous, mononematous, subcylindrical, subclavate to flexuous, sinuous, rarely septate, pale brown to brown, smooth, with conspicuous conidial scars, 42– 83.5×5.5 – $7.5\,\mu m$. Conidial scars refractive, intercalary or terminal, occasionally protuberant. Conidia formed singly, ellipsoid-obovoid, subcylindric, 0–1-euseptate, hyaline to slightly pigmented, smooth, rounded at the apex, with the base rounded to subtruncate, 17.5– 32×8.5 – $12.5\,\mu m$ (avg. = $25.3 \times 10.4\,\mu m$, n = 30); hilum sometimes slightly thickened, hardly refractive.

Note: The present fungus is characterized by its conidial morphology and conidial scars and clearly belongs to the genus *Pseudodidymaria* U. Braun (Braun 1998). The genus *Pseudodidymaria* includes three species: *P. aeschynomenes*

U. Braun & Crous found on Aeschynomene (Fabaceae), P. clematidis U. Braun & Rogerson found on Clematis (Ranunculaceae), and P. wyethiae (Ellis & Everh.) U. Braun found on Helianthella (Asteraceae) and Wyethia (Asteraceae) (Braun 1998; Braun et al. 2002). The present fungus subsists on Symplocarpus (Araceae). Regarding the genus Symplocarpus, only one species, Cercospora symplocarpi Peck, has hitherto been discovered for Cercospora and its related genera (Pollack 1987). Pseudodidymaria symplocarpi was obviously different from C. symplocarpi based on the shape and size of its conida. Therefore, a new species, Pseudodidymaria symplocarpi, was described.

Acknowledgments We are grateful to Emi Masuda and Mayumi Arai for their excellent technical assistance during the study. During the survey of plant-inhabiting fungi on Okinoerabu and Amami Island, Tomomi Sato, Yuri Hirooka, Takashi Nishikawa, and Toshiko Furukawa assisted us in collecting the plant materials and in taking photographs. We would especially like to thank Taku Moriguchi, who passed away at the age of 31 in 2004. He was a great help during all our research activities.

Fig. 5. *Pseudodidymaria symplocarpi* Yas. Ono & Tak. Kobay. Conidia and cross section of a conidioma on *Symplocarpus nipponicus*. *Bar* 20μm



References

Braun U (1998) A monograph of *Cercosporella*, *Ramularia* and allied genera (Phytopathogenic Hyphomycetes), vol 2. IHW, Munich

Braun U, Crous PW, Pons N (2002) Annotated list of *Cercospora* species (epithets a-b) described by C. Chupp. Feddes Repert 113:112–127

Deighton FC (1990) Observations on *Phaeoisariopsis*. Mycol Res 94:1096-1102

Dennis RWG (1986) Fungi of the Hebrides. Royal Botanic Gardens, Kew, Surrey

Ellis MB (1961) Dematiaceous Hyphomycetes. III. Mycol Pap 82:1–55 Ellis MB (1963) Dematiaceous Hyphomycetes. IV. Mycol Pap 87:1–42 Ellis MB (1971) Dematiaceous Hyphomycetes. Commonwealth Mycological Institute, Kew, Surrey

Ginns JH (1986) Compendium of plant disease and decay fungi in Canada 1960–1980. Research Branch Publications 1813. Agriculture Canada, Ottawa Matsushima T (1989) Matsushima Mycological Memoirs No. 6. Matsushima Fungus Collection, Kobe

Ono Y, Kobayashi T (2001) Notes on new and noteworthy plantinhabiting fungi from Japan (1). Mycoscience 42:439–446

Ono Y, Kobayashi T (2003) Notes on new and noteworthy plantinhabiting fungi from Japan (2): *Griphosphaerioma zelkovicola* sp. nov. with *Sarcostroma* anamorph isolated from bark of *Zelkova serrata*. Mycoscience 44:109–114

Onofri S (1984) A new species of the genus $\it Flosculomyces$. Mycotaxon 19:385–388

Pollack FG (1987) An annotated compilation of *Cercospora* names. Mycol Mem 12:1–212

Reid D (1985) An annotated list of some fungi from the Channel Islands, mostly from Jersey. Trans Br Mycol Soc 84:709–714

Sutton BC (1978) New and interesting Hyphomycetes from Tampa, Florida. Mycologia 70:784–801

Sutton BC (1980) The Coelomycetes. Commonwealth Mycological Institute, Kew, Surrey

Yokoyama T, Ito T (1989) Descriptive catalogue of IFO fungus collection XI. IFO Res Commun 14:156–161