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FULL PAPER

Yuko Kurihara · Nampiah Sukarno · Muhammad Ilyas
Erny Yuniarti · Wibowo Mangunwardoyo
Ju-Young Park · Rasti Saraswati · Yantyati Widyastuti
Katsuhiko Ando

Indonesian Kickxellales: two species of *Coemansia* and *Linderina*

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Abstract Four species belonging to Kickxellales (Kickxellomycotina) isolated from soil of Indonesia are described and illustrated. Two new species of *Coemansia*, *C. asiatica* and *C. javaensis*, were discovered in South Sulawesi and West Java, and two known species of *Linderina*, *L. pennispora* and *L. macrospora*, were discovered in East Kalimantan and South Sulawesi, respectively. These four species are newly added to the Indonesian mycobiota. A technique for inducing sporulation of *C. javaensis* and *L. macrospora* by adding substances derived from invertebrates such as aphids, nereids, or cladocerans to culture media is described.

Key words *Coemansia asiatica* · *Coemansia javaensis* · Indonesia · *Linderina macrospora* · *Linderina pennispora*

Y. Kurihara[1] (✉) · J.-Y. Park · K. Ando
NITE Biological Resource Center (NBRC), National Institute of Technology and Evaluation, Kisarazu, Chiba, Japan

N. Sukarno
Biology Department, Bogor Agricultural University (IPB), Bogor, Indonesia

M. Ilyas
Research Center for Biology, Indonesian Institute of Sciences (LIPI), Cibinong, Indonesia

E. Yuniarti · R. Saraswati
Soil Biology Division, Soil Research Institute, Bogor, Indonesia

W. Mangunwardoyo
Department of Biology, University of Indonesia, Depok, Indonesia

Y. Widyastuti
Research Center for Biotechnology, LIPI, Cibinong, Indonesia

[1]*Present address:*
Ishigaki Laboratory, OP BIO FACTORY Co., ltd, Tonoshiro 965-3-102, Ishigaki, Okinawa 907-0004, Japan
Tel. +81-980-88-1715; Fax +81-980-88-1716
e-mail: kurihara@opbio.com

Introduction

The order Kickxellales R.K. Benj. (Kickxellomycotina) contains one family and nine genera (Kurihara and Degawa 2006). Although the Kickxellales had been classified traditionally within the class Zygomycetes, recently it was segregated from other orders of this class to establish the subphylum Kickxellomycotina with Harpellales Lichtw. & Manier, Asellariales Manier ex Manier & Lichtw., and Dimargaritales R.K. Benj. (Hibbett et al. 2007). Within the Kickxellomycotina, Kickxellales is phylogenetically closest to the harpellalean genus *Orphella* L. Léger & M. Gauthier (White 2006; White et al. 2006), an inhabitant of stonefly nymph guts (Misra and Lichtwardt 2000). Most species of Kickxellales have been isolated from mammal excrement, soil, rhizosphere soil, other fungi, and cadavers and excrement of soil-dwelling arthropods (Linder 1943; Kwaśna et al. 1999; Benny et al. 2001; Kurihara et al. 2001, 2004; Ho and Hsu 2005; Kurihara and Degawa 2006). Most kickxellalean species are rarely found, except for several species of *Coemansia* (Benjamin 1958, 1979; Kwaśna et al. 1999; Benny et al. 2001).

Coemansia Tiegh. & G. Le Monn. is the largest and the most common genus within the Kickxellales (Kirk 1993). However, few tropical species of *Coemansia* have been described, and most of them have never been rediscovered after their original descriptions, whereas some temperate species appear to be prevalent (Chien 1994; Kurihara et al. 2000, 2008). For example, *C. ceylonensis* Linder, *C. guatemalensis* Thaxt. ex Linder, and *C. kamerunensis* Linder were described from tropical countries (Sri Lanka, Guatemala, and Cameroon) (Linder 1943), and they have never been recollected.

In Indonesia, only 2 species of *Coemansia* have been recorded (Boedijn 1958) among all 34 known species of Kickxellales (Index Fungorum, www.indexfungorum.org, accessed on Oct. 11, 2007); these include *C. erecta* Bainier and *C. reversa* Tiegh. & G. Le Monn. (Boedijn 1958). In this article, we add 4 kickxellalean species, including 2 new species of *Coemansia*, to the Indonesian mycobiota; these

include *C. asiatica* Kurihara & Sukarno, *C. javaensis* Sukarno & Kurihara, *Linderina pennispora* Raper & Fennell, and *L. macrospora* Chang. Herein we describe and illustrate these species, and describe a technique to induce sporulation of *C. javaensis* and *L. macrospora* by adding invertebrate-derived substances to the culture media.

Materials and methods

To isolate kickxellalean species in Indonesia, 77 soil samples were collected from four areas in August and September 2005 and July 2006. The four sampling areas were located in Kupang, West Timor (S 9°48′–10°10′, E 123°49′–124°15′, alt. 26–815 m), Enrekang, South Sulawesi (S 3°16′–3°35′, E 119°45′–119°50′, alt. 72–734 m), Cibinong, West Java (S 6°29′, E 106°50′–106°52′, alt. 171–180 m), and Kutai National Park, East Kalimantan (N 0°22′–0°31′, E 117°27′–117°28′, alt. 26–94 m). The number of samples was 20, 24, 6, and 27, respectively. These soil samples were collected from various environments including tropical savannas, hill forests, arable fields, and lowland tropical rainforests.

The soil samples were treated by the direct inoculation method using crustacean medium or carrot medium, a crustacean baiting method, or an enrichment method (Kurihara et al. 2008). Samples were incubated at 28°–30°C under light/dark conditions (L:D = 12:12). Pure cultures of kickxellalean species that grew from the samples were cultured by transferring a spore mass to a Miura's agar medium plate (Miura and Kudo 1970) with a fine needle. In addition to the Indonesian isolates, a Japanese isolate of *Coemansia* from rabbit excrement was provided by Ms. Kimi Sakai of Odawara-shi and Dr. Yousuke Degawa of Kanagawa Prefectural Museum of Natural History.

All the kickxellalean isolates were identified mainly by optical microscopy. For morphological observations, the isolates were incubated on Miura's or half-strength malt extract-yeast extract agar medium (1/2 ME-YE; Kurihara et al. 2001). When the isolates did not sporulate on Miura's or 1/2 ME-YE media, Miura's medium supplemented with invertebrate-derived substances such as aphids, nereids, or cladocerans was employed to induce sporulation. Among these media, the one that provided the best sporulation of each isolate was used for morphological observation.

To supplement Miura's medium with invertebrate-derived substances, the invertebrates were prepared as follows. Two species of aphids [*Acyrthosiphon pisum* (Harris) and *Aphis craccivora* Koch], which live on *Vicia angustifolia* L., were collected in Chiba, Japan, and stored at –20°C. The aphids were sterilized by dry heat at 160°C for 10 min, and placed onto the surface of Miura's medium plates before inoculating a fungus. A half-teaspoonful of aphids (about 0.07 g) was used for a 9-cm plate. A living nereid [*Cirriformia tentaculata* (Montagu)], which is sold as fishing bait, was autoclaved, dehydrated in a dry heat sterilizer at 50°C, broken into pieces, and then stored at –20°C. Before use, the nereid was sterilized by dry heat and used as same as the aphids. A dried cladoceran (Cladocera, Arthropoda), which is sold as goldfish food, was sterilized by autoclaving or dry heating, and used same as the aphids. Other commercially available invertebrates including dried sakura shrimps (*Sergia lucens* Hansen), mealworm larvae (*Tenebrio* sp.), and krill (Euphausiacea) were prepared as the cladocerans. To induce sporulation, the surface of the agar plates was cut into pieces with a sterilized needle 1 month after inoculation. The treatment is not always essential but often effective for sporulation of some kickxellalean species.

All isolates were incubated at 26°C under light/dark conditions (L:D = 15:9), excluding the isolate of *C. javaensis*, which was incubated at 30°C. Preparation of slides and measurements of each morphological feature were conducted as described in Kurihara et al. (2000).

Based on these studies, two isolates of *Coemansia* and three isolates of *Linderina* Raper & Fennel were recovered from 5 of the 77 soil samples. Morphological examination of the five Indonesian isolates indicated that they represented two undescribed species of *Coemansia*, *L. pennispora*, and *L. macrospora*. The two undescribed species of *Coemansia* were morphologically distinct from all currently known species of the genus. These two *Coemansia* species, *C. asiatica* and *C. javaensis*, and the two *Linderina* species were newly recorded in Indonesia. The Japanese isolate of *Coemansia* was identified as *C. asiatica*. Partial sequences of the nuclear large (LSU) and small subunits of ribosomal DNA (rDNA) of these isolates have been deposited at DDBJ/EMBL/GenBank (accession numbers: AB287996, AB295424–AB295429).

In Indonesia, Boedijn (1958) reported two *Coemansia* species from mammal excrement: *C. erecta* from bat excrement and *C. reversa* from *Isaria* Fr. that was growing on rabbit excrement. In contrast, all the species discovered in this study were isolated from soil.

Taxonomy

Coemansia asiatica Kurihara & Sukarno, sp. nov.

Figs. 1, 2

Coloniae in agaro 1/2 ME-YE luteolae. Hyphae vegetativae hyalinae, septatae. Sporangiophora erecta, septata, 7–19.5 µm lata, simplicia, furcata vel aliquantum trifurcata, asperula. Sporocladia primo divergentia, mox fere parallela ad sporangiophorum, cymbiformia, fere recta vel curvula, asperula, ex stipitibus brevivus 4–9 × 4–7 µm evolventia, 5–9-cellularia, praeter stipites 19–37 × 5–9.5 µm; cellula apicalis sterilis et curvata, 2–9.5 × 2–5 µm. Pseudophialides lageniformes, e cellulis fertilibus sporocladii lateraliter orientes, latrorsae, 5–7.5 × 2–3.5 µm. Sporangiola monospora, incolorata, fusiformia, ventricosa, 8–14(–16) × 2.5–3.5(–4) µm. Sporangiosporae fusiformes, ventricosae, sursum attenuatae, 7–13.5(–15) × 2.5–3.5 µm.

Zygosporae ingnotae.

Holotypus: BO22543, colonia exsiccata in cultura ex solo, Anggeraja, Enrekang, South Sulawesi, Indonesia, Aug. 29, 2005, K. Ando, Y. Widyastuti, R. Saraswati, Y.

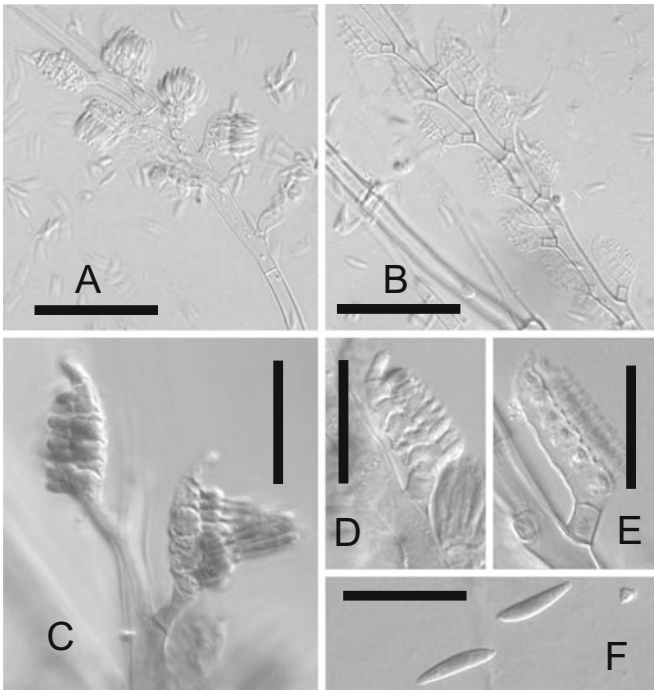

Fig. 1. *Coemansia asiatica* (BTCC-F31). **A** Sporangiophore with sporocladia bearing mature sporangiospores. **B** Sporangiophore with sporocladia after separation of sporangiospores. **C** Sporocladium with immature pseudophialides (*above*) and sporocladium with developing sporangiospores formed on pseudophialides (*below*). **D** Sporocladium after separation of sporangiospores (*above*) and sporocladia with nearly mature sporangiospores (*below*). **E** Sporocladium and pseudophialides after separation of sporangiospores. **F** Sporangiospores. *Bars* **A, B** 50 µm; **C–F** 20 µm

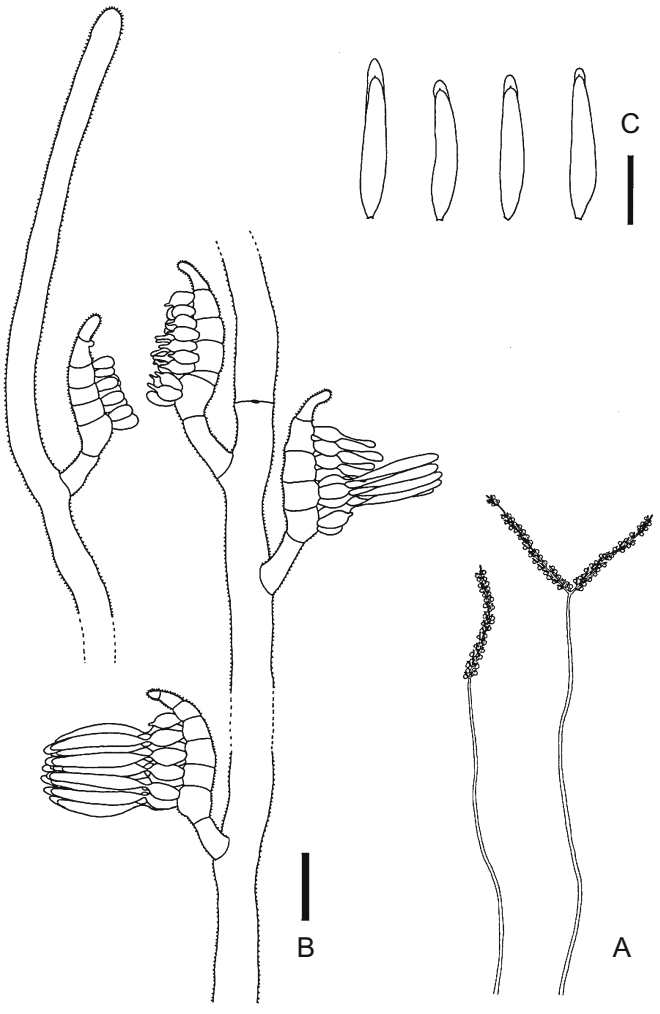

Fig. 2. *Coemansia asiatica* (BTCC-F31). **A** Sketch of the habit of two sporangiophores (not to scale). **B** Upper portion of a sporangiophore bearing four sporocladia with pseudophialides and developing sporangiospores. **C** Four sporangiospores enveloped in sporangiola. *Bars* **B** 20 µm; **C** 10 µm

Lestari, R. Ridwan, S. Ratnakomala, H. Yamamura, J.-Y. Park, et Y. Kurihara leg., Y. Kurihara isol., in Herbario Bogoriensi conservata; BTCC-F31 (= ID05-F0205), cultura viva.

Etymology: Latin *asiaticus*, referring to its localities.

Colonies on 1/2 ME-YE pale yellow. Vegetative hyphae hyaline, septate. Sporangiophores erect, septate, 7–19.5 µm wide, simple, furcate or sometimes trifurcate, asperulate, fertile branches of sporangiophore producing sporocladia laterally, often with intervals that lack sporocladia. Sporocladia divergent to nearly parallel to sporangiophores, boat-shaped, nearly straight to curved shortly, asperulate, stalk short 4–9 × 4–7 µm, composed of 5–9 cells excluding the stalks, 19–37 × 5–9.5 µm, the apical cell sterile and curved, 2–9.5 × 2–5 µm. Pseudophialides flask-shaped, 5–7.5 × 2–3.5 µm, positioned laterally in transverse rows on sporocladia. Sporangiola monosporic, hyaline, fusiform, ventricosus, 8–14(–16) × 2.5–3.5(–4) µm, easily detached at maturity, forming a spore mass. Sporangiospores fusiform, ventricosus, slightly tapering to the apex, 7–13.5(–15) × 2.5–3.5 µm, surrounded by a sporangiole. Zygospores not observed.

Holotype: BO22543 (dried culture), Anggeraja, Enrekang, South Sulawesi, Indonesia, Aug. 29, 2005, isolated from soil under *Cassia fistula* L., recovered by the direct inoculation method using crustacean medium, coll. K. Ando, Y. Widyastuti, R. Saraswati, Y. Lestari, R. Ridwan, S. Ratnakomala, H. Yamamura, J.-Y. Park, and Y. Kurihara, isol. Y. Kurihara, deposited in Herbarium Bogoriense; BTCC-F31 (= ID05-F0205), ex-holotype strain, deposited in Biotechnology Culture Collection Institution, Research Center for Biotechnology-LIPI.

Specimen examined: Iryuda, Odawara, Kanagawa, Japan, Mar. 4, 2003, isolated from rabbit excrement collected from an orchard of *Citrus unshiu* Marc., incubated in a moist chamber, coll. K. Sakai, isol. K. Sakai and Y. Degawa; NBRC 102546 (living culture).

Notes: BTCC-F31 grew and sporulated well on 1/2 ME-YE and Miura's medium with or without the invertebrate-derived supplements (especially with sakura shrimps or mealworm larvae), but slightly degenerated on Sabouraud glucose agar medium (SGA) (Uchiyama 1999) and Miura's medium with aphids. *Coemansia asiatica* resembles *C. mojavensis* R.K. Benj. in producing slightly curved sporangio-

spores. However, sporangiola and sporangiospores of *C. asiatica* are not as conspicuously pointed as those of *C. mojavensis*, and sporocladia of *C. asiatica* are significantly more curved than those of *C. mojavensis*.

The morphology of isolate NBRC 102546 could not be fully characterized because the isolate sporulated poorly on agar media; nevertheless, we identified NBRC 102546 as *C. asiatica*. Because the partial sequence of domains D1 and D2 of the LSU rDNA of BTCC-F31 and NBRC 102546 were concordant with each other (596 bps), and they showed 100% bootstrap affinities by neighbor-joining and most parsimony analyses using known species of Kickxellales (data not shown), although they showed slight differences in the partial sequences of the nuclear small subunit rDNA (1 position in 2478 bp) and internal transcribed spacer regions including 5.8S rDNA (3 positions in 870 bp).

Coemansia species often produce delicate fruity aromas when they are incubated on 1/2 ME-YE and other agar media. The cultures of NBRC 102546 on 1/2 ME-YE produced a narcissus-like fragrance, while those of BTCC-F31 produced no discernible odor.

Coemansia javaensis Sukarno & Kurihara, sp. nov.

Figs. 3, 4

Coloniae in agaro Miurae cum cladoceranti luteolae. Hyphae vegetativae hyalinae, septatae. Sporangiophora erecta, septata, 4.5–12 µm lata, simplicia vel ramosa, asperula. Sporocladia divergentia e sporangiophoris, cymbiformia, asperula, ex stipitibus brevivus 3–6.5(–10) × 3–4.5(–5.5) µm evolventia, 2–5(–7)-cellularia, praeter stipites (14–)16–27(–31.5) × 5–7 µm; cellula apicalis longa, 0–1-septata, (3.5–)4.5–12.5 × (2–)2.5–4(–5) µm, saepe fertilis. Pseudophialides lageniformes, e cellulis fertilibus sporocladii lateraliter orientes, latrorsae, 4–7.5 × 2.5–4 µm. Sporangiola monospora, longa, cylindrica, incolorata, (11–)14–24(–27) × 2–3 µm. Sporangiosporae longae, cylindricae, rectiusculae, (8.5–)12.5–19.5(–24) × 1.5–2.5(–3) µm.

Zygosporae ingnotae.

Holotypus: BO22544, colonia exsiccata in cultura ex solo, Cibinong, West Java, Indonesia, Sept. 1, 2005, K. Ando, R. Ridwan, D.R. Waltam, H. Yamamura, et J.-Y. Park leg., Y. Kurihara isol., in Herbario Bogoriensi conservata; BTCC-F33 (= ID05-F0237), cultura viva.

Etymology: Latin, *javaensis* = relating to Java, referring to the type locality.

Colonies on Miura's medium supplemented with cladocerans pale yellow. Vegetative hyphae hyaline, septate. Sporangiophores erect below, septate, 4.5–12 µm wide, simple or branched, asperulate, sparsely producing sporocladia sympodially. Sporocladia divergent from sporangiophores, boat-shaped, asperulate, with a short stalk of 3–6.5(–10) × 3–4.5(–5.5) µm, composed of 2–5(–7) cells excluding the stalks, (14–)16–27(–31.5) × 5–7 µm, the apical cell long, 0–1 septate, (3.5–)4.5–12.5 × (2–)2.5–4(–5) µm, often fertile. Pseudophialides flask-shaped, 4–7.5 × 2.5–4 µm, produced laterally in transverse rows on sporocladia. Sporangiola monosporic, hyaline, cylindrical, (11–)14–24(–27) × 2–3 µm, easily detached at maturity, forming a spore mass. Sporangiospores long, cylindrical, nearly straight, (8.5–)12.5–19.5 (–24) × 1.5–2.5(–3) µm, surrounded by a sporangiole. Secondary sporangiospores often produced directly from the tip of apical cells of sporocladia without pseudophialides. Zygospores were not observed.

Holotype: BO22544, Cibinong, West Java, Indonesia, Sept. 1, 2005, isolated from soil under an evergreen tree in an experimental garden using the crustacean baiting method, coll. K. Ando, R. Ridwan, D.R. Waltam, H. Yamamura, and J.-Y. Park, isol. Y. Kurihara, deposited in Herbarium Bogoriense; BTCC-F33 (= ID05-F0237), ex-

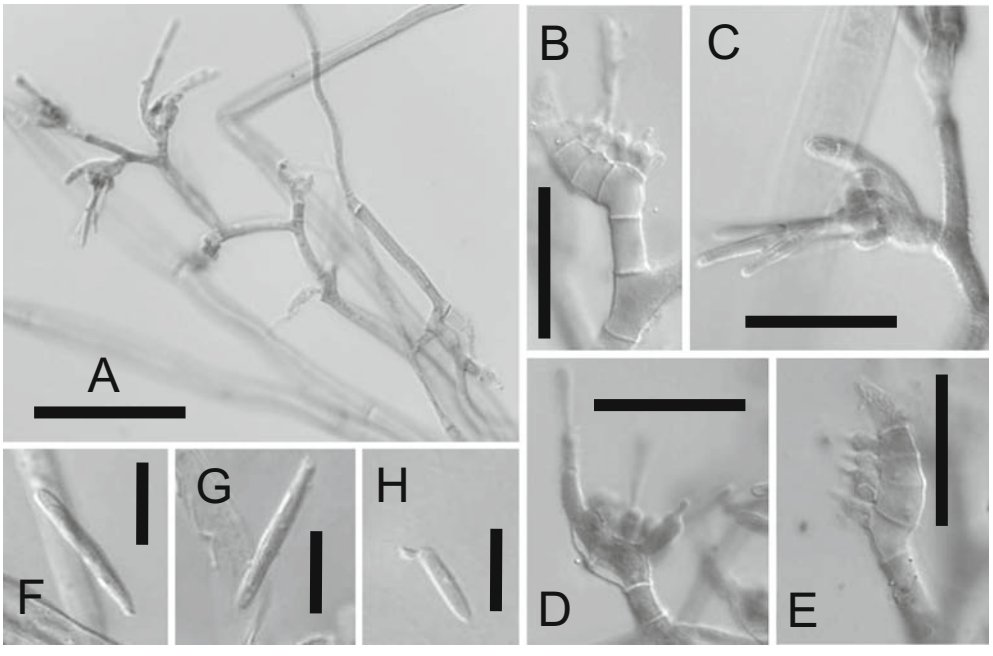

Fig. 3. *Coemansia javaensis* (BTCC-F33). **A** Sporangiophore bearing sporocladia. **B** Sporocladium bearing pseudophialides and a developing sporangiospore. **C** Sporocladium with immature sporangiospores. **D** Sporocladium producing an irregular sporangiospore from the tip of the apical cell. **E** Sporocladium and pseudophialides after separation of sporangiospores. **F–H** Sporangiospores. **A, C, D, F, G** cultured on Miura's medium supplemented with cladocerans; **B, E** on Miura's medium supplemented with aphids; **H** on 1/2 ME-YE medium. *Bars* **A** 50 µm; **B–E** 20 µm; **F–H** 10 µm

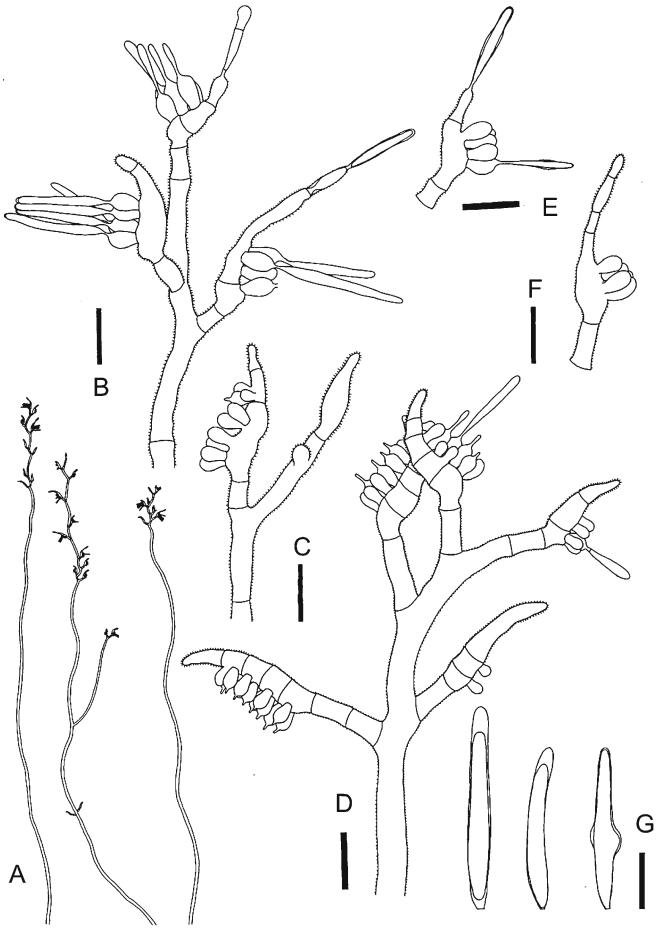

Fig. 4. *Coemansia javaensis* (BTCC-F33). **A** Sketches of the habit of three sporangiophores, the *two on the left* on Miura's medium supplemented with cladocerans, and the *one on the right* on Miura's medium supplemented with aphids (not to scale). **B** Upper portion of a sporangiophore bearing somewhat degenerated sporocladia, the uppermost and the lowermost sporocladia bear irregular sporangiospores from the tips. **C** Two immature sporocladia. **D** Upper portion of a sporangiophore with sporocladia. **E** Two-celled and degenerate sporocladium bearing an irregular sporangiospore. **F** Degenerate sporocladium with an elongated three-celled apical cell. **G** Two mature sporangiospores (*two on left*) and a germinating sporangiospore (*one on right*). **B, C, E–G** incubated on Miura's medium supplemented with cladocerans; **D** incubated on Miura's medium supplemented with aphids. *Bars* **B–F** 20 μm; **G** 10 μm

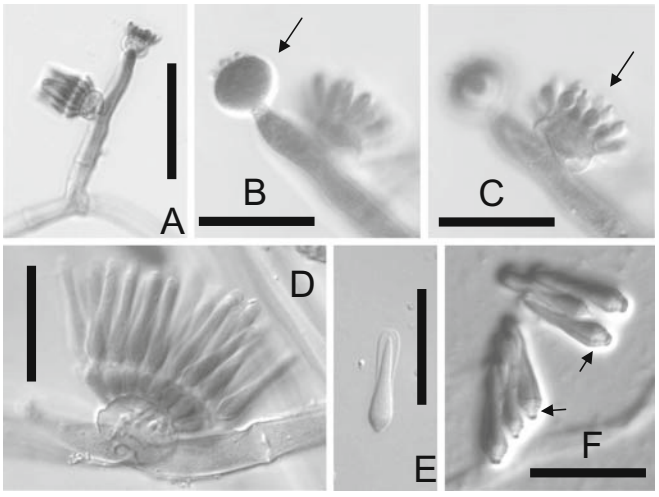

Fig. 5. *Linderina pennispora* (BTCC-F34). **A** Sporangiophore bearing two immature sporocladia. **B** Sporocladium with immature pseudophialides (*arrow*). **C** Sporocladium with developing pseudophialides (*arrow*). **D** Sporocladium with nearly mature sporangiospores. **E** Sporangiospore enveloped in a fin-like sporangiole. **F** Six sporangiospores showing transverse lines of minute spines on sporangiospores (*arrows*). *Bars* **A** 50 μm; **B–F** 20 μm

holotype strain, deposited in Biotechnology Culture Collection Institution, Research Center for Biotechnology-LIPI.

Notes: The ex-type strain (BTCC-F33) grew very slowly, and the colony diameter reached about 2 mm after 2-week incubation at 30°C on 1/2 ME-YE. Although this strain did not sporulate on 1/2 ME-YE, Miura's, and Miura's supplemented with krill, it sporulated abundantly on Miura's with cladocerans or aphids, and sporulated modestly on Miura's with sakura shrimps or mealworms.

Coemansia javanensis resembles *C. ceylonensis* Linder in producing sporocladia composed of small numbers of cells. For example, sporocladia are 2–5(–7)-celled in *C. javanensis*, and 3–5-celled in *C. ceylonensis* (Linder 1943). However, *C. javanensis* differs from *C. ceylonensis* in producing taller sporangiophores, sporocladia that are sharply angled to sporangiophores, longer sporangiospores (11–13 × 2–2.5 μm in *C. ceylonensis*), and abundant production of irregular sporangiospores that develop from the tip apical cell of sporocladia.

Coemansia javaensis frequently produced sporangiospores from the tip apical cell of sporocladia. These irregular sporangiospores were produced directly by sporocladia without pseudophialides, although the tips of the apical cells of sporocladia were delimited, and they functioned as pseudophialides. *Coemansia javaensis* produced the irregular sporangiospores abundantly on Miura's medium supplemented with cladocerans. We observed that nearly half of the sporocladia bore irregular sporangiospores. In contrast, irregular sporangiospores are only rarely produced in old and degenerated cultures of other species of *Coemansia* that were cultivated on rich media such as potato dextrose agar (PDA; Nissui Pharmaceutical, Tokyo, Japan).

Linderina pennispora Raper & Fennell, Am. J. Bot. 38:83, 1962. Figs. 5, 6

Colonies on Miura's medium pale yellow to yellow, nearly 5 mm high. Vegetative hyphae hyaline, septate. Sporangiophores erect, septate, 7.5–12.5 μm wide, branched, asperulate. Sporocladia subglobose to globose, sessile, slightly asperulate when immature, (7–)9–15 × 10–16.5(–19.5) μm, producing pseudophialides on the upper surface. Pseudophialides flask-shaped, 5.5–9(–9.5) × 2.5–3.5(–4) μm, producing a sporangiole apically. Sporangiola monosporic, hyaline, elongate-obclavate to very narrow pyriform, 18–21.5 × 3.5–5 μm, easily detached at maturity, producing a spore mass. Sporangiospores elongate-obclavate, truncate at the base, 16.5–20.5 × 3.5–5 μm, surrounded by a sporangiole; the surface with transverse lines of minute spines. Zygospores not observed.

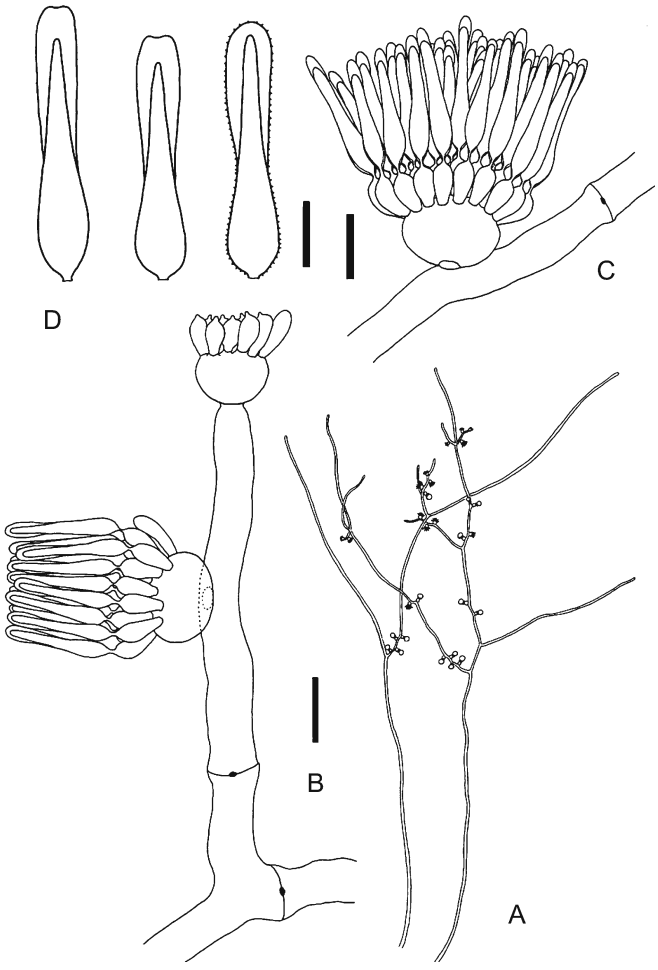

Fig. 6. *Linderina pennispora* (BTCC-F34). **A** Sketch of the habit of two sporangiophores (not to scale). **B** Upper portion of a sporangiophore bearing two sporocladia. Sporocladia with immature pseudophialides (*above*) and developing sporangiospores (*below*). **C** Sporocladia with nearly mature sporangiospores. **D** Three sporangiospores enveloped in fin-like sporangiola. *Bars* **B, C** 20 µm; **D** 10 µm

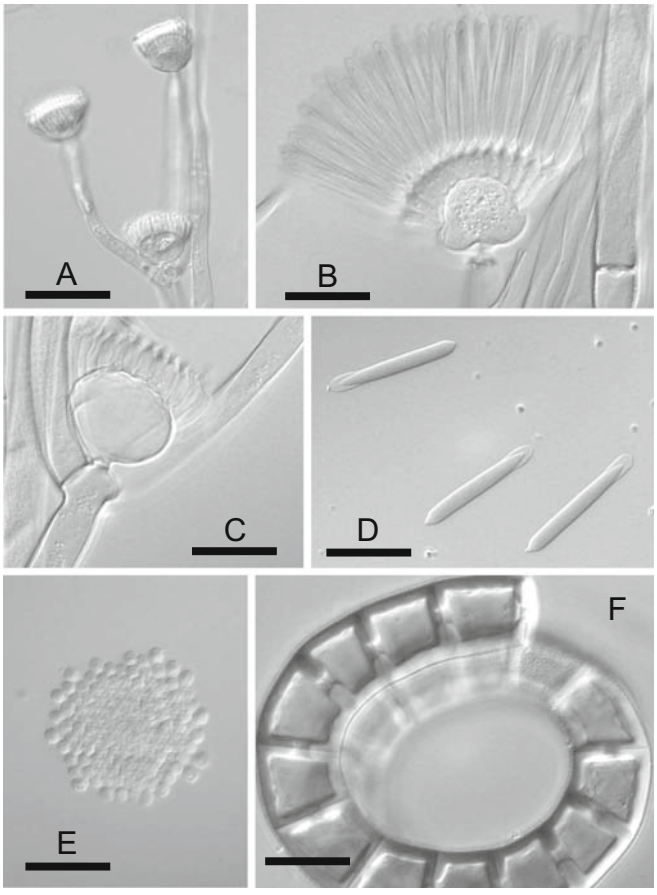

Fig. 7. *Linderina macrospora* (BTCC-F30). **A** Branched sporangiophore bearing three sporocladia after separation of sporangiospores. **B** Sporocladium bearing nearly mature sporangiospores on pseudophialides. **C** Sporocladium after separation of sporangiospores. **D** Three sporangiospores. **E** Mass of sporangiospores, the view from *above*. **F** Thick-walled helicoid structure that may represent a degenerate sporangiophore. *Bars* **A** 50 µm; **B**–**F** 20 µm

Specimen examined: Kutai National Park, East Kalimantan, Indonesia, July 20, 2006, isolated from soil of a lowland tropical rainforest using the crustacean baiting method, coll. K. Ando, Y. Widyastuti, Gina Kartika, R. Ridwan, J.-Y. Park, T. Tamura, and Y. Kurihara, isol. Y. Kurihara; BTCC-F34 (= ID06-F0278), living culture.

Notes: BTCC-F34 grew and sporulated well on Miura's medium with no supplement. The isolate grew well and reached nearly 1 cm high on 1/2 ME-YE, but it sporulated less than on Miura's medium. Two species of *Linderina* have been described with *L. pennispora* representing the type species. *Linderina pennispora* has been found in Liberia, India, and Malaysia (Raper and Fennell 1952; Baijal 1963; Loh et al. 2001), which suggests that it may be a tropical species. Based on published descriptions, the morphology of isolate BTCC-F34 is nearly concordant with that of the ex-type strain from Liberia (Raper and Fennell 1952; Benjamin 1959) and an isolate from Malaysia (Loh et al. 2001) except in the size of its sporocladia. Our isolate produces smaller sporocladia than these two strains; the sizes of sporocladia of the ex-type strain and the Malaysian isolate were 12–25 × 15–30 µm and 13–20(–28) × 22–28(–35) µm, respectively (Benjamin 1959; Loh et al. 2001). However, we emphasisized the size and shape of sporangiospores and identified BTCC-F34 as *L. pennispora*.

Linderina macrospora Chang, Trans. Br. Mycol. Soc. 50: 312, 1967. Figs. 7, 8

Colonies on Miura's medium supplemented with aphids pale yellow to yellow, nearly 1 cm high. Vegetative hyphae hyaline, septate. Sporangiophores erect, septate, 6–18 µm wide, branched, asperulate, producing sporocladia sympodially. Sporocladia subglobose to globose, sessile, asperulate, 13.5–27.5 × 17–35.5 µm, producing pseudophialides on the upper surface. Pseudophialides flask-shaped, asperulate especially when immature, 6.5–14.5 × 2.5–4 µm, producing a sporangiole apically. Sporangiola monosporic, hyaline, cylindrical, 26–32 × 3–4 µm, easily detached at maturity to produce a spore mass. Sporangiospores cylindrical, tapering

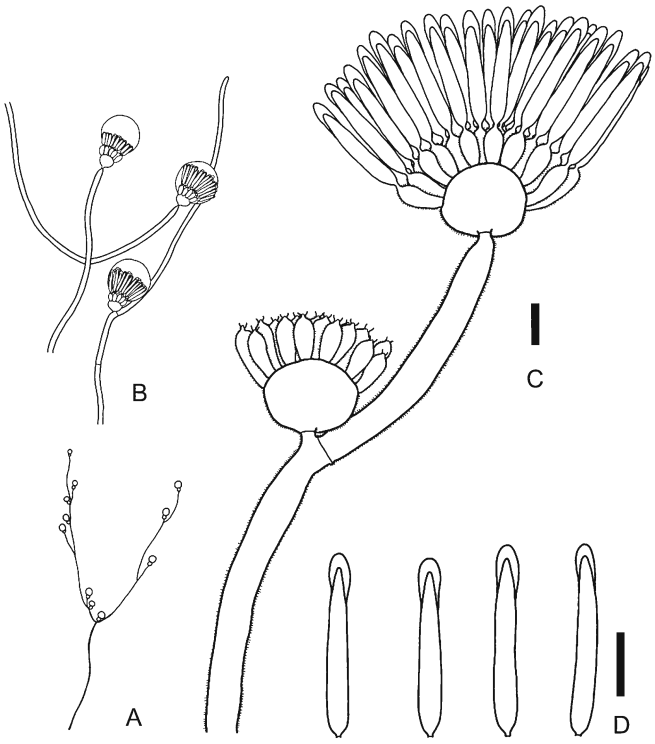

Fig. 8. *Linderina macrospora* (BTCC-F30). **A** Sketch of the habit of a sporangiophore (not to scale). **B** Sketch of two sporangiophores bearing three spore droplets on sporocladia (not to scale). **C** Upper portion of a sporangiophore bearing two sporocladia. Sporocladium bearing immature sporangiospores on pseudophialides (*above*) and a sporocladium and pseudophialides after separation of sporangiospores (*below*). **D** Four sporangiospores within sporangiola. *Bars* 20 μm

to the round apex, truncate at the base, 24–30 × 3–4 μm, surrounded by a sporangiole. Zygospores not observed.

Specimens examined: Anggeraja, Enrekang, South Sulawesi, Indonesia, Aug. 29, 2005, isolated from soil under sugar palm (*Arenga pinnata* Merr.) by the direct inoculation method using crustacean medium, coll. K. Ando, Y. Widyastuti, R. Saraswati, Y. Lestari, R. Ridwan, S. Ratnakomala, H. Yamamura, J.-Y. Park, and Y. Kurihara, isol. Y. Kurihara; BTCC-F30 (= ID05-F0180), living culture. Alla, Enrekang, South Sulawesi, Indonesia, Aug. 30, 2005, isolated from soil under teak (*Tectona grandis* L.f.) by the direct inoculation method using crustacean medium, coll. K. Ando, Y. Widyastuti, R. Saraswati, Y. Lestari, R. Ridwan, S. Ratnakomala, H. Yamamura, J.-Y. Park, and Y. Kurihara, isol. Y. Kurihara; BTCC-F32 (= ID05-F0214), living culture.

Notes: *Linderina macrospora* was originally discovered in Hong Kong (Chang 1967), and rediscovered in southeastern United States (Chien 1971). The morphology of isolate BTCC-F30 was nearly identical to that of the ex-type strain (Chang 1967), although BTCC-F30 produced more abundant sporocladia per sporangiophore and shorter sporangiola and sporangiospores than the ex-type strain.

Adding invertebrate-derived substances to the culture media can be effective for inducing sporulation of *L. macrospora*, although this technique is not always successful. Cultures of two *L. macrospora* isolates (BTCC-F30 and BTCC-F32) grew well on PDA, Miura's medium, cornmeal agar medium (CMA; Nissui Pharmaceutical), and SGA, but sporulated poorly on these media. Sporulation of these two isolates has not been observed on CMA and SGA after incubation for 13 months. Poor sporulation of BTCC-F30 was observed on PDA and Miura's medium after incubation for 5 months and 13 months, respectively. Similarly, BTCC-F32 sporulated poorly on Miura's medium after incubation for 9 months. By way of contrast, BTCC-F30 and BTCC-F32 sporulated well on Miura's medium after incubation for 2 months on Miura's medium medium supplemented with aphids (BTCC-F30) or nereids (BTCC-F32). The substances that induced sporulation differed between the isolates.

In addition, dual cultures of these two isolates also induced sporulation of BTCC-F30. BTCC-F30 sporulated abundantly when it was cultivated with BTCC-F32 on CMA, Miura's medium, or malt extract agar (Nissui Pharmaceutical); however, BTCC-F32 did not sporulate on these media.

Twisted to helicoid structures composed of thick-walled, bright ochre-yellow cells of 10–28.5 × 14–26 μm were often observed on most media used in this study (Fig. 7F). These structures appear to represent degenerate sporangiophores.

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