New species of *Acremonium*, *Cylindrocarpon* and *Verticillium* from soil in the Bonin (Ogasawara) Islands, Japan

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Three new soil fungi from the Bonin (Ogasawara) Islands, Japan are described: Acremonium macroclavatum, characterized by large clavate guttulate conidia; Cylindrocarpon boninense, characterized by 3-7-septate clavate macroconidia, terminal or intercalary chlamydospores, rarely produced unicellular clavate microconidia; and Verticillium hahajimaense, characterized by conidial heads bearing cylindrical conidia, and catenulate chlamydospores.

Key Words——Acremonium macroclavatum; Cylindrocarpon boninense; Hahajima; soil fungi; Verticillium hahajimaense.

During studies on lignocellulose and dioxin-decomposing fungi, a total of 370 soil isolates obtained from the Bonin islands were examined, and several new species were found including a *Dactylella* species, as reported previously (Watanabe et al., 2001 a, b).

In this study, new species of *Acremonium* Link: Fr., *Cylindrocarpon* Wollenw. and *Verticillium* Nees from Hahajima, the Bonin (Ogasawara) Islands soil are reported.

The Acremonium species is characterized by simple conidiophores with terminal spore heads containing large clavate, guttulate conidia; the *Cylindrocarpon* species by 3-7-septate clavate macroconidia, clavate microconidia and chlamydospores; and the *Verticillium* species by erect, verticillate conidiophores with the terminal spore heads containing eguttulate cylindrical conidia and catenulate chlamydospores.

These fungi may be differentiated from all previously described and accepted species of the respective genera, and thus, they are described and illustrated as new in this study.

Materials and Methods

The new Acremonium species was isolated by the direct inoculation method (Watanabe, 1989), the Cylindrocarpon species by the bait method with both cucumber seeds (Watanabe, 1981, 1994) and toothpicks (Watanabe et al., 2001c, d), and the Verticillium species by the bait method with cucumber seeds.

Colony colors (upper surface and reverse) in plates were determined according to Ridgway (1912).

Living cultures were deposited at Bioconsortia Program Laboratory, National Institute of Advanced Industrial Science and Technology (AIST), Ministry of Economy, Trade and Industry, and the Gene Bank, Ministry of Agriculture, Forestry and Fisheries (MAFF), National Institute of Agrobiological Sciences in Tsukuba, Japan.

Results and Discussion

All fungi studied grew well on the conventional agar media tested including both home-made PDA and commercial PDA "Nissui" (Nissui Pharmaceutical Co., Ltd, Tokyo, Japan), Difco corn meal agar (CMA), Difco malt agar (MA) and Difco YM agar(YM). Formation of teleomorphs or other types of sporulation was not observed in these cultures or in water agar cultures with inocula originated from these rich cultures.

Acremonium macroclavatum Ts. Watanabe, sp. nov.

Figs. 1, 2, 10

Coloniae in agaro decocto tuberosum pallide aurantiacae, zonatae, mycelio aerio praesentae vel prostratae. Conidiophora erecta, plerumque simplicia, raro aliquoties ramosa, hyalina, plerumque 2-3-septata, 20-200 μ m longa, prope basin 2-4(-6) μ m et ad apicem 1-2 μ m lata, sursum attenuata. Cellulae conidiogenae terminales, phialidicae. Conidia apicales, in capitulis mucidis 12-40 μ m diam conglomerata, clavata, cylindrica vel ellipsoidea, hyalina, aseptata, guttulata, (5-)8-14× 2.0-3.6(-4.0) μ m. Chlamydosporae absunt.

Holotypus: AIST 0050, colonia exsiccata in cultura ex solo, Chibusayama, Hahajima, Bonin Insula, Tokyo,



Figs. 1, 2. Acremonium macroclavatum (MAFF 238162). 1. Conidiophores and masses of conidia. 2. Conidia and smooth and crustose hyphae. Scale bar = 10 μm

- Figs. 3–6. Cylindrocarpon boninense (MAFF 238163). 3. Habit showing aggregated sporulation. 4. Conidiophore and masses of macroconidia. Macroconidia, microconidia and an apical part of conidiophore in the inset. 5. Aggregate of macroconidia. 6. Chlamydospore. Scale bars: 3=20 μm; 4–6=10 μm.
- Figs. 7–9. Verticillium hahajimaense (MAFF 238172). 7. Sporulation habit. 8. Phialides and conidia. 9. Chlamydospores in chains. Scale bars: 7=20 μm; 8, 9=10 μm.



Fig. 10. Acremonium macroclavatum. A. Simple and branched conidiophores. B. Conidia. C. Apexes of unproliferated and proliferated conidiophores. Scale bar=10 μ m.

Japonia, 14 Jan. 2000, T. Watanabe. Cultura viva: MAFF 238162 (=TW 00-50).

Etymology: On the basis of the larger clavate conidia.

PDA colonies slightly aerial, prostrate, slightly zonate, pale yellow orange, reverse pale yellow orange (Ridgway, 1912). Conidiophores (phialides) erect, most-ly simple or rarely branched, usually 2-3-septate, hyaline, gradually tapering towards apexes usually $20-200 \,\mu\text{m}$ long, $2-4(-6) \,\mu\text{m}$ wide basally, $1-2 \,\mu\text{m}$ apically with terminal slimy conidial heads that are $12-40 \,\mu\text{m}$ in diam, occasionally proliferated from the apices or conidial heads. Conidia phialosporous, clavate, cylindrical or ellipsoidal, hyaline, 1-celled, mostly bi-guttulate, $(5-)8-14 \times 2.0-3.6(-4.0) \,\mu\text{m}$. Hyphae occasionally crustaceous, $1.8-6.0 \,\mu\text{m}$ broad. Chlamydospores absent.

This fungus grew and sporulated well on CMA, MA and YM, and the latter two cultures resemble a pale yellow orange-pigmented PDA colony. On CMA, the mycelia are thin, semi-transparent and slightly colored.

The fungus resembles *A. obclavatum* W. Gams (Gams et al., 1984) in forming exclusively solitary phialides and clavate conidia in the section Albo-lanosa (Morgan-Jones and Gams, 1982), but differs from the latter in forming conidial heads and larger guttulate conidia ($(5-)8-14 \times 2.0-3.6(-4.0) \mu$ m) (Figs. 1, 2, 10A, B). The conidia of *A. obclavatum* are arranged in imbricate chains and are smaller ($2.0-4.5 \times 1-2 \mu$ m). In addition, the conidia of *A. macroclavatum* are larger than those of various *Acremonium* species in the previous

reports (Gams, 1971, 1975; Samuels, 1976).

Cylindrocarpon boninense Ts. Watanabe, sp. nov.

Figs. 3–6, 11

Coloniae in agaro decocto tuberosum luteo-brunneae vel pallide vinaceae, mycelio aerio praesentae; reversum vinaceum. Conidiophora (phialides) simplicia, hyalina, erecta, recta, $60-200 \ \mu m$ longa, ad basim $3.2-5.0 \ \mu m$ et ad apicem $1.2-1.6 \ \mu m$ lata, sursum attenuata. Conidia phialosporica, hyalina, mucosa, in globulis agglutinata, dimorphica: macroconidia clavata cylindrica, ellipsoidea vel fusiformia, recta vel raro curvata, basi truncata, 3-7 septata, $(20-)34-72(-98) \times 5-7 \ \mu m$; microconidia clavata, aseptata, basi apiculata, $7-30 \times 2.6-4.0 \ \mu m$. Chlamydosporae globsae vel subglobosae, granulatae, $9-18 \ \mu m$ diam.

Holotypus: AIST 0062, colonia exsiccata in cultura ex solo: Miyukigahama, Hahajima, Bonin Insula, Tokyo, Japonia, 15 Jan. 2000, T. Watanabe, cultura viva: MAFF 238163(=TW 00-62).

Etymology: From the name of The Bonin islands, referring to the type locality.

PDA colony aerial, Dresden brown, buckthorn brown or ochraceus tawny, reverse Dresden brown, buckthorn brown or ochraceus tawny (Ridgway, 1912). Conidiophores (phialides) simple, hyaline, erect, straight, 60–200 μ m tall or more, 3.2–5.0 μ m wide basally, 1.2–1.6 μ m wide apically, gradually tapering toward apexes with



Fig. 11. Cylindrocarpon boninense. A. Conidiophore and macroconidia. B. Macroconidia. C. Microconidia. D. Conidiophores. E. Chlamydospores. Scale bar=10 μ m.

conidial masses. Conidia phialosporous, slimy, agglutinated into globules at the apices of phialides, hyaline, dimorphic; (1) macroconidia clavate or cylindrical, straight or occasionally slightly curved, truncate basally 3-7-septate, (20–)34–72(–98) × 5–7 μ m; (2) microconidia rarely formed, clavate, one-celled, apiculate at one end, 7–30 × 2.6–4.0 μ m. Chlamydospores globose or subglobose, granulate, terminal or intercalary, 9–18 μ m in diam. Hyphae 4.0–5.6 μ m broad.

This fungus grew well on CMA, MA and YMA, and the latter two cultures resemble a yellowish brown or greenish brown PDA colony covered with white mycelia. On CMA, the mycelia are non-aerial, thin, and semi-transparent.

Among *Cylindrocarpon* species with chlamydospores and more than 3-septate macroconidia, this fungus resembles *C. reteaudii* Bugnic. with cylindrical macroconidia (80–110 μ m long), *C. tonkinense* Bugnic. with clavate macroconidia (18–40 μ m long), *C. decombeus* Wollenw. with curved, cylindrical to fusoid macroconidia (5–5.5 μ m wide), and *C. olidum* (Wollenw.) Wollenw. with curved, cylindrical to claviform macroconidia (7–10 μ m wide) (Booth, 1966). However, the macroconidia ((20–)34–72(–98)×5–7 μ m) (Figs. 4, 5, 11A, B) of this fungus are shorter, longer, wider or narrower than those of the other species.

Verticillium hahajimaense Ts. Watanabe, sp. nov.

Figs. 7-9, 12

Coloniae in agaro decocto tuberosum brunneae, mycelio aerio praesentae; reversum brunneum. Conidiophora erecta, hyalina, recta, 220–350 μ m alta, ad basim 2.6–4 μ m lata, plerumque ramosis, multos verticillos phialidum ferentia. Phialides 20–50 μ m longae, sursum angustatae, collaretta inconspicua. Conidia phialosporica, in capitulis 10–12 μ m diam conglomerata, hyalina, cylindrica, 8–10×2.0–2.4 μ m. Chłamydosporae brunneae, globosae, granulatae, catenulatae, 9–16 μ m diam.

Holotypus: AIST 0065, colonia exsiccata in cultura ex solo, Minamizaki, Hahajima, Bonin Insula, Tokyo, Japonia, 15 Jan. 2000, T. Watanabe. Cultura viva: MAFF 238172 (=TW 00-65).

Etymology: From the name of Hahajima, the type locality.

PDA colony aerial, Sudan brown or snuff brown, reverse Sudan brown or snuff brown (Ridgway, 1912). Conidiophores erect, hyaline, straight, $220-350 \times 2.6-4.0 \,\mu\text{m}$, mostly branched with verticillate phialides, bearing terminal spore masses, mostly $10-12 \,\mu\text{m}$ in diam, phialides gradually tapering toward tips, $20-50 \,\mu\text{m}$ long, collarette inconspicuous. Conidia hyaline, cylindrical, $8-10 \times 2-2.4 \,\mu\text{m}$. Chlamydospores brown, globose, granulate, formed in chains, $9-16 \,\mu\text{m}$ in diam.

This fungus grew well on CMA, MA and YMA, and the latter two cultures resemble a pale brown to brown PDA colony covered with white mycelia. On CMA, the mycelia are thin and semi-transparent.

This fungus is characterized by chlamydospores in chains (Figs. 9, 12D) and asymmetrical subglobose or



Fig. 12. Verticillium hahajimaense. A. Conidiophore with verticillate phialides and conidial masses. B. Apical part of conidiophore with verticillate phialides and conidial masses.
C. Conidia. D. Chlamydospores in chains. Scale bars: A, B, D=10 μm; C=5 μm.

various-shaped spore heads (Figs 7, 12A, B) composed of cylindrical conidia (Figs. 8, 12C). It resembles *V. guttulatum* W. Gams (Gams and Zaayen, 1982) in conidial shape and dimensions, but differs in forming eguttulate conidia and chlamydospores in chains.

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