

## Two new heterothallic *Neosartorya* from African soil

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*Neosartorya nishimurae* and *N. otanii*, two new heterothallic species isolated from African forest soil, are described and illustrated. *Neosartorya nishimurae* is characterized by its rapid growth on Czapek and malt extract agars, white cleistothecia, lenticular ascospores with two broad equatorial crests and echinulate convex surfaces, and large ellipsoidal conidia with a lobate-reticulate wall. *Neosartorya otanii* is characterized by its rapid growth on Czapek and malt extract agars, lenticular ascospores with two widely separated equatorial crests and tuberculate or lobate-reticulate convex surfaces, and globose to broadly ellipsoidal conidia with a micro-tuberculate wall.

Key Words—Ascomycetes; *Aspergillus*; heterothallic *Neosartorya*; soil fungi; systematics.

Most members of the ascomycete genus *Neosartorya* Malloch & Cain (Malloch and Cain, 1972) in the Eurotiales are soil-fungi with cosmopolitan distributions, abundantly occurring nearly everywhere, and are active in decomposing organic materials; some species predominantly cause spoilage in heat processed food products.

In the course of a survey of thermophilic and thermotolerant soil-borne fungi, two heterothallic *Neosartorya* have been isolated from forest soil in Kenya. Although *Neosartorya* includes more than 20 species, only 3 were only known to be heterothallic: *N. fennelliae* Kwon-Chung & Kim (Kwon-Chung and Kim, 1974; Takada et al., 1986), *N. spathulata* Takada & Udagawa (Takada and Udagawa, 1985); and *N. udagawae* Horie et al. (Horie et al., 1995).

The first species, *N. fennelliae*, isolated from eyeballs of laboratory rabbits in USA, is characterized by lenticular ascospores with two broad equatorial crests and delicately reticulate convex walls due to the shallow ridges (Takada et al., 1986; Samson et al., 1990; Horie, 1992; Fig. 14 in this paper). The second species, *Neosartorya spathulata*, was isolated from Taiwan soil and is distinguished from *N. fennelliae* by its yellow ascomata and lenticular ascospores possessing almost smooth convex walls, as opposed to reticulate walls in *N. fennelliae* (Takada and Udagawa, 1985; Samson et al., 1990; Horie, 1992; Fig. 15 in this paper). In addition *N. spathulata* has two types of conidiophores and its conidia are much larger than those of *N. fennelliae*. The third species, *Neosartorya udagawae* Horie et al., was species isolated from plantation soil in Brazil. It differs from the

above two species in its rather spherical ascospores with an irregular ornamentation composed of two indistinct narrow equatorial crests and several ribs in a somewhat reticulate pattern (Horie et al. 1995; Fig. 16 in this paper).

Two phylogenetic analyses of these heterothallic *Neosartorya* have been reported. Geiser et al. (1998) recently reported from their molecular study on members of *Aspergillus* sect. *Fumigati* that “forcing *N. spathulata* and *N. fennelliae* to form a single clade did not result in a significantly less likely tree,” and that, consequently, evolution of heterothallism in the *Aspergillus* sect. *Fumigati* may represent a single event. More recently Varga et al. (2000) showed that *N. udagawae* belongs to a different clade than the other two heterothallic species, *N. spathulata* and *N. fennelliae*. According to their data, the large genetic distances observed between *N. udagawae* and *N. fennelliae* (0.092), and between *N. udagawae* and *N. spathulata* (0.095) as compared to those observed, e.g., between *N. udagawae* and *N. aureola* (0.036) or between *N. fennelliae* and *Neosartorya* sp. NRRL 4179 (0.041) also indicate that these species are not closely related.

The two isolates from Kenya differ sufficiently from the three known species of heterothallic *Neosartorya* and are described here as new species. The type specimens studied and living cultures derived from the types are deposited at the Natural History Museum and Institute, Chiba (CBM).

### Taxonomy

*Neosartorya nishimurae* Takada, Horie & Abliz, sp. nov.  
Figs. 1, 3–7

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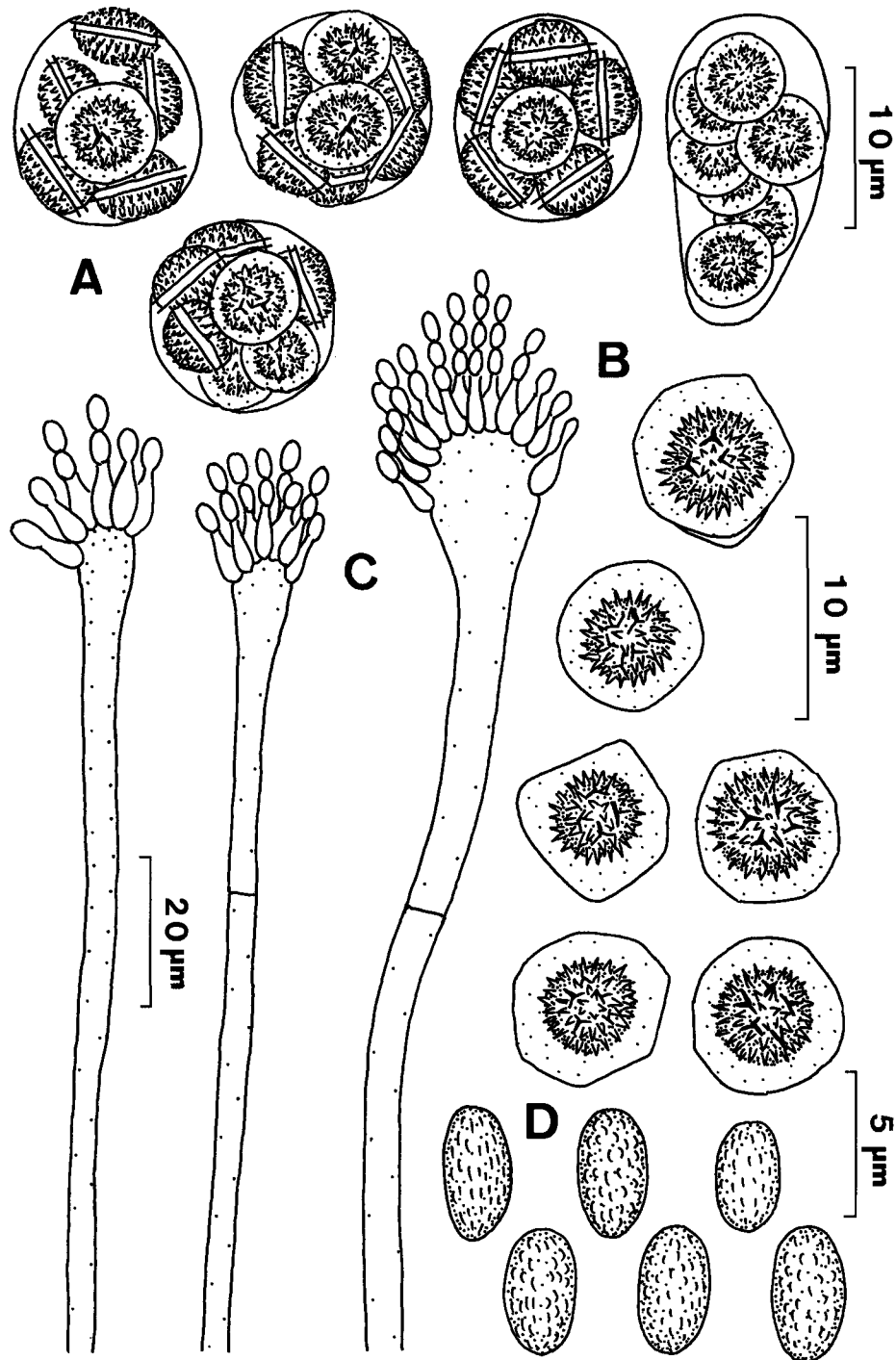


Fig. 1. *Neosartorya nishimurae*. A. Asci. B. Ascospores. C. Aspergilla. D. Conidia.

Fungus heterothallicus. Cleistothecia alba, globosa vel subglobosa vel ovoidea,  $290\text{--}370 \times 200\text{--}370 \mu\text{m}$ , cum hyphis aeriis laxè intricatis circumdata; peridium tenue, membranaceum, ex cellulis angularibus  $2.5\text{--}12.5 \mu\text{m}$  diam compositum. Asci octospori, globosi vel ovoidei vel late ellipsoidei,  $14\text{--}19 \times 10\text{--}14 \mu\text{m}$ , evanescentes. Ascosporae hyalinae vel dilute flavo-brunneae, late lenticulares, praeter cristas  $4\text{--}5.5 \times 4\text{--}5 \mu\text{m}$ , duabus cristis aequatorialibus  $0.8\text{--}2 \mu\text{m}$  latis

praeditae; superficies convexae echinulatae. Status anamorphus: *Aspergillus nishimurae*.

***Aspergillus nishimurae*** Takada, Horie & Abliz, anam. nov.

Capitula conidica brevi-columnaria vel columnaria. Conidiophora ex mycelio basali vel hyphis aeriis orientia; stipes hyalini vel dilute flavo-brunnei, usque  $270 \mu\text{m}$  longi, medio  $3\text{--}5 \mu\text{m}$  crassi, leves; vesiculae hemisphaericae vel ampulliformes,  $4.5\text{--}17 \mu\text{m}$  diam. Aspergilla

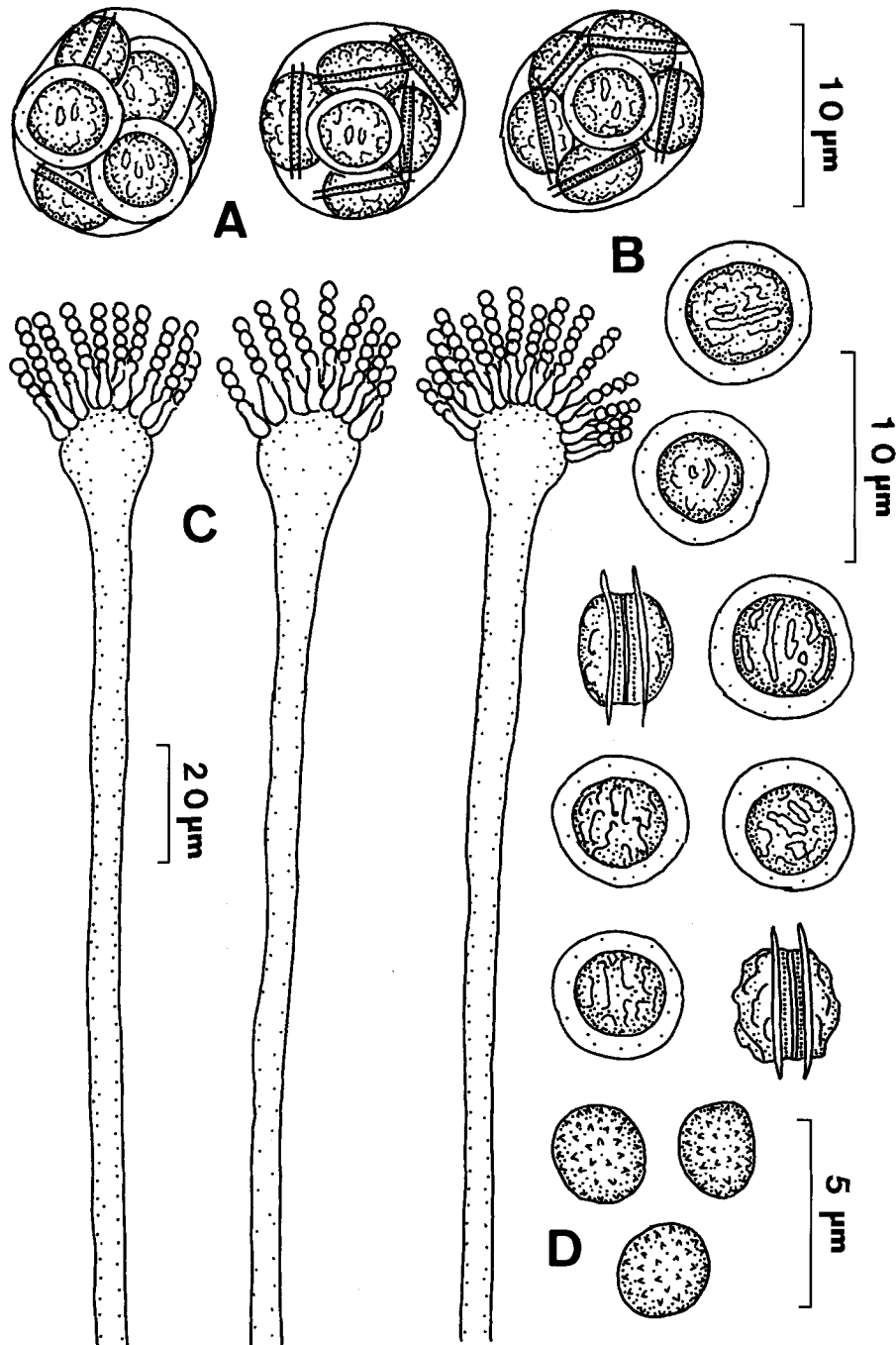


Fig. 2. *Neosartorya otanii*. A. Asci. B. Ascospores. C. Aspergilla. D. Conidia.

uniserialia; phialides dimidium superiore vesiculae obducentes, ampulliformes,  $6-8 \times 2-3 \mu\text{m}$ . Conidia hyalina, in massa viridula, ellipsoidea,  $3.5-4(-5) \times 2-3 \mu\text{m}$ , lobatim reticulata. Status teleomorphus: *Neosartorya nishimurae*.

Coloniae in agar Czapekii celeriter crescentes, viridi-griseae vel griseo-virides; conidiogenesis abundantia; reversum griseo-aurantiacum vel brunneo-aurantiacum.

Coloniae in agaro maltoso celeriter crescentes, albae vel flavo-albae; conidiogenesis dispersa; reversum griseo-aurantiacum vel brunneo-aurantiacum.

Holotypus: CBM-FA-919, colonia exsiccata ex cruce CBM-FA-910 (A)  $\times$  CBM-FA-911 (a), utraque ex solo sylve, in Nairobi, Kenya, Africa, 10 VIII 1974, a M. Takada isolata et ea collectione fungorum Musei et Instituti Historiae Naturalis Chiba (CBM) conservata.

Etymology: named after Professor Kazuko Nishimura, Research Center for Pathogenic Fungi and Microbial Toxicoses, Chiba University, eminent medical mycologist.

Colonies on Czapek agar (CzA) spreading broadly, attaining a diam of 76–80 mm in 14 d at 25°C, Greenish

Grey (30B2, after Kornerup and Wanscher, 1978) to Greyish Green (29B3), floccose, consisting of a thin mycelial felt; conidiogenesis abundant; reverse Greyish Orange (5B3) to Brownish Orange (5C4).

Colonies on malt extract agar (MEA) spreading broadly, attaining a diam of 80–84 mm in 14 d at 25°C, white to Yellowish White (4A2), floccose, consisting of a thin mycelial felt; white cleistothecia abundantly produced after pairing; conidiogenesis sparse; reverse Greyish Orange (5B3) to Brownish Orange (5C4).

Heterothallic. Cleistothecia produced only when dissimilar mating types are paired, superficial, white, globose to subglobose or ovoid, 290–370 × 200–370 µm, surrounded by a loose covering of hyaline to pale yellowish brown aerial hyphae; peridium hyaline to pale yellowish brown, thin, membranaceous, consisting of angular, 2.5–12.5 µm diam cells. Asci 8-spored, globose to ovoid or broadly ellipsoidal, 14–19 × 10–14 µm, evanescent at maturity. Ascospores hyaline to pale yellowish brown, broadly lenticular, spore body 4–5.5 × 4–5 µm, provided with two broad equatorial crests measuring 0.8–2 µm wide; convex surfaces echinulate (LM and SEM).

Conidial heads grayish green to dull green, columnar to loose columnar, 100–155 × 40–85 µm. Conidiophores arising from the basal mycelium or aerial hyphae, hyaline to pale yellowish brown, smooth-walled, up to 270 µm long, 3–5 µm wide at the middle; vesicles hyaline to pale grayish green, hemispherical to flask-shaped, 4.5–17 µm in diam. *Aspergilla uniseriate*; phialides pale grayish green to pale grayish olive, covering the upper half of the vesicle, flask-shaped, 6–8 × 2–3 µm. Conidia hyaline, grayish green to dull green in mass, ellipsoidal, 3.5–4(–5) × 2–3 µm, lobate-reticulate (LM and SEM).

At 37°C, growth is more rapid than at 25°C.

Isolation: forest soil, Nairobi, Kenya, 10 August 1974, coll. M. Takada.

Holotype: The type specimen, CBM-FA-919, is a dried culture from a pair of the isolates, CBM-FA-910 (A) × CBM-FA-911 (a). Deposited in herb. CBM.

This fungus is readily distinguished from other species of heterothallic *Neosartorya* by the echinulate ornamentation of its ascospore convex walls and its large ellipsoidal conidia with a lobate-reticulate wall.

***Neosartorya otanii*** Takada, Horie & Abliz, sp. nov.

Figs. 2, 8–13

Fungus heterothallicus. Cleistothecia alba vel flavo-alba, globosa vel subglobosa, 150–270 µm diam, cum hyphis aeriis laxe intricatis circumdata; peridium tenue, membranaceum, ex cellulis angularibus 3–15 µm diam compositum. Asci octospori, globosi vel subglobosi vel late ellipsoidei, 10–13 × 8–11 µm, evanescentes. Ascospores hyalinae vel dilute flavo-brunneae, late lenticulares, praeter cristas 4–5 × 3.5–4.5 µm, duabus cristis aequatorialibus 1 µm, latis praeditae, superficies convexae tuberculatae vel lobatim reticulatae. Status anamorphus: *Aspergillus otanii*.

***Aspergillus otanii*** Takada, Horie & Abliz, anam. nov.

Capitula conidica griseo-viridia, radiantia vel brevi-

columnaria. Conidiophora ex mycelio basali vel hyphis aeriis orientia; stipites hyalini vel dilute flavo-brunnei, usque 410 µm longi, medio 4–7 µm crassi, leves; vesiculae hemisphaericae vel ampulliformes, 13–16 µm diam. *Aspergilla uniseriata*; phialides dimidium superiore vesiculae obducentes, ampulliformes, griseo-virides vel dilute virides, 4–7 × 1.5–2.5 µm. Conidia griseo-viridia vel dilute viridia, globosa vel subglobosa vel late ellipsoidea, 2–3.5 × 2–3 µm, micro-tuberculata. Status teleomorphus: *Neosartorya otanii*.

Coloniae in agaro Czapekii celeriter crescentes, albae vel flavo-albae; conidiogenesis sparsa; reversum brunneo-aurantiacum.

Coloniae in agaro maltoso celeriter crescentes, griseo-virides vel herbeo-virides; conidiogenesis dispersa; reversum griseo-flavum.

Holotypus CBM-FA-914, colonia exsiccata ex cruce, CBM-FA-912 (A) × CBM-FA-913 (a), utraque ex solo sylvae, in Nairobi, Kenya, Africa, 15 XI 1974, a M. Takada isolata et ea collectione fungorum Musei et Instituti Historiae Naturalis Chiba (CBM) conservata.

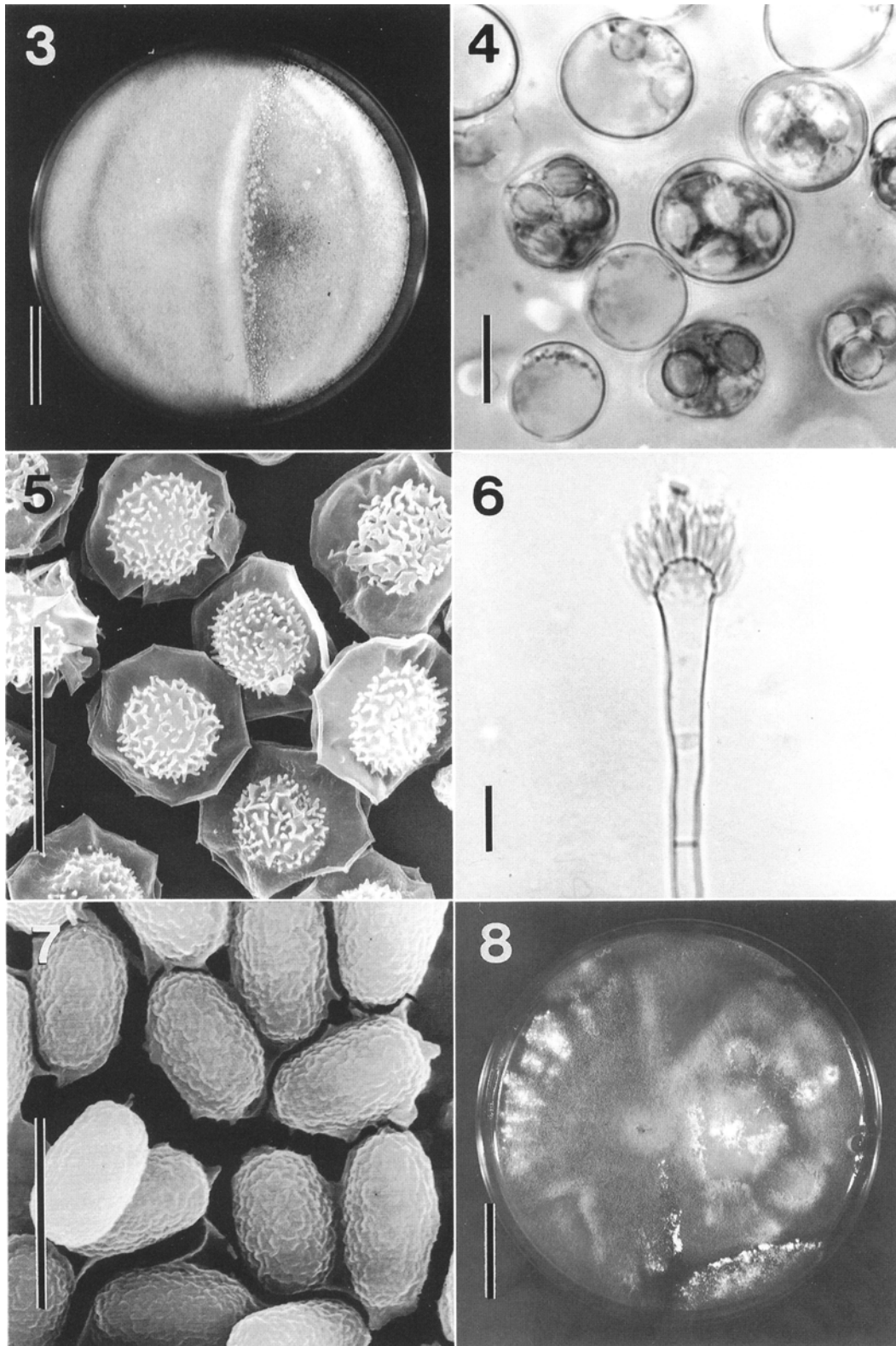
Etymology: named in memory of the late Dr. Yoshio Otani, National Science Museum Tokyo, eminent mycologist, whose research contributed so much to the describing of the Asian mycoflora in ascomycete systematics.

Colonies on CzA spreading broadly, attaining a diam of 81–82 mm in 14 d at 25°C, white, floccose, consisting of a thin mycelial felt; conidiogenesis sparse; reverse Brownish Orange (5C4).

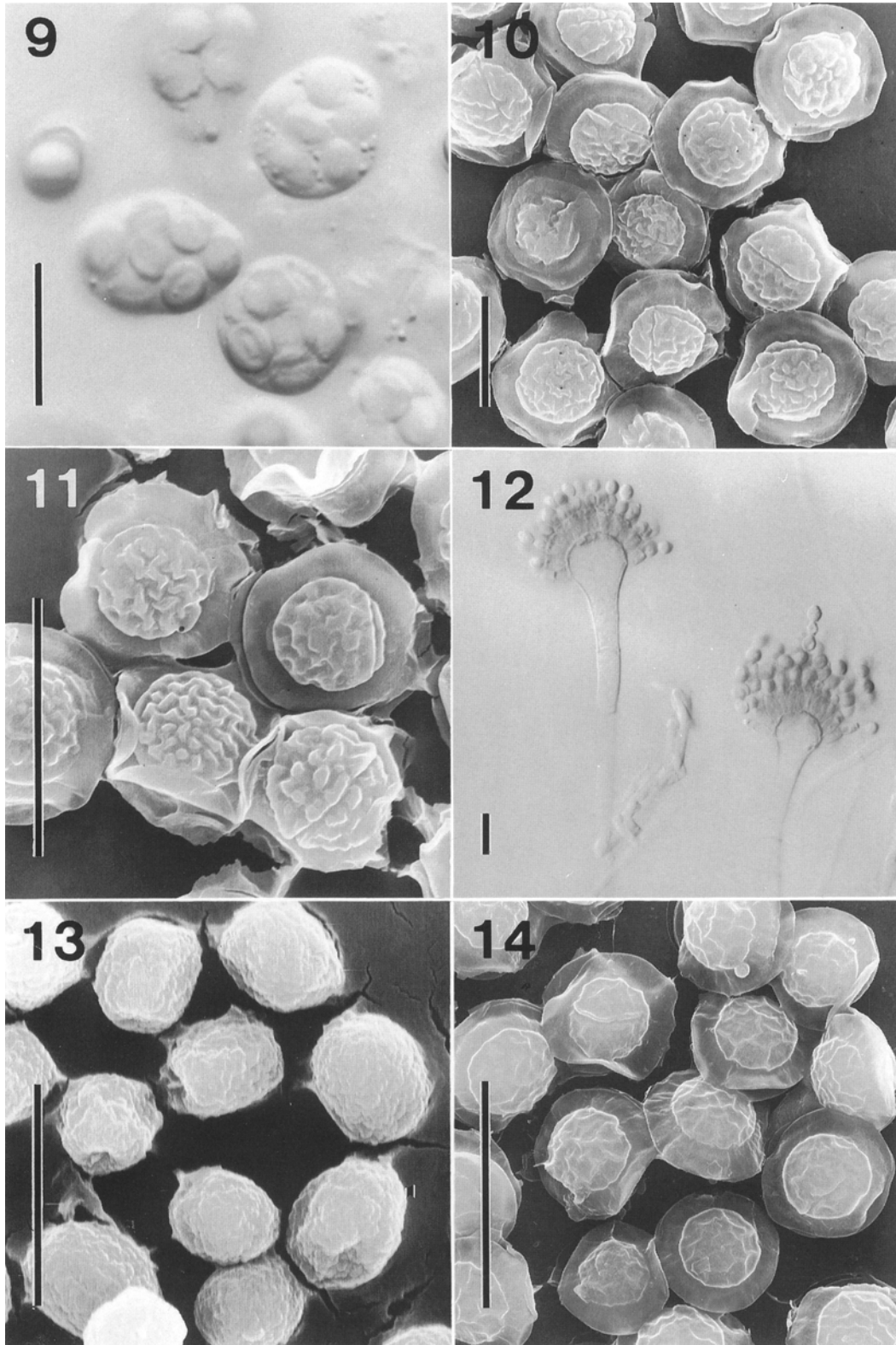
Colonies on MEA spreading broadly, attaining a diam of 80–84 mm in 14 d at 25°C, greyish green (30C3) to dull green (30D3), floccose, consisting of a thin mycelial felt; conidiogenesis sparse; reverse greyish yellow (4B3 to 4C3).

Heterothallic. Cleistothecia produced only when dissimilar mating types are paired, superficial, white to yellowish white, globose to subglobose, 150–270 µm in diam, surrounded by a loose covering of hyaline to pale yellowish brown aerial hyphae; peridium hyaline to pale yellowish brown, thin, membranaceous, consisting of angular cells, 3–15 µm in diam. Asci 8-spored, globose to subglobose or broadly ellipsoidal, 10–13 × 8–11 µm, evanescent at maturity. Ascospores hyaline to pale yellowish brown, broadly lenticular, spore body 4–5 × 3.5–4.5 µm, provided with two widely separated equatorial crests, tuberculate or lobate-reticulate convex surfaces (LM and SEM).

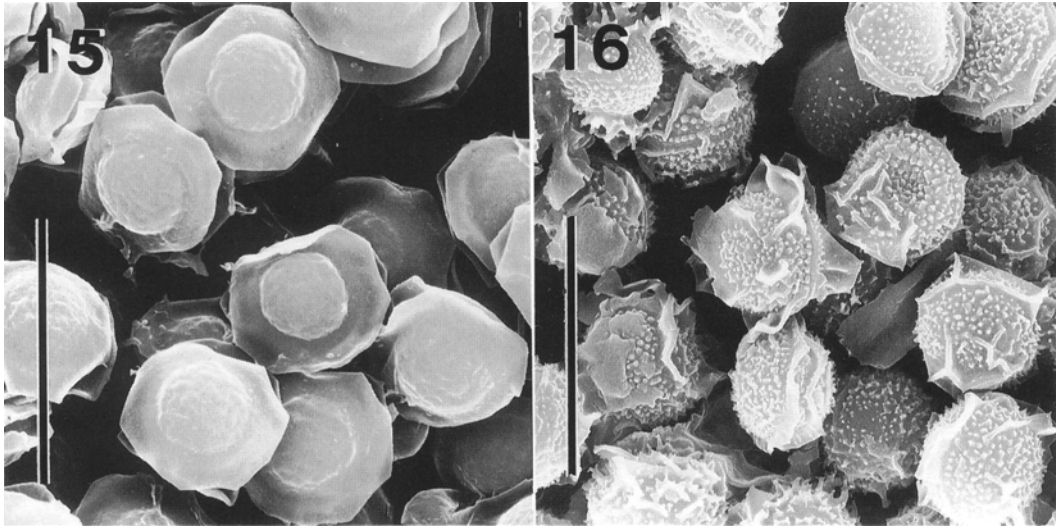
Conidial heads grayish green, short columnar to columnar, 55–100 × 30–40 µm. Conidiophores arising from the basal mycelium or aerial hyphae, hyaline to pale yellowish brown, smooth-walled, up to 410 µm long, 4–7 µm wide at the middle; vesicles pale yellowish brown to pale olive-brown, hemispherical to flask-shaped, 13–16 µm in diam. *Aspergilla uniseriate*; phialides pale grayish green, covering the upper half of the vesicle, flask-shaped, 4–7 × 1.5–2.5 µm. Conidia hyaline, grayish green to dull green in mass, globose to subglobose or broadly ellipsoidal, 2–3.5 × 2–3 µm, micro-tuberculata (LM and SEM).



Figs. 3–7. *Neosartorya nishimurae*; Fig. 8. *Neosartorya otanii*. 3. Paired culture. 4. Asci. 5. Ascospores (SEM). 6. Aspergilla. 7. Conidia (SEM). 8. Paired culture. Scales: 3, 8=10 mm; 4–6=10  $\mu$ m; 7=5  $\mu$ m.



Figs. 9–13. *Neosartorya otanii*; Fig. 14. *Neosartorya fennelliae*. 9. Asci. 10, 11. Ascospores (SEM). 12. *Aspergilla*. 13. Conidia (SEM). 14. Ascospores (SEM). Scales: 9, 10, 11, 12, 14=10 μm; 13=5 μm.



Figs. 15, 16. *Neosartorya* spp. 15. *Neosartorya spathulata* ascospores (SEM). 16. *Neosartorya udagawae* ascospores (SEM). Scales: 15, 16 = 10  $\mu$ m.

At 37°C, growth is more rapid than at 25°C.

Isolation: forest soil, Nairobi Kenya, 15 November 1974, coll. M. Takada.

Holotype: The type specimen, CBM-FA-914, is a dried culture from a pair of the isolates, CBM-FA-912 (A)  $\times$  CBM-FA-913 (a). Deposited in herb. CBM.

The structural pattern and general dimensions of the anamorph of this fungus are strikingly suggestive of *N. fennelliae*. Morphology of the ascospores in the two species is, however, clearly different. Those of *N. fennelliae* are ornamented with two closely appressed equatorial crests and delicately reticulate convex walls, appearing "cerebriform".

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