

A new variety of *Talaromyces wortmannii* and some observation on *Talaromyces assiutensis*

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Accepted for publication 20 December 1993

A new variety of *Talaromyces*, *T. wortmannii* var. *sublevisporus*, is described and illustrated. It is characterized by producing nearly smooth-walled ascospores. The other characters are almost identical with those of the type variety. Based on observations of several strains including the authentic ones, *T. assiutensis* is considered a finely rough-spored variety of *T. trachyspermus*.

Key Words—*Talaromyces*; *Penicillium*; Indonesia; Japan; Nepal.

During our continuing survey of soil-borne microfungi in Japan, Indonesia and Nepal, two species of *Talaromyces* C. R. Benjamin (Trichocomaceae, Eurotiales, Ascomycotina), *T. wortmannii* (Klöcker) C. R. Benjamin (anam. *Penicillium kloeckeri* Pitt) and *T. trachyspermus* (Shear) Stolk et Samson (anam. *P. lehmanii* Pitt), have frequently been isolated. Most of these are identical with the descriptions of both species in the monographs (Stolk and Samson, 1972; Pitt, 1979). However, some isolates have shown a discrepancy in the ascospore ornamentation; namely, almost smooth ascospores were constantly observed under an interference phase contrast light microscope. Scanning electron microscopical (SEM) examination of the ascospores more clearly showed that such unusual isolates can be readily recognized on the basis of ascospore morphology (Fig. 1).

Although there are no differences in the profiles of secondary metabolites between the representative strains of the taxa and the unusual isolates, the ascospore ornamentation is considered to be reliable as a key character for separation of taxa in the genus at least at the varietal level. Hence we are proposing a taxonomical revision of the two species.

***Talaromyces wortmannii* C. R. Benjamin, var. *sublevisporus* Yaguchi et Udagawa, var. nov. Figs. 1-B, 2**

A typo differt ascosporis fere levibus.

Holotypus PF 1130, colonia exsiccata in cultura ex solo, Sanada-machi, Chiisagata-gun, Nagano, in Japonia, 20. xi. 1990, a T. Yaguchi isolata et ea collectione fungorum, Musei et Instituti Historiae Naturalis Chiba (CBM) conservata.

Etymology: from Latin, *sub*=somewhat, *levisporus*=smooth-walled ascospores, referring to the character of ascospores.

Anamorph: *Penicillium kloeckeri* Pitt pro parte.

Colonies on Czapek agar growing rather restrictedly,

attaining a diam of 13–20 mm in 7 days at 25°C, velvety or somewhat floccose, consisting of a thin basal felt, Light Yellow (M. 3A5, after Kornerup and Wanscher, 1978) or Pure Yellow (Rayner, 1970), producing ascomata slowly; conidiogenesis absent or very inconspicuous; reverse Greyish Orange (M. 5B6) or Orange (Rayner). Colonies on Czapek yeast extract agar (CYA) growing rather restrictedly, attaining a diam of 16–22 mm in 7 days at 25°C, more or less floccose, plane, consisting of a thick basal felt, producing abundant ascomata, Reddish Yellow to Greyish Yellow (M. 4A6-3B6) or Amber to Pale Luteous (Rayner); conidiogenesis moderate, Dull Green (M. 29E3) or Greenish Grey (Rayner); exudate pale yellow; reverse Reddish Brown to Reddish Orange (M. 8E4-7B7) or Apricot to Dark Brick (Rayner). Colonies on malt extract agar (MEA) growing rather restrictedly, attaining a diam of 14–17 mm in 7 days at 25°C, plane, floccose, consisting of a thin basal felt, producing abundant ascomata, Yellow (M. 3A6) or Pure Yellow (Rayner); conidiogenesis sparse, inconspicuous; exudate orange; reverse Greyish Orange (M. 6B6) or Orange to Fulvous (Rayner). Colonies on oatmeal agar growing rather rapidly, attaining a diam of 25 mm in 7 days at 25°C, plane, granular in appearance due to the production of abundant ascomata, Reddish Yellow (M. 4A6) or Pure Yellow to Amber (Rayner); conidiogenesis inconspicuous; exudate small, yellow to orange; reverse Brownish Orange (M. 7C3) or Vinaceous Buff (Rayner). Colonies on cornmeal agar growing restrictedly, attaining a diam of 13–20 mm in 7 days at 25°C, thin, plane, Yellow (M. 3A6) or Pure Yellow (Rayner), producing abundant ascomata on the substratum; conidiogenesis absent or sparse; reverse uncolored.

Ascomata discrete or confluent, non-ostiolate, yellow to orange, globose to subglobose, 80–300 µm in diam, maturing within 14 days; ascomatal wall consisting of a loose network of interwoven hyphae, surrounded

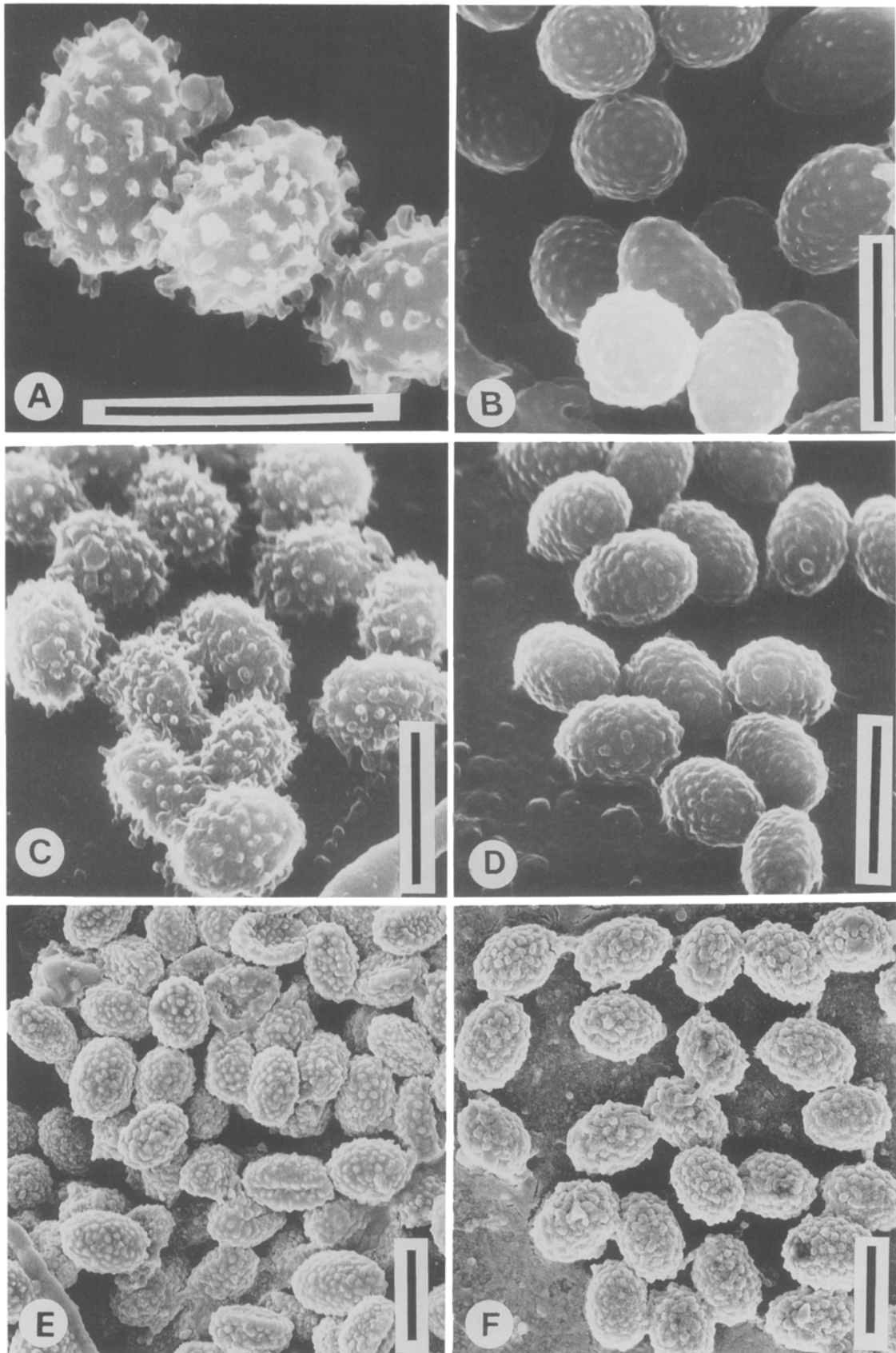


Fig. 1.

by radiating hyphae. Ascumatal initials like the variety *wortmannii*. Asci 8-spored, subglobose to ellipsoidal, $7.5\text{--}11 \times 7\text{--}9 \mu\text{m}$, borne in short chains, evanescent. Ascospores hyaline to pale yellow, ellipsoidal, $3.5\text{--}5 (-5.5) \times 2.5\text{--}3.5 \mu\text{m}$, with walls almost smooth to very finely roughened (under SEM).

Conidiophores arising from the basal mycelium; stipes $250\text{--}350 \times 2.5\text{--}3.5 \mu\text{m}$, smooth-walled, septate, hyaline. Penicilli typically biverticillate. Metulae in verticils of 4–8, divergent at angles of up to 60° , $(9.5\text{--}) 13.5\text{--}16 \times 3\text{--}4 \mu\text{m}$. Phialides 4–6 per metula, acerose, $9.5\text{--}12 \times 2.5\text{--}3 \mu\text{m}$, with long collula. Conidia hyaline, subglobose to ellipsoidal, $2.5\text{--}4 \times 1.5\text{--}3.5 \mu\text{m}$, smooth-walled, in long chains.

Major ubiquinone: Q-10(H₂).

At 37°C , growth is nil.

Specimens examined: PF 1130 (holotype), in dried culture isolated from soil, Sanada-machi, Chiisagata-gun, Nagano-ken, Japan, November 20, 1990, coll. T. Yaguchi; NE 137-2, in culture isolated from soil in paddy field, Pikhel, Kathmandu, Nepal, September 22, 1986, coll. S. Udagawa; NE 125, in culture isolated from soil in paddy field, Surjebinayak, Kathmandu, Nepal, September 21, 1986, coll. S. Udagawa; NE 119, in culture isolated from soil in paddy field, Dokathali, Kathmandu, Nepal, September 19, 1986, coll. S. Udagawa; and NE 97, in culture isolated from soil in paddy field, Baudha, Kathmandu, Nepal, August 26, 1986, coll. S. Udagawa. The holotype has been deposited with the Natural History Museum and Institute, Chiba, Japan.

Talaromyces wortmannii var. *sublevisporus* is readily distinguished from the type variety by difference in ascospore ornamentation, but there are the following minor discrepancies: somewhat larger ascumata ($80\text{--}300 \mu\text{m}$ vs. $100\text{--}200 \mu\text{m}$ fide Pitt (1979)), larger conidiophore stipes ($250\text{--}350 \times 2.5\text{--}3.5 \mu\text{m}$ vs. $50\text{--}200 \times 2.2\text{--}2.5 \mu\text{m}$ fide Pitt (1979) or up to $150 \mu\text{m}$ in length fide Stolk and Samson (1972)), and larger metulae ($(9.5\text{--}) 13.5\text{--}16 \times 3\text{--}4 \mu\text{m}$ vs. $8\text{--}12 \times 2\text{--}2.5 \mu\text{m}$ fide Pitt (1979) and Stolk and Samson (1972)).

***Talaromyces trachyspermus* (Shear) Stolk et Samson, var. *assiutensis* (Samson et Abdel-Fattah) Yaguchi et Udagawa, comb. nov. Figs. 1-D~F, 3**

Basionym: *Talaromyces assiutensis* Samson et Abdel-Fattah, *Persoonia*, **9**: 501. 1978.

= *Talaromyces gossypii* Pitt, The genus *Penicillium* and its teleomorphic states *Eupenicillium* and *Talaromyces*, p. 500. 1979.

Anamorph: *Penicillium lehmanii* Pitt pro parte.

= *Penicillium assiutense* Samson et Abdel-Fattah, *Persoonia* **9**: 501. 1978.

= *Penicillium gossypii* Pitt, The genus *Penicillium* and its teleomorphic states *Eupenicillium* and *Talaromyces*, p. 500. 1979.

Specimens examined: CBS 147.78, ex type of *Talaromyces assiutensis*, in culture isolated by dilution plating from soil which had been amended with crushed buffalo hooves and incubated for 5 months at 35°C by H. M. Abdel-Fattah, Assiut, Egypt, 1977; IMI 198365, ex type of *Talaromyces gossypii*, isolated from *Gossypium* sp., India, by R. B. Somani, 1975; PF 1124, in culture isolated from soil, Hahajima Island, Ogasawara-mura, Tokyo-to, Japan, December 12, 1991, coll. T. Yaguchi; and PF 1123, in culture isolated from soil, Jawa Timur, Indonesia, August 8, 1991, coll. T. Yaguchi.

Our description based on strain PF 1124 follows:

Colonies on CYA growing rather restrictedly, attaining a diam of 20 mm in 7 days at 25°C , velvety to floccose, plane, consisting of a thin basal felt, producing limited ascumata on the felt, loosely covered by somewhat funiculose hyphae, White to Yellowish White (M. 4A2) or Rosy Buff (Rayner); conidiogenesis sparse, inconspicuous, Greyish Green (M. 25D5) or Glaucous Grey (Rayner); odor strongly musty; reverse Yellowish White to Light Brown (M. 4A2-6D4) or Rosy Buff (Rayner). Colonies on MEA growing rather rapidly, attaining a diam of 20–25 mm in 7 days at 25°C , floccose to funiculose, plane, consisting of a thin basal felt, producing limited ascumata on the felt, similar to CYA in appearance; conidiogenesis absent or sparse, inconspicuous; reverse uncolored to faintly Light Brown (M. 6D4) or Fawn (Rayner). Colonies on oatmeal agar growing rather restrictedly, attaining a diam of 20 mm in 7 days at 25°C , more or less funiculose, lightly wrinkled, consisting of a thin basal felt, white; ascumata slowly but abundantly produced on the felt; conidiogenesis absent or sparse, inconspicuous, Greyish Green (M. 27B3-26C3) or Greenish Glaucous to Pistachio Green (Rayner); reverse uncolored to Yellowish White (M. 4A2) or Buff (Rayner).

Ascumata cream to pale yellow, globose to subglobose, $80\text{--}320 \mu\text{m}$, maturing within 14–21 days; ascumatal wall consisting of a loose network of interwoven hyphae, surrounded by radiating hyphae. Ascumatal initials like the variety *trachyspermus*. Asci 8-spored, subglobose to ovoid, $6.5\text{--}8 \times 6\text{--}7 \mu\text{m}$, borne in short chains, evanescent. Ascospores hyaline, ellipsoidal, $3\text{--}4 \times 2\text{--}2.5 \mu\text{m}$, almost smooth to very finely roughened (under SEM).

Conidiophores arising from aerial hyphae; stipes

Fig. 1. Scanning electron micrographs, representing ascospores of *Talaromyces wortmannii*, *T. trachyspermus*, *T. assiutensis* and *T. gossypii*.

A. *Talaromyces wortmannii* var. *wortmannii*, PF 1141.

B. *T. wortmannii* var. *sublevisporus*, PF 1130.

C. *T. trachyspermus* var. *trachyspermus*, PF 1142.

D. *T. trachyspermus* var. *assiutensis*, PF 1124.

E. *T. trachyspermus* var. *assiutensis* (*T. assiutensis*), CBS 147.78.

F. *T. trachyspermus* var. *assiutensis* (*T. gossypii*), IMI 198365. All scale bars = $5 \mu\text{m}$.

short, $5-20 \times 2-2.5 \mu\text{m}$, smooth-walled, septate, hyaline. Penicilli mostly biverticillate or monoverticillate. Metulae appressed or sometimes divergent, in verticils of 2-3, $9-15 \times 2-2.5 \mu\text{m}$. Phialides 2-6 per metula, acerose, $10-15 \times 1.5-2 \mu\text{m}$. Conidia hyaline, ellipsoidal, $2.5-3 \times 1.5-2.5 \mu\text{m}$, smooth-walled, in disordered chains.

Major ubiquinones: Q-(10(H₂) (49%)+10(H₄) (51%)).

At 37°C, colony diam on CYA in 7 days 35 mm; ascospores produced.

Talaromyces assiutensis, described in 1978 by Samson and Abdel-Fattah as a new species in the section *Talaromyces* of the genus, was based upon one strain:

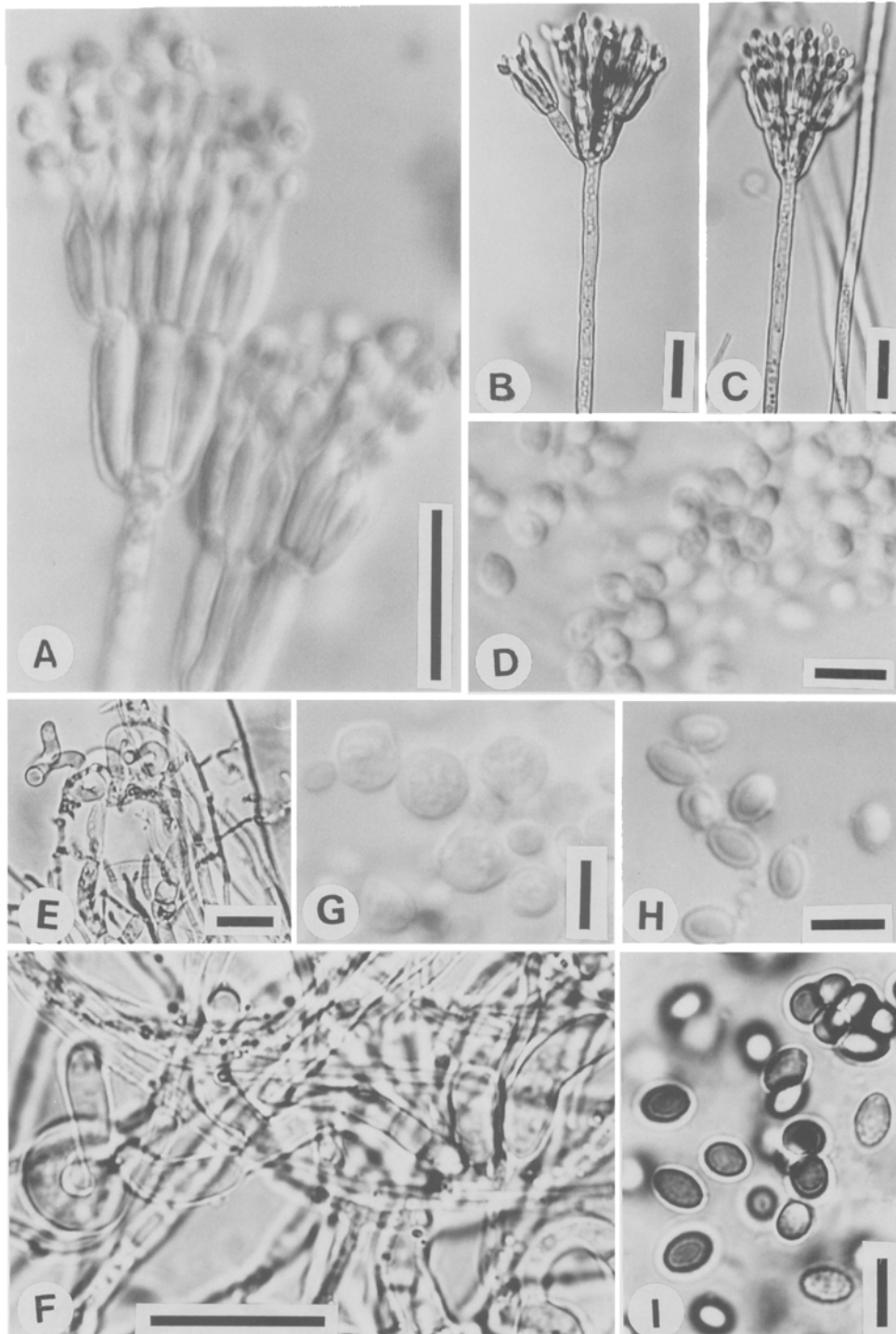


Fig. 2. *Talaromyces wortmannii* var. *sublevisporus*, PF 1130.

A-C. Penicilli. D. Conidia. E, F. Ascomatal initials. G. Asci. H, I. Ascospores. Scale bars; A-C, E, F = 10 μm . D, G-I = 5 μm .

CBS 147.78, isolated by Abdel-Fattah from soil in Egypt (Samson and Abdel-Fattah, 1978). The species is characterized by the white ascomata and the small, smooth-walled to finely roughened ascospores. The similar fungus, *T. gossypii*, was independently described by Pitt (1979). Since there are no significant differences in morphology between *T. assiutensis* and *T. gossypii*,

they were placed in synonymy. A chemotaxonomic comparison of the authentic cultures of both species proved their identity (Frisvad et al., 1990), although Kuraishi et al. (1991) distinguished *T. gossypii* from *T. assiutensis* by ubiquinone analysis, viz. a mixture of Q-10(H₂) and Q-10(H₄) for *T. assiutensis* vs. Q-10(H₄) as a major component for *T. gossypii*. The discrepancy in the ubi-

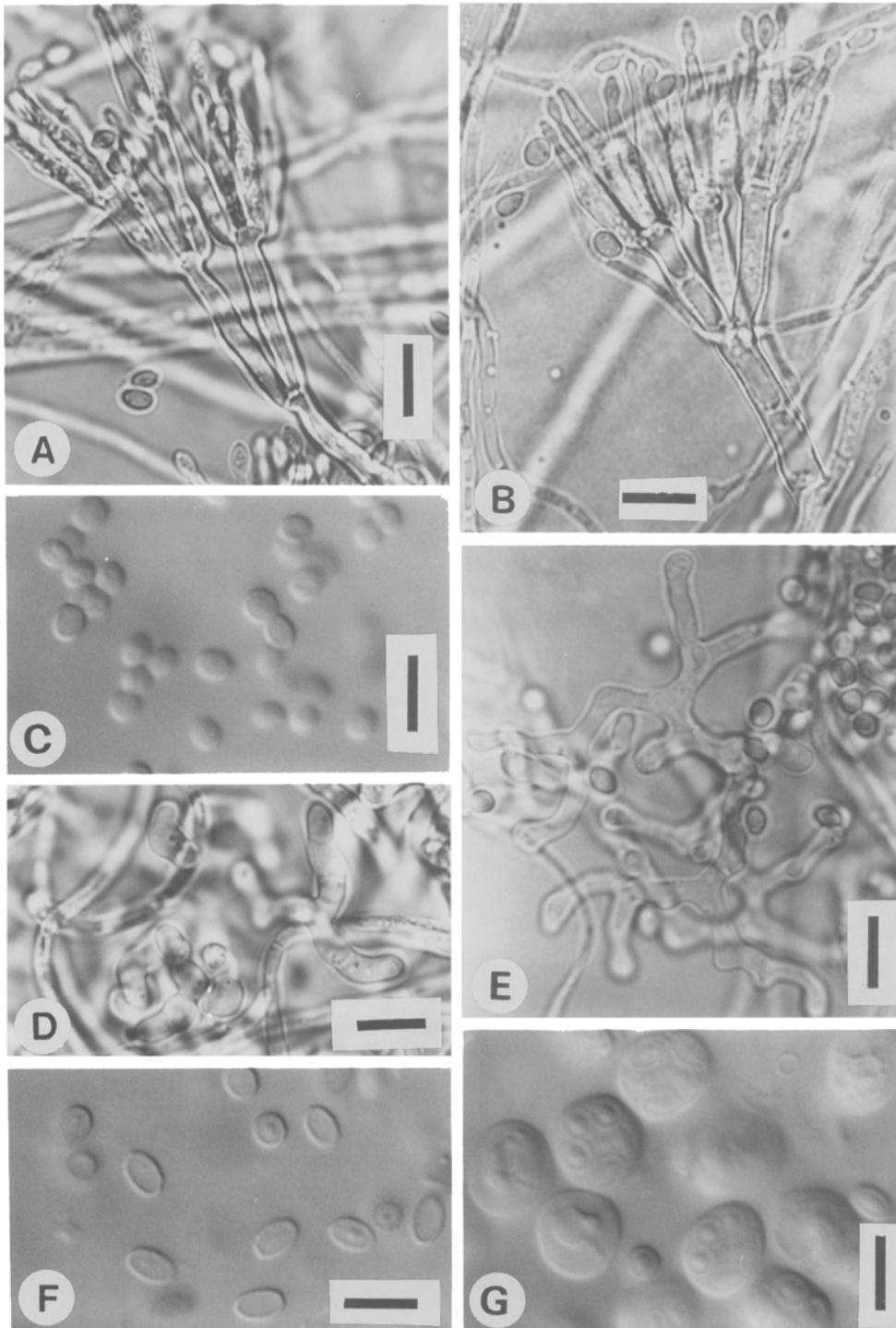


Fig. 3. *Talaromyces trachyspermus* var. *assiutensis*, PF 1124. A, B. Penicilli. C. Conidia. D, E. Ascomatal initials. F. Ascospores. G. Asci. All scale bars = 5 µm.

Table 1. Comparison of some diagnostic characters between *T. trachyspermus* and *T. assiutensis* (including *T. gossypii* and additional isolate).

Character	<i>T. trachyspermus</i>	<i>T. assiutensis</i>	<i>T. gossypii</i>	PF 1124
Growth on CYA (7d, 25°C)	15–20 mm	11–15 mm	15–20 mm	20 mm
Growth on MEA (7d, 25°C)	20–25 mm	21–25 mm	25–30 mm	20–25 mm
Colony color (CYA)	white	white to pale yellow	white to pale yellow or pale brown	white to yellowish white
Ascomata (μm)	yellowish white 50–350 or 300–500(–700)	white to pale yellow 250–500	white or cream 400–800	cream to pale yellow 80–320
Asci (μm)	6–8 × 6–7	7–10 × 5.5–7	7–8	6.5–8 × 6–7
Ascospores (μm)	ellipsoidal 3.5–4.5 × 2.2–2.5 spinulose	ellipsoidal 3–3.5 × 2–2.5 smooth to finely roughened	ellipsoidal 3–3.5 × 2.2–2.5 smooth to finely roughened	ellipsoidal 3–4 × 2–2.5 almost smooth to finely roughened
Ascoma initials	gnarled	gnarled	gnarled	gnarled
Conidiophores (μm)	5–20 × 1.8–2.2, smooth	25–40 × 2.5–3, smooth	20–40(–80) × (1.5–) 1.8–2.2, smooth	5–20 × 2–2.5, smooth
Penicilli	biverticillate	biverticillate	biverticillate	biverticillate
Conidia (μm)	ellipsoidal or fusiform 2.5–4.5 × 2–2.5, smooth	ellipsoidal 2.2–4 × 1.5–2.2 smooth	subspheroidal to ellipsoidal 2–3 × 1.8–2.5, smooth	ellipsoidal 2.5–3 × 1.5–2.5 smooth
Growth at 37°C	rapid	rapid	rapid	rapid
Ubiquinones	Q-10(H ₂) + 10(H ₄)	Q-10(H ₂) + 10(H ₄)	Q-10(H ₂) + 10(H ₄)	Q-10(H ₂) + 10(H ₄)
Secondary metabolites	I: spiculisporic acid II: glauconic acid	II: glauconic acid	II: glauconic acid	I: spiculisporic acid

quinone profiles is, however doubtful, because we found on careful examination of the authentic strain of *T. gossypii* (IMI 198365) that its ubiquinone is a mixture of Q-10(H₂) (30%) and Q-10(H₄) (70%). Further comparison of these two strains of *T. assiutensis* and our unusual isolates of *T. trachyspermus* also shows that the teleomorphs as well as the anamorphs are congeneric (Table 1). Besides the characters of ascospore ornamentation, the morphological and chemotaxonomic profiles demonstrated no significant differences between the strains of *T. assiutensis* including our isolates and the representative ones of *T. trachyspermus* (Table 1). Thus, like *T. wortmannii* var. *sublevisporus*, we propose that *T. assiutensis* should be considered as a variety of *T. trachyspermus*.

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