Mycoscience 41: 411–414, 2000

Note

Materials for the fungus flora of Japan (56)* *Mariannaea camptospora* and *M. elegans* var. *punicea* from Japan

Toru Okuda and **Kozo Yamamoto**

Tanabe Seiyaku Co., Ltd., Discovery Research Laboratory, 2–2–50 Kawagishi, Toda, Saitama 335–8505, Japan

Accepted for publication 18 June 2000

One additional species and a variety of *Mariannaea, M. camptospora* and *M. elegans* var. *punicea*, were recorded for the first time in Japan. *Mariannaea camptospora* formed two types of conidiophores. One type was characterized by simple verticillate phialides sometimes with punctuate walls at the base, producing long oblique conidial chains, and symmetrical spindle-shaped conidia. The other type was characterized by more crowded and shorter phialides with small conidial droplets and hemispherical to concave smaller conidia. *Mariannaea elegans* var. *punicea* was characterized by distinct red purple pigmentation in agar media.

Key Words——*Mariannaea camptospora*; *Mariannaea elegans* var. *punicea*.

Mariannaea camptospora Samson, Stud. Mycol. **6**: 78, 1974

Cultural properties—On oatmeal agar (OA), 2% malt extract agar (2%MA), or Miura medium (LCA), colonies grew rapidly, attaining a diameter of 21–28 mm in 7 d at 25°C, showing flat velutinous appearance. When sporulated, the surface of the colonies was white to cream or ivory yellow (Munsell 5Y9/2) (anon. 1977). Reverse side of the colonies was dull yellow (Munsell 5Y8/4) to pastel yellow (Munsell 5Y9/4).

Morphological characteristics—Two types of conidiophores were observed: *Verticillium* Nees-type reminiscent of *Verticillium* (Figs. 1A, C, E) and *Trichoderma* Pers.-type (Figs. 1B, D, F). *Verticillium*-type conidiophores were mainly borne from the agar surface. Stipes were 160–400 μm long. The basal cells were thick, 4–9 μm in width, with rough walls. Mono- to terverticillate branches were characteristically produced along the uppermost parts of the *Verticillium*-type conidiophores. Three to 8 phialides were borne on the terminal verticils, while 2–4 phialides were borne verticillately along slightly lower parts of stipes. Terminal verticils often possessed 1–3 metulae, and sometimes a short branch occurred. The verticils were rather convergent. Phialides were long tapered acerose to long flask-shaped, and often punctuated under SEM (Fig. 1E). They measured 15.5–28.5 × 3.0–4.5 μm, L/W 3.8–8.3 (average 19.6 × 3.6 μm, L/W 5.6) on OA. Terminal phialides tended to be the longest. Conidia were fusiform and symmetrical (Fig. 1G), forming oblique columns, sometimes slipping down (Figs. 1A, E). They measured 5.5–7.0 × 2.5–3.5 μm with L/W 1.9–2.6 (average 6.4 × 2.9 μm, L/W 2.2) on OA. On the other hand, *Trichoderma*-type conidiophores were borne from chromophilic hyphal strands or as short branches from the lower parts of *Verticillium*-type conidiophores. The stipes were 20–100 μm long with basal cells 3–4 μm wide. Phialides were formed solitary or as 2–5 verticils from irregular branches of conidiophores, smooth-walled under SEM, flexuous to sinuous, 9.0–13.5 × 2.5–3.5 μm, L/W 2.7–5.2 (average 11.7 × 3.0 μm, L/W 3.9) on OA (Figs. 1D, F). The verticils were mostly divergent and near perpendicular to their bearers. Conidia were concave or short lunate, and asymmetrical (Fig. 1H), forming irregular clusters or masses (Figs. 1B, F). They measured 2.5–5.5 × 2.0–3.5 μm, L/W 1.1–2.2 (average 4.0 × 2.7 μm, L/W 1.5) on OA.

Strain examined: TC 1287 from soil, near Kanpire Fall, Iriomote Island, Taketomi, Okinawa, Japan, 3 July 1997; isolated by a washing/filtration method slightly modified from the method of Bills and Polishook (1994).

Mariannaea camptospora has so far been isolated from soil, humus layer with low pH values, dead fallen trunk of *Quercus*, and *Podocarpus* sp. (Samson, 1974; anon., 1999). The locations where the substrates were collected were restricted to the Netherlands and Chile. This does not necessarily mean that the species is rare: it may be readily neglected. We compared our strain with *M. camptospora* CBS 209.73, the culture derived from type, which revealed the identity. *Mariannaea camptospora* formed two distinct types of conidiophores:

* (55): Udagawa, S. and Uchiyama, S., Mycoscience **41**: 263–267, 2000.

** Present address: Tamagawa University Research Institute, 6–1–1 Tamagawa-Gakuen, Machida, Tokyo 194–8610, Japan

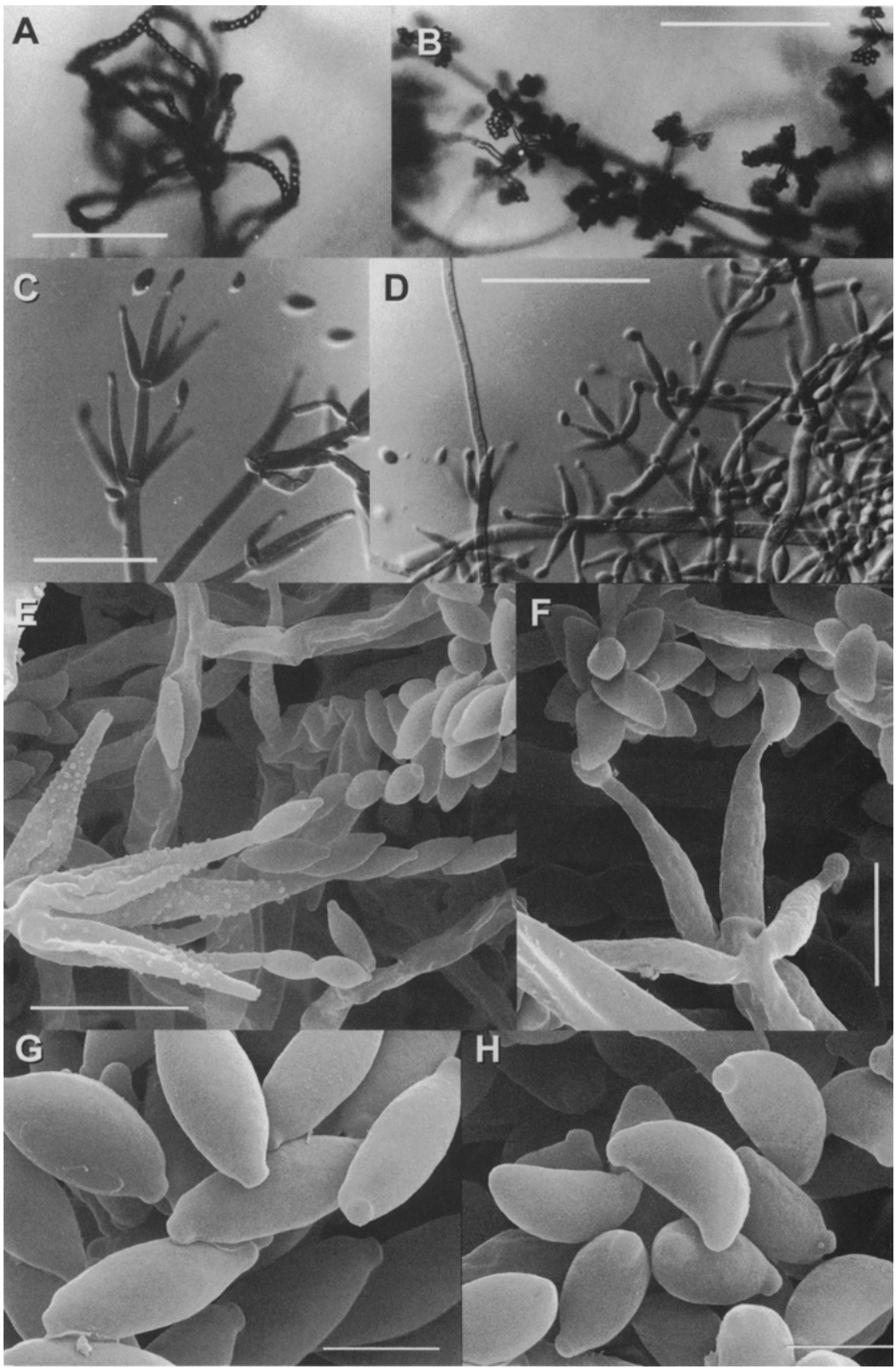

Fig. 1. *Mariannnaea camtospora* TC 1287. A. Long oblique conidial chains from typical *Verticillium*-type conidiophores on 2%MA, bar=100 μm. B. Conidial mass and short compact phialides on LCA, bar=100 μm. C. Phialides from *Verticillium*-type conidiophores on LCA, bar=50 μm. D. Phialides from complicated branching, bar=50 μm. E. Punctuated phialides of *Verticillium*-type conidiophore on OA, SEM, bar=10 μm. F. Short phialides with smooth walls on OA, SEM, bar=6 μm. G. Symmetrical spindle-shaped conidia from long phialides on OA, SEM, bar=3 μm. H. Lunate conidia from short phialides on OA, SEM, bar=3 μm.

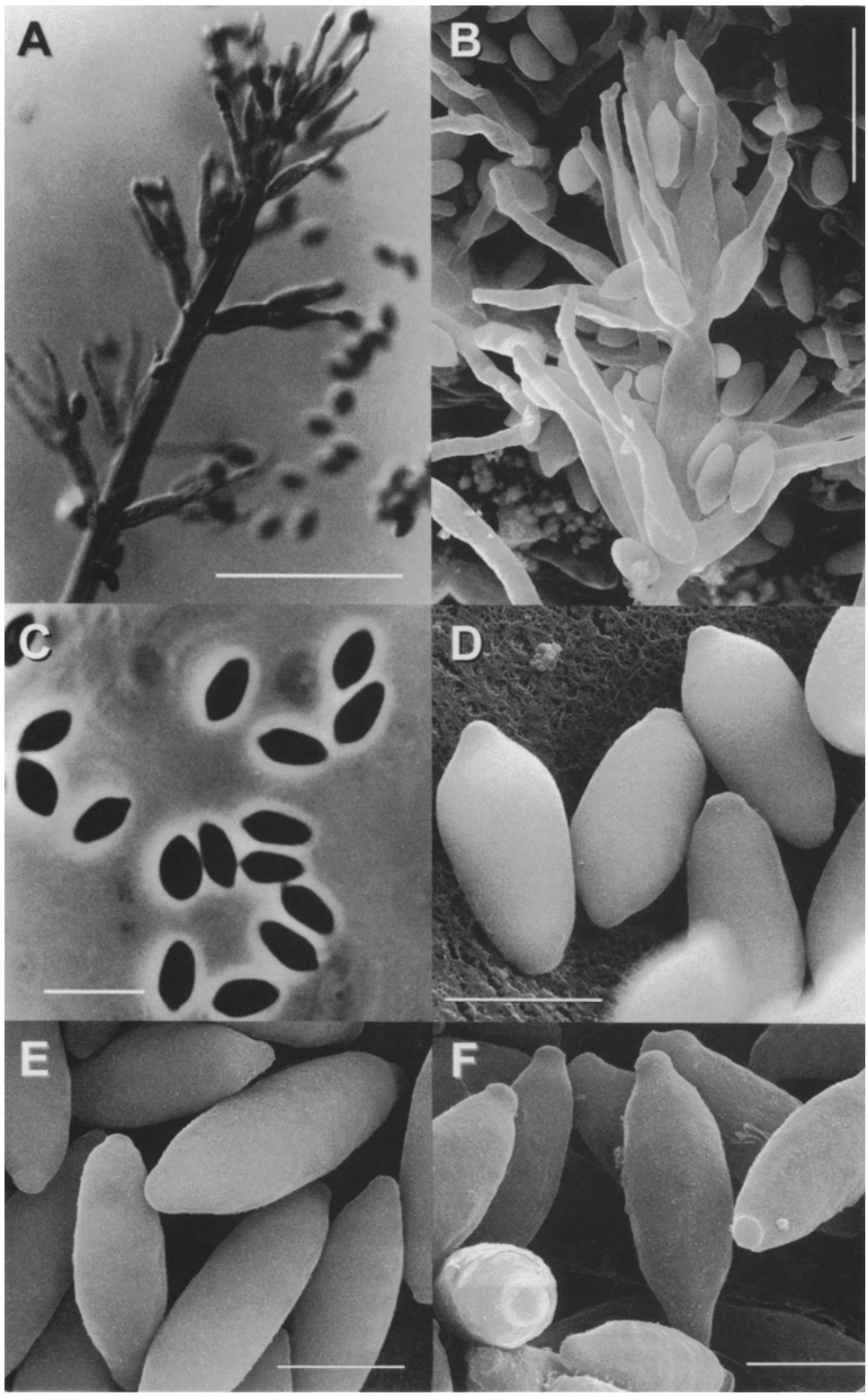

Fig. 2. *Mariannaea elegans* var. *punicea* TC 1322 and *Mariannaea elegans* var. *elegans* TC 1323 & TC 1318. A. conidiophore on LCA, bar = 50 μm. B. SEM of conidiophore on OA, bar = 10 μm. C. Conidia on OA, bar = 10 μm. D. SEM of conidia on OA, bar = 3 μm. E. SEM of conidia of *Mariannaea elegans* var. *elegans* TC 1323 on OA for comparison, bar = 3 μm. F. SEM of conidia of *Mariannaea elegans* var. *elegans* TC 1318 on OA for comparison, bar = 3 μm.

Verticillium-type and typical conidiophores of *Mariannaea* G. Arnaud ex Samson. Samson (1974) did not clearly state this, although his drawings show both types. The *Verticillium*-type conidiophores formed straight phialides with long imbricate chains of fusiform conidia, whereas the other type was like the complicated branching system of *Trichoderma harzianum* Rifai (hence we call it *Trichoderma*-type here) and consisted of more crowded branches and phialides forming an irregular mass of Chinese-dumpling-shaped conidia. Two types of conidia or conidiophores are observed in the genus *Clonostachys* Corda (Schroers et al., 1999; Okuda et al., 2000). The *Verticillium*-type and *Trichoderma*-type in *M. camptospora* may correspond to the *Verticillium*-type (primary conidiophores) and penicillate conidiophores (secondary conidiophores) in *Clonostachys rosea* (Link: Fr.) Schroers, Samuels, Seifert & W. Gams (Domsch et al., 1980; Schroers et al., 1999), respectively. Schroers et al. (1999) mentioned that asymmetrical conidia form imbricate conidial columns as in *Clonostachys*, while symmetrical conidia form drops of conidial mass as in *Gliocladium* Corda sensu stricto. In *M. camptospora*, however, due to the strongly asymmetrical shape, the conidia from *Trichoderma*-type conidiophores easily slipped down, becoming very short chains or small irregular clusters. On the other hand, spindle-shaped conidia formed long imbricate columns as in *Clonostachys rosea*. The conidia appeared from the apex of phialides obliquely, and the spindle-shaped conidia had a shoulder-like protuberance near the truncate base. These events were responsible for the imbricate conidial chains.

Mariannaea elegans (Corda) Samson var. ***punicea*** Samson, Stud. Mycol. **6**: 78, 1974.

Cultural properties—On 2%MA, colonies grew rapidly, attaining a diameter of 27–28 mm in 7 d at 25°C. Since sporulation was poor, the substrate mycelium is responsible for the color of the surface, deep yellow to mustard (Munsell 5Y7/12). The reverse was therefore deep yellow to mustard (Munsell 5Y7/12). On OA or LcA, colonies grew rapidly, attaining a diameter of 30–34 mm in 7 d at 25°C. Conidial area was white to cream (Munsell 5Y9/2) with zonate powdery appearance. The reverse side of the colonies was striking, geranium red to rose red (Munsell 10RP5/12).

Morphological characteristics—Conidiophores mainly arose from the agar surface but also from chromophilic hyphal strands. The stipes were 400–800 × 6–12 μm, straight, stiff, chromophilic near the base, rough and granulated, gradually tapering toward the apex, bearing regular verticillate branching. Phialides were borne in a verticil of 2 to 6 directly attached to the stipe or from short verticillate branches (Figs. 2A, B). They were slender flask-shaped with a long neck or simply acerose, and straight, 12.0–20.5 × 3.5–4.0 μm, L/W 3.1–5.5 (average 15.6 × 3.7 μm, L/W 4.2) on OA. Conidia were ellipsoidal to fusiform, widest at the upper one-fourth and abruptly tapered to the papillate apex, forming long divergent oblique chains (Figs. 2C, D). They measured 4.0–6.0 × 1.5–3.5 μm, L/W 1.2–2.7 (average 5.1 × 2.7 μm, L/W 1.9) on OA.

Strain examined: TC 1344 from soil, near Hisaka Shrine, Shuzenji, Tagata, Shizuoka, Japan; October 21, 1996, collected and isolated by a washing/filtration method.

Mariannaea elegans var. *punicea* is characterized by its distinct red-purple coloration in agar and the reverse (Samson, 1974). However, the pigment production seems to be lost easily because the living culture derived from type, CBS 239.56 did not show such purple color. Except for the striking color, in fact, other characteristics were similar to those of *M. elegans* (Corda) Arnaud ex Samson var. *elegans*. But, the shape of conidia was slightly different. We compared it with *Mariannaea elegans* var. *elegans* TC 1318 and TC 1323, which were isolated from tree sap collected in Hanno, Saitama, Japan, 22 January 1996, and from soil collected in Shuzenji, Tagata, Shizuoka, Japan, 21 October 1996, respectively. In *M. elegans* var. *punicea*, the conidia were fusiform to long obovoid and widest in the one-fourth part from the apex, while *M. elegans* var. *elegans* formed mostly slender fusiform conidia (Figs. 3E, F). The conidia of variety *punicea* (4.5–6.0 μm long) were slightly shorter than those of the latter species (5.0–7.5 μm long), and hence showed a smaller L/W ratio (1.6–2.2 vs. 1.7–2.6). The phialides were also slightly shorter in variety *punicea*, and hence showed a smaller L/W ratio. Two types of conidiophores were sometimes observed in both varieties. However, the distinction was not so clear as in *M. camptospora*.

Acknowledgement—We thank Noboru Kishi for his technical assistance.

Literature cited

Anonymous 1977. Munsell Color System. Japan Color Enterprise Co., 15 pp Tokyo.

Anonymous 1999. Filamentous fungi database on Search the CBS databases in the CBS website. http://www.cbs.knaw.nl/

Bills, G. F. and Polishook, J. D. 1994. Abundance and diversity of microfungi in leaf litter of a lowland rain forest in Costa Rica. Mycologia **86**: 187–198.

Domsch, K. H., Gams, W. and Anderson, T.-H. 1980. Compendium of soil fungi, vol. 1, pp. 368–377. Academic Press, London.

Miura, K. and Kudo, Y. Y. 1970. An agar-medium for aquatic Hyphomycetes. Trans. Mycol. Soc. Japan **11**: 116–118. (In Japanese.)

Okuda, T., Kohno, J., Kishi, N., Asai, Y., Nishio, M. and Komatsubara, S. 2000. Production of TMC-151, TMC-154 and TMC-171, a new class of antibiotics is specific to '*Gliocladium roseum*' group. Mycoscience **41**: 239–253.

Samson, R. A. 1974. *Paecilomyces* and some allied hyphomycetes. Stud. Mycol. **6**: 1–119.

Schroers, H.-J., Samuels, G. J., Seifert, K. A. and Gams, W. 1999. Classification of the mycoparasite *Gliocladium roseum* in *Clonostachys* as *C. rosea*, its relationship to *Bionectria ochroleuca*, and notes on other *Gliocladium*-like fungi. Mycologia **91**: 365–385.