Neotropical Ascomycetes 12. *Mirannulata samuelsii* gen. et sp. nov. and *M. costaricensis* sp. nov., new taxa from the Caribbean and elsewhere

Sabine M. Huhndorf[1]*, Fernando A. Fernández[1], Andrew N. Miller[1,2] & D. Jean Lodge[3]

[1] Field Museum of Natural History, Botany Department, Chicago, Illinois 60605–2496, USA
[2] University of Illinois at Chicago, Department of Biological Sciences, Chicago, Illinois 60607–7060, USA
[3] Center for Forest Mycology Research, USDA-Forest Service, Forest Products Laboratory, P.O. Box 1377, Luquillo, PR 00773-1377, USA

Huhndorf, S. M., F. A. Fernández, A. N. Miller & D. J. Lodge (2003). Neotropical Ascomycetes 12. *Mirannulata samuelsii* gen. et sp. nov. and *M. costaricensis* sp. nov., new taxa from the Caribbean and elsewhere. – Sydowia 55 (2): 172–180.

A terrestrial wood-inhabiting pyrenomycete was collected numerous times in the Neotropics. It possesses superficial, clustered ascomata, large, distinctive ascus rings and strongly guttulate, fusiform ascospores. A second similar pyrenomycete was collected once in Costa Rica. They could not be placed into any known genus, so a new genus, *Mirannulata*, is described. *Mirannulata samuelsii* has hyaline, fusiform, three-septate ascospores while *M. costaricensis* has brown, fusiform, one-septate ascospores. *Mirannulata* has a very large, chitinoid, double-layered ascus apical ring similar to that of *Annulatascus* and *Jobellisia*.

Keywords: *Annulatascus*, Annulatascaceae, *Jobellisia*, Neotropics.

A terrestrial wood-inhabiting pyrenomycete was collected numerous times in the Neotropics. It possesses superficial, clustered ascomata, large, distinctive ascus rings and strongly guttulate, fusiform ascospores. A second, similar pyrenomycete was collected once in Costa Rica. They could not be placed into any known genus so a new genus is described.

Material and methods

Ascomata were mounted first in water, which was then replaced with lactophenol containing azure A. All measurements were made in water. Ascomata were sectioned at 5 µm for light microscopy using the techniques of Huhndorf (1991) and structures were examined using bright field (BF), phase contrast (PH) and differential inter-

* e-mail address: shuhndorf@fieldmuseum.org

ference microscopy (DIC). A minimum of 30 asci, paraphyses and ascospores were measured in water. Images were captured and photographic plates were produced following the methods of Huhndorf & Fernández (1998). Abbreviations for collectors are SMH = S. M. Huhndorf, DJL = D. J. Lodge, FF = F. Fernández, ANM = A. N. Miller and GJS = G. J. Samuels. When no collector is listed, the collector's initials are given with the specimen number. All SMH collections are deposited at F. Latitude and longitude are given in degrees or calculated decimal equivalents. All specimens were collected from decorticated wood unless otherwise noted and dimensions given for the substrates are diameters.

Results

Mirannulata Huhndorf, F. A. Fernández, A. N. Mill. & Lodge, **gen. nov.**

Ascomata obpyriformia vel ovoidea, superficialia, papillata. Paraphyses angustae. Asci cylindracei, brevi-stipitati, annulo apicali bipartito. Ascosporae fusiformes, septatae, hyalinae vel brunneae, guttulatae.

Ascomata obpyriform to ovoid; superficial; papillate. – Paraphyses narrow. – Asci cylindrical, short-stipitate, with large, bipartite ring. – Ascospores fusiform, septate, hyaline or brown, guttulate.

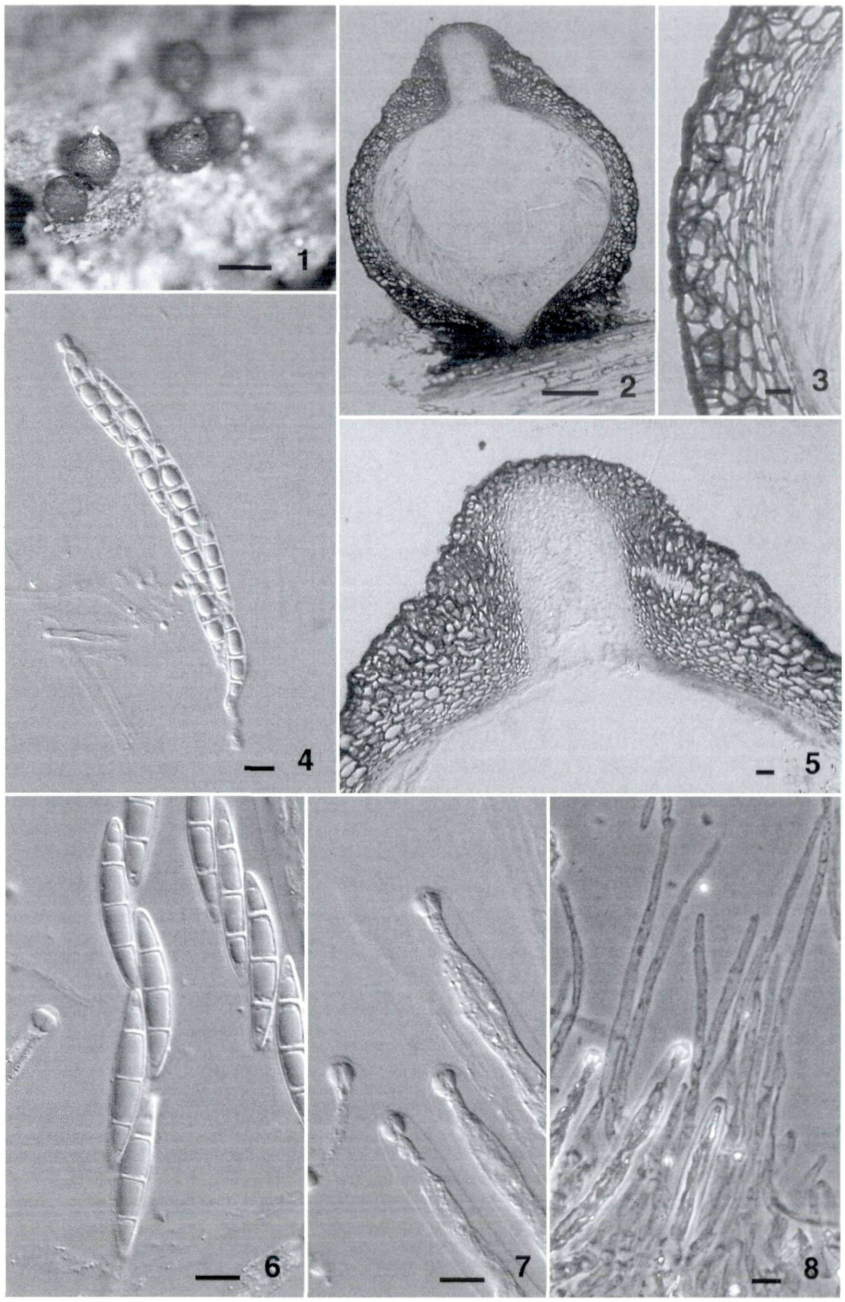
Type species. – *Mirannulata samuelsii* Huhndorf, F. A. Fernández, A. N. Mill. and Lodge

Etymology. – *mirus* = remarkable, *annulatus* = having a ring, refers to the prominent ascus apical ring.

Mirannulata samuelsii Huhndorf, F. A. Fernández, A. N. Mill. & Lodge, **sp. nov.** - Figs. 1–8.

Ascomata obpyriformia vel ovoidea, 480–525 µm diametro, 550–630 µm alta, papillata, pagina ascomatis glabrata. Paries ascomatis superficialis textura angularis-globosa, in sectione longitudinali 45–95 µm crassus, bistratosus. Papilla conica, 100–150 µm alta, 120–300 µm lata, periphysibus induta. Paraphyses angustae abundantes. Asci cylindracei, 140–185 × 13–18 µm, brevi-stipitati, octospori, biseriati, annulo apicali bipartito. Ascosporae fusiformes, 33.6–43.3(–48.4) × 5.4–7.3(–8.0) µm, triseptatae, hyalinae, guttulatae, sine vagina vel appendicibus.

Holotype. – UNITED STATES, PUERTO RICO: El Verde Research Area, Luquillo Mts., 16-ha Grid, base quadrat 02.01.41, 18° 19' 26 N, 65° 49' 1 W, [18.3167, -65.8167], 372 m, 14. – I. 1996, on 25 cm diam standing stump of *Tetragastris balsamifera* (Sw.) Kuntze (Burseraceae), SMH1880 (Holotype: F); Paratype. – near quadrat 01.05.42, 18° 19' 28 N, 65° 49' 3 W, 362 m, 28. – IV. 1995, on 25 cm diam log, SMH1211 (Paratype: F).



Figs. 1–8. *Mirannulata samuelsii*. – 1. Ascomata on substrate. 2. – Longitudinal section through ascoma. 3. – Longitudinal section through ascomatal wall. – 4. Ascus. – 5. Longitudinal section through neck wall. – 6. Ascospores. – 7. Ascus apex with refractive ring. – 8. Paraphyses. – Figs. 1 = macroscopic view; 2–7 = DIC; 8 = PH. Scale bars: 1 = 0.5 mm; 2 = 100 µm; 3–8 = 10 µm. Figs. 1, 4, 6–8 from SMH 1880; 2, 3, 5 from SMH 1211.

Anamorph. – None known.

Ascomata obpyriform to ovoid; not collapsing when dried; 480–525 µm diameter, 550–630 µm high; numerous; separate; superficial; papillate; surface glabrous, slightly roughened; dark brown appearing black. – Ascomatal wall of textura *angularis-globosa* in surface view; in longitudinal section 45–70 µm thick at the sides, thicker, up to 95 µm, at the sides of the base and near the apex, 2-layered, composed of polygonal to elongate, pseudoparenchymatic cells (9–25 × 6–10 µm diameter), with an external melanized crust; at the base smaller and isodiametric. – Ascomatal apex bluntly conical to broadly rounded; 100–150 µm high, 120–130 µm wide at the apex, 250–300 µm wide at the base; ostiole circular, 75–90 µm wide, periphysate. – Paraphyses tapering, narrowing toward the apex; 3–4.5 µm wide; abundant; persistent; without gelatinous coating. – Asci cylindrical; 140–185 × 13–18 µm; short-stalked; numerous; basal and lateral, lining the peripheral wall of the centrum; unitunicate; apex rounded, with large, bipartite ring, 4.8–6.0 µm high, 5.0–6.5 µm wide; with 8 biseriate ascospores. – Ascospores fusiform; 33.6–43.3(–48.4) × 5.4–7.3(–8.0) µm; straight to slightly curved; hyaline; smooth; 3-septate, without constrictions at septa, with prominent guttule in each cell; without sheath or appendages; spores collecting as a white droplet at the ascomal apex.

Etymology. – In honor of Gary J. Samuels.

Habitat. – On decorticated wood.

Known distribution. – Costa Rica, Ecuador, French Guiana, Puerto Rico.

Other material examined. – Costa Rica: San Jose, San Gerardo de Dota, Albergue de Montana, Savegre, trail to waterfall along Rio Savegre, 2150 m, [9.5439, -83.8142], 11. V. 1996, on 5 cm diam branch, SMH, FF, SMH2378; Puntarenas, La Amistad Pacifico, Las Tablas, 1700 m, [8.9492, -82.7772], 16. I. 1999, FF, SMH4028; Cartago, Cantón Paraiso, District Orosi, Parque Nacional Tapanti, Oropendola trail, 1300 m, [9.7517, -83.7908], 27. VI. 2000, on 10 cm log, FF, SMH4303; Alajuela, Cantón Upala, District Bijagua, Heliconias Station, Heliconias trail, 1190 m, [10.7081, -85.0453], 12. VII. 2001, on wood fragment, SMH, FF, ANM, M. P. DaRin, SMH4479, SMH4483. Ecuador: Orellana Prov, Yasuni National Park; Ceiba trail, [-.6713, -77.4005], 6. III. 2001, on 15 cm log, FF, ANM, R. Briones, SMH4357; Peru trail, 4. III. 2001, on rotten wood, FF, ANM, R. Briones, SMH4369; Mirador trail, 10. III. 2001, on 15 cm log, FF, ANM, R. Briones, SMH4419; Ceiba trail, 10. III. 2001, on wood fragment, FF, ANM, R. Briones, SMH4436. French Guiana: St-Laurent-du-Maroni Arrondissement: Canton de Maripasoula, Commune de Maripasoula, Upper Marouini River, 2 km N of Oumanfou-Lange Soula, 150 m, 12-14. VIII. 1987, on very rotten wood, GJS, J.-J. deGranville, L. Allorge, W. Hahn, M. Hoff, (NY, GJS5769); 2.5 km W of Monpe Soula between the Marouini River and a large granitic rock ca. 15 min walk W of river, 250 m, 26. VIII. 1987, on decorticated wood, GJS, J.-J. deGranville, L. Allorge, W. Hahn, M. Hoff,

A. Weitzman, (NY, GJS6049); Commune de Saül, Saül, ca. 7 km SW of Saül (03° 60' N, 53° 20' W) toward Mt. Galbao (03° 10 50' N, 53° 30' W), 450-500 m, 11. I. 1986, on log, GJS, J. R. Boise, (NY, GJS2535); Saül, ca. 10 km SW of Saül toward Mt. Galbao, ca. 200 m, I. II. 1986, on log, GJS, J. R. Boise (NY, GJS2804); ca. 7 km SW of Saül on trail to Mt. Galbao, 'Cambrouze', 200-300 m, 11-13. II. 1986, on decorticated wood, (NY, GJS3688); Monts La Fumée, dry primary forest, ca. 400 m, 4-6. II. 1986, on well rotted log, (NY, GJS3464); Eaux Claires, 5 km NE along the Sentier Botanique, 200 m, [3.7, -53.2], 1. IX. 1994, on decorticated wood, SMH742; S along Route de Belizon, 6. IX. 1994, SMH838; 10. IX. 1994, SMH937; headwater of Crique St. Eloi 5 km NE on Sentier Botanique, 7. IX. 1994, SMH872; at Crique Tortue ca. 1 km E on Sentier Botanique, 12. IX. 1994, SMH970, SMH972, SMH983; on decaying bark, SMH981; 7. XI. 1997, SMH3698; Cayenne Arrondissement: Canton de Sinnamary, Commune de Sinnamary, at km 16 on Route de Saint-Elie, 'ECEREX', ORSTOM research area, II-III. 1986, on well rotted wood, (NY, GJS3999); at km 103 along Hwy N1, 30 m, [5.3833, -53.0667], 18. IX. 1994, on decorticated wood, SMH1063; Canton de Matoury, Commune de Matoury, ca. 0.5 km NW of Aeroport de Rochambeau, [4.8333, -52.35], 1. XI. 1997, on old stump, SMH3641. United States, Puerto Rico: El Verde Research Area, Luquillo Mts., 16-ha Grid, 350 to 425 m, [18.3167, -65.8167], 27. IX. 1995, on 20 cm log, SMH1599; 25. I. 1996, on 25 cm log, SMH2042; 26. I. 1996, on 22 cm log, SMH2066; on 50 cm log, SMH2069; 29. I. 1996, on <1 cm twig, SMH2110; 30. I. 1996, on 20 cm log, SMH2152; 13. I. 1997, on 30 cm log, SMH, FF, SMH2910; 14. I. 1997, on stump, SMH2957; 15. I. 1997, on branch, SMH2968; on 30 cm log, SMH2971; 16. I. 1997, on wood fragment, SMH3010; 18. I. 1997, on 5 cm vine fragment, SMH3020; Bisley Watershed 3, Luquillo Mts., 220 m, [18.3167, -65], 8. V. 1995, on 30 cm log, SMH, DJL, SMH1389; 27. I. 1997, on 30 cm log, FF, SMH3139; on 1m log, SMH3144; on 40 cm log, SMH3151. (All F, except otherwise noted).

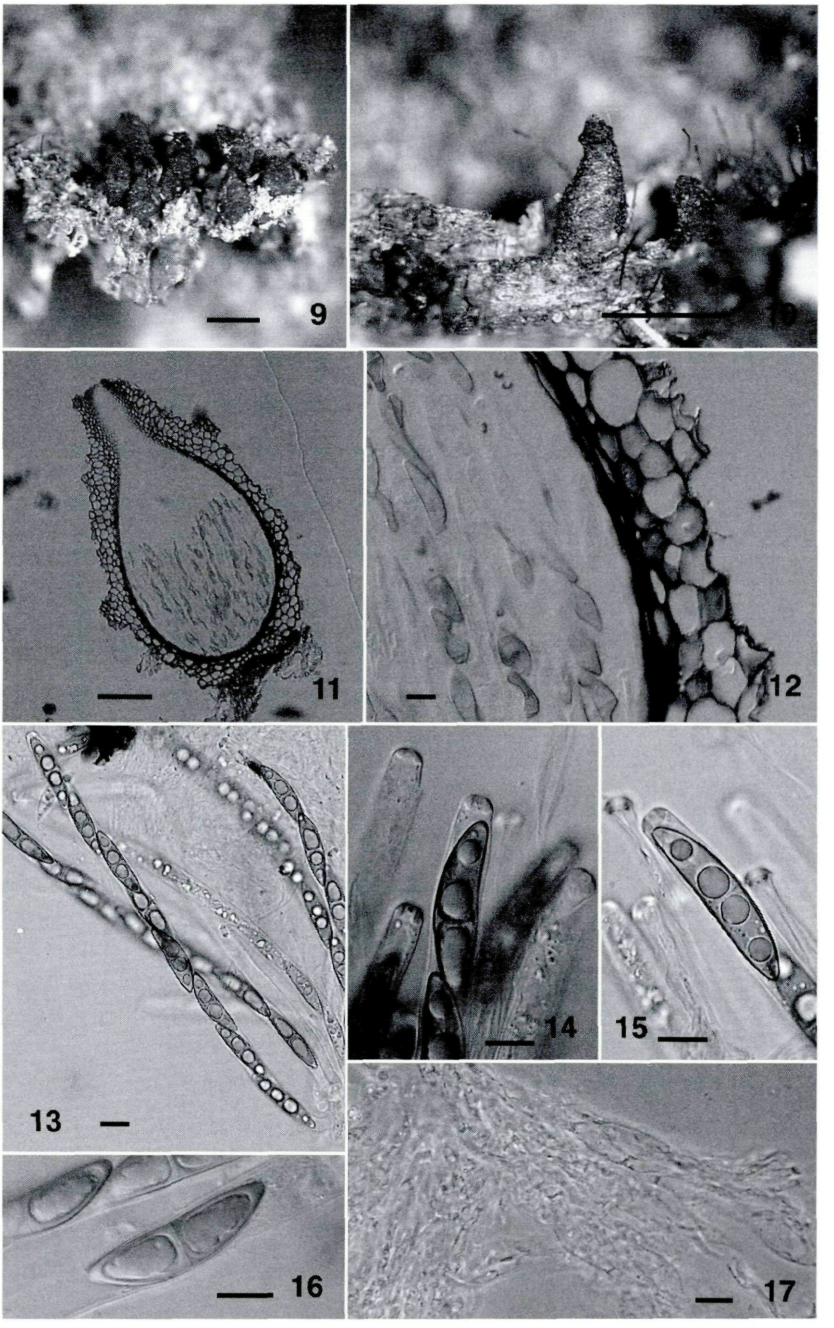
Mirannulata costaricensis Huhndorf, F. A. Fernández, A. N. Mill. & Lodge, **sp. nov**. - Figs. 9–17.

Similis *M. samuelsii* sed ascomata 300–400 µm diametro, 500–625 µm alta, collapsa. Paries ascomatis in sectione longitudinali 30–45 µm µm crassus, bistratosus. Papilla 130–140 µm alta, 80–180 µm lata, periphysibus induta. Paraphyses angustae sparsae. Asci 250–280 × 11–14 µm. Ascosporae fusiformes, 35–40 × 8.0–9.2 µm, uniseptatae, brunneae.

Holotype. – Costa Rica: San Jose, San Gerardo de Dota, Albergue de Montana, Savegre, Sendero la Quebrada, 2300 m, [9.5333, -83.8], 14. V. 1996, on wood fragment, SMH, FF, SMH2477 (Holotype USJ; Isotype F).

Anamorph. – None known.

Ascomata obpyriform to ovoid; collapsing somewhat laterally when dried; 300–400 µm diameter, 500–625 µm high; numerous; separate; superficial; papillate; surface irregularly, coarsely verrucose. – Ascomatal wall of textura *angularis-globosa* in surface view; in longitudinal section uniformly 30–45 µm thick, 2-layered, composed of polygonal, pseudoparenchymatic cells, without external melanized crust. – Ascomatal apex conical; 130–140 µm high, 80–90 µm wide at the apex, 160–180 µm wide at the base; ostiole circular, 40–50 µm wide, with periphyses. – Paraphyses tapering



Figures 9–17. *Mirannulata costaricensis*. – 9, 10. Ascomata on substrate. – 11. Longitudinal section through ascoma. – 12. Longitudinal section through ascomal wall. – 13. Ascus. – 14, 15. Ascus apex with refractive ring. – 16. Ascospores. – 17. Paraphyses. – Figs. 9, 10 = macroscopic view; 11, 12, 14, 16 = DIC; 13, 15 = BF; 17 = PH. Scale bars: 9, 10 = 0.5 mm; 11 = 100 µm; 12–17 = 10 µm. All from SMH 2477.

toward the apex; 2.5–4 µm wide; sparse; persistent; without gelatinous coating. – Asci cylindrical; 250–280 × 11–14 µm; short-stalked; numerous; basal and lateral, lining the peripheral wall of the centrum; unitunicate; apex rounded, with large, bipartite, refractive ring, 2.0–4.0 µm high, 5.5–6.5 µm wide; with 8 biseriate ascospores. – Ascospores fusiform, 35–40 × 8.0–9.2 µm; straight to slightly curved; brown; smooth; 1-septate, without constriction at septum, with prominent guttule in each cell; without sheath or appendages.

Etymology. – Refers to the collection locality.

Habitat. – On decorticated wood.

Known distribution. – Costa Rica, known only from type collection.

Mirannulata has a large, double-layered ascus apical ring similar to those found in *Annulatascus* K. D. Hyde and *Jobellisia* Barr. Hyde (1992) described the primarily aquatic genus *Annulatascus* for two taxa possessing this striking morphological feature: a large, bipartite apparatus in the ascus apex. Subsequently, further taxa were added to the genus and additional, predominantly aquatic genera were described that possessed similar large ascal rings but differed in other morphological features (Hyde, 1995; Hyde, 1996; Hyde & al., 1999; Wong & al., 1999). *Cataractispora* K. D. Hyde, S. W. Wong & E. B. G. Jones is one aquatic genus that has a similar ring and can occur superficially on the substrate. Huhndorf & al. (1999) also found a similar ascal ring in several species of the terrestrial genus *Jobellisia*. Prominent ascal apical rings also occur in other superficially occurring, wood-inhabiting genera such as *Ceratosphaeria* Niessl and *Ceratostomella* Sacc.

Mirannulata differs from *Jobellisia* in having short-stipitate asci and large, fusiform ascospores and from *Annulatascus* in its large, superficial, obpyriform ascomata. In *Annulatascus* the ascomata are completely immersed in the substrate, with more-or-less elongate, erumpent necks. Ascospore morphology differentiates *Cataractispora, Ceratosphaeria* and *Ceratostomella* from *Mirannulata*. *Cataractispora* has long, thread-like, polar appendages on the ascospores, *Ceratostomella* has one-celled, ellipsoid, hyaline or pale brown ascospores and *Ceratosphaeria* has filiform, hyaline ascospores. In addition, the ring in *Ceratosphaeria* and *Ceratostomella* is not bipartite. One structure that *Ceratosphaeria* does share with *Mirannulata* is the *textura globosa* nature of the outer ascal wall, a feature also found in other unrelated taxa such as *Lasiosphaeria raciborskii* (Penz. & Sacc.) G. C. Carroll & Munk.

Mirannulata samuelsii was found repeatedly in several tropical collecting localities in Costa Rica, Ecuador, Puerto Rico and French Guiana, and is probably quite common throughout the lowland forests in the Neotropics. *Mirannulata costaricensis* was found only once in Costa Rica. The two species share the feature of the ascus ring but differ in their ascospore morphology. *Mirannulata samuelsii* has hyaline, fusiform, three-septate ascospores, while *M. costaricensis* has brown, fusiform, one-septate ascospores. Both species have superficial ascomata.

There is evidence that the presence of a large ascus ring alone does not define a monophyletic group (Reblova & Winka, 2001). Huhndorf & al. (in press) include *Jobellisia, Annulatascus* and *Ceratosphaeria* LSU sequences in an analysis of the Sordariales and related taxa and the three genera resolve in separate clades in the tree. Sequences of *M. samuelsii* are included in a large analysis of taxa in the Sordariomycetes (Miller unpubl., data not shown) and it is determined to be unrelated to the other large-ringed taxa sampled. However its placement in the class is unresolved so we place *Mirannulata* in the Sordariomycetes inc. sed. for the present.

Acknowledgments

The production of the manuscript was supported in part by NSF PEET Grants (Partnerships for Enhancing Expertise in Taxonomy, DEB-9521926 and DEB-0118695) to SMH through the Field Museum of Natural History. Support for SMH's 1995-96 fieldwork was provided by the National Research Council Resident Research Associate Post-doctoral Program in cooperation with the USDA Forest Service, Forest Products Laboratory, Madison, WI. We thank Drs. Jill Thompson and Jess Zimmerman for access to the forest grid at El Verde Field Station. The 1997 collecting trip to French Guiana for SMH was supported by a National Geographic Society Grant (No. 5769-96) awarded to Dr. Scott A. Mori, New York Botanical Garden. Two collecting trips to Ecuador were supported by a NGS grant (No. 6914-00) awarded to FF. The New York Botanical Garden is thanked for the loan of the Samuels collections.

References

Huhndorf, S. M. (1991). A method for sectioning ascomycete herbarium specimens for light microscopy. – Mycologia 83: 520–524.

—— & F. A. Fernández. (1998). Neotropical Ascomycetes 7. *Caudatispora biapiculatis* sp. nov. from Puerto Rico. – Sydowia 50 (2): 200–204.

——, —— & D. J. Lodge. (1999). Neotropical Ascomycetes 9. *Jobellisia* species from Puerto Rico and elsewhere. – Sydowia 51 (2): 183–196.

Hyde, K. D. (1992). Tropical Australian freshwater fungi. II. *Annulatascus velatispora* gen. et sp. nov., *A. bipolaris* sp. nov. and *Nais aquatica* sp. nov. (Ascomycetes). – Australian Systematic Botany 5: 117–124

—— (1995). Tropical Australian freshwater fungi. VII. New genera and species of Ascomycetes. – Nova Hedwigia 61: 119–140.

—— (1996). Tropical Australian freshwater fungi. X. *Submersisphaeria aquatica* gen. et sp. nov. – Nova Hedwigia 62: 171–175.

Hyde, K. D., S. W. Wong, & E. B. G. Jones. (1999). *Cataractispora* gen. nov. with three new freshwater lignicolous species. – Mycol. Res. 103: 1019-1031.

Réblová M, & K. Winka (2001). Generic concepts and correlations in ascomycetes based on molecular and morphological data: *Lecythothecium duriligni* gen. et sp. nov. with a *Sporodesmium* anamorph, and *Ascolacicola austriaca* sp. nov. – Mycologia 93: 478–493.

Wong, S. W., K. D. Hyde, E. B. G. Jones & S. T. Moss. (1999). Ultrastructural studies on the aquatic ascomycetes *Annulatascus velatisporus* and *A. triseptatus* sp. nov. – Mycol. Res. 103: 561–571.

(Manuscript accepted 6th June 2003)