

**ACROSPERMUM CHILENSE SP. NOV. FROM CHILE AND THE  
ACROSPERMALES ORD. NOV.\***

D.W. MINTER<sup>1</sup>, H.L. PEREDO<sup>2</sup> and ANNA T. WATSON<sup>3</sup>

**Summary:** *AcrospERMUM chilense* sp. nov. on living stems of *Chusquea quila* from southern Chile is described, illustrated and discussed. The asci and ascospores, more than 1000 µm long, may be the longest ever found in the ascomycetes. The ordinal position of the family AcrospERMACEAE is discussed, and the order AcrospERMALLES ord. nov. is erected to accommodate it.

**Key words:** AcrospERMACEAE, AcrospERMALLES, ascospores, *Chusquea*, Chile.

**Resumen:** Se describe, ilustra y discute *AcrospERMUM chilense* sp. nov. sobre tallos de *Chusquea quila* del Sur de Chile. Los ascos y ascosporas, de más de 1000 µm de longitud, pueden ser las más largas halladas en ascomicetes. La posición ordinal de la familia AcrospERMACEAE es discutida y el orden AcrospERMALLES ord. nov. es propuesto para ubicarla.

**Palabras clave:** AcrospERMACEAE, AcrospERMALLES, ascosporas, *Chusquea*, Chile.

**Dedication**

The authors dedicate this work to that most distinguished Argentine mycologist, Prof. Irma Gamundí, on the occasion of her birthday. The senior author, in particular, wishes to thank her for companionable field excursions together, and for fruitful collaboration. Although this beautiful new species was collected in Chile, the type locality is no more than a few kilometres across the border from Argentina's Puerto Blest, one of her favourite foraging areas. The authors therefore collectively delight in challenging her and her colleagues to find this fungus on her side of the Andes!

**INTRODUCTION**

During fieldwork by two of the authors (DWM & HLP) in the Puyehue National Park, southern Chile in October 1995, a remarkable fungus was encountered on living stems of *Chusquea quila* Kunth, a native bamboo. That fungus is described and illustrated in the present paper, with a discussion of the taxonomic position of the family in which it is placed.

**MATERIALS AND METHODS**

Fresh specimens were collected on 22 October 1995 from *Chusquea quila* stems growing by the side of the track from the Puyehue National Park

Administration Building at Aguas Calientes (72°19'W, 40°44'S, alt. 440 m) to Antillanca in the Puyehue National Park in southern Chile. Most fruitbodies were found within 2 km of Aguas Calientes, on the south side of the track. The fungus had been previously observed by HLP, but not collected, in Puyehue National Park at the Mirador El Pionero, and at San Pablo de Tregua in the Cordillera de Pangipulli. Unsuccessful searches were also made for this fungus in October 1995 by DWM & HLP in the national parks of Conguillio, Laja, Nahuelbuta and Pérez Rosales, and by DWM at Puerto Blest, near Bariloche in Argentina. Sections 10 µm thick of the ascomata were made using a freezing microtome, then mounted in cotton blue in lactic acid, and lactofuchsin. Temporary squash mounts in erythrosin in ammonia, and semi-permanent squash mounts in cotton blue and lactic acid were also prepared. The fungus was then examined with an Olympus BH2 compound light microscope.

\*Trabajo publicado en homenaje a la Dra Irma J. Gamundí en conmemoración de su 80° aniversario.

<sup>1</sup>CABI, Bakeham Lane, Egham, Surrey, TW20 9TY, UK.

<sup>2</sup>Instituto Silvicultura, Facultad de Ciencias Forestales, Universidad Austral de Chile, Casilla 567, Valdivia, Chile.

<sup>3</sup>11 Hall Croft, Skipton, North Yorkshire, BD23 1PG, UK.

## RESULTS

### *Field observations*

With the unaided eye, and using a hand lens, the fungus was observed on apparently healthy young stems, towards their tips, at about 1.5–2 m above ground level. Only a small number of plants were infected, but where the fungus was present, it was frequently on more than one stem of the plant. Infected plants were sometimes surrounded by uninfected plants; occasionally two or more infected plants were adjacent. One to six (but most often three) elongated, clavate fruitbodies projected from the stem, usually within 5 mm of the base of a leaf lamina. The fruitbodies were black, often rather flattened in cross-section, and measured approximately 1–4 mm from base to tip. They were frequently bent, making the main axis of many fruitbodies not perpendicular to the main axis of the bamboo stem (through this was highly variable in extent). In some cases the fruitbodies appeared to be growing superficially on the stem, but more frequently they were erumpent from a small crack in the plant tissue. Even close to the fruitbody the stem showed no marked lesion or discoloration.

### *Microscopic observations*

Viewed with a dissecting microscope, each fruitbody was black, matt but rather velvety, 1–4 mm long, 0.2–0.4 mm wide at the mid point. Most were twisted helically along the main axis, and gentle pressure from one side caused them to bend rather than snap. Some had transverse circumferential ridges in groups at the base and/or singly further up. In some cases, in the lower part, the fruitbodies appear to become abruptly wider distally, though this may have been accentuated by the helical twisting. This widening suggested that the fruitbody was composed of a single ascoma on a stalk. In other cases the change in width from presumed stalk to ascoma was so gradual that it was not possible to tell from the outside where the stalk ended and the ascoma began. The stalk was approximately 1–2 mm long, and the ascoma was about 1.8–2.1 mm long. Viewed from above, a white circular ostiolar disk 70–150  $\mu\text{m}$  diam with a thin and possibly crescent-shaped pore could be seen at the tip of some fruitbodies.

In vertical section the fruitbody comprised a single long thin ascoma supported by a stalk. The ascoma consisted of a single chamber in which asci, ascospores and paraphyses were produced, with a small opening at the apex. The cavity ended before

the fruitbody base, where the structure narrowed to become the stalk. Twisting and bending of the fruitbodies made it impossible to obtain a single section showing all of both the ascoma and the stalk. Because material was scarce only one ascoma was examined in its entirety in vertical section. The stalk was composed of colourless cells 2–4  $\mu\text{m}$  wide, with thick refractive walls, forming a *textura intricata* of uniform density and pigmentation, but with black or dark brown deposits in and around the walls of the outermost cells. The ascomatal wall was 45–70  $\mu\text{m}$  thick along its sides, and had two layers. The outer layer was 30–50  $\mu\text{m}$  thick, and composed of tightly packed colourless cylindrical cells, 2–4  $\mu\text{m}$  wide, with thick refractive walls, which usually ran parallel to the ascoma main axis, forming a *textura oblita*. These cells gradually merged with the stalk cells towards the base of the ascoma, and formed a thicker region at the ascomatal apex. The outermost cells of this outer layer were covered with black or dark brown deposits, with a slightly wrinkled outer surface which may account for the velvety external appearance of the fruitbody. As a result, at times, it almost seemed as though there were another outermost layer of dark walled *textura angularis*. The inner layer was 15–20  $\mu\text{m}$  thick, and composed of tightly packed colourless cylindrical cells, 1–2  $\mu\text{m}$  wide, with thin walls, also usually running parallel to the ascoma main axis, also thickening towards the apex, and also forming a *textura oblita*. Although it intergraded to some extent with the outer wall, in some sections the action of the knife had caused it to separate from that outer layer. Asci lay in the inner cavity of the ascoma, parallel to its longitudinal axis.

In squash preparations asci were narrowly cylindrical, up to at least 1300  $\mu\text{m}$  long and 6–8  $\mu\text{m}$  wide, with a wall uniformly about 1  $\mu\text{m}$  thick except at the apex where it was slightly thicker with a poorly-defined apical nasse. Other than this slight thickening and nasse, no apical pore or other special opening mechanism was noted, and no open asci were observed. In broken asci, the ascus wall was not observed to separate into different functional layers, but this feature was in any case not easy to see. Their extreme length made it difficult to locate bases of individual asci, and no observations were made of this part of the ascus. Each ascus contained at least 4 and probably 8 ascospores, the tips of these spores being usually about 20–30  $\mu\text{m}$  below the ascus apex. The ascospores were arranged in a bundle within the ascus, and were not observed to coil markedly.

Ascospores were colourless, thin-walled, smooth, filiform with a rounded apex, and apparently slightly shorter than the asci. Their extreme length made them difficult to measure accurately, but they were at least 1000 µm long by 1–1.5 µm wide, with no mucous sheath observed. At least some ascospores had many transverse septa while still inside the ascus, though fragmentation into part spores was not observed. Paraphyses, which were observed in only some of the preparations, were colourless, thin-walled, smooth, up to about 1 µm wide. It was not clear whether they had branches and septa.

## DISCUSSION

### *Taxonomic position of the current fungus*

The most remarkable feature of this fungus is the great length of its asci and ascospores. A quick search was made through various general publications about ascomycetes, particularly those with filiform ascospores (Breitenbach & Kränzlin, 1981; Dennis, 1978; Eriksson, 1981; Sherwood, 1977) where the largest asci reported were about 600 µm in length. The present fungus appears therefore to have asci more than twice as long as any commonly known ascomycete. Long thin asci and filiform ascospores occur in only a small number of ascomycete groups. The orders Clavicipitales (part of the Hypocreales in Hawksworth *et al.*, 1995), Hysteriales (part of the Dothideales in Hawksworth *et al.*, 1995), Ostropales and Rhytismatales probably together hold most of these fungi.

In the present fungus, the ascomata are long, thin, vertically-oriented and external to the tissue of the associated organism. These characteristics are not found in the Ostropales or Rhytismatales. In the Hysteriales, such ascomata exist, but their walls are shiny and often extremely brittle (described as «carbonaceous» in many accounts), whereas those of the present fungus are matt, velvety and softer in texture. Ascomata of the Hysteriales also open by a clearly-defined split. In the present fungus, however, it is not clear whether the apical aperture is a split or a circular ostiole. If it is a split, it is certainly not clearly-defined.

The association of the present fungus with living stems of grass is strongly reminiscent of the Clavicipitales, which has many members parasitic on this plant family. The ascomata of the Clavicipitales are, however, generally of a bright colour, and their asci have characteristically thickened apices in which

a narrow pore can easily be observed. In the present fungus, in contrast, the ascomata are darker, and the ascus apices, while slightly thickened, are not at all reminiscent of the Clavicipitales. Indeed the ascus apices of the present fungus, with the poorly defined nasse, are more similar in general appearance to the «bitunicate» asci of the Dothideales, although two functional wall layers could not be distinguished in this study, and it is hard to imagine how a jack-in-the-box mechanism of ascus extension could be engineered in such long thin asci.

If the keys to the families of fungal phyla in the 8<sup>th</sup> edition of *Ainsworth & Bisby's Dictionary of the Fungi* (Hawksworth *et al.*, 1995) are used with the current fungus, one arrives at couplet 47. In this couplet a distinction is made between fungi having asci with one wall layer and those having asci with two wall layers. If the option for two wall layers is followed, it leads to no obvious match. Following the other option, one arrives in time at couplet 171. This couplet distinguishes fungi with brightly coloured ascomata from those with black ascomata. Taking the option for black ascomata again leads to no obvious match. The option for brightly coloured ascomata, however, leads to the family Acrospermaceae, another ascomycete group with long thin asci.

The family Acrospermaceae, as circumscribed in the *Dictionary*, has two genera *Oomyces* Berk. & Broome and *Acrospermum* Tode. *Oomyces* contains one currently accepted species, *O. carnealbus* (Lib.) Berk. & Broome, and seven other names. The genus has brightly coloured ascomata which are occasionally united to form a stroma containing more than one locule (Eriksson, 1981). *Acrospermum* Tode in contrast contains several currently accepted species, and many other names. There appear to be no accounts of ascomata of *Acrospermum* species uniting to form multilocular stromata, and its ascomata vary in colour from bright yellows to black. Eriksson (1981) reported that more than one wall layer could be discerned in asci of *Acrospermum*, but that they did not appear to open by a jack-in-the-box mechanism. Although asci with more than one wall layer were not detected in the current work, examination of descriptions of *Acrospermum* species and of specimens identified as *Acrospermum* in herb. IMI, made it clear that the current fungus belongs in this genus.

*Comparison of the current fungus with other species of Acrospermum*

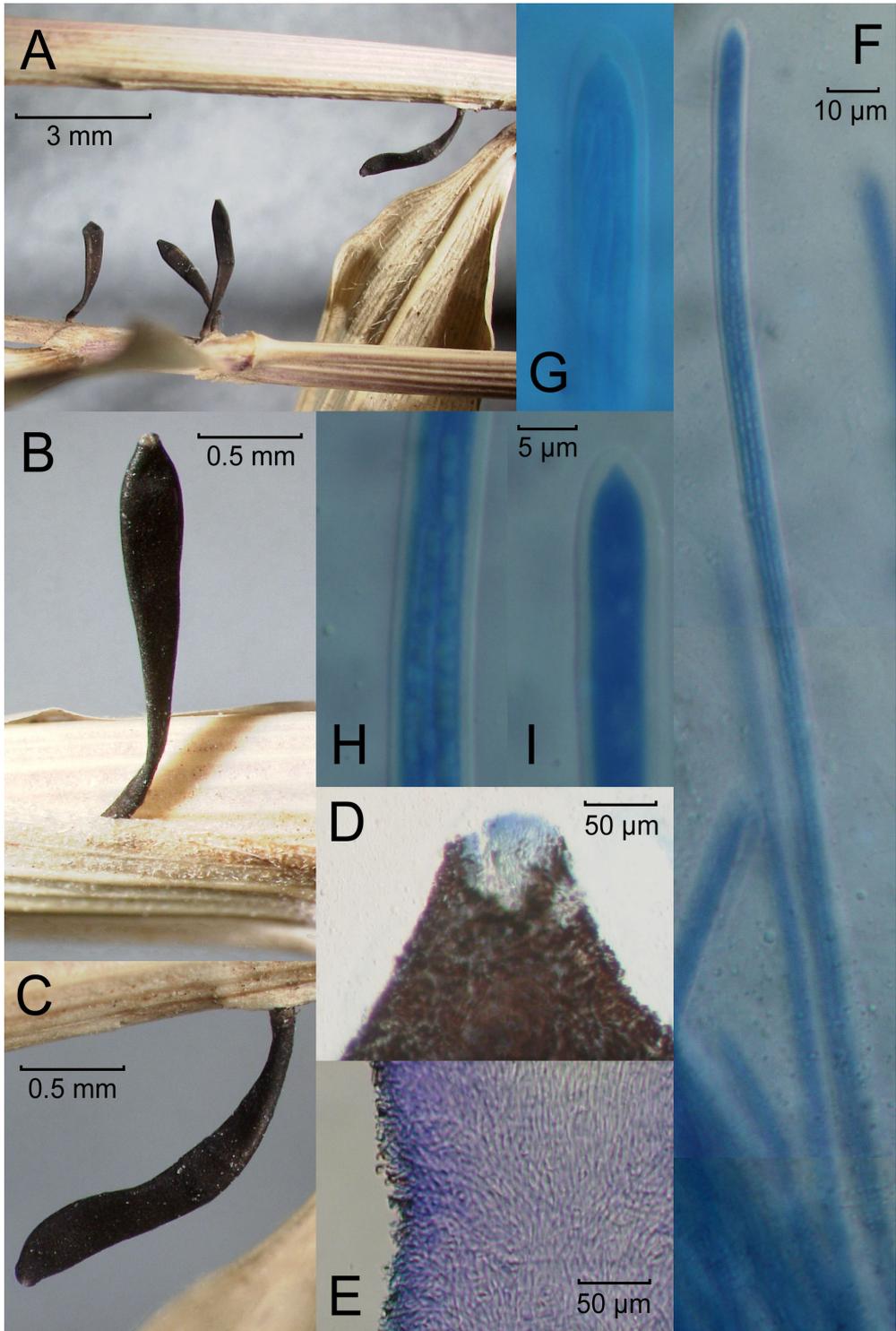
A search for names in *Acrospermum* was made in *Index Fungorum* ([www.indexfungorum.org](http://www.indexfungorum.org)), the on-line nomenclatural database (collectively containing over 405,000 different fungus names). This revealed 52 specific epithets, and eight subspecific epithets. Wherever possible, the protologue was examined for each of these names. In cases where the protologue was unavailable, in all cases for older names, the description in Saccardo's *Sylloge Fungorum* was used. In many cases the substratum or place of collection mentioned in the protologue enabled the name to be eliminated as a potential candidate for the current species.

The following *Acrospermum* species and varieties were described originally from South America: *Acrospermum antarcticum* Speng. (Argentina: Tierra del Fuego, Staten Island, Port Cook), *A. antennariicola* Speng. (Argentina), *A. bignoniicola* Henn. (Brazil), *A. bromeliacearum* Theiss. (Brazil: San Leopoldo), *A. coniforme* Speng. (Argentina), *A. corrugatum* Ellis (Chile: Straits of Magellan, Punta Arenas), *A. minutum* Henn. (Brazil), *A. ochraceum* Syd. & P. Syd. (Brazil: Rio de Janeiro, Tijuca), *A. puiggarii* Speng. (Brazil: Apiahy), *A. syconophilum* Speng. (Argentina: Misiones, Puerto León). The following were described from grasses, including bamboos: *A. conicum* Pers. (Gramineae), *A. coniforme* (*Diplachne latifolia*), *A. decipiens* Pass. (*Brachypodium sylvaticum*), *A. graminum* Lib. (*Poa*, *Festuca*, *Calamagrostis*), *A. graminum* var. *robustius* Fuckel (*Brachypodium sylvaticum*), *A. ochraceum* (*Bambusa*). Only two *Acrospermum* species, therefore, have previously been described from grasses in South America: *A. coniforme* from Argentina on *Diplachne*, and *A. ochraceum* from Brazil on *Bambusa*. Although the exact location of collection of *A. coniforme* is not cited by Farr (1973), *Diplachne* is a grass genus of the tropics and warm temperate regions, and is not a bamboo. Although *A. ochraceum* was described from a bamboo (the only existing species specifically associated with this plant), the original collection was also from the tropics, and is specifically described in the protologue as being «in foliis emortuis vel putrescentibus». The present fungus, from cool-temperate rain forest, on living bamboos, is clearly different from both of these species, which were the two most similar in habitat preferences. It was also different from these and all other existing species in the size of its asci and ascospores: in no other case was an ascus length reported greater than 600 µm.

#### *Ordinal position of the Acrospermaceae*

The ordinal position of the Acrospermaceae has never properly been settled. In the 9<sup>th</sup> edition of *Ainsworth & Bisby's Dictionary of the Fungi* (Kirk *et al.*, 2001), and in the preceding edition, the family was listed as of uncertain affinity. In the 1<sup>st</sup> and 2<sup>nd</sup> editions it was allocated to «Hysteriales or Dothideales»; in the 3<sup>rd</sup> to 5<sup>th</sup> editions it was placed in the Dothideales; in the 6<sup>th</sup> it was put in the Ostropales; in the 7<sup>th</sup>, it was placed tentatively in the Clavicipitales. In all editions of «Outline of the Ascomycetes» in *Systema Ascomycetum* (eg Eriksson & Hawksworth, 1991), the Acrospermaceae is listed as a family of uncertain affinity. Brandriff (1936) speculated that it might be related to the Coryneliales or Pseudosphaeriales (ie Dothideales). The family was placed rather doubtfully in the Pyrenulales by Eriksson (1982). Sherwood (1977) discussed its ordinal position briefly, stating that it was not close to the Ostropales, but speculating a relationship with the Clavicipitales. Barr (1990) discussed its ordinal position in more detail, and placed it in the Xylariales.

The Pyrenulales, Dothideales and Hysteriales (if different from the Dothideales) can all be rejected as potential homes for this family because their asci have two functional wall layers, with some sort of jack-in-the-box mechanism. As already noted, the Hysteriales typically open by an elongated apical split, and are black and very brittle. The Dothideales tend to have longitudinally asymmetrical ascospores constricted at the primary septum (Hawksworth *et al.*, 1995). The Pyrenulales are usually lichenized and their ascomata are often immersed and rather flattened in the horizontal plane (Hawksworth *et al.*, 1995). Recent work has shown that asci in the Coryneliales have two functional wall layers, of which the outer splits at a very early stage in development (Johnston & Minter, 1989), a form of development very unlike that of the Acrospermaceae. If Sherwood's (1977) widely accepted concept of the Ostropales is followed, it is hard to see how members of the Acrospermaceae, being largely superficial and perithecial, can be fitted into an order containing immersed discomycetous fungi. The Clavicipitales, synonymized with Hypocreales by Hawksworth *et al.* (1995), have asci with very characteristic thickened apices quite unlike those of the Acrospermaceae. Barr's (1990) placement of the family in the Xylariales, apparently on the basis of free ended paraphyses, is «no more probable than any of the others» (Eriksson & Hawksworth, 1991).



**Fig. 1.** *Acrospermum chilense*. **A.** Habit on living stem. **B & C.** Ascomata on stems viewed with stereo microscope. **D.** Ostiole viewed with compound microscope. **E.** Ascomatal stalk in vertical section. **F.** Upper part of ascus. **G.** Ascus apex with ascospore apices. **H.** Part of median portion of ascus. **I.** Apex of young ascus without ascospores. **G, H & I** all same magnification.

## CONCLUSIONS

The current fungus appears to be a new and undescribed species of *Acrospermum* and is formally described as such below. A new order, the Acrospermales, is needed to accommodate the

Acrospermaceae, and this is also formally described below.

The following modification to couplet 171 of the keys to ascomycetes in Hawksworth *et al.* (1995, page 511) is required, to take into account the black fruitbodies of the current species.

171 (170) Not lichenized; stromata (if present) and ascomata brightly coloured, usually yellow, orange or red (or, if black, then with exceptionally long, thin, cylindrical asci)

172

Lichenized or not lichenized; stromata (if present) and ascomata black (at least partially) at maturity, but not with exceptionally long, thin, cylindrical asci

176

***Acrospermum chilense*** Minter, Peredo & A.T. Watson, **sp. nov.** (Fig. 1)

Fungus ad genus *Acrospermi* pertinens et ascis et ascosporis plus quam 1000 µm longitudinis respectu gaudet. **Holotypus**: on living stems of *Chusquea quila*, Chile, Región de Los Lagos: Puyehue National Park, within 2 km of Aguas Calientes (72°19'W, 40°44'S, alt. 440 m), on the track to Antillanca, 22 October 1995, leg. H.L. Peredo & D.W. Minter (IMI 365246). **Etymology**: the specific epithet, *chilense*, celebrates the country in which this fungus was collected: in their extreme length, the remarkable asci bear resemblance to the geographical shape of Chile.

**Acrospermales** Minter, Peredo & A.T. Watson, **ordo nov.**

Ascomycetes sunt, superficiales vel erumpentes, stipibus induti, stromatici, peritheciales, ostiolati vel parvis cum rimis dehiscentes. Habent ascos cylindricos, saepius longissimos, quorum paries (licet cum microscopo et luce videatur) unicum stratum habet. Horum fungorum ascosporae quoque longissimae saepius et cylindricae sunt, et septis multis gaudent. Apex cuiusque asci incrassata est. **Typus**: Acrospermaceae Focke.

## ACKNOWLEDGEMENTS

CONAF and The British Council are warmly thanked for financial and other assistance.

## BIBLIOGRAPHY

BARR, M.E. 1990. Prodromus to nonlichenized,

pyrenomycetous members of class Hymenoascomycetes. *Mycotaxon* 39: 43-184.

BRANDRIFF, H. 1936. The development of the ascocarp of *Acrospermum compressum*. *Mycologia* 28: 228-235.

BREITENBACH, J. & F. KRÄNZLIN. 1981. *Champignons de Suisse 1, Les Ascomycetes*. 310 pp., Switzerland, Lucerne, Edition Mykologia.

DENNIS, R.W.G. 1978. *British Ascomycetes*. Edn 3, 585 pages, Liechtenstein, Vaduz, J. Cramer.

ERIKSSON, O. 1981. The families of bitunicate ascomycetes. *Opera Botanica* 60: 1-220.

ERIKSSON, O. 1982. Outline of the ascomycetes - 1982. *Mycotaxon* 15: 203-248.

ERIKSSON, O. & D.L. HAWKSWORTH. 1991. Notes on ascomycete systematics. Nos 1128-1251. *Systema Ascomycetum* 10 (1): 27-67.

FARR, M.L. 1973. An annotated List of Spegazzini's Fungus Taxa. *Bibliotheca Mycologica* 35 (1): 1661 pp.

HAWKSWORTH, D.L., P.M.; KIRK, B.C SUTTON & D.N. PEGLER. 1995. *Ainsworth & Bisby's Dictionary of the Fungi*. 8<sup>th</sup> Edn, 616 pp., UK, Wallingford, CAB International.

JOHNSTON, P.R. & D.W. MINTER. 1989. Structure and taxonomic significance of the ascus in the Coryneliaceae. *Mycological Research* 92 (4): 422-430.

KIRK, P.M., P.F. CANNON, J.C DAVID & J.A STALPERS. 2001. *Ainsworth & Bisby's Dictionary of the Fungi*. 9<sup>th</sup> Edn, 655 pp., UK, Wallingford, CAB International.

SHERWOOD, M.A. 1977. The ostropalean fungi. *Mycotaxon* 5 (1): 1-277.

Recibido el 14 de Febrero de 2007, aceptado el 09 de Abril de 2007.