Cryptogamie, Mycologie, 2003, 24 (1): 13-20 © 2003 Adac. Tous droits réservés

Nigramammilla calami gen. et sp. nov. and Arecomyces calami, A. licualae and Pseudohalonectria palmae spp. nov. from palms

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Abstract – *Nigramammilla calami, Arecomyces calami, A. licualae* and *Pseudohalonectria palmae* are four new ascomycete species described from palms. *Nigramammilla* cannot be accommodated in any other genus and is introduced here as a new genus. Each new species is described and illustrated and compared with related taxa.

Ascomycetes / new genera / new species / palm fungi

INTRODUCTION

There has been considerable interest in the last decade in fungi associated with palms, as this substrate appears to support an high diversity of fungi (Fröhlich & Hyde, 2000; Hyde *et al.*, 2000; Yanna *et al.*, 2001; 2002). In this paper we describe three new species, two in the genus *Arecomyces* and one in *Pseudohalonectria*. We also introduce a new genus *Nigramammilla* to accommodate one new species that cannot be included in an existing genus. All taxa are compared with similar taxa and brief discussions of each of the genera are also provided.

TAXONOMY

Arecomyces calami K.D. Hyde & J. Fröhl., sp. nov.

(Figs 1-8)

Ascomata 277-340 × 205-268 µm, subglobosa, immersa, ostiolata, solitaria vel gregaria. Asci 75-100 × (8.8-)10-12.5 µm, 8-spori, cylindrici, breve pedicellati, unitunicati, truncati, apparatu apicali ca 0.5 µm alti, 2.5 µm diametro praediti. Ascosporae 14-17.5 × 4.5-7 µm, bi-seriatae, ellipsoideae vel fusiformis, unicellulae, hyalinae, guttulae, tunica gelatinosa praeditae.

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Figs 1-8. *Arecomyces calami* (from holotype). 1: Appearance of ascomata on host surface. Note that only the ostioles are visible (arrowed). 2: Vertical section through the ascoma. 3: Vertical section through the central ostiole. 4: Vertical section through the peridium. 5: Paraphyses. 6,7: Asci. 8: Ascospores. Note the mucilaginous sheaths. (Scale bars: $1, 2 = 100 \mu m$; $3-8 = 10 \mu m$).

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Etymology: In reference to the host.

Ascomata 277-340 µm wide, 205-268 µm high, subglobose, immersed beneath a pale, but otherwise unaltered area of host tissue with a central, periphysate ostiole, 50-55 µm in diameter, solitary, scattered (Figs 1-3). Peridium 5-15 µm thick, composed of 3-4 layers of flattened cells with thin, very-dark brown walls (Fig. 4). Paraphyses numerous, 1.25-2.5 µm wide, septate, filamentous and tapering (Fig. 5). Asci 75-100 × (8.8-)10-12.5 µm ($\bar{\mathbf{x}} = 89.2 \times 10.9$ µm, n = 25), 8spored, cylindrical, broadest in the centre, short pedicellate, unitunicate, with a truncate apex and a non-amyloid, refractive, apical ring ca 2.5 µm in diameter, 0.5 µm high (Figs 6, 7). Ascospores 14-17.5 × 4.5-7 µm ($\bar{\mathbf{x}} = 15.7 \times 5.7$ µm, n = 50), biseriate, ellipsoidal to fusiform with one slightly more rounded, and one slightly more pointed end, unicellular, hyaline, guttulate and with a mucilaginous sheath (Fig. 8).

Host species: Calamus conirostris.

Known distribution: Brunei Darussalam.

Material examined: BRUNEI DARUSSALAM: Temburong, Batu Apoi Forest Reserve, Sungai Belalong, Kuala Belalong Field Studies Centre (KBFSC), Baki Tributary, on dead rattan of *Calamus conirostris*, July 1993, J. Fröhlich (HKU(M)JF 72, **holotype designated here**).

Arecomyces licualae K.D. Hyde & J. Fröhl., sp. nov. (Figs 9-16) Ascomata 296-324 × 65-87.5 μm, lenticularis, immersa, ostiolata. Asci 67-102.5 × (10-)12-19.5 μm, 8-spori, fusiformes, breve pedicellati, unitunicati, apparatus subapicali 0.5-1 μm alti, 2-3.6 μm diametro praediti. Ascosporae 16.3-22.5 × 4.5-6.5(-7) μm, 2-3-seriatae, naviculae, unicellulae, hyalinae, tunica gelatinosa praeditae.

Etymology: In reference to the host.

Ascomata 296-324 µm wide, 65-87.5 µm high, lenticular, clustered in small groups of 2-6, immersed beneath slightly raised areas of host epidermis which are concolourous with the surrounding tissue, with an inconspicuous central, pore-like ostiole (Figs 9-11). *Peridium ca* 10 µm thick, composed of 1-4 layers of flattened, irregular cells with very thin, hyaline walls (Fig. 12). *Paraphyses* 2.5-3 µm wide, numerous, septate, filamentous, tapering, of similar length to, or slightly longer than the asci (Fig. 16). *Asci* 67-102.5 × (10-)12-19.5 µm ($\bar{\mathbf{x}} = 83.2 \times 13.3$ µm, n = 25), 8-spored, fusiform, widest in the centre, short pedicellate, unitunicate, with a non-amyloid, refractive, subapical ring, 0.5-1 µm high, 2-3.6 µm in diameter (Figs 14, 15). *Ascospores* 16.3-22.5 × 4.5-6.5(-7) µm ($\bar{\mathbf{x}} = 18.3 \times 5.3$ µm, n = 50), 2-3-seriate, navicular, unicellular, hyaline, smooth and with a thin, inconspicuous mucilaginous sheath (Fig. 13).

Host species: Licuala sp.

Known distribution: Brunei Darussalam.

Material examined: BRUNEI DARUSSALAM: Temburong, Batu Apoi Forest Reserve, Sungai Belalong, Kuala Belalong Field Studies Centre (KBFSC), Ruth Levy's Plot, on dead petiole of *Licuala* sp. 1, July 1993, J. Fröhlich (HKU(M)JF 57, **holotype designated here**).

Remarks: These two species could be accommodated in *Arecomyces*, *Hyponectria* or *Physalospora*. *Arecomyces* was introduced by Hyde (1996) for taxa from palms and comprises species with ascomata often immersed beneath a pseudostroma or clypeus, broadly cylindrical, short pedicellate asci, with a J-, discoid, refractive apical ring and unicellular ascospores surrounded by a mucilaginous sheath (Hyde *et al.*, 2000). *Hyponectria* and the *Hyponectriaceae* have been discussed by Wang & Hyde (1999). In *Hyponectria* ascomata lack a clypeus, asci



Figs 9-16. Arecomycyes licualae (from holotype). 9, 10: Appearance of ascomata on host surface. 11: Vertical section through the ascoma. 12: Vertical section through the corner of the ascoma illustrating the horizontal orientation of the asci and the thin peridium. 13: Ascospores. 14: Ascus. 15: Ascus tip illustrating the refractive, non-amyloid, subapical ring. 16: Paraphyses. (Scale bars: 9-11 = 100 μ m; 12-14, 16 = 10 μ m; 15 = 1 μ m).

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have an indistinct apical ring and ascospores are unicellular. In *Physalospora* asci are cylindrical, short-stalked, and evanescent (Sivanesan & Shivas, 2002). The two new fungi are best accommodated in *Arecomyces* on account of the refractive discoid apical rings in the asci, however there is considerable overlap in diagnostic characters of these three genera and molecular studies and anamorphic data are needed to resolve their phylogeny.

Arecomyces epigeni K.D. Hyde and A. tetrasporus K.D. Hyde are most similar to A. calami and A. licualae (Hyde, 1996) in ascospore shape. In A. epigeni ascomata are visible as raised conical areas and the clypeus is blackened, while A. calami lacks a clypeus. Ascospores are also smaller in A. epigeni (12-16 × 4-4.5 µm in A. epigeni vs 14-17.5 × 4.5-7 µm in A. calami vs 16.3-22.5 × 4.5-6.4(-7.2) µm in A. licualae). Arecomyces tetrasporus has 4-spored asci as compared to 8-spored asci in A. calami and A. licualae.

Nigramammilla K.D. Hyde & J. Fröhl., gen. nov.

Ascomata erumpentes vel superficialia, mammiformes, nigra, ostiolata, papillata, paraphysaticum. Asci 8-spori, cylindrici, breve pedicellati, unitunicati, apparatus subapicale. Ascosporae 3-4-seriatae, cylindricae-fusiformis, 3-septatae, hyalinae, laevis vel curvae, appendiculatae.

Etymology: *Nigromammilla* from *nigra* = black, in reference to the black ascomata, plus *mamilla* in reference to the ascomata shape.

Ascomata erumpent to superficial, mammiform, black, with a central, papillate ostiole, scattered. *Peridium* comprising a few layers of brown-walled compressed cells. *Paraphyses* 1.5-2 µm wide, sparse, septate, tapering slightly distally. *Asci* 8-spored, cylindrical, short pedicellate, unitunicate, with an inconspicuous refractive subapical apparatus. *Ascospores* 3-4-seriate, cylindrical-fusiform, 3-septate, hyaline, straight or very slightly curved, tapering gently at both apices with short mucilaginous appendages.

Nigramammilla calami K.D. Hyde & J. Fröhl., sp. nov. (Figs 17-23) Ascomata 113-205 μm alta × 158-238 μm diam., erumpentes vel superficialia, mammiformes, nigra, ostiolata, papillata, paraphysaticum. Asci 78-96 × 8.5-12 μm, 8-spori, cylindrici, breve pedicellati, unitunicati, apparatus apicale, ca 0.5 μm alta, 2 μm diam. Ascosporae 35-41.5 × 3.5-4.5 μm, 3-4-seriatae, cylindricae-fusiformis, 3-septatae, hyalinae, laevis vel curvae, appendiculatae.

Etymology: In reference to the host.

Ascomata 113-205 µm high × 158-238 µm diam., erumpent to superficial, mammiform, black, with a central papillate ostiole 25-40 µm in diameter, scattered (Fig. 17). *Peridium* comprising a few layers of brown-walled compressed cells. *Paraphyses* 1.5-2 µm wide, sparse, septate, tapering slightly distally (Fig. 18). Asci 78-96 × 8.5-12 µm, 8-spored, cylindrical, short-pedicellate, unitunicate, with an inconspicuous refractive apical apparatus, *ca* 0.5 µm high × 2 µm diam. (Figs 19, 20). Ascospores 35-41.5 × 3.5-4.5 µm, 3-4-seriate, cylindrical-fusiform, 3-septate, hyaline, straight or very slightly curved, tapering gently at both apices, with short (2.5-4.5 µm long) mucilaginous appendages (Figs 21-23).

Material examined: HONG KONG, New Territories, Fan Ling, Wo Hop Shek Stream, on dead rattan sheath of *Daemonorops margaritae*, August 1995, J. Fröhlich (HKU(M)JF 863, holotype designated here); ECUADOR, Oriente, *Reserva de Produccion Faunistica Cuyabeno* (Cuyabeno Reserve), Rio Cuyabeno, forest near the Laguna Grande, Canangucho, Path A, on dead petiole of *Mauritia flexuosa*, August 1993, K.D. Hyde (HKU(M)JF 212).

Host substrate: Daemonorops margaritae, Mauritia flexuosa.



Figs 17-23. *Nigramammilla calami* (from holotype). 17: Ascomata. 18: Paraphyses. 19: Asci. 20: Ascus tip illustrating apical ring. 21-23: Ascospores. Note the polar appendages (arrowed in 22). (Scale bars: $17 = 100 \ \mu m$; 18-23 = 10 μm).

Known distribution: Ecuador, Hong Kong.

Remarks: Nigramammilla is most typical of the *Lasiosphaeriaceae*, but differs as the ascoma are mammiform and thin-walled. The species was originally thought to be a species of *Lasiosphaeria*, but asci are atypical in having short pedicels and a rather distinctive apical ring (Figs 19, 20).

Given their geographic distance, these two collections are remarkably similar in morphology and size. Their hosts are in the same subfamily of palms.

Pseudohalonectria taylorii K.D. Hyde & J. Fröhl., sp. nov. (Figs 24-34)

Ascomata 292-552 × 192-328 μ m, ellipsoidea, immersa, gregaria. Collum 720-1024 × 72-90 μ m. Asci 60-76 × 6-8 μ m, 8-spori, cylindrici, pedicellati, apparatus apicale ca. 2 × 2.5-4 μ m praediti. Ascosporae 40-61 × 2-3 μ m, hyalinae, filiformes, septatae.

Etymology: In honour of J.E. Taylor for her work on palm fungi.

Ascomata 292-552 × 192-328 µm, ellipsoidal, completely immersed with long, erumpent, brown neck, texture soft, gregarious (Fig. 24). Neck 720-1024 × 72-90 µm, central, cylindrical, periphysate with uniform width. *Peridium* 10-36 µm, even throughout the ascoma with thickening at the base of the neck, 4-layered: outer layer pale brown, surrounding the ascomata and neck, thicker around the ascomata, *textura epidermoidea*; second layer thinner at neck, *textura porrecta* 12-46 µm, third layer brown, comprised of 4-5 layers of compressed globular, brown walled cells; inner layer hyaline, comprised of 1-2 layers of compressed cells. At neck region, 2-layered, outer layer brown, comprised of 8-10 layers of dark walNigramammilla calami and Arecomyces calami, A. licualae and Pseudohalonectria palmae 19



Figs 24-34. *Pseudohalonectria taylorii* (from holotype).24: Section of ascoma.25-27: Sections of ascoma. Note the four wall layers of the peridium.28: Ascus tip illustrating apical ring typical of *Pseudohalonectria*.29: Paraphyses.30, 31: Asci.32-34. Ascospores. (Scale bars: $24 = 100 \mu m$; 25-34 = $10 \mu m$).

led, parallel, compressed cells; inner layer hyaline, comprised of 3-4 layers of parallel, compressed cells (Figs 25-27). *Paraphyses* hyaline, septate, broad at the apex, tapering towards the base, numerous, forming clumps, embedded in a gelatinous matrix (Fig. 29). *Asci* 60-76 × 6-8 µm (($\bar{x} = 67.4 \times 6.9 \mu$ m, n = 30), 8-spored, cylindrical, slightly curved to sigmoid, apex rounded, base conical, apedicellate, apical ring cylindrical, J-, ca.2 × 2.5-4 µm (Figs 28, 30, 31). *Ascospores* 40-61 × 2-3 µm (($\bar{x} = 50 \times 2.3 \mu$ m, n = 30), fasciculate, hyaline, straight to falcate, filiform, septate, tapering towards the ends, frequently released by deliquescence of the basal half of ascus (Figs 32-34).

Material examined: ECUADOR, Cuyabeno, Rio Cuyabeno: Botanists' plot", on Jessenia sp., August 1993, K.D. Hyde, E15 (HKU(M) 2642, holotype designated here).

Remarks: This new taxon is typical of *Pseudohalonectria* in having ascomata with a 4-layered coloured peridium, apedicellate cylindrical asci and filiform ascospores. Two species of *Pseudohalonectria* are known from palms and both differ in the size of their ascospores (Hyde et al., 1999). In P. eubenangeensis K.D. Hyde, Joa. E. Taylor & J. Fröhl. ascospores are longer (70-98 µm), while in P. palmicola K.D. Hyde, Joa, E. Taylor & J. Fröhl, they are wider (4.4.5 um). All aquatic species of *Pseudohalonectria* have narrower ascospores (Shearer, 1989).

Acknowledgements. Helen Leung is thanked for technical assistance. J. Taylor is thanked for her comments on the manuscript.

REFERENCES

FRÖHLICH J. & HYDE K.D., 2000 – Palm Microfungi. Fungal Diversity Research Series 3, 1-393.

- HYDE K.D. 1996 Fungi from palms. XXXII. Arecomyces gen. nov., with seven new species. Sydowia 48: 224-240.
- K.D., TAYLOR J.E. & FRÖHLICH J., 1999 Two new species of HYDE Pseudohalonectria from palms. Mycologia 91: 520-524.

HYDE K.D., TAYLOR J.E. & FRÖHLICH J., 2000 – Genera of Ascomycetes from Palms. Fungal Diversity Research Series 2, 1-247.

SHEARER C.A. 1989. - Pseudohalonectria (Lasiosphaeriaceae) an antagonistic genus from wood in freshwater. Can. J. Bot.67: 1944-1955.

SIVANESAN A. & SHIVAS R.G., 2002. - New species from each of the pyrenomycete genera Hyponectria, Physalospora and Trichosphaeria from Queensland, Australia. Fung. Divers.9: 169-174.

WANG Y.Z. & HYDE K.D., 1999. - Hyponectria buxi with notes on the Hyponectriaceae. Fung. Divers.3: 159-172.

YANNA HO W.H., HYDE K.D. & GOH T.K., 2001. - Occurrence of fungi on tissues of Livistona chinensis. Fung. Divers.6: 167-180.

YANNA, HO W.H. & HYDE K.D., 2002. - Fungal succession on fronds of Phoenix hanceana in Hong Kong. Fung. Divers.10: 185-211.