

***Pseudohalonectria miscanthicola* sp. nov.
and three interesting fungi from tropics**

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ABSTRACT – *Pseudohalonectria miscanthicola* sp. nov. is reported from senescent culms of *Miscanthus floridulus* (Gramineae) from Lung Fu Shan, Hong Kong. This fungus differs from other *Pseudohalonectria* species in having smaller ascospores which are 0-1-septate. It is described, illustrated and compared with other species in the genus *Saccardoella aquatica*, first described on submerged wood in Hong Kong and South Africa, is reported here for the first time from dead terrestrial wood at Mount Nicholson, Hong Kong. We also report *Massariothea themedae* from senescent culms of an unidentified grass from Kudremukh National Park, Karnataka, India. This species is described and illustrated. Important morphological characters of 8 known species of *Massariothea* are tabulated and a key to all the species is provided. *Oxydothis oraniopsis* is reported on senescent fronds of *Calamus* sp. collected from Chorle Ghat, Goa, India. This is the first record of ascomycetous genus *Oxydothis* from India.

Ascomycetes / biodiversity / coelomycete / graminicolous / new species / saprobes / taxonomy / wood

INTRODUCTION

The magnitude of fungal diversity has been estimated at 1.5 M species (Hawksworth, 1991, 2001). The highest proportions of new fungal species are found either in the tropics or in poorly investigated ecological niches (e.g. Das *et al.*, 2004; Desjardin *et al.*, 2003; Vánky, 2004). Hong Kong has a rich fungal diversity (Hyde, 2003). Lu *et al.* (2000) published a checklist of 2125 fungi reported from Hong Kong and there has been a rapid increase in the number of fungi reported from Hong Kong since 2000 (e.g. Wong & Hyde, 2001a, b; Jeewon *et al.*, 2003; Tang *et al.*, 2003a, b; Taylor & Hyde, 2003; Zhou *et al.*, 2003; Cai *et al.*, 2004; Kodsueb *et al.*, 2004; Shenoy *et al.*, 2004; Tsui & Hyde, 2004). During our study on graminicolous fungi, a new species of *Pseudohalonectria* Minoura & T. Muroi was observed on senescent culms of *Miscanthus floridulus* L. (Gramineae) from Lung Fu Shan, Hong Kong. *Pseudohalonectria miscanthicola* sp. nov. is described, illustrated and compared with similar taxa. *Saccardoella aquatica* K.M. Tsui,

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K.D. Hyde, Hodgkiss & Goh is reported on senescent wood collected from Mount Nicholson, Hong Kong. This fungus was first described by Tsui *et al.* (1998) on submerged wood in streams in Hong Kong and South Africa. This is the first report of the fungus from a terrestrial habitat.

The highest number of unique fungal species has been recorded from India (Hawksworth, 2001). Here two interesting fungi are reported from India. *Massariothea themedae* Syd. is reported from senescent culms of an unidentified grass collected at Kudremukh National Park, Karnataka. Important morphological characters of 8 known species of *Massariothea* are tabulated and a key to all species is provided. *Oxydothis oraniopsis* J. Fröhl. & K.D. Hyde is also described from senescent fronds of *Calamus* sp. collected at Chorle Ghat, Goa, India. This is the first record of the genus from India.

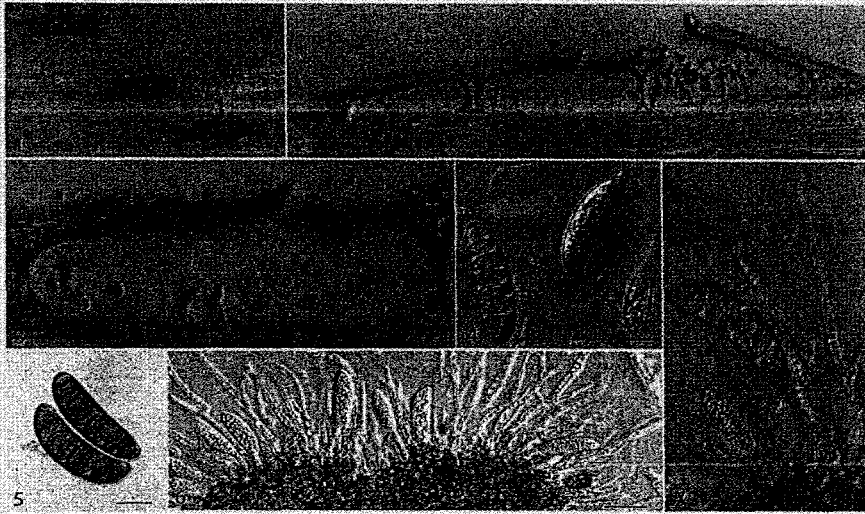
MATERIALS AND METHODS

Senescent culms of *Miscanthus floridulus* L. (Site: Lung Fu Shan, Hong Kong), senescent wood pieces (Site: Mount Nicholson, Hong Kong), senescent culms of an unidentified grass (Site: Kudremukh National Park, Karnataka, India) and senescent fronds of *Calamus* sp. (Site: Chorle Ghat, Goa, India) were collected. The collected specimens were placed into plastic "zip-locked" bags and returned to the laboratory. Then the samples were incubated at room temperature (25°C) individually with moist tissue to allow fungal fruiting bodies to develop. They were examined under a stereo-microscope with a cool light source (to prevent dehydration). The fruiting bodies were cut open with a sharp razor blade and their contents were scooped out and placed in a drop of sterile water on a microscope slide using fine-tip forceps. Melzer's reagent was used to stain the ascus apical structures. India ink was added to visualize the shape of the mucilaginous sheath. The specimens were numbered and deposited in the herbarium of the Centre for Research in Fungal Diversity, the University of Hong Kong (HKU (M)). Attempts were made to grow single-spore isolates of the fungus in culture (Choi, 1999) (water agar; potato dextrose agar), but each time they proved unsuccessful.

TAXONOMY

Massariothea themedae Syd. (Figs 1-7)

Conidiomata eustromatic, solitary, sparse, dark brown to black, immersed, globose to collabent, in section 250-590 μm diam \times 50-100 high, multilocular, often with separate ostioles, peridermal, the upper wall composed of dark brown thick-walled *textura angularis* and the lower thinner wall of paler cells, beak conical to cylindrical. *Paraphyses* branched, septate, hyaline, filiform. *Conidiogenous cells* discrete, monoblastic, lageniform to cylindrical, hyaline, smooth, with a wide channel and moderate periclinal thickening, 18-19 \times 2.5-2.6 μm , formed from the inner cells of locular walls. *Conidia* (30-)32-46 \times (11-)12-15(-16) μm (\bar{x} = 39.8 \times 13.3 μm , n = 60), (3-)4-7-distoseptate, each cell with a conspicuously reduced lumen, brown,



Figs 1-7. *Massariothea themedae* 1. Conidiomata on the host surface with conidia accumulated (arrowed). 2. Vertical section of a pycnidium showing three locules. 3. Vertical section a pycnidium showing an ostiole. 4, 5. Distoseptate conidia. 6, 7. Squash mount of contents of a conidioma, showing conidiogenesis (arrowed). Scale bars: 1 = 200 μm ; 2, 3 = 50 μm ; 4-7 = 20 μm .

clavate to fusoid-clavate, straight or moderately curved, outer wall thickened and darkened at the apex and base, periclinal wall paler and thinner in transverse band immediately below the thickened apex and the base (Fig. 5), truncate and darker at the base.

Specimens examined: India, Kudremukh National Park, Karnataka, on senescent culms of an unidentified grass, June 2004, B.D. Shenoy, HKU(M) 17490, *ibid.* HKU (M) 17491.

Oxydothis oraniopsis J. Fröhl. & K.D. Hyde. (Figs 17-20)

Ascomata forming under slightly raised dome-shaped regions on the host surface, solitary or in small groups; papilla at one end, curving upwards and piercing the host cuticle. Asci 160-190 \times 16 μm , long cylindrical, ring subapical, J+, 2 μm high, 3 μm diam. Ascospores (48-)56-62 \times 7-8 μm , fusiform, 1-septate, hyaline, gradually tapering from the central septum to long narrow processes.

Material examined: India, Chorle Ghat, Goa, on senescent culms of *Calamus* sp. June 2004, B.D. Shenoy, HKU(M) 17492.

Pseudohalonectria miscanthicola B.D. Shenoy, R. Jeewon & K.D. Hyde, sp. nov. (Figs 8-16)

Ascomata immersa vel erumpentia, globosa vel subglobosa, solitaria vel gregaria, paraphysata. Collum 1.4-2.2 mm longum, 0.1-0.4 mm diam. Asci 76-94 \times 10-16 μm , 8-spori, cylindricacei vel clavati, apparatus apicalis 3-4 μm diam., 3 μm



Figs 8-16. *Pseudohalonectria miscanthicola* (from holotype). 8. Ascoma partially immersed in the host surface. 9. Apical portion of neck showing deposition of ascospores/asci (arrowed). 10. Section of an ascoma. 11. Vertical section of neck. 12. Paraphyses. 13, 14. Ascospores. 15. An empty ascus showing an apical refractive ring (arrowed). 16. A non-amyloid (refractive) apical ring. Figs. 17-20. *Oxydothis oraniopsis*. 17, 18. Surface view of fruiting structures on host (arrowed). 19. Squash mount of contents of an ascoma. 20. An ascospore showing long narrow processes.

Scale bars: 8 = 500 μ m; 9 = 800 μ m; 10, 11 = 400 μ m; 12 = 30 μ m; 13-15 = 40 μ m; 16 = 70 μ m; 17 = 100 μ m; 18 = 80 μ m; 19 = 200 μ m; 20 = 30 μ m.

altus. Ascospores (20-)22-33 \times (5-)6-8 μ m, biseriate, ellipsoideae, 0-1-euseptate, hyalinae.

Etymology: Latin – living on the grass genus *Miscanthus*.

Ascomata immersed to partly immersed, erumpent, yellow to orange, globose to subglobose, solitary to gregarious. Neck cylindrical, periphysate, yellowish to orange, 1.4-2.3 mm long, 0.1-0.12 mm diam at the tip, 0.3-0.4 mm diam at the base (Figs 8, 9). Paraphyses numerous, up to 140-180 μ m long, 8-10 μ m wide at the base, 4-5 μ m wide at the apex, filamentous, tapering distally, septate, hyaline, unbranched (Fig. 12). Asci 76-94 \times 10-16 μ m (\bar{x} = 85 \times 13 μ m, n = 35), 8-spored, cylindrical to clavate, with a refractive, thimble-shaped, non-amyloid apical apparatus, 3 μ m diam, 3-4 μ m high. Ascospores (20-)22-33 \times (5-)6-8 μ m (\bar{x} = 29 \times 6.8 μ m, n = 37), biseriate, overlapping, ellipsoidal, 0-1-septate, hyaline, apex rounded, surrounded by a mucilaginous sheath.

Holotype: Hong Kong, Lung Fu Shan, on senescent culms of *Miscanthus floridulus*, Aug. 2004, B.D. Shenoy, HKU(M) 17487.

Paratypes: From the same origin: HKU(M) 17488 and HKU(M) 17489, PDD 78748.

***Saccardoella aquatica* K.M. Tsui, K.D. Hyde, Hodgkiss & Goh**

Ascomata up to 1 mm in diam, perithecioid, globose to subglobose, immersed to erumpent, black, gregarious, ostiolate, neck black. Pseudoparaphyses numerous, 2-4- μ m wide. Asci 170-208 \times 8 μ m (\bar{x} = 188.6 \times 8 μ m, n = 10), 8-spored, long-cylindrical, short-pedicellate, with a faint ring-like apical apparatus. Ascospores 20-30 \times 8 μ m (\bar{x} = 27.8 \times 8 μ m, n = 25), overlapping uniseriate, fusiform, apices rounded, hyaline, 3-septate, slightly constricted at the septa, surrounded by inconspicuous mucilaginous sheath.

Specimen examined: HONG KONG, Mount Nicholson, on a senescent wood, Aug. 2004, B.D. Shenoy, HKU(M) 17497.

DISCUSSION

Massariothea themedae

Massariothea Syd. was introduced by Sydow (1939) to accommodate the type species, *M. themedae* Syd. possessing stromatic pycnidia and brown-coloured phragmoconidia. There are 8 known species of *Massariothea* (Table 1) and a key to the species was provided by Alcorn (1993). He considered *Massariothea themedae* to have unilocular pycnidia whereas Subramanian & Muthumary (1979), who studied the type specimens, described the same to have pycnidia with one to three locules in section i.e., multilocular. In our collection, the fungus, in section had multilocular (1-3 locules) pycnidia and 4-7-distoseptate conidia. Therefore a new key to the 8 species is given below.

Key to all known *Massariothea* species (Modified from Alcorn, 1993)

1. Conidia < 20 μ m long *M. scotica*
1. Conidia > 20 μ m long 2
2. Conidia commonly 3-septate, septa placed asymmetrically *M. triseptata*
2. Conidia 5-septate *M. botulispora*
2. Conidia up to 7 or more septate 3
3. Conidiomata uni- or multilocular 4
3. Conidiomata multilocular, up to 650 μ m diam. *M. shawiae*
3. Conidiomata unilocular 5
4. Conidia (7.5-)9.5-11 μ m wide, 6-8 distoseptate *M. attenuata*
4. Conidia 10-16 μ m wide, (3-)4-7 distoseptate *M. themedae*
5. Conidiomata up to 200 μ m diam., Conidia 48-69 \times 12.5 μ m *M. paspali*
5. Conidiomata up to 600 μ m diam, Conidia 27-68 \times 7.5-14 μ m *M. similis*

Massariothea is similar to *Paramassariothea* Subramanian & Muthumary (1979). The latter differs in having conidia which are hyaline, straight to falcate with a truncate base and tapering ends, and the length/width ratio of 10:1 with numerous septa (Subramanian & Muthumary, 1979).

Oxydothis oraniopsis

There are 67 species of *Oxydothis* (Shenoy *et al.*, 2004). *Oxydothis oraniopsis* was first described by Fröhlich & Hyde (1994) on a living leaf of *Oraniopsis*

Table 1. Synopsis of the genus *Massariothea*; all the values are in μm .

Species	Conidiomata: size (diam. \times high)	Conidiomata: Uni/multilocular	Conidiogenous cells (l \times w)	Conidia (l \times w)	No. of distosepta	Host	Distribution	Reference
<i>M. attenuata</i> B. Sutton & Alcorn	upto 5 diam.	Uni or multilocular	9-16 (-27) \times 2.5-4.5	(25-) 40-53 (-63) \times (7.5-) 9.5-11	6-8	Leaves of <i>Eragrostis interrupta</i>	Australia	Sutton & Alcorn, 1985
<i>M. botulispora</i> (Teng) B. Sutton	300-450 diam.	?	9-19 (-27) \times 2.5-4.5 (‘conidiophores’)	40-52 \times 12-14	5	Leaves of <i>Phragmites communis</i>	P. R. China	Sutton, 1980
<i>M. paspali</i> (Ellis & Everh.) B. Sutton	upto 200 diam.	Unilocular	6-10 \times 5-10	48-69 \times 12.5	6-7	Culms of <i>Paspalum platycaule</i>	USA	Sutton, 1975
<i>M. scotica</i> B. Sutton & Rizwi	upto 165 \times 90	?	4.5-10 \times 2-3.5	15-19.5 \times 5.5-6.5	3	Dead barks of <i>Quercus</i> sp.	UK	Sutton & Rizwi, 1980
<i>M. shawiae</i> (B. Sutton) B. Sutton	upto 650 diam.	Multilocular	upto 50 \times 2-3	31-52 \times 7.5-8.5	3-7	<i>Zea mays</i>	India	Sutton, 1978
						<i>Sorghum vulgare</i>	Papua New Guinea	Sutton, 1975
<i>M. similis</i> Alcorn	525-1000 \times 290-600	Unilocular	12-21 \times 3-5	27-68 \times 7.5-14	3-11	Leaves of <i>Heteropogon triticeus</i>	Australia	Alcorn, 1993
<i>M. themedae</i> Syd.	250-590 \times 50-100	Multilocular	18-19 \times 2.5-2.6	(30-) 32-46 \times (11-) 12-15 (-16)	(3-) 4-7	Culms of unidentified grass	India	This paper
	up to 600 diam.	Unilocular	6-13 \times 4-5	31.5-44 \times 10.5-12	6-7	Culms of <i>Sorghum graminicola</i> , <i>S. vulgare</i> , <i>Themeda</i> sp.	India	Sutton, 1978
	160-250 diam.	Unilocular	5 \times 2-4 (‘conidiophores’)	32-55 \times 10-16.5	5-7	Leaves of <i>Themeda gigantea</i>	Philippines	Sydow, 1939
						Leaves of <i>Sorghum plumosum</i> , <i>Neyraudia</i> sp., <i>Themeda</i> sp.	Hong Kong, India	Sutton, 1980
	300-350 \times 250-300 (in unilocular pycnidia)							
	200-350 \times 250-300 (in multilocular pycnidia)	Uni to multilocular	5-15 \times 4-8	40-50 \times 14-16	4-7	Leaves of <i>Themeda gigantea</i>	Philippines	Subramanian & Muthumary, 1979
<i>M. triseptata</i> Alcorn	500-950 \times 370-550	Unilocular	10-28 \times 3.5-5 (-8 at the base)	25-38 \times 10-12.5	3	Leaves of <i>Sorghum plumosum</i>	Australia	Alcorn, 1993

appendiculata from north Queensland, Australia. This fungus has also been recorded on a living leaf of *Laccospadix australasicus* and on dead petioles of *Licuala ramsayi* from Australia (Fröhlich & Hyde, 1994; Fröhlich & Hyde, 2000). We observed this fungus on senescent fronds of *Calamus* sp. collected from Chorle Ghat, Goa, India. This is the first record of genus *Oxydothis* from India.

Pseudohalonectria miscanthicola

Pseudohalonectria was described by Minoura and Muroi (1978) on wood submerged in a Japanese lake. The genus is characterised by bright yellow to light orange perithecia with protruding, cylindrical, periphysate necks; unitunicate, cylindrical to clavate asci with a J- cylindrical apical apparatus; tapering paraphyses; and smooth, hyaline to slightly pale, cylindrical to filiform ascospores. *Pseudohalonectria* is most similar to *Ophioceras* and they are compared based on morphological characters by Hyde *et al.* (2000) and Promptutha *et al.* (2004).

There are 11 accepted species of *Pseudohalonectria* (Table 2). *Pseudohalonectria miscanthicola* should be compared to *P. fuxianii* L. Cai, K.M. Tsui, K.Q. Zhang & K.D. Hyde as both have ellipsoidal ascospores. *Pseudohalonectria miscanthicola* differs from *P. fuxianii* in having narrower asci (10-16 μm vs. 17.5-30 μm), shorter ascospores ((20-)22-33 vs. 30-52.5) and fewer ascospore septa (0-1 vs. 3-5) (Cai *et al.*, 2002b). *Pseudohalonectria miscanthicola* is similar to *P. adversaria* Shearer in ascospore shape but differs in having shorter asci (76-94 μm vs. 120-150 μm) and, ascospores ((20-)22-33 μm vs. 33.5-49 μm) and fewer ascospore septa (0-1 vs. 5-7) (Shearer, 1989).

Shearer *et al.* (1999) noted that species of *Pseudohalonectria* and *Ophioceras* were morphologically similar to species of Magnaporthaceae and temporarily transferred these two genera to that family. Inderbitzin *et al.* (2001) investigated the phylogenetic relationships based on SSU rDNA of *Pseudohalonectria lignicola* and *Ophioceras leptosporum* and found that both were closer to the Magnaporthaceae and did not group with *Sordaria fimicola*. Kirk *et al.* (2001) placed these two genera in Magnaporthaceae.

Saccardoella aquatica

The genus *Saccardoella* was described by Spegazzini (1879). There are 16 accepted species (Cai *et al.*, 2002a). The characteristics of this genus include large, immersed to erumpent ascomata, long cylindrical asci with a faint ring-like apical apparatus and uniseriate ascospores having several transverse septa and sheaths or appendages (Petraik, 1962; Hyde, 1992; Barr, 1994). *Saccardoella* species have been reported from freshwater, marine, and terrestrial habitats (Cai *et al.*, 2002a). *Saccardoella aquatica* was first described by Tsui *et al.* (1998). They reported this fungus on wood submerged in streams in Hong Kong and South Africa. This species is distinct from other in *Saccardoella* in having fusiform, 3-septate ascospores. We observed this fungus on a decaying wood from a terrestrial habitat. This is the first record of *Saccardoella* from dual habitats (i.e. fresh water and terrestrial).

Acknowledgements : This research was funded by the university of Hong Kong (CRCG 10205773). B.D.S, D.V. and C.L. thank The University of Hong Kong for the award of a Postgraduate studentship. Subramanya Rao (Department of Microbiology, F.M. Kariappa, College, Madikeri, India), Sanil Deepak (Department of Applied Zoology, Mangalore University, India), Krishna (Department of Commerce, Vijaya College, Mulki, India) are thanked for their assistance and guidance during sample collection in Kudremukh National Park, Kudremukh, India. Suraksha, G. Puja and J. Pratibha (Goa

Table 2. Synopsis of known *Pseudohalonectria* spp.

<i>Taxa</i>	<i>Ascomata colour when young</i>	<i>Ascomata neck ((l × d μm)</i>	<i>Asci (l × w μm)</i>	<i>Apical ring (h × d μm)</i>	<i>Ascospore shape</i>	<i>Ascospores (l × w μm)</i>	<i>Ascospore septation</i>	<i>Reference</i>
<i>P. adversaria</i>	Orange	79-248 × 109-158	120-150 × 13-20	2.5-5 × 2.5-3	Ellipsoidal	33.5-49 × 4.5-7	5-7	Shearer, 1989
<i>P. aomoriensis</i>	Yellow to yellowish-brown	900-1400 × 160-180	112-145 × 8-11	not given	Cylindrical	49-57 × 3-4.5	5-7	Ono & Kobayashi, 2001
<i>P. eubenangeensis</i>	Yellow	135-175 × 90-125	80-120 × 8-11.5	2-2.5 × 1.5-2	Filiform or Cylindrical	70-80 × 2.5-3.5	3-5 (-7)	Hyde <i>et al.</i> , 1999
<i>P. falcata</i>	Light yellow or light brown	81-162 × 81-108	106-244 × 14.4-21.6	not given	Falcate	97-166 × 4.2-7.2	6-16	Shearer, 1989
<i>P. fuxianii</i>	Orange-brown	430-570 × 70-110	90-187.5 × 17.5-30	2-2.5 × 2.5-3	Ellipsoidal	30-52.5 × 7.5	3-5	Cai <i>et al.</i> , 2002b
<i>P. figinicola</i>	Pale Yellow	170-621	90-132 × 11-17.5	not given	Cylindrical	38.4-74.8 × 3.5-6.5	5-11	Shearer, 1989
<i>P. longirostrum</i>	Bright yellow	1683-3712 × 118-168	94-130 × 8.5-12	3-5 × 2	Filiform	84-105.5 × 3.8-4.0	4-8	Shearer, 1989
<i>P. lutea</i>	Yellowish brown	300-600 × 160-200	122-192 × 14.4-18	not given	Cylindrical	99-168 × 4.8-8.4	5	Shearer, 1989
<i>P. palmicola</i>	Dark brown	1600 × 132-152	120-156 × 13-15	3.5-4 × 2.5-4	Filiform	74-83 × 4-4.5	3-6	Hyde <i>et al.</i> , 1999
<i>P. phialidica</i>	Yellow	614-1940 × 89-129	82-99 × 5-7.9	3.2-5 × 2-3.5	Filiform	64.5-79 × 2	0-4	Shearer, 1989
<i>P. miscanthicola</i>	Yellow to Orange	1400-2200 × 100-400	76-94 × 10-16	3 × 4	Ellipsoidal	(20-)22-33 × (5-)6-8	0-1	This paper
<i>P. suthepensis</i>	Dark brown	400-800 × 120-180	135-170 × 5-8.5	2.5 × 2-3	Filiform	85-137 × 2.5	4-7	Promptutha <i>et al.</i> , 2004

University, Goa, India) are thanked for their assistance and guidance during sample collection in Chorle Ghat, Goa, India. W.H. Ho, E.H.C. McKenzie are thanked for stimulating discussion on *Massariothea*. I. Promputtha, L.Y.M. Helen, S. Thongkantha, K. Rampai, (Chiang Mai University, Thailand) are thanked for technical and photographic assistance.

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