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Biodiversity of Intertidal Estuarine Fungi on *Phragmites* at Mai Po Marshes, Hong Kong

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Intertidal decaying stems and leaf sheaths of *Phragmites australis* were randomly collected and their mycota examined. Sixty one species of fungi were associated with the decaying stems and leaf sheaths, including *Antiptodera* spp., *Ligníncola laevis*, *Phomatospora phragmiticola* and *Zopfiella latipes*. The following new species are described, *Halosarpheia phragmiticola*, *Massarina phragmiticola*, *Phomatospora phragmiticola* and *Cytoplacosphaeria phragmiticola*. The fungal communities associated with decaying *Phragmites australis* permanently submerged in the gei wai (tidal shrimp farms) differ from those in the intertidal region. The diversity of these fungi are discussed in relation to the biodiversity of fungi in mangrove communities in Hong Kong and with those fungi of other salt marsh communities.

Introduction

Temperate coastal wetlands are often dominated by salt marsh grasses, such as species of *Spartina* and *Juncus*. Most research on the associated mycota of these temperate plants in based on *Spartina* (Johnson and Sparrow 1961, Meyers *et al.* 1970, Meyers 1974, Gessner 1976, 1977, Gessner and Kohlmeyer 1976, Kohlmeyer and Kohlmeyer 1979). There is also some information on the fungi associated with *Juncus roemerianus* Scheele (Kohlmeyer and Volkmann-Kohlmeyer 1993 a, b, 1995, Volkmann-Kohlmeyer and Kohlmeyer 1993, 1994, Kohlmeyer *et al.* 1995 a, b, c, 1966).

Mangrove vegetation is the tropical counterpart of temperate tidal salt marshes (Kohlmeyer and Kohlmeyer 1979). There is a large body of information on mangrove fungi, with most data on biodiversity (Hyde and Jones 1988, Hyde *and al.* 1986, 1992, 1993, Hyde 1992, Hyde and Nakagiri 1992, Jones and Agerer 1992, Sadaba *et al.* 1995).

In subtropical regions, salt marsh grasses such as *Phragmites australis* (Cav.) Trin. *ex* Steud [also known as *Phragmites communis* (L.) Trin.] coexist with the mangrove tree species. The tidal shrimp ponds (gei wai) of Mai Po were originally excavated from native mangrove habitats with communities of *Kandelia candel* (L.) Druce, *Avicennia marina* (Forsk.) Vierh. and *Aegiceras corniculatum* (L.) Blanco. Traditional gei wais at Mai Po are characterised by repeated draining and flooding and details of operational methods can be found in Macintosh (1983). Mangroves (mostly *Kandelia candel*) were originally the chief primary producers in Mai Po Marshes. However, due to progressive change in the operation of the gei wais from traditional shrimp catching to fish culture, this has resulted in some gei wais (e. g. gei wai no. 12) being kept undrained for several months. Semi-permanent flooding had resulted in death of mangroves which are subsequently replaced by *Phragmites australis*, which colonises the banks and edges of gei wais and elevated mud platforms. *Phragmites australis* has now acquired co-dominance in the gei wais with *Kandelia candel*, and is one of the main primary producers (Lee 1990). It was estimated that about 40% of open or elevated mud platforms in the gei wais were covered with *P. australis* in early 1988 (Lee 1990).

Fungi have long been recognised as one of the major decomposers of salt marsh plants, beside bacteria (e. g. Meyers *et al.* 1970, Gessner 1977, Newell 1993, 1996). Recent research on the dynamics of microorganisms and the change in the chemical composition of decomposing *Phragmites australis* in Japan, have shown that fungi are the main decomposers of decaying leaves before and after submergence in seawater, while bacteria were only dominant decomposers within the short period following submergence (Tanaka 1991). Tanaka (1991) suggested that the decrease in the fungal populations during the short time following submergence can be attributed to the inability of terrestrial fungi to survive in the saline aquatic environment.

As crucial decomposers of a major primary producer, in a highly productive subtropical estuarine region, fungi warrant more attention to their biodiversity and ecology than has previously been given. Unfortunately, the mycota of *Phragmites australis*, is even less well investigated than either *Spartina alterniflora* Loisel. or *Juncus roemerianus*. Only two marine fungi, *Phaeosphaeria albopunctata* (West.) Shoemaker *et* C. E. Babc. and *Cirrenalia fusca* I. Schmidt, have been reported from *P. australis* by previous researchers (Kohlmeyer and Kohlmeyer 1979). *Massariosphaeria typhicola* (Karst.) Leucht. has been found on *P. australis* in freshwater locations in Finland (Karsten 1873). Several terrestrial mitosporic fungi have also been reported on leaves and stems and leaf sheaths of *P. australis* (e. g. Cunnell 1958, Sutton and Alcorn 1974). In this study an assessment of the biodiversity of the mycota found on decaying stems and leaf sheaths of *P. australis* at the Mai Po Marshes is made.

Materials and Methods

Study area

All sampling was conducted along the edge of a tidal shrimp pond ('gei wai', no. 12) and on both sides of the sluice gate of gei wai no. 12 outside the security fence, in Mai Po Marshes. Mai Po Marshes are situated along the northwestern coast of the New Territories facing the Pearl River estuary (22°29' N, 114°02' E).

Sampling site

Two sampling sites were chosen to initiate a comparison between the fungi found in two different habitats.

Outside security fence: *Phragmites australis* is subjected to natural tidal inundation with the basal portion (ca. 30 cm) of the plant submerged regularly during high tides.

Along the edge of gei wai: The amount of time for the basal to middle portion of plant being inundated is regulated solely by the operation method of gei wai. During the period of this study, up to one half (ca. 70 cm) of the plant had been immersed for several months.

Collection of samples

In order to collect saprophytic fungi responsible for the decompositions of decaying *Phragmites australis*, dead stems and leaf sheaths were randomly collected from within one to two meters of the shore at the two sites. These were lying on the mangrove floor outside the security fence, or standing upright with the lower part submerged in the gei wai, and were naturally decomposing samples. One hundred and twenty five decaying stem and leaf sheath segments (ca. 15 cm) were collected from outside the security fence and 144 samples were collected from the gei wai, with ca. half of the samples collected in August and the other half in November 1995. Samples were placed in plastic bags, sealed and returned to the laboratory. Samples were then incubated on moist tissue paper, in clear plastic boxes (25 × 12 × 10 cm) at room remperature and normal lighting conditions, for what was found to be an optimum period of 1–3 weeks to induce sporulation of fungi. The fungi were identified by using a Leica MZ 12 dissecting microscope at 15–20 × magnification. Voucher slides and/or dried material of the fungi found were prepared and are held in the mycological herbarium of The University of Hong Kong [HKU(M)].

Analyses

Frequency of occurrence of fungi collected is expressed as the number of collections of a species at each site divided by the total number of samples examined from this site. Based on these figures, fungi collected are classified as 'very frequent' (> 20%), 'fequent' (10–20%) and 'infrequent' (< 10%) species, as adopted by Tan and Leong (1989). It was not feasible to identify all taxa to species level, as some genera lack modern treatments or up to date keys and may contain numerous species. However, each unique taxon is named (e. g. *Farrowia* sp. represents one species of *Farrowia*, *Phomopsis* sp. 1 and *Phomopsis* sp. 2 represent two species of *Phomopsis*. They are reported here in order to give an estimation of fungal diversity occurring on *Phragmites australis* at Mai Po Marshes.

Water temperatures at the time of collection was between 24–31 °C, while the salinity range outside security fence and along edge of gei wai no. 12 was 2.5–19‰, and 2–15‰ respectively. *In situ* ammonium concentration (NH_4^+) of water outside security fence and along edge of gei wai no. 12 was 15–20 ppm, and 22–26 ppm respectively.

Results

Sixty one taxa of higher fungi were found associated with decaying stems and leaf sheaths of *Phragmites australis* and the results are presented in Table I. Forty one taxa were collected from 125 decaying plant samples from outside security fence, and 47 taxa were found on 144 plant samples along the edge of the gei wai. The ratio of ascomycetes to coelomycetes to hyphomycetes was roughly equal (17 : 19 : 25). *Lignincola laevis* was overall the most common species (22.7%), while another very frequent taxon was *Colletotrichum* sp. (21.2%). Frequent taxa were *Phomopsis* sp. 1 (19.3%), *Aniptodera phragmiticola* (15.2%), *Fusarium* sp. (14.9%), *Cladosporium* sp. (13%), *Trichoderma* sp. (12.3%), *Cytoplea* sp. (11.9%) and *Rhinocladiella* sp. (11.2%), while other taxa were infrequent.

Outside the security fence, *Lignincola laevis* (37.6%) was the most common species. Other very frequent taxa were *Trichoderma* sp. (26.4%), *Antiptodera phragmiticola* (23.2%) and *Collectotrichum* sp. (22.4%). Along the edge of the gei wai, *Fusarium* sp. (28.8%), *Cladosporium* sp. (24.3%), *Phomopsis* sp. 1 (21.5%) and *Colletotrichum* sp. (20.1%) were very frequent.

Twenty seven taxa including *Colletotrichum* sp., *Phomopsis* sp. 1, *Rhinocladiella* sp., *Lignincola laevis* and *Aniptodera phragmiticola* were found common to both sites. The majority of taxa (34), however, occurred at a single site. Thirteen taxa were found only outside the security fence, such as *Trichoderma* sp., *Phomatospora marina* and *Phragmitensis marina*. Twenty taxa were also found only along the edge of the gei wai, such as *Fusarium* sp., *Cladosporium* sp. and *Macrophomina* sp.

Saprophytic fungi occurring on *Phragmites australis*

The lower parts of the standing decaying stems and leaf sheaths (up to 30 cm) of *Phragmites australis* found outside the security fence, are subjected to daily tidal inundation. Along the edge of the gei wai, up to one half of the standing decaying stems and leaf sheaths of *P. australis* (ca 70 cm) are immersed for several months. Fungi occurring on these lower plant parts can be considered to be intertidal fungi, while those occurring on the upper parts of the stems and leaf sheaths of *P. australis* are terrestrial. Middle portions of the plants are occasionally submerged or exposed to salt spray and hence colonised by mostly intertidal fungi.

Intertidal fungi. Differences in intertidal fungal composition between the two sites (outside the security fence and along the edge of the gei wai) were observed. Eleven intertidal fungi were confined to the decaying stems and leaf sheaths lying on the mangrove floor, collected outside the security fence. *Lignincola laevis* (37.6%) and *Aniptodera phragmiticola* (23.2%) were the most dominant intertidal species here, while *Gaeumannomyces* sp. was an infrequent ascomycete. Three coelomycetes (*Chaetasbolisia* sp., *Microsphaeropsis* sp., *Stauronema* sp.) and two hyphomycetes (*Pithomyces maydiscus* and *Spegazzinia tessarthra*) were infrequent.

Six hyphomycetes (*Acremonium* sp. 2, *Alternaria alternata*, *Arthrobotrys conoides*, *Dactylaria* sp., *Gliomastix* sp. 1, *Sarocladium* sp.) were confined to the plant samples collected at the water/air interface, along the side of the gei wai. The most common taxa being the hypohomycetes *Cladosporium* sp. (24.3%), *Sarocladium* sp. (8.3%) and *Arthrobotrys conoides* (4.9%). One coelomycete, *Dinemasporium strigosum*, and one ascomycete, *Massarina thalassiae* also occurred on these lower portions of plant samples.

Most intertidal fungi were found at both sites, although some exhibited a clear affinity towards a specific site. The ascomycetes *Aniptodera phragmiticola*, *Lignincola laevis* and *Massarina phragmiticola* were mostly found outside the security fence. The coelomycetes *Cytoplascosphaeria phragmiticola*, *Phoma* sp., *Phomopsis* sp. 1, *Sclerostagonospora* sp. and *Septoriella* sp.) were common to both sites, although *Cytoplacosphaeria phragmiticola* and *Septoriella* sp. were more common along the edge of the gei wai. The hyphomycetes *Arthrinium* state of *Apiospora* sp., *Drechslera hawaiiensis*, *Phaeoisaria* sp., *Rhinocladiella* sp., *Stachybotrys* sp., and *Tetraploa aristata* were also common to both sites, but *Tetraploa aristata* was more frequently recovered outside the security fence.

Terrestrial fungi. Sixteen terrestrial fungi were found associated with the decomposition of the apical to middle portion of decaying stems and leaf sheaths of *Phragmites australis*. Four of these were common to both sites. *Chaetomium globosum* was the only terrestrial ascomycete found on the upper levels of the plant samples. This species was common at both sites, but was more frequently recovered outside the security fence. *Cytoplea* sp. and the *Arthrinium* state of *Apiospora montagnei* were found on plant samples, both outside the security fence and along the edge of the gei wai.

Outside the security fence, *Trichoderma* sp. (26.4%) and *Chaetomium globosum* (6.4%) were the most frequently recorded terrestrial fungi. *Phomopsis* sp. 2, *Tetranacrium* sp., *Dendrostilbella* sp. and *Trichoderma* sp. were only collected here.

Along the edge of the gei wai, *Fusarium* sp. (28.8%) was the most commonly recorded taxon. Eight less frequent terrestrial deuteromycetes (*Cladosporium* sp. *Macrophomina* sp. *Neottiosporina* sp. *Pestalotiopsis* sp. *Stagonospora* sp. *Acremonium* sp. 1, *Gliomastrix* sp. 2 and *Paecilomyces* sp.) were limited to plant samples along the edge of the gei wai.

Descriptions of fungi

An account of selected saprophytic fungi found on decaying stems and leaf sheaths of *Phragmites australis* during this study is given in the following section.

Ascomyetes

Halosarpheia phragmiticola O. K. Poon *et* K. D. Hyde, sp. nov.
Figs 1–10

Etymology: In reference to *Phragmities australis* the host.

Diagnosis

Ascomata ca *350µm in diameter.*, ca *400 µm alta, globosa vel subglobosa, immersa vel superficialia, ostiolata, papillata, coriacea, nigra, solitaria.* Asci *105–167.5 × 32.5–47.5 µm, 8-spori, clavati, pedicellati, apparatu apicali praediti.* Ascosporae *25–35 × 7.7–10.5 µm, ellipsoido-fusiformes, 1-septatae, appendiculatae.*

Ascomata 350–400 µm in diameter, globose to subglobose, immersed to superficial, ostiolate, coriaceous, black, solitary, with a long black cylindrical neck (Fig. 1). *Neck* up to 700 µm long, 70 µm in diameter, cylindrical, superficially covered with short hyaline hairs, periphysate. *Peridium* up to 10 µm thick, two layered; outer layer comprising black thick-walled cells; inner layer comprising hyaline

Table I. Estuarine saprophytic fungi associated with decaying stems and leaf sheaths of *Phragmites australis* collected in Mai Po Marshes in August and November 1995.

Fungi	Outside fence		Along edge of gei wai		Total no. of collections	% occurrence
	No. of collections	% occurrence	No. of collections	% occurrence		
ASCOMYCETES (17 taxa)						
Aniptodera chesapeakensis Shearer *et* Miller	6	4.8	2	1.4	8	3
Aniptodera phragmiticola O. K. Poon *et* K. D. Hyde	29	23.2	12	8.3	41	15.2
Chaetonium globosum Kunze *ex* Steud.	8	6.4	1	0.7	9	3.3
Farrowia sp.	0	0	1	0.7	1	0.4
Gaeumannomyces sp.	1	0.8	0	0	1	0.4
Halosarpheia unicaudata (E. B. G. Jones *et* LeCampion–Alsumard) R. G. Johnson, E. B. G. Jones *et* S. T. Moss *ex* Kohlm. *et* Volkm.-Kohlm.	0	0	13	9	13	4.8
Leptosphaeria sp.	1	0.8	8	5.6	9	3.3
Lignincola laevis Höhnk	47	37.6	14	9.7	61	22.7
Massarina phragmiticola O. K. Poon *et* K. D. Hyde	11	8.8	1	0.7	12	4.4
Massarina thalassiae Kohlm. *et* Volkm.-Kohlm.	0	0	1	0.7	1	0.4
Nectria haematococca Berk. *et* Broome	2	1.6	0	0	2	0.7
Phomatospora phragmiticola O. K. Poon *et* K. D. Hyde	20	15.9	0	0	20	7.4
Phragmitensis marina M. Wong, O. K. Poon *et* K. D. Hyde	4	3.2	0	0	4	7.4
Pleospora spartinae (Webster *et* Lucas) Apinis *et* Chesters	1	0.8	12	8.3	13	4.8
Pseudohalonectria falcata Shearer	3	2.4	2	1.3	5	1.9
Verruculina enalia (Kohlm.) Kohlm. *et* Volkm.-Kohlm.	0	0	2	1.3	2	0.7
Zopfiella latipes (Lundqvist) Malloch *et* Cain	2	1.6	0	0	2	0.7
DEUTEROMYCETES (44 species)						
Coelomycetes (19 species)						
Chaetasbolisia sp.	1	0.8	0	0	1	0.4
Chaetospermum camelliae Agnihothr.	1	0.8	2	1.3	3	1.1
Colletotrichum sp.	28	22.4	29	20.1	57	21.2
Cytoplacosphaeria phragmiticola O. K. Poon *et* K. D. Hyde	1	0.8	15	10.4	16	5.9
Cytoplea sp.	15	12	17	11.8	32	11.9
Dinemasporium strigosum Pers. *ex* Fr.	0	0	1	0.7	1	0.4
Macrophomina sp.	0	0	16	11.1	16	5.9
Microsphaeropsis sp.	9	7.2	1	0.7	10	3.7
Neottiosporina sp.	0	0	1	0.7	1	0.4
Pestalotiopsis sp.	0	0	2	1.3	2	0.7
Phoma sp.	5	4	4	2.8	9	3.3
Phomopsis sp. 1	21	16.8	31	21.5	52	19.3
Phomopsis sp. 1	2	1.6	0	0	2	0.7
Pseudorobillarda phragmitis (Cunnell) Morelet	1	0.8	0	0	1	0.4
Sclerostagonospora sp.	5	4	6	4.2	11	4.1
Septoriella sp.	1	0.8	21	14.6	22	8.2
Stagonospora sp.	0	0	2	1.3	2	0.7
Stauronema sp.	2	1.6	0	0	2	0.7
Tetranacrium sp.	1	0.8	0	0	1	0.4
Hyphomycetes (25 species)						
Acremonium sp. 1	0	0	7	4.9	7	2.6
Acremonium sp. 2	0	0	1	0.7	1	0.4

Table I. Continued.

Fungi	Outside fence		Along edge of gei wai		Total no. of collections	% occurrence
	No. of conditions	% occurrence	No. of collections	% occurrence		
Alternaria alternata Nees	0	0	1	0.7	1	0.4
Arthrinium state of *Apiospora* sp.	5	4	8	5.6	13	4.8
Arthrinium state of *Apiospora montagnei* Sacc.	11	8.8	8	5.6	19	7.1
Arthrobotrys sp.	16	12.8	3	2.1	19	7.1
Arthrobotrys conoides Drechsler	0	0	7	4.9	7	2.6
Cladosporium sp.	0	0	35	24.3	35	13
Dactylaria sp.	0	0	11	7.6	11	4.1
Dendrostilbella sp.	3	2.4	0	0	3	1.1
Drechslera hawaiiensis (Bugnicourt) Subram. *et* Jain *ex* M. B. Ellis	2	1.6	2	1.3	4	1.4
Ellisembia sp.	0	0	4	2.8	4	1.5
Fusarium sp.	0	0	40	28.8	40	14.9
Gliomastix sp. 1	3	2.4	9	6.3	12	4.5
Gliomastix sp. 2	0	0	5	3.5	5	1.9
Paecilomyces sp.	0	0	6	4.2	6	2.2
Penicillium sp.	9	7.2	3	2.1	12	4.5
Phaeosiaria sp.	1	0.8	3	2.1	4	1.4
Pithomyces maydicus (Sacc.) M. B. Ellis	3	2.4	0	0	3	1.1
Rhinocladiella sp.	13	10.4	17	11.8	30	11.2
Sarocladium sp.	0	0	12	8.3	12	4.5
Spegazzinia tessarthra (Berk. *et* Curt.) Sacc.	1	0.8	0	0	1	0.4
Stachybotrys sp.	4	3.2	2	1.3	6	2.2
Tetraploa aristata Berk. *et* Broome	14	11.2	1	0.7	15	5.6
Trichoderma sp.	33	26.4	0	0	33	12.3
No. of samples examined	125		144		269	

thin-walled cells, both in the form of *textura angularis* (Fig. 2). *Catenophyses* 5–15 × 27.5–145 μm septate, hyaline, developing from pseudoparenchyma of the centrum. *Asci* 105–167.5 × 32.5–47.5 μm, 8-spored, clavate, pedicellate, with an apical pore, IKI-negative, thin-walled, unitunicate, cytoplasm constricted below the apex, persistent, maturing successively on the ascogenous tissue at base of locule (Figs 3–8). *Ascospores* 25–35 × 7.7–10.5 μm, ellipsoid-fusiform, 1-septate, not constricted at septa, provided with a large lipid guttule in each cell, bearing one appendage at each end; appendages hamate, filamentous, hyaline (Figs 9, 10).

Mode of life: Saprobic.

Habitat: Stems and leaf sheaths of *Phragmites australis*.

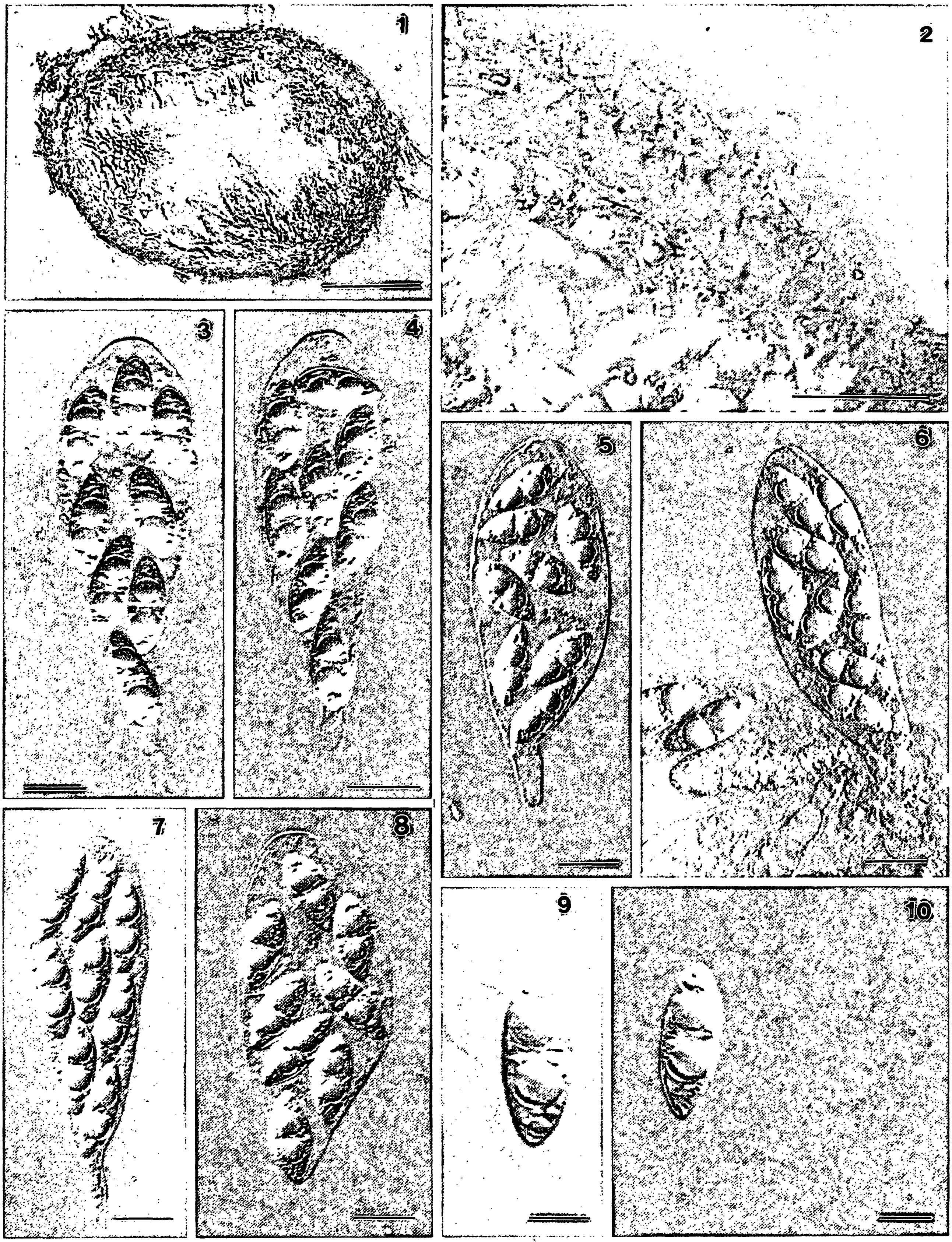
Known distribution: Hong Kong.

Material examined: Decaying stems and leaf sheaths of intertidal *P. australis*, Mai Po Marshes, Hong Kong (22°29′ N, 114°02′ E), 17 Aug. 1995, O. K. Poon [HKU(M) 5186, holotype]; decaying stems and leaf sheaths of intertidal *P. australis*, Mai Po Marshes, Hong Kong (22°29′ N, 114°02′ E), 16 Nov. 1995, O. K. Poon [HKU(M) 5187].

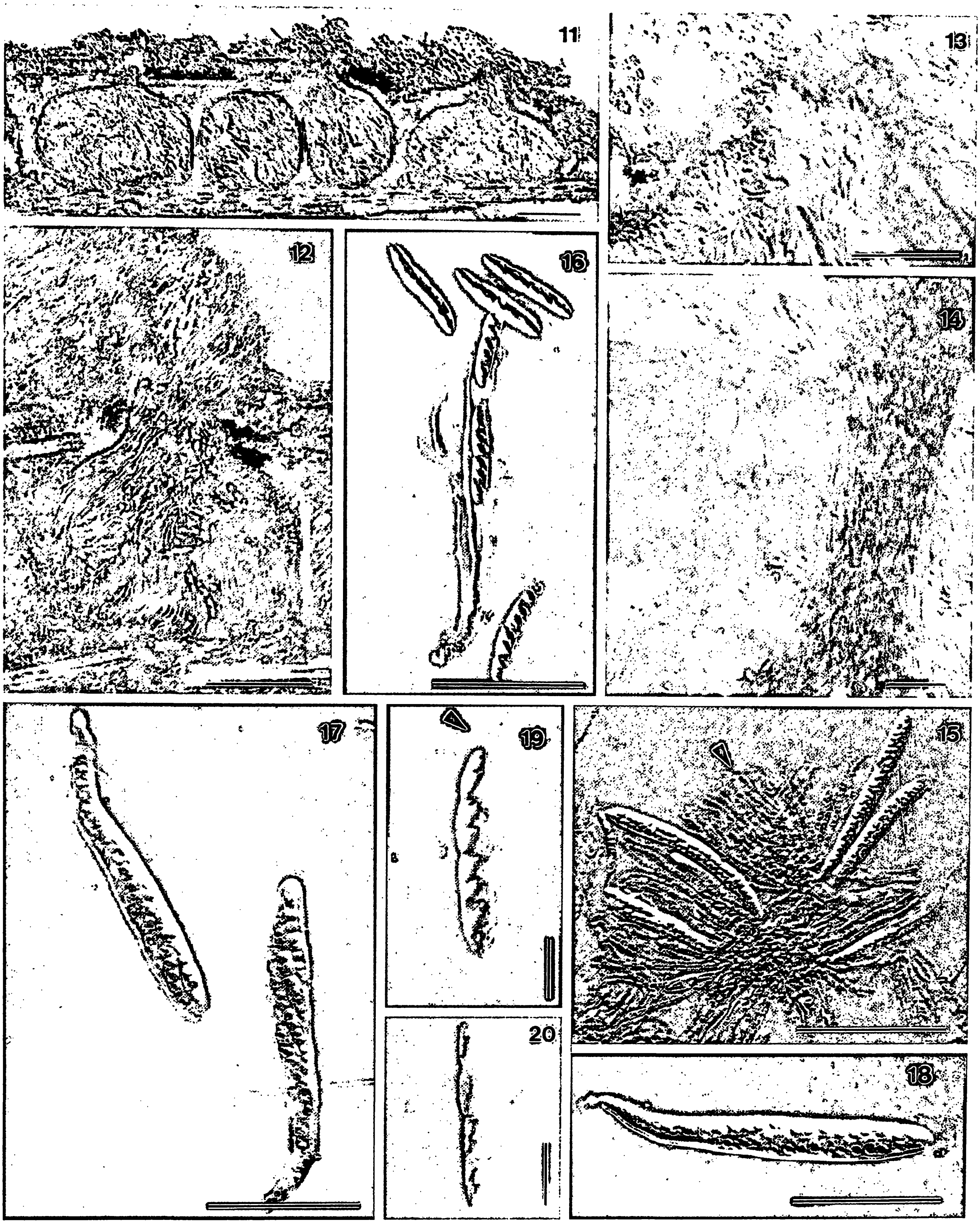
This collection on decaying stems and leaf sheaths of *P. australis* in the intertidal region is superficially very similar to *Halosarpheia culmiperda* Kohlm., Volkm.-Kohlm. *et* O. E. Erikss. (Kohlm. *et al.* 1995 c). The species, however differs, as *H. culmiperda* has wider ascospores (9–13 μm) with subapical cap-like appendages, narrower clavate asci (23.5–30 μm wide) and tawny ascomata. In *Halosarpheia phragmiticola* ascospores are 7.7–10.5 μm wide, appendages are large and hamate, asci are saccate-clavate and 32.5–47.5 μm wide, and ascomata are black. *Halosarpheia phragmiticola* should be compared with *Aniptodera juncicola* Volkm.-Kohlm. *et* Kohlm., from *Juncus roemerianus* (Volkmann-Kohlmeyer and Kohlmeyer 1994). *Aniptodera juncicola* lacks appendaged ascospores, and has greyish brown to fuscous ascomata. The ascopores also differ in shape from those of *Halosarpheia viscosa* (I. Schmidt) Shearer *et* Crane.

Massarina phragmiticola O. K. Poon *et* K. D. Hyde, sp. nov.
Figs 11–20

Etymology: In reference to the host *Phragmites australis*.



Figs 1−10. *Aniptodera phragmiticola* (from holotype).
Fig. 1. Section of ascoma (bar = 100 μm). Fig. 2. Section of ascoma, illustrating peridium (bar = 10 μm). Figs 3−8. Mature asci (bars = 20 μm). Figs 9−10. Mature ascospores with filamentous appendages at both ends (bars = 10 μm).



Figs 11–20. *Massarina phragmiticola* (from holotype).
Fig. 11. Section of aggregated ascomata (bar = 100 μm). Fig. 12. Section of individual ascoma (bar = 100 μm). Fig. 13. Section through papilla (bar = 50 μm). Fig. 14. Section of peridium (bar = 10 μm). Fig. 15. Hypha-like septate pseudoparaphyses (arrow) in a gelatinous matrix (bar = 100 μm). Figs 16–18. Mature asci (bars = 50 μm) fissitunicate in 16. Figs 19–20. Mature ascospores (bars = 10 μm). Fig. 19. Mature ascospore with distal appendage (arrow) markedly smaller than proximal appendage.

Diagnosis

Ascomata *180−280 µm in diameter., 150−200 µm alta, solitaria vel aggregata, immersa, subglobosa vel ellipsoidea, nigra, glabra, ostiolata, breve papillata.* Asci *102.5−133 × 12.5−16.3 µm, cylindrici vel clavati, fissitunicati, 8-spori, pedicellati.* Ascosporae *28−37.5 × 4.7−6.5 µm (x = 33.5 × 5.3 µm, n = 25), hyalinae, bicellulares, cylindricae, utrinque obtusae, guttulatae, appendiculatae.*

Ascomata 180−280 µm in diameter, 150−200 µm high, solitary or aggregated, immersed, subglobose to ellipsoidal, black, glabrous, ostiolate, short papillate (Figs 11−13). *Peridium* 11.8−19.7 µm thick comprising several layers of compressed, brown-walled angular cells (Fig. 14). *Pseudoparaphyses* ca 85 µm long, 2.5−3 µm in diameter at the base and 1.5−2.5 µm in diameter at the apex, filiform, hypha-like, septate, numerous, mostly free-ended, unbranched or branched at the base, markedly shorter than the asci, invested in mucilage (Fig. 15). *Asci* 102.5−133 × 12.5−16.3 µm, 8-spored, cylindrical to clavate, fissitunicate, pedicellate, invested in mucilage, developing from base of locule (Figs 15−18). *Ascospores* 28−37.5 × 4.7−6.5 µm (x = 33.5 × 5.3 µm, n = 25), 2−3 seriate, 2-celled, hyaline, obtuse at both ends, uni-septate, constricted at septa; apical cell irregularly cylindrical, straight, basal cell cylindrical, straight or slightly curved at the end; guttulate, appendaged at both ends; appendages cupulate, mucilaginous; apical appendage 3.7−6.3 µm in diameter, markedly smaller than basal appendage; basal appendage 7.5−9.5 µm in diameter (Figs 19, 20).

Mode of life: Saprobic.

Habitat: On decaying stems and leaf sheaths of intertidal *Phragmites australis*.

Known distribution: Hong Kong.

Material examined: Decaying stems and leaf sheaths of *Phragmites australis*, Mai Po Marshes, Hong Kong (22°29' N, 114°02' E), 16 Nov. 1995, O. K. Poon [HKU(M) 5188, holotype].

Massarina phragmiticola was found to be an infrequent intertidal species occurring on dead *P. australis* stems and leaf sheaths. It can readily be distinguished from the *Massarina ricifera* Kohlm., Volkm.-Kohlm. *et* O. Erikss. and *M. carolinensis* Kohlm., Volkm.-Kohlm. *et* O. Erikss. from *Juncus roemerianus* (Kohlmeyer *et al.* 1995b, 1996) as *M. ricifera* has smaller ascospores (19−25 × 5.5−7 µm) surrounded by a spreading mucilaginous sheath. In *M. carolinensis* ascospores are also smaller (16.5−21 × 4.5−6.5 µm) and are totally surrounded by a gelatinous sheath. *Massarina phragmiticola* and *M. ricifera* are marine species, while *M. carolinensis* is a terrestrial species.

Phomatospora phragmiticola O. K. Poon *et* K. D. Hyde, sp. nov.
Figs 21−35

Etymology: In reference to the host *Phragmites australis*.

Diagnosis

Ascomata *155−175 µm in diameter., 125−133 µm alta, immersa, globosa vel subglobosa, membranacea, nigra, ostiolata, breve papillata, solitaria.* Asci *85−125 × 5−7.5 µm, 8-spori, cylindrici, unitunicati, pedicellati, apparatu apicali praediti.* Ascosporae *7.5−11.3 × 3−5 µm (x = 9.3 × 4.2 µm; n = 50), unicellulares, ellipsoideae, hyalinae, striatae, appendiculatae.*

Ascomata 155−175 µm in diameter, 125−133 µm high, immersed in outer soft tissue of stem, globose to subglobose, membranous, black, ostiolate, short papillate, solitary (Fig. 21). *Neck* 33−38 µm long, 32−45 µm in diameter, cylindrical, papilla central, periphysate (Fig. 22). *Peridium* 6−7 µm thick, comprising several layers of compressed cells, hyaline inwardly (Fig. 23). *Paraphyses* 7−8 µm in diameter at the base, hypha-like, straight to flexous, septate, tapering distally, hyaline, numerous, developing from the base of the ascoma (Fig. 24). *Asci* 85−125 × 5−7.5 µm, eight-spored, cylindrical, unitunicate, thin-walled, pedicellate, with a refractive apical ring which stains in methylene blue, asci developing from base of ascoma (Figs 25−29). *Ascospores* 7.5−11.3 × 3−5 µm (x = 9.3 × 4.2 µm; n = 50), unicellular, ellipsoidal, hyaline, mostly uniseriate, longitudinally striate, with a bifurcate mucilaginous appendage, 5−8 µm in diameter, provided at each end; appendage does not stain in methylene blue (Figs 30−35).

Mode of life: Saprobic.

Habitat: Stems and leaf sheaths of *Phragmites australis*.

Known distribution: Hong Kong.

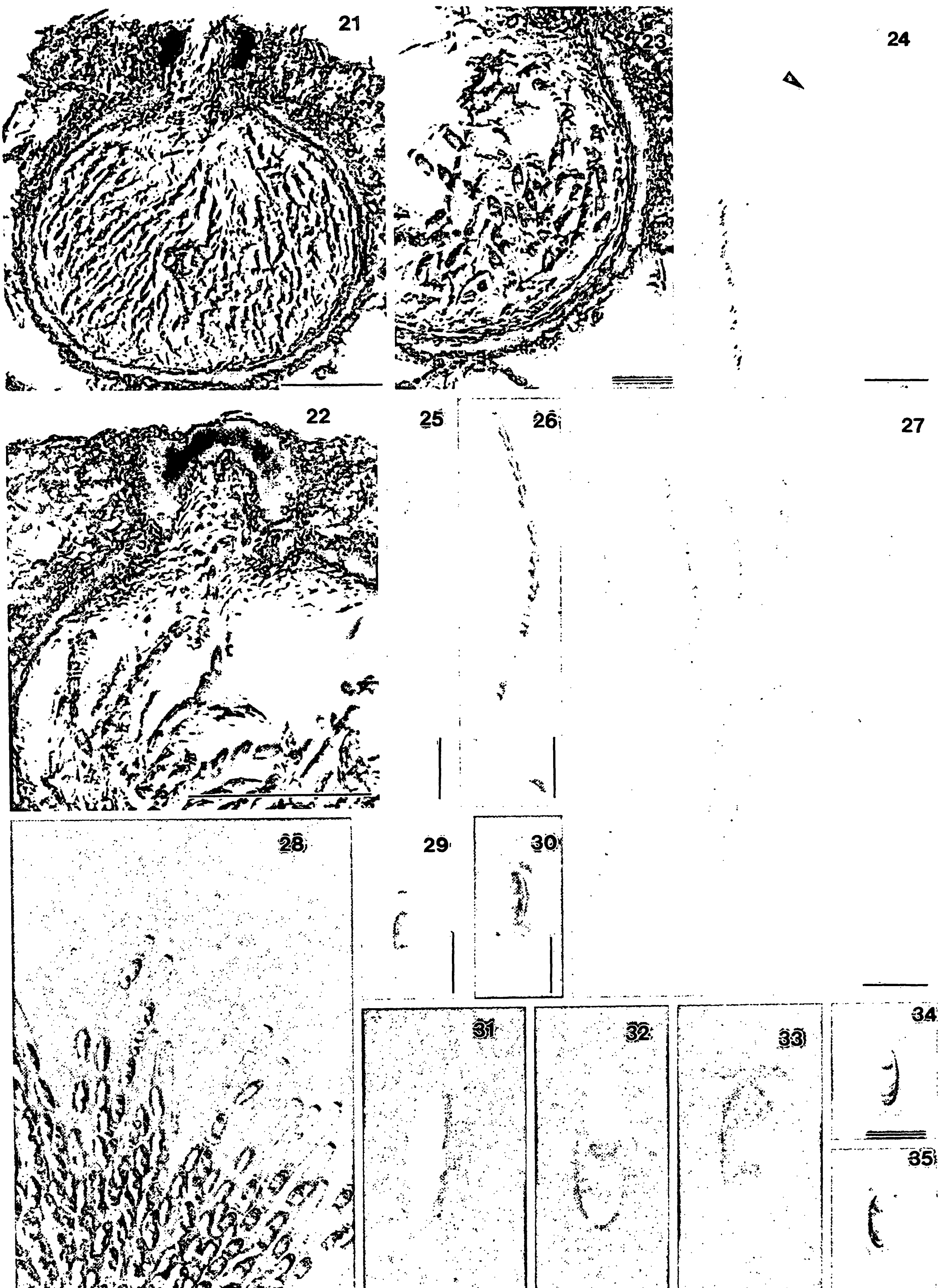
Material examined: Decaying stems and leaf sheaths of intertidal *P. australis*, Mai Po Marshes, Hong Kong (22°29' N, 114°02' E), 17 Aug. 1995, O. K. Poon [HKU(M) 5189, holotype], decaying stems and leaf sheaths of intertidal *P. australis*, Mai Po Marshes, Hong Kong (22°29' N, 114°02' E), 16 Nov. 1995, O. K. Poon [HKU(M) 5190].

Phomatospora phragmiticola can be considered to be an obligate marine fungus as it has the highest affinity in the intertidal region. *Phomatospora phragmiticola* can be confused with *P. bellaminuta* Kohlm., Volkm.-Kohlm. *et* O. E. Erikss. (Kohlmeyer *et al.* 1995b) which was found at the lower parts of decaying *Juncus roemarianus* in the U. S. A. The differences between these species and other marine species are given in Table II. *Phragmites phragmiticola* differs from *P. berkelyi* Sacc, which has smaller ascospores (Kirk 1984).

Cytoplacosphaeria phragmiticola O. K. Poon *et* K. D. Hyde, sp. nov.
Figs 36−45

Diagnosis

Conidiomata *ca 800 µm in diameter., 290 µm alta, eustromatica, immersa vel erumpentia, brunnea, ellip-*



Figs 21–35. *Phomatospora phragmiticola* (from holotype).
Fig. 21. Section of ascoma (bar = 50 μm.). Fig. 22. Section through neck (bar = 50 μm). Fig. 23. Section through peridium (bar = 10 μm). Fig. 24. Hypha-like hyaline paraphyses (bar = 20 μm). Figs 25–28. Mature asci (bars = 20 μm). Fig. 29. Apex of mature ascus illustrating apical ring (bar = 10 μm). Figs 30, 34 and 35. Mature ascospores (bars = 10 μm). Fig. 39. Mature ascospore, showing longitudinal striations. Figs 31–33. Mature ascospores with bifurcate appendages (bars = 5 μm).

Table II. Synopsis of marine species of *Phomatospora*.

	P. acrostichi K. D. Hyde	*P. bellaminuta* Kohlm. *et* Volkm.-Kohlm.	*P. kandeliae* K. D. Hyde	*P. phragmiticola* Kohlm. *et* Volkm.-Kohlm.
Ascomata (μm)	106–274 high 176–330 diam.	115–125 high 125–160 diam.	145–260 high 195–325 diam.	125–133 high 155–175 diam.
Ascospore size (μm)	5.7–7.1 × 2–2.8	10.1–12.5 (–13) × 3.7–4.6 (x = 11.3 × 4.1, n = 67)	11.5 × 16 × 5.5–8	7.5–11.3 × 3.3–5 (x = 9.3 × 4.2, n = 50)
Appendages	Bifurcate, at one end	Gelatinous caps, staining blue in methylene blue	Caps which may become filamentous	Bifurcate, not staining blue in methylene blue
Ascospore arrangement in ascus	Uniseriate	1–2 seriate	Uni-or-overlapping uniseriate	Uniseriate
Ascospore shape	Ellipsoidal with rounded ends	Ellipsoidal with rounded ends	Ellipsoidal with rounded ends	Ellipsoidal with somewhat acute ends
Habitat	*Acrostichum*, intertidal	*Juncus*, lower part of stems and leaf sheaths	*Kandelia*, intertidal	*Phragmites*, lower parts of stems and leaf sheats
Reference	Hyde 1988	Kohlmeyer and Volkmann-Kohlmeyer 1995 a	Hyde 1992	This paper

soidea vel lenticularia. Condiophora *nullae.* Cellulae conidiogenae *9.8–12.5 × 7.3–8 μm, enteroblasticae, phialidicae, discretae, determinatae, ampulliformes vel doliiformes, laeves.* Conidia *17.5–75 × 2.5–5 μm, cylindrica, laevia, curva, hyalina, 0–5 septata.*

Conidiomata ca 800 μm in diameter, 290 μm high, eustromatic, loosely aggregated into stromata with 1–5 locules, immersed, brown, ellipsoidal to lenticular, scarcely erumpent (Figs 36, 37). *Peridium* outer wall consists of several layers of thick-walled dark brown cells, in the form of *textura angularis*; becoming thinner and paler towards the conidiogenous region. *Ostioles* indistinct, dehiscence possibly by rupture of the upper wall (Fig. 38). *Conidiophores* absent. *Conidiogenous cells* 9.8–12.5 × 7.3–8 μm, enteroblastic, phialidic, discrete, determinate, ampulliform to doliiform, smooth, with apical or lateral apertures, collarette clearly visible, channels comparatively wide, hyaline, developing from inner cells of locules (Figs 39–42). *Conidia* 17.5–75 × 2.5–5 μm, cylindrical, straight, curved or irregular, thin-walled, minutely guttulate, smooth, hyaline, 0–5 septate, not constricted at septa (Figs 43–45).

Mode of life: Saprobic.

Habitat: On decaying stems and leaf sheaths of *Phragmites australis*.

Known distribution: Hong Kong.

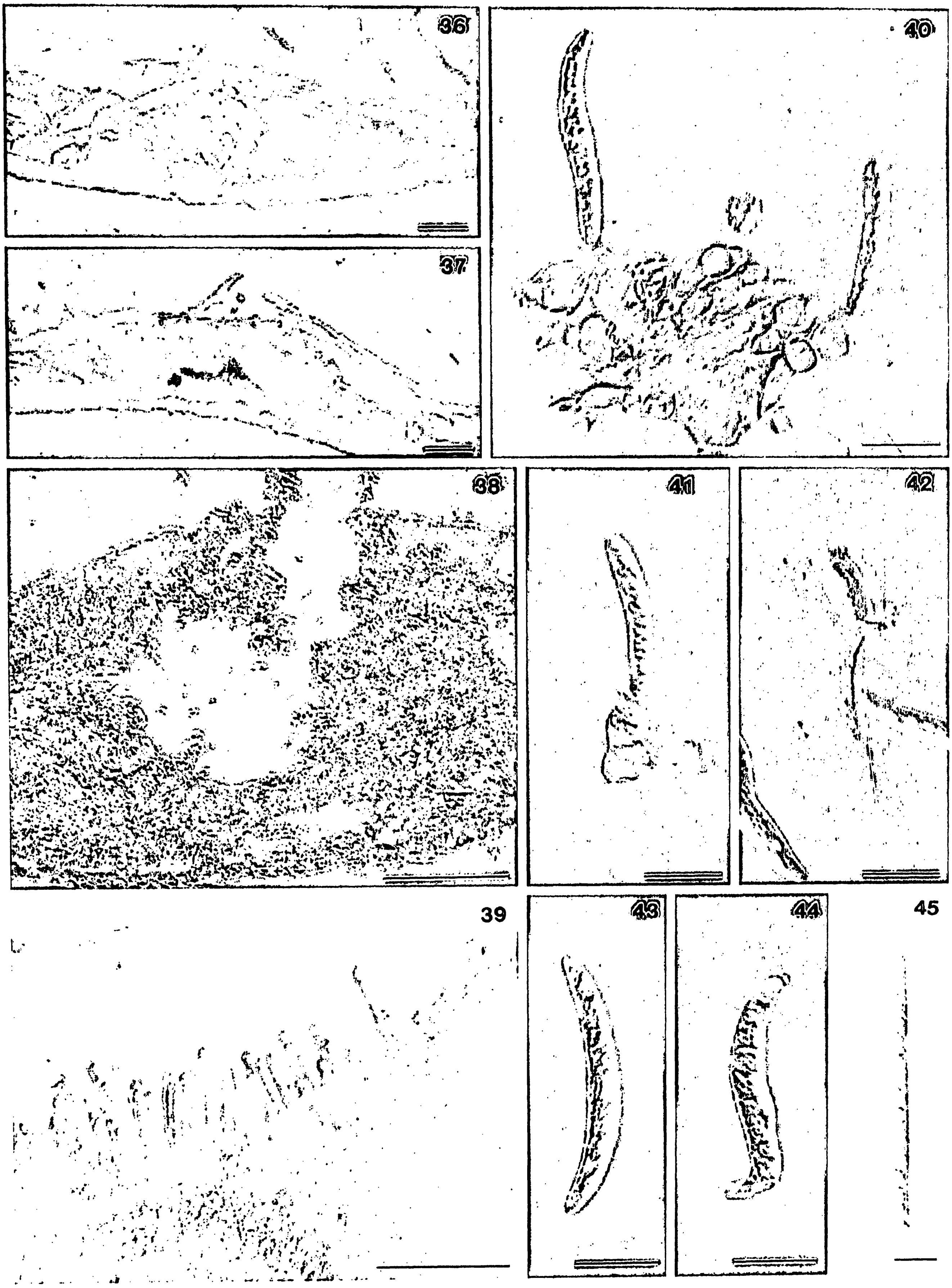
Material examined: Decaying stems of leaf sheaths of *P. australis*, Mai Po Marshes, Hong Kong (22°29' N, 114°02' E), 16 Nov. 1995, O. K. Poon [HKU(M) 5191, holotype].

Cytoplacosphaeria phragmiticola is a facultative brackish water species frequently occurring on *Phragmites australis* stems and leaf sheaths collected from the sides of the gei wai. It has a high affinity around 10–20 cm above the water-air interface, which is the upper to middle portion of *P. australis* stems and leaf sheaths. *Cytoplacosphaeria phragmiticola* was recovered once on the basal portion of *P. australis* stem and leaf sheath in the intertidal region outside the gei wai.

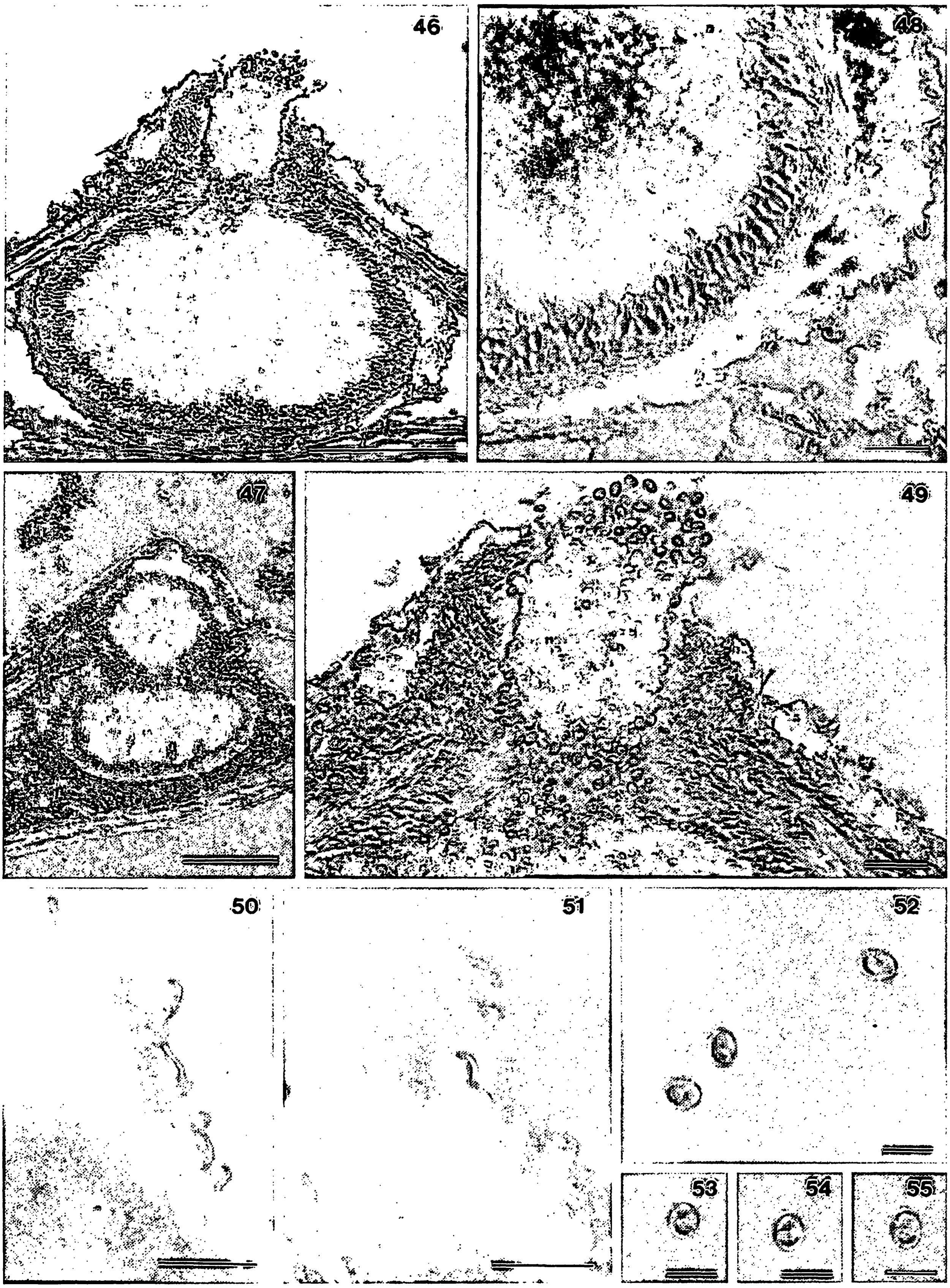
This species is similar to the type species *Cytoplacosphaeria rimosa* (oud.) Petr., which occurs on stems of leaf sheaths of *Phragmites australis*, *P. vulgaris* Crepin and *Phragmites* sp., in Latvia, Czechoslovakia and England (Sutton 1980). However, *C. rimosa* has conidiogenous cells with a distinct collarette and shorter eguttulate conidia (13–20 × 3 μm). This makes *C. phragmiticola* readily distinguishable from *C. rimosa*. The ostiole in *C. rimosa* is reported to be single, circular and papillate, whereas it appears that *C. phragmiticola* dehisces by rupture of the upper wall.

Microsphaeropsis sp.
Figs 46–55

Conidiomata pycnidial, ampulliform, occasionally subglobose, black, immersed, papillate, mostly unilocular, occasionally bilocular, *ca* 280 μm in diameter, 240 μm high (Figs 46, 47). *Peridium* 11–23 μm thick, outer wall of 1–3 layers of thick-walled dark brown cells, in the form of *textura angularis*; middle wall of 3–5 layers of thick-walled pale brown cells,



Figs 36–45. *Cytoplacosphaeria phragmiticola* (from holotype).
Figs 36–38. Sections of conidiomata (bars = 100 µm). Fig. 39. Conidia arising from conidiogenous cells developing from base of locule (bar = 50 µm. Figs 40, 41. Conidiognous cells with developing conidia (bars = 20 µm). Fig. 42. Conidiogenous cell (bar = 20 µm). Figs 43–55. Mature conidia (bars = 5 µm).



Figs 46–55. *Microsphaeropsis* sp.
Figs 46, 47. Sections of conidiomata (bars = 100 μm). Fig. 48. Section through peridium (bar = 20 μm). Fig. 49. Section through papilla (bar = 20 μm). Figs 50, 51. Condiophores with developing conidia (bars = 10 μm). Figs 52–55. Mature conidia (bars = 5 μm).

in the form of *textura porrecta*; inner wall of single layer of hyaline thin-walled cells, in the form of *textura angularis* (Fig. 48). *Papilla ca* 80 µm long, 130 µm wide (Fig. 49). *Conidiophores* absent. *Conidiogenous cells* enteroblastic, phialidic, determinate, discrete, doliiform to cylindrical, collarette minute, smooth, hyaline, developing from inner cells of pycnidial wall (Figs 50, 51). *Conidia* 3.7–5 × 3.7–4.5 µm, 1-celled, brown, thick-walled, ellipsoidal, biguttulate at both ends, aseptate, enclosed in a thin mucilagenous sheath (Figs 52–55).

This is a marine species which occurred only at the basal portion of decaying standing stems and leaf sheaths of *Phragmites australis* and on decaying stems and leaf sheaths of *P. australis* submerged in brackish water. No species from this genus is reported on salt marsh plants. There are 800 names in *Coniothyrium* and many of them probably belong in *Microsphaeropsis* (Sutton 1980). A major revision of the group is needed and therefore it would be unwise to name this species beyond generic level. *Coniothyrium obiones* Jaap is a marine species which has been reported on dead stems and leaf sheaths and moribund propagules of a coastal salt marsh plant *Halimione portulacoides* (L.) Aellen collected in England and Germany (Kohlmeyer and Kohlmeyer 1979). *Microsphaeropsis* sp. collected from *Phragmites australis*, has non-tapered conidiogenous cells and conidia enclosed in mucilaginous sheaths, and differs from *Coniothyrium obiones* Jaap.

Discussion

Biodiversity of fungi found in Hong Kong mangroves

The results of these studies are preliminary observations on the fungi involved in the decay of stems and leaf sheaths of *Phragmites australis* at Mai Po Marshes in Hong Kong. Because of the relatively small sample sizes based on two site visits in August and November 1995 the data on frequency and composition must be treated with caution as additional collections at different times may result in different compositions. However, the results do represent the first data on the biodiversity of fungi on grasses in the tropics.

Similar studies on biodiversity of fungi on other grasses in Hong Kong are unavailable for comparison. However, there have been several reports on lignicolous fungi from mangroves in Hong Kong (Vrijmoed 1990, Vrijmoed *et al.* 1994, Sadaba *et al.* 1995).

In the study of Vrijmoed (1990), Vrijmoed *et al.* (1994) and Sadaba *et al.* (1994), and in this study, *Lignincola laevis* was the most frequently identified fungus on *Acanthus ilicifolius* L., *Avicennia marina*, *Kandelia candel*, *Phragmites australis* and driftwood. *Aniptodera chesapeakensis* was also commonly found on *Acanthus ilicifolium*, *Avicennia marina* and *Phragmites australis*. *Halosarpheia* sp. and *Lulworthia* sp. were common to both *Kandelia candel* and driftwood. Species of *Leptosphaeria* were found on *Acanthus ilicifolium* and *Phragmites australis* and driftwood. *Halonectria milfordensis* was found on *Aegiceras corniculatum* and *Kandelia candel*.

The number of taxa recorded in this study (61) is higher than reported by Sadaba *et al.* (1995) on *Acanthus ilicifolium*. Only 44 higher fungi were found associated with decaying standing parts of *Acanthus ilicifolium* (Sadaba *et al.* 1995). There were differences in fungal composition on *Phragmites australis* and *Acanthus ilicifolium*. In the intertidal region of Mai Po Marshes, the most common taxa on standing parts of *Acanthus ilicifolium* were *Acremonium* sp. (55%), *Colletotrichum* sp. (42.5%), *Phoma* sp. (42.5%), *Fusarium* sp. (25%) and *Tubercularia* sp. (24.2%) (Sadaba *et al.* 1995). On the other hand, the common taxa found on dead stems and leaf sheaths of *Phragmites australis* were *Lignincola laevis* (22.7%), *Colletotrichum* sp. (21.2%), *Phomopsis* sp. 1 (19.3%), *Aniptodera phragmiticola* (15.2%), *Fusarium* sp. (14.9%), *Cladosporium* sp. (13%) and *Trichoderma* sp. (12.3%).

The differences in mycota reported may be due to different host species studied. Sadaba *et al.* (1995) only collected *Acanthus ilicifolium* samples from the intertidal region of Mai Po Marshes (outsides the security fence), while *Phragmites australis* were collected from both the intertidal region and along the edge of the gei wai. There appears to be a group of fungi which are only found inside or along the edge of the gei wai. Nonetheless, two frequent taxa (*Colletotrichum* sp. and *Fusarium* sp.) are common to both hosts.

Few mitosporic fungi are reported by Vrijmoed (1990), since most mitosporic fungi are terrestrial species and are unlikely to occur on the decaying plant samples found in the aquatic environment. The large numer of fungi recorded from *Phragmites australis* and *Acanthus ilicifolius* reflected the intensive study of these plants. Further study is needed concerning the host specificity of fungi associated with mangrove plants in Hong Kong.

Biodiversity of salt marsh fungi found on grasses

The diversity of fungi recorded from other salt marsh grasses, such as species of *Spartina* is lower than those found in this study. *Phragmites australis* has the greatest number of fungal species recorded (63), followed by *Spartina alterniflora* (49), *Spartina townsendii* H. Groves *et* J. Groves (39), an unidentified species of *Spartina* (30), *Spartina patens* (Aiton.) Muhl. (16), *Spartina cynosuroides* (L.) Roth (14), *Spartina foliosa* Trin. (3), *Spartina maritima* (Curtis) Fernald (2), and *Spartina anglica* C. E. Hubbard (1) (Gessner and Kohlmeyer 1976).

The most common species found on these species of *Spartina* are *Buergenerula spartinae* Kohlm. *et*

Gessner, *Claviceps purpurea* (Fr.) Tul., *Phaeosphaeria albopunctata*, *Leptosphaeria marina* Ellis *et* Everh., *Passeriniela obiones* (Crouan *et* Crouan) K. D. Hyde *et* Mouzouras, species of *Lulworthia*, *Phaeosphaeria spartinicola* Leucht., *Puccinia sparganioides* Ellis *et* Barth., *Phoma* sp., and *Stagonospora* sp. (Gessner and Kohlmeyer 1976). The most common taxa on *Phragmites australis* are *Aniptodera phragmiticola*, *Cladosporium* sp., *Colletotrichum* sp., *Fusarium* sp., *Lignincola laevis*, *Phomopsis* sp. 1 and *Trichoderma* sp.

Gessner (1977) found a characteristic group of fungi associated with *Spartina alterniflora* along the east coasts of North and South America. Differences in fungal taxa found on *Phragmites australis* and species of *Spartina* can be attributed to temperature, host specificity and location. Perhaps it is unlikely for a group of fungi adapted to decomposing *S. alterniflora* to be found on decaying stems and leaf sheaths of *P. australis*. Further investigation is required regarding host specificity of saprophytic fungi on grasses.

Saprophytic fungi occurring on *Phragmites australis* in Mai Po

The differences in fungal composition at the two sites in Mai Po may be due to the preference of fungi towards certain inundation periods. Salinity difference is not a factor contributing to these differences. Salinities in intertidal region and gei wai are nearly the same due to the connection of two water bodies via the sluice gate. There are also differences in species composition of terrestrial fungi between the two sites. This is not expected since terrestrial fungi are not affected by the differences of inundation period as in aquatic species.

There are also differences between the intertidal and terrestrial fungi. With the exception of *Chaetomium globosum* all ascomycetes are intertidal fungi (e. g. *Phomatospora phragmiticola* and *Lignincola laevis*). On the other hand, most terrestrial taxa were mitosporic fungi, such as *Fusarium* sp., *Gliomastix* sp. 2. sp., *Macrophomina* sp. and *Trichoderma* sp. This may be due to the fact that mitosporic fungi, especially hyphomycetes are better adapted to the terrestrial environment at the upper level of the dead standing plant. Rapid colonisation of substrates by production of large amount of light conidia is characteristic of most hyphomycetes.

Terrestrial fungi (e. g. *Chaetomium globosum* and *Dendrostilbella* sp.) were not recovered from lower portions of standing decaying stems and leaf sheaths of *Phragmites australis* as they are probably not adapted to periodic submergence in seawater. This was also observed in the succession pattern of decomposers on *P. australis* in Japan (Tanaka 1991).

In conclusion, fungal communities associated with decaying *Phragmites australis* permanently submerged in the gei wai, are different to those in the intertidal region. The diversity of fungal taxa found in this study is probably an underestimate, since the identification of species depends on the reproductive structures being produced, and a relatively short study was carried out.

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