



Novel fungi from an ancient niche: cercosporoid and related sexual morphs on ferns

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Key words

biodiversity
Cercospora
frond spot
multilocus sequence typing (MLST)
Mycosphaerella
phylogeny
Pteridophyta
systematics

Abstract The fern flora of the world (*Pteridophyta*) has direct evolutionary links with the earliest vascular plants that appeared in the late Devonian. Knowing the mycobiota associated to this group of plants is critical for a full understanding of the Fungi. Nevertheless, perhaps because of the minor economic significance of ferns, this niche remains relatively neglected by mycologists. Cercosporoid fungi represent a large assemblage of fungi belonging to the *Mycosphaerellaceae* and *Teratosphaeriaceae* (*Ascomycota*) having cercospora-like asexual morphs. They are well-known pathogens of many important crops, occurring on a wide host range. Here, the results of a taxonomic study of cercosporoid fungi collected on ferns in Brazil are presented. Specimens were obtained from most Brazilian regions and collected over a 7-yr period (2009–2015). Forty-three isolates of cercosporoid and mycosphaerella-like species, collected from 18 host species, representing 201 localities, were studied. This resulted in a total of 21 frond-spotting taxa, which were identified based on morphology, ecology and sequence data of five genomic loci (actin, calmodulin, ITS, LSU and partial translation elongation factor 1- α). One novel genus (*Clypeosphaerella*) and 15 novel species (*Cercospora samambaiae*, *Clypeosphaerella sticheri*, *Neoceratosperma alsophila*, *N. cyatheae*, *Paramycosphaerella blechni*, *Pa. cyatheae*, *Pa. dicranopteridis-flexuosae*, *Pa. sticheri*, *Phaeophleospora pteridivora*, *Pseudocercospora brackenicola*, *Ps. paranaensis*, *Ps. serpocaulonicola*, *Ps. trichogena*, *Xenomycosphaerella diplazii* and *Zasmidium cyatheae*) are introduced. Furthermore, 11 new combinations (*Clypeosphaerella quasiparkii*, *Neoceratosperma yunnanensis*, *Paramycosphaerella aeryhyalinosporum*, *Pa. dicranopteridis*, *Pa. gleicheniae*, *Pa. irregularis*, *Pa. madeirensis*, *Pa. nabiacense*, *Pa. parkii*, *Pa. pseudomarksii* and *Pa. vietnamensis*) are proposed. Finally, nine new host associations are recorded for the following known fungal species: *Cercospora conioigrammes*, *Cercospora* sp. Q, *Ps. abacopteridicola*, *Ps. lygodiicola* and *Ps. thelypteridis*.

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INTRODUCTION

Cercosporoid fungi are well-known plant pathogens that are etiological agents of leaf spot diseases of many important crops (Agrios 2005). Major diseases include angular leaf spot of bean (*Pseudocercospora griseola*), black leaf streak of banana (*Ps. fijiensis*) and leaf spots on many other hosts including grapevine (*Ps. vitis*), celery (*Cercospora apii*) and sugarbeet (*C. beticola*), to name but a few (Braun et al. 2013).

Since the seminal monograph of Chupp (1954) on the genus *Cercospora*, several studies were aimed at investigating this group and dividing cercospora-like fungi into more natural genera. Of special relevance are the publications prepared with that intent (Deighton 1965, 1967, 1971, 1974, 1976, 1979, 1983, 1987, 1990, Pons & Sutton 1988, Braun 1993a–c, 1995, 1998, Crous & Braun 1996, Braun & Mel'nik 1997, Crous et al. 2000). Crous & Braun (2003) also revisited Chupp's work and, using morphological criteria, consolidated the generic circumscription of *Cercospora*, reducing the number of taxa from 3000 to 659 species names. Additionally, numerous stud-

ies dealing with cercosporoid fungi found in different countries have been published, e.g. Brazil (Viégas 1945), Japan (Katsuki 1965), Singapore and the Malay Peninsula (Yen & Lim 1980), Taiwan (Hsieh & Goh 1990), China (Guo & Hsieh 1995, Guo et al. 2003, 2005), South Africa (Crous & Braun 1996), Russia and adjacent countries (Braun & Mel'nik 1997), Korea (Shin & Kim 2001), Laos (Phengsintham et al. 2013a) and Thailand (Phengsintham et al. 2013b). Unfortunately, all of these regional studies of cercosporoids were only based on morphological, ecological and host specificity data for species delimitation, and in many instances, this has proven inadequate (Halleen et al. 2004, Lee et al. 2004, Réblová et al. 2004, Verkley et al. 2004a, b, Crous et al. 2006a, b, 2007a, b, 2009a, b, Arzanlou et al. 2007, Phillips et al. 2008, Shivas et al. 2009).

The tradition of naming fungi in the absence of molecular data remains dominant in published literature, despite the limitations of this approach rendering data-driven comparisons difficult to impossible, especially in groups with known wide host ranges. Of the fungal species described in 2013, 65 % still lacked DNA data (Crous et al. 2015a). The lack of DNA barcodes is still further complicated by the lack of ex-type cultures, which are frequently not deposited in publicly available biological resource centres. This is true for fungi in general, but in the case of the cercosporoid fungi in particular, the situation is further complicated by the fact that they are often only found as asexual morphs (Goodwin et al. 2001). When the sexual morph is present, cercosporoid taxa have traditionally been classified in entirely different genera, with few morphological characters that can be used to facilitate accurate identification (Braun et al. 2013, 2014, 2015). Moreover, many species (especially in

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the tropics and subtropics) are known only from their asexual morphs, and may exhibit considerable morphological variation due to environmental conditions, encouraging mycologists to mistakenly recognise them as distinct genera. As a result, numerous asexual genera, which may eventually prove to be artificial, have been introduced e.g. *Cercodeuterospora*, *Centrospora*, *Heterosporium* and others (Chupp 1954). On the other hand, once these groups are subjected to molecular phylogenetic comparisons, it has frequently also led to a high number of generic lineages that previously were not discernable based on morphology alone, e.g. *Paracercospora*, *Phaeocercospora* and in the *Teratosphaeriaceae* (Crous et al. 2013b, Quaedvlieg et al. 2014).

With DNA sequencing becoming widely available for use by mycologists as a reliable source of information (Taylor et al. 2000), a more concrete classification of fungi was initiated, and several studies have since been published on cercosporoid fungi (Arzanlou et al. 2007, Crous et al. 2007a, 2009b, d, 2013a, Braun et al. 2013, Groenewald et al. 2013, Bakhshi et al. 2014, 2015, Nguanhom et al. 2015). These studies have shown that some morphology-based genera were largely monophyletic, e.g. *Pseudocercospora* and *Ramularia* (Crous et al. 2013a, Groenewald et al. 2013, Bakhshi et al. 2014, 2015, Videira et al. 2015) whereas others like *Passalora* and other genera not recognised as cercosporoids, were clearly polyphyletic, e.g., *Phloeospora*, *Phoma*, *Pseudocercospora*, *Septoria* and *Stagonospora* (Aveskamp et al. 2010, Frank et al. 2010, De Gruyter et al. 2013, Quaedvlieg et al. 2013).

Despite the intense effort by mycologists over the last two centuries at describing the world's mycobiota, this task is far from being complete (Crous et al. 2015a). Several niches harbouring unique fungi that may be of relevance for understanding fungal phylogeny, have been mostly neglected. One case in point is fungi associated with ferns. Ferns are members of the division *Pteridophyta* (= '*Monilophyta*'). In recent classifications (e.g., Smith et al. 2008) the division includes 37 families, approximately 300 genera and more than 9 000 species. Although there are presently c. 1 110 species known from Brazil, it has been estimated that this number may be far greater (Forzza et al. 2015). Approximately 60 different species of fungi have been recorded on ferns in Brazil, from which two are cercosporoid (Viégas 1961, Farr & Rossman 2015, Mendes & Urben 2015). In Brazil and elsewhere, ferns have probably been poorly collected because of the lack of economic importance of most species. One exception in the general absence of monographic treatments of fungi on ferns is the recent publications by Braun et al. (2013, 2014, 2015), a series of works aiming at congregating all cercosporoid taxa by host. Braun et al. (2013) redescribed and discussed 44 cercosporoid species occurring on 47 different fern hosts. One of these (*Pseudocercospora davalliicola*) was originally described from Brazil. Such significant morphological revisions based on previously published species, provide a solid foundation to facilitate future DNA phylogenetic studies.

Early results of the survey for plant pathogenic fungi occurring on ferns in Brazil indicated a plethora of novel taxa to exist in this niche. Two of the preliminary findings, namely two taxa in the *Parmulariaceae*, have already been published: the new genus *Rhagadolobiosis* (Guatimosim et al. 2014a) and the new species *Inocyclus angularis* (Guatimosim et al. 2014b). Similarly, another research group in Asia has been studying fungi on ferns and have recently described the new species *Venustosynnema reniformisporum* and *Zasmidium dicranopteridis* (Kirschner & Liu 2014). Furthermore, the phylogenetic placement of the monotypic class *Mixomyces* was recently elucidated based on the study of *Mixia osmundae*, which is an intracellular parasite of ferns (Toome et al. 2014).

The present work aims to present part of the results of a broad survey of the mycobiota of ferns in Brazil, with particular reference to the cercosporoid and related fungi which were collected in association with frond spots on members of the *Pteridophyta* collected in Brazil. Additionally, this work aims at partially supplementing the initiative of Braun et al. (2013) with robust DNA data, in order to promote a precise taxonomic classification of the cercosporoid fungi within *Mycosphaerellaceae*. In a recent study, Quaedvlieg et al. (2014) proposed employing a Consolidated Species Concept, aiming to integrate ecology, morphology, cultural characteristics and multilocus DNA phylogenetic data in order to appropriately verify species boundaries. The same approach was adopted in the present publication for the cercosporoids occurring on ferns in Brazil.

MATERIALS AND METHODS

Specimens and isolates

Frond samples bearing fungal colonies were collected in Brazil from different biomes, including natural ecosystems in the Amazon, the Atlantic rainforest, the Caatinga and the Cerrado, as well as ruderal areas and gardens between 2009 and 2015. These were dried in a plant press and later examined under a dissecting microscope to detect fungal structures. Such fungal structures, preferably spores, were scraped from a single frond spot, and whenever possible, single conidial colonies were established on potato carrot agar (PCA) (Crous et al. 2009e). In the case of ascospores-producing structures being present, excised lesions were placed in distilled water for approximately 2 h, after which they were placed at the bottom of Petri dish lids, over which the plate containing PCA was placed. Ascospore germination patterns were recognised using the different modes of ascospore germination proposed by Crous (1998). Freehand sections of fungal colonies were prepared and fungal structures mounted in clear lactic acid, lactophenol, lactofuchsin, and/or Melzer's reagent. When necessary, sections were made using a Microm HM 520 freezing microtome. Observations were made with a Nikon SMZ1500 stereo-microscope and with a Nikon Eclipse 80i light microscope using differential interference contrast (DIC) illumination and a Nikon DS-Fi1 camera and NIS-Elements imaging software. Colony descriptions were made on 2 % malt extract agar (MEA), potato dextrose agar (PDA), PCA and oatmeal agar (OA) (Crous et al. 2009e), in the dark at 25 °C and under a 12 h light/dark regime. Colony colours (surface and reverse) were rated according to the colour charts of Rayner (1970). Representative fungarium specimens were deposited in the Fungarium of the Universidade Federal de Viçosa (VIC) and the Fungarium of the CBS-KNAW Fungal Biodiversity Centre, Utrecht, The Netherlands (CBS H). Axenic cultures were deposited in the working collection of P.W. Crous (CPC), housed at CBS, and in the Coleção Octávio de Almeida Drumond (COAD), housed at the Universidade Federal de Viçosa. A complete list of the species and isolates included in this study is presented in Table 1.

Scanning electron microscopy

Samples of dried material containing fungal structures were mounted on stubs with double-sided adhesive tape and gold-coated using a Balzer's FDU 010 sputter coater. A Carl-Zeiss Model LEO VP 1430 scanning electron microscope (SEM) was used to analyse and generate images from the samples.

DNA isolation, amplification and sequencing

Isolates were grown on MEA plates for 20 d at 25 °C. Genomic DNA was extracted from mycelium using the Wizard® Genomic DNA Purification Kit (Promega Corporation, WI, USA) following the manufacturer's instructions. The DNA samples were

Table 1 Collection details and GenBank accession numbers of isolates included in this study. New generated sequences are in bold.

Species	Culture accession numbers ^{1,2}	Host/isolation source	Host family	Country	Collector	ITS	<i>tef1</i>	<i>act</i>	<i>cmdA</i>	LSU
<i>Amycosphaerella africana</i>	CBS 110500 ^{ET} of <i>Mycosphaerella aurantia</i>	<i>Eucalyptus globulus</i>	Myrtaceae	Australia	A. Maxwell	KF901516	KF903115	KF903395	–	KF901837
	CBS 110843 = CPC 850 ^{ET} of <i>Mycosphaerella ellipsoides</i>	<i>Eucalyptus cladocalyx</i>	Myrtaceae	South Africa	P.W. Crous	KF901702	KF903118	KF903407	–	KF902049
	CBS 116154 = CPC 794 ^{ET} of <i>Mycosphaerella africana</i>	<i>Eucalyptus viminalis</i>	Myrtaceae	South Africa	P.W. Crous	KF901700	KF903116	KF903480	–	KF902047
	CBS 680.95 = CPC 796 ^{ET} of <i>Mycosphaerella africana</i>	<i>Eucalyptus viminalis</i>	Myrtaceae	South Africa	P.W. Crous	KF901701	KF903117	KF903589	–	KF902048
<i>Cercospora apii</i>	CBS 116455 = CPC 11556 ^{ET}	<i>Apium graveolens</i>	Apiaceae	Germany	K. Schrammeyer	AY840519	AY840486	AY840450	AY840417	–
	CBS 121.31 = CPC 5073	<i>Beta vulgaris</i>	Chenopodiaceae	Austria	E.W. Schmidt	AY343371	AY343334	AY840444	AY840411	–
	CBS 536.71 = CPC 5087	<i>Apium graveolens</i>	Apiaceae	Romania	O. Constantinescu	AY752133	AY752166	AY752194	AY752225	–
<i>C. apicola</i>	CBS 563.71 = CPC 5083	<i>Plumbago europaea</i>	Plumbaginaceae	Romania	O. Constantinescu	DQ233320	DQ233344	DQ233370	DQ233396	–
	CBS 116457 = CPC 10267 ^{ET}	<i>Apium</i> sp.	Apiaceae	Venezuela	N. Pons	AY840536	AY840503	AY840467	AY840434	–
	CBS 132644 = CPC 10248	<i>Apium</i> sp.	Apiaceae	Venezuela	N. Pons	AY840539	AY840506	AY840470	AY840437	–
	CPC 10220	<i>Apium</i> sp.	Apiaceae	Venezuela	N. Pons	AY840538	AY840505	AY840469	AY840436	–
<i>C. celosiae</i>	CPC 10265	<i>Apium</i> sp.	Apiaceae	Venezuela	N. Pons	AY840540	AY840507	AY840471	AY840438	–
	CBS 132600 = CPC 10660	<i>Celosia argentea</i> var. <i>cristata</i>	Amaranthaceae	South Korea	H.D. Shin	JX143570	JX143326	JX143080	JX142834	–
	CBS 119395 = CPC 12682	<i>Musa</i> sp.	Musaceae	Bangladesh	I. Buddenhagen	EU514222	JX143335	JX143089	JX142843	–
	CBS 132669 = CPC 12683	<i>Musa</i> sp.	Musaceae	Bangladesh	I. Buddenhagen	EU514223	JX143336	JX143090	JX142844	–
<i>C. cf. citrulina</i>	MUCC 576 = MAFF 237913	<i>Citrullus lanatus</i>	Cucurbitaceae	Japan	T. Kobayashion et al.	JX143579	JX143337	JX143091	JX142845	–
	MUCC 577 = MAFF 238205	<i>Momordica charantia</i>	Cucurbitaceae	Japan	E. Imaizumi & C. Nomi	JX143580	JX143338	JX143092	JX142846	–
	MUCC 584 = MAFF 305757	<i>Psophocarpus tetragonolobus</i>	Fabaceae	Japan	–	JX143581	JX143339	JX143093	JX142847	–
	MUCC 588 = MAFF 239409	<i>Ipomoea pes-caprae</i>	Convolvulaceae	Japan	–	JX143582	JX143340	JX143094	JX142848	–
<i>C. conioagrammes</i>	CBS 132634 = CPC 17017 ^{ET}	<i>Conioagramme japonica</i>	Cryptogramma- ceae	Australia	P.W. Crous	JX143583	JX143341	JX143095	JX142849	–
	CPC 24661 = COAD 1067	<i>Macrothyloperis torresiana</i>	Thelypteridaceae	Brazil	R.W. Barreto	KT037509	KT037469	KT037591	KT037458	KT037550
<i>C. cf. physalidis</i>	CPC 24669 = COAD 1093	<i>Macrothyloperis torresiana</i>	Thelypteridaceae	Brazil	R.W. Barreto	KT037512	KT037472	KT037594	KT037461	KT037553
	CPC 24672 = COAD 1089	<i>Macrothyloperis torresiana</i>	Thelypteridaceae	Brazil	R.W. Barreto	KT037513	KT037473	KT037595	KT037462	KT037554
	CPC 24706 = COAD 1997	<i>Macrothyloperis torresiana</i>	Thelypteridaceae	Brazil	E. Guatimosim	KT037507	KT037467	KT037589	KT037456	KT037548
	CPC 25070 = COAD 1769	<i>Hypolepis mitis</i>	Dennstaedtiaceae	Brazil	R.W. Barreto	KT037517	KT037477	KT037599	KT037466	KT037558
	CBS 131.32 = CPC 5076	<i>Nicotiana tabacum</i>	Solanaceae	Indonesia	H. Diddens and A. Jaarsveld	DQ835073	DQ835099	DQ835119	DQ835146	–
	CBS 132632 = CPC 15918	<i>Glycine max</i>	Fabaceae	Mexico	Ma. de Jesús Yáñez-Morales	JX143631	JX143390	JX143144	JX142988	–
	CBS 570.69 = CPC 5075	<i>Nicotiana tabacum</i>	Solanaceae	Nigeria	S.O. Alasoadura	DQ835074	DQ835100	DQ835120	DQ835147	–
	CBS 765.79	<i>Solanum tuberosum</i>	Solanaceae	Peru	L.J. Turkensteen	JX143633	JX143392	JX143146	JX142900	–
	CBS 132607 = CPC 10749 ^{ET}	<i>Pilea pumila</i>	Urticaceae	South Korea	H.D. Shin	JX143634	JX143393	JX143147	JX142901	–
	CBS 132647 = CPC 10693	<i>Pilea hamaoi</i>	Urticaceae	South Korea	H.D. Shin	JX143635	JX143394	JX143148	JX142902	–
<i>C. pseudochenopodii</i>	CPC 11369	<i>Pilea pumila</i>	Urticaceae	South Korea	H.D. Shin	JX143636	JX143395	JX143149	JX142903	–
	CBS 136022 = CCTU 1038 ^{ET}	<i>Chenopodium</i> sp.	Chenopodiaceae	Iran	M. Bakhshi	KJ886516	KJ886355	KJ886033	KJ885872	–
	CCTU 1045	<i>Chenopodium</i> sp.	Chenopodiaceae	Iran	M. Arzanlou	KJ886517	KJ886356	KJ886034	KJ885873	–
	CCTU 1176	<i>Chenopodium album</i>	Chenopodiaceae	Iran	M. Arzanlou	KJ886518	KJ886357	KJ886035	KJ885874	–
	CBS 132594 = CPC 10304	<i>Chenopodium ficifolium</i>	Chenopodiaceae	South Korea	H.D. Shin	JX143572	JX143328	JX143082	JX142836	–
	CBS 132677 = CPC 15599	<i>Chenopodium</i> sp.	Chenopodiaceae	Mexico	Ma. de Jesús Yáñez-Morales	JX143573	JX143329	JX143083	JX142837	–
	CPC 12450	<i>Chenopodium ficifolium</i>	Chenopodiaceae	South Korea	H.D. Shin	JX143574	JX143330	JX143084	JX142838	–
	CPC 15763	<i>Chenopodium</i> sp.	Chenopodiaceae	Mexico	Ma. de Jesús Yáñez-Morales	JX143575	JX143331	JX143085	JX142839	–
	CPC 15859	<i>Chenopodium</i> sp.	Chenopodiaceae	Mexico	Ma. de Jesús Yáñez-Morales	JX143576	JX143332	JX143086	JX142840	–
	CPC 15862	<i>Chenopodium</i> sp.	Chenopodiaceae	Mexico	Ma. de Jesús Yáñez-Morales	JX143577	JX143333	JX143087	JX142841	–
<i>C. samambaiae</i>	CPC 24673 = COAD 1090 ^{ET}	<i>Thelypteris dentata</i>	Thelypteridaceae	Brazil	R.W. Barreto	KT037514	KT037474	KT037596	KT037463	KT037555
	CPC 24727 = COAD 1427	<i>Pteris deflexa</i>	Pteridaceae	Brazil	E. Guatimosim	KT037508	KT037468	KT037590	KT037456	KT037549
	CBS 132618 = CPC 12062	<i>Zea mays</i>	Poaceae	South Africa	P. Caldwell	DQ185071	DQ185083	DQ185095	DQ185107	–
	CBS 115518 = CPC 5360	<i>Bidens frondosa</i>	Asteraceae	New Zealand	C.F. Hill	JX143681	JX143441	JX143195	JX142949	–
<i>Cercospora</i> sp. A	CPC 5438	<i>Salvia viscosa</i>	Lamiaceae	New Zealand	C.F. Hill	JX143682	JX143442	JX143196	JX142950	–
	CBS 115205 = CPC 5116	<i>Dichondra repens</i>	Convolvulaceae	New Zealand	C.F. Hill	JX143683	JX143443	JX143197	JX142951	–
<i>Cercospora</i> sp. F	CPC 5438	<i>Salvia viscosa</i>	Lamiaceae	New Zealand	C.F. Hill	JX143682	JX143442	JX143196	JX142950	–
<i>Cercospora</i> sp. G	CPC 5438	<i>Salvia viscosa</i>	Lamiaceae	New Zealand	C.F. Hill	JX143683	JX143443	JX143197	JX142951	–
<i>Cercospora</i> sp. H	CPC 11620	<i>Chamaelaucium uncinatum</i>	Myrtaceae	Argentina	S. Wolcan	JX143684	JX143444	JX143198	JX142952	–

<i>Cercospora</i> sp. I	CBS 114815 = CPC 5364 CBS 114816 = CPC 5363 CBS 114817 = CPC 5365 CBS 114818 = CPC 5362 CBS 115117	<i>Deutzia purpurascens</i> <i>Deutzia × rosea</i> <i>Fuchsia procumbens</i> <i>Deutzia crenata</i> <i>Archontophoenix cunilinghamiana</i>	New Zealand New Zealand New Zealand New Zealand New Zealand	C.F. Hill C.F. Hill C.F. Hill C.F. Hill C.F. Hill	JX143685 JX143686 JX143687 JX143688 JX143689	JX143445 JX143446 JX143447 JX143448 JX143449	JX143199 JX143200 JX143201 JX142955 JX142956 JX142957	
<i>Cercospora</i> sp. Q	CBS 115121 CBS 132597 = CPC 10615 CBS 132643 = CPC 10138 CPC 10616 CPC 5440 CBS 132656 = CPC 11536 CPC 10551 CPC 11539 CPC 10550 CBS 113997 = CPC 5325 CBS 115410 = CPC 5331 CBS 115411 = CPC 5332 CBS 115412 = CPC 5333 CBS 115536 = CPC 5329 CBS 115537 = CPC 5330 CBS 132663 = CPC 11636	<i>Gunnera tinctoria</i> <i>Coreopsis verticillata</i> <i>Ajuga multiflora</i> <i>Coreopsis verticillata</i> <i>Nicotiana</i> sp. <i>Acacia mangium</i> <i>Acacia mangium</i> <i>Acacia mangium</i> <i>Acacia mangium</i> <i>Cajanus cajan</i> <i>Cajanus cajan</i> <i>Cajanus cajan</i> <i>Cajanus cajan</i> <i>Cajanus cajan</i> <i>Dioscorea esculenta</i>	New Zealand New Zealand South Korea New Zealand New Zealand Thailand Thailand Thailand South Africa South Africa South Africa South Africa South Africa South Africa Papua New Guinea	C.F. Hill C.F. Hill H.D. Shin C.F. Hill C.F. Hill K. Pongpanich K. Pongpanich K. Pongpanich L. van Jaarsveld L. van Jaarsveld L. van Jaarsveld L. van Jaarsveld L. van Jaarsveld L. van Jaarsveld J. Peters & A.N. Jama	JX143690 JX143691 JX143692 JX143693 JX143694 JX143723 AY752140 JX143729 AY752172 AY752139 JX143717 JX143718 JX143719 JX143720 JX143721 JX143722 JX143725	JX143450 JX143451 JX143452 JX143453 JX143454 JX143482 AY752173 JX143488 AY752172 JX143476 JX143477 JX143478 JX143479 JX143233 JX143234 JX143481 JX143484	JX143204 JX143205 JX142960 JX142961 JX142962 JX142990 AY752232 JX143242 AY752200 JX143230 JX142984 JX142985 JX142987 JX142988 JX142989 JX142992	
		<i>Dioscorea rotundata</i>	Papua New Guinea	J. Peters & A.N. Jama	JX143724	JX143483	JX143237	JX142991
		<i>Dioscorea rotundata</i>	Papua New Guinea	J. Peters & A.N. Jama	JX143730	JX143489	JX143243	JX142997
		<i>Euphorbia</i> sp.	Mexico	Ma. de Jesús Yáñez-Morales	JX143727	JX143486	JX143240	JX142994
		<i>Euphorbia</i> sp.	Mexico	Ma. de Jesús Yáñez-Morales	JX143731	JX143490	JX143244	JX142998
		<i>Phaseolus vulgaris</i>	Mexico	Ma. de Jesús Yáñez-Morales	JX143726	JX143485	JX143239	JX142993
		<i>Taraxacum</i> sp.	Mexico	Ma. de Jesús Yáñez-Morales	JX143728	JX143487	JX143241	JX142995
		<i>Thelypteris dentata</i>	Brazil	R.W. Barreto	KT037510	KT037470	KT037592	KT037459
		<i>Macrothelypteris torresiana</i>	Brazil	R.W. Barreto	KT037511	KT037471	KT037593	KT037460
		<i>Cyathea delgadii</i>	Brazil	R.W. Barreto	KT037515	KT037475	KT037597	KT037464
		<i>Lygodium volubile</i>	Brazil	R.W. Barreto	KT037516	KT037476	KT037598	KT037465
	<i>C. zeae-maydis</i>	<i>Zea mays</i>	USA	B. Fleener	DQ185074	DQ185086	DQ185110	DQ185108
		<i>Zea mays</i>	USA	B. Fleener	DQ185072	DQ185084	DQ185096	DQ185108
		<i>Zea mays</i>	USA	B. Fleener	DQ185073	DQ185085	DQ185097	DQ185109
		<i>Zea mays</i>	USA	B. Fleener	DQ185075	DQ185087	DQ185099	DQ185111
	<i>C. zebrina</i>	<i>Hebe</i> sp.	New Zealand	C.F. Hill	JX143746	JX143508	JX143262	JX143016
		<i>Trifolium subterraneum</i>	Australia	M.J. Barbetti	JX143748	JX143510	JX143264	JX143018
<i>C. zeina</i>	<i>Lotus pedunculatus</i>	New Zealand	C.F. Hill	JX143754	JX143516	JX143204	JX143024	
	<i>Zea mays</i>	South Africa	P. Caldwell	DQ185081	DQ185093	DQ185105	DQ185117	
	<i>Zea mays</i>	South Africa	P. Caldwell	DQ185082	DQ185094	DQ185106	DQ185118	
<i>C. cf. zinniae</i>	<i>Zinnia elegans</i>	South Africa	H.D. Shin	JX143756	JX143518	JX143272	JX143026	
	–	Brazil	A.C. Alfenas	JX143757	JX143519	JX143273	JX143027	
	<i>Zinnia elegans</i>	Japan	J. Nishikawa	JX143758	JX143520	JX143274	JX143028	
	<i>Zinnia elegans</i>	Japan	S. Uematsu	JX143759	JX143521	JX143275	JX143029	
<i>Clypeosphaerella quasiparkii</i>	<i>Eucalyptus</i> sp.	Thailand	P. Suwannawong	KF901771	KF903113	KF903543	KF902128	
	CBS 123243 = CPC 154109 ^{ET} of <i>Myosphaerella quasiparkii</i>							
<i>Cl. sticheri</i>	<i>Sticherus bifidus</i>	Brazil	R.W. Barreto	KT037546	KT037505	KT037610	KT037588	
	<i>Sticherus bifidus</i>	Brazil	E. Guatimosim	KT037536	KT037495	KT037609	KT037577	
<i>Neoceratosperma alsophilae</i>	<i>Alsophila</i> sp.	Brazil	R.W. Barreto	KT037543	KT037502	KT037616	KT037585	
<i>N. cyatheae</i>	<i>Cyathea delgadii</i>	Brazil	R.W. Barreto	KT037539	KT037498	KT037624	KT037580	
	<i>Cyathea delgadii</i>	Brazil	R.W. Barreto	KT037541	KT037500	KT037625	KT037583	

Table 1 (cont.)

Species	Culture accession numbers ^{1,2}	Host/isolation source	Host family	Country	Collector	GenBank accession numbers ³				ITS	LSU
						<i>tef1</i>	<i>act</i>	<i>cmdA</i>			
<i>N. cyathea</i> (cont.)	CPC 24704 ^{ET}	<i>Cyathea delgadii</i>	Cyatheaceae	Brazil	E. Guatimosim	KT037545	KT037504	KT037626	–	KT037587	
	CPC 24712 = COAD 2002	<i>Cyathea delgadii</i>	Cyatheaceae	Brazil	E. Guatimosim	KT037527	KT037487	KT037617	–	KT037568	
	CPC 24724 = COAD 2007	<i>Cyathea delgadii</i>	Cyatheaceae	Brazil	E. Guatimosim	KT037529	KT037489	KT037618	–	KT037570	
	CPC 24726 = COAD 1426	<i>Cyathea delgadii</i>	Cyatheaceae	Brazil	E. Guatimosim	KT037531	KT037491	KT037619	–	KT037572	
	CPC 24728 = COAD 2008	<i>Cyathea delgadii</i>	Cyatheaceae	Brazil	E. Guatimosim	KT037532	KT037492	KT037620	–	KT037573	
	CPC 24732 = COAD 2011	<i>Cyathea delgadii</i>	Cyatheaceae	Brazil	E. Guatimosim	KT037535	KT037494	KT037622	–	KT037576	
	CPC 24744	<i>Cyathea delgadii</i>	Cyatheaceae	Brazil	E. Guatimosim	KT037537	KT037496	KT037623	–	KT037578	
	CPC 24729 = COAD 1428	<i>Cyathea delgadii</i>	Cyatheaceae	Brazil	E. Guatimosim	KT037533	KT037493	KT037621	–	KT037574	
	CBS 137998 = CPC 23465 ^{ET}	<i>Eucalyptus</i> sp.	Myrtaceae	Thailand	R. Cheewangkoon	KJ869153	–	–	–	KJ869210	
	CBS 119975 = CMW 23443 = MUCC 410 ^{ET} of <i>Mycosphaerella yunnanensis</i>	<i>Eucalyptus urophylla</i>	Myrtaceae	China	B. Dell	KF901628	KF903375	KF903515	–	KF901962	
<i>Paramycosphaerella aeorhyalinospomum</i>	CBS 125011 = CPC 14636 ^{ET}	<i>Eucalyptus tectifera</i>	Myrtaceae	Australia	B.A. Summrell	KF901605	KF903376	KF903576	KF902788	KF901930	
	CPC 24698 = COAD 1183 ^{ET}	<i>Blechnum serrulatum</i>	Blechnaceae	Brazil	R.W. Barreto	KT037544	KT037503	KT037611	–	KT037586	
<i>Pa. brachystegia</i>	CBS 136436 = CPC 21137, CPC 21136 ^{ET}	<i>Brachystegia</i> sp.	Fabaceae	Zimbabwe	J. Roux	KF777178	KT037506	KT037612	–	KF777230	
	CPC 24730 ^{ET} = COAD 2009	<i>Cyathea delgadii</i>	Cyatheaceae	Brazil	E. Guatimosim	KT037534	–	KT037613	–	KT037575	
<i>Pa. dicranopteridis</i>	BCRC FU30234 ^{ET} of <i>Zasmidium dicranopteridis</i>	<i>Dicranopteris linearis</i>	Gleicheniaceae	Taiwan	R. Kirschner	KJ201941	–	–	–	–	
	CPC 24743 ^{ET} = COAD 2016	<i>Dicranopteris flexuosa</i>	Gleicheniaceae	Brazil	P.B. Schwartzburd	KT037538	KT037497	KT037614	–	KT037579	
<i>Pa. gleicheniae</i>	RoKi 3613	<i>Dicranopteris linearis</i>	Gleicheniaceae	Taiwan	R. Kirschner	KJ201929	–	–	–	–	
	RoKi 3945	<i>Dicranopteris linearis</i>	Gleicheniaceae	Taiwan	R. Kirschner	KJ201930	–	–	–	–	
<i>Pa. intermedia</i>	CBS 114356 = CPC 10902	<i>Eucalyptus saligna</i>	Myrtaceae	New Zealand	M. Dick	KF901681	KF903142	KF903466	–	KF902026	
	CBS 114415 = CPC 10922	<i>Eucalyptus saligna</i>	Myrtaceae	New Zealand	M. Dick	KF901682	KF903143	KF903468	–	KF902027	
<i>Pa. irregularis</i>	CBS 123242 = CPC 15408 ^{ET}	<i>Eucalyptus globulus</i>	Myrtaceae	Thailand	R. Cheewangkoon	KF901769	KF903107	KF903542	–	KF902126	
	CBS 112301 = CPC 3747 ^{ET}	<i>Eucalyptus globulus</i>	Myrtaceae	Portugal	S. Denman	KF901688	KF903108	KF903453	–	KF902033	
<i>Pa. madeirensis</i>	CBS 112895 = CPC 3745 = CMW 14458	<i>Eucalyptus globulus</i>	Myrtaceae	Portugal	S. Denman	KF901675	KF903109	–	–	KF902017	
	CBS 110750 = CPC 822 = CMW 14778	<i>Eucalyptus grandis</i>	Myrtaceae	South Africa	G. Kemp	KF901709	KF903149	KF903404	–	KF902056	
<i>Pa. parkii</i>	CBS 110920 = CPC 935	<i>Eucalyptus botryoides</i>	Myrtaceae	Australia	A.J. Carnegie	KF901520	KF903145	KF903410	–	KF901842	
	CBS 110963 = CPC 4632	<i>Musa</i> sp.	Musaceae	South Africa	K. Surridge	KF901707	KF903146	KF903411	–	KF902054	
<i>Pa. nabiaceae</i>	CBS 110964 = CPC 4633	<i>Musa</i> sp.	Musaceae	South Africa	K. Surridge	KF901708	KF903147	KF903412	–	KF902055	
	CBS 110981 = CPC 1073	<i>Eucalyptus</i> sp.	Myrtaceae	Tanzania	M.J. Wingfield	KF901749	KF903148	KF903417	–	KF902103	
<i>Pa. parkii</i>	CBS 125010 = CPC 12748 ^{ET} of <i>Zasmidium nabiaceae</i>	<i>Eucalyptus</i> sp.	Myrtaceae	Australia	A.J. Carnegie	KF901608	KF903391	KF903575	–	KF901933	
	CBS 387.92 = CPC 353 ^{ET} of <i>Zasmidium parkii</i>	<i>Eucalyptus grandis</i>	Myrtaceae	Brazil	M.J. Wingfield	KF901785	KF903392	KF903585	–	KF902143	
<i>Pa. pseudomarksii</i>	CBS 123241 = CPC 15410 ^{ET} of <i>Mycosphaerella pseudomarksii</i>	<i>Eucalyptus</i> sp.	Myrtaceae	Thailand	R. Cheewangkoon	KF901770	KF903111	KF903541	–	KF902127	
	CPC 24720 = COAD 1422 ^{ET}	<i>Sticherus penniger</i>	Gleicheniaceae	Brazil	E. Guatimosim	KT037528	KT037488	KT037615	–	KT037569	
<i>Pa. vietnamensis</i>	CBS 119974 = CMW 23441 = MUCC 66 ^{ET} of <i>Mycosphaerella vietnamensis</i>	<i>Eucalyptus grandis</i> hybrid	Myrtaceae	Vietnam	T.I. Burgess	KF901809	KF903114	KF903514	–	KF902171	
	CBS 129524 = CPC 18480 ^{ET}	<i>Eucalyptus saligna</i>	Myrtaceae	Brazil	P.W. Crous & A.C. Alfenas	KF901613	KF903153	KF903445	–	KF901938	
<i>Passalora eucalypti</i>	CBS 111318 = CPC 1457 ^{ET}	<i>Eucalyptus leptophlebia</i>	Myrtaceae	Brazil	P.W. Crous, A.C. Alfenas, R. Alfenas & O.L. Pereira	KF901614	KF903155	KF903580	–	KF901939	
	CBS 112970 = CPC 1228 ^{ET}	<i>Eucalyptus globulus</i>	Myrtaceae	Zambia	T. Coutinho	KF901811	KF903157	KF903458	–	KF902175	
<i>Phaeophloeospora eugeniae</i>	CBS 112971 = CPC 1227 ^{ET}	<i>Eucalyptus globulus</i>	Myrtaceae	Zambia	T. Coutinho	KF901810	KF903156	KF903459	–	KF902174	
	CPC 15143	<i>Eugenia uniflora</i>	Myrtaceae	Brazil	A.C. Alfenas	KF901615	KF903160	KF903674	–	KF901940	
<i>Ph. gregaria</i>	CPC 15159	<i>Eugenia uniflora</i>	Myrtaceae	Brazil	A.C. Alfenas	KF901742	KF903159	KF903675	–	KF902095	
	CBS 110501	<i>Eucalyptus globulus</i>	Myrtaceae	Australia	A. Maxwell	KF901524	KF903161	KF903396	–	KF901846	
	CBS 111166 = CPC 1224	<i>Eucalyptus cladocalyx</i>	Myrtaceae	South Africa	A.R. Wood	KF901710	KF903162	KF903433	–	KF902057	

<i>Ph. gregaria</i> (cont.)	CBS 111167 = CPC 1225	<i>Eucalyptus cladocalyx</i>	Myrtaceae	South Africa	A.R. Wood	KF901711	KF903163	KF903434	–	KF902058
	CBS 111519 = CPC 1191	<i>Eucalyptus</i> sp.	Myrtaceae	South Africa	P.W. Crous	KF901712	KF903164	KF903448	–	KF902059
	CBS 114662 = CPC 1193 ^{ET}	<i>Eucalyptus</i> sp.	Myrtaceae	South Africa	P.W. Crous	KF901713	KF903165	KF903470	–	KF902060
<i>Ph. hymenocallidicola</i>	CBS 139912 = CPC 25014 ^{ET}	unknown fern	Polypodiaceae	Thailand	P.W. Crous	–	–	–	–	KR476772
<i>Ph. hymenocallidis</i>	CBS 139911 = CPC 25018 ^{ET}	unknown fern	Polypodiaceae	Thailand	P.W. Crous	KR476740	–	–	–	KR476773
<i>Ph. pteridivora</i>	CPC 24683 = COAD 1182 ^{ET}	<i>Serpocaulon triseriale</i>	Polypodiaceae	Brazil	R.W. Barreto	KT037547	KT037499	KT037631	–	KT037582
<i>Ph. scytalidii</i>	CBS 118493 = CPC 10998 ^{ET}	<i>Eucalyptus urophylla</i>	Myrtaceae	Colombia	M.J. Wingfield	KF901631	KF903493	KF903493	–	KF901966
	CBS 516.93 = CPC 653	<i>Eucalyptus globulus</i>	Myrtaceae	Brazil	P.A. Ferreira	KF901616	KF903166	KF903588	–	KF901941
<i>Ph. stonei</i>	CBS 120830 = CPC 13330 ^{ET}	<i>Eucalyptus</i> sp.	Myrtaceae	Australia	P.W. Crous & J. Stone	KF901525	KF903168	KF903645	–	KF901847
<i>Ph. stramentii</i>	CBS 118909 = CPC 11545 ^{ET}	leaf litter of <i>Eucalyptus</i> sp.	Myrtaceae	Brazil	A.C. Alfenas	KF901617	KF903169	KF903506	–	KF901942
<i>Pseudocercospora abacopteridicola</i>	CPC 24709 = COAD 2009	<i>Adiantum</i> sp.	Pteridaceae	Brazil	E. Guatimosim	KT037518	KT037478	KT037600	–	KT037559
<i>Ps. angolensis</i>	CBS 149.53	<i>Citrus sinensis</i>	Rutaceae	Angola	T. de Carvalho & O. Mendes	JQ324975	JQ324988	JQ325011	–	JQ324941
<i>Ps. araliiae</i>	CBS 112933 = CPC 4118	<i>Citrus</i> sp.	Rutaceae	Zimbabwe	M.C. Pretorius	GU269836	GU384548	JQ325010	–	–
	CPC 10154	<i>Aralia elata</i>	Araliaceae	South Korea	H.D. Shin	GU269652	GU384370	GU320360	–	GU253701
	MUCC 873 ^{ET}	<i>Aralia elata</i>	Araliaceae	Japan	T. Kobayashi & C. Nakashima	GU269653	GU384371	GU320361	–	GU253702
<i>Ps. assamensis</i>	CBS 122467 ^{ET}	<i>Musa cultivar</i>	Musaceae	India	I. Buddenhagen	GU269656	GU384374	GU320364	–	GU253705
<i>Ps. atromarginalis</i>	CBS 132010 = CPC 11372	<i>Solanum nigrum</i>	Solanaceae	South Korea	H.D. Shin	GU269657	GU384375	JX902130	–	GU214671
<i>Ps. balsaminiae</i>	CBS 131882 = CPC 10044	<i>Impatiens textori</i>	Balsaminaceae	South Korea	H.D. Shin	GU269660	GU384379	GU320367	–	GU253708
<i>Ps. basiramifera</i>	CMW 5148	<i>Eucalyptus pellita</i>	Myrtaceae	Thailand	AF309595	DQ211677	DQ147607	–	–	DQ204761
<i>Ps. basitruncata</i>	CBS 114664 = CPC 1202	<i>Eucalyptus grandis</i>	Myrtaceae	Colombia	M.J. Wingfield	DQ211675	DQ147622	–	–	DQ253710
	CBS 112280 = CMW 14785	<i>Eucalyptus grandis</i>	Myrtaceae	Colombia	M.J. Wingfield	DQ211676	DQ147621	–	–	DQ204760
<i>Ps. brackenicola</i>	CPC 24695 ^{ET} = COAD 1991	<i>Pteridium arachnoidium</i>	Dennstaedtiaceae	Brazil	R.W. Barreto	KT037524	KT037484	KT037606	–	KT037565
<i>Ps. chengtuentensis</i>	CBS 131924 = CPC 10696	<i>Lyium chinense</i>	Solanaceae	South Korea	H.D. Shin	GU269673	GU384390	GU320379	–	JQ324942
<i>Ps. contraria</i>	CBS 132108 = CPC 14714	<i>Dioscorea quinqueloba</i>	Dioscoreaceae	South Korea	H.D. Shin	GU269677	GU384394	GU320385	–	JQ324945
<i>Ps. cordiana</i>	CBS 114685 = CPC 2552 ^{ET}	<i>Cordia goeldiana</i>	Boraginaceae	Brazil	P.W. Crous & R.L. Benchimol	GU269681	GU384398	GU320387	–	GU214472
<i>Ps. crocea</i>	CBS 126004 = CPC 11668 ^{ET}	<i>Pilea hamaoi</i>	Urticaceae	South Korea	H.D. Shin	GU269792	GU384502	GU320493	–	JQ324947
<i>Ps. cruenta</i>	CBS 132021 = CPC 10846	<i>Vigna</i> sp.	Fabaceae	Trinidad	H. Booker	GU269688	GU384404	JQ325012	–	GU214673
<i>Ps. cyathicicola</i>	CBS 129520 = CPC 17047 = CPC 17048 ^{ET}	<i>Cyathea australis</i>	Cyatheaceae	Australia	P.W. Crous & R.G. Shivas	JF951139	KT072761	KT072760	–	JF951159
<i>Ps. cymbidiiicola</i>	CBS 115132 ^{ET}	<i>Cymbidium</i> sp.	Orchidaceae	New Zealand	C.F. Hill	GU269692	GU384408	GU320397	–	GU253733
<i>Ps. dendrobii</i>	MUCC 596	<i>Dendrobium</i> sp.	Orchidaceae	Japan	C. Nakashima & K. Motohashi	GU269696	GU384412	GU320401	–	GU253737
<i>Ps. dianellae</i>	CBS 117746	<i>Dianella caerulea</i>	Liliaceae	New Zealand	C.F. Hill	GU269695	GU384411	GU320400	–	GU253736
<i>Ps. eucalyptorum</i>	CBS 116371 = CPC 10507	<i>Eucalyptus nitens</i>	Myrtaceae	New Zealand	P.W. Crous	GU269687	JQ324989	GU320393	–	JQ324990
	CBS 132309 = CPC 12568	<i>Eucalyptus nitens</i>	Myrtaceae	Australia	C. Mohammed	GU269796	GU384506	GU320497	–	GU253814
	CBS 132032 = CPC 12802	<i>Eucalyptus globulus</i>	Myrtaceae	Portugal	A. Phillips	JQ324976	JQ324990	GU320466	–	GU253789
	CBS 132035 = CPC 13769	<i>Eucalyptus punctata</i>	Myrtaceae	South Africa	P.W. Crous	GU269659	GU384378	GU320366	–	GU253707
	CBS 132114 = CPC 13816	<i>Eucalyptus glaucescens</i>	Myrtaceae	United Kingdom	S. Denman	GU269801	JQ324992	GU320504	–	GU253819
<i>Ps. eupatoriella</i>	CBS 113372	<i>Chromolaena odorata</i>	Asteraceae	Jamaica	M.J. Morris	GU269704	GU384420	GU320408	–	GU253743
<i>Ps. fori</i>	CBS 132113 = CPC 14880	<i>Eucalyptus</i> sp.	Myrtaceae	South Africa	P.W. Crous	GU269806	GU384517	GU320506	–	GU253824
<i>Ps. fuliginea</i>	CBS 132017 = CPC 12296	<i>Lycopodium</i> sp.	Myrtaceae	Thailand	Z. Mersha	GU269711	GU384427	GU320415	–	JQ324953
<i>Ps. haiwensis</i>	CBS 131584 = CPC 14084 ^{ET}	<i>Eucalyptus</i> sp.	Myrtaceae	China	X. Zhou	GU269803	GU384514	GU320506	–	GU253821
<i>Ps. humuli</i>	MUCC 742 ^{ET}	<i>Humulus lupulus</i> var. <i>lupulus</i>	Cannabaceae	Japan	C. Nakashima & I. Araki	GU269725	GU384439	GU320428	–	GU253758
<i>Ps. humuli-japonici</i>	CPC 11462 ^{ET}	<i>Plectranthus</i> sp.	Lamiaceae	Republic of Korea	H.D. Shin	JX901784	JX902139	–	–	JX901892
<i>Ps. humulicola</i>	CBS 131883 = CPC 10049	<i>Humulus scandens</i>	Cannabaceae	South Korea	H.D. Shin	GU269724	JQ324996	JQ325018	–	JQ324955
<i>Ps. indonesiana</i>	CBS 122474	<i>Musa cultivar</i>	Musaceae	Indonesia	I.W. Buddenhagen	EU514283	JQ324997	JQ325019	–	JQ324957
<i>Ps. jussiaeae</i>	CBS 132117 = CPC 14625	<i>Ludwigia prostrata</i>	Onagraceae	South Korea	H.D. Shin	JQ324977	JQ324998	JQ325020	–	JQ324958
<i>Ps. kaki</i>	MUCC 900	<i>Diospyros kaki</i>	Ebenaceae	Japan	S. Uematsu & C. Nakashima	GU269729	GU384442	GU320431	–	GU253761
<i>Ps. lilacis</i>	CBS 132031 = CPC 12767	<i>Ligustrum japonicum</i>	Oleaceae	USA	C. Hodges	GU269737	GU384449	GU320439	–	GU253767
<i>Ps. lonicericola</i>	MUCC 889 ^{ET}	<i>Lonicera gracilipes</i> var. <i>glabra</i>	Caprifoliaceae	Japan	T. Kobayashi	GU269736	JQ324999	GU320438	–	GU253766
<i>Ps. luzardii</i>	CPC 2556 ^{ET}	<i>Hancornia speciosa</i>	Apocynaceae	Brazil	A.C. Alfenas	GU269738	GU384450	GU320440	–	GU214477
<i>Ps. lygodiiicola</i>	CPC 25755 = COAD 1745	<i>Lygodium volubile</i>	Lythraceae	Brazil	R.W. Barreto	KT037526	KT037486	KT037608	–	KT037567
<i>Ps. lythri</i>	CBS 132115 = CPC 14588 ^{ET}	<i>Lythrum salicaria</i>	Lythraceae	South Korea	H.D. Shin	GU269742	GU384454	GU320444	–	GU253771
	MUCC 865	<i>Lythrum salicaria</i>	Lythraceae	Japan	I. Araki & M. Harada	GU269743	GU384455	GU320445	–	GU253772
<i>Ps. macrospora</i>	CBS 114696 = CPC 2553 ^{ET}	<i>Bertholletia excelsa</i>	Legythraceae	Brazil	P.W. Crous & R.L. Benchimol	GU269745	GU384457	GU320447	–	GU214478
<i>Ps. mazandaransensis</i>	CCTU 1102 = CBS 136115 ^{ET}	<i>Nerium oleander</i>	Oleaceae	Iran	M. Bakhshi	KM452854	KM452876	KM452831	–	–

Table 1 (cont.)

Species	Culture accession numbers ^{1,2}	Host/isolation source	Host family	Country	Collector	ITS	GenBank accession numbers ³	LSU
							tef1 act cmdA	
<i>Ps. mazandaranensis</i> (cont.)	CCTU 1146	<i>Nerium oleander</i>	Oleaceae	Iran	M. Bakhshi	KM452855	KM452877	–
<i>Ps. metrosideri</i>	CBS 118795 ^{ET}	<i>Metrosideros collina</i>	Myrtaceae	New Zealand	C.F. Hill	GU269746	GU384458	GU320448
<i>Ps. natalensis</i>	CBS 111069 = CPC 1263	<i>Eucalyptus nitens</i>	Myrtaceae	South Africa	T. Coutinho	DQ303077	JK325000	DQ147620
<i>Ps. nephrolepidis</i>	CBS 11912 ^{1ET}	<i>Nephrolepis auriculata</i>	Oleaceae	Taiwan	R. Kirschner	GU269751	GU384462	GU320453
<i>Ps. nogalesii</i>	CBS 115022	<i>Chamaecytisus proliferus</i>	Fabaceae	New Zealand	C.F. Hill	GU269752	GU384463	GU320454
<i>Ps. norchiensis</i>	CBS 120738 ^{ET}	<i>Eucalyptus</i> sp.	Myrtaceae	Italy	W. Gams	GU269753	GU384464	GU320455
	CCTU 1009	<i>Rubus</i> sp.	Rosaceae	Iran	M. Bakhshi	KM452856	KM452878	KM452833
	CCTU 1019	<i>Rubus</i> sp.	Rosaceae	Iran	M. Bakhshi	KM452857	KM452879	KM452834
	CCTU 1032	<i>Rubus</i> sp.	Rosaceae	Iran	M. Bakhshi	KM452858	KM452880	KM452835
<i>Ps. ocimi-basilici</i>	CPC 10283 ^{ET}	<i>Ocimum basilicum</i>	Lamiaceae	Mexico	M.E. Palm	GU269754	GU384465	GU320456
<i>Ps. oenotherae</i>	CBS 131885 = CPC 10290	<i>Oenothera odorata</i>	Onagraceae	South Korea	H.D. Shin	GU269856	GU384567	GU320559
<i>Ps. palaeobrunnea</i>	CBS 124771 = CPC 13387 ^{ET}	<i>Syzygium</i> sp.	Myrtaceae	Australia	P.W. Crous	GQ303288	GU384460	GU320500
<i>Ps. pallida</i>	CBS 131889 = CPC 10776	<i>Campsis grandiflora</i>	Bignoniaceae	South Korea	H.D. Shin	GU269758	GU384469	GU320459
<i>Ps. pancrati</i>	CBS 137.94	–	–	Cuba	R.F. Castaneda	DQ267602	DQ211680	DQ147606
<i>Ps. paraguayensis</i>	CBS 111286 = CPC 1459	<i>Eucalyptus nitens</i>	Myrtaceae	Brazil	P.W. Crous	GU269759	GU384470	GU320460
<i>Ps. paranaensis</i>	CPC 24680 ^{ET} = COAD 1987	<i>Cyathea atrovirens</i>	Cyatheaceae	Brazil	R.W. Barreto	KT037522	KT037482	KT037604
	COAD 1180	<i>Cyathea atrovirens</i>	Cyatheaceae	Brazil	R.W. Barreto	KT037523	KT037483	KT037605
<i>Ps. parapseudarthriae</i>	CBS 137996 = CPC 23449 ^{ET}	<i>Pseudarthria hookeri</i>	Leguminosae	South Africa	A.R. Wood	KJ869151	KJ869238	KJ869208
<i>Ps. pouzolziae</i>	CBS 122280	<i>Gonostegia hirta</i>	Urticaceae	Taiwan	R. Kirschner	GU269761	GU384472	GU320462
<i>Ps. profusa</i>	CPC 10042	<i>Acalypha australis</i>	Euphorbiaceae	South Korea	H.D. Shin	GU269762	GU384497	GU320488
	CBS 132306 = CPC 10055	<i>Acalypha australis</i>	Euphorbiaceae	South Korea	H.D. Shin	GU269762	GU384473	GU320463
<i>Ps. proteae</i>	CBS 131587 = CPC 15217 ^{ET}	<i>Protea mundii</i>	Proteaceae	South Africa	F. Roets	GU269808	GU384519	GU320511
<i>Ps. prunicola</i>	CBS 132107 = CPC 14511	<i>Prunus yedoensis</i>	Rosaceae	South Korea	H.D. Shin	GU269676	GU384393	GU320382
<i>Ps. punctata</i>	CBS 132116 = CPC 14734 ^{ET}	<i>Syzygium</i> sp.	Myrtaceae	Madagascar	P.W. Crous	GU269765	GU384477	GU320468
<i>Ps. puniceae</i>	CBS 136111 = CCTU 1125	<i>Punica granatum</i>	Lythraceae	Iran	M. Bakhshi	KM452859	KM452881	KM452836
	CCTU 1169	<i>Punica granatum</i>	Lythraceae	Iran	M. Bakhshi	KM452860	KM452882	KM452837
<i>Ps. purpurea</i>	CBS 114163 = CPC 1664	<i>Persea americana</i>	Lauraceae	Mexico	P.W. Crous	GU269783	GU384494	GU320486
<i>Ps. pyracanthae</i>	MUCC 892	<i>Pyracantha angustifolia</i>	Rosaceae	Japan	T. Kobayashi & C. Nakashima	GU269767	GU384479	GU320470
<i>Ps. rhabdothamni</i>	CBS 114872 ^{ET}	<i>Rhabdothamnium solandri</i>	Gesneriaceae	New Zealand	M. Fletcher	GU269768	GU384480	GU320471
<i>Ps. rhannellae</i>	CBS 131590 = CPC 12500 ^{ET}	<i>Rhannella franguloides</i>	Rhamnaceae	South Korea	H.D. Shin	GU269795	GU384505	GU320496
<i>Ps. rumohrae</i>	CBS 117747	<i>Marattia salicina</i>	Marattiaceae	New Zealand	C.F. Hill	GU269774	GU384486	GU320477
<i>Ps. rubi</i>	MUCC 875	<i>Rubus allegheniensis</i>	Rosaceae	Japan	T. Kobayashi & C. Nakashima	GU269773	GU384485	GU320476
<i>Ps. schizobolii</i>	CBS 120029 = CPC 12962 ^{ET}	<i>Schizobolium parahyba</i>	Fabaceae	Ecuador	M.J. Wingfield	KF251322	KF253269	KF253628
<i>Ps. serpocaulonicola</i>	CPC 25077 = COAD 1866 ^{ET}	<i>Serpocaulon triseriale</i>	Polypodiaceae	Brazil	R.W. Barreto	KT037525	KT037485	KT037607
<i>Ps. sophoricola</i>	CBS 136020 = CCTU 1037 ^{ET}	<i>Sophora alopecuroides</i>	Fabaceae	Iran	M. Bakhshi	KM452861	KM452883	KM452838
<i>Ps. sordida</i>	MUCC 913	<i>Campsis radicans</i>	Bignoniaceae	Japan	C. Nakashima & E. Imaizumi	GU269777	GU384488	GU320480
<i>Pseudocercospora</i> sp.	CBS 110998 = CPC 1054	<i>Eucalyptus grandis</i>	Myrtaceae	South Africa	M.J. Wingfield	GU269778	GU384489	GU320481
<i>Pseudocercospora</i> sp. A	CBS 136113 = CCTU 1165	<i>Phaseolus vulgaris</i>	Fabaceae	Iran	M. Bakhshi	KM452863	KM452885	KM452840
	CCTU 1166	<i>Phaseolus vulgaris</i>	Fabaceae	Iran	M. Bakhshi	KM452864	KM452886	KM452841
<i>Pseudocercospora</i> sp. B	CCTU 1066	<i>Phaseolus vulgaris</i>	Ebenaceae	Iran	M. Bakhshi	KM452865	KM452887	KM452842
	CCTU 1191	<i>Diospyros lotus</i>	Ebenaceae	Iran	M. Bakhshi	KM452866	KM452888	KM452843
	CBS 136114 = CCTU 1206	<i>Diospyros lotus</i>	Ebenaceae	Iran	M. Bakhshi	KM452867	KM452889	KM452844
<i>Ps. thelypteridis</i>	CPC 24676 ^{ET} = COAD 1985	<i>Thelypteris</i> sp.	Thelypteridaceae	Brazil	R.W. Barreto	KT037521	KT037481	KT037603
<i>Ps. trichogena</i>	CPC 24670 = COAD 1088 ^{ET}	<i>Deparia petersenii</i>	Athyriaceae	Brazil	R.W. Barreto	KT037520	KT037480	KT037602
	CPC 24664 = COAD 1087	<i>Macrothelypteris torresiana</i>	Thelypteridaceae	Brazil	R.W. Barreto	KT037519	KT037479	KT037601
<i>Ps. udagawana</i>	CBS 131931 = CPC 10799	<i>Hovenia dulcis</i>	Rhamnaceae	South Korea	H.D. Shin	GU269824	GU384537	GU320527
<i>Pseudoramichloridium henryi</i>	CBS 124775 = CPC 13121 ^{ET}	<i>Corymbia henryi</i>	Myrtaceae	Australia	A.J. Carnegie	KF901535	KF903227	KF903559
	CPC 13122	<i>Corymbia henryi</i>	Myrtaceae	Australia	A.J. Carnegie	KF901533	KF903226	KF903639
<i>Ramularia endophylla</i>	CBS 113265 ^{ET}	dead leaf of <i>Quercus robur</i>	Fagaceae	Netherlands	G. Verkleij	KF901725	KF903240	KF903461
<i>R. eucalypti</i>	CBS 120726 = CPC 13043 ^{ET}	<i>Eucalyptus grandiflora</i>	Myrtaceae	Italy	W. Gams	KF901666	KF903241	KF903525
<i>Septoria eucalyptorum</i>	CBS 118505 = CPC 11282 ^{ET}	leaf litter of <i>Eucalyptus</i> sp.	Myrtaceae	India	W. Gams & M. Arzaniou	KF901651	KF903265	KF903501

<i>Sonderhenia eucalypticola</i>	CPC 11251	<i>Eucalyptus globulus</i>	Myrtaceae	Spain	M.J. Wingfield	KF901746	KF903266	KF903596	–	KF902099
	CPC 11252	<i>Eucalyptus globulus</i>	Myrtaceae	Spain	M.J. Wingfield	KF901747	KF903268	KF903597	–	KF902100
	CBS 112502 = CPC 3749	<i>Eucalyptus</i> sp.	Myrtaceae	Spain	P.W. Crous	KF901677	KF903267	KF903454	–	KF902019
	CBS 118910 = CPC 12226 ^{ET}	<i>Eucalyptus</i> sp.	Myrtaceae	France	P.W. Crous	KF901649	KF903269	KF903507	–	KF901988
	CBS 120061 = CPC 13055 ^{ET}	<i>Eucalyptus robusta</i>	Myrtaceae	Australia	B.A. Summerell	KF901552	KF903270	KF903517	KF902693	KF901874
	CPC 24691 ^{ET} = COAD 1990	<i>Diplazium</i> sp.	Athyriaceae	Brazil	R.W. Barreto	KT037542	KT037501	KT037627	–	KT037584
	CBS 120735 = CPC 13378 ^{ET}	<i>Eucalyptus camaldulensis</i> × <i>urophylla</i>	Myrtaceae	Venezuela	M.J. Wingfield	KF901808	KF903374	KF903528	–	KF902170
<i>Zasmidium cellare</i>	CBS 146.36 ^{ET}	wine cellar	–	–	H. Schanderl	EU041821	–	–	–	EU041878
<i>Z. citri</i>	CBS 116366 = CPC 10522 = CMW 11730	<i>Acacia mangium</i>	Fabaceae	Thailand	K. Pongpanich	KF901780	KF903386	–	–	KF902138
<i>Z. cyatheae</i>	CPC 15291	<i>Citrus</i> sp.	Rutaceae	USA	–	KF901793	KF903382	KF903676	–	KF902152
<i>Z. eucalyptigenum</i>	CBS 138860 = CPC 2425 ^{1ET}	<i>Cyathea delgadoii</i>	Cyatheaceae	Brazil	E. Guatimosim	KT037530	KT037490	–	–	KT037571
<i>Z. eucalyptorum</i>	CBS 118500 = CPC 11174 ^{ET}	<i>Eucalyptus urophylla</i>	Myrtaceae	Mozambique	M.J. Wingfield	KP004458	–	KT037630	–	KP004486
<i>Z. pseudoparkii</i>	CBS 110999 = CPC 1087 ^{ET}	<i>Eucalyptus grandis</i>	Myrtaceae	Indonesia	M.J. Wingfield	KF901652	KF903101	KF903495	–	–
	CBS 110988 = CPC 1090	<i>Eucalyptus grandis</i>	Myrtaceae	Colombia	M.J. Wingfield	KF901642	KF903273	KF903419	–	KF901977
	CBS 111049 = CPC 1089	<i>Eucalyptus grandis</i>	Myrtaceae	Colombia	M.J. Wingfield	KF901640	KF903271	KF903418	–	KF901975
	CPC 24679 = COAD 1178	<i>Blechnum serrulatum</i>	Myrtaceae	Colombia	M.J. Wingfield	KF901641	KF903272	KF903426	–	KF901976
<i>Zasmidium</i> sp.	CBS 111185 = CPC 1300 ^{ET}	<i>Eucalyptus grandis</i>	Blechnaceae	Brazil	R.W. Barreto	KT037540	–	KT037628	–	KT037581
<i>Z. xenoparkii</i>			Myrtaceae	Indonesia	M.J. Wingfield	KF901663	KF903274	KF903438	–	KF902002

¹ BCRC: Bioresource Collection and Research Center, Hsinchu, Taiwan; CBS: CBS-KNAW Fungal Biodiversity Centre, Utrecht, The Netherlands; CCTU: Culture Collection of the Forestry and Agricultural Biotechnology Institute, Pretoria, South Africa; COAD: Coleção Octávio de Almeida Drumond, Vitória, Minas Gerais, Brazil; CPC: Culture collection of Pedro Crous, housed at CBS; MUCC: Culture Collection, Laboratory of Plant Pathology, Mie University, Tsu, Mie Prefecture, Japan; Roki: R. Kirschner, dried specimen deposited in National Museum of Natural Science, Taichung, Taiwan; WAC: Department of Agriculture Western Australia Plant Pathogen Collection, Perth, Australia.

² ET: ex-type; EET: ex-epitype.

³ ITS: internal transcribed spacers and intervening 5.8S rDNA; *tef1*: translation elongation factor 1alpha; *act*: actin; *cmdA*: calmodulin; LSU: 28S rRNA gene.

subsequently diluted 50–100 times in preparation for further DNA amplification reactions. Four partial nuclear genes were initially targeted for PCR amplification and sequencing, namely 28S nrRNA gene (LSU), internal transcribed spacer regions and intervening 5.8S nrRNA gene (ITS) of the nrDNA operon, actin (*act*), and translation elongation factor 1- α (*tef1*). Additionally, for the *Cercospora* strains, a part of the calmodulin gene (*cmdA*) was amplified. The primers employed are listed in Table 2. The PCR amplifications were performed in a total volume of 12.5 μ L solution containing 10–20 ng of template DNA, 1 \times PCR buffer, 0.63 μ L DMSO (99.9 %), 1.5 mM MgCl₂, 0.5 μ M of each primer, 0.25 mM of each dNTP, 1.0 U BioTaq[®] DNA polymerase (Bioline GmbH Luckenwalde, Germany). PCR conditions for ITS and LSU were set as follows: an initial denaturation (95 °C; 5 min), 35 cycles amplification (95 °C, 30 s; annealing (Table 2), 30 s; 72 °C, 1 min) and a final extension (72 °C, 6 min). PCR conditions for *tef1* were set as an initial denaturation (94 °C, 5 min), 45 cycles amplification (94 °C, 45 s; annealing (Table 2), 30 s; 72 °C, 90 s) and a final extension (72 °C, 6 min). For *cmdA*, the PCR conditions were set as an initial denaturation (94 °C, 5 min) 45 cycles amplification (94 °C, 24 s; annealing (Table 2) 40 s; 72 °C, 40 s) and a final extension (72 °C, 5 min). For *act*, a touchdown protocol was used and set as an initial denaturation (94 °C, 5 min), 13 amplification cycles (94 °C, 30 s; 65 °C, 30 s; 72 °C, 30 s); 25 amplification cycles (94 °C, 30 s; 56 °C, 30 s; 72 °C, 30 s) and a final extension (72 °C, 7 min). The resulting fragments were sequenced using the PCR primers and the BigDye[®] Terminator Cycle Sequencing Kit v. 3.1 (Applied Biosystems[™], Foster City, CA, USA) following the protocol of the manufacturer. DNA sequencing amplicons were purified through Sephadex[®] G-50 Superfine columns (Sigma Aldrich, St. Louis, MO) in MultiScreen HV plates (Millipore, Billerica, MA). Purified sequence reactions were run on an ABI Prism 3730xl DNA Analyzer (Life Technologies, Carlsbad, CA, USA).

DNA sequence data were analysed in MEGA (Molecular Evolutionary Genetics Analysis) v. 6.0 (Tamura et al. 2013). Consensus sequences were generated and imported into MEGA v. 6.0 for initial alignment and the construction of sequence datasets. Initially, sequences obtained from the datasets of Schoch et al. (2009, TreeBASE S10245), Groenewald et al. (2013, TreeBASE S13645), Crous et al. (2013a, TreeBASE S12805), from GenBank (www.ncbi.nlm.nih.gov) and the novel sequences generated during this study, were aligned using MAFFT v. 7 (<http://mafft.cbrc.jp/alignment/server/index.html>; Katoh & Standley 2013) and whenever necessary, manually improved in MEGA v. 6.0. After a preliminary analysis, the datasets were trimmed down to Brazilian isolates and the direct neighbours.

Phylogenetic analyses

Appropriate gene models were selected using MrModeltest v. 2.3 (Nylander 2004) and applied to each gene partition. Based on the results of MrModeltest, a Bayesian phylogenetic analysis was performed with MrBayes v. 3.2.1 (Ronquist et al. 2012) applying different substitution models for each locus as listed in Table 3. *Sphaerulina cercidis* (CBS 118910) served as outgroup for the phylogenetic analyses of *Cercospora* species, *Passalora eucalypti* (CBS 111318) for *Pseudocercospora* species and *Staninwardia suttonii* (CBS 120061) served as outgroup for the mycosphaerella-like species. Posterior probabilities were determined by Markov Chain Monte Carlo sampling (MCMC) in MrBayes v. 3.2.1. Six simultaneous Markov chains were run for 10 000 000 generations and trees were sampled every 100th generation, until convergence (stopval = 0.01) was reached. A heating parameter ('temp') of 0.30 was used for the *Cercospora* analysis and 0.15 for the *Pseudocercospora* and mycosphaerella-like taxa analyses. Sequences derived in this study were lodged in GenBank, the alignments and trees in

Table 2 Details of primers used in this study for the PCR amplification and sequencing of different genes.

Gene ¹	Primer name	Sequence 5'→3'	Annealing temperature (°C)	Orientation	Reference
<i>act</i>	ACT-512F	ATG TGC AAG GCC GGT TTC GC	65→56	Forward	Carbon & Kohn 1999
	ACT-783 R	TAC GAG TCC TTC TGG CCC AT	65→56	Reverse	Carbon & Kohn 1999
<i>cmdA</i>	CAL-228F	GAG TTC AAG GAG GCC TTC TCC C	58	Forward	Carbon & Kohn 1999
	CAL-737R	CAT CTT TCT GGC CAT CAT GG	58	Reverse	Carbon & Kohn 1999
ITS	ITS5	GGA AGT AAA AGT CGT AAC AAG G	52	Forward	White et al. 1990
	ITS4	TCC TCC GCT TAT TGA TAT GC	52	Reverse	White et al. 1990
LSU	LR0R	ACC CGC TGA ACT TAA GC	52	Forward	Vilgalys & Hester 1990
	LR5	TCC TGA GGG AAA CTT CG	52	Reverse	Vilgalys & Hester 1990
<i>tef1</i>	EF-728F	CAT CGA GAA GTT CGA GAA GG	52	Forward	Carbon & Kohn 1999
	EF2Fd	GAT CTA CCA GTG CGG TGG	52	Forward	Groenewald et al. 2013
	EF-2	GGA RGT ACC AGT SAT CAT GTT	52	Reverse	O'Donnell et al. 1998

¹ *act*: actin gene; *cmdA*: calmodulin gene; ITS: internal transcribed spacer regions and intervening 5.8S nrRNA gene of the nrDNA operon; LSU: 28S nrRNA gene; *tef1*: translation elongation factor 1- α .

Table 3 Substitution models applied to the different phylogenetic analyses performed in this study.

	Locus ¹				
	ITS	<i>tef1</i>	<i>act</i>	<i>cmdA</i>	LSU
<i>Cercospora</i> spp.	SYM+I	HKY+G	K80+G	HKY+I+G	
<i>Pseudocercospora</i> spp.	SYM+G	HKY+I+G	SYM+I+G		
mycosphaerella-like spp.	GTR+I+G	HKY+I+G	HKY+I+G		GTR+I+G

¹ Substitution models used in the studies. GTR: General Time Reversible; HKY: Hasegawa-Kishino-Yano; K80: Kimura 2-parameter; SYM: symmetrical model; Non-uniformity of evolutionary rates among sites were modeled by using a discrete Gamma distribution (+G) alone and with five rate categories and by assuming that a certain fraction of sites are evolutionarily invariable (+I).

TreeBASE (<http://www.treebase.org>; S17948), and taxonomic novelties in MycoBank (www.Mycobank.org; Crous et al. 2004a).

RESULTS

Phylogenetic analyses

The three datasets consisted of 1 265 characters, representing 92 taxa for the *Cercospora* tree, including the outgroup (*act*: 183, *tef1*: 315, ITS: 476 and *cmdA*: 291), 1 114 characters, representing 94 taxa for the *Pseudocercospora* tree, including the outgroup (*act*: 217, *tef1*: 394 and ITS: 503) and 1 944 characters, representing 84 taxa for the mycosphaerella-like tree, including the outgroup (*act*: 232, *tef1*: 435, ITS: 507 and LSU: 758).

The respective alignments included 351 unique site patterns for the *Cercospora* tree (*act*: 76, *tef1*: 125, ITS: 41 and *cmdA*: 109), 351 unique site patterns for the *Pseudocercospora* tree (*act*: 79, *tef1*: 200 and ITS: 72) and 723 unique site patterns for the mycosphaerella-like tree (*act*: 127, *tef1*: 226, ITS: 221 and LSU: 149).

After topological convergence of the Bayesian runs, the following numbers of trees were generated and subsequently sampled (using a burn-in fraction of 0.25 and indicated after the slash) in order to generate the three Bayesian phylogenies: 2948/2140 for *Cercospora* (Fig. 1), 4465/3572 for *Pseudocercospora* (Fig. 2) and 1710/1368 for mycosphaerella-like taxa (Fig. 3). The resulting phylogenetic trees of all three individual combined datasets showed consistent clustering of all taxa over each one of the trees, and the results are treated below. Bayesian posterior probabilities (PP) are presented on the left of each node, on each tree.

TAXONOMY

The Consolidated Species Concept was employed in this study to distinguish species, revealing a rich diversity among the cercosporoid fungi on ferns in Brazil. Forty-three isolates of cer-

cosporoid and mycosphaerella-like species, collected from 18 host species representing 201 localities, were studied. The Bayesian analysis resulted in a total of 20 frond-spotting taxa, which belong to eight genera including *Cercospora*, *Clypeosphaerella*, *Neoceratosperma*, *Paramycosphaerella*, *Phaeophleospora*, *Pseudocercospora*, *Xenomycosphaerella* and *Zasmidium*. Three of these were assigned to an existing species name, one more could not be named unequivocally, a further 15 were described as new, and one novel species, as well as one new genus, are introduced below for the remaining taxon.

Cercospora Fresen., Beitr. Mykol. 3: 91. 1863

Cercospora coniogrammes Crous & R.G. Shivas, Stud. Mycol. 75: 151. 2013 — Fig. 4

Description & Illustration — Groenewald et al. (2013).

Specimens examined. BRAZIL, Rio de Janeiro, Nova Friburgo, Fazenda Barreto II, garden, on fronds of *Macrothelypteris torresiana*, 7 Aug. 2010, R.W. Barreto (VIC 42537, CBS H-22063, cultures CPC 24661, COAD 1067); Rio de Janeiro, Nova Friburgo, Alto do Micheis, Riograndina, reforestation area, on fronds of *M. torresiana*, 13 June 2011, R.W. Barreto (VIC 42545, CBS H-22064, cultures CPC 24669, COAD 1093); Rio de Janeiro, Gávea, Atlantic rainforest, on fronds of *M. torresiana*, 12 Oct. 2011, R.W. Barreto (VIC 42554, CBS H-22065, cultures CPC 24672, COAD 1089); Minas Gerais, Araponga, Pedra Dourada, Atlantic rainforest, on fronds of *M. torresiana*, 19 Nov. 2011, E. Guatimosim (VIC 42464, CBS H-22073, cultures CPC 24706); Rio de Janeiro, Nova Friburgo, Macaé de Cima, roadside, on fronds of *Hypolepis mitis*, 10 May 2014, R.W. Barreto (cultures CPC 25070, COAD 1769).

Cercospora samambaiae Guatimosim, R.W. Barreto & Crous, *sp. nov.* — MycoBank MB812771; Fig. 5

Etymology. Name refers to the common name used for ferns in Brazil, or of native Indian Tupi language origin – samambaia.

Description in planta — *Frond spots* irregular, starting on the edges of the pinnulets, extending to encompass whole pinnulets and sometimes leading to the necrosis of the entire pinnule. Starting centrally, pale brown, becoming pale brown to red at the edges, coalescing, turning dark brown to black. *Caespituli*

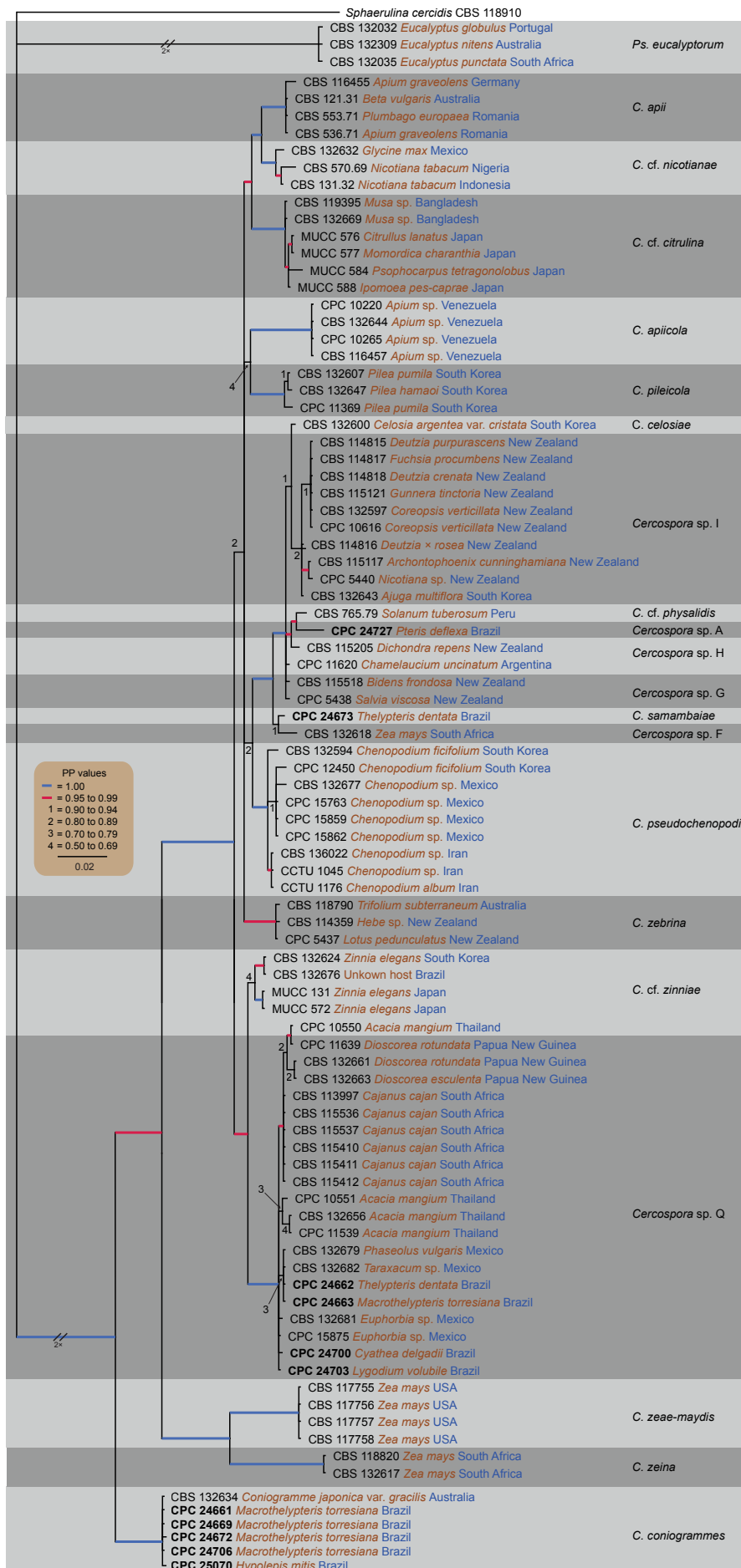


Fig. 1 Consensus phylogram (50 % majority rule) of *Cercospora* species, from a Bayesian analysis of the combined 4-gene sequence alignment (ITS, *tef1*, *act*, *cmdA*). Bayesian posterior probabilities are indicated with colour-coded branches and numbers (see legend) and the scale bar indicates 0.02 expected changes per site. Isolates from Brazil are indicated in **bold**. Hosts and countries of origin are indicated in brown and blue text, respectively. The tree was rooted to *Sphaerulina cercidis* (isolate CBS 118910).

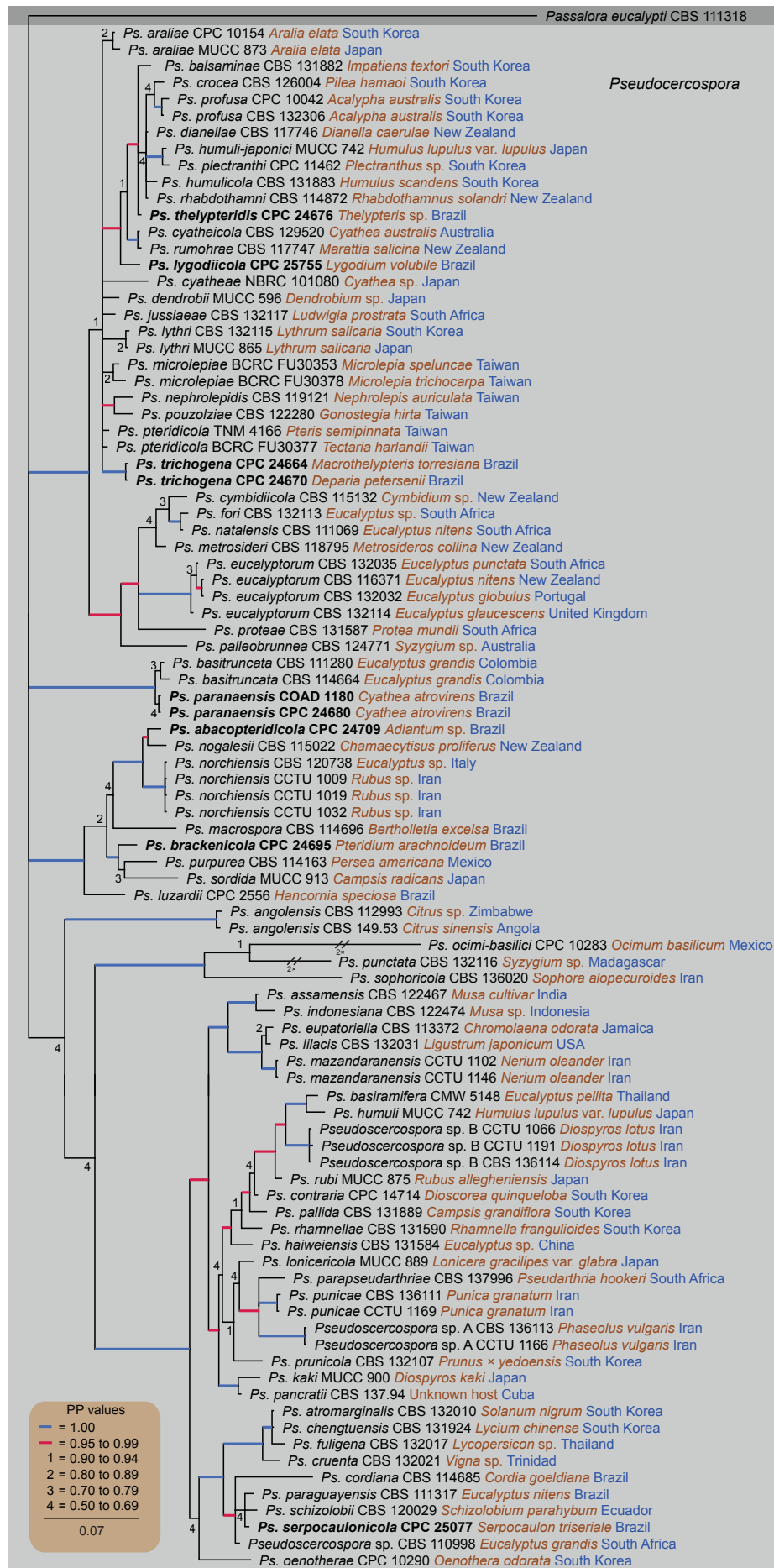


Fig. 2 Consensus phylogram (50 % majority rule) of *Pseudocercospora* species, from a Bayesian analysis of the combined 3-gene sequence alignment (ITS, *act*, *tef1*). Bayesian posterior probabilities are indicated with colour-coded branches and numbers (see legend). The scale bar indicates 0.07 expected changes per site. Isolates from Brazil are indicated in bold. Hosts and countries of origin are indicated in brown and blue text, respectively. The tree was rooted to *Passalora eucalypti* (isolate CBS 111318).

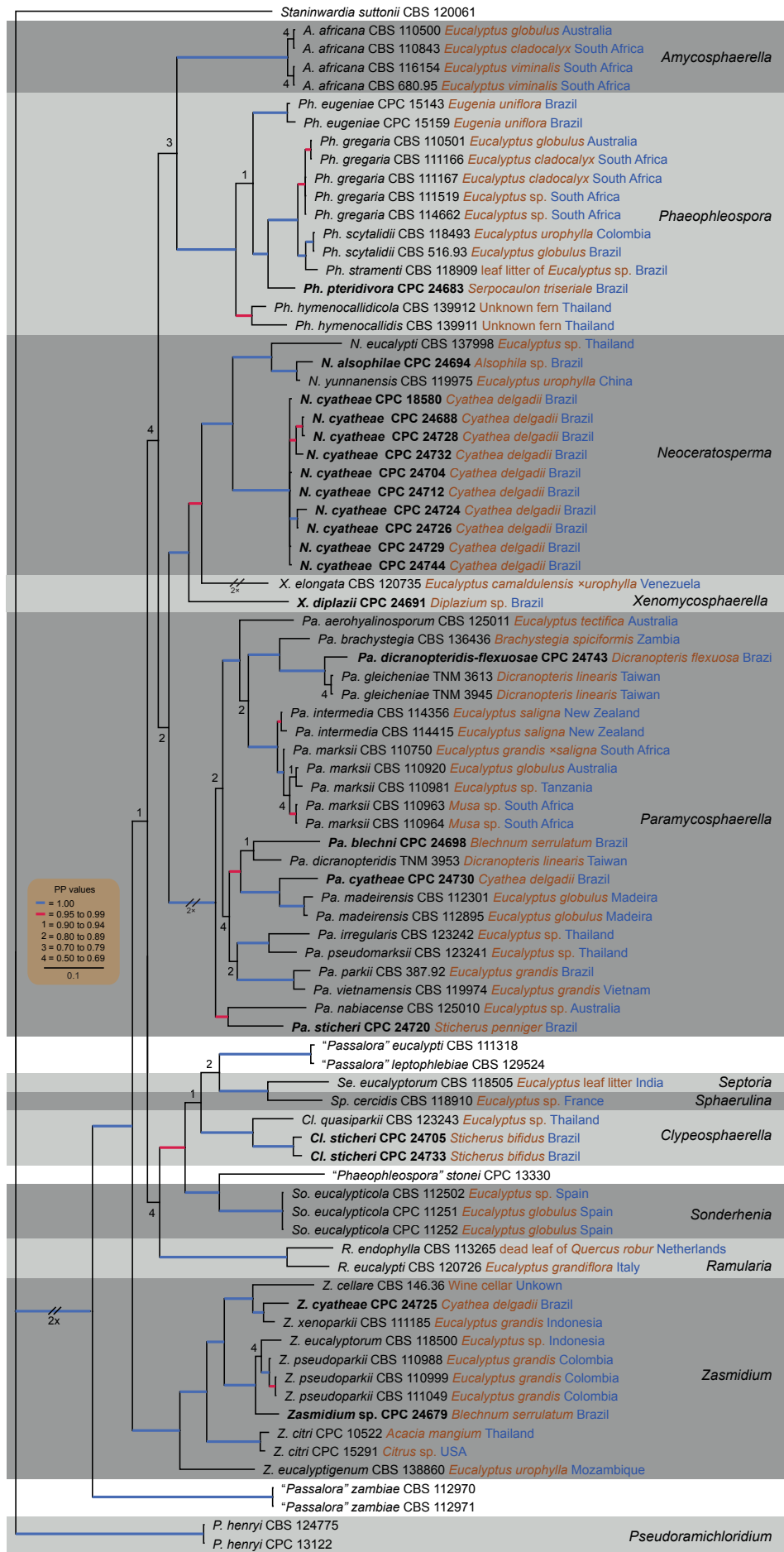


Fig. 3 Consensus phylogram (50 % majority rule) of mycosphaerella-like species, from a Bayesian analysis of the combined 4-gene sequence alignment (*act*, *tef1*, ITS, LSU). Bayesian posterior probabilities are indicated with colour-coded branches and numbers (see legend). The scale bar indicates 0.1 expected changes per site. Isolates from Brazil are indicated in bold. Hosts and countries of origin are indicated in brown and blue text, respectively. The tree was rooted to *Staninwardia suttonii* (isolate CBS 120061).

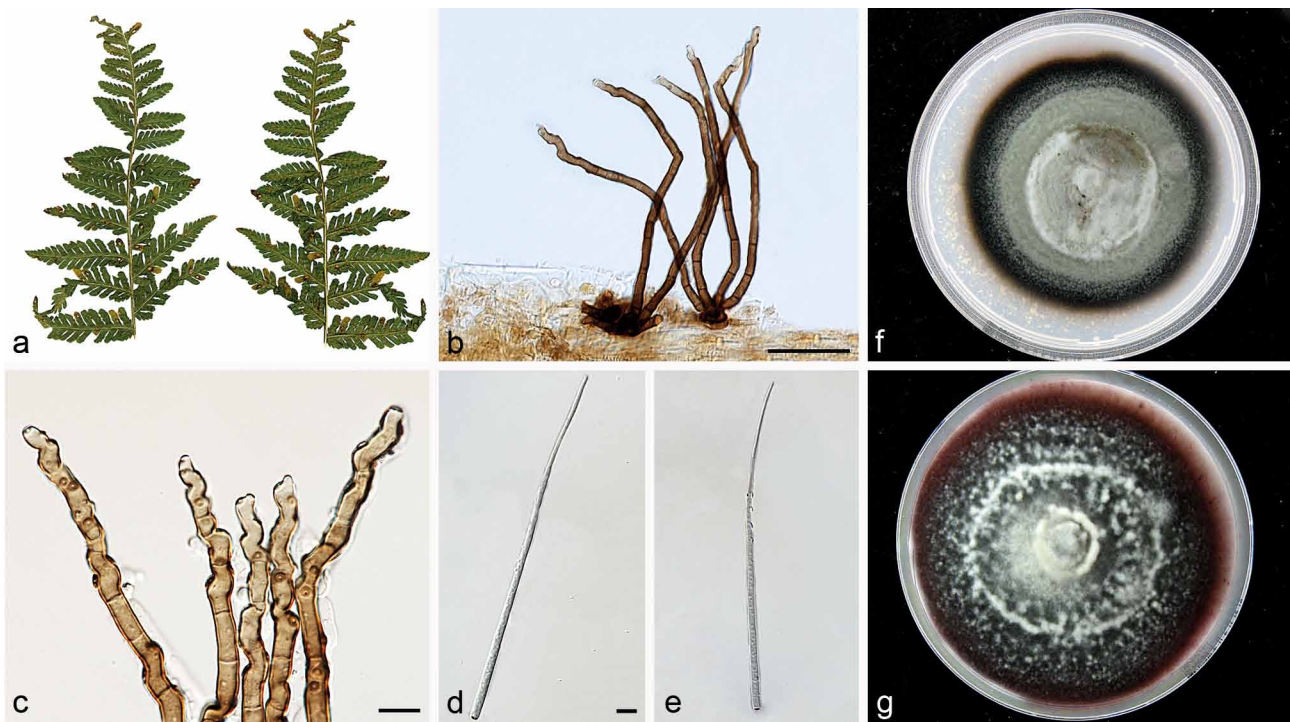


Fig. 4 *Cercospora coniogrammes* (CPC 24661). a. Frond spots on *Marcothelypteris torresiana*; b. c. conidiophores; d. e. conidia; f. culture on PDA; g. culture on PCA. — Scale bars: b = 50 μ m; c, d = 10 μ m.

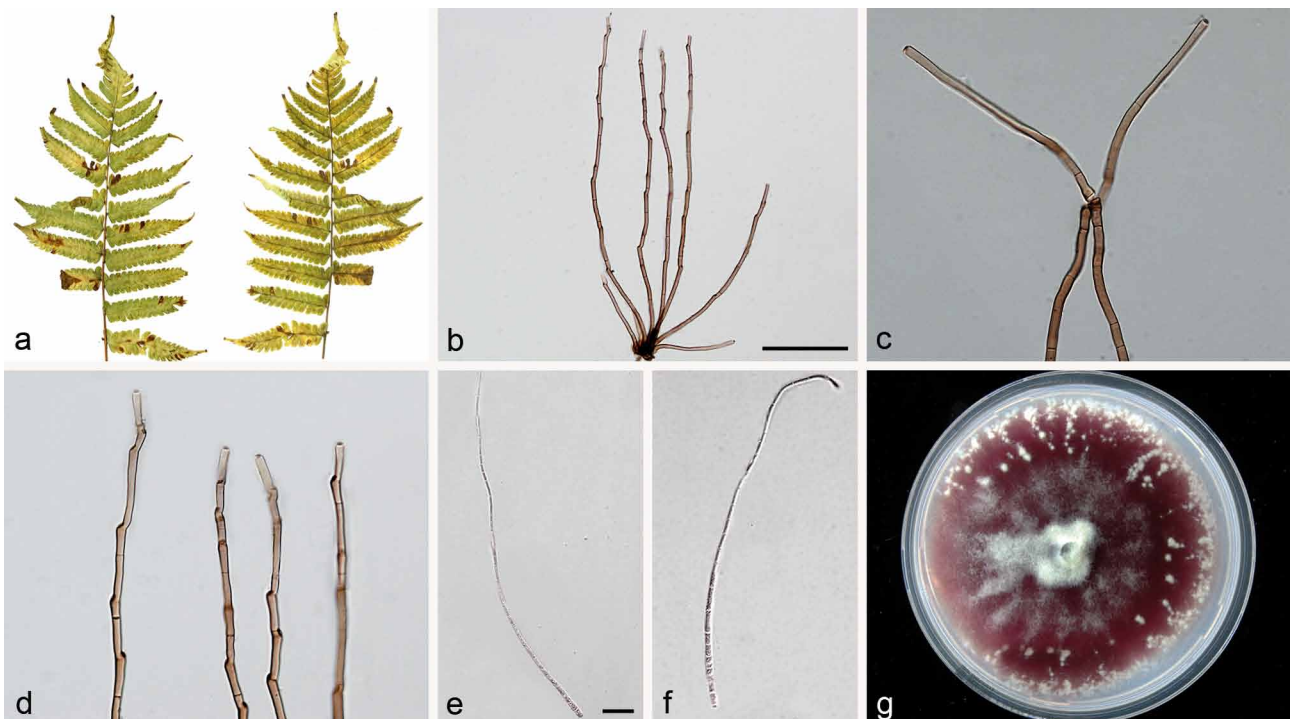


Fig. 5 *Cercospora samambaiae* (CPC 24673). a. Frond spots on *Thelypteris dentata*; b–d. conidiophores; e. f. conidia; g. culture on PCA. — Scale bars: b = 100 μ m; e = 50 μ m.

hypophyllous, abundant. *External hyphae* absent. *Internal hyphae* indistinct. *Stromata* rudimentary, irregular, composed of *textura globulosa*, dark brown. *Conidiophores* rising through the stomata, hypophyllous, forming fascicles (6–11 stalks per fascicle), subcylindrical, straight to curved, geniculate, (92–)140–320(–509) \times 5–6 μ m, unbranched, 3–15-septate, guttulate, pale brown becoming paler at the apex, smooth. *Conidigenous cells* terminal, integrated, holoblastic, subcylindrical, predominantly sympodial, 40–95 \times 4–6 μ m, pale to olivaceous brown, scars conspicuous, 1–3 per cell, 1.5–4 μ m, thickened, darkened. *Conidia* solitary, acicular, straight to slightly curved, (134–)218–282(–320) \times 2–3 μ m, apex acute, base subtrun-

cate, 2.5–4.5 μ m diam at the base, (13–)16–21(–34)-septate, guttulate, hyaline, smooth; hila thickened, darkened, refractive, 2–4 μ m diam.

Culture characteristics — Colonies on PCA slow-growing, 80 mm diam after 28 d; flat, with sparse aerial mycelium, mouse grey centrally, lavender grey to white at periphery, pigmenting the medium to livid red; reverse livid red.

Specimens examined. BRAZIL, Minas Gerais, Itabirito, Posto Esperança, garden, on fronds of *Thelypteris dentata*, 23 Oct. 2011, R.W. Barreto (holotype CBS H-22071, isotype VIC 42555, cultures ex-type CPC 24673, COAD 1090).

Notes — In the *tef1*, and *cmdA* phylogeny, isolates of *C. samambaiae* and *Cercospora* sp. F (sensu Groenewald et al. 2013) cluster together in a distinct well-supported clade. In the *act* phylogeny, *C. samambaiae* forms a distinct clade, whereas *Cercospora* sp. F cannot be distinguished from *Cercospora* sp. Q (sensu Groenewald et al. 2013), nor from *C. conioagrammes* (data not shown). The different *act* sequences explain the basal position of *Cercospora* sp. F to the *C. samambaiae* clade in the combined phylogeny (Fig. 1). Two *Cercospora* species are known to cause frond spots on species of *Thelypteridaceae*, namely *C. abacopteridis* and *C. cyclosori*. *Cercospora abacopteridis* is morphologically quite distinct from *C. samambaiae* in having much smaller and narrower conidiophores (15–120 × 4–5 µm), rising directly from the internal hyphae. Additionally, *C. abacopteridis* is only known from Singapore, causing leaf spots on *Abacopteris urophylla* (Braun et al. 2013). *Cercospora cyclosori*, described on *Cyclosorus* spp. from India and Taiwan, is even more distinct from *C. samambaiae* in having shorter and wider conidia (50–110 × 3–4 µm) and shorter and narrower conidiophores (25–160 × 4–5 µm) (Braun et al. 2013).

Cercospora sp. A

Culture characteristics — Colonies on PCA slow-growing, 60 mm diam after 28 d; flat, with sparse aerial mycelium, pale mouse grey centrally, mouse grey to olivaceous grey at periphery; reverse leaden black.

Specimens examined. BRAZIL, Paraná, Curitiba, BR 116 road to Rio Negro, roadside, on fronds of *Pteris deflexa*, 14 Apr. 2013, E. Guatimosim (CBS H-22070, VIC 42529, cultures CPC 24727, COAD 1427).

Notes — Fungarium specimens of this fungus were in poor condition and no conidia were seen. Isolation was performed by conidiophore transfer only. Phylogenetically, this specimen has *C. cf. physalidis* (CBS 765.79) as sister clade (Fig. 1), but differs from the latter by having the following number of variable sites: 11 for *act*, 5 for *cmdA* and 1 for *tef1*. Once no conidia were seen and all attempts to promote sporulation in vitro proved to be unsuccessful, it is not possible to determine the species boundaries of this isolate.

Cercospora sp. Q sensu Groenewald et al. (2013) — Fig. 6

Description in planta — *Frond spots* amphigenous, irregular, starting at the apex of the pinnulets, spreading to the base of the pinnule, coalescing, leading to complete necrosis of the pinnule. *Caespituli* hypophyllous, abundant. *Internal hyphae* septate, intra- and intercellular, frequently branched, 2–4 µm wide, pale brown, smooth. *Stromata* rudimentary, globular, composed of *textura globulosa*, dark brown. *Conidiophores* rising through the stomata, hypophyllous, forming loose fascicles (3–7 stalks per fascicle), subcylindrical, straight or slightly curved to sinuose, geniculate, (96–)141–230(–326) × 4–5 µm, unbranched, 3–9-septate, olivaceous brown, thin-walled, smooth. *Conidiogenous cells* terminal, rarely integrated, holo-



Fig. 6 *Cercospora* sp. Q (CPC 24662). a. Frond spots on *Lygodium volubile*; b. frond spots on *Cyathea delgadii*; c. frond spots on *Thelypteris dentata*; d. e. sporulation on the pinnule; f–h. conidiophores; i–m. conidia. — Scale bars: f = 10 µm; h = 50 µm; i = 15 µm.

blastic, subcylindrical, tapering to a flat-tipped apex, with numerous tightly aggregated apical conidiogenous loci, proliferating sympodially, $(26\text{--}38\text{--}71\text{--}102) \times 4\text{--}5 \mu\text{m}$, pale brown, smooth, scars conspicuous, protruding, $2.5\text{--}4 \mu\text{m}$ diam, thickened, darkened. *Conidia* solitary, acicular, sinuous to slightly curved, $(142\text{--})192\text{--}256\text{--}(303) \times 2\text{--}3 \mu\text{m}$, apex acute, base subtruncate, $(10\text{--})18\text{--}28\text{--}(31)$ -septate, rarely guttulate, hyaline, thin-walled, smooth; hila thickened, darkened, refractive, $2\text{--}4 \mu\text{m}$ diam.

Specimens examined. BRAZIL, Minas Gerais, Viçosa, Sítio Cristais, from a garden, on fronds of *Thelypteris dentata*, 10 May 2011, R. W. Barreto (CBS H-22067, VIC 42538, cultures CPC 24662, COAD 630); Rio de Janeiro, Nova Friburgo, Alto do Micheis, Riograndina, reforestation area, on fronds of *M. torresiana*, 13 June 2011, R. W. Barreto (CBS H-22068, VIC 42540, cultures CPC 24663, COAD 322); Goiás, Pirenópolis, Fazenda Bom-sucesso, Cerrado biome, on fronds of *Cyathea delgadii*, 26 Sept. 2013, R. W. Barreto

(CBS H-22069, VIC 42601, cultures CPC 24700, COAD 1418); Minas Gerais, Viçosa, Sítio Cristais, from a garden, on fronds of *Lygodium volubile*, 4 Feb. 2014, R. W. Barreto (CBS H-22066, culture CPC 24703).

Notes — Four Brazilian isolates, from different hosts and families, cluster within this clade, to which different names can be applied. As stated by Groenewald et al. (2013) and Bakhshi et al. (2015), to resolve their taxonomy, fresh collections authentic for the names, based on host and country, need to be recollected and included in future studies. Morphologically, the isolates from Brazil are indistinguishable from *C. apii*, but the hosts on which they cause disease are significantly different, e.g. all isolates included in *Cercospora* sp. Q so far, were obtained from angiosperms, while the Brazilian isolates in this study, are from three different orders of *Pteridophyta*, (*Cyatheaales*, *Polypodiales* and *Schizaeales*). Phylogenetically,



Fig. 7 *Clypeosphaerella sticheri* (CPC 24705). a–c. Frond spots on *Sticherus bifidus*; d. erumpent subcuticular ascomata, fruiting epiphyllous; e, f. vertical section of the ascoma, note the thicker upper part of the ascoma, resembling a pseudoclypeus; g, h. asci; i. ascospores; j. germinating ascospores; k. culture on MEA; l. culture on OA; m. culture on PDA. — Scale bars = 10 μm .

the isolates included in *Cercospora* sp. Q clade differ from the other species by their position in the *cmdA* and *tef1* phylogeny; while in the *act* phylogeny they cannot be distinguished from *Cercospora* sp. F (data not shown). Based on the genes studied here, and five other different loci studied by Groenewald et al. (2013), the species boundaries of all isolates included in this clade could not be clarified.

Clypeosphaerella Guatimosim, R.W. Barreto & Crous, *gen. nov.*
— MycoBank MB812820

Type species. *Clypeosphaerella sticheri* Guatimosim, R.W. Barreto & Crous.

Etymology. Named after the thickened wall of the ascomata, resembling a pseudoclypeus.

Frondicolous, plant pathogenic. *Ascomata* pseudothecial, epiphyllous, solitary, subcuticular to erumpent, globose, walls of 2–3 layers of brown to dark brown *textura angularis*, ostiole central. *Asci* bitunicate, aparaphysate, fasciculate, subsessile, 8-spored, obpyriform to ovoid, hyaline, smooth. *Ascospores* inordinate, overlapping, fusoid, straight, 1-septate, slightly constricted at the septum, biguttulate, hyaline, thin-walled, smooth. *Ascospores germinating* at both ends, remaining hyaline, germ tubes following the main axis of the spore.

Notes — *Clypeosphaerella* is morphologically similar to species of *Mycosphaerella* s.lat., differing by having the thicker upper wall of the ascomata, resembling a *pseudoclypeus*. Additionally, the former genus is phylogenetically distinct from other mycosphaerella-like fungi (Fig. 3).

Clypeosphaerella quasiparkii (Cheew. et al.) Guatimosim, R.W. Barreto & Crous, *comb. nov.* — MycoBank MB812821

Basionym. *Mycosphaerella quasiparkii* Cheew. et al., *Persoonia* 21: 85. 2008.

Description & Illustration — Cheewangkoon et al. (2008).

Specimen examined. THAILAND, Burirum, on leaves of *Eucalyptus* sp., July 2007, *P. Suwannawong* (holotype CBS H-20132, cultures ex-type CBS 123243, CPC 15433, CPC 15434).

Clypeosphaerella sticheri Guatimosim, R.W. Barreto & Crous, *sp. nov.* — MycoBank MB812822; Fig. 7

Etymology. Name refers to the host genus from which it was isolated, *Sticherus*.

Description in planta — *Frond spots* epiphyllous, affecting almost all the pinnulets, starting as small dark brown areas, irregular, usually close to the main vein of the pinnae, spreading through the pinnulet, becoming fertile, confluent and necrotic. *Internal hyphae* intra- and intercellular, 1.5–3.5 µm wide, branched, septate, subhyaline, smooth. *Ascomata* pseudothecial, epiphyllous, mostly congregated at the basis of the pinnae, solitary, subcuticular to erumpent, globose, 40–71 × 43–83 µm, walls of 2–3 layers of brown to dark brown *textura angularis*, cells 4–8 × 1.5–5 µm, ostiole central, 10–24 µm diam. *Asci* bitunicate, aparaphysate, fasciculate, subsessile, 8-spored, obpyriform to ovoid, straight or slightly curved, 20–34 × 10–14 µm, hyaline, smooth. *Ascospores* inordinate, overlapping, fusoid, straight, 9–13 × 2–4 µm, 1-septate, slightly constricted at the septum, tapering towards rounded ends, narrower towards the lower end, biguttulate, hyaline, thin-walled, smooth. *Ascospores germinating* at both ends, remaining hyaline, germ tubes following the main axis of the spore, while the spore becomes distorted and constricted at the septum (Type F, Crous 1998). *Asexual morph* not known.

Culture characteristics — Colonies on MEA slow-growing, 22 mm diam after 24 d; raised, aerial mycelium velvety, laven-

der grey centrally and pale vinaceous at periphery, vinaceous buff reverse. On OA, aerial mycelium sparse, mouse grey centrally, buff periphery; dark mouse grey with rosy buff periphery reverse. On PDA pale mouse grey centrally, white periphery; smoke with rosy buff periphery reverse; cultures sterile.

Specimens examined. BRAZIL, Rio de Janeiro, Nova Friburgo, Fazenda Barreto II, Riograndina, ruderal, on fronds of *Sticherus bifidus*, 11 Feb. 2014, R.W. Barreto (holotype CBS H-22088, isotype VIC 42607, culture ex-type CPC 24705); Minas Gerais, Araponga, Parque Estadual da Serra do Brigadeiro, path to Pico do Pato, Atlantic rainforest, on fronds of *S. bifidus*, 21 Feb. 2014, E. Guatimosim (CBS H-22089, VIC 42516, culture CPC 24733).

Notes — Morphologically, *Cl. sticheri* is most similar to *Cl. quasiparkii* described on *Eucalyptus* sp. from Thailand (Cheewangkoon et al. 2008), but can be distinguished from it by having smaller and wider asci (45–50 × 8.5–9 µm in the later), larger ascospores (10–11 × 3–3.5 µm in the latter) and by the germination of the ascospores – following the main axis, regular in width, not distorted in *Cl. sticheri* (Type F, Crous 1998) whereas in *Cl. quasiparkii* germ tubes arise from the polar ends, develop firstly parallel to the main axis, and later grow perpendicularly, becoming distorted (Type D, Crous 1998) (Cheewangkoon et al. 2008). Additionally, it is also phylogenetically distinct (Fig. 3).

Neoceratosperma Crous & Cheew., *Persoonia* 32: 255. 2014
— MycoBank MB808935

Notes — *Neoceratosperma* has thus far been known only from its type species, *N. eucalypti*, isolated on *Eucalyptus* sp. (*Myrtaceae*) from Thailand (Crous et al. 2014). *Neoceratosperma eucalypti* is asexual and zasmidium-like in morphology. In the present study, we expanded the generic concept by including three additional species, two of which are known from their sexual morphs, being mycosphaerella-like in morphology.

Neoceratosperma alsophilae Guatimosim, R.W. Barreto & Crous, *sp. nov.* — MycoBank MB812816; Fig. 8

Etymology. Name refers to the host genus from which it was isolated, *Alsophila*.

Description in planta — *Frond spots* random on pinnules, amphigenous, irregular, initially pale brown with cream central area at the tips the pinnulets, spreading through the base of the pinnulet, becoming necrotic with a fertile cream to pale brown centre and distinct dark brown to black halo. *Internal hyphae* intra- and intercellular, 1.5–3 µm wide, septate, branched, subhyaline, smooth. *External hyphae* absent. *Ascomata* pseudothecial, epiphyllous, solitary, subcuticular to erumpent, globose, 61–91 × 64–112 µm, walls of 2–3 layers of pale to dark brown *textura angularis*, cells 5–8 × 3–5 µm, ostiole central, 17–32 µm diam. *Asci* bitunicate, aparaphysate, fasciculate, subsessile, 8-spored, obovoid to broadly ellipsoidal, straight or slightly curved, 29–42 × 9–18 µm, hyaline, smooth. *Ascospores* inordinate, overlapping, fusoid, straight or slightly curved, 10–17 × 2–4 µm, medianly 1-septate, wider in middle of apical cell, tapering toward rounded ends, biguttulate, hyaline, thin-walled, smooth. *Asexual morph* not known.

Culture characteristics — Colonies on MEA, slow-growing, 26 mm diam after 24 d; centrally raised, with lobate, smooth margins, aerial mycelium velvety, olivaceous grey centrally, and mouse grey in the outer region; leaden black in reverse. On OA, colony radially striate with lobate margins, aerial mycelium cottony, pale mouse grey centrally and mouse greenish grey in the outer region; leaden black in reverse. On PDA colony centrally elevated, aerial mycelium sparse to absent, mouse grey centrally and producing a black halo in the outer region; leaden black in reverse; cultures sterile.



Fig. 8 *Neoceratosperma alsophilae* (CPC 24694). a, b. Frond spots on *Alsophila* sp.; c, d. erumpent subcuticular ascomata, fruiting epiphyllous; e, f. vertical section of the ascoma; g. asci; h. ascospores; i. culture on MEA; j. culture on OA; k. culture on PDA. — Scale bars = 10 μ m.

Specimens examined. BRAZIL, Minas Gerais, Capitólio, Furnas, roadside next to Rio do Turvo Inn, on fronds of *Alsophila* sp., 9 Nov. 2012, E. Guatimosim (holotype CBS H-22075, isotype VIC 42586, cultures ex-type CPC 24694, COAD 1181).

Notes — Morphologically and phylogenetically, *N. alsophilae* is closely related to *N. yunnanensis* described on *Eucalyptus urophylla*, restricted to the southwest of China (Burgess et al. 2007). It can be distinguished from *N. yunnanensis* by having narrower, obclavate to broadly ellipsoidal asci (ovoid to obclavate, 27–38 \times 7–11 μ m in *N. yunnanensis*) and ascospores (10–12.5 \times 2.5–3 μ m in *N. yunnanensis*). Moreover, *N. yunnanensis* is phylogenetically distinct from *N. alsophilae* (Fig. 3).

Neoceratosperma cyatheae Guatimosim, R.W. Barreto & Crous, sp. nov. — MycoBank MB812817; Fig. 9

Etymology. Name refers to the host genus from which it was isolated, *Cyathea*.

Description in planta — *Frond spots* random on pinnulets, amphigenous, irregular to angular, starting on the edges of the pinnulets and spreading along the centre, 3–9 \times 3–5 mm, leading to entire pinnule necrosis and, at the final stages, the entire pinnae being affected. Becoming chlorotic (under high humidity conditions), sometimes leading to complete necrosis of the pinnae tip, together with distinct cinnamon to yellow-brown areas, appearing at the pinnae bases. *Internal hyphae* intra- and intercellular, 2–3 μ m wide, septate, branched, subhyaline

to pale brown, smooth. *External hyphae* hypophyllous, arising through stomata and covering the entire lesion, 2–3 μ m wide, septate, branched, pale brown to brown, strongly verruculose. *Conidiophores* arising singly from superficial hyphae, reduced to conidiogenous cells obcuneiform, straight, proliferating sympodially, 4–19 \times 2–6 μ m, unbranched, aseptate, pale brown, smooth, scars conspicuous, several per cell, terminal, crowded, darkened, thickened. *Conidia* solitary, subcylindrical, straight, curved or sinuous, (40–)95–160(–280) \times 3–5 μ m, apex obtuse, base subtruncate, distoseptate when young, indistinctly 5–19-septate at maturity, strongly guttulate, pale to dark brown, strongly verruculose; hila 1–3 μ m wide, thickened, darkened and refractive. *Sexual morph* not known.

Culture characteristics — Colonies on MEA and OA slow-growing, 20 mm diam after 24 d; raised, with lobate, feathery margins and velvety aerial mycelium, lavender grey centrally, leaden black mixed with lavender grey areas at periphery; iron-grey reverse. On PDA, colony humid centrally, pale mouse grey centrally, mouse grey periphery; green-black reverse; cultures sterile.

Specimens examined. BRAZIL, Rio de Janeiro, Fazenda Barreto II, Rio-grandina, on fronds of *C. delgadii*, 11 Feb. 2014, R.W. Barreto (holotype CBS H-22074, isotype VIC 42605, culture ex-type CPC 24704); Rio de Janeiro, Nova Friburgo, Macaé de Cima, on fronds of *C. delgadii*, 11 July 2009, R.W. Barreto (CBS H-22078, VIC 42533, cultures CPC 18580, COAD 573); Rio Grande do Sul, Ituporanga, highway to Alfredo Wagner, roadside, on fronds of *C. delgadii*, 15 Apr. 2013, E. Guatimosim (CBS H-22083, VIC 42520, cultures CPC 24729, COAD 1428); São Paulo, Eldorado, vicinities of



Fig. 9 *Neoceratosperma cyatheae* (CPC 24704). a, b. Frond spots on *Cyathea delgadii*; c. SEM of the conidia and conidiophore, note the smooth conidiophore reduced to conidiogenous cell; d. detail of the external hyphae arising through the stoma; e. conidiophores arising through hyphae, reduced to conidiogenous cells; f–k. conidia; l. culture on MEA; m. culture on OA; n. culture on PDA. — Scale bars = 10 μ m.

Parque Caverna do Diabo, Atlantic rainforest, on fronds of *C. delgadii*, 13 Apr. 2013, *E. Guatimosim* (CBS H-22084, culture CPC 24724); São Paulo, Barra do Turvo, highway Regis Bitancourt, roadside, on fronds of *C. delgadii*, 13 Apr. 2013, *E. Guatimosim* (CBS H-22081, VIC 42527, culture CPC 24726); São Paulo, Iporanga, highway to Barra do Turvo, roadside, 13 Apr. 2013, *E. Guatimosim* (CBS H-22082, VIC 42530, cultures CPC 24728); Minas Gerais, Araponga, Parque Estadual da Serra do Brigadeiro, Atlantic rainforest, on fronds of *C. delgadii*, 21 Feb. 2014, *E. Guatimosim* (CBS H-22080, VIC 42524, culture CPC 24732); *ibid.*, 23 Feb. 2014, *E. Guatimosim* (CBS H-22079, VIC 42461, culture CPC 24744); Rio de Janeiro, road between Macaé de Cima and Lumiar, riverside, on fronds of *C. delgadii*, 29 Apr. 2012, *R.W. Barreto* (CBS H-22077, VIC 42578, cultures CPC 24688, COAD 1238); Rio Grande do Sul, Itaporanga, highway to Rio do Sul, roadside, on fronds of *C. delgadii*, 15 Apr. 2013, *E. Guatimosim* (CBS H-22085, VIC 42477, culture CPC 24712).

Notes — *Neoceratosperma cyatheae* is phylogenetically different from all other species in this clade (Fig. 3). It was not possible to compare *N. cyatheae* with *N. alsophilae* and *N. yunnanensis* since the latter species are only known from their sexual morphs (Burgess et al. 2007, this study). In contrast for *N. cyatheae* only the asexual morph was found, which resembles zasmidium-like fungi, which are known to be polyphyletic (Crous et al. 2009a, b). Morphologically, *N. cyatheae* is similar to *N. eucalypti*, but differs from the latter by having smooth conidiophores reduced to conidiogenous cells (1–15-septate, verruculose, up to 100 μ m long in *N. eucalypti*) and solitary conidia (solitary to catenulate in *N. eucalypti*) (Crous et al. 2014). The distoseptation in young conidia, a characteristic

feature for *Neoceratosperma*, can easily be overlooked due to the abundant, large guttules.

Neoceratosperma yunnanensis (Barber & T.I. Burgess) Guatimosim, R.W. Barreto & Crous, *comb. nov.* — MycoBank MB813444

Basionym. *Mycosphaerella yunnanensis* Barber & T.I. Burgess, *Fung. Diversity* 24: 150. 2007.

= *Xenomycosphaerella yunnanensis* Quaedvlieg & Crous, *Persoonia* 33: 24. 2014.

Description & Illustration — Burgess et al. (2007).

Specimen examined. CHINA, Yunnan, Lancang, leaves of *Eucalyptus urophylla*, May 2005, *B. Dell* (holotype MURU 407, culture ex-type CBS 119975 = CMW 23443).

Paramycosphaerella Crous, *Persoonia* 31: 245. 2013. — MycoBank MB805850

Notes — The genus *Paramycosphaerella* is based on *Pa. brachystegia*, which occurs on *Brachystegia* sp. (*Fabaceae*) from Zimbabwe (Crous et al. 2013b). Thus far, only sexual morphs were known from this genus, which contains mycosphaerella-like species. In a previous study, Quaedvlieg et al. (2014) restricted their analyses to two species of *Paramycosphaerella*, relying on phylogenetic inferences to allocate species to this

genus. In the present study, we expanded the genus by also including additional phylogenetically related taxa.

Paramycosphaerella aerohyalinosporum (Crous & Summerell) Guatimosim, R.W. Barreto & Crous, *comb. nov.* — MycoBank MB509762

Basionym. *Zasmidium aerohyalinosporum* Crous & Summerell, *Persoonia* 23: 142. 2009.

Description & Illustration — Crous et al. (2009c).

Specimen examined. AUSTRALIA, New South Wales, road to Robin Falls, on leaves of *Eucalyptus tectifica*, 23 Sept. 2007, B.A. Summerell (holotype CBS H-20274, cultures ex-type CBS 125011, CPC 14636, CPC 14637).

Paramycosphaerella blechni Guatimosim, R.W. Barreto & Crous, *sp. nov.* — MycoBank MB812773; Fig. 10

Etymology. Name refers to the host genus from which it was isolated, *Blechnum*.

Description in planta — *Fron*d spots amphigenous, starting on the pinnule as pale brown random spots, vein-delimited, with a pale brown central area, coalescing with age, becoming irregular, with a central pale brown necrotic area surrounded with a distinct dark brown halo where ascomata are produced. *Internal hyphae* branched, septate, intra- and intercellular, 1.5–3.5 µm wide, subhyaline to pale brown, smooth. *Ascomata*

pseudothecial, epiphyllous, solitary, subcuticular to erumpent, globose to subglobose, 52–90 × 58–76 µm, walls of 2–3 layers of brown to dark brown *textura angularis*, cells 3.5–7 × 2–3.5 µm, black, ostiole central, 17–28 µm diam. *Asci* bitunicate, paraphysate, fasciculate, sessile, 8-spored, obpyriform to ovoid, straight or slightly curved, 22–52 × 7.5–14 µm, hyaline, smooth. *Ascospores* inordinate, overlapping, fusoid, straight to slightly curved, 12.5–19 × 2–4.5 µm, medianly 1-septate, apical cell wider, tapering towards both ends, but more prominently towards the upper end, guttulate, hyaline, thin-walled, smooth. *Ascospore germination* not seen. *Asexual morph* not known.

Culture characteristics — Colonies on MEA and PDA slow-growing, 42 mm diam after 24 d; raised with lobate margins, sparse feathery aerial mycelium in centre, immersed mycelium at periphery, humid, lavender grey to white in centre, iron-grey at periphery; reverse iron-grey. On OA, colony entirely lavender grey; leaden grey with amber zones in reverse; cultures sterile.

Specimen examined. BRAZIL, Paraná, Curitiba, highway to Joinville, roadside, on fronds of *Blechnum serrulatum*, 14 Nov. 2012, E. Guatimosim (holotype CBS H-22090, isotype VIC 42593, culture ex-type CPC 24698, COAD 1183).

Notes — Morphologically, *Pa. blechni* is rather similar to *Pa. dicranopteridis-flexuosae* described on *Dicranopteris flexuosa* from Brazil (this study), but can be distinguished from it by having narrower obpyriform to ovoid asci (pyriform to narrowly ellipsoid, 10–18 µm wide in *Pa. dicranopteridis-flexuosae*).



Fig. 10 *Paramycosphaerella blechni* (CPC 24698). a–c. Frond spots on *Blechnum serrulatum*; d, e. vertical section of the ascoma; f. asci; g. ascospores; h. culture on MEA; i. culture on OA; j. culture on PDA. — Scale bars = 10 µm.

Phylogenetically, *Pa. blechni* is related to *Pa. dicranopteridis*, which is only known from its asexual morph. Both species differ from other species within this clade (Fig. 3). *Paramycosphaerella dicranopteridis* is presently only known from its ITS DNA sequence data (Kirschner & Liu 2014). Nevertheless, the two species differ on 33 bp for the ITS region.

Paramycosphaerella cyatheae Guatimosim, R.W. Barreto & Crous, *sp. nov.* — MycoBank MB812775; Fig. 11

Etymology. Name refers to the host genus from which it was isolated, *Cyathea*.

Description in planta — *Fron*d spots randomly affecting individual pinnules, irregular, initially necrotic along the main vein of the pinnulet, pale brown, with a cream central area where ascospores are formed, becoming dark brown. *Internal hyphae* branched, septate, intra- and intercellular, 2.5–4.5 µm wide, subhyaline, smooth. *Ascospores* pseudothecial, epiphyllous, solitary, subcuticular to erumpent, globose, (36–)50–82(–101) × 62–90 µm, walls of 2–3 layers of brown to dark brown *textura angularis*, cells 5–10 × 2–6 µm, black, ostiole central, 11–23 µm diam. *Asci* bitunicate, paraphysate, fasciculate, sessile, 8-spored, obpyriform, straight or slightly curved, 26–54 × 9–20 µm, hyaline, smooth. *Ascospores* inordinate, overlapping, fusoid, straight, 10–15 × 2.5–4 µm, unequally 1-septate, constricted at the septum, upper cell shorter, tapering towards

rounded ends, with two large opposed guttules, hyaline, thin-walled, smooth. *Ascospores germinating* from both ends, remaining hyaline after germination, germ tubes growing along the main axis of ascospore, germ tubes irregular in width, not to slightly distorted, spores becoming slightly constricted at the septum (Type C, Crous 1998). *Asexual morph* not known.

Culture characteristics — Colonies on MEA, OA and PDA slow-growing, 14 mm diam after 24 d; raised, with discrete margins, and dense cottony aerial mycelium, smoke grey centrally, iron at periphery, humid; iron-grey in reverse. On OA, slightly pigmenting the media, olivaceous grey; cultures sterile.

Specimen examined. BRAZIL, Minas Gerais, Araponga, Parque Estadual da Serra do Brigadeiro, path to Pico do Pato, on fronds of *Cyathea delgadii*, 22 Feb. 2014, E. Guatimosim (holotype CBS H-22092, isotype VIC 42519, culture ex-type CPC 24730).

Notes — Morphologically, *Pa. cyatheae* is rather similar to *Pa. madeirae* described on *Eucalyptus* sp. from Madeira (Crous et al. 2004b) and to *Pa. sticheri*, described on *Sticherus penningeri* from Brazil (this study), but can be distinguished by having wider asci (8–12 µm wide in *Pa. madeirae*) and smaller ascospores (14–20 × 3–5.5 µm in *Pa. sticheri*). Phylogenetically, *Pa. cyatheae* has *Pa. madeirae* as sister clade (Fig. 3). These two species, however, differ from each other by having the following number of variable sites for each locus: 23 bp for *act* and 17 bp for ITS.



Fig. 11 *Paramycosphaerella cyatheae* (CPC 24730). a, b. Frond spots on *Cyathea delgadii*; c. erumpent subcuticular ascospores, fruiting epiphyllous; d. vertical section of the ascoma; e. asci; f. ascospores; g. germinating ascospores; h. culture on MEA; i. culture on OA; j. culture on PDA. — Scale bars = 10 µm.

Paramycosphaerella dicranopteridis (R. Kirschner) Guatimosim, R.W. Barreto & Crous, *comb. nov.* — MycoBank MB812807

Basionym. *Zasmidium dicranopteridis* R. Kirschner, *Phytotaxa* 176: 319. 2014.

Description & Illustration — Kirschner & Liu (2014).

Specimen examined. TAIWAN, Taipei City, Wenshan District, Maokong, on fronds of *Dicranopteris linearis* var. *linearis*, 20 Oct. 2013, R. Kirschner (holotype TNM 3953, culture ex-type RoKi 3953).

Paramycosphaerella dicranopteridis-flexuosae Guatimosim, R.W. Barreto & Crous, *sp. nov.* — MycoBank MB812776; Fig. 12

Etymology. Name refers to the host species epithet, *Dicranopteris flexuosa*.

Description in planta — *Frond spots* amphigenous, irregular, starting as small dark brown spots, with a white centre adaxially, leading to the chlorosis of the pinnulet (particularly at the apex), and subsequently its necrosis, which become entirely brown to black, deformed, and often brittle, ascomata produced adaxially in a grey well-delimited area, coalescing and leading to the blight of entire pinnae. *Internal hyphae* branched, septate, intra- and intercellular, 1.5–5 µm wide, subhyaline to pale brown, smooth. *Ascomata* pseudothecial, epiphyllous, solitary,

subcuticular to erumpent, globose, (46–)74–98(–114) × (55–)84–95(–109) µm, walls of 3–4 layers of pale to dark brown *textura angularis*, cells 4–11.5 × 1.5–3.5 µm, ostiole central, 9–17 µm diam. *Asci* bitunicate, aparaphysate, fasciculate, sessile, 8-spored, obclavate to narrowly ellipsoid, straight or slightly curved, 24–51 × 10–18 µm, hyaline, smooth. *Ascospores* inordinate, overlapping, fusoid, straight, 10–19 × 2–4.5 µm, medianly 1-septate, tapering toward both rounded ends, guttulate, hyaline, thin-walled, smooth. *Ascospore germination* mostly from both ends, remaining hyaline, extending at an angle in reference to main ascospore apex, irregular in width, slightly distorted (mixture of Type G and K, Crous 1998).

Culture characteristics — Colonies on MEA, OA and PDA slow-growing, 23 mm diam after 24 d; raised, with lobate, undulate, feathery margins, and cottony aerial mycelium, iron-grey centrally, lavender grey at periphery; leaden black in reverse; On OA and PDA, slightly pigmenting the media, rosy vinaceous; cultures sterile.

Specimens examined. BRAZIL, Minas Gerais, Ouro Preto, Parque Municipal das Andorinhas, on fronds of *Dicranopteris flexuosa*, 25 Jan. 2014, P.B. Schwartzburd (holotype CBS H-22091, isotype VIC 43118, culture ex-type CPC 24743); *ibid.*, vicinity of the Parque Estadual do Itacolomi, on fronds of *Dicranopteris flexuosa*, 8 June 2013, E. Guatimosim, VIC 42475.

Notes — Morphologically, *Pa. dicranopteridis-flexuosae* is quite similar to *Pa. gleicheniae*, recorded on *D. linearis* from India, Malaysia and Taiwan (Kirschner & Liu 2014), but can



Fig. 12 *Paramycosphaerella dicranopteridis-flexuosae* (CPC 24743). a–c. Frond spots on *Dicranopteris flexuosa*; d. vertical section of the ascoma; e. asci; f. ascospores; g. germinating ascospores; h. culture on MEA; i. culture on OA; j. culture on PDA. — Scale bars = 10 µm.

be distinguished from the latter by having longer and wider asci (24–51 × 10–18 µm in *Pa. dicranopteridis-flexuosae* and 18–33 × 9–15 µm in *Pa. gleicheniae*) (Ramakrishnan & Ramakrishnan 1950). In fact, the two hosts, *D. flexuosa* and *D. linearis*, are also very similar and retained as two geographical entities: the former occurring only in the Neotropics, and the latter in the Paleotropics (Mickel & Smith 2004, Bingyang et al. 2013). Phylogenetically, only ITS sequence data is available for *Pa. gleicheniae* (Kirschner & Liu 2014), from which only 5 bp are different from *Pa. dicranopteridis-flexuosae*. Nevertheless, the tree produced in this study (Fig. 3) demonstrated that *Pa. gleicheniae* is quite distinct from *Pa. dicranopteridis-flexuosae*. Additional loci should be sequenced for the former species, aiming at clarifying the true species boundaries. At present, based on the host species, geographical distribution, and until additional loci have been studied, we decided to maintain them as distinct taxa. An asexual stigmata-like morph was observed on different specimens, collected in different seasons at the same place, being associated with similar symptoms to those caused by *Pa. dicranopteridis-flexuosae*. However, no cultures were obtained from this fungus and the connection between these two morphs needs to be confirmed.

Paramycosphaerella gleicheniae (T.S. Ramakr. & K. Ramakr.) Guatimosim, R.W. Barreto & Crous, *comb. nov.* — MycoBank MB812808

Basionym. *Mycosphaerella gleicheniae* T.S. Ramakr. & K. Ramakr., Proc. Indian Acad. Sci., B 32: 205. 1950.

Specimens examined. INDIA, Coonoor, Nilgiris, Tamil Nadu, on fronds of *Dicranopteris linearis* (= *Gleichenia linearis*), 29 May 1948, T.S. Ramakrishnan & K. Ramakrishnan (holotype presumably lost). — TAIWAN, New Taipei City, Yingge, trail to Yingge Rock, on fronds of *D. linearis*, 11 Apr. 2012, R. Kirschner (TNM 3613, culture RoKi 3613); Taoyuan County, Dasi (Daxi) Township, Weilliao Old Trail, 29 Sept. 2013, R. Kirschner (TNM 3945, culture RoKi 3945).

Notes — *Paramycosphaerella gleicheniae* was described from India, the holotype of which has presumably been lost (Aptroot 2006). The specimens examined here are from the same host, but from a different country (Taiwan), therefore inadequate to be used as neotype. However, despite the ascospores from the Taiwanese material being somewhat different from the type (Kirschner & Liu 2014), it is probable that they are conspecific. *Paramycosphaerella gleicheniae* still awaits neotypification.

Paramycosphaerella irregularis (Cheew. et al.) Guatimosim, R.W. Barreto & Crous, *comb. nov.* — MycoBank MB812824

Basionym. *Mycosphaerella irregularis* Cheew. et al., Persoonia 21: 82. 2008, as '*irregulari*'.

Description & Illustration — Cheewangkoon et al. (2008).

Specimen examined. THAILAND, Udonthani, on leaves of *Eucalyptus* sp., July 2007, R. Cheewangkoon (holotype CBS H-20135, culture ex-type CBS 123242).

Paramycosphaerella madeirensis (Crous & Denman) Guatimosim, R.W. Barreto & Crous, *comb. nov.* — MycoBank MB812825

Basionym. *Mycosphaerella madeirensis* Crous & Denman, Stud. Mycol. 50: 204. 2004, as '*madeirae*'.

Description & Illustration — Crous et al. (2004b).

Specimen examined. MADEIRA, Party Farm, on leaves of *Eucalyptus globulus*, Apr. 2000, S. Denman (holotype CBS H-9898, cultures ex-type CBS 112895, CBS 112301).

Paramycosphaerella nabiacense (Crous & Carnegie) Guatimosim, R.W. Barreto & Crous, *comb. nov.* — MycoBank MB812809

Basionym. *Zasmidium nabiacense* Crous & Carnegie, Persoonia 23: 142. 2009.

Description & Illustration — Crous et al. (2009c).

Specimen examined. AUSTRALIA, New South Wales, Nabiac, on leaves of *Eucalyptus* sp., 30 Nov. 2005, A.J. Carnegie (holotype CBS H-20273, cultures ex-type CBS 125010, CPC 12749, 12750).

Paramycosphaerella parkii (Crous et al.) Guatimosim, R.W. Barreto & Crous, *comb. nov.* — MycoBank MB812810

Basionym. *Mycosphaerella parkii* Crous et al., Mycol. Res. 97: 582. 1993. = *Stenella parkii* Crous & Alfenas, Mycologia 87: 121. 1995.

≡ *Zasmidium parkii* (Crous & Alfenas) Crous & U. Braun, Schlechtendalia 20: 102. 2010.

Descriptions & Illustrations — Crous et al. (1993), Crous & Alfenas (1995).

Specimen examined. BRAZIL, Aracruz Florestal nursery, on leaves of *Eucalyptus grandis*, 24 Feb. 1990, M.J. Wingfield (holotype PREM 50668, culture ex-type CBS 387.92, CMW 14775, STE-U 353).

Notes — The link between the sexual (PREM 50668, culture CBS 387.92) and asexual morph (PREM 51713) was based on morphology, and never corroborated by DNA sequence data. Because subsequent studies have revealed '*Mycosphaerella parkii*' to be a species complex (Crous et al. 2006b, Cheewangkoon et al. 2008), fresh collections are required to resolve the status of *Zasmidium parkii*.

Paramycosphaerella pseudomarksii (Cheew. et al.) Guatimosim, R.W. Barreto & Crous, *comb. nov.* — MycoBank MB812811

Basionym. *Mycosphaerella pseudomarksii* Cheew. et al., Persoonia 21: 83. 2008.

Description & Illustration — Cheewangkoon et al. (2008).

Specimen examined. THAILAND, Chiang Mai, Mae Tang, on leaves of *Eucalyptus* sp., June 2007, R. Cheewangkoon (holotype CBS H-20134, culture ex-type CBS 123241).

Paramycosphaerella sticheri Guatimosim, R.W. Barreto & Crous, *sp. nov.* — MycoBank MB812777; Fig. 13

Etymology. Name refers to the host genus from which it was isolated, *Sticherus*.

Description in planta — *Fron*d spots amphigenous, irregular, initially small and vein delimited along the pinnulets, black and dark brown intermixed areas, growing and leading to complete necrosis of the pinnula, sometimes causing blight of entire pinnule. *Internal hyphae* branched, septate, intra- and intercellular, 2–2.5 µm wide, subhyaline to pale brown, smooth. *Ascomata* pseudothecial, amphigenous, more abundant abaxially, solitary, subcuticular to erumpent, globose, (51–)60–96(–106) × 45–94 µm, walls of 2–3 layers of brown to dark brown *textura angularis*, cells 2.5–4 × 2–3 µm, black, ostiole central, 16–30 µm diam. *Asci* bitunicate, paraphysate, fasciculate, sessile, 8-spored, obpyriform, straight or slightly curved, 24–58 × 11–20 µm, hyaline, smooth. *Ascospores* inordinate, overlapping, fusoid, straight, 14–20 × 3–5.5 µm, medianly 1-septate, not to slightly constricted at the septum, tapering towards rounded ends, but more prominently towards the lower end, guttulate, hyaline, thin-walled, smooth. *Ascospores* germinating from both ends, remaining hyaline, germ tubes following the long axis of the spore, germ tubes irregular in width, slightly distorting, spores



Fig. 13 *Paramycosphaerella sticheri* (CPC 24720). a. Frond spots on *Sticherus penniger*; b. erumpent subcuticular ascomata, fruiting epiphyllous; c. vertical section of the ascoma; d. e. asci; f. ascospores; g. germinating ascospores; h. culture on MEA; i. culture on OA; j. culture on PDA. — Scale bars = 10 μ m.

becoming constricted at the septum (Type C, Crous 1998). *Asexual morph* not known.

Culture characteristics — Colonies on MEA and PDA slow-growing, 19 mm diam after 24 d; dome-shaped, lobate, with sharp margins and velvety aerial mycelium, pale mouse grey centrally, mouse grey at periphery; olivaceous grey reverse. On OA, surface pale mouse grey centrally, outer region lavender grey, with a distinct leaden black margin; greenish grey reverse; cultures sterile.

Specimen examined. BRAZIL, Santa Catarina, São Pedro de Alcântara, roadside, on fronds of *Sticherus penniger*, 17 Apr. 2013, E. Guatimosim (holotype CBS H-22093, isotype VIC 42498, culture ex-type CPC 24720, COAD 1422).

Notes — Morphologically, *Pa. sticheri* is rather similar to *Pa. dicranopteridis-flexuosae*, recorded on *Dicranopteris flexuosa* from Brazil (this study). Nevertheless, it can be distinguished from the latter species by having slightly narrower ascospores (2–4.5 μ m in the latter). Moreover, they are phylogenetically quite distinct from each other according to the following number of variable sites for each locus: 28 bp for *act*, 43 bp for ITS, 101 bp for *tef1* and 8 bp for LSU. Additionally, based on multi-gene phylogenetic inference (Fig. 3), *Pa. sticheri* grouped basal to other taxa in the genus, having *Pa. nabiacense* as sister clade.

Paramycosphaerella vietnamensis (Barber & T.I. Burgess)

Guatimosim, R.W. Barreto & Crous, *comb. nov.* — MycoBank MB812812

Basionym. *Mycosphaerella vietnamensis* Barber & T.I. Burgess, Fung. Diversity 24: 148. 2007.

Description & Illustration — Burgess et al. (2007).

Specimen examined. VIETNAM, South East Forestry Institute nursery, from leaves of *Eucalyptus grandis*, 6 July 2004, T.I. Burgess (holotype MURU411, ex-culture CBS 119974, CMW 23441).

Phaeophleospora Rangel, Arq. Mus. Nac. Rio de Janeiro 18: 162. 1916. — MycoBank MB9311

Phaeophleospora pteridivora Guatimosim, R.W. Barreto & Crous, *sp. nov.* — MycoBank MB812826; Fig. 14

Etymology. Name refers to the high degree of damage caused by the fungus on infected fronds.

Description in planta — *Frond spots* amphigenous, irregular, affecting almost all the pinnulets. Starting as small pale brown areas, usually close to the apex of the pinnulets, affecting the edges, which becomes distorted and brittle, spreading and becoming confluent, necrotic, leading to complete necrosis of the pinnulet. *External hyphae* absent. *Internal hyphae* branched, septate, intra- and intercellular, 1.5–3 μ m wide, dark brown,



Fig. 14 *Phaeophleospora pteridivora* (CPC 24683). a, b. Frond spots on *Serpocaulon triseriale*; c. erumpent subcuticular ascomata, fruiting epiphyllous; d. e. vertical section of the ascoma; f. conidiophores arising from the stroma; g. conidia; h. culture on MEA; i. culture on OA; j. culture on PDA. — Scale bars = 10 µm.

smooth. *Ascomata* pseudothecial, hypophyllous, solitary, subcuticular to erumpent, globose, $44\text{--}64 \times 42\text{--}61$ µm, wall of 3–4 layers of brown to dark brown *textura angularis* cells, $2\text{--}11 \times 2\text{--}8$ µm, black, ostiole central, $10\text{--}22$ µm diam. *Asci* bitunicate, aparaphysate, fasciculate, sessile, 8-spored, ellipsoidal to ovoid, straight or slightly curved, $15\text{--}25 \times 6\text{--}8$ µm, hyaline, smooth. *Ascospores* inordinate, overlapping, fusoid, straight, $1.5\text{--}12 \times 1\text{--}8$ µm, medianly 1-septate, not constricted at the septum, tapering towards rounded ends, with two large opposed guttules, hyaline, thin-walled, smooth. *Ascospore germination* not seen. *Asexual morph* cercosporoid, hypophyllous. *Stromata* subcuticular, erumpent, globose, $40\text{--}46 \times 50\text{--}54$ µm, composed of an aggregation of *textura angularis*, cells $4\text{--}5 \times 2\text{--}5$ µm, brown to dark brown, smooth. *Conidiophores* sporodochial, arising from the stroma, restricted to the conidiogenous cells, subcylindrical to ampuliform, straight, $5\text{--}25 \times 2\text{--}5$ µm, unbranched, aseptate, subhyaline to pale brown, smooth. *Conidiogenous cells* terminal, determined, unbranched, tapering to the apex, subhyaline to pale brown, smooth, scars inconspicuous, one per cell, not thickened, nor darkened. *Conidia* solitary, subcylindrical, curved to sinuous, $70\text{--}107 \times 2\text{--}3$ µm, tapering toward the acute apex, base truncate, $1.5\text{--}2.5$ µm diam at the base, 6–9-septate, guttulate, pale brown to olivaceous brown, smooth, scars not thickened, nor darkened.

Culture characteristics — Colonies on MEA slow-growing, 46 mm diam after 24 d; undulated, spreading, with lobate, fea-

they margins and sparse aerial mycelium, mouse grey centrally, pale mouse grey at periphery with a distinct narrow white external rim; greenish grey reverse. On OA, cream with a honey to buff periphery; iron-grey centrally with amber periphery reverse. On PDA, mouse grey with lavender grey periphery; mouse grey reverse centrally, amber periphery; cultures sporulating moderately on OA, producing conidia.

Specimen examined. BRAZIL, Rio de Janeiro, Cláudio Coutinho path, Praia Vermelha, Urca, humid rocks, on fronds of *Serpocaulon triseriale*, 3 Feb. 2012, R.W. Barreto (holotype CBS H-22097, isotype VIC 42559, culture ex-type CPC 24683, COAD 1182).

Notes — The genus *Phaeophleospora*, which is based on *Ph. eugeniae*, was collected from *Eugenia uniflora* (Myrtaceae) in Brazil (Crous et al. 1997) and clusters within *Mycosphaerellaceae* (Crous et al. 2007a). In the past, this genus included species that are presently accommodated in *Teratosphaeria* (= *Kirramyces*) and have pycnidial asexual morphs (Walker et al. 1992, Andjic et al. 2007). The new species described on *Serpocaulon triseriale* (Polypodiaceae) was based on material producing both the sexual and asexual morphs. Surprisingly, its asexual morph is a sporodochial hyphomycete (Fig. 3). Given the recent conidiomatal species with aseptate conidia described from ferns collected in Thailand (Crous et al. 2015b), the genus *Phaeophleospora* as presently defined based on DNA phylogeny, is morphologically rather diverse.

Pseudocercospora Speg., Anales Mus. Nac. Hist. Nat. Buenos Aires, Ser. 3, 13: 437. 1911

Pseudocercospora abacopteridicola J.M. Yen & Lim, Cah. Pacifique 17: 97. 1973. — Fig. 15

Description in planta — *Fron*d spots amphigenous, starting as minute, vein-delimited, pale brown spots, affecting random pinnales, leading to an extensive necrosis of entire pinnae, which then become dark brown to black, with a central area white to grey. *Caespituli* hypophyllous, abundant. *External hyphae* branched, septate, arising from the stomata, 1.6–2.5 µm wide, pale to medium brown, smooth. *Internal hyphae* indistinct. *Stromata* absent. *Conidiophores* arising from the hyphae, hypophyllous, restricted to the conidiogenous cells. *Conidiogenous cells* terminal, holoblastic, subcylindrical, straight, geniculate, 5–5.5 × 2–2.5 µm, unbranched, aseptate, pale brown, smooth, scars indistinct. *Conidia* solitary, subcylindrical, straight or curved, (25–)45–66(–77) × 1.8–3 µm, rounded apex, base subtruncate, 2–8-septate, guttulate, pale brown, smooth; hila not thickened, nor darkened, 1–3 µm diam.

Culture characteristics — Colonies on MEA slow-growing, 26 mm diam after 20 d in the dark; surface smooth, raised with dense aerial mycelium and even margins, olivaceous grey in the centre, followed by a pale olivaceous grey ring and greenish black periphery; iron-grey reverse; cultures sterile.

Specimen examined. BRAZIL, Minas Gerais, Cachoeira do Campo, Café Retiro Novo, on fronds of *Adiantum* sp., 12 Nov. 2012, E. Guatimosim (CBS H-22098, culture CPC 24709).

Notes — *Pseudocercospora abacopteridicola* was only known from the type specimen, collected on *Abacopteris urophylla* (*Thelypteridaceae*) from Singapore (Yen & Lim 1980, Braun et al. 2013). The specimen collected in Brazil was found on a distantly related host – *Adiantum* sp. (*Pteridaceae*). However, as morphology and biometric data are indistinguishable, instead of describing the fungus from Brazil as new, we prefer to place it in *Ps. abacopteridicola* until DNA of the fungus from Singapore becomes available for a molecular comparison.

Pseudocercospora brackenicola Guatimosim, R.W. Barreto & Crous, sp. nov. — MycoBank MB812813; Fig. 16

Etymology. Name refers to bracken – the common English name for species of *Pteridium*.

Description in planta — *Fron*d spots, amphigenous, irregular, starting as small, dark brown vein delimited spots at pinnulet margins, spreading and becoming black with age and occasionally reaching the entire pinnulet. *Caespituli* hypophyllous, abundant. *External hyphae* hypophyllous, arising from a tuft through the stomata and spreading, slightly branched, septate, pale brown, smooth. *Internal hyphae* intra- and intercellular, septate, branched, 1.4–3.5 µm, subhyaline to pale brown, smooth. *Stromata* rudimentary, inside the stomatal cavity, irregular, 24.5–56.5 × 11.5–25.5 µm, composed of a few globose cells, pale brown. *Conidiophores* hypogenous, arising through the stomata, producing dense fascicles, up to 20 conidiophores per fascicle, subcylindrical, straight to curved, often geniculate at the tip, 11–29.5 × 2–3 µm, branched, mostly aseptate, rarely 1–2-septate, eguttulate, pale brown, smooth. *Conidiogenous cells* terminal, integrated, holoblastic, subcylindrical, sympodial, 4.5–17 × 2–3 µm, pale brown, smooth, scars indistinct, 1 per cell, discoid, c. 2 µm diam, not thickened, nor darkened. *Conidia* solitary, obclavate to subcylindrical, straight, curved, or sinuous, 20–77 × 1–2 µm, rounded apex, base truncate, 1–6-septate, guttulate, pale brown, smooth; hila not thickened, nor darkened, 1–2 µm diam.

Culture characteristics — Colonies on MEA slow-growing, 30 mm diam after 20 d in the dark; raised with velvety aerial mycelium, pale greenish grey centrally, and mouse grey at periphery; olivaceous grey in reverse; cultures sterile.

Specimens examined. BRAZIL, Minas Gerais, Capitólio, Furnas, Rio do Turvo Inn, in front of the announcement board of Clube Náutico, on fronds of *Pteridium arachnoideum*, 9 Nov. 2012, R.W. Barreto (holotype CBS H-22101, isotype VIC 42588, culture ex-type CPC 24695).

Notes — Phylogenetically, *Ps. brackenicola* clusters with *Ps. purpurea* and *Ps. sordida* as sister clade (Fig. 2), but differs from them by having the following number of variable sites for each locus: *Ps. purpurea* (7 bp for ITS, 9 bp for *act*, 24 bp

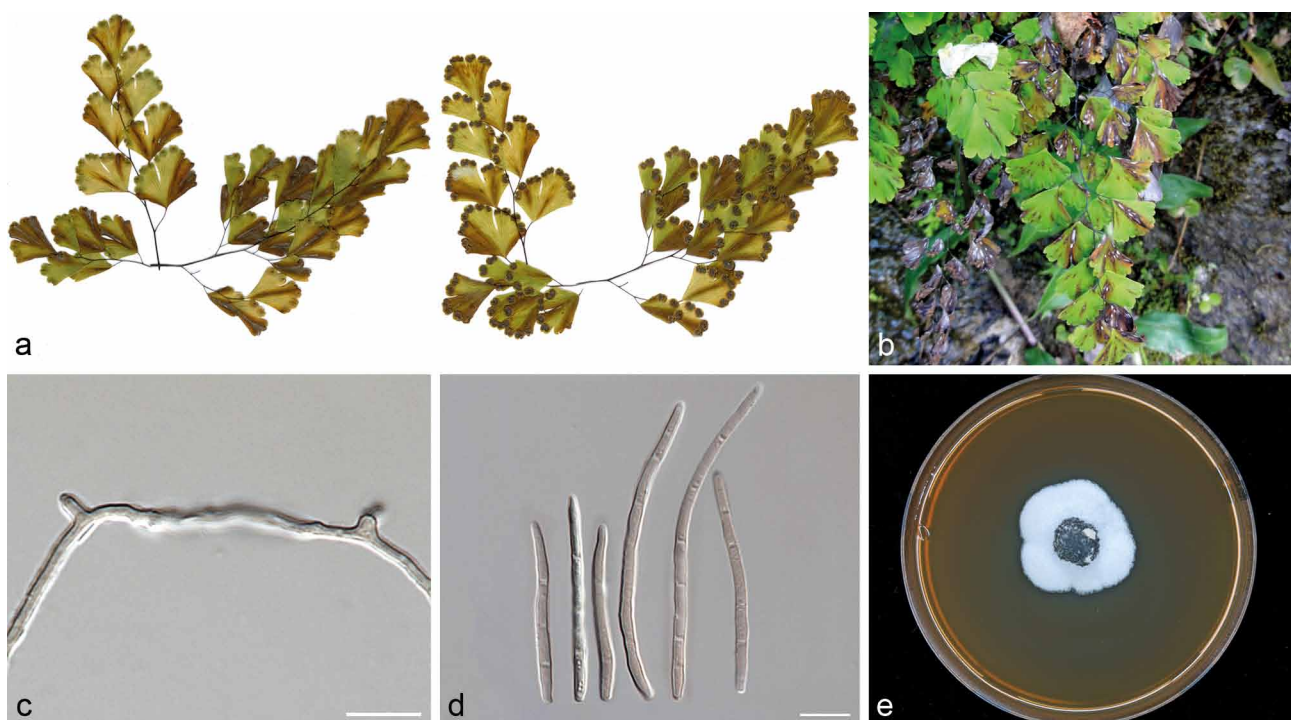


Fig. 15 *Pseudocercospora abacopteridicola* (CPC 24709). a, b. Frond spots on *Adiantum* sp.; c. conidiophores restricted to the conidiogenous cells, arising from the hyphae; d. conidia; e. culture on MEA. — Scale bars = 10 µm.

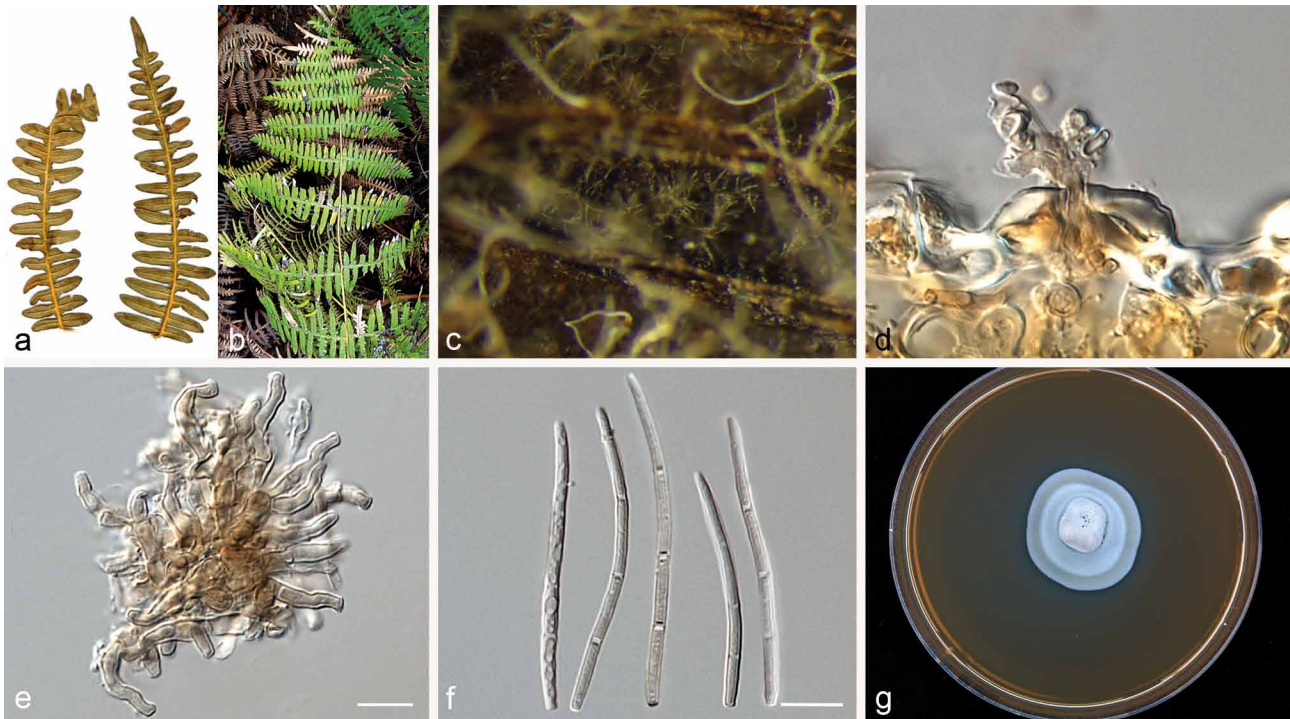


Fig. 16 *Pseudocercospora brackenicola* (CPC 24709). a, b. Frond spots on *Pteridium arachnoideum*; c. conidia sporulating abaxially; d. detail of conidiophores arising through the stoma; e. conidiophores; f. conidia; g. culture on MEA. — Scale bars = 10 μ m.

for *tef1*) and *Ps. sordida* (8 bp for ITS, 14 bp for *act*, 33 bp for *tef1*). Morphologically, both species are clearly different from *Ps. brackenicola* by having larger conidiophores (20–200 \times 3.5–4.5 μ m in *Ps. purpurea* and 20–90 \times 3.5–5 μ m in *Ps. sordida*) and larger conidia (20–100 \times 2–4.5 μ m in *Ps. purpurea* and 20–165 \times 3–5.5 μ m in *Ps. sordida*) (Chupp 1954, Guo & Hsieh 1995). Additionally, the hosts of *Ps. purpurea* and *Ps. sordida* are higher plant families in the *Perseaceae* and *Bignoneaceae*, respectively (Farr & Rossman 2015). *Pseudocercospora brackenicola* is similar to *Ps. davallicola* (described on *Davallia fejeensis* from Brazil) and to *Ps. lonchitidis* (described on *Lonchitis hirsuta* from Venezuela) (Braun et al. 2013). Molecular data are lacking for both species, but there are various morphological differences that distinguish them. Firstly, the conidia in *Ps. davallicola* can be formed in short chains (absent in *Ps. brackenicola*), and the conidiophores of *Ps. davallicola* are solitary, whereas on *Ps. brackenicola* they form fascicles emerging from stomata, through stomata (Braun et al. 2013). Secondly, *Ps. lonchitidis* has erumpent, well-developed stomata (loosely dense, emerging through the stoma in *Ps. brackenicola*), straight and thicker conidiophores, 3–5 μ m wide in *Ps. davallicola* (curved to sinuous, 2–3 μ m wide in *Ps. brackenicola*), and conidiogenous loci are subdenticulate (inconspicuous in *Ps. brackenicola*) (Braun et al. 2013). This is the first record of a *Pseudocercospora* sp. on the genus *Pteridium*. *Pseudocercospora brackenicola* causes a damaging disease on its host (bracken), which is a highly noxious weed. Further investigations are required to determine its potential role as biological control agent.

Pseudocercospora lygodiicola Y.L. Guo & U. Braun, IMA
Fungus 4: 317. 2013. — Fig. 17

Description in planta — *Frond spots* amphigenous, irregular, starting from the main vein and spreading until the edges of the pinnulets, becoming centrally cream and necrotic, with a distinct dark brown to black halo. *Caespituli* hypophyllous, abundant. *External hyphae* absent. *Internal hyphae* intra- and intercellular, 1.5–3.5 μ m wide, septate, branched, pale brown, smooth. *Stomata* rudimentary, arising from the stomatal cavity, subglobose,

composed of *textura angularis*, 22–70 μ m diam, dark brown, cells 3–7 \times 2.5–3 μ m. *Conidiophores* arising from stomata, hypophyllous, forming small fascicles (up to 15), subcylindrical, sinuous or curved, geniculate towards the apex, 26–80 \times 3–5 μ m, unbranched, 3–6-septate, eguttulate, pale brown, smooth. *Conidiogenous cells* terminal, holoblastic, subcylindrical, attenuated at the tip, 3–18 \times 2–4 μ m, subhyaline, smooth, scars inconspicuous, 1 per cell, subdenticulate, 1–3.5 μ m, not thickened, nor darkened. *Conidia* solitary, obclavate, curved or sinuous, 43–117 \times 2.5–4.5 μ m, tapering toward rounded apex, base obconically truncate, 6–12-septate, guttulate, pale brown, smooth; hila not thickened, nor darkened, 1–4 μ m diam.

Culture characteristics — Colonies on MEA slow-growing, reaching 32 mm diam after 20 d in the dark; centrally raised, and flat at periphery, aerial mycelium cottony, dry, iron-grey combined with olivaceous grey areas centrally, olivaceous grey towards periphery; reverse olivaceous black centrally and olivaceous grey at periphery; cultures sterile.

Specimen examined. BRAZIL, Rio de Janeiro, BR-116 Highway, near to Parque Nacional Serra dos Órgãos, roadside, on fronds of *Lygodium volubile*, 14 June 2014, R. W. Barreto (VIC 42917, cultures CPC 25755, COAD 1745).

Notes — There are four species of *Pseudocercospora* known from *Lygodium*, namely *Ps. lygodi* (on *L. japonicum* from Taiwan), *Ps. lygodiicola* (on *L. japonicum* from China), *Ps. lygodiigena* and *Ps. polypodiacearum* (both on *Lygodium* sp. from India) (Braun et al. 2013). Species boundaries among these taxa are based on morphological and biometric characters, which could be considered as tentative, as the host and distribution range of these taxa are quite similar. Currently there are no records of ex-type cultures or DNA information on any of these taxa.

The fungus isolated from *L. volubile* in Brazil has morphological and biometric data similar to *P. lygodiicola*, but until the latter has been epitypified, we decided to extend its host range, rather than propose a new name for the Brazilian collection. Phylogenetically, *Ps. lygodiicola* clusters in the same clade with three other species isolated from ferns, namely *Ps. cyatheicola*, *Ps. rumohrae* and *Ps. thelypteridis* (Fig. 2).

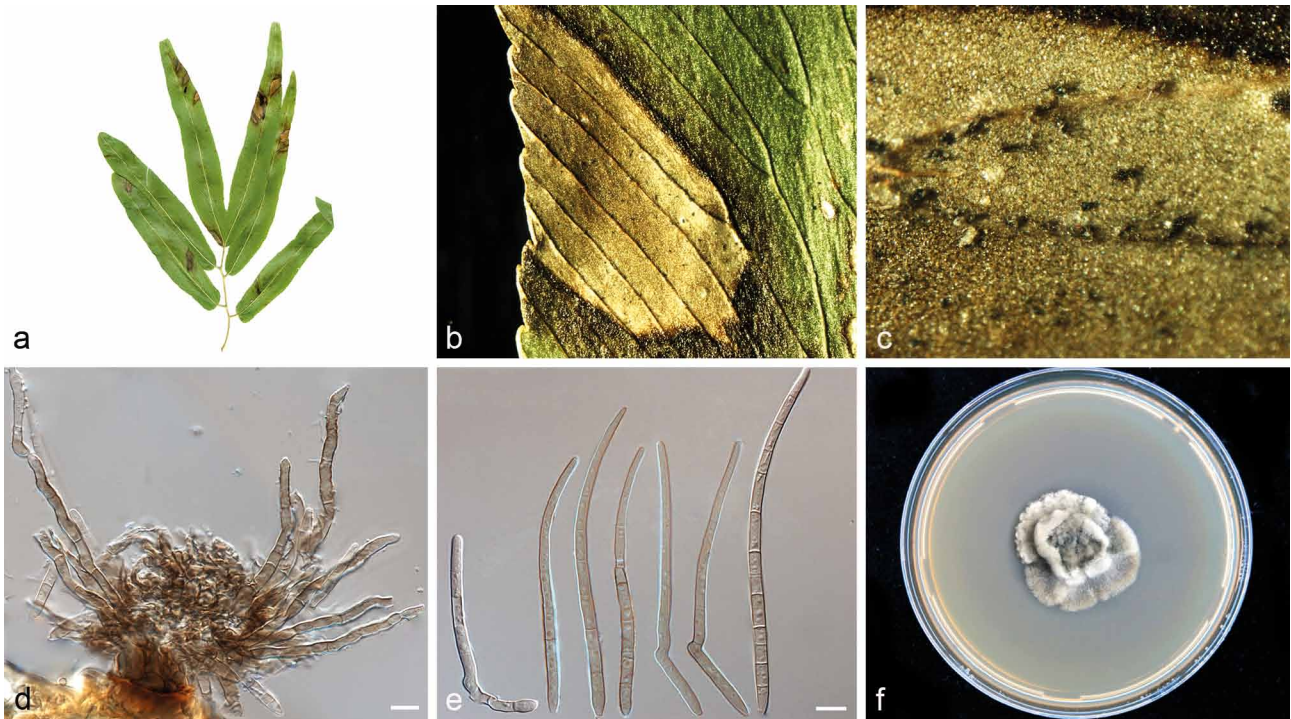


Fig. 17 *Pseudocercospora lygodiicola* (CPC 25755). a, b. Frond spots on *Lygodium volubile*; c. conidiophores sporulating adaxially; d. conidiophores arising from the stroma through the stoma; e. conidia; f. culture on MEA. — Scale bars = 10 µm.

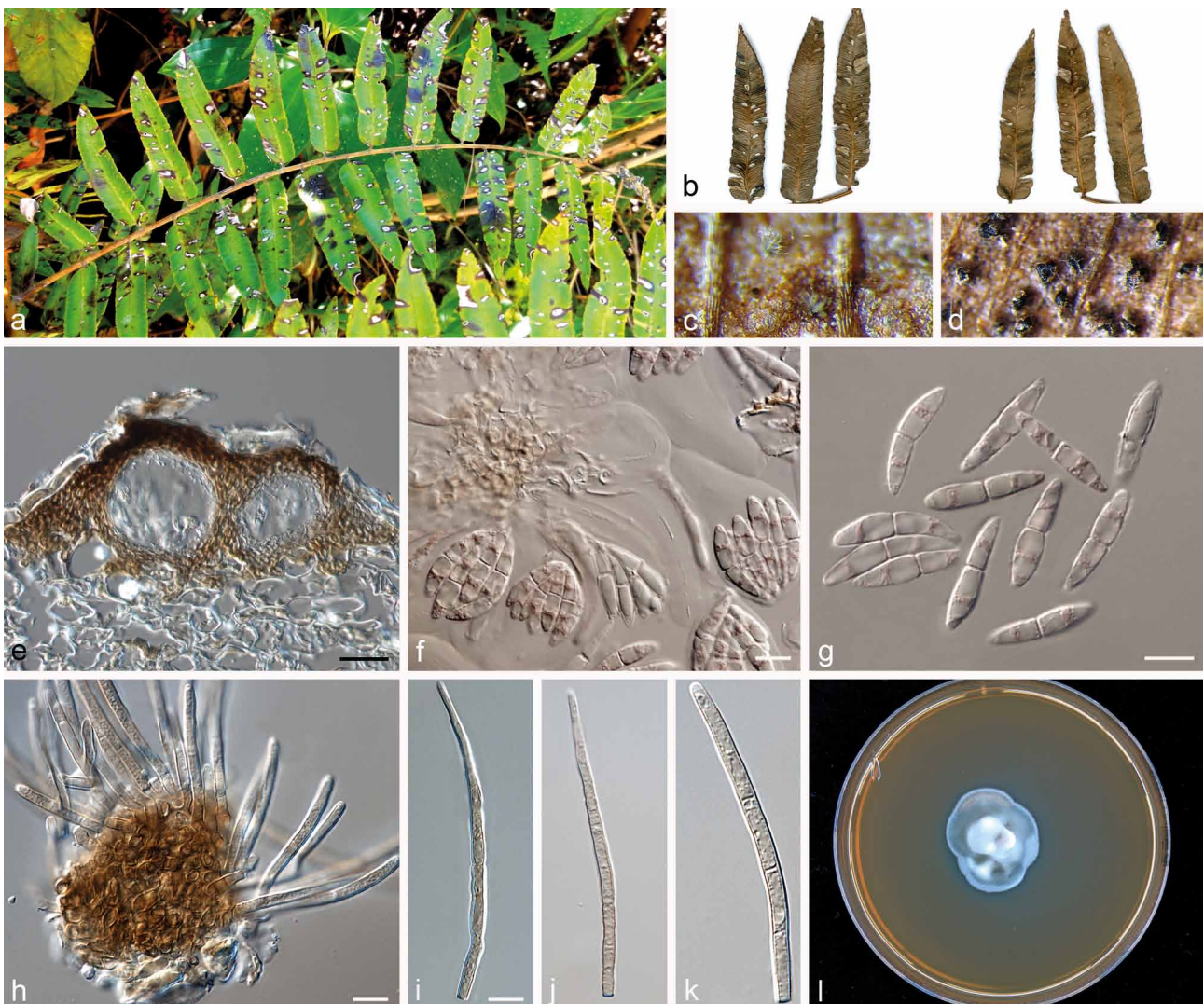


Fig. 18 *Pseudocercospora paranaensis* (asexual morph COAD 1180, sexual morph CPC 24680). a, b. Frond spots on *Cyathea atrovirens*; c. conidia sporulating abaxially; d. erumpent subcuticular ascomata, fruiting epiphyllous; e. vertical section of the ascoma; f. asci; g. ascospores; h. conidiophores arising from the stroma; i–k. conidia; l. culture on MEA. — Scale bars = 10 µm.

Pseudocercospora paranaensis Guatimosim, R.W. Barreto & Crous, *sp. nov.* — MycoBank MB812814; Fig. 18

Etymology. Name refers to the state in Brazil from where the fungus was collected, Paraná.

Fronde spots amphigenous, firstly irregular, vein delimited, pale brown to black, distributed along the pinnules, becoming circular, white to grey at the centre, with a brown to black halo sometimes perforated centrally leading to necrosis of the whole pinnule, and occasionally whole pinnae. *External hyphae* absent. *Internal hyphae* intra- and intercellular, septate, branched, 1–2 µm wide, hyaline, smooth. *Ascomata* pseudothecial, hypophyllous, solitary to confluent, subepidermal to erumpent, globose to subglobose, 40–80 × 45–73.5 µm, walls of 2–3-layers of *textura angularis*, medium brown to dark, 9.5–32 µm thick, ostiole central, c. 39 µm diam. *Asci* bitunicate, paraphysate, fasciculate, sessile, 8-spored, fusoid-ellipsoidal when immature and pyriform at maturity, straight or slightly curved, 40–75 × 13–30 µm, hyaline, smooth. *Ascospores* biserial to inordinate, overlapping, fusoid, straight, 18–27 × 3.5–6 µm, unequally 1-septate, slightly constricted at the septum, tapering towards rounded ends, with two large opposed guttules, hyaline, thin-walled, smooth. *Ascospore germination* not observed. *Asexual morph:* *Caespituli* hypophyllous, abundant. *Stromata* subsuperficial, globose, composed of dark brown *textura globulosa*, 26–39 × 15–31.5 µm. *Conidiophores* arising from the stroma, hypophyllous, sporodochial, restricted to the conidiogenous cells, ampuliform, swollen at the base, 7–11 × 1.5–2 µm, unbranched, aseptate, eguttulate, pale brown, smooth; scars, 2 µm wide, neither thickened, nor darkened. *Conidia* solitary, subcylindrical or obclavate, curved or rarely straight, 79–99 × 2–3 µm, rounded to obtuse apex, base truncate, 3–9-septate, guttulate, pale brown, smooth; hila not thickened, nor darkened, sometimes slightly darkened and slightly refractive, 1–2 µm diam.

Culture characteristics — Colonies on MEA slow-growing, 28 mm diam after 20 d in the dark; smooth with even margins, raised, aerial mycelium velvety, surface olivaceous grey, mixed with pale olivaceous grey; iron-grey in reverse; cultures sterile.

Specimens examined. BRAZIL, Paraná, Piraquara, Mananciais da Serra, on fronds of *Cyathea atrovirens*, sexual morph, 2 Feb. 2012, R.W. Barreto (holotype CBS H-22099, isotype VIC 42559, culture ex-type CPC 24680); *ibid.*, asexual morph, 2 Feb. 2012, R.W. Barreto (VIC 42558, culture COAD 1180).

Notes — Both morphs (sexual and asexual) were found occurring hypophyllously, on different fronds from the same host. *Pseudocercospora paranaensis* clusters in an isolated clade (Fig. 2), having *Ps. basitruncata* as sister clade. Besides, *Ps. basitruncata* is known to be an extremely variable species, some features remaining relatively constant such as the irregular annellations on the conidiogenous cells, and the conidial shape. Smaller conidia tend to be cylindrical, whereas larger conidia are tapered to more obtuse apices (Crous 1998). *Pseudocercospora paranaensis* does not have any annellations on its conidiogenous cells, which proliferate sympodially instead. Additionally, *Ps. paranaensis* differs from *Ps. basitruncata* by having significantly smaller conidiophores (7–11 µm in the former and 12–60 µm in the latter) and longer conidia (79–99 µm in the former and 45–70 µm in the latter). Finally, *Ps. basitruncata* is only known from an unrelated species of *Eucalyptus* (Hunter et al. 2011, Crous et al. 2013a).

Two other species of *Pseudocercospora* have already been recorded on members of *Cyatheaceae*, namely *Ps. cyatheae* described on *Cyathea* sp. from Japan and *Ps. cyatheicola* on *Cyathea australis* from Australia (Braun et al. 2013). With regards to *Ps. cyatheae*, the only sequence available in GenBank for this species is of the ITS region. *Pseudocercospora paranaensis*

differs from *Ps. cyatheae* in ITS and clusters in a separate and highly supported clade (data not shown). Nevertheless, morphological criteria alone clearly separate the two species. *Pseudocercospora cyatheae*, in contrast to *Ps. paranaensis*, has epiphyllous caespituli, its conidiogenous cells have a rim-like thickening at the scars, and it also has thicker, cylindrical to obclavate conidia (30–50 × 3.7–5.5 µm) with rounded bases (Nakashima et al. 2006). *Pseudocercospora cyatheicola* is different from *Ps. paranaensis* both phylogenetically – grouping in a different clade of the tree (Fig. 2) – and morphologically – having amphigenous stromata, larger conidiophores (30–70 × 2–3 µm), and percurrently proliferating conidiogenous cells (Crous et al. 2011).

Pseudocercospora serpocauloncola Guatimosim, R.W.

Barreto & Crous, *sp. nov.* — MycoBank MB812815; Fig. 19

Etymology. Name refers to the host genus from which it was isolated, *Serpocaulon*.

Description in planta — *Fronde spots* amphigenous, irregular, firstly concentrated next to the main vein and progressively spreading towards the margins of the pinnule, centrally pale brown, becoming dark brown towards the periphery. *Caespituli* epiphyllous, abundant. *External hyphae* absent. *Internal hyphae* intra- and intercellular, 1–2.5 µm wide, branched, septate, subhyaline to pale brown, smooth. *Stromata* rudimentary, subcuticular, composed of pale brown *textura angularis*, 15–36.5 µm wide, pale brown, smooth. *Conidiophores* restricted to the conidiogenous cell, arising from the stromata, epiphyllous, forming loose fascicles with up to 15 stalks, subcylindrical, attenuated at the tip, sinuous, often geniculate, 7–22 × 2–3.5 µm, unbranched, 0–1-septate, eguttulate, subhyaline to pale brown, smooth, scars inconspicuous, 1 per cell, not thickened, nor darkened. *Conidia* solitary, subcylindrical to obclavate, straight or curved, 31–75 × 2–3.5 µm, apex attenuated, base obconically truncate, 2–7-septate, guttulate, pale brown, smooth; hila not thickened, nor darkened, 2–4 µm diam.

Culture characteristics — Colonies on MEA slow-growing, 31 mm diam after 20 d in the dark; flat, aerial mycelium cottony, with water droplets at periphery, pale olivaceous-grey combined lavender grey areas centrally, greenish grey towards periphery; olivaceous black centrally and olivaceous grey at periphery in reverse; cultures sterile.

Specimens examined. BRAZIL, Rio de Janeiro, Gávea, Parque da Cidade, on fronds of *Serpocaulon triseriale*, 14 June 2014, R.W. Barreto (holotype CBS H-22105, cultures ex-type CPC 25077, COAD 1866).

Notes — *Pseudocercospora serpocauloncola* clustered within a new clade, together with an isolate recorded on *Eucalyptus grandis* from South Africa (CBS 110998), *Ps. cordiana*, *Ps. paraguayensis* and *Ps. schizolobii* (Fig. 2), but differs from them by having the following variable sites for each locus: *Pseudocercospora* sp. (9 bp for ITS), *Ps. cordiana* (30 bp for ITS, 1 bp for *act*, 1 bp for *tef1*), *Ps. paraguayensis* (6 bp for ITS, 1 bp for *act*, 4 bp for *tef1*) and *Ps. schizolobii* (5 bp for ITS, 5 bp for *act*, 4 bp for *tef1*). Morphologically, it was not possible to compare the present collection to *Pseudocercospora* sp. (CBS 110998), as the fungarium specimen was in poor condition, and neither conidiophores nor conidia were seen. Moreover, the cultures proved to be sterile. Two other *Pseudocercospora* species known on ferns (for which no DNA data are available in GenBank) have a similar morphology to *Ps. serpocauloncola*. These are *Ps. microsori* on *Microsorium pustulatum* from Australia, and *Ps. phyllitidis*, which occurs on various ferns belonging to different families, and has a cosmopolitan distribution (Shivas et al. 2010, Braun et al. 2013). *Pseudocercospora microsori* differs from *Ps. serpocauloncola* by having well-developed stromata (20–60 µm wide), longer (30–65 × 3–5

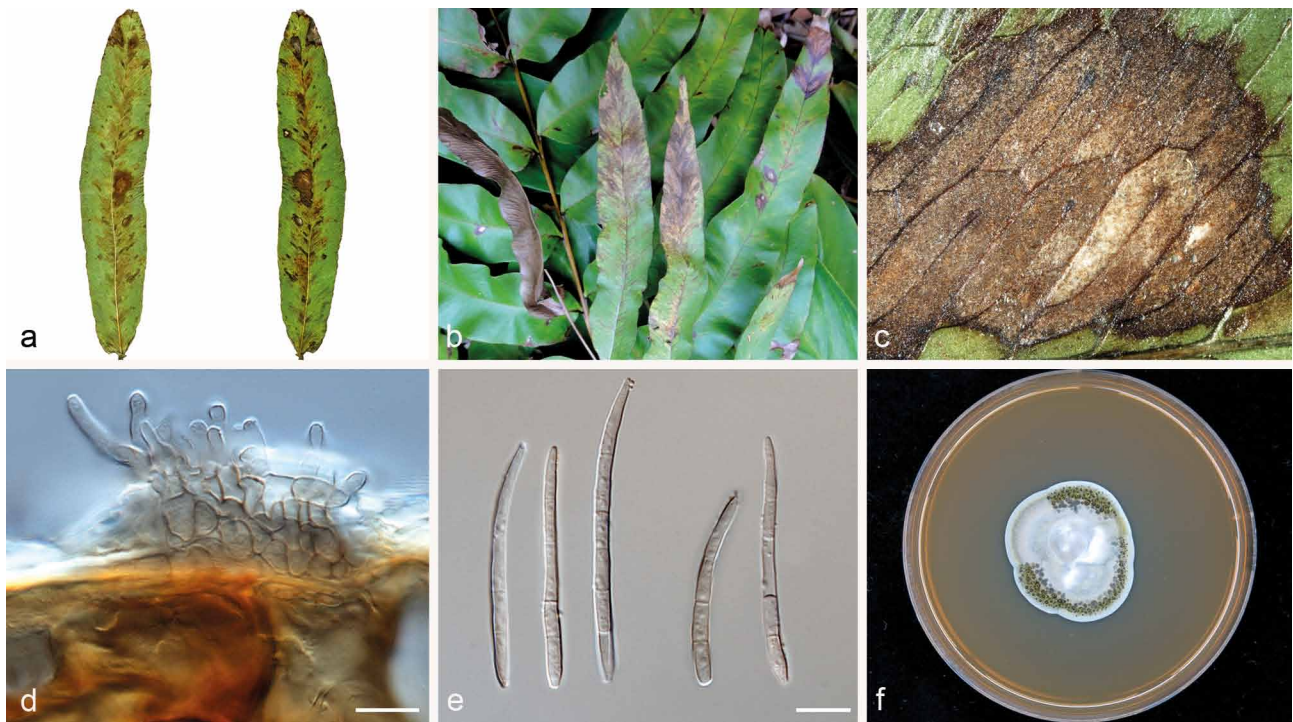


Fig. 19 *Pseudocercospora serpocaulonicola* (CPC 25077). a–c. Frond spots on *Serpocaulon triseriale*; d. conidiophores arising through the stoma; e. conidia; f. culture on MEA. — Scale bars = 10 μ m.

μ m), densely fasciculate (5–30 stalks per fascicle), red-brown conidiophores, and moderately wide (2.5–4 μ m), curved to flexuous conidia (Shivas et al. 2010). On the other hand, *Ps. phyllitidis* is known to be an extremely variable species and probably is polyphyletic. However, one distinctive feature that remains relatively constant for specimens belonging to this species is the persistency of the conidia, which remain attached to the conidiogenous cells for a long time (Braun et al. 2013). This feature is absent in *Ps. serpocaulonicola*. Additionally, *Ps. phyllitidis* has immersed stromata (ill-formed and subcuticular in *Ps. serpocaulonicola*) and moderately wider conidiophores

(1.5–4 μ m), compared to *Ps. serpocaulonicola* (2–3.5 μ m) (Braun et al. 2013). This is the first record of a fungus causing disease on *S. tritseriale*.

Pseudocercospora thelypteridis Goh & W.H. Hsieh, Trans. Mycol. Soc. Repub. China 4: 30. 1989. — Fig. 20

Description in planta — *Frond spots* amphigenous, irregular, starting from the main vein and spreading until the edges of the pinnulets, dark brown to black, sometimes reaching the entire pinnule. *Caespituli* hypophyllous, abundant. *External*



Fig. 20 *Pseudocercospora thelypteridis* (CPC 24676). a–d. Frond spots on *Thelypteris* sp.; e. conidiophores arising from the stroma; f–h. conidia; i. culture on MEA. — Scale bars = 10 μ m.

hyphae absent. *Internal hyphae* intra- and intercellular, septate, branched, subhyaline, smooth. *Stromata* subepidermal, discoid, composed of *textura angularis*, $19 \times 44.5 \mu\text{m}$, pale to dark brown. *Conidiophores* arising from stromata, reduced to the conidiogenous cells, hypophyllous, forming dense fascicles (more than 40 stalks per fascicle), subcylindrical, attenuated at the tip, straight, $14\text{--}23 \times 2.5\text{--}4 \mu\text{m}$, unbranched, aseptate, eguttulate, subhyaline, smooth, scars inconspicuous, 1 per cell, $2\text{--}2.5 \mu\text{m}$, not thickened, nor darkened. *Conidia* solitary, subcylindrical to acicular, straight or slightly curved, $65\text{--}96 \times 2.5\text{--}4 \mu\text{m}$, obtuse to round apex, base truncate, 5–8-septate, guttulate, subhyaline, smooth; hila not thickened, nor darkened, $2\text{--}2.5 \mu\text{m}$ diam.

Culture characteristics — Colonies on MEA slow-growing, 41 mm diam after 20 d in the dark; surface smooth with even margins, flat, cottony aerial mycelium, surface olivaceous grey mixed with zones of pale olivaceous grey; iron-grey reverse; cultures sterile.

Specimen examined. BRAZIL, Rio de Janeiro, Nova Friburgo, Mury, near a waterfall, growing over humid rocks, on fronds of *Thelypteris* sp., 5 Nov. 2011. R.W. Barreto (VIC 42569, CBS H-22102, culture CPC 24676).

Notes — *Pseudocercospora thelypteridis* clusters basal to a clade including several species of *Pseudocercospora*, e.g. *Ps. balsaminae*, *Ps. crocea*, *Ps. dianellae*, *Ps. humuli-japonici*, *Ps. humulicola*, *Ps. plectranthi*, *Ps. profusa* and *Ps. rhabdothamni*, while *Ps. cyatheicola* and *Ps. rumohrae* clusters basal to *Ps. thelyp-*

teridis (Fig. 2). However, *Ps. cyatheicola* is different from *Ps. thelypteridis* by having erumpent and amphigenous stromata, longer and narrower conidiophores ($30\text{--}70 \times 2\text{--}3 \mu\text{m}$), percurrently proliferating conidiogenous cells, and pale brown conidia (Crous et al. 2011). *Pseudocercospora rumohrae* differs from the new species by the absence of stromata, with conidiophores arising directly from the hyphae, as well as longer and narrower conidia ($60\text{--}120 \times 3\text{--}3.5 \mu\text{m}$) (Braun et al. 2013).

Pseudocercospora thelypteridis is known from the type material on *Thelypteris laxa* from Taiwan and China, and on *Nephrolepis* sp. from Brunei (Braun et al. 2013). However, as the morphology and biometric data are quite similar, we chose not to introduce a novel species for the fungus found in Brazil. This is the first record of *P. thelypteridis* from Brazil.

Pseudocercospora trichogena Guatimosim, R.W. Barreto & Crous, *sp. nov.* — MycoBank MB812827; Fig. 21

Etymology. Name derived from the trichomata habit of the species.

Description in planta — *Frond spots* on *Deparia petersenii*, amphigenous, evident adaxially, irregular, pale brown with necrotic fertile centre and distinctive black halo. *Ascromata* pseudothecial, epiphyllous, solitary, subepidermal to erumpent, globose to subglobose, $42\text{--}81 \times 37\text{--}60 \mu\text{m}$, walls of 2–3 layers of brown to dark brown *textura angularis*, cells $3\text{--}4 \times 2\text{--}3 \mu\text{m}$, black, ostiole central, $12\text{--}25 \mu\text{m}$ diam. *Asci* bitunicate, paraphysate,



Fig. 21 *Pseudocercospora trichogena* (asexual morph CPC 24664, sexual morph CPC 24670). a. Frond spots on *Deparia petersenii*; b. frond spots on *Macrothelypteris torresiana*; c. erumpent subcuticular ascromata, fruiting epiphyllous; d. conidia sporulating on a trichoma, hypophyllous; e. asci; f. ascospores; g. detail of the external hyphae arising through the stoma, and growing along the trichoma; h. conidiophores; i. j. conidia; k. culture on MEA. — Scale bars = 10 μm .

sessile, 8-spored, fusoid-ellipsoidal when immature, pyriform at maturity, curved, $26\text{--}42 \times 8\text{--}14 \mu\text{m}$, hyaline, smooth. *Ascospores* biseriate to inordinate, overlapping, fusoid, straight, $9\text{--}15 \times 2\text{--}4 \mu\text{m}$, 1-septate, with one cell larger than the other, tapering towards rounded ends, guttulate, hyaline, thin-walled, smooth. *Ascospore* germination not observed. *Asexual morph*: Frond spots on *Macrothelypteris torresiana*, amphigenous, irregular, starting from the main vein of the pinnulet, and spreading towards the edge, initially pale brown, becoming dark and necrotic. *Caespituli* hypophyllous, abundant on trichomata. *External hyphae* hypophyllous, abundant, often erupting through the cuticle, rarely arising through the stoma, and growing along the trichoma, spreading and covering the entire lesion, $2\text{--}3 \mu\text{m}$ wide, branched, septate, pale brown, smooth. *Internal hyphae* intra- and intercellular, abundant, $1\text{--}3 \mu\text{m}$ wide, prominently branched, septate, subhyaline, smooth. *Stromata* absent. *Conidiophores* arising from external hyphae, hypophyllous, often reduced to conidiogenous cells, formed in groups on trichomata, subcylindrical, attenuated at the tip, straight or sinuous, $19\text{--}74 \times 5\text{--}6 \mu\text{m}$, often branched, 1–5-septate, eguttulate, pale brown to brown, smooth. *Conidiogenous cells* terminal, integrated, holoblastic, subcylindrical, determinate, $10\text{--}35 \times 5\text{--}6 \mu\text{m}$, pale brown to brown, smooth, scars inconspicuous, 1 per cell, $1\text{--}2 \mu\text{m}$, not thickened, nor darkened. *Conidia* solitary, obclavate, straight or curved, $72\text{--}147 \times 3\text{--}5 \mu\text{m}$, apex rounded, base truncate, 4–13-septate, guttulate, pale brown, smooth; hila not thickened, nor darkened, $1\text{--}2 \mu\text{m}$ diam.

Culture characteristics — Colonies on MEA slow-growing, $10\text{--}23 \text{ mm}$ diam after 20 d in the dark; smooth to folded or concentrically folded, raised, aerial mycelium cottony or velvety, mouse grey, pale olivaceous grey or lavender grey; purplish grey or iron-grey in reverse; cultures sterile.

Specimens examined. BRAZIL, Rio de Janeiro, Nova Friburgo, Limeira, on fronds of *Macrothelypteris torresiana*, asexual morph, 13 June 2011, R.W. Barreto (holotype CBS H-22104, isotype VIC 42542, cultures ex-type CPC 24664, COAD 1087); Rio de Janeiro, Faz. Barreto II, Alto do Micheis, Riograndina, reforestation area, on fronds of *Deparia petersenii*, sexual morph, 13 June 2011, R.W. Barreto, (CBS H-22103, VIC 42546, cultures CPC 24670, COAD 1088).

Notes — Sexual and asexual morphs of *Ps. trichogena* were found in the same region but on different hosts. However, based on DNA phylogenetic analyses, there is no doubt that they belong to the same species. Phylogenetically, *Ps. trichogena* clusters in a highly diverse clade, differing from all species within it (Fig. 2). Morphologically, *Ps. trichogena* is similar to three other species recorded on *Thelypteridaceae*, namely *Ps. abacopteridicola* on *Abacopteris urophylla* from Singapore, *Ps. pteridophytophila* on *Cyclosorus acuminatus* from Asia and *Ps. thelypteridis* on *Nephrolepis* sp. and *Thelypteris laxa* from Asia (Braun et al. 2013, Farr & Rossman 2015). Among those, *Ps. pteridophytophila* is the only species for which there is molecular data available in GenBank (Kirschner & Liu 2014), though the ITS region differs from *Ps. trichogena* by 8 bp. Additionally,

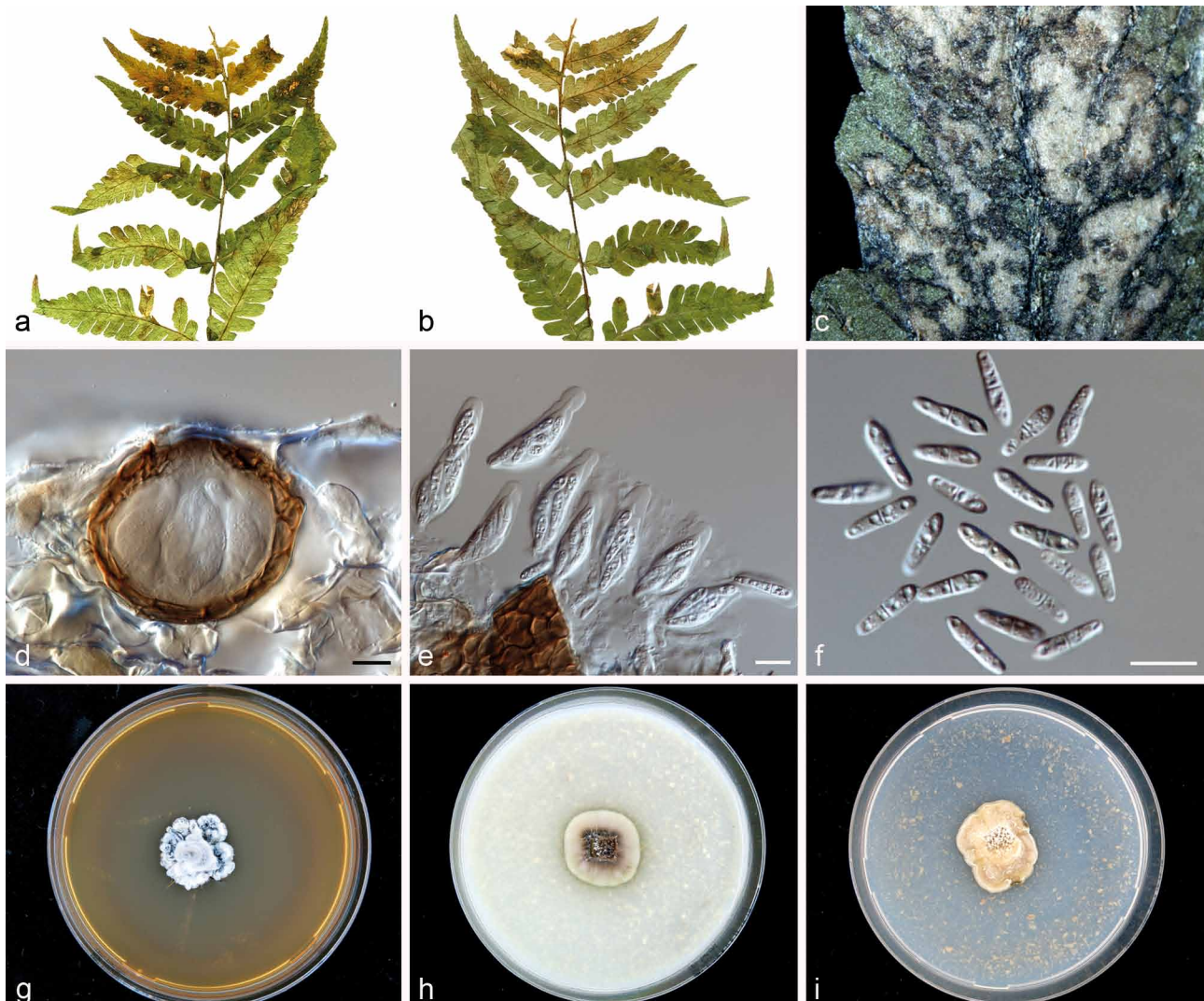


Fig. 22 *Xenomycosphaerella diplazii* (CPC 24691). a, b. Frond spots on *Diplazium* sp.; c. erumpent subcuticular ascomata, fruiting epiphyllous; d. vertical section of the ascoma; e. asci; f. ascospores; g. culture on MEA; h. culture on OA; i. culture on PDA. — Scale bars = $10 \mu\text{m}$.

Ps. pteridophytophila and *Ps. thelypteris* differ from *Ps. trichogena* by having well-developed stromata, arising from the stomata with narrower conidiophores, 2–5 µm and 2–3 µm, respectively (Hsieh & Goh 1990), while *Ps. abacopteridicola* has narrower and smaller conidia (30–80 × 2–3 µm) and conidiophores (5–15 × 2.5–3 µm) (Yen & Lim 1980). *Pseudocercospora trichogena* is the first species of *Pseudocercospora* with a trichomatose habit recorded on ferns.

Xenomycosphaerella Quaedvlieg & Crous, *Persoonia* 33: 24. 2014

Notes — The genus *Xenomycosphaerella* is based on *X. elongata*, which occurs on *Eucalyptus camaldulensis* × *urophylla* from Venezuela (Crous et al. 2007b). So far, only sexual morphs are known for the genus, and because they are morphologically similar to *Mycosphaerella*, they were allocated to *Xenomycosphaerella* based solely on phylogenetic inference (Quaedvlieg et al. 2014).

Xenomycosphaerella diplazii Guatimosim, R.W. Barreto & Crous, *sp. nov.* — MycoBank MB812818; Fig. 22

Etymology. Name refers to the host genus from which it was isolated, *Diplazium*.

Description in planta — *FronD spots* random on pinnulets, but more intense on the pinnule apices, amphigenous, irregular, starting as a dark brown spot at the main vein of the pinnule, expanding towards the margins of the pinnulets, becoming centrally necrotic, with a fertile cream central area with a distinct dark brown to black halo. *External hyphae* absent. *Internal hyphae* intra- and intercellular, 2–4 µm wide, septate, branched, subhyaline, smooth. *Ascomata* pseudothecial, epiphyllous, solitary, subcuticular to erumpent, globose, 50–55 × 55–128 µm, walls of 1–2 layers of pale to dark brown *textura angularis*, cells 7–12 × 4–7 µm, ostiole central, 9–22 µm diam. *Asci* bitunicate, paraphysate, fasciculate, sessile, 8-spored, obovoid to broadly ellipsoidal, straight or slightly curved, 28–42 × 9–13 µm, hyaline, smooth. *Ascospores* inordinate, overlapping, fusoid, straight or slightly curved, 7–13 × 1.5–3 µm, medianly 1-septate, tapering towards rounded ends, narrower towards the lower end, guttulate, hyaline, thin-walled, smooth. *Asexual morph* not known.

Culture characteristics — Colonies on MEA slow-growing, 25 mm diam after 24 d; raised, crustose, with lobate, feathery margins and cottony aerial mycelium at periphery, lavender grey centrally, and lavender grey mixed with leaden grey at periphery; leaden black reverse. On OA flat, aerial mycelium sparse, olivaceous grey centrally, buff to rosy buff periphery; cinnamon reverse. On PDA raised, yeast-like, rosy buff centrally, buff at the periphery; buff reverse; cultures sterile.

Specimen examined. BRAZIL, Rio de Janeiro, Macaé de Cima, road to Fazenda Ouro Verde, on fronds of *Diplazium* sp., 29 Apr. 2012, R.W. Barreto (holotype CBS H-22076, isotype VIC 42565, culture ex-type CPC 24691).

Notes — Based on morphology, *X. diplazii* is similar to *X. elongata*, but differs from the latter by having smaller asci (45–60 µm long in *X. elongata*) and smaller and narrower ascospores, not constricted at the septum (20–25 × 4–5 µm, constricted at the septum in *X. elongata*) (Crous et al. 2007b). Phylogenetically (Fig. 3), *X. diplazii* differs from *X. elongata* by 51 bp for *act*, 69 bp for ITS, 26 bp for LSU and 96 bp for *tef1*. All attempts to induce sporulation of *X. diplazii* have thus far proven unsuccessful. Currently, members of the genus *Xenomycosphaerella* are restricted to South America (Brazil and Venezuela).

Zasmidium Fr., *Summa Veg. Scand.*, section Post. (Stockholm): 407. 1849

Notes — The genus *Zasmidium*, based on *Z. cellare*, comprises species with conspicuously thickened, darkened conidiogenous loci and hila, as typical of *Stenella* (Braun et al. 2013). However, *Stenella* has wide, flat conidial hila and scars, and clusters within *Teratosphaeriaceae*, while *Zasmidium* has planate and somewhat thickened and darkened conidial hila and scars, and clusters within *Mycosphaerellaceae* (Arzanlou et al. 2007, Braun et al. 2013, Quaedvlieg et al. 2014).

Zasmidium sp.

Culture characteristics — Colonies on MEA slow-growing, 53 mm diam after 24 d; flat, with undulate, lobate, feathery margins, mycelium centrally immersed, and velvety aerial mycelium periphery, vinaceous buff centrally, pale mouse grey periphery; isabelline centrally and iron-grey periphery reverse. On OA and PDA lavender grey with iron-grey periphery; olivaceous grey reverse; cultures sterile.

Specimen examined. BRAZIL, Paraná, Guaraguaçu, sand dune area, on fronds of *Blechnum serrulatum*, 1 Feb. 2012, R.W. Barreto (CBS H-22087, culture CPC 24679, COAD 1178).

Notes — Fungarium specimens of this fungus were in poor condition and no conidia were seen. Isolation was performed by conidiophore transfer only. All attempts to promote sporulation in vitro proved to be unsuccessful. It appears that this taxon is a cryptic lineage closely related to *Zasmidium australiensis*, described on the same host, *Blechnum serrulatum*, from Australia (Mulder 1989, Braun et al. 2013). Presently, there are no sequences or known cultures available for *Z. australiensis*.

Zasmidium cyatheae Guatimosim, R.W. Barreto & Crous, *sp. nov.* — MycoBank MB812819; Fig. 23

Etymology. Name refers to the host genus from which it was isolated, *Cyathea*.

Description in planta — *FronD spots* amphigenous, irregular, affecting random pinnulets, starting at the apex of the pinnulets leading firstly to dark brown to black necrosis of the pinnule apex, then spreading to the base, where a cream area appears causing a necrosis of entire pinnulets, and occasionally of the pinnae. *External hyphae* absent. *Internal hyphae* intra- and intercellular, 1.5–2 µm wide, branched, septate, subhyaline to pale brown, smooth. *Ascomata* pseudothecial, epiphyllous, solitary, subcuticular to erumpent, globose, 33–59 × 21–52 µm, walls of 2–3 layers of brown to dark brown *textura angularis*, cells 5–9 × 3–7 µm, ostiole central, 10–18 µm diam. *Asci* bitunicate, paraphysate, fasciculate, sessile, 8-spored, obpyriform, straight, 30–46 × 12–16 µm, hyaline, smooth. *Ascospores* inordinate, overlapping, fusoid, straight, 14–22 × 3–6 µm, medianly 1-septate, tapering towards both rounded ends, narrower towards the lower end, guttulate, hyaline, thin-walled, smooth. *Ascospore germination* not seen. *Asexual morph* not observed.

Culture characteristics — Colonies on MEA and PDA slow-growing, 31 mm diam after 24 d; raised, with smooth, feathery margins, aerial mycelium velvety, pale mouse grey centrally, iron-grey periphery, iron-grey reverse. On OA, aerial mycelium absent, centrally black, periphery of velvety mouse grey aerial mycelium, olivaceous grey reverse; cultures sterile.

Specimen examined. BRAZIL, São Paulo, Eldorado, vicinities of Parque Caverna do Diabo, Atlantic rainforest, on fronds of *Cyathea delgadii*, 13 Apr. 2013, E. Guatimosim (holotype CBS H-22086, isotype VIC 42526, cultures ex-type CPC 24725, COAD 1425).



Fig. 23 *Zasmidium cyatheae* (CPC 24725). a, b. Frond spots on *Cyathea delgadii*; c. erumpent subcuticular ascomata, fruiting epiphyllous; d. vertical section of the ascoma; e. asci; f. ascospores; g. culture on MEA; h. culture on OA; i. culture on PDA. — Scale bars = 10 μ m.

Notes — Phylogenetically, *Z. cyatheae* clustered with *Z. xenoparkii* as sister clade (Fig. 3). *Zasmidium xenoparkii* was described on *Eucalyptus grandis* from Indonesia (Crous et al. 2006b). *Zasmidium cyatheae* is clearly different from *Z. xenoparkii* by having the following number of variable sites for each locus: 11 bp for *act*, 24 bp for *tef1* and 23 bp for ITS. The sexual morph (having mycosphaerella-like structures) is known for only two of the seven species of *Zasmidium* included in this study. These are *Z. citri* (described on *Citrus* sp. from USA) (Huang et al. 2015) and *Z. eucalyptorum* (collected on *Eucalyptus* sp. from Indonesia) (Whiteside 1972, Quaedvlieg et al. 2014). However, the ascospores of *Z. cyatheae* (14–22 \times 3–6 μ m) are longer and wider than those of *Z. citri* (6–11 \times 2–3 μ m) and *Z. eucalyptorum* (12–17 \times 3.5–4.5 μ m) (Whiteside 1972, Crous et al. 2006b). This is the first record of a *Zasmidium* species from *Cyatheaceae*.

DISCUSSION

The present survey presents a phylogenetic overview of the cercosporoid taxa and related sexual morphs that were collected during a systematic survey of fern fungi from Brazil. Quaedvlieg et al. (2014) recently provided a phylogenetic overview of fungi clustering in the *Teratosphaeriaceae*. In the latter study, the authors focused on pathogens of *Eucalyptus*, which makes it interesting to compare to the Brazilian fern fungi, as this could provide an insight into the question if the fungi occurring on

ferns are somehow related to those attacking distant related taxa, such as *Eucalyptus*, or if they evolved independently with their fern hosts.

Forty-four cercosporoid species are known causing frond spots of *Pteridophyta* worldwide: 13 *Cercospora* spp., two *Passalora* spp., 28 *Pseudocercospora* spp. and one *Zasmidium* sp. (Braun et al. 2013). Although no pathogenicity tests were done, all species described on the present study were found associated with frond spot symptoms, indicating their probable habit as pathogens. However, further studies are necessary to clarify the pathogenicity of these fungi on ferns.

Most *Cercospora* species are morphologically very similar to taxa occurring in the *C. apii* species complex (Braun et al. 2013). In the present study, we were able to identify one new *Cercospora* species, and demonstrate that the host range of *C. conigrammes* is wider than previously known, including ferns belonging to two additional families. Plant hosts from *Pteridophyta* represent some of the oldest lineages of vascular plants (Smith et al. 2008). It is interesting to note that *C. conigrammes* is on one hand proving to have a wider host range within the *Pteridophyta* and, on the other hand, found to be basal in the phylogeny of the genus *Cercospora* (Groenewald et al. 2013; Fig. 1).

As for *Pseudocercospora*, a long list of names have been published for which there are no DNA data and ex-type cultures available (Braun et al. 2013), complicating a better understand-

ing of the taxonomy of the genus. Recollecting and epitypifying all these species is a challenging, but necessary task for mycologists dealing with cercosporoid fungi. Three examples of taxonomic decisions that are still pending even after the present study involve *Ps. abacopteridicola*, *Ps. lygodiicola* and *Ps. thelypteridis*. Although we suspect that these Brazilian collections may in fact represent novel species, this can only be confirmed after the recollection of fresh materials from the type localities (Singapore, China and Taiwan, respectively – Yen & Lim 1980, Braun et al. 2013), followed by epitypification and a phylogenetic comparison.

Historically, the taxonomy of cercosporoid fungi has been based upon morphological and ecological features, including conjectured host specificity (Chupp 1954, Deighton 1965, 1971, 1973, 1974, 1976, Pons & Sutton 1988, Braun 1993a, b, c, 1995, 1998, Crous & Braun 1996, Braun & Mel'nik 1997, Crous et al. 2000, Braun et al. 2013, 2014, 2015). It is now widely accepted that this was an inadequate basis for the taxonomy of this complex plethora of fungi. Two examples of potentially mistaken conclusions based on morphology, symptoms and host-association alone were provided by our results:

1. *Zasmidium cyatheae* (only sexual morph found) and *Neoceratosperma cyatheae* (only asexual morph found), co-occurred on the same frond spot, on the fern *Cyathea delgadii*. Without pure cultures and access to molecular data the mistaken conclusion would be that *Z. cyatheae* was the sexual morph of *N. cyatheae*.
2. A similar situation occurred for *Paramycosphaerella sticheri* and *Clypeosphaerella sticheri*. Both were found attacking two different species in the same host genus *Sticherus* causing similar disease symptoms. It is likely that many conjectured connections between asexual and sexual morphs have been mistakenly made for cercosporoids and other fungal groups. Efforts towards clarifying these connections with modern criteria should be continued in order to generate an appropriate and consolidated taxonomy of cercosporoids and other fungal groups (Taylor et al. 2000, Crous & Groenewald 2005, Crous et al. 2009f, 2015b, Quaedvlieg et al. 2014).

In the past, mycologists have hypothesized that plant pathogenic fungi associated with primitive plants were also evolutionarily basal to the evolution of fungi. Thus, Savile (1971) proposed that primitive plant hosts, such as ferns, would have primitive rust genera. Later, phylogenetic studies involving rust species in different genera have proven this hypothesis wrong. For example, *Hemileia* and *Marvalia* – sister genera at the base of the *Pucciniales* phylogenetic tree (Wingfield et al. 2004, Aime 2006) – are pathogens of higher plant taxa, especially in the *Rubiaceae* and *Asclepiadaceae*, respectively.

As for the cercosporoid and mycosphaerella-like species documented here, there is some evidence that the fungal species associated with ferns are evolutionarily basal to the evolution of their relatives. In the *Cercospora* phylogeny (Fig. 1), *C. coniogrammes* (recorded only from ferns) is basal to the evolution of all other *Cercospora* species, whilst the same pattern is reproduced in the *Pseudocercospora* phylogeny (Fig. 2), where *Ps. cyatheicola*, *Ps. lygodiicola*, *Ps. rumohrae* and *Ps. thelypteridis*, all isolated from ferns, appear to be evolutionarily basal in the clade where they cluster; in the phylogeny of mycosphaerella-like taxa (Fig. 3), a basal position was observed for *Phaeophleospora hymenocallidis*, *Ps. hymenocallidicola* and *Ps. pteridivora* (all from ferns), appearing evolutionarily basal to all other species in the genus for which sequence data were available.

As more sequence data become available for cercosporoids associated with ferns, this preliminary evidence may become

stronger and allow for an elucidation of further cercosporoid genealogies and, hence, should permit a better understanding of the co-evolutionary history of this fungal group and its association with host plants.

The present study has significantly expanded our knowledge of cercosporoid and mycosphaerella-like fungi associated with frond spots in Brazilian *Pteridophyta*. Previously, only one cercosporoid and one mycosphaerella-like species (*Ps. davallicola* and '*Mycosphaerella*' *tocoyenae*, respectively) were known to be associated with diseases on ferns in Brazil (Farr & Rossman 2015, Mendes & Urben 2015). The present work has expanded this number significantly by adding one new genus (*Clypeosphaerella*) and 15 new species to this list. Here we also provide novel molecular information that may be useful to obtain a better understanding of the evolution of cercosporoid and mycosphaerella-like fungi. We also hope that further exploration of these cultures will contribute in the future to a more robust phylogeny of these fungi across various families of host plants, and help establishing a better understanding of their host specificity and evolution. The clear abundance of novel taxa collected on ferns in Brazil, also underlines the scientific value of host or host-group based surveys as a source of mycological novelties. Finally, our findings confirm that mycologists in the tropics have thus far given little attention to fungi occurring on plant hosts with apparent limited economic relevance, such as ferns. Fern fungi in Brazil and other tropical regions are likely to represent an important source of a highly diverse mycobiota that still awaits discovery.

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