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Botryosphaeriaceae associated with diseases of mango (*Mangifera indica*)

T. Trakunyingcharoen · R. Cheewangkoon · C. To-anun · P. W. Crous · J. M. van Niekerk · L. Lombard

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Abstract Fungal species of *Botryosphaeriaceae* have a cosmopolitan distribution and are important pathogens of a wide range of plant hosts. This study aims to use phylogenetic inference to review the geographical distribution of botryosphaeriacous species that have been associated with diseases of mango (*Mangifera indica*) globally. The phylogenetic analyses were performed based on the combined sequence datasets of the internal transcribed spacer (ITS) region of the nuclear rDNA and a partial region of the translation elongation factor 1-alpha (EF1-α) gene. The phylogenetic study revealed seven clades with distinct morphological characters from several countries, including Australia, Brazil, Egypt, Iran, Mali, Peru, South Africa, Taiwan and Thailand. *Lasiodiplodia theobromae* appears to be a dominant species on mango with the largest geographical distribution, whereas *L. crassispora* and *Barriopsis iraniana* have only been reported on mango in Brazil and Iran, respectively. These finding indicate that most of the species reported from mango are not restricted to specific geographical regions, although some genera appear to have a limited distribution.

Keywords *Botryosphaeriaceae* · EF1-α · ITS · Mango · *Mangifera indica*

T. Trakunyingcharoen · R. Cheewangkoon · C. To-anun (✉)
Department of Entomology and Plant Pathology, Faculty of Agriculture, Chiang Mai University, Chiang Mai 50200, Thailand
e-mail: chaiwat.toanun@gmail.com

P. W. Crous · L. Lombard
CBS-KNAW Fungal Biodiversity Centre, Uppsalalaan 8, 3584 CT Utrecht, The Netherlands

P. W. Crous
Microbiology, Department of Biology, Utrecht University, Padualaan 8, 3584 CH Utrecht, The Netherlands

P. W. Crous
Laboratory of Phytopathology, Wageningen University and Research Centre (WUR), Droevendaalsesteeg 1, 6708 PB Wageningen, The Netherlands

J. M. van Niekerk
Westfalia Technological Services, P.O. Box 1103, Tzaneen 0850, South Africa

Introduction

Mango (*Mangifera indica*, *Anacardiaceae*) is an economically important tropical fruit, which is produced in at least 90 countries around the world (Evans 2008). In 2007, Asia accounted for the largest global mango production (77 %) followed by the Americas (13 %) and Africa (9 %), with Thailand representing the third largest producer in the world (FAOSTAT 2007), resulting in mango export to most of the Southeast Asian market (Evans 2008). In 2008–2009, Asia was the largest global mango producer, especially in India, where 31.5 million metric tons per year were produced. The United States and European Union accounted for 75 % of the world's mango importation according to the FAO's 2009 Food Market Analysis of Tropical Fruits (Tanzania Agriculture Productivity Programme 2010).

Botryosphaeriaceae is a species-rich family of ascomycetous fungi (Crous et al. 2006; Slippers et al. 2013), known as either endophytes or opportunistic pathogens of various living plant hosts (Phillips et al. 2013). As opportunistic pathogens, they display the ability to remain as latent infections of healthy plants, but cause disease when the host plant comes under stress (Denman et al. 2000; Slippers and Wingfield 2007). These fungi have a cosmopolitan distribution and wide host ranges including mango. Several genera play a key role as pathogens of mango, and are responsible for pre- and post-harvest diseases causing canker, dieback, panicle brown rot, fruit rot and stem-end rot (Abdalla et al. 2003; Abdollahzadeh et al. 2010; de Oliveira Costa et al. 2010; Sakalidis et al. 2011;

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Ismail et al. 2012). These diseases pose a serious restriction to mango production and their global marketability, as has been reported from Australia (Sakalidis et al. 2011), Brazil (de Oliveira Costa et al. 2010), Egypt (Ismail et al. 2012), Peru (Javier-Alva et al. 2009), Taiwan (Ni et al. 2012a, b) and Thailand (Jitareerat et al. 2005).

Recent studies have highlighted an increase in disease incidences on mango attributed to these fungi as shown in the study of Marques et al. (2013a, b). The authors were able to illustrate that *Lasiodiplodia crassispora*, *L. egyptiacae*, *L. hormozganensis*, *L. iraniensis* and *L. pseudotheobromae* were responsible for branch dieback and black leaf spots on mango in Brazil. Furthermore, new records of *Botryosphaeriaceae* on mango have surfaced in several countries. According to Ni et al. (2012a, b), *L. theobromae*, *Fusicoccum aesculi*, *Neofusicoccum mangiferae* and *N. parvum* were associated with mango fruit rot in Taiwan. In Australia, *Neoscytalidium hyalinum* (= *N. dimidiatum*) and *Ne. novaehollandiae* have been associated with dieback and stem cankers of mango trees (Ray et al. 2010). *Lasiodiplodia egyptiacae*, *L. pseudotheobromae* and *L. theobromae* have been found as causal agents of dieback and black leaf spots on leaves of mango in Egypt (Ismail et al. 2012). In Iran, *Barriopsis iraniana* (Abdollahzadeh et al. 2009), *L. hormozganensis* and *L. iraniensis* (Abdollahzadeh et al. 2010; Phillips et al. 2013) were isolated from dieback, cankers, fruit rot and others necrotic symptoms of mango.

Prior to the introduction of molecular techniques, morphological characteristics were primarily used for fungal characterisation and identification of this group of fungi. Denman et al. (2000) divided *Botryosphaeria* asexual morphs into two groups based on conidium colour. The first group, characterised by hyaline conidia, represented *Fusicoccum*, and the second group, producing pigmented conidia, were regarded as *Diplodia* species. However, some overlapping morphological characters were found between closely related species (Slippers et al. 2005), and Alves et al. (2007) reported that conidial pigmentation and septation were influenced by the state of conidium maturity, resulting in taxonomic confusion. It was only in later studies that this complex was shown to represent up to 19 different genera (Crous et al. 2006; Alves et al. 2008; de Oliveira Costa et al. 2010; Sakalidis et al. 2011; Ismail et al. 2012; Liu et al. 2012; Abdollahzadeh et al. 2013; Phillips et al. 2013), clarifying the morphological variation observed in the hyaline and pigmented clades depicted by Denman et al. (2000).

Given the fact that the genera associated with the *Botryosphaeriaceae* have finally been clarified (Phillips et al. 2013), the primary aim of this review was to determine which genera and species occur on mango, and also to clarify the geographical distribution of the species using phylogenetic inference. In addition, a number of previously unidentified isolates obtained from mango collected in Thailand and South Africa were also included.

Materials and methods

Isolation and morphology investigation

Asymptomatic twigs were collected from a commercial mango plantation in Chiang Mai province, Thailand and incubated in moist chambers at room-temperature for 7–14 days. Similarly, various plant parts (fruits, leaves and shoots) were collected from mango trees grown in Limpopo province, South Africa, and also incubated in moist chambers as above. After sporulation, direct isolations were made from the plant material onto 2 % Potato Dextrose Agar (PDA) according to Crous et al. (2009). The morphology of *Botryosphaeriaceae* isolates was investigated by inducing sporulation on sterile pine needles placed on 2 % (w/v) water agar (WA) under near UV light at 25 °C for 14–30 day as described by Smith et al. (1996). Gross morphological characters were determined by mounting fungal structures in clear lactic acid. Measurements of 30 conidia and other fungal structures were performed at ×1,000 magnification using an Olympus BX51 (Olympus, USA) microscope with differential interference contrast.

DNA isolation, PCR amplification, and sequencing

Total genomic DNA was extracted from 7 to 10-day-old cultures maintained at 25 °C, using the Ultraclean® Microbial DNA Isolation Kit (MO-BIO Laboratories, Inc, Carlsbad, USA) following the manufacturer's instructions. Sequences of the internal transcribed spacers and 5.8 s gene (ITS) region of the nuclear rDNA and partial sequences of the translation elongation factor 1-alpha (EF1-α) gene region were amplified. The amplifications were conducted in a thermal cycler using the following cycling steps: initial denaturation at 95 °C for 5 min, followed by 35 cycles of denaturation at 95 °C for 30 s, annealing at 52 °C for 30 s, extension at 72 °C for 1 min, and a final extension step at 72 °C for 7 min. The ITS region was amplified using primers ITS5 and ITS4 (White et al. 1990). The partial EF1-α region was amplified using primers EF1-728 (Carbone and Kohn 1999) and EF2 (O'Donnell et al. 1998). However for some isolates in the genus *Lasiodiplodia*, this region was amplified using primers EF1-688 and EF1-1251 (Alves et al. 2008).

The amplified fragments were used as templates for sequencing reactions in both directions using the same primer pairs in an ABI PRISM™ 3730 DNA automated sequencer (Perkin-Elmer Applied BioSystems, Foster City, CA, USA) together with Big Dye terminator sequencing kit v. 3.1 following the manufacturer's instructions.

Phylogenetic analysis

The generated nucleotide sequences were edited and manually adjusted as necessary using MEGA v. 5.1 (Tamura et al. 2011).

The generated sequences were aligned with other sequences obtained from GenBank, which represented past studies on botryosphaeriacous fungi on mango, using MAFFT v. 7 (http://mafft.cbrc.jp/alignment/server/). Maximum Parsimony (MP) and Bayesian Inference (BI) analyses were performed on a combined datasets of ITS and EF1-α sequences using PAUP v. 4.0b10 (Swofford 2003) and MrBayes v. 3.2.1 (Ronquist and Huelsenbeck 2003), respectively. The phylogenetic analyses were rooted using *Phyllosticta citricarpa* (CBS 111.20), a member of the sister family, *Phyllostictaceae* (Wikee et al. 2013) as outgroup.

The MP analysis was conducted using the heuristic search option with random stepwise addition using a 1,000 replicates, tree bisection and reconnection (TBR) as branch swapping algorithm (Swofford and Begle 1993) and random taxon addition sequences for the construction of MP trees. All characters were unordered and had equal weight and gaps were treated as missing data. Branches of zero length were collapsed and all multiple equally parsimonious trees were saved. The robustness of the equal parsimonious trees was calculated using 1,000 bootstrap replications (Hillis and Bull 1993). Calculated values for parsimony included tree length (TL), consistency index (CI), retention index (RI), rescaled consistency index (RC), and homoplasy index (HI).

Bayesian analyses were conducted by two independent runs of Markov Chain Monte Carlo (MCMC) (Larget and Simon 1999) algorithms to construct the phylogenetic tree. The analyses were performed using four MCMC chains running simultaneously applying a general time-reversible (GTR) (Rodriguez et al. 1990) substitution model with rate variation of gamma-distribution (G), and proportion of invariable site (I) for both ITS and EF 1-α, respectively, determined using MrModel test v. 2.2 (Nylander 2004). The analyses were run until the average standard deviation of split frequencies came below 0.01 within 100,000,000 generations and trees were saved every 1,000 generations. The first 25 % of saved trees were discarded as the "burn-in" phase and posterior probabilities (Rannala and Yang 1996) were calculated from the remaining trees.

Results

Isolation and morphology investigation

Isolates obtained from asymptomatic mango twigs collected in Thailand represented species of *Lasiodiplodia*, based on morphology. Isolates CPC 22783, 22787, 22793 and 22794 were identified as *L. pseudotheobromae* forming superficial and solitary or aggregated pycnidia producing large, ellipsoidal conidia (23–32×13–15 μm), and hyaline paraphyses, mostly aseptate with rounded tips (25–62×1.5–2 μm). Isolate CPC 22795 closely resembled *L. iraniensis*, forming semi-immersed and solitary pycnidia, producing larger ovoid-ellipsoid conidia (22–26×13–16 μm), and shorter hyaline, septate paraphyses with rounded tips (25–51×1–1.5 μm). Isolate CPC 22800 closely resembled *L. viticola*, producing larger ellipsoidal conidia (23–29×13–14 μm) and hyaline paraphyses with rounded tips (40–82×1.5–2 μm) (Abdollahzadeh et al. 2010; Alves et al. 2008; Úrbez-Torres et al. 2012). Although these isolates were obtained from asymptomatic twigs, they were shown to be pathogenic on mango woody tissues and fruits causing canker and rot diseases, respectively (Trakunyingcharoen et al. 2013).

The South African isolates obtained from mango represented three different genera, namely *Lasiodiplodia*, *Neofusicoccum* and *Pseudofusicoccum*. The *Lasiodiplodia* isolate CPC 23311 obtained from an asymptomatic shoot represented *L. theobromae*, having large, ovoid to ellipsoid conidia (22–30×12–15 μm) and hyaline, septate paraphyses with rounded tips (46–55×2–3 μm). The *Neofusicoccum* isolates CPC 23297 obtained from an asymptomatic shoot and CPC 23298 obtained from an asymptomatic shoot and fruit panicle tissue represented species belonging to the *N. parvum*/*N. ribis* complex, forming fusoid-ellipsoid conidia (13–22×4–5 μm) that became darker with age. A single isolate CPC 23304 obtained from mature fruit surface which was not associated with rot symptoms represented a species in the genus *Pseudofusicoccum*, producing large, solitary pycnidia, which in turn formed hyaline, aseptate conidia (27–35×8–11 μm) surrounded by a mucoid sheath, and lacking paraphyses, closely resembling *P. olivaceum* (Mehl et al. 2011).

Molecular and phylogenetic analysis

Amplicons generated of the ITS region were approximately 570 bp using the ITS5 and ITS4 primer combination. Amplicons generated for the partial EF1-α gene region were approximately 500 bp using primers EF1-728 and EF2 and approximately 700 bp using primers EF1-688 and EF1-1251. All sequences generated during this study were deposited into GenBank as indicated in Table 1.

The combined datasets, representing 144 taxa, consisted of 971 characters, including alignment gaps, of which 412 characters were constant, 139 characters were variable and parsimony-uninformative and 420 characters were parsimony-informative. The MP analysis yielded 1,000 trees (TL=1,348 steps, CI=0.705, RI=0.973, RC=0.687, and HI=0.295). The Bayesian consensus tree, presented as Fig. 1, confirmed both the tree topology and bootstrap support of the strict consensus tree obtained with maximum-parsimony and posterior probability stringent of the branch nodes.

The Bayesian tree revealed seven well supported clades corresponding to genera. Clade I represents species in the genus *Pseudofusicoccum*, Clade II species of *Neofusicoccum* and Clade III species of *Botryosphaeria*, which closely relates

Table 1 Isolates of *Botryosphaeriaceae* used in this study

Identity	Isolate no.	Locality	Collector	GenBank accession no.	
				ITS	EF1-α
Barriopsis iraniana	IRAN 1448C*	Iran	J. Abdollahzadeh & A. Javadi	FJ919663	FJ919652
B. iraniana	IRAN 1453C	Iran	J. Abdollahzadeh & A. Javadi	FJ919664	FJ919653
B. fusca	CBS 174.26*	Cuba	N.E. Stevens	EU673330	EU673296
Botryosphaeria dothidea	B922	Taiwan	H.F. Ni	GQ421485	GU002162
B. dothidea	B964	Taiwan	H.F. Ni	GQ861429	GU002157
B. dothidea	CMM 1327	Brazil	V.S.O. Costa	EU938338	
B. dothidea	CMW 7020	Australia	G.I. Johnson	AY615191	
B. dothidea	CMW 7027	Australia	G.I. Johnson	AY615192	
B. dothidea	CMM 3937	Brazil	M.W. Marques	JX513643	JX513622
B. dothidea	CMM 3938	Brazil	M.W. Marques	JX513645	JX513624
B. dothidea	CMW 8000*	Switzerland	B. Slippers	AY236949	AY236898
B. fabicercianum	B811	Taiwan	H.F. Ni	GU453689	GU002164
B. fabicercianum	B844	Taiwan	H.F. Ni	GU453690	GU002163
B. fabicercianum	B833	Taiwan	H.F. Ni	GQ861430	GU002161
B. fabicercianum	B801	Taiwan	H.F. Ni	GQ861431	GU002160
B. fabicercianum	CMM 3899	Brazil	M.W. Marques	JX513646	JX513625
B. fabicercianum	CMM 3905	Brazil	M.W. Marques	JX513642	JX513621
B. fabicercianum	CMW 27094*	China	M.J. Wingfield	HQ332197	HQ332213
B. scharifii	IRAN 1529C*	Iran	J. Abdollahzadeh	JQ772020	JQ772057
B. scharifii	IRAN 1543C	Iran	J. Abdollahzadeh & A. Javadi	JQ772019	JQ772056
Cophinforma mamane	CMM 1390	Brazil	M.W. Marques	KC184893	JX513627
C. mamane	CMM 3941	Brazil	M.W. Marques	KC184892	JX513626
C. mamane	GS 97-59*	Hawaii	D. Gardner	AF246930	
Lasiodiplodia crassispora	CMM 3982	Brazil	M.W. Marques	JX464058	JX464015
L. crassispora	CMW 13488*	Venezuela	S. Mohali	DQ103552	DQ103559
L. egyptiacae	BOT 10*	Egypt	A.M. Ismail	JN814397	JN814424
L. egyptiacae	BOT 29	Egypt	A.M. Ismail	JN814401	JN814428
L. egyptiacae	CMM 3981	Brazil	M.W. Marques	JX464072	JX464035
L. egyptiacae	CMM 1485	Brazil	V.S.O. Costa	JX464076	JX464044
L. hormozganensis	IRAN 1498C	Iran	J. Abdollahzadeh & A. Javadi	GU945356	GU945344
L. hormozganensis	CJA57	Iran	J. Abdollahzadeh & A. Javadi	GU945357	GU945345
L. hormozganensis	CMM 1546	Brazil	V.S.O. Costa	JX464077	JX464053
L. hormozganensis	CMM 3983	Brazil	M.W. Marques	JX464069	JX464033
L. hormozganensis	IRAN 1500C*	Iran	J. Abdollahzadeh & A. Javadi	GU945355	GU945343
L. iraniensis	IRAN 921C	Iran	N. Khezrinejad	GU945346	GU945334
L. iraniensis	IRAN 1519C	Iran	J. Abdollahzadeh & A. Javadi	GU945350	GU945338
L. iraniensis	WAC 13290	WA	J. Ray	GU172381	HM218824
L. iraniensis	WAC 13297	WA	J. Ray	GU172379	HM218823
L. iraniensis	CMM 3990	Brazil	M.W. Marques	JX464067	JX464028
L. iraniensis	CMM 1483	Brazil	V.S.O. Costa	JX464073	JX464023
L. iraniensis	CMM 3993	Brazil	M.W. Marques	JX464068	JX464029
L. iraniensis	CMM 3995	Brazil	M.W. Marques	JX464097	JX464046
L. iraniensis	IRAN 1520C*	Iran	J. Abdollahzadeh & A. Javadi	GU945348	GU945336
Lasiodiplodia sp.	**CPC 22795**	Thailand	T. Trakunyingcharoen	*KJ193637*	*KJ193681*
L. pseudotheobromae	BOT 1	Egypt	A. M. Ismail	JN814375	JN814402
L. pseudotheobromae	BOT 13	Egypt	A. M. Ismail	JN814377	JN814404
L. pseudotheobromae	BOT 16	Egypt	A. M. Ismail	JN814379	JN814406

Table 1 (continued)

Identity	Isolate no.	Locality	Collector	GenBank accession no.	
				ITS	EF1-α
L. pseudotheobromae	BOT 18	Egypt	A. M. Ismail	JN814381	JN814408
L. pseudotheobromae	BOT 2	Egypt	A. M. Ismail	JN814382	JN814409
L. pseudotheobromae	BOT 11	Egypt	A. M. Ismail	JN814383	JN814410
L. pseudotheobromae	BOT 17	Egypt	A. M. Ismail	JN814385	JN814412
L. pseudotheobromae	BOT 12	Egypt	A. M. Ismail	JN814386	JN814413
L. pseudotheobromae	WAC 13281	WA	J. Ray	GU172380	GU172412
L. pseudotheobromae	**CPC 22783**	Thailand	T. Trakunyingcharoen	*KJ193638*	*KJ193682*
L. pseudotheobromae	**CPC 22787**	Thailand	T. Trakunyingcharoen	*KJ193639*	*KJ193683*
L. pseudotheobromae	**CPC 22793**	Thailand	T. Trakunyingcharoen	*KJ193640*	*KJ193684*
L. pseudotheobromae	**CPC 22794**	Thailand	T. Trakunyingcharoen	*KJ193641*	*KJ193685*
L. pseudotheobromae	CBS 116459*	Costa Rica	J. Carranza-Velazquez	EF622077	EF622057
L. theobromae	BOT 5	Egypt	A. M. Ismail	JN814376	JN814403
L. theobromae	BOT 9	Egypt	A. M. Ismail	JN814392	JN814419
L. theobromae	BOT 4	Egypt	A. M. Ismail	JN814395	JN814422
L. theobromae	BOT 7	Egypt	A. M. Ismail	JN814396	JN814423
L. theobromae	IRAN 1496C	Iran	J. Abdollahzadeh & A. Javadi	GU973869	GU973861
L. theobromae	IRAN 1499C	Iran	J. Abdollahzadeh & A. Javadi	GU973870	GU973862
L. theobromae	B961	Taiwan	H.F. Ni	GQ502453	GQ979999
L. theobromae	B965	Taiwan	H.F. Ni	GQ502454	GQ980000
L. theobromae	B838	Taiwan	H.F. Ni	GQ502456	GQ980001
L. theobromae	CMM 1476	Brazil	M.W. Marques	JX464083	JX464057
L. theobromae	CMM 1481	Brazil	M.W. Marques	JX464095	JX464021
L. theobromae	CMM 1517	Brazil	M.W. Marques	JX464060	JX464054
L. theobromae	CMM 4019	Brazil	M.W. Marques	JX464096	JX464026
L. theobromae	CMM 4039	Brazil	M.W. Marques	JX464065	JX464041
L. theobromae	CMM 4041	Brazil	M.W. Marques	KC184891	JX464042
L. theobromae	CMM 4042	Brazil	M.W. Marques	JX464070	JX464017
L. theobromae	CMM 4046	Brazil	M.W. Marques	JX464091	JX464027
L. theobromae	CMM 4048	Brazil	M.W. Marques	JX464093	JX464048
L. theobromae	CMM 4021	Brazil	M.W. Marques	JX464064	JX464047
L. theobromae	**CPC 23311**	SA	J.M. van Niekerk	*KJ193642*	*KJ193686*
Lasiodiplodia sp.1	WAC 13280	WA	J. Ray	GU172377	GU172408
Lasiodiplodia sp.2	WAC 13300	WA	J. Ray	GU172378	GU172409
Lasiodiplodia sp.	**CPC 22795**	Thailand	T. Trakunyingcharoen	*KJ193637*	*KJ193681*
Lasiodiplodia sp.	CMM 4014	Brazil	M.W. Marques	JX464098	JX464031
Lasiodiplodia sp.	CMM 4015	Brazil	M.W. Marques	JX464063	JX464049
Lasiodiplodia sp.	**CPC 22800**	Thailand	T. Trakunyingcharoen	*KJ193643*	*KJ193687*
L. viticola	CBS 128313*	USA	R.D. Cartwright & W.D. Gubler	HQ288227	HQ288269
Neofusicoccum brasiliense	CMM 1285	Brazil	M.W. Marques	JX513628	JX513608
N. brasiliense	CMM 1338*	Brazil	M.W. Marques	JX513630	JX513610
N. mangiferae	B809	Taiwan	H.F. Ni	GQ848323	GQ998898
N. mangiferae	B793	Taiwan	H.F. Ni	GQ848320	GQ998900
N. mangiferae	CBS 118532	Australia	G.I. Johnson	AY615186	DQ093220
N. mangiferae	CBS 118531*	Australia	G.I. Johnson	AY615185	DQ093221
N. mediterraneum	IRAN 1549C	Iran	J. Abdollahzadeh & A. Javadi	JQ772053	JQ772090
N. mediterraneum	IRAN 1550C	Iran	J. Abdollahzadeh & A. Javadi	JQ772047	JQ772084
N. mediterraneum	IRAN 1554C	Iran	J. Abdollahzadeh & A. Javadi	JQ772048	JQ772085

Table 1 (continued)

Identity	Isolate no.	Locality	Collector	GenBank accession no.	
				ITS	EF1-α
N. mediterraneum	CJA 144	Iran	J. Abdollahzadeh & A. Javadi	JQ772051	JQ772088
N. mediterraneum	CBS 121718*	Greece	Crous, Wingfield & Phillips	GU251176	GU251308
N. parvum	B845	Taiwan	H.F. Ni	GQ861434	GQ985316
N. parvum	B946	Taiwan	H.F. Ni	GQ861432	GQ985313
N. parvum	B794	Taiwan	H.F. Ni	GQ861433	GQ985312
N. parvum	B1260	Taiwan	H.F. Ni	GU073287	GU121432
N. parvum	B1001	Taiwan	H.F. Ni	GQ861435	
N. parvum	CMW 7025	Australia	G.I. Johnson	AY615181	
N. parvum	CMW 7026	Australia	G.I. Johnson	AY615182	
N. parvum	CMM 1317	Brazil	V.S.O. Costa	EU938333	
N. parvum	CMM 1291	Brazil	M.W. Marques	JX513633	JX513613
N. parvum	CMM 1465	Brazil	M.W. Marques	JX513634	JX513614
N. parvum	A4	Peru		FJ528596	FJ528597
N. parvum	**CPC 23283**	SA	J.M. van Niekerk	*KJ193644*	*KJ193688*
N. parvum	**CPC 23285**	SA	J.M. van Niekerk	*KJ193645*	*KJ193689*
N. parvum	**CPC 23286**	SA	J.M. van Niekerk	*KJ193646*	*KJ193690*
N. parvum	**CPC 23287**	SA	J.M. van Niekerk	*KJ193647*	*KJ193691*
N. parvum	**CPC 23288**	SA	J.M. van Niekerk	*KJ193648*	*KJ193692*
N. parvum	**CPC 23289**	SA	J.M. van Niekerk	*KJ193649*	*KJ193693*
N. parvum	**CPC 23290**	SA	J.M. van Niekerk	*KJ193650*	*KJ193694*
N. parvum	**CPC 23291**	SA	J.M. van Niekerk	*KJ193651*	*KJ193695*
N. parvum	**CPC 23292**	SA	J.M. van Niekerk	*KJ193652*	*KJ193696*
N. parvum	**CPC 23293**	SA	J.M. van Niekerk	*KJ193653*	*KJ193697*
N. parvum	**CPC 23294**	SA	J.M. van Niekerk	*KJ193654*	*KJ193698*
N. parvum	**CPC 23295**	SA	J.M. van Niekerk	*KJ193655*	*KJ193699*
N. parvum	**CPC 23296**	SA	J.M. van Niekerk	*KJ193656*	*KJ193700*
N. parvum	**CPC 23297**	SA	J.M. van Niekerk	*KJ193657*	*KJ193701*
N. parvum	**CPC 23298**	SA	J.M. van Niekerk	*KJ193658*	*KJ193702*
N. parvum	**CPC 23299**	SA	J.M. van Niekerk	*KJ193659*	*KJ193703*
N. parvum	**CPC 23300**	SA	J.M. van Niekerk	*KJ193660*	*KJ193704*
N. parvum	**CPC 23301**	SA	J.M. van Niekerk	*KJ193661*	*KJ193705*
N. parvum	**CPC 23302**	SA	J.M. van Niekerk	*KJ193662*	*KJ193706*
N. parvum	**CPC 23303**	SA	J.M. van Niekerk	*KJ193663*	*KJ193707*
N. parvum	**CPC 23305**	SA	J.M. van Niekerk	*KJ193664*	*KJ193708*
N. parvum	**CPC 23306**	SA	J.M. van Niekerk	*KJ193665*	*KJ193709*
N. parvum	**CPC 23307**	SA	J.M. van Niekerk	*KJ193666*	*KJ193710*
N. parvum	**CPC 23308**	SA	J.M. van Niekerk	*KJ193667*	*KJ19371l*
N. parvum	**CPC 23309**	SA	J.M. van Niekerk	*KJ193668*	*KJ193712*
N. parvum	**CPC 23310**	SA	J.M. van Niekerk	*KJ193669*	*KJ193713*
N. parvum	**CPC 23312**	SA	J.M. van Niekerk	*KJ193670*	*KJ193714*
N. parvum	**CPC 23313**	SA	J.M. van Niekerk	*KJ193671*	*KJ193715*
N. parvum	**CPC 23314**	SA	J.M. van Niekerk	*KJ193672*	*KJ193716*
N. parvum	**CPC 23315**	SA	J.M. van Niekerk	*KJ193673*	*KJ193717*
N. parvum	**CPC 23316**	SA	J.M. van Niekerk	*KJ193674*	*KJ193718*
N. parvum	**CPC 23317**	SA	J.M. van Niekerk	*KJ193675*	*KJ193719*
N. parvum	**CPC 23318**	SA	J.M. van Niekerk	*KJ193676*	*KJ193720*
N. parvum	**CPC 23319**	SA	J.M. van Niekerk	*KJ193677*	*KJ193721*

Table 1 (continued)

Identity	Isolate no.	Locality	Collector	GenBank accession no.	
				ITS	EF1-α
N. parvum	**CPC 23320**	SA	J.M. van Niekerk	*KJ193678*	*KJ193722*
N. parvum	CMW 9081*	New Zealand	G.J. Samuels	AY236943	AY236888
N. ribis	CMW 7772*	New York	B. Slippers & G. Hudler	AY236935	AY236877
Neoscytalidium hyalinum	WAC 13301	WA	J. Ray	GU172384	GU172416
Ne. hyalinum	WAC 13287	WA	J. Ray	GU172385	GU172417
Ne. hyalinum	WAC 13277	WA	J. Ray	GU172388	GU172420
Ne. hyalinum	WAC 13305	WA	J. Ray	GU172390	GU172422
Ne. hyalinum	CBS 499.66	Mali	J. Brun	AY819727	EU144063
Ne. hyalinum	**CBS 312.90***	Netherlands	R. Benne	*KJ193679*	*KJ193723*
Ne. novaehollandiae	WAC 12691	WA	J. Ray	EF585542	EF585574
Ne. novaehollandiae	WAC 12688	WA	J. Ray	EF585543	EF585575
Ne. novaehollandiae	WAC 13273	WA	J. Ray	GU172397	GU172429
Ne. novaehollandiae	WAC 13275	WA	J. Ray	GU172400	GU172432
Ne. novaehollandiae	CBS 122071*	WA	T.I. Burgess & M.J. Wingfield	EF585540	EF585580
Pseudofusicoccum sp.	CMW 7022	Australia	A.W. Cooke	AY615188	
Pseudofusicoccum sp.	CMW 7802	Australia	A.W. Cooke	AY615190	
P. adansoniae	WAC 12689	WA	J. Ray	EF585534	EF585567
P. adansoniae	MUCC 525	WA	T. I. Burgess	EF585527	EF585573
P. adansoniae	WAC 13292	WA	J. Ray	GU172401	GU172433
P. adansoniae	WAC 13278	WA	J. Ray	GU172402	GU172434
P. adansoniae	WAC 13295	WA	J. Ray	GU172403	GU172435
P. adansoniae	CBS 122055*	WA	T.I. Burgess & M.J. Wingfield	EF585523	EF585571
P. ardesiacum	WAC 13294	WA	J. Ray	GU172405	GU172437
P. ardesiacum	CBS 122062*	WA	T.I. Burgess & M.J. Wingfield	EU144060	EU144075
P. kimberleyense	WAC 13293	WA	J. Ray	GU172406	GU172438
P. kimberleyense	WAC 13298	WA	J. Ray	GU172407	GU172439
P. kimberleyense	CBS 122058*	WA	T.I. Burgess & M.J. Wingfield	EU144057	EU144072
P. olivaceum	**CPC 23304**	SA	J.M. van Niekerk	*KJ193680*	*KJ193724*
P. olivaceum	CBS 124939*	SA	J. Roux	FJ888459	FJ888437
P. stromaticum	CMM 3953	Brazil	M.W. Marques	JX464102	JX464109
P. stromaticum	CMM 3961	Brazil	M.W. Marques	JX464103	JX464110
P. stromaticum	CMW 13434*	Venezuela	S. Mohali	AY693974	AY693975
Phyllosticta citricarpa	CBS 111.20*	Australia		FJ538314	FJ538372

New isolates obtained from mango in Thailand and South Africa are indicated in **bold**; novel sequences generated in this study are indicated in *italic*; * represented ex-type

Abbreviation of isolates and culture collection: *B* Department of Plant Pathology and Microbiology, National Taiwan University, Taipei, Taiwan, *BOT* Plant Pathology Research Institute, Giza, Egypt, *CBS* CBS-KNAW Fungal Biodiversity Centre, Utrecht, Netherlands, *CJA* and *IRAN* Iranian Research Institute of Plant Protection, Tehran, Iran, *CMM* Phytopathogenic Fungi of the Universidade Federal Rural dePernambuco, *CMW* Forest and Agricultural Biotechnology Institute, University of Pretoria, Pretoria, South Africa, *CPC* Culture Collection of P. W. Crous, housed at CBS, *MUCC* Murdoch University Culture Collection, Perth, Australia, *WA* Western Australia, *WAC* Department of Agriculture Western Australia Plant Pathogen Collection, Perth, Australia, *SA* South Africa

with *Cophinforma mamane* of Clade IV. Clade V represents species belonging to the genus *Neoscytalidium*, Clades VI species of *Lasiodiplodia*, which divides in seven subclades and Clade VII species of *Barriopsis*, respectively. The isolates obtained from mango in Thailand clustered in Clade VI with Subclades I (*L. pseudotheobromae*), IV (*Lasiodiplodia* sp. which closely relates to *L. iraniensis*), and V (*L. viticola*). Isolates obtained from South Africa clustered in Clade II (*Neofusicoccum parvum*), Clade VI with Subclade VI (*L. theobromae*) and Clade I (*Pseudofusicoccum olivaceum*).

Based on the tree, the global distribution of botryosphaeriacous fungi associated with mango are

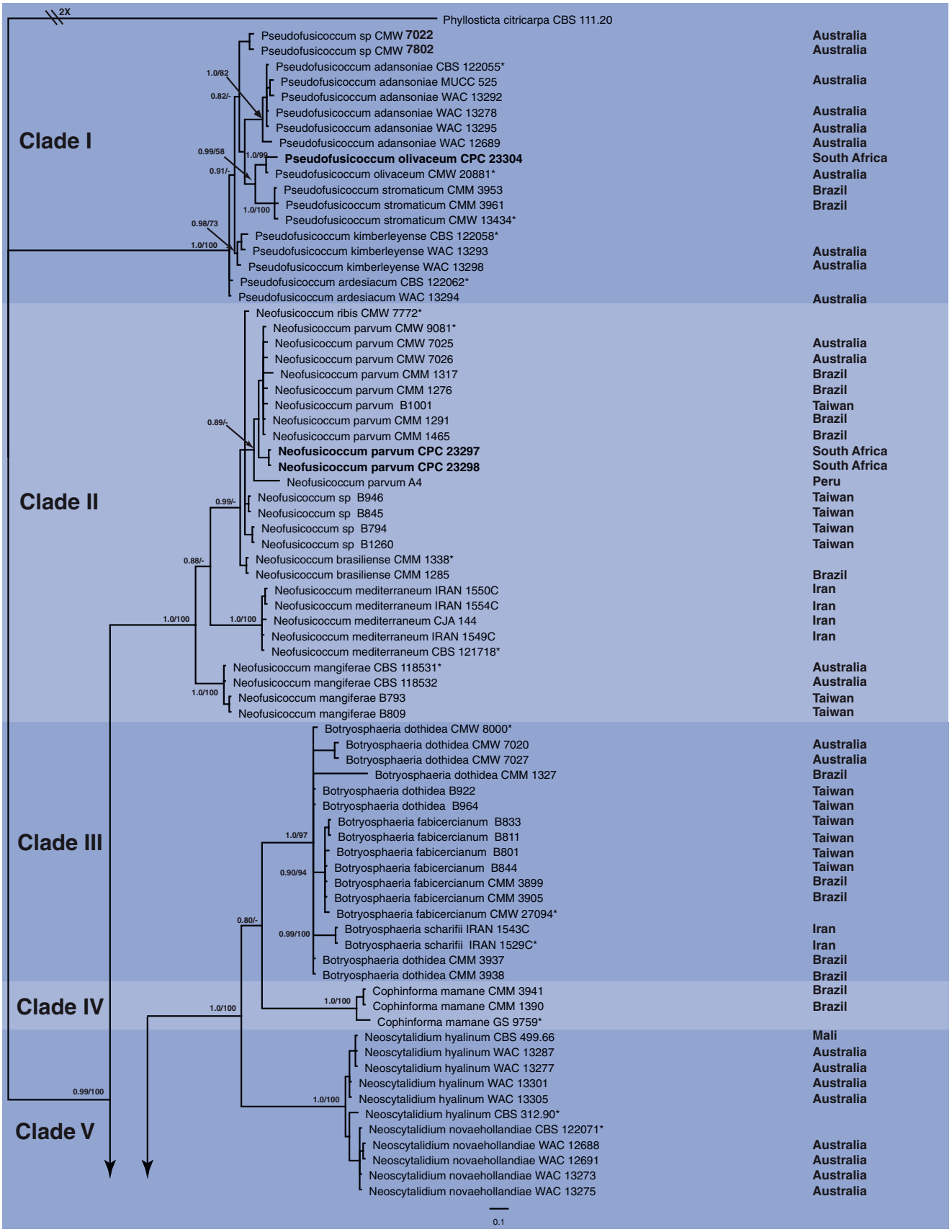

Fig. 1 The Bayesian consensus tree representing the phylogenetic relationship based on the combined sequence datasets of ITS and EF1-α with posterior probability/ bootstrap values indicated at the branches. Isolates from Thailand and South Africa are indicated in *bold* and *asterisk* represents ex-type isolates

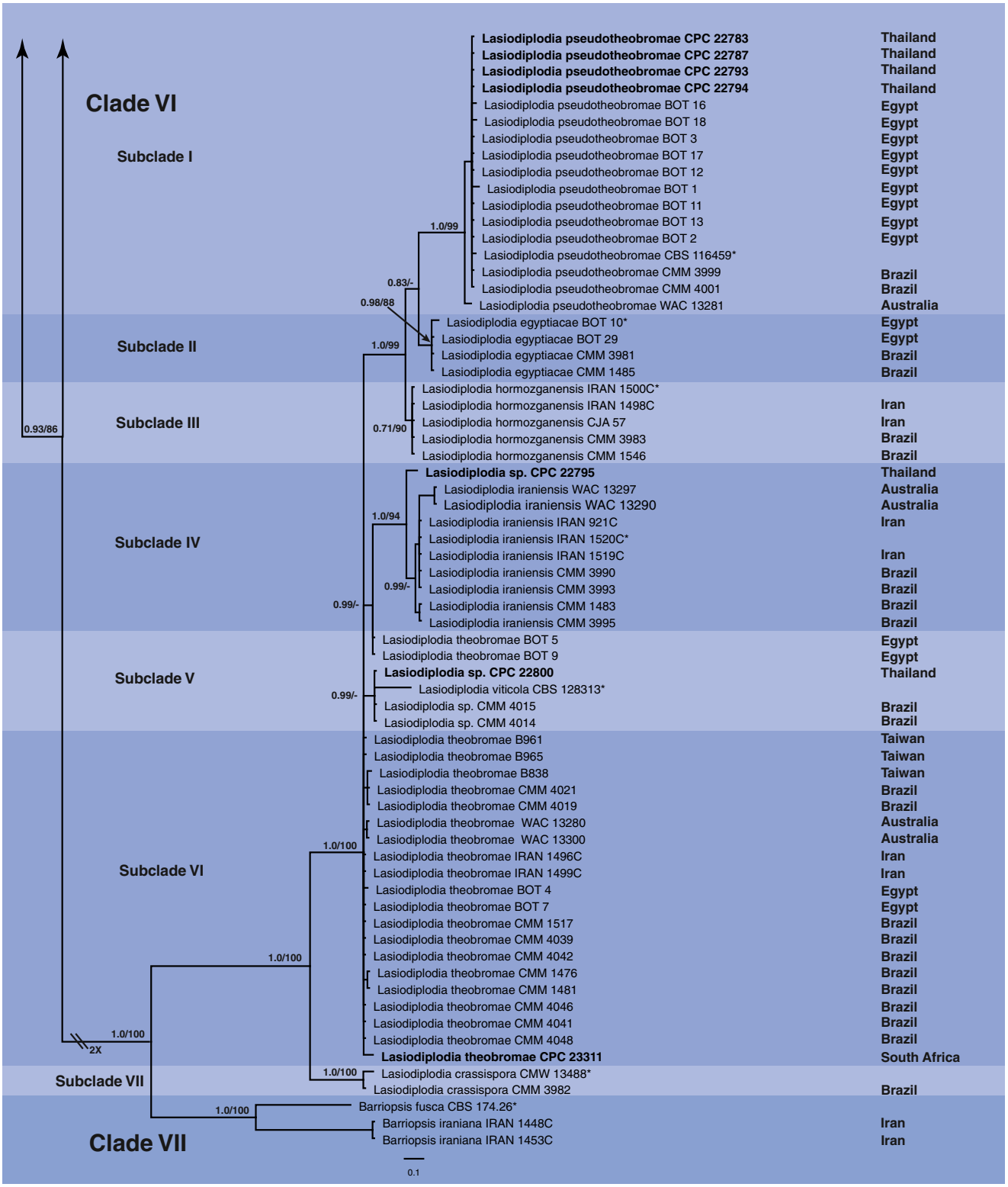

Fig. 1 (continued)

distributed over nine countries which included Australia, Brazil, Egypt, Iran, Mali, Peru, South Africa, Taiwan and Thailand (Table 2). The genera *Neofusicoccum* has been reported in Australia, Brazil, Iran, Peru, South Africa and Taiwan. The genera *Botryosphaeria* has been reported in Australia, Brazil, Iran and Taiwan. The genera *Neoscytalidium*, *Cophinforma* and *Barriopsis* represent the narrowest distribution, with *Neoscytalidium* reported in

Table 2 Distribution of *Botryosphaeriaceae* reported on mango

Species	Country	Reference
Barriopsis iraniana	Iran	Abdollahzadeh et al. 2009
Botryosphaeria dothidea	Australia	Slippers et al. 2005
	Brazil	Marques et al. 2013b
	Taiwan	Ni et al. 2010, 2012a,b
B. fabicercianum	Brazil	Marques et al. 2013a, b
	Taiwan	Ni et al. 2012b
B. scharifii	Iran	Abdollahzadeh et al. 2013
Cophinforma mamane	Brazil	Marques et al. 2013b
Lasiodiplodia sp.	Brazil	Marques et al. 2013a
	Thailand	This study
L. crassispora	Brazil	Marques et al. 2013a
L. egyptiacae	Brazil	Ismail et al. 2012
	Egypt	Marques et al. 2013a
L. hormozganensis	Brazil	Abdollahzadeh et al. 2010
	Iran	Marques et al. 2013a
L. iraniensis	Australia	Sakalidis et al. 2011
	Brazil	Marques et al. 2013a
	Iran	Abdollahzadeh et al. 2010
L. pseudotheobromae	Australia	Sakalidis et al. 2011
	Brazil	Marques et al. 2013a
	Egypt	Ismail et al. 2012
	Thailand	This study
L. theobromae	Australia	Sakalidis et al. 2011
	Brazil	Marques et al. 2013a
	Egypt	Ismail et al. 2012
	Iran	Abdollahzadeh et al. 2010
	South Africa	This study
	Taiwan	Ni et al. 2012b
Neofusicoccum brasiliense	Brazil	Marques et al. 2013b
N. mangiferae	Australia	Slippers et al. 2005
	India	Phillips et al. 2013
	Taiwan	Ni et al. 2012b
N. mediterraneum	Iran	Abdollahzadeh et al. 2013
N. parvum	Australia	Slippers et al. 2005
	Brazil	Marques et al. 2013b
	Peru	Slippers et al. 2005
	South Africa	This study
	Taiwan	Ni et al. 2012b
Neoscytalidium hyalinum	Australia	Sakalidis et al. 2011
	Mali	Pavlic et al. 2008
	Niger	Reckhaus 1987
Ne. novaehollandiae	Australia	Sakalidis et al. 2011
Pseudofusicoccum adansoniae	Australia	Sakalidis et al. 2011
P. ardesiacum	Australia	Sakalidis et al. 2011
P. kimberleyense	Australia	Sakalidis et al. 2011
P. olivaceum	South Africa	This study
P. stromaticum	Brazil	Marques et al. 2013b

Australia and Mali, and *Cophinforma* and *Barriopsis* only reported in Brazil and Iran, respectively. The genera *Lasiodiplodia* represented the largest distribution, reported in countries which included Australia, Brazil, Egypt, Iran, South Africa, Taiwan and Thailand. The members of the genus *Pseudofusicoccum* has been reported in Australia, Brazil and South Africa.

Discussion

Species of *Botryosphaeriaceae* are known to have a cosmopolitan distribution, and to occur on a wide range of plant hosts. On mango these fungi are commonly associated with fruit and stem-end rot, leaf and panicle dieback, black leaf spots, branch cankers and dieback (Slippers et al. 2005; Javier-Alva et al. 2009; Ismail et al. 2012; Ni et al. 2012a, b; Abdollahzadeh et al. 2013; Marques et al. 2013a, b). These fungi have thus far been associated with disease of mango in countries such as Australia, Brazil, Egypt, Iran, Mali, Peru, South Africa, Taiwan and Thailand, where they represent a serious limiting factor for the production and export of mango fruit. Based on the phylogenetic inference in this study, these botryosphaeriacous fungi could be separated into seven genus-specific clades, which in turn clearly highlighted their distribution.

Pavlic et al. (2008) introduced several species of *Pseudofusicoccum*, including *P. adansoniae*, *P. ardesiacum* and *P. kimberleyense*, which were originally isolated from acacia, baobab, *Eucalyptus* and fig. Additionally, they have been reported from mango in Australia (Sakalidis et al. 2011). *Pseudofusicoccum olivaceum* has subsequently been described from wild teak by Mehl et al. (2011), but has not yet been reported from mango. The present study represents the first report of *P. olivaceum* on mango in South Africa, however, the pathogenicity still need to be determined to confirm its pathogenic status on mango. Mohali et al. (2006) initially described *Pseudofusicoccum stromaticum* on branches of *Eucalyptus* in Venezuela, while Marques et al. (2013b) later reported it on mango in Brazil. These species can be distinguished morphologically, as conidia of *P. ardesiacum* and *P. kimberleyense* are larger than *P. adansoniae*, *P. olivaceum* and *P. stromaticum*, whereas conidia of *P. olivaceum* are longer than that of *P. adansoniae* (Pavlic et al. 2008; Mehl et al. 2011).

Species of *Neofusicoccum* that have been reported on mango include *N. brasiliense* from Brazil (Marques et al. 2013b), and *N. mangiferae,* which has been found in Australia (Slippers et al. 2005) and Taiwan (Ni et al. 2012a, b). However, although known to occur in Europe and the USA (Crous et al. 2007; Trouillas et al. 2010; Martin et al. 2011) *N. mediterraneum* has thus far only been reported on mango in Iran (Abdollahzadeh et al. 2013). *Neofusicoccum parvum* is distributed in Australia (Slippers et al. 2005), Brazil (de Oliveira Costa et al. 2010; Marques et al. 2013b), Peru

(Javier-Alva et al. 2009), and Taiwan (Ni et al. 2012a, b). This study represents the first record of this fungal species on mango in South Africa (SA). However, the pathogenicity on mango woody tissues and fruits still need to be determined, especially since all South African isolates were isolated from asymptomatic tissues which included leaves, shoots, fruit panicles or fruits. The study by Ni et al. (2012a,b) reported isolates of *Neofusicoccum* (B794, B845, B946 and B1260) on mango in Taiwan as *N. parvum* based on separate single region analyses of ITS, β-tubulin (TUB) and EF1-α. In contrast, they could only be identified as *Neofusicoccum* sp. belonging to the *N. parvum/ribis* complex in this study based on the combined datasets of ITS and EF1-α. This observation is supported by Slippers and Wingfield (2007), who reported that single gene phylogenies frequently leads to an underestimation of the true species diversity among closely related or cryptic species within the *Botryosphaeriaceae*. Therefore, these isolates should be regarded as yet undescribed members of the *N. parvum/ribis* complex and not *N. parvum* as reported previously.

Botryosphaeria dothidea (*Fusicoccum aesculi*) (Crous et al. 2006), was initially believed to be the dominant species on mango in Australia (Slippers et al. 2005), Brazil (de Oliveira Costa et al. 2010; Marques et al. 2013b), and Taiwan (Ni et al. 2012a, b). However, based on the results of this study, this seems to not be the case. During a survey of mango in Iran by Abdollahzadeh et al. (2013), a new *Botryosphaeria* species, *B. scharifii*, was identified and described. This species can be distinguished from *B. dothidea* based on its smaller conidia, and septate conidiophores (Slippers et al. 2004; Abdollahzadeh et al. 2013). *Botryosphaeria fabicercianum* (= *F.fabicercianum*) was firstly described from *Eucalyptus* in China (Chen et al. 2011), but has subsequently also been observed on mango in Brazil (Marques et al. 2013b).

Cophinforma mamane (= *Botryosphaeria mamane*) was first described from witches' broom of flowering plants in the *Fabaceae* (*Sophora chrysophylla*) in Hawaii (Gardner 1997), and it was later reported in Brazil for the first time associated with mango (Marques et al. 2013b). The phylogenetic inference in this study showed that *C. mamane* is clearly separate from *B. dothidea* and other related species as presented in previous studies (Phillips et al. 2006; Chen et al. 2011; Mehl et al. 2011; Marques et al. 2013b). However, *C. mamane* was closely related to the genus *Neoscytalidium* with low bootstrap support which is in agreement with the finding of Phillips et al. (2013) who placed it in *Cophinforma*.

Crous et al. (2006) introduced the genus *Neoscytalidium*, with *N. hyalinum* (= *N. dimidiatum*) as type, to accommodate fusicoccum-like fungi which produce a *scytalidium*-like synasexualmorph, originally isolated from mango. This genus includes two species, *N. hyalinum* and *N. novaehollandiae* (Pavlic et al. 2008; Phillips et al. 2013), of which the latter species appears to be restricted to Australia (Pavlic et al. 2008). Both species have a large plant host range (Farr et al. 1989; Elshafie and Ba-Omar 2001; Pavlic et al. 2008; Polizzi et al. 2009), which also includes mango. Thus far, both species have been reported from Australia (Ray et al. 2010; Sakalidis et al. 2011) associated with cankers and dieback of mango, with *N. hyalinum* also reported from Mali (Pavlic et al. 2008) and Niger (as *Hendersonula toruloidea*; Reckhaus 1987). During the survey on mango conducted by Ray et al. (2010), *N. hyalinum* was also isolated from fig shrubs (*Ficus carica*) neighbouring a mango plantation, indicating that this fungal pathogen can easily move between different plant hosts, though it has also been reported as opportunistic human pathogen (de Hoog et al. 2004).

Based on a survey of the literature and the phylogenetic inference in this study, the genus *Lasiodiplodia* represents the dominant genus of *Botryosphaeriaceae* causing diseases of mango worldwide. *Lasiodiplodia theobromae* is the dominant species having been reported from Australia (Sakalidis et al. 2011), Brazil (Marques et al. 2013a), Egypt (Ismail et al. 2012), Iran (Abdollahzadeh et al. 2010) and Taiwan (Ni et al. 2012a, b). This study also provides the first report of *L. theobromae* on mango in South Africa. A further six *Lasiodiplodia* species are known from mango, including *L. crassispora* (Brazil; Marques et al. 2013a), *L. egyptiacae* (Brazil and Egypt; Marques et al. 2013a; Ismail et al. 2012), *L. homozganensis* (Brazil, Iran; Marques et al. 2013a; Abdollahzadeh et al. 2010), *L. iraniensis* (Australia, Brazil, Egypt and Iran; Sakalidis et al. 2011; Marques et al. 2013a; Ismail et al. 2012; Abdollahzadeh et al. 2010), *L. pseudotheobromae* (Australia, Brazil, Egypt and Thailand; Sakalidis et al. 2011; Marques et al. 2013a; Ismail et al. 2012; Trakunyingcharoen et al. 2013) and *L. viticola* (Brazil; Marques et al. 2013a).

Since *L. pseudotheobromae* was introduced by Alves et al. (2008), there has been some confusion in distinguishing *L. pseudotheobromae* and *L.theobromae* as their morphological characters overlap, resulting in several misidentifications. Alves et al. (2008) distinguished *L. pseudotheobromae* from *L. theobromae* based on their conidia, with *L. pseudotheobromae* having larger and more ellipsoid conidia than those of *L. theobromae*, which are ovoid shape and have a stronger tapering base. Additionally, *L. pseudotheobromae* produces a dark pink pigment on PDA at 35 °C and is able to grow at 10 °C while *L. theobromae* produce no pigment and cannot grow at 10 °C. Phylogenetic inference employing multiple gene sequences is needed, however, to definitively distinguish between these species (Abdollahzadeh et al. 2010; Ismail et al. 2012; Marques et al. 2013a).

Lasiodiplodia pseudotheobromae has been found on mango in Australia (Sakalidis et al. 2011), Brazil (Marques et al. 2013a) and Egypt (Ismail et al. 2012). To our knowledge,

species of *Lasiodiplodia* associated with mango diseases in Thailand have been mainly reported as *L. theobromae* (Jitareerat et al. 2005; Fahrungsang et al. 2011). The study of Trakunyingcharoen et al. (2013) represented the first report of *L. pseudotheobromae* associated with mango in Thailand.

According to our phylogenetic results (Fig. 1), *L. hormozganensis* and *L. egyptiacae* are closely related species associated with mango diseases. However, they can be distinguished based on their conidia and paraphyses (Ismail et al. 2012). *Lasiodiplodia hormozganensis* has been found on mango in Brazil (Marques et al. 2013a) and Iran (Abdollahzadeh et al. 2010). *Lasiodiplodia egyptiacae* was first described as a new species on mango in Egypt (Ismail et al. 2012), and later found in Brazil (Marques et al. 2013a). This indicates that the distribution of *Lasiodiplodia* species on mango is still unresolved, and might still change depending on the number of mango regions being sampled internationally.

Lasiodiplodia iraniensis has been reported on mango in Australia (Sakalidis et al. 2011), Brazil (Marques et al. 2013a) and Iran (Abdollahzadeh et al. 2010). Isolate CPC 22795, isolated from asymptomatic twigs of mango in Thailand, is shown to be closely related to *L. iraniensis*. However, morphological characters of this isolate indicate that it might be distinct from *L. iraniensis*, as the conidial dimensions ((20–)22–25(–26)×(12–)13–15(–16) μm) overlap with those of *L. iraniensis* ((15.3–)17–23(–29.7)×11–14 μm) whereas the paraphyses were shorter and narrower (25–51×1–1.5 μm) than those of *L. iraniensis* (127×2–4 μm). Therefore, more isolates are needed to determine whether this isolate represents a new species, or variation within *L. iraniensis*.

Lasiodiplodia viticola was described as a new species by Úrbez-Torres et al. (2012) from grapevine dieback in the USA. Marques et al. (2013a) reported a *Lasiodiplodia* sp. on mango in Brazil, which grouped with *L. viticola*. In this study, this same species (CPC 22800) was found on asymptomatic twigs of mango in Thailand. It exhibits similar conidial dimensions ((23–)24–27(–29)×(12–)13–14 μm) to the *Lasiodiplodia* sp. (25.1–27.3×14.1–15 μm) reported in the study of Marques et al. (2013a). This is the first report of this undescribed *Lasiodiplodia* sp. which is closely related to *L. viticola* and associated with mango in Thailand.

Since *Lasiodiplodia crassispora* has been introduced by Burgess et al. (2006) from sandal wood canker in Australia, and as endophyte of *Eucalyptus* in Venezuela, it has been reported on various other plant hosts including grapevine (van Niekerk et al. 2010; Úrbez-Torres et al. 2010) and mango (Marques et al. 2013a). Interestingly, this fungus has only been reported on mango in Brazil (Marques et al. 2013a) and it is clearly separate to other species of *Lasiodiplodia*. Morphological characters showed that the cell wall of immature conidia was notably thicker, striations were wider, and the cytoplasm wart-like in appearance differentiating it from other *Lasiodiplodia* species (Burgess et al. 2006).

Barriopsis iraniana was described as an endophytic fungus from citrus, olive, and also occurs on mango in Iran (Abdollahzadeh et al. 2009). Unlike *Lasiodiplodia*, conidia of *B. iraniana* have obvious striations on immature hyaline conidia even while still attached to their conidiogenous cells (Abdollahzadeh et al. 2009; Phillips et al. 2013).

Results in the present study showed that most genera and species in the *Botryosphaeriaceae* reported from mango were not restricted by geographical boundaries, which could be elucidated by the expansion of horticultural trade around the world. However, it is known that some genera and species of *Botryosphaeriaceae* could be limited in their distribution based on environmental factors prevalent in different regions (Slippers and Wingfield 2007; van Niekerk et al. 2011; Abdollahzadeh et al. 2013). In the genus *Neofusicoccum*, *N. parvum*, has a large distribution on mango in numerous countries, while *N. ribis*, *N. brasiliense* and *N. mediterraneum* have been reported only in Taiwan, Brazil and Iran, respectively. The genera *Botryosphaeria* has been reported on mango in several countries as shown for *B. dothidea*, whereas, *B. fabicercianum* have been reported only in Brazil and *B. scharifii* in Iran (Abdollahzadeh et al. 2013). The genera *Neoscytalidium*, *Cophinforma* and *Barriopsis* represent the narrowest distribution on mango around the world; *C. mamane* and *B. iraniana* have been reported only in Brazil and Iran, respectively. Although, most of *Neoscytalidium* spp. known only from Australia, there has been a single isolate of *N. hyalinum* reported in Mali (Crous et al. 2006). The genus *Pseudofusicocum* is mainly distributed in Australia which include *P. adansoniae*, *P. ardesciacum* and *P. kimberleyense*, however, species of *P. olivaceum* and *P. stromaticum* have now been reported on mango in South Africa and Brazil, respectively.

The genus *Lasiodiplodia* represent the largest geographical distribution reported on mango worldwide; *L. theobromae* appears to be the dominant species followed by *L. pseudotheobromae*, *L. iraniensis* and *Lasiodiplodia* sp. resembling *L. viticola*. However *L. crassispora* appears to be limited to Brazil only. It is surprising that *L. crassispora* was not recovered from mangoes in Kununurra, Australia (Sakalidis et al. 2011) as this is the region from which this fungus was first described on plantation sandalwood (*Santalum album*; Burgess et al. 2006). Although, *L. egyptiacae* and *L. hormozganensis* are known from mango in Egypt (Ismail et al. 2012) and Iran (Abdollahzadeh et al. 2010), respectively, they have also been reported in Brazil (Marques et al. 2013a). This illustrates that the geographical distribution of botryosphaeriacous species on mango is still largely unknown, and might change as more mango growing regions are surveyed and horticulture trade in mangoes increase internationally. A better understanding of the ecology and epidemiology of botryosphaeriacous fungi on mango is required to find effective management strategies for mango

plantation diseases and quarantine, which would significantly reduce the economic loss experienced by the international mango industry.

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