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New geographical records of *Neopestalotiopsis* and *Pestalotiopsis* species in Guangdong Province, China

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Abstract

A study of monocotyledon inhabiting fungi in Guangdong Province, China resulted in the collection of several pestaloid taxa. Evidence from multi-locus phylogenies using ITS, BT and tef $1-\alpha$, together with morphology revealed *Neopestalotiopsis alpapicalis*, *Pestalotiopsis diploclisiae* and *P. parva* from living leaves of *Phoenix roebelenii*. *Pestalotiopsis parva* was also found on a dead petiole of *Phoenix* sp. and *P. diploclisiae* on dead leaves of *Butia* sp. *Pestalotiopsis foedans*, *P. lawsoniae*, *P. macadamia* and *P. virgatula* have been reported in Guangdong Province, and *Pestalotiopsis parva* and *P. diploclisiae* reported for the first time. This *Neopestalotiopsis alpapicalis* collection is the first species of the genus collected from this province. We provide descriptions and illustrations for these three isolates. Additionally, we provide a list of *Pestalotiopsis* and *Neopestalotiopsis* species recorded from China.

Key words – Appendage bearing conidia – Coelomycetes – Monocotyledons – Saprobes – Sporocadaceae

Introduction

Pestalotiopsis Steyaert was introduced to accommodate pestaloid species with 5-celled conidia (Steyaert 1949). Maharachchikumbura et al. (2014) re-examined *Pestalotiopsis* at the morphological and molecular levels and introduced two new genera, *Neopestalotiopsis* and *Pseudopestalotiopsis*. Currently, these three genera placed in Sporocadaceae (Amphisphaeriales) (Wijayawardene et al. 2018, Hyde et al. 2020). *Neopestalotiopsis* typified by *N. protearum* (Crous & L. Swart) Maharachch., K.D. Hyde & Crous, is morphologically distinguished from other pestaloid genera by its varicolored median cells and indistinct conidiophores which are often reduced to conidiogenous cells. *Pestalotiopsis* typified by *P. guepinii* (Desm.) Steyaert and is easily

distinguished from other pestaloid genera as its conidia have concolourous median cells (Maharachchikumbura et al. 2014).

Species in both *Pestalotiopsis* and *Neopestalotiopsis* commonly occur as endophytes in leaves (Hu et al. 2007, Liu et al. 2010, Maharachchikumbura et al. 2012a, Debbab et al. 2013, Chen et al. 2018, Norphanphoun et al. 2019), saprobes on dead leaves (Ariyawansa & Hyde 2018, Tsai et al. 2018), bark and twigs (Ellis & Ellis 1997) or human and animal pathogens (Monden et al. 2013). Some species found from soil, fabrics, wools and some are in the extreme environments (Guba 1961, Strobel et al. 1996, Tejesvi et al. 2007). Some *Pestalotiopsis* species can degrade plastics (Russell et al. 2011). Pestaloid endophytes produce chemical compounds, which use in therapeutic applications and agriculture (Aly et al. 2010, Xu et al. 2010, 2014). Therefore, investigation of novel pestaloid taxa and their chemical properties are of importance.

In this study, we collected three pestaloid taxa from Shenzhen, Guangdong Province, China, and their identifications, and phylogenetic relationships are investigated based on morphology and DNA sequence data of the internal transcribed spacer (ITS), β -tubulin (BT) and partial translation elongation factor 1- α gene (tef 1– α). Additionally, a list of *Pestalotiopsis* and *Neopestalotiopsis* species recorded from China is provided.

Materials & Methods

Sample collection and fungal isolation

Samples were collected in a survey of monocotyledon inhabiting fungi during 2018–2019 in Guangdong Province, China. The samples were brought to the laboratory in paper bags. They were examined and photographed using a Carl Zeiss Discovery V8 stereomicroscope fitted with Axiocam. The morphological characters were photographed using a Nikon Eclipse 80i compound microscope fitted with a Canon 450D digital camera. All microscopic measurements were made with Tarosoft image framework (v. 0.9.0.7). Colony characters were recorded from cultures grown on potato dextrose agar (PDA).

Single conidia isolation was carried out following the method described by Senanayake et al. (2018). Germinated conidia were aseptically transferred into fresh PDA plates, and incubated at 16°C to obtain pure cultures. Cultures were later transferred to PDA slants and stored at 4°C for further studies. All the voucher specimens are deposited in the fungaria of Mae Fah Luang University (MFLU), and living cultures are deposited at the Culture Collection of Kunming Institute of Botany (KUMCC).

DNA extraction, PCR amplification and DNA sequencing

Fungal mycelium grown on PDA for two weeks at 16°C in the dark and fruit bodies directly picked from the specimens were used for DNA extraction using M5 fungal Genomic DNA extraction kit. PCR reactions were carried out using ITS1/ITS4 for internal transcribed spacer nrDNA (ITS) (White et al. 1990), BT2a/BT2b for β -tubulin (BT) (Glass & Donaldson 1995), and EF1-728F/EF2 for translation elongation factor 1- α (tef 1– α) (Rehner 2001, Liu et al. 2017) genes according to the same protocol of Maharachchikumbura et al. (2014).

The amplification reactions were carried out with the following protocol: 25 μ L reaction volume containing 1 μ L of DNA template, 1 μ L of each forward and reverse primers, 12.5 μ L of 2×PCR Master Mix and 9.5 μ L of double-distilled sterilized water (ddH₂O). The PCR products were observed on 1% agarose electrophoresis gel stained with ethidium bromide. Purification and sequencing of PCR products were carried out at the Sunbiotech Company, Beijing, China. Sequence quality was checked and sequences were concatenated with DNASTAR Lasergene v.7.1. Sequences derived in this study were deposited in the GenBank, and accession numbers were obtained (Table 1).

Sequence alignment and phylogenetic analyses

BLASTn searches were made using the newly generated sequences to assist taxon sampling

analyses. All sequences obtained from GenBank for phylogenetic and used bv Maharachchikumbura et al. (2014, 2016), Liu et al. (2017), Nozawa et al. (2017), Ariyawansa & Hyde (2018), Chen et al. (2018), Tibpromma et al. (2018), Tsai et al. (2018), Watanabe et al. (2018), Norphanphoun et al. (2019), are listed in Table 1. DNA sequence data of the ITS, BT and tef 1– α sequence alignments were done using default settings of MAFFT v.7 (Katoh et al. 2017) and manually adjusted using BioEdit 7.1.3 (Hall 1999) to allow maximum alignment and minimum gaps. The evolutionary models for phylogenetic analyses were determined by MrModeltest v. 2.3 under the Akaike Information Criterion (AIC) was implemented in PAUP v. 4.0b10 (Nylander 2004).

Maximum likelihood analysis was performed by RAxML (Stamatakis & Alachiotis 2010) implemented in raxmlGUIv.1.3 (Silvestro & Michalak 2012). The search strategy was set to rapid bootstrapping, and the analysis carried out using the GTRGAMMAI model of nucleotide substitution with 1,000 replicates.

For the Bayesian inference (BI) analyses of the individual loci and concatenated ITS, BT and tef $1-\alpha$ alignment, the above-mentioned model test was used to determine the best fitting nucleotide substitution model settings for MrBayes v. 3.0b4. Dirichlet base frequencies and the GTR+I+G model with inverse gamma-distributed rate were predicted by the MrModeltest analysis for all three data partitions and used in the Bayesian analysis.

The Markov Chain Monte Carlo sampling (MCMC) resulted in MrBayes v. 3.0b4 (Huelsenbeck et al. 2003) was used to calculate Posterior probability values (Zhaxybayeva & Gogarten 2002). Four simultaneous Markov chains were initially run for 10,000,000 generations, and every 500th generation was sampled. The distribution of log-likelihood scores was observed to check whether sampling is in stationary phase or not and Tracer v1.5 was used to check if further runs were required to reach convergence or not (Rambaut & Drummond 2007).

The Bayesian analysis lasted 10,000,000 generations (average standard deviation of split frequencies value = 0.0098), and the consensus tree and posterior probabilities were calculated after discarding the first 20% of sampled trees as burn-in. The remaining trees were used for calculating posterior probabilities in the majority rule consensus tree. The bootstrap values equal to or greater than 0.9 are given below or above each node (Figs 1, 2). The phylogram was visualized in FigTree v. 1.2.2 (Rambaut & Drummond 2008).

Taxon Culture accession numb		Genbank number		
		ITS β-tubulin tef 2		tef 1–α
Neopestalotiopsis acrostichi	MFLUCC 17-1754	MK764272	MK764338	MK764316
Neopestalotiopsis alpapicalis	MFLUCC 17-2544	MK357772	MK463545	MK463547
Neopestalotiopsis alpapicalis	KUMCC 20-0036	MT222276	MT135199	MT175375
Neopestalotiopsis alpapicalis	KUMCC 20-0037	MT222277	MT135200	MT175376
Neopestalotiopsis aotearoa	CBS 367.54	KM199369	KM199454	KM199526
Neopestalotiopsis asiatica	MFLUCC 12-0286	JX398983	JX399018	JX399049
Neopestalotiopsis australis	CBS 114159	KM199348	KM199432	KM199537
Neopestalotiopsis brachiata	MFLUCC 17-1555	MK764274	MK764340	MK764340
Neopestalotiopsis brasiliensis	PA10	N/A	MK286948	MK253112
Neopestalotiopsis chiangmaiensis	MFLUCC 18-0113	N/A	MH412725	MH388404
Neopestalotiopsis chrysea	MFLUCC 12-0261	JX398986	JX399021	JX399052
Neopestalotiopsis clavispora	MFLUCC 12-0281	MN121843	MN121844	MN121845
Neopestalotiopsis cocoes	MFLU 15-0220	NR-156312	N/A	N/A
Neopestalotiopsis coffea-arabicae	HGUP 4015	KF412647	KF412641	KF412644
Neopestalotiopsis cubana	CBS 600.96	KM199347	KM199438	KM199521
Neopestalotiopsis egyptiaca	PEST1	KP943747	KP943746	KP943748
Neopestalotiopsis ellipsospora	MFLUCC 12-0283	JX398981	JX399015	JX399046
Neopestalotiopsis eucalypticola	CBS 264.37	KM199376	KM199431	KM199551
Neopestalotiopsis foedans	CGMCC 3.9123	JX398987	JX399022	JX399053
Neopestalotiopsis formicarum	CBS 362.72	KM199358	KM199455	KM199517

Table 1 Details of the isolates used in the phylogenetic tree. Newly generated sequences are bold.

Table 1 Continued.

Taxon	Culture accession number	Genbank number		
1 4 2 0 1	Culture accession number	$\frac{1}{1} \frac{1}{\alpha}$		
Neopestalotiopsis honoluluana	CBS 111535	N/A	KM199461	KM199546
Neopestalotiopsis honoluluana	CBS 114495	KM199364	KM199457	KM199548
Neopestalotiopsis nonotatiana Neopestalotiopsis iraniensis	P815	N/A	N/A	N/A
Neopestalotiopsis trainensis Neopestalotiopsis javaensis	CBS 257.31	KM199357	KM199457	KM199548
Neopestalotiopsis javaensis Neopestalotiopsis keteleeria	MFLUCC 13-0915	KJ503820	KJ503821	KJ503822
Neopestalotiopsis keleleettä Neopestalotiopsis macadamiae	BRIP 63737c	NR-161002	KX186654	KX186627
Neopestalotiopsis macaaamae Neopestalotiopsis magna	MFLUCC 12-0055	KF582795	KF582793	KF582791
Neopestalotiopsis magna Neopestalotiopsis mesopotamica	CBS 464.69	KM199353	KM199436	N/A
Neopestalotiopsis mesopolamica Neopestalotiopsis musae	MFLUCC 15-0776	NR-156311	KX789686	KX789685
	CBS 138.41	NR-156288	KM199466	KM199552
Neopestalotiopsis natalensis	KUMCC 17-0175	N/A	MH412720	MH388389
Neopestalotiopsis pandanicola	RV01	KJ792466	N/A	N/A
Neopestalotiopsis pernambucana	MFLUCC 17-1737	MK764276	N/A MK764342	MK764320
Neopestalotiopsis petila	MFLUCC 18-0119	MH388354	MH412721	MK704320 MH388390
Neopestalotiopsis phangngaensis	CBS 225.30		KM199451	мп388390 КМ199535
Neopestalotiopsis piceana		KM199371		
Neopestalotiopsis piceana	CBS 394.48	KM199368	KM199453	KM199527
Neopestalotiopsis protearum	CBS 114178	JN712498	KM199463	KM199542
Neopestalotiopsis rhizophorae	MFLUCC 17-1550	MK764277	MK764343	MK764321
Neopestalotiopsis rosae	CBS 124745	KM199360	KM199430	KM199524
Neopestalotiopsis rosicola	CFCC 51992	KY885239	KY885245	KY885243
Neopestalotiopsis samarangensis	CBS 115451	KM199365	KM199447	KM199556
Neopestalotiopsis saprophytica	CBS 115452	KM199345	KM199433	KM199538
Neopestalotiopsis saprophytica	MFLUCC 12-0282	JX398982	JX399017	JX399048
Neopestalotiopsis sonneratae	MFLUCC 17-1744	MK764280	MK764346	MK764324
Neopestalotiopsis sp.	CBS 266.37	KM199349	KM199459	KM199547
Neopestalotiopsis sp.	CBS 323.76	KM199350	KM199458	KM199550
Neopestalotiopsis sp.	FMB 0127	N/A	MH460876	MH523647
Neopestalotiopsis sp.	FMB 0128	N/A	MH460875	MH523646
Neopestalotiopsis sp.	CBS 119.75	KM199356	KM199439	KM199531
Neopestalotiopsis sp.	LC3318	KX894964	KX895296	KX895181
Neopestalotiopsis sp.	LC6285	KX895013	KX895346	KX895232
Neopestalotiopsis sp.	LC6471	KX895019	KX895352	KX895238
Neopestalotiopsis sp.	LPS61	MF379331	N/A	N/A
Neopestalotiopsis sp.	SC2A3	KU252210	KU252477	KU252390
Neopestalotiopsis sp.	SC2A4	KX146639	KX146757	KX146698
Neopestalotiopsis sp.	SC3A3	KU252211	KU252478	KU252391
Neopestalotiopsis sp.	SC5A9	KU252212	KU252479	KU252392
Neopestalotiopsis sp.	YN1A5	KU252216	KU252483	KU252396
Neopestalotiopsis sp.	ZJ1A2	KU252215	KU252482	KU252395
Neopestalotiopsis sp.	CBS 274.29	KM199375	KM199448	KM199534
Neopestalotiopsis sp.	CBS 322.76	KM199366	KM199446	KM199536
Neopestalotiopsis sp.	CBS 360.61	KM199346	KM199440	KM199522
Neopestalotiopsis sp.	CBS 110.20	KM199342	KM199442	KM199540
Neopestalotiopsis sp.	CBS 164.42	KM199367	KM199434	KM199520
Neopestalotiopsis sp.	URM7148	N/A	N/A	KU306740
Neopestalotiopsis steyaertii	IMI 192475	KF582796	KF582794	KF582792
Neopestalotiopsis sucjuenti Neopestalotiopsis surinamensis	CBS 111494	N/A	KM199462	KM199530
Neopestalotiopsis surinamensis	CBS 450.74	KM199351	KM199465	KM199518
Neopestalotiopsis surmanensis Neopestalotiopsis thailandica	MFLUCC 17-1730	MK764281	MK764347	MK764325
Neopestalotiopsis indianatea	MFLUCC 12-0285	JX398984	JX399019	JX399050
Neopestalotiopsis unbrinospora Neopestalotiopsis vitis	JZB340018	KU140694	KU140685	KU140676
	CBS 111495	JX556231	KU140085 KM199456	KM199545
Neopestalotiopsis zimbabwana Pastalotiopsis adusta	ICMP 6088	JX356251 JX399006	XM199456 JX399037	XM199545 JX399070
Pestalotiopsis adusta	MFLUCC 10-0146	JX399006 JX399007	JX399037 JX399038	JX399070 JX399071
Pestalotiopsis adusta				
Pestalotiopsis aggestorum	LC6301	KX895015	KX895348	KX895234

Table 1 Continued.

Taxon	Culture accession number	Genbank number		
		ITS β-tubulin tef 1–α		
Pestalotiopsis aggestorum	LC8186	KY464140	KY464160	KY464150
Pestalotiopsis anacardiacearum	IFRDCC 2397	KC247154	KC247155	KC247156
Pestalotiopsis arceuthobii	CBS 434.65	KM199341	KM199427	KM199516
Pestalotiopsis arengae	CBS 331.92	KM199340	KM199426	KM199515
Pestalotiopsis australasiae	CBS 114126	KM199297	KM199409	KM199499
Pestalotiopsis australasiae	CBS 114141	KM199298	KM199410	KM199501
Pestalotiopsis australis	CBS 114193	KM199334	KM199385	KM199477
Pestalotiopsis australis	CBS 119350	KM199333	KM199384	KM199476
Pestalotiopsis biciliata	CBS 124463	KM199308	KM199399	KM199505
Pestalotiopsis biciliata	CBS 790.68	MH859228	KM199400	KM199507
Pestalotiopsis brachiata	LC2988	KX894933	KX895265	KX895150
Pestalotiopsis brachiata	LC8188	KY464142	KY464162	KY464152
Pestalotiopsis brassicae	CBS 170.26	KM199379	N/A	KM199558
Pestalotiopsis camelliae	CBS 443.62	KM199336	KM199424	KM199512
Pestalotiopsis camelliae	MFLUCC 12-0277	KY319138	KY363542	KY342366
Pestalotiopsis chamaeropis	CBS 113604	KM199323	KM199389	KM199471
Pestalotiopsis chamaeropis	CBS 186.71	KM199325	KM199390	KM199472
Pestalotiopsis chinensis	LC3013	KX894939	KX895271	KX895156
Pestalotiopsis clavata	MFLUCC 12-0268	JX398990	JX399025	JX399056
Pestalotiopsis colombiensis	CBS 118553	KM199307	KM199421	KM199488
Pestalotiopsis colombiensis	ICMP 5434	KP781879	KP781883	N/A
Pestalotiopsis diploclisiae	CBS 115587	KM199314	KM199416	KM199485
Pestalotiopsis diploclisiae	KUMCC 20–0035	MT222272	N/A	MT175371
Pestalotiopsis distincta	LC3232	KX894961	KX895293	KX895178
-	MFLUCC 12-0287	NR_120187	JX399040	JX399073
Pestalotiopsis diversiseta	MFLUCC 10-0149	KP781877	JA399040 N/A	KP781880
Pestalotiopsis dracontomelon				
Pestalotiopsis ericacearum	OP023	KC537807	KC537821	KC537814
Pestalotiopsis formosana	NTUCC 17-0010	MH809382	MH809386	MH809390
Pestalotiopsis formosana	NTUCC 17-0009	MH809381	MH809385	MH809389
Pestalotiopsis funerea	ML4DY	EF055197	EF055234	N/A
Pestalotiopsis furcata	MFLUCC 12-0054	JQ683724	JQ683708	JQ683740
Pestalotiopsis gaultheria	IFRD 411-014	KC537805	KC537819	KC537812
Pestalotiopsis gibbosa	Pes6	LC311589	LC311590	LC311591
Pestalotiopsis grevilleae	CBS 114127	KM199300	KM199407	KM199504
Pestalotiopsis hawaiiensis	CBS 114491	KM199339	KM199428	KM199514
Pestalotiopsis hollandica	CBS 265.33	KM199328	KM199388	KM199481
Pestalotiopsis humus	CBS 115450	KM199319	KM199418	KM199487
Pestalotiopsis humus	CBS 336.97	KM199317	KM199420	KM199484
Pestalotiopsis inflexa	MFLUCC 12-0270	JX399008	JX399039	JX399072
Pestalotiopsis intermedia	MFLUCC 12-0259	JX398993	JX399028	JX399059
Pestalotiopsis italiana	MFLUCC 12-0657	KP781878	KP781882	KP781881
Pestalotiopsis jesteri	CBS 109350	KM199380	KM199468	KM199554
Pestalotiopsis jiangxiensis	LC4399	KX895009	KX895341	KX895227
Pestalotiopsis jinchanghensis	LC6636	KX895028	KX895361	KX895247
Pestalotiopsis jinchanghensis	LC8190	KY464144	KY464164	KY464154
Pestalotiopsis kenyana	CBS 442.67	KM199302	KM199395	KM199502
Pestalotiopsis kenyana	CBS 911.96	KM199303	KM199396	KM199503
Pestalotiopsis knightiae	CBS 111963	KM199311	KM199406	KM199495
Pestalotiopsis knightiae	CBS 114138	KM199310	KM199408	KM199497
Pestalotiopsis licualacola	HGUP 4057	KC492509	KC481683	KC481684
Pestalotiopsis linearis	MFLUCC 12-0271	JX398992	JX399027	JX399058
Pestalotiopsis longiappendiculata	LC3013	KX894939	KX895271	KX895156
Pestalotiopsis lushanensis	LC4344	KX895005	KX895337	KX895223
Pestalotiopsis lushanensis	LC8182	KY464136	KY464156	KY464146
Pestalotiopsis macadamiae	BRIP 63738b	KX186588	KX186680	KX186621

Table 1 Continued.

Taxon	Culture accession number	(Genbank number		
		ITS	β-tubulin	tef 1–α	
Pestalotiopsis malayana	CBS 102220	KM199306	KM199411	KM199482	
Pestalotiopsis microspora	UMAS P15	KT337388	N/A	N/A	
Pestalotiopsis monochaeta	CBS 144.97	KM199327	KM199386	KM199479	
Pestalotiopsis monochaeta	CBS 440.83	KM199329	KM199387	KM199480	
Pestalotiopsis montellica	MFLUCC 12-0279	JX399012	JX399043	JX399076	
Pestalotiopsis neglecta	1100	AB482220	LC311599	LC311600	
Pestalotiopsis neolitseae	NTUCC 17-011	MH809383	MH809387	MH809391	
Pestalotiopsis novae-hollandiae	CBS 130973	KM199337	KM199425	KM199511	
Pestalotiopsis oryzae	CBS 111522	KM199294	KM199394	KM199493	
Pestalotiopsis oryzae	CBS 171.26	MH854881	KM199397	KM199494	
Pestalotiopsis paeoniicola	TR40	N/A	KY930634	N/A	
Pestalotiopsis paeoniicola	TR41	N/A	KY930635	N/A	
Pestalotiopsis pallidotheae	110	N/A	LC311584	LC311585	
Pestalotiopsis papuana	CBS 331.96	KM199321	KM199413	KM199491	
Pestalotiopsis papuana	CBS 887.96	KM199318	KM199415	KM199492	
Pestalotiopsis parva	KUMCC 20-0038	MT222274	MT135197	MT175373	
Pestalotiopsis parva	MFLU 20-0060	MT222275	MT135198	MT175374	
Pestalotiopsis parva	CBS 265.37	KM199312	KM199404	KM199508	
Pestalotiopsis parva	CBS 278.35	KM199313	KM199405	KM199509	
Pestalotiopsis portugalica	CBS 393.48	KM199335	KM199422	KM199510	
Pestalotiopsis rhizophorae	MFLUCC 17-0417	MK764284	MK764350	MK764328	
Pestalotiopsis rhododendri	CBS 144024	MH554109	MH554782	MH554543	
Pestalotiopsis rhododendri	OP086	KC537804	KC537818	KC537811	
Pestalotiopsis rosea	MFLUCC 12-0258	JX399005	JX399036	JX399069	
Pestalotiopsis scoparia	CBS 176.25	KM199330	KM199393	KM199478	
Pestalotiopsis shorea	MFLUCC 12-0314	KJ503811	KJ503814	KJ503817	
Pestalotiopsis sp.	UMAS 1705	KT337373	N/A	N/A	
Pestalotiopsis sp.	CBS 263.33	KM199316	KM199414	KM199489	
Pestalotiopsis sp.	CBS 264.33	KM199322	KM199412	KM199490	
Pestalotiopsis sp.	HGUP 4057	KC492509	KC481683	KC481684	
Pestalotiopsis spathulata	CBS 356.86	KM199338	KM199423	KM199513	
Pestalotiopsis telopeae	CBS 113606	KM199295	KM199402	KM199498	
Pestalotiopsis telopeae	CBS 114161	KM199301	KM199469	KM199559	
Pestalotiopsis thailandica	MFLUCC 17-1616	MK764285	MK764351	MK764329	
Pestalotiopsis theae	CMU-ELA1	JX205216	N/A	N/A	
Pestalotiopsis theae	CPO Pe	JQ619652	N/A	N/A	
Pestalotiopsis trachicarpicola	CBS 111507	MH553960	MH554619	MH554378	
Pestalotiopsis trachicarpicola	HN 56.2	N/A	MK360941	MK512494	
Pestalotiopsis unicolor	MFLUCC 12-0275	JX398998	JX399029	MK512494	
Pestalotiopsis unicolor	MFLUCC 12-0276	JX398999	JX399030	N/A	
Pestalotiopsis verruculosa	MFLUCC 12-0274	JX398996	N/A	JX399061	
Pestalotiopsis yanglingensis	LC3067	KX894949	KX895281	KX895166	
Pestalotiopsis yanglingensis	LC4553	KX895012	KX895345	KX895231	

Abbreviations: BRIP: The Plant Pathology Herbarium, Queensland, Australia; CBS: Culture collection of the Centraalbureau voor Schimmelcultures, Fungal Biodiversity Centre, Utrecht, The Netherlands; CFCC: Chinese Forestry Culture Collection Center, Chinese Academy of Sciences, Beijing, China; CGMCC: China General Microbiological Culture Collection Center, Institute of Microbiology, Chinese Academy of Sciences, Beijing, China; HGUP: The Plant Pathology Herbarium of Guizhou University, China; ICMP: International Collection of Microorganisms from Plants, Auckland, New Zealand; IFRDCC: International Fungal Research & Development Centre Culture Collection, China; IMI: Culture collection of CABI Europe UK Centre, Egham, UK; KUMCC: Culture Collection of Kunming Institute of Botany, Chinese Academy of Sciences, Kunming, China; MFLU: Mae Fah Luang University Herbarium, Chiang Rai; Thailand; MFLUCC: Mae Fah Luang University Culture Collection, Taiwan; UMAS: Department of Plant Science and Environmental Ecology, Faculty of Resource Science and Technology, University Malaysia Sarawak

Results

Phylogenetic inferences

The first combined BT, ITS and tef $1-\alpha$ sequence dataset comprised 102 strains of *Pestalotiopsis*, and *Neopestalotiopsis* sp. (CBS 119.75) was the outgroup taxon. The second combined BT, ITS and tef $1-\alpha$ sequence data set comprised 72 sequences of *Neopestalotiopsis* with *Pestalotiopsis parva* (CBS 265.37) as the outgroup taxon. Both concatenated data matrixes comprised 1527 characters (ITS: 566, BT: 469 and tef $1-\alpha$: 490). All individual trees generated under different criteria. Single-gene datasets were essentially similar in topology and not significantly different from the tree generated from the concatenated dataset (not discussed herein).

Maximum likelihood analysis for *Pestalotiopsis* with 1,000 bootstrap replicates yielded a tree with the likelihood value of ln: -13138.225580 and the following model parameters: alpha: 0.550988; $\Pi(A)$: 0.239550, $\Pi(C)$: 0.287070, $\Pi(G)$: 0.215467 and $\Pi(T)$: 0.257914. Maximum likelihood analysis for *Neopestalotiopsis* with 1,000 bootstrap replicates yielded a tree with the likelihood value of ln -6466.001103 and the following model parameters: alpha: 0.731256; $\Pi(A)$: 0.234344, $\Pi(C)$: 0.266539, $\Pi(G)$: 0.216132 and $\Pi(T)$: 0.282985. The ML analyses also resulted in similar tree topologies to those obtained in the Bayesian analyses. The best scoring RAxML trees derived from the analyses of the concatenated datasets for *Pestalotiopsis (*ingroup) and *Neopestalotiopsis (*ingroup) are shown in Figs 1, 2, respectively. Maximum likelihood bootstrap values \geq 50% and Bayesian inference (BI) \geq 0.9 are given at each node.

In our concatenated ML analyses, one *Pestalotiopsis* isolate (KUMCC 20–0035) form a distinct subclade with *P. diploclisiae* (CBS 115449) with high statistical support (Fig. 1). In addition, the other two isolates of *Pestalotiopsis* (KUMCC 20–0038 and MFLU 20–0060) form a separate, high statistical supported lineage with *Pestalotiopsis parva* (CBS 278.35 and CBS 265.37). Hence, *Pestalotiopsis* strains of KUMCC 20–0038/MFLU 20–0060 is confirmed as *Pestalotiopsis parva* (Fig. 1). In the second dataset, two strains of *Neopestalotiopsis* form a distinct subclade with *Neopestalotiopsis alpapicalis* and *N. rhizophorae* with high statistical support. Hence, this collection is proposed here as *Neopestalotiopsis alpapicalis* (Fig. 2).

Taxonomy

Pestalotiopsis diploclisiae Maharachch., K.D. Hyde & Crous, in Maharachchikumbura, Hyde, Groenewald, Xu & Crous, Stud. Mycol. 79: 160 (2014) Fig. 3

Facesoffungi number: FoF 06982

Saprobic, associated with dead leaves of *Butia* sp. Sexual morph: Undetermined. Asexual morph: *Conidiomata* 500–900 µm diam., pycnidial, globose, blackish brown, immersed on substrate, semi-immersed in PDA, releasing conidia as a black, slimy, globose, glistening mass on culture media. *Conidiophores* indistinct or reduced to conidiogenous cells. *Conidiogenous cells* 5– $20 \times 2-3 \mu m$ ($\overline{x} = 11 \times 2.8 \mu m$, n = 20), discrete, lageniform, hyaline, smooth-walled, annellidic, proliferating 2–3 times percurrently, collarette present, may not appears as flared. *Conidia* 18–25 × 5–7 µm ($\overline{x} = 23 \times 6 \mu m$, n = 20), fusiform to clavate, straight to slightly curved, wall of one side curved than other side, 4-euseptate; basal cell obconic with a truncate base, hyaline or sometimes greenish brown, thick-and smooth-walled, 2–3.5 µm long ($\overline{x} = 2.9 \mu m$, n = 20); three median cells ± equal, each 4–5 µm long ($\overline{x} = 4 \mu m$), doliiform or trapezoid, concolorous, pale brown, septa and periclinal walls darker than rest of the cell, wall smooth; apical cell 3–5 µm long ($\overline{x} = 3.9 \mu m$, n = 20), hyaline, conic to acute with truncate base; apical appendages 8–13 × 0.5–1 µm ($\overline{x} = 11.5 \times 0.5 \mu m$, n = 40), 3–4 (mostly 3), tubular, inserted at different loci but in a crest at the apex of the apical cell, unbranched, flexuous, rough; single basal appendage, tubular, unbranched, centric, 3–6 µm long ($\overline{x} = 4.9 \mu m$, n = 20).

Culture characteristics – Colonies on PDA reaching 2 cm diam., after 1 week at 18°C, under dark, colonies circular, medium dense, aerial mycelium on surface raised, white from above and reverse; fruiting bodies appears as black slimy bubbles.

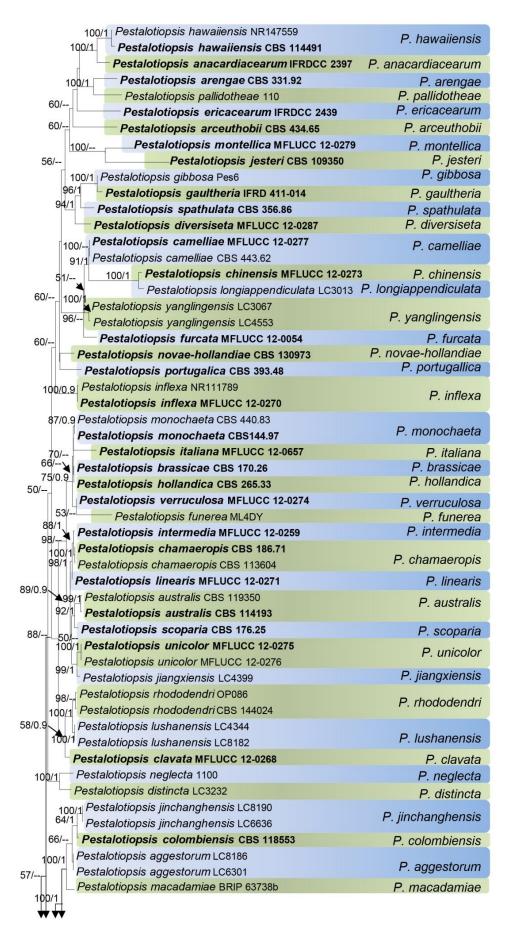


Fig. 1 – Phylogram generated from maximum likelihood analysis based on combined ITS, BT and tef 1– α sequence data. Bootstrap support values for ML greater than 50% and Bayesian posterior

probabilities greater than 0.9 are given near nodes respectively. The tree is rooted with *Neopestalotiopsis* sp (CBS 119.75). Ex-type strains are in black bold and the newly generated sequences are indicated in blue bold.

96/1 Pestalotiopsis diploclisiae KUMCC 20–0035	P. diploclisiae
72/0.9 Pestalotiopsis diploclisiae CBS 115587	F. diplociisiae
Pestalotiopsis humus CBS 336.97	
^{50/} Pestalotiopsis humus CBS 115450	P. humus
58/ 88/1 Pestalotiopsis sp UMAS 1705	
60/ Pestalotiopsis microspora UMAS P15	P. microspora
52/ Pestalotiopsis malayana CBS 102220	P. malayana
64/- Pestalotiopsis sp CBS 264.33	
67// Pestalotiopsis sp CBS 263.33	
^{69/-} Pestalotiopsis papuana CBS 887.96	P. papuana
96/1 Pestalotiopsis papuana CBS 331.96	r : papaana
87/1 Pestalotiopsis adusta ICMP6088	P. adusta
^{87/1} Pestalotiopsis adusta MFLUCC 10-0146	
52/ Pestalotiopsis neolitseae NTUCC 17-011	P. neolitseae
Pestalotiopsis shorea MFLUCC 12-0314	P. shorea
Pestalotiopsis digitalis ICMP 5434	P. digitalis
91/1 <i>Pestalotiopsis parva</i> KUMCC 20-0038	
71/+- Pestalotiopsis parva MFLU 20-0060	P. parva
Pestalotiopsis parva CBS 278.35	
99/1 Pestalotiopsis parva CBS 265.37	
97/1 Pestalotiopsis formosana NTUCC 17-009	P. formosana
82/ Pestalotiopsis formosana NTUCC 17-0010	
Pestalotiopsis rhizophorae MFLUCC 17-0417 91/1 Pestalotiopsis thailandica MFLUCC 17-1616	P. rhizophorae
100/1] Pestalotiopsis knightiae CBS 111963	P. thailandica
59/ Pestalotiopsis knightiae CBS 114138	P. knightiae
Pestalotiopsis dracontomelon MFLUCC 10-0149	P. dracontomelon
57/ Pestalotiopsis rosea MFLUCC 12-0258	P. dracomonielon P. rosea
Pestalotiopsis grevilleae CBS 114127	
	Parevillege
	P. grevilleae
100/1 Pestalotiopsis brachiata LC8188	P. grevilleae P. barachiata
100/1 Pestalotiopsis brachiata LC8188 Pestalotiopsis brachiata LC2988 Pestalotiopsis biciliata CBS 124463	P. barachiata
100/1 Pestalotiopsis brachiata LC8188 Pestalotiopsis brachiata LC2988 Pestalotiopsis biciliata CBS 124463	
100/1 Pestalotiopsis brachiata LC8188 Pestalotiopsis brachiata LC2988	P. barachiata P. biciliata
100/1 Pestalotiopsis brachiata LC8188 Pestalotiopsis brachiata LC2988 Pestalotiopsis biciliata CBS 124463 98/1 Pestalotiopsis biciliata CBS 790.68	P. barachiata
100/1 Pestalotiopsis brachiata LC8188 Pestalotiopsis brachiata LC2988 Pestalotiopsis biciliata CBS 124463 98/1 Pestalotiopsis biciliata CBS 790.68 100/1 Pestalotiopsis australasiae CBS 114141	P. barachiata P. biciliata P. australasiae
100/1 Pestalotiopsis brachiata LC8188 Pestalotiopsis brachiata LC2988 98/1 Pestalotiopsis biciliata CBS 124463 98/1 Pestalotiopsis biciliata CBS 790.68 100/1 Pestalotiopsis australasiae CBS 114141 Pestalotiopsis australasiae CBS 114141 Pestalotiopsis telopeae CBS 114161 70/- Pestalotiopsis telopeae CBS 113606	P. barachiata P. biciliata
100/1 Pestalotiopsis brachiata LC8188 Pestalotiopsis brachiata LC2988 98/1 Pestalotiopsis biciliata CBS 124463 98/1 Pestalotiopsis biciliata CBS 124463 98/1 Pestalotiopsis biciliata CBS 124463 98/1 Pestalotiopsis australasiae CBS 114141 Pestalotiopsis australasiae CBS 114141 Pestalotiopsis telopeae CBS 114161 84/- Pestalotiopsis telopeae CBS 113606 61/ Pestalotiopsis macadamiae BRIP 63738b	P. barachiata P. biciliata P. australasiae
100/1 Pestalotiopsis brachiata LC8188 Pestalotiopsis brachiata LC2988 98/1 Pestalotiopsis biciliata CBS 124463 98/1 Pestalotiopsis biciliata CBS 790.68 100/1 Pestalotiopsis australasiae CBS 114141 Pestalotiopsis australasiae CBS 114141 Pestalotiopsis telopeae CBS 114126 84/1 Pestalotiopsis telopeae CBS 113606 61/ Pestalotiopsis macadamiae BRIP 63738b Pestalotiopsis oryzae CBS 111522	P. barachiata P. biciliata P. australasiae P. telopeae P. macadamiae
100/1 Pestalotiopsis brachiata LC8188 Pestalotiopsis brachiata LC2988 98/1 Pestalotiopsis biciliata CBS 124463 98/1 Pestalotiopsis biciliata CBS 790.68 100/1 Pestalotiopsis australasiae CBS 114141 Pestalotiopsis australasiae CBS 114141 Pestalotiopsis telopeae CBS 114161 84/- Pestalotiopsis telopeae CBS 114161 70/- Pestalotiopsis macadamiae BRIP 63738b 99/1 Pestalotiopsis oryzae CBS 11522 92/ 99/1	P. barachiata P. biciliata P. australasiae P. telopeae
100/1 Pestalotiopsis brachiata LC8188 Pestalotiopsis brachiata LC2988 98/1 Pestalotiopsis biciliata CBS 124463 98/1 Pestalotiopsis biciliata CBS 790.68 100/1 Pestalotiopsis australasiae CBS 114141 Pestalotiopsis australasiae CBS 114141 Pestalotiopsis telopeae CBS 114161 84, Pestalotiopsis telopeae CBS 114161 70, Pestalotiopsis oryzae CBS 113606 99/1 Pestalotiopsis oryzae CBS 111522 99/1 Pestalotiopsis kenyana CBS 442.67	P. barachiata P. biciliata P. australasiae P. telopeae P. macadamiae
100/1 Pestalotiopsis brachiata LC8188 Pestalotiopsis brachiata LC2988 98/1 Pestalotiopsis biciliata CBS 124463 98/1 Pestalotiopsis biciliata CBS 790.68 100/1 Pestalotiopsis australasiae CBS 114141 Pestalotiopsis australasiae CBS 114141 Pestalotiopsis telopeae CBS 114161 84/- Pestalotiopsis telopeae CBS 114161 99/1 Pestalotiopsis oryzae CBS 111522 99/1 Pestalotiopsis kenyana CBS 442.67 86/ Pestalotiopsis kenyana CBS 911.96	P. barachiata P. biciliata P. australasiae P. telopeae P. macadamiae P. oryzae P. kenyana
100/1 Pestalotiopsis brachiata LC8188 Pestalotiopsis biciliata CBS 124463 98/1 Pestalotiopsis biciliata CBS 790.68 100/1 Pestalotiopsis australasiae CBS 114141 Pestalotiopsis australasiae CBS 114126 84/ Pestalotiopsis telopeae CBS 114161 70/- Pestalotiopsis telopeae CBS 113606 61/ Pestalotiopsis oryzae CBS 111522 99/1 Pestalotiopsis kenyana CBS 442.67 86/ Pestalotiopsis kenyana CBS 911.96	P. barachiata P. biciliata P. australasiae P. telopeae P. macadamiae P. oryzae
 Pestalotiopsis brachiata LC8188 Pestalotiopsis brachiata LC2988 Pestalotiopsis biciliata CBS 124463 Pestalotiopsis biciliata CBS 790.68 Pestalotiopsis australasiae CBS 114141 Pestalotiopsis australasiae CBS 114126 Pestalotiopsis telopeae CBS 114161 Pestalotiopsis telopeae CBS 114161 Pestalotiopsis oryzae CBS 111522 Pestalotiopsis kenyana CBS 442.67 Pestalotiopsis kenyana CBS 911.96 Pestalotiopsis trachicarpicola HN56 2.1 	P. barachiata P. biciliata P. australasiae P. telopeae P. macadamiae P. oryzae P. kenyana
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100/1 Pestalotiopsis brachiata LC8188 Pestalotiopsis brachiata LC2988 Pestalotiopsis biciliata CBS 124463 98/7 Pestalotiopsis australasiae CBS 114161 Pestalotiopsis australasiae CBS 114161 Pestalotiopsis telopeae CBS 114161 99/7 Pestalotiopsis telopeae CBS 113606 61/ Pestalotiopsis oryzae CBS 111522 99/7 Pestalotiopsis oryzae CBS 353.69 99/7 Pestalotiopsis kenyana CBS 442.67 99/7 Pestalotiopsis kenyana CBS 911.96 99/7 Pestalotiopsis trachicarpicola HN56 2.1 Pestalotiopsis trachicarpicola IFRDCC 2440 Pestalotiopsis paeoniicola TR41 100/0.9 Pestalotiopsis paeoniicola TR41	P. barachiata P. biciliata P. australasiae P. telopeae P. macadamiae P. oryzae P. kenyana P. kenyana P. trachicarpicola P. paeoniicola
100/1 Pestalotiopsis brachiata LC8188 Pestalotiopsis brachiata LC2988 Pestalotiopsis biciliata CBS 124463 98/ Pestalotiopsis australasiae CBS 114161 Pestalotiopsis telopeae CBS 114161 Pestalotiopsis telopeae CBS 114161 99/ Pestalotiopsis telopeae CBS 113606 61/ Pestalotiopsis oryzae CBS 111522 99/ Pestalotiopsis oryzae CBS 353.69 Pestalotiopsis kenyana CBS 442.67 Pestalotiopsis trachicarpicola HN56 2.1 Pestalotiopsis trachicarpicola HN56 2.1 Pestalotiopsis paeoniicola TR41 100/0-9 Pestalotiopsis paeoniicola TR41 100/0-9 Pestalotiopsis theae CMU-ELA1	P. barachiata P. biciliata P. australasiae P. telopeae P. macadamiae P. oryzae P. kenyana P. kenyana P. trachicarpicola
100/1 Pestalotiopsis brachiata LC8188 Pestalotiopsis brachiata LC2988 Pestalotiopsis biciliata CBS 124463 98/1 Pestalotiopsis australasiae CBS 114161 Pestalotiopsis australasiae CBS 114161 Pestalotiopsis telopeae CBS 113606 61/ Pestalotiopsis oryzae CBS 113606 61/ Pestalotiopsis oryzae CBS 111522 92/ 99/1 Pestalotiopsis oryzae CBS 353.69 Pestalotiopsis kenyana CBS 442.67 Pestalotiopsis trachicarpicola HN56 2.1 Pestalotiopsis trachicarpicola HN56 2.1 Pestalotiopsis trachicarpicola IFRDCC 2440 Pestalotiopsis paeoniicola TR41 Pestalotiopsis paeoniicola TR40 100/ Pestalotiopsis theae CMU-ELA1 Pestalotiopsis theae CPO Pe Pestalotiopsis theae CPO Pe	P. barachiata P. biciliata P. australasiae P. telopeae P. macadamiae P. oryzae P. kenyana P. rhododendri P. trachicarpicola P. paeoniicola
100/1 Pestalotiopsis brachiata LC8188 Pestalotiopsis brachiata LC2988 Pestalotiopsis biciliata CBS 124463 98/ Pestalotiopsis australasiae CBS 114161 Pestalotiopsis telopeae CBS 114161 Pestalotiopsis telopeae CBS 114161 99/ Pestalotiopsis telopeae CBS 113606 61/ Pestalotiopsis oryzae CBS 111522 99/ Pestalotiopsis oryzae CBS 353.69 Pestalotiopsis kenyana CBS 442.67 Pestalotiopsis trachicarpicola HN56 2.1 Pestalotiopsis trachicarpicola HN56 2.1 Pestalotiopsis paeoniicola TR41 100/0-9 Pestalotiopsis paeoniicola TR41 100/0-9 Pestalotiopsis theae CMU-ELA1	P. barachiata P. biciliata P. australasiae P. telopeae P. macadamiae P. oryzae P. kenyana P. kenyana P. trachicarpicola P. paeoniicola

Fig. 1 – Continued.

63/0.9	Neopestalotiopsis sp LC6471
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100/0.9 Neopestalotiopsis sp YN1A5

^{100/0.9} Neopestalotiopsis sp YN1A5	
64/ Neopestalotiopsis asiatica	FLUCC 12-0286 N. asiatica
65/ Neopestalotiopsis umbrinosp	ra MFLUCC 12-0285 N. umbrinospora
Neopestalotiopsis chrysea M	LUCC 12-0261 N. chrysea
	37
Neopestalotiopsis musae	MFLUCC 15-0776 N. musae
- Neopestalotiopsis egyptiaca P	IST1 N. egyptiaca
70/+ Neopestalotiopsis australis Cl	s 114159 N. australis
¹ Neopestalotiopsis vitis JZB34001	N. vitis
91/0.9 Neopestalotiopsis sp FMB 0128	
Neopestalotiopsis sp FMB 0127	
96/0.9 Neopestalotiopsis sp CBS 323	76
96/0.9 Neopestalotiopsis zimbabw	ana CBS 111495 N. zimbabwana
94/0.9 Neopestalotiopsis honolulu	ana CBS 114495
Neopestalotiopsis honoluluar	NI honoluluono
Neopestalotiopsis eucalyptico	
Neopestalotiopsis rosae CBS 12474	
Neopestalotiopsis pernambucana F	
Neopestalotiopsis javaensis CBS	257.31 N. javaensis
Neopestalotiopsis chiangmai	
Neopestalotiopsis sp CBS 164.42	N. Chiangmaichsis
_{03/0} 9 <i>Neopestalotiopsis</i> sp LC6285	
Neopestalotiopsis sp LPS61	
Neopestalotiopsis mesopotamica	CBS 464.69 N. mesopotamica
0/1 Neopestalotiopsis saprophytica CBS	115452
Neopestalotiopsis saprophytica	N sanronhytics
/09 Neopestalotiopsis sp CBS 360.61	
Neopestalotiopsis sp CBS 119.75	
Neopestalotiopsis cubana CBS 6	0.96 N. cubana
Neopestalotiopsis mag	
Neopestalotiopsis pandar	
- Neopestalotiopsis foedans CGMC	
Neopestalotiopsis brasiliensis PA10	N. brasiliensis
86/1 Neopestalotiopsis	
Neopestalotio	sis iraniensis P815 N. iraniensis
64/ Neopestalotiopsis surinamensis	BS 111494
Neopestalotiopsis surinamens	s CBS 450.74 N. surinamensis
Neopestalotiopsis acrostichi MF	LUCC 17-1754 N. acrostich
Neopestalotiopsis protearum CB	114178 N. protearum
Neopestalotiopsis thailandica M	
68/ Neopestalotiopsis coffea-arab	
68/ <u>Neopestalotiopsis coffea-arab</u> Neopestalotiopsis sonneratae M	
	LUCC 17-1744 N. sonneratae
Neopestalotiopsis sonneratae M	LUCC 17-1744 N. sonneratae
Neopestalotiopsis sonneratae M Neopestalotiopsis macadamiae E	FLUCC 17-1744 N. sonneratae RIP 63737c N. macadamiae

Fig. 1 – Continued.

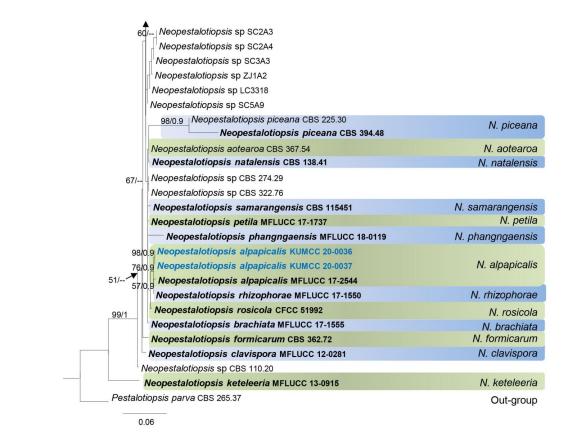


Fig. 2 – Phylogram generated from maximum likelihood analysis based on combined ITS, BT and tef 1– α sequence data. Bootstrap support values for ML greater than 50% and Bayesian posterior probabilities greater than 0.9 are given near nodes respectively. The tree is rooted with *Pestalotiopsis parva* (CBS 265.37). Ex-type strains are in black bold and the newly generated sequences are indicated in blue bold.

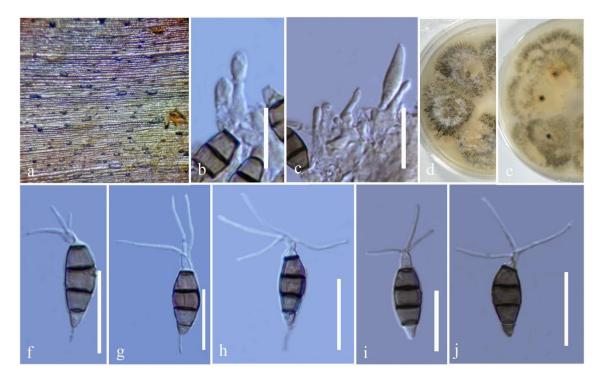


Fig. 3 – *Pestalotiopsis diploclisiae* (MFLU 20–0059). a Conidiomata on substrate. b-c Conidiogenous cells attached to conidia. d Upper surface of culture on PDA. e Lower surface of culture on PDA. f-j Conidia. Scale bars: b-c = $50 \mu m$, f-j = $15 \mu m$.

Material examined – CHINA, Guangdong Province, Shenzhen, Nanshan District, Mountain Yangtai Forest Park, 22°39′21.26″N 113°57′18.53″E, dead leaves of *Butia* sp. (Arecaceae), 5 September 2018, I. C. Senanayake, SI 66, (MFLU 20–0059; living culture KUMCC 20–0035).

Notes – Phylogenetically, our *Pestalotiopsis diploclisiae* collection (KUMCC 20–0035) is closely related to *P. diploclisiae* (CBS 115449) with high bootstrap support. Type strains of *Pestalotiopsis diploclisiae* were collected from fruits of *Diploclisia glaucescens* and *Psychotria tutcheri* in Hong Kong, while our *P. diploclisiae* strain (KUMCC 20–0035) collected from a dead leaf of *Butia* sp. in Shenzhen (China) closer to Hong Kong. The prologue provided by Maharachchikumbura et al. (2014) for *Pestalotiopsis diploclisiae* was based on the morphology derived from cultures. However, our description and illustration are based on the morphology derived from the specimen. Our strain produces smaller $(5-11 \times 2-3 \mu m)$, discrete, lageniform conidiogenous cells and smaller $(17-24 \times 6-7 \mu m)$, fusiform to clavate conidia with conic to acute apical cell and 3–4 apical appendages. Except for the size variation in conidia and conidiogenous cells, our collection of *Pestalotiopsis diploclisiae* (KUMCC 20–0035) is morphologically identical to its holotype. Comparison of the ITS regions DNA sequence of *Pestalotiopsis diploclisiae* (KUMCC 20–0035) with *P. diploclisiae* (CBS 115449) gives 0.94% base pair differences and therefore, our strain assigned as *Pestalotiopsis diploclisiae*.

Pestalotiopsis parva Maharachch., K.D. Hyde & Crous, in Maharachchikumbura, Hyde, Groenewald, Xu & Crous, Stud. Mycol. 79: 175 (2014) Fig. 4

Facesoffungi number: FoF 07749

Saprobic, associated with dead petiole of *Phoenix* sp. Appears as black spots coming out from plant epidermis surface. Sexual morph: Undetermined. Asexual morph: *Conidiomata* pycnidial, globose, immersed in substrate, semi-immersed on PDA, brown, releasing conidia in a black, slimy, globose, glistening mass. *Conidiophores* reduced to conidiogenous cells. *Conidiogenous cells* $7-10 \times 2-3$ ($\overline{x} = 8.9 \times 2.4 \mu m$, n = 20), discrete, subcylindrical to lageniform, hyaline, smooth, thin-walled, annellidic, proliferating once percurrently. *Conidia* $17-21 \times 6-7 \mu m$, ($\overline{x} = 18.9 \times 6.8 \mu m$, n = 20), fusiform to mostly globose, straight, 4-septate; basal cell conic to acute with a truncate base, hyaline or sometimes pale brown, thin and smooth-walled, 2.5-4 µm long ($\overline{x} = 3 \mu m$, n = 20); three median cells ± equal, each 3–5 µm long ($\overline{x} = 4 \mu m$, n = 20), doliiform, pale brown, septa darker than rest of the cell, concolorous, wall rugose; apical cell 3–4.5 µm long ($\overline{x} = 3.5 \mu m$, n = 20), hyaline, subcylindrical to obconic, with 2–3 tubular appendages on apical cell, arising from the apical crest, unbranched, flexuous, 9–18 × 0.6–0.9 µm ($\overline{x} = 13 \times 1 \mu m$, n = 20); basal appendage single, tubular, unbranched, centric, 3–4.5 µm long, ($\overline{x} = 3.6 \mu m$).

Culture characters – Colonies on PDA reaching 2.5 cm diam., within 1 week at 20°C, under dark, circular with several layers, medium dense, aerial mycelium clots concentrated along the colony margin, flat, filiform margin, white from above and reverse; fruiting bodies did not appear on cultures.

Material examined – CHINA, Guangdong Province, Shenzhen, Nanshan, Nanhai Avenue, Shenzhen University, dead petiole of *Phoenix* sp. (Arecaceae), 28 August 2018, I.C. Senanayake, SI 9, (MFLU 20–0060, living culture KUMCC 20–0038).

Notes – One of our *Pestalotiopsis* strain (KUMCC 20–0038) clusters with the type species of *P. parva* (CBS 265.37) with moderate bootstrap support in the phylogenetic analysis. *Pestalotiopsis parva* was introduced based on only two strains as CBS 265.37 and CBS 278.35 (Maharachchikumbura et al. 2014). However, the collected localities of those strains are unknown. Maharachchikumbura et al. (2014) described *Pestalotiopsis parva* based on the morphology derived from cultures. However, we obtained morphological characters of our *Pestalotiopsis* strain directly from the specimen, not from culture. Comparison of the ITS sequence of *Pestalotiopsis parva* KUMCC 20–0038 with *P. parva* CBS 265.37 and CBS 278.35 revealed that the base pair differences between them are less than 1% (0.88% and 0.88% respectively) which propose our strain as an existing species (Jeewon & Hyde 2016).

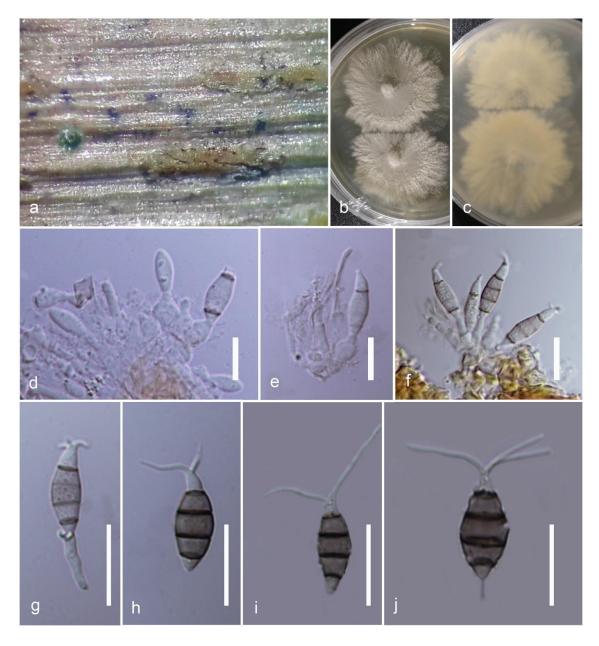


Fig. 4 – *Pestalotiopsis parva* (MFLU 20–0060). a Conidiomata on substrate. b Upper surface of culture on PDA. c Lower surface of culture on PDA. d-f Conidiogenous cells attached to conidia. g-j Conidia. Scale bars: $d-j = 20 \mu m$.

Neopestalotiopsis alpapicalis Vin. Kumar, Gentekaki & K.D. Hyde, in Kumar, Cheewangkoon, Gentekaki, Maharachchikumbura, Brahmange & Hyde, Phytotaxa 393(3): 253 (2019) Fig. 5

Facesoffungi number: FoF 05753

Saprobic or pathogenic, associated with living leaves of Phoenix roebelenii. Sexual morph: Undetermined. Asexual morph: Appears as swollen areas with split barks. Conidiomata 25–80 µm diam., pycnidial, globose, black, immersed in substrate, superficial in PDA, releasing conidia as a black, slimy, globose, mass on culture media. Conidiophores reduced to conidiogenous cells. Conidiogenous cells 4–6 × 3–4 µm ($\bar{x} = 5 \times 4$ µm, n = 20), discrete, annellidic, globose to umbonate, short, hyaline, smooth-walled, simple, wide at the base. Conidia 24–28 × 9–11 µm ($\bar{x} = 26 \times 10$ µm, n = 20), ellipsoid, straight to slightly curved, 4–(6)-septate; basal cell conic to obconic with a truncate base, hyaline, thin-and smooth-walled, 3–4 µm long ($\bar{x} = 3.8$ µm, n = 20); 3–(5) median cells, each 4.5–7 µm long ($\bar{x} = 5.5$ µm, n = 20), 4–7 µm long ($\bar{x} = 5.6$ µm, n = 20), 4–6 µm long ($\bar{x} = 4.6$ µm, n = 20), doliiform, concolorous, pale brown, septa and periclinal walls darker than rest of the cell, wall rugose; apical cell 3–6 µm long ($\bar{x} = 4.6$ µm, n = 20), long, hyaline, conic

to obtuse with truncate base; apical appendages 7–12 μ m long ($\bar{x} = 10 \mu$ m, n = 20), short, 1–4, more tubular, inserted at different loci but in a crest at the apex of the apical cell, unbranched, flexuous; single basal appendage, tubular, unbranched, rarely branched, centric, 4–6 μ m long ($\bar{x} = 4.7 \mu$ m, n = 20).

Culture characteristics – Colonies on PDA reaching 2 cm diam., within 10 days at 18°C, under dark, circular, medium dense, aerial mycelium clots scattered on PDA, flat, filiform margin, white from above and reverse; black, globose, sporulate on cultures after 4 weeks incubate at 20°C in dark.

Material examined – CHINA, Guangdong Province, Shenzhen, Luohu District, Fairy-lake botanical garden, 22°34′43.10″N 114°09′55.98″E, living leaves of *Phoenix roebelenii* O'Brien (Arecaceae), 26 July 2018, I.C. Senanayake, SI 100, (MFLU 20–0061, living culture KUMCC 20–0037); CHINA, Guangdong, Shenzhen, Luohu District, Fairy-lake botanical garden, 22°34′43.10″N 114°09′55.98″E, living leaves of *Musa* sp. (Musaceae), 26 July 2018, I.C. Senanayake, SI 103, (MFLU 20–0058, living culture KUMCC 20–0036).

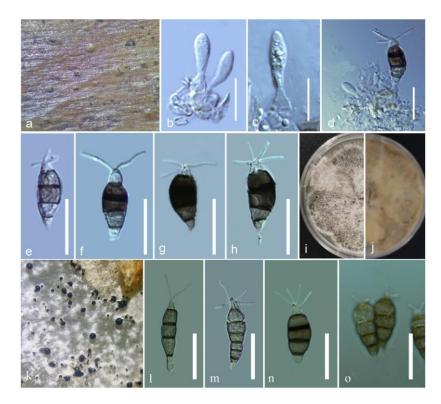


Fig. 5 – *Neopestalotiopsis alpapicalis* (MFLU 20–0058). a Conidiomata on substrate. b-d Conidiogenous cells attached to conidia. e-h Conidia from fruit bodies in substrate. i upper surface of culture on PDA. j Lower surface of culture on PDA. k Conidioma on PDA. l-o Conidia derived from culture (l; unusual basal cell, m; conidia with six cells, o; wall ornamentations). Scale bars: b-h, $1-o = 25 \mu m$.

Notes – In the phylogenetic analysis (Fig. 2), our *Neopestalotiopsis* strain forms a distinct subclade basal to *N. alpapicalis* (MFLUCC 17–2544), and *N. rhizophorae* (MFLUCC 17–1550) with moderate bootstrap support. There are 1.27%, 1.07% and 1.02% base pair differences of the ITS (566bp), BT (469bp) and tef 1– α (490bp) sequences of our *Neopestalotiopsis* strains with *N. alpapicalis* (MFLUCC 17–2544), and these values are 1.59%, 0.85%, and 1.02% for *N. rhizophorae* (MFLUCC 17–1550). However, multi-locus gene regions use in this study may not enough to separate *Neopestalotiopsis* species well and there are no more gene regions available in GenBank. Therefore, determination of taxonomy of *Neopestalotiopsis* strains is challenging.

However, morphologically our *Neopestalotiopsis* collection is similar to *N. alpapicalis* more than *N. rhizophorae* in having highly pigmented, conidia with shorter, tubular, apical appendages

which are attached to the tip of apical cell. It is difficult to clarify and compare the morphological characters of fungi grown in different media and different growth conditions. Our *Neopestalotiopsis* strain is a saprobe collected from a terrestrial, monocotyledon plant in China, while *Neopestalotiopsis alpapicalis* collected with leaf spots of mangrove plants in Thailand. Therefore, based on available molecular data, morphology and ecological data we named this species as *N. alpapicalis*.

Discussion

Pestalotiopsis parva and Neopestalotiopsis alpapicalis collected from Phoenix are mostly dominant in northern and central Africa, Southeastern Europe, Southern Asia and east to Southern China (Chase et al. 2000). Fruits of some Phoenix species are edible and used as raw materials in the sugar industry. Phoenix roebelenii is widely grown for its ornamental value and its fruit used as food for livestock and poultry (Riffle & Craft 2003). Phoenix species have some resistant to pests and tolerance to soil variation and drought. Therefore, Phoenix species used for reforestation in swamps, deserts and mangrove coasts. Pestalotiopsis diploclisiae collected from dead leaves of Butia, which is an ornamental genus (Faria et al. 2011) and fruits of Butia species are used as foods, such as juices, liquor, marmalades and ice cream, while seeds are used to extract oil. Musa species are also important as a food source.

Even though fungal diversity associated with these plants have been studied, the micro-fungi inhabiting them are poorly known in Guangdong Province (Chobba et al. 2013, Shen et al. 2014, Wei et al. 2007, Zakaria & Aziz 2018). In this study, we collected several pestaloid taxa on Butia, Phoenix and Musa species and identified them through morpho-phylogenetic studies. Here, we provide taxonomic details for them.

Colonies of *Pestalotiopsis parva* grew faster on PDA than *Neopestalotiopsis alpapicalis* and *Pestalotiopsis diploclisiae* and did not sporulate in culture. *Pestalotiopsis diploclisiae* and *Neopestalotiopsis alpapicalis* sporulate in culture after four weeks of incubation at 20°C in the dark. Both *Pestalotiopsis diploclisiae* and *Neopestalotiopsis alpapicalis* initially formed copious aerial mycelia clots and those clots disappear with the formation of conidiomata on the PDA.

A checklist of *Neopestalotiopsis* and *Pestalotiopsis* fungi in China is given in Table 2. This includes seven species of *Neopestalotiopsis* and 69 species of *Pestalotiopsis*. Pestaloid fungi are common phytopathogens that cause a variety of diseases, including canker lesions, shoot dieback, leaf spots, needle blight, tip blight, grey blight, scabby canker, severe chlorosis, fruit rots and various post-harvest diseases (Crous et al. 2011, Zhang et al. 2013, Maharachchikumbura et al. 2014). *Pestalotiopsis clavispora* and *P. anacardiacearum* have been reported to cause grey leaf spots and associated with the mango tip borer by *Penicillaria jocosatrix*, respectively in China. *Pestalotiopsis camelliae* was associated with grey leaf blight of *Camellia japonica* and *Pestalotiopsis ericacearum* with leaf spots of *Rhododendron delavayi*.

Taxon	Host/substrate	Province	References
Neopestalotiopsis asiatica	leaves of tree	Hunan	Maharachchikumbura et al. (2014)
N. chrysea	dead leaves	Guangxi	Maharachchikumbura et al. (2014)
N. clavispora	dead leaves of <i>Magnolia</i> sp.	Guangxi	Maharachchikumbura et al. (2014)
N. ellipsospora	dead plant material	Yunnan	Maharachchikumbura et al. (2014)
N. foedans	mangrove plant leaves	Hainan	Maharachchikumbura et al. (2014)
N. saprophytica	leaves of Magnolia	Yunnan	Maharachchikumbura et al. (2014)
N. umbrinospora	dead leaves	Guangxi	Maharachchikumbura et al. (2014)
Pestalotiopsis adusta	leaves of <i>Podocarpus</i> macrophyllus	Guangxi	Wei et al. (2007)
P. affinis	unknown	Yunnan	Chen et al. (2002)

Table 2 Checklist of Neopestalotiopsis and Pestalotiopsis fungi in China

Table 2 Continued.

Taxon	Host/substrate	Province	References
P. aggestorum	leaves of Camellia	Yunnan	Liu et al. (2017)
	sinensis		
P. alpiniae	leaves of Alpinia	Guangxi	Chen et al. (2002)
	galanga		
P. anacardiacearum	living leaf of Mangifera	Yunnan	Maharachchikumbura et al. (2013)
	indica	~ .	
P. antiaris	leaves of Antiaris	Guangxi	Chen et al. (2002)
During	toxicaria	D ::	U
P. apiculata	trunk and leaves of <i>Cunninghamia</i>	Fujian	Huang (1983)
	lanceolata		
P. brideliae	living leaves of <i>Bridelia</i>	China	Chen & Wei (1997)
1. Dridelide	morcica	China	
P. briosiana	twigs of <i>Camellia</i>	Yunnan	Wei et al. (2007)
	sasanqua		
P. camelliae	leaves of <i>Camellia</i>	Yunnan	Zhang et al. (2012a)
	japonica		5
P. canarii	living leaves of	Guangxi	Chen et al. (2003)
	Canarium album	C	
P. chinensis	leaves of Taxus	Yunnan	Maharachchikumbura et al. (2012b)
P. clavata	leaf of Buxus	Yunnan	Maharachchikumbura et al. (2014)
P. coffeae-arabicae	living leaves of Coffea	Hainan	Song et al. (2013)
	arabica		
P. crassiuscula	leaves of Podocarpus	Zhejiang	Wei et al. (2007)
5 HU I	macrophyllus		
P. dilleniae	leaves of Dillenia	Guangxi	Chen et al. (2002)
	turbinata	T::	$L_{1} = 1$ (2017)
P. dilucida	leaves of Camellia sinensis	Jiangxi	Liu et al. (2017)
P. diospyri			Wei et al. (2007)
r. alospyri	leaves of <i>Podocarpus</i> macrophyllus		wei et al. (2007)
P. disseminata	leaves of <i>Podocarpus</i>		Wei et al. (2007)
1. aussemmana	macrophyllus		() of of all (2007)
P. diversiseta	leaves of <i>Rhododendron</i>		Maharachchikumbura et al. (2014)
P. dracaenae	leaves of Dracaena		Ariyawansa et al. (2015)
	fragrans		
P. ericacearum	leaves of Rhododendron		Zhang et al. (2013)
	delaveyi		
P. foedans	twigs of Podocarpus	Guangdong	Wei et al. (2007)
	massoniana		
P. gaultheriae	on Gaultheria	Yunnan	Maharachchikumbura et al. (2014)
P. hainanensis	stem of <i>Podocarpus</i>	Hainan	Liu et al. (2007)
	macrophyllus	71	
P. heterocornis	fruit and bark of	Zhejiang	Wei et al. (2007)
	Podocarpus		
P. inflexa	<i>macrophyllus</i> leaf of tree	Hunan	Maharachchikumbura et al. (2014)
P. intermedia	dead leaf of tree	Hubei	Maharachchikumbura et al. (2014)
P. jiangxiensis	on <i>Camellia</i>	Jiangxi	Liu et al. (2017)
P. jinchanghensis	on leaves of <i>Camellia</i>	Yunnan	Liu et al. (2017)
j	sinensis		
P. keteleeriae	on leaves of <i>Keteleeria</i>	Guizhou	Song et al. (2014)
	pubescens		
P. kunmingensis	leaves of Podocarpus	Yunnan	Wei & Xu (2004)
	macrophyllus		

Taxon	Host/substrate	Province	References
P. kwangsiensis	leaves of	Guangxi	Chen et al. (2002)
Ū	Sinopimelodendron	e	
	kuwangsiensis		
P. lawsoniae	leaves of <i>Pinus</i>	Guangxi	Wei et al. (2007)
1.1400501140	massoniana; twigs of	Guangdong	((ef et al. (2007))
	0	Guanguong	
	Podocarpus		
	massoniana		G (2012)
P. licualicola	living leaves of Licuala	Hainan	Geng et al. (2013)
	grandis		
P. lijiangensis	unknown	Yunnan	Zhou et al. (2008)
P. linearis	leaves of	Yunnan	Maharachchikumbura et al. (2014)
	Trachelospermum		
P. longiappendiculata	Camellia sinensis	Fujian	Liu et al. (2017)
P. lushanensis	<i>Camellia</i> sp	Jiangxi	Liu et al. (2017)
P. macadamii	living leaves of	Guangdong	Akinsanmi et al. (2017)
	Macadamia integrifolia	Cambrong	
P. menezesiana	leaves of <i>Podocarpus</i>	Guangxi	Wei et al. (2007)
1. menezestana	macrophyllus	Guangxi	Wei et al. (2007)
D :	1 5	с ·	$W_{1} = (1)(2007)$
P. microspora	twigs of <i>Podocarpus</i>	Guangxi	Wei et al. (2007)
	macrophyllus		
P. nattrassioides	unknown	Yunnan	Zhao & Zhao (2012)
P. neglecta	twigs of <i>Podocarpus</i>	Guangxi	Wei et al. (2007)
	nagi		
P. nelumbonis	leaves of Nelumbo	Guangxi	Chen et al. (2002)
	nucifera	U	
P. olivacea	leaves of Podocarpus	Yunnan	Wei et al. (2007)
	nagi		(101 00 dat (2007))
P. oxyanthi	leaves of <i>Podocarpus</i>	Zhejiang	Wei et al. (2007)
1 . <i>Oxyanını</i>	-	Zhejiang	Wei et al. (2007)
	macrophyllus	V	C_1 (1)(2002)
P. pachirae	living leaves of	Yunnan	Chen et al. (2003)
	Pachira macrocarpa		
P. phaii	living leaves of <i>Phaius</i>	Yunnan	Chen et al. (2003)
	tankervilleae		
P. photiniae	twigs of <i>Camellia</i>	Guangxi	Wei et al. (2007)
	japonica, Camellia	Zhejiang,	
	sasanqua, Leaves of	Yunnan	
	Podocarpus		
	massoniana		
P. photiniicola	leaves of <i>Photinia</i>	Guizhou	Chen et al. (2017)
1. pholinicold	serrulata	Guizilou	
D mlauna aminita		Vummen	$7h_{22} \approx 7h_{22} (2012)$
P. pleurocrinita	unknown	Yunnan	Zhao & Zhao (2012)
P. rhododendri	dead parts of leaves of	Yunnan	Maharachchikumbura et al. (2014)
	Rhododendron		
	sinogrande		
P. rhodomyrtus	living leaves of	Guangxi	Zhang et al. (2013)
	Rhodomyrtus		
	tomentosa		
P. rosea	isolated from leaves of	Yunnan	Maharachchikumbura et al. (2014)
	Pinus		
P. schimae	leaves of <i>Schima</i>	Guangxi	Chen et al. (2002)
I. SUITHING	superba	JuangAl	Chen et al. (2002)
D auhaharaa	1	Cuarani	A minimum $a = a = 1$ (2015)
P. subshorea	leaves of <i>Michelia</i>	Guangxi	Ariyawansa et al. (2015)
b	hedyosperma	** '	
P. synsepali	leaves of Synsepalum	Hainan	Chen et al. (2002)
	dulcificum		

Table 2 Continued.

Taxon	Host/substrate	Province	References
P. theae	leaves of Camellia	Zhejiang,	Wei et al. (2007)
	sinensis, Camellia	Guangxi	
	reticulata and Camellia	Yunnan	
	nitidissima, twigs of	Zhejiang	
	Podocarpus macrophyllus		
P. trachycarpicola	leaves of Trachycarpus fortune, Podocarpus macrophyllus	Yunnan	Zhang et al. (2012b)
P. unicolor	leaf of Rhododendron	Hunan	Maharachchikumbura et al. (2012b)
P. verruculosa	leaf of Rhododendron	Yunnan	Maharachchikumbura et al. (2012b)
P. virgatula	leaves of Podocarpus	Zhejiang	Wei et al. (2007)
	macrophyllus, twigs of	Guangdong	
	Podocarpus		
	massoniana		
P. vismiae	unknown	Guangxi, Yunnan	Zhang et al. (2003)
P. yanglingensis	on Camellia sinensis	Jiangxi	Liu et al. (2017)
P. yunnanensis	twigs of <i>Podocarpus</i> macrophyllus and grown on leaf segments of <i>Dianthus</i>	Yunnan	Wei et al. (2013)
_	caryophyllus		
P. zonata	fruits of <i>Podocarpus</i> macrophyllus	Zhejiang	Wei et al. (2007)

Table 2 Continued.

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References

- Akinsanmi OA, Nisa S, Jeff-Ego OS, Shivas RG, Drenth A. 2017 Dry flower disease of Macadamia in Australia caused by *Neopestalotiopsis macadamiae* sp. nov. and *Pestalotiopsis* macadamiae sp. nov. Plant Disease 101(1), 45–53.
- Aly AH, Debbab A, Kjer J, Proksch P. 2010 Fungal endophytes from higher plants: a prolific source of phytochemicals and other bioactive natural products. Fungal Diversity 41, 1–16.
- Ariyawansa HA, Hyde KD. 2018 Additions to *Pestalotiopsis* in Taiwan. Mycosphere 9, 999–1013.
- Ariyawansa HA, Hyde KD, Jayasiri SC, Buyck B et al. 2015 Fungal diversity notes 111–252 taxonomic and phylogenetic contributions to fungal taxa. Fungal Diversity 75, 27–274.
- Chase MW, Soltis DE, Soltis PS, Rudall PJ et al. 2000 Higher-level systematics of the monocotyledons: An assessment of current knowledge and a new classification. Monocots: Systematics and Evolution 3–16.
- Chen YX, Wei G, Chen WP, Wang ZW, Lu ZH. 2003 Three new species of *Pestalotiopsis* in China. Journal of Guangxi Agricultural and Biological Science 22, 1–4.
- Chen YX, Wei G, Chen WP. 2002 New species of Pestalotiopsis. Mycosystema. 21, 316–323.

- Chen YX, Wei G. 1997 Continuous notes on congeners of *Pestalotiopsis* in China. Journal of Guangxi Agricultural University 16: 1–9.
- Chen YX, Zeng L, Shu N, Jiang M et al. 2018 *Pestalotiopsis* like species causing gray blight disease on *Camellia sinensis* in China. Plant Disease 102, 98–106.
- Chen YY, Maharachchikumbura SSN, Liu JK, Hyde KD et al. 2017 Fungi from Asian Karst formations I. *Pestalotiopsis photinicola* sp. nov., causing leaf spots of *Photinia serrulata*. Mycosphere 8(1), 103–110.
- Chobba IB, Elleuch A, Ayadi I, Khannous L et al. 2013 Fungal diversity in adult date palm (*Phoenix dactylifera* L.) revealed by culture-dependent and culture-independent approaches. Journal of Zhejiang University Science B 14, 1084–1099.
- Crous PW, Summerell BA, Swart L. 2011 Fungal pathogens of Proteaceae. Persoonia 27, 20-45.
- Debbab A, Aly AH, Proksch P. 2013 Mangrove derived fungal endophytes a chemical and biological perception. Fungal Diversity 61, 1–27.
- Ellis MB, Ellis JP. 1997 Microfungi on land plants. an identification handbook. Richmond Publishing, England.
- Faria JP, Siqueira E, Vieira R, Agostini-Costa TS. 2011 Fruits of *Butia capitata* (Mart.) Becc as good sources of β -carotene and provitamina. Revista Brasileira de Fruticultura 33, 612–617.
- Geng K, Zhang B, Song Y, Hyde KD et al. 2013 A new species of *Pestalotiopsis* from leaf spots of *Licuala grandis* from Hainan, China. Phytotaxa 88(3): 49–54.
- Glass NL, Donaldson GC. 1995 Development of primer sets designed for use with the PCR to amplify conserved genes from filamentous ascomycetes. Applied and Environmental Microbiology 61, 1323–1330.
- Guba EF. 1961 Monograph of *Pestalotia* and *Monochaetia*. Harvard University Press, Cambridge.
- Hall TA. 1999 BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. Nucleic Acids Symposium Series 41, 95–98.
- Hu HL, Jeewon R, Zhou DQ, Zhou TX, Hyde KD. 2007 Phylogenetic diversity of endophytic *Pestalotiopsis* species in *Pinus armandii* and *Ribes* spp.: evidence from rDNA and β-tubulin gene phylogenies. Fungal Diversity 24, 1–22.
- Huang TZ. 1983 A preliminary report on dieback (shoot) disease in Chinese fir. Journal of North-Eastern Forestry Institute China 11(3), 45–50.
- Huelsenbeck JP, Ronquist F, Hall B. 2003 MrBayes: a program for the Bayesian inference of phylogeny. Version 3.0 b4.
- Hyde KD, Norphanphoun C, Maharachchikumbura SSN, Bhat DJ et al. 2020 Refined Families of Sordariomycetes. Mycosphere 11(1): 305–1059.
- Jeewon R, Hyde KD. 2016 Establishing species boundaries and new taxa among fungi: recommendations to resolve taxonomic ambiguities. Mycosphere 7, 1669–1677.
- Katoh K, Rozewicki J, Yamada KD. 2017 MAFFT online service: multiple sequence alignment, interactive sequence choice and visualization. Briefings in bioinformatics. Doi 10.1093/bib/bbx108
- Liu AR, Chen SC, Wu SY, Xu T et al. 2010 Cultural studies coupled with DNA based sequence analyses and its implication on pigmentation as a phylogenetic marker in *Pestalotiopsis* taxonomy. Molecular Phylogenetics and Evolution 57, 528–535.
- Liu AR, Xu T, Guo LD. 2007 Molecular and morphological description of *Pestalotiopsis hainanensis* sp. nov., a new endophyte from a tropical region of China. Fungal Diversity 24, 23–36.
- Liu F, Hou LW, Raza M, Cai L. 2017 *Pestalotiopsis* and allied genera from *Camellia*, with description of 11 new species from China. Scientific Reports 7(866), 1–19.
- Maharachchikumbura SSN, Guo LD, Cai L, Chukeatirote E et al. 2012a A multi-locus backbone tree for *Pestalotiopsis*, with a polyphasic characterization of 14 new species. Fungal Diversity 56, 95–129.

- Maharachchikumbura SSN, Guo LD, Lei C, Chukeatirote E et al. 2012b A multi-locus backbone tree for *Pestalotiopsis*, with a polyphasic characterization of 14 new species. Fungal Diversity 56(1), 95–129.
- Maharachchikumbura SSN, Guo LD, Liu ZY, Hyde KD. 2016 *Pseudopestalotiopsis ignota* and *Ps. camelliae* spp. nov. associated with grey blight disease of tea in China. Mycological Progress 15, 22.
- Maharachchikumbura SSN, Hyde KD, Groenewald JZ, Xu J, Crous PW. 2014 *Pestalotiopsis* revisited. Studies Mycology 79, 121–186.
- Maharachchikumbura SSN, Zhang YM, Hyde KD. 2013 *Pestalotiopsis anacardiacearum*. sp. nov. has an intricate relationship with the mango tip borer. Phytotaxa 99, 49–57.
- Monden Y, Yamamoto S, Yamakawa R. 2013 First case of fungal keratitis caused by *Pestalotiopsis clavispora*. Clinical Ophthalmology 7, 2261–2264.
- Norphanphoun C, Jayawardena RS, Chen Y, Wen TC. 2019 Morphological and phylogenetic characterization of novel pestalotioid species associated with mangroves in Thailand. Mycosphere 10, 531–578.
- Nozawa S, Yamaguchi K, Van Hop D, Phay N et al. 2017 Identification of two new species and asexual morph from the genus *Pseudopestalotiopsis*. Mycoscience 58, 328–337.
- Nylander JAA. 2004 MrModeltest 2.0. Program distributed by the author. Evolutionary Biology Centre, Uppsala University.
- Rambaut A, Drummond A. 2008 FigTree: Tree figure drawing tool, version 1.2. 2. Institute of Evolutionary Biology, University of Edinburgh.
- Rambaut A, Drummond AJ. 2007 Tracer v1, 4. Available from: http://beast. bio. ed. ac. uk/Tracer (Accessed on November 1, 2019).
- Rehner S. 2001 Primers for elongation factor $1-\alpha$ (EF1- α). Available from:
- http. ocid. nacas. org/research/deephyphae/EF1primer. (Accessed on January 1, 2020).
- Riffle RL, Craft P. 2003 An Encyclopedia of Cultivated Palms. Portland: Timber Press.
- Russell JR, Huang J, Anand P, Kucera K et al. 2011 Biodegradation of Polyester Polyurethane by Endophytic Fungi. Applied and Environmental Microbiology 77(17), 6076–6084.
- Senanayake IC, Jeewon R, Chomnunti P, Wanasinghe DN et al. 2018 Taxonomic circumscription of Diaporthales based on multigene phylogeny and morphology. Fungal Diversity 93, 241– 443.
- Shen HF, Zhang JX, Lin BR, Pu XM. 2014 First Report of *Pestalotiopsis microspora* Causing Leaf Spot of Oil Palm (*Elaeis guineensis*) in China. Plant Disease 98, 1429.
- Silvestro D, Michalak I. 2012 raxmlGUI: a graphical front-end for RAxML. Organisms Diversity and Evolution 12, 335–337.
- Song Y, Geng K, Zhang B, Hyde KD et al. 2013 Two new species of *Pestalotiopsis* from Southern China. Phytotaxa 126(1), 22–30.
- Song Y, Maharachchikumbura SSN, Jiang YL, Hyde KD, Wang Y. 2014 Pestalotiopsis keteleeria sp. nov., isolated from Keteleeria pubescens in China. Chiang Mai Journal of Science 41(4), 885–893.
- Stamatakis A, Alachiotis N. 2010 Time and memory efficient likelihood-based tree searches on phylogenomic alignments with missing data. Bioinformatics 26, 1132–1139.
- Steyaert RL. 1949 Contributions al'etude monographique de Pestalotia de Not. et Monochaetia Sacc. (Truncatella gen. nov. et Pestalotiopsis gen. nov.). Bulletin Jardin Botanique Etat Bruxelles 19, 285–354.
- Strobel G, Yang XS, Sears J, Kramer R et al. 1996 Taxol from *Pestalotiopsis microspora*, an endophytic fungus of *Taxus wallachiana*. Microbiology 142, 435–440.
- Tejesvi MV, Nalini MS, Mahesh B, Prakash HS et al. 2007 New hopes from endophytic fungal secondary metabolites. Boletín de la Sociedad Química de México 1, 19–26.
- Tibpromma S, Hyde KD, McKenzie EHC, Bhat DJ et al. 2018 Fungal diversity notes 840–928: micro-fungi associated with *Pandanaceae*. Fungal Diversity 92, 1–160.

- Tsai I, Maharachchikumbura SSN, Hyde KD, Ariyawansa HA. 2018 Molecular phylogeny, morphology and pathogenicity of *Pseudopestalotiopsis* species of *Ixora* in Taiwan. Mycological Progress 17, 941–952.
- Watanabe K, Nozawa S, Hsiang T, Callan B. 2018 The cup fungus *Pestalopezia brunneopruinosa* is *Pestalotiopsis gibbosa* and belongs to Sordariomycetes. PloS one 13, 6.
- Wei JG, Phan CK, Wang L, Xu T et al. 2013 Pestalotiopsis yunnanensis sp. nov., an endophyte from Podocarpus macrophyllus (Podocarpaceae) based on morphology and ITS sequence data. Mycological Progress 12, 563–568.
- Wei JG, Xu T, Guo LD, Liu AR et al. 2007 Endophytic *Pestalotiopsis* species associated with plants of Podocarpaceae, Theaceae and Taxaceae in southern China. Fungal Diversity 24, 55–74.
- Wei JG, Xu T. 2004 *Pestalotiopsis kunmingensis* sp. nov., an endophyte from *Podocarpus macrophyllus*. Fungal Diversity 15, 247–254.
- White TJ, Bruns T, Lee SJWT, Taylor J. 1990 Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. PCR protocols: a guide to methods and applications 18, 315–322.
- Wijayawardene NN, Hyde KD, Lumbsch HT, Liu JK et al. 2018 Outline of Ascomycota 2017. Fungal Diversity 88, 167–263.
- Xu J, Ebada SS, Proksch P 2010 *Pestalotiopsis* a highly creative genus: chemistry and bioactivity of secondary metabolites. Fungal Diversity 44, 15–31.
- Xu J, Yang X, Lin Q. 2014 Chemistry and biology of Pestalotiopsis-derived natural products. Fungal Diversity 66, 37–68.
- Zakaria L, Aziz WNW. 2018 Molecular Identification of Endophytic Fungi from Banana Leaves (*Musa* spp.). Trop Life Sci Res 29, 201–211.
- Zhang J, Xu T, Ge Q. 2003 Notes on *Pestalotiopsis* from southern China. Mycotaxon 85, 91–99.
- Zhang YM, Maharachchikumbura SSN, McKenzie EHC, Hyde KD. 2012a *Pestalotiopsis* camelliae sp. nov. associated with grey blight of *Camellia japonica* in China. Mycoscience 64, 335–344.
- Zhang YM, Maharachchikumbura SSN, Mckenzie EHC, Hyde KD. 2012b A novel species of *Pestalotiopsis* causing leaf spots of *Trachycarpus Fortunei*. Cryptogamie, Mycologie 33(3), 311–318.
- Zhang YM, Maharachchikumbura SSN, Tian Q, Hyde KD. 2013 *Pestalotiopsis* species on ornamental plants in Yunnan Province, China. Sydowia 65(1), 113–128.
- Zhao GC, Zhao RL. 2012 The higher microfungi from forests of Yunnan Province. book, 1–572.
- Zhaxybayeva O, Gogarten JP. 2002 Bootstrap, Bayesian probability and maximum likelihood mapping: exploring new tools for comparative genome analyses. BMC genomics 3, 4.
- Zhou YK, Li FP, Hou CL. 2018 *Pestalotiopsis lijiangensis* sp. nov., a new endophytic fungus from Yunnan, China. Mycotaxon 133(3), 513–522.