Received: 28 August 2023 | Revised: 30 October 2023 | Accepted: 13 November 2023

DOI: 10.1111/efp.12839

Forest Pathology WILEY

ORIGINAL ARTICLE

First report of phyllode rust on *Acacia crassicarpa* outside its native range

Michael J. Wingfield[1] | Seonju Marincowitz[2] | Irene Barnes[2] | Marthin Tarigan[1,3] | Myriam Solís[2] | Alvaro Durán[3] | Nam Q. Pham[1]

[1]Department of Plant and Soil Sciences, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, South Africa

[2]Department of Biochemistry, Genetics and Microbiology, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, South Africa

[3]Plant Health Program, Research and Development, Asia Pacific Resources International Holdings Ltd., Pangkalan Kerinci, Indonesia

Correspondence
Nam Q. Pham, Department of Plant and Soil Sciences, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, South Africa.
Email: nam.pham@fabi.up.ac.za

Abstract

Acacia crassicarpa is a tropical tree species native to Australia, West Papua and Papua New Guinea, which has been widely used to establish plantations in the lowland humid tropics of Sumatra and Kalimantan. These trees, able to grow on sites having relatively poor nutrition, have been relatively free of serious disease problems. A rust disease infecting the phyllodes of *A. crassicarpa* has recently been encountered in plantations in various areas of Indonesia and Malaysia where they are not native. In this study, the rust was collected and identified as *Endoraecium violae-faustiae* using DNA sequence analysis based on the internal transcribed spacer (ITS) region of the rDNA. This is the first record of the rust outside its native range. Damage at present appears to be relatively mild but the pathogen could become important in the future.

KEYWORDS
acacia, fungal pathogen, introduced pathogen, plantation forestry, rust disease, Southeast Asia

1 | INTRODUCTION

Various Australian *Acacia* spp. have been utilized to establish plantations in countries of the tropics and Southern Hemisphere. Of these, *Acacia mearnsii* has been planted in temperate areas such as in parts of South Africa and Brazil, where it tolerates relatively low winter temperatures (Moreno Chan et al., 2015; Moreno Chan & Isik, 2021; Richardson et al., 2015). Two other species, *Acacia mangium* and *A. crassicarpa*, have been planted over much greater areas with tropical climates and particularly in the humid tropics of Southeast Asia (Griffin et al., 2011; Harwood et al., 2015; Midgley & Turnbull, 2003).

In all areas of the world where tree species are planted outside their native range, they have increasingly been challenged by diseases (Burgess & Wingfield, 2016; Wingfield et al., 2011, 2015). These include those that are native to the areas where the trees have been established and others that have been gradually introduced over time (Burgess & Wingfield, 2016; Wingfield et al., 2015). This is equally true for *Acacia* spp. as it is for other trees widely planted in the tropics and Southern Hemisphere such as *Eucalyptus*, *Pinus* and *Populus* (Burgess & Wingfield, 2016; Hurley et al., 2023; Roux & Wingfield, 2009; Wingfield et al., 2011). Broadly, the situation in all cases is that diseases as well as insect pests are some of the most economically important factors threatening the long-term sustainability of plantation forestry utilizing non-native tree species internationally (Healy et al., 2022; Payn et al., 2015; Wingfield et al., 2015).

Rust diseases are amongst those that have caused serious damage to non-native trees such as *Eucalyptus* and *Acacia* grown over relatively short rotations. The best known of these is myrtle rust caused by *Austropuccinia psidii* (Sphaerophragmiaceae, Pucciniales),

which has a wide host range in the Myrtaceae and can seriously damage plantation-grown *Eucalyptus* trees (Coutinho et al., 1998; Glen et al., 2007). In the case of *Acacia* spp., phyllode rust caused by *Endoraecium digitatum* (Endoraeciaceae, Pucciniales) is a well-known disease of *A. mangium* in Southeast Asian countries including Indonesia, Malaysia, and Vietnam (Berndt, 2011; Doungsa-ard et al., 2018; Mohd Farid et al., 2023; Old et al., 2000). In recent years, rust caused by *Uromycladium acaciae* (Raveneliaceae, Pucciniales) has emerged as a threat to *A. mearnsii* in South African plantations (Fraser et al., 2021; McTaggart, Doungsa-ard, Wingfield, & Roux, 2015; Morris et al., 1988) and most recently also in Ethiopia (Pham et al., 2023). In all cases, these *Acacia* rust pathogens, *Endoraecium* and *Uromycladium*, are known on their native hosts in Australia (Doungsa-ard et al., 2018).

Acacia crassicarpa, native to Australia, West Papua and Papua New Guinea, is amongst the most widely established plantation tree species in Southeast Asia, where it is well suited to grow on nutrient-poor sites (Griffin et al., 2011; Harwood et al., 2015). Relatively little work has been conducted on the pests and pathogens of these trees outside their native range, despite their importance to the commercial forestry sector. One of the first serious diseases to appear on *A. crassicarpa* was caused by the leaf and shoot pathogen *Pleopassalora perplexa* (syn. *Passalora perplexa*) (Beilharz et al., 2004). These trees can also be infected by *Ceratocystis manginecans* that has devastated plantations of *A. mangium* in Southeast Asia (Hurley et al., 2023; Tarigan et al., 2011, b; Wingfield et al., 2023), although they have substantially lower levels of susceptibility to that pathogen (Barnes et al., 2023; Tarigan, Roux, et al., 2011). Most recently, it has been recognized that these trees, particularly where they are under stress, can be severely infested and damaged by the wood-boring ambrosia beetle *Euwallacea perbrevis* (Lynn et al., 2020, 2021).

Lesions reminiscent of rust infections have been observed on the phyllodes (modified petioles) of *A. crassicarpa* trees in Indonesia (Riau, Kalimantan) and Malaysia since 2019. The aim of this study was to identify the pathogen using DNA sequence data and morphological characteristics.

2 | MATERIALS AND METHODS

2.1 | Sample collections

Infected *A. crassicarpa* leaves showing symptoms of rust (Figure 1) were collected from Kalimantan and Riau in Indonesia, and Sabah in Malaysia (Table 1), and stored in paper bags. These samples were kept in a fridge before they were brought into the laboratory for investigation or dried at room temperature for storage. Additionally, sori on the leaves were either scraped off or added directly to cryovials containing RNAlater (Thermo Fisher Scientific, Waltham, MA, USA), or pieces of leaf material with the sori were placed in these vials for downstream analyses. The specimens were deposited in the H.G.W.J. Schweickerdt Herbarium (PRU), University of Pretoria, South Africa (Table 1).

2.2 | DNA sequencing, PCR amplification and sequencing

DNA was extracted from spore masses or infected plant material using DNeasy® Plant Pro Kit (QIAGEN, Hilden, Germany) following the manufacturer's protocols. The internal transcribed spacer regions 1 and 2 (ITS), including the 5.8S rRNA region, were amplified using primers ITS5u and ITS4rust (Beenken et al., 2012; Pfunder et al., 2001). DNA was amplified by PCR with FastStart™ Taq DNA Polymerase (Sigma-Aldrich, St. Louis, MO, USA). The thermal cycling included an initial denaturation at 95°C for 5 min followed by 10 primary amplification cycles of 30 s at 95°C, 30 s at 53°C, and 60 s at 72°C, then 30 additional cycles of the same reaction sequence, with a 5 s increase in the annealing step per cycle, and the reactions were completed with a final extension at 72°C for 10 min. Amplified fragments were purified using ExoSAP-IT™ PCR Product Cleanup Reagent (Thermo Fisher Scientific, Waltham, MA, USA). Amplicons were sequenced in both directions using the BigDye Terminator v 3.1 Cycle Sequencing kit (Thermo Fisher Scientific, Waltham, MA,

FIGURE 1 *Acacia crassicarpa* infected with *Endoraecium violae-faustiae* in Riau, Indonesia, resulted in heavy defoliation of lower branches seen below these trees (a), and a phyllode with sori (scale bar: 1 mm) (b).

TABLE 1 Collection details for the *Endoraecium* specimens and GenBank accessions included in the phylogenetic analysis.

Species	Specimen	Host	Country	ITS accession number	Reference
E. auriculiformis	BRIP 56548[a]	*A. auriculiformis*	Australia	KJ862355	McTaggart, Doungsa-ard, Geering, et al. (2015)
	BRIP 56549	*A. auriculiformis*	Australia	KJ862356	McTaggart, Doungsa-ard, Geering, et al. (2015)
E. carnegiei	BRIP 57926	*A. dealbata*	Australia	KJ862357	McTaggart, Doungsa-ard, Geering, et al. (2015)
E. disparrimae	BRIP 55626[a]	*A. disparrima*	Australia	KJ862358	McTaggart, Doungsa-ard, Geering, et al. (2015)
	BRIP 55632	*A. disparrima*	Australia	KJ862359	McTaggart, Doungsa-ard, Geering, et al. (2015)
E. falciformis	BRIP 57583[a]	*A. falciformis*	Australia	KJ862360	McTaggart, Doungsa-ard, Geering, et al. (2015)
E. irroratae	BRIP 57279	*A. irrorata*	Australia	KJ862365	McTaggart, Doungsa-ard, Geering, et al. (2015)
	BRIP 57286[a]	*A. irrorata*	Australia	KJ862366	McTaggart, Doungsa-ard, Geering, et al. (2015)
E. maslinii	BRIP 57872[a]	*A. daphnifolia*	Australia	KJ862367	McTaggart, Doungsa-ard, Geering, et al. (2015)
E. parvum	BRIP 57524	*A. leiocalyx*	Australia	KJ862369	McTaggart, Doungsa-ard, Geering, et al. (2015)
	BRIP 57631	*A. leiocalyx*	Australia	KJ862373	McTaggart, Doungsa-ard, Geering, et al. (2015)
E. peggii	BRIP 55602[a]	*A. holosericia*	Australia	KJ862361	McTaggart, Doungsa-ard, Geering, et al. (2015)
	BRIP 55631	*A. holosericia*	Australia	KJ862362	McTaggart, Doungsa-ard, Geering, et al. (2015)
E. phyllodiorum	BRIP 57516	*A. aulacocarpa*	Australia	KJ862378	McTaggart, Doungsa-ard, Geering, et al. (2015)
	BRIP 57578	*A. aulacocarpa*	Australia	KJ862381	McTaggart, Doungsa-ard, Geering, et al. (2015)
E. podalyriifoliae	BRIP 57294	*A. podalyriifolia*	Australia	KJ862386	McTaggart, Doungsa-ard, Geering, et al. (2015)
	BRIP 57576[a]	*A. podalyriifolia*	Australia	KJ862387	McTaggart, Doungsa-ard, Geering, et al. (2015)
E. tierneyi	BRIP 27880	*A. harpophylla*	Australia	KJ862389	McTaggart, Doungsa-ard, Geering, et al. (2015)
	BRIP 27887	*A. harpophylla*	Australia	KJ862390	McTaggart, Doungsa-ard, Geering, et al. (2015)
E. tropicae	BRIP 56555	*A. tropica*	Australia	KJ862391	McTaggart, Doungsa-ard, Geering, et al. (2015)
	BRIP 56557[a]	*A. tropica*	Australia	KJ862392	McTaggart, Doungsa-ard, Geering, et al. (2015)
E. violae-faustiae	BRIP 55601	*A. aulacocarpa*	Australia	KJ862393	McTaggart, Doungsa-ard, Geering, et al. (2015)
	BRIP 55629	*A. aulacocarpa*	Australia	KJ862394	McTaggart, Doungsa-ard, Geering, et al. (2015)
	BRIP 56544	*A. difficillis*	Australia	KJ862395	McTaggart, Doungsa-ard, Geering, et al. (2015)
	BRIP 55616	*A. difficillis*	Australia	KJ862396	McTaggart, Doungsa-ard, Geering, et al. (2015)
	BRIP 55611	*A. difficillis*	Australia	KJ862397	McTaggart, Doungsa-ard, Geering, et al. (2015)
	BRIP 56539	*A. difficillis*	Australia	KJ862398	McTaggart, Doungsa-ard, Geering, et al. (2015)
	BRIP 56540	*A. difficillis*	Australia	KJ862399	McTaggart, Doungsa-ard, Geering, et al. (2015)
	BRIP 56545	*A. difficillis*	Australia	KJ862400	McTaggart, Doungsa-ard, Geering, et al. (2015)
	BRIP 56547	*A. difficillis*	Australia	KJ862401	McTaggart, Doungsa-ard, Geering, et al. (2015)
	PRU(M) 4555	*A. crassicarpa*	Sabah, Malaysia	OR481949	This study
	PRU(M) 4556	*A. crassicarpa*	Riau, Indonesia	OR481950	This study
	PRU(M) 4557	*A. mangium* × *auriculiformis*	Riau, Indonesia	OR481951	This study
	PRU(M) 4558	*A. crassicarpa*	Kalimantan, Indonesia	OR481952	This study
Diorchidium woodii	ZT Myc 582	*Millettia grandis*	South Africa	KM217352	Beenken and Wood (2015)

Note: Specimens collected in this study are represented in bold. BRIP: Queensland Plant Pathology Herbarium, Queensland, Australia; PRU(M): H.G.W.J. Schweickerdt Herbarium, University of Pretoria, Pretoria, South Africa. ZT Myc: Zürich Herbaria Z + ZT, ETH Zürich, Switzerland.

[a]Holotype.

USA). Sequences were obtained by running the products on an ABI PRISM™ 3100 DNA sequencer (Applied Biosystems, Waltham, MA, USA) at the Sequencing Facility of the Faculty of Natural and Agricultural Sciences, University of Pretoria, South Africa. Geneious Prime 2023.0.1 was used to assemble and edit the raw sequences (https://www.geneious.com). All sequences generated in this study were deposited in GenBank (http://www.ncbi.nlm.nih.gov) (Table 1).

2.3 | Phylogenetic analyses

Reference sequences for species closely related to those emerging from this study were downloaded from GenBank database (Table 1). All sequences were aligned using MAFFT v. 7 (http://mafft.cbrc.jp/alignment/server/) (Katoh & Standley, 2013) and then confirmed manually in MEGA v. 7 (Kumar et al., 2016), where necessary. Maximum likelihood (ML) analysis was performed using RaxML v. 8.2.4 on the CIPRES Science Gateway v. 3.3 (Stamatakis, 2014) with default GTR substitution model and 1000 rapid bootstraps. Sequence data for *Diorchidium* woodii (ZT Myc 255) was used as the outgroup taxon. The resulting tree was viewed using MEGA v. 7.

2.4 | Morphology

Cut pieces of the phyllodes containing sori were boiled for a few seconds. Spore masses were scraped from the surface of phyllodes using a hypodermic needle or with an agar piece at the tip of the needle. The spores were mounted on a microscope slide in water that was replaced with 85% lactic acid, in which measurements and micrographs were made. Nikon microscopes (SMZ18, Eclipse Ni, Tokyo, Japan) and cameras (Nikon DS-Ri2, Japan) mounted on the microscopes, and an image processing program (NIS Elements, Nikon, Japan) were used to capture images, make measurements and examine morphological structures.

The colonization of the rust within the phyllode tissues was studied in vertically sectioned tissues. The boiled phyllodes containing lesions infected with rust were cut into small pieces (5 mm^2) and mounted in a freezing medium (Tissue freezing medium, Leica, UK). These tissues were cut into sections of 10–12 μm thickness using a Leica Cryomicrotome (Leica Biosystem, Germany) and mounted in 85% lactic acid for observation. Up to fifty measurements were made for characteristic structures when available and are presented as minimum–maximum (average ± standard deviation). The nomenclature used to describe spores and sori followed the suggestions of Laundon (1967) and McTaggart, Doungsa-ard, Geering, et al. (2015).

For Scanning Electron Microscopy (SEM), tissues containing lesions were cut into 1 cm^2 and fixed in 2.5% glutaraldehyde/formaldehyde (50% v/v) for 24 h. Samples were dehydrated using a graded ethanol series at 30%, 50%, 70% and 90% for 15 min each, followed by four dehydration steps in 100% ethanol, three times for 15 min each and once for 30 min. The dehydrated samples were placed in hexamethyldisilazane (HMDS) and subsequently mounted on aluminium stubs. Samples were coated with carbon using a Quorum Q150T Coating Unit (Quorum, UK) and were observed using a 540 Gemini Ultra Plus FEG SEM (Zeiss, Germany) at the Laboratory for Microscopy and Microanalysis, University of Pretoria, South Africa.

3 | RESULTS

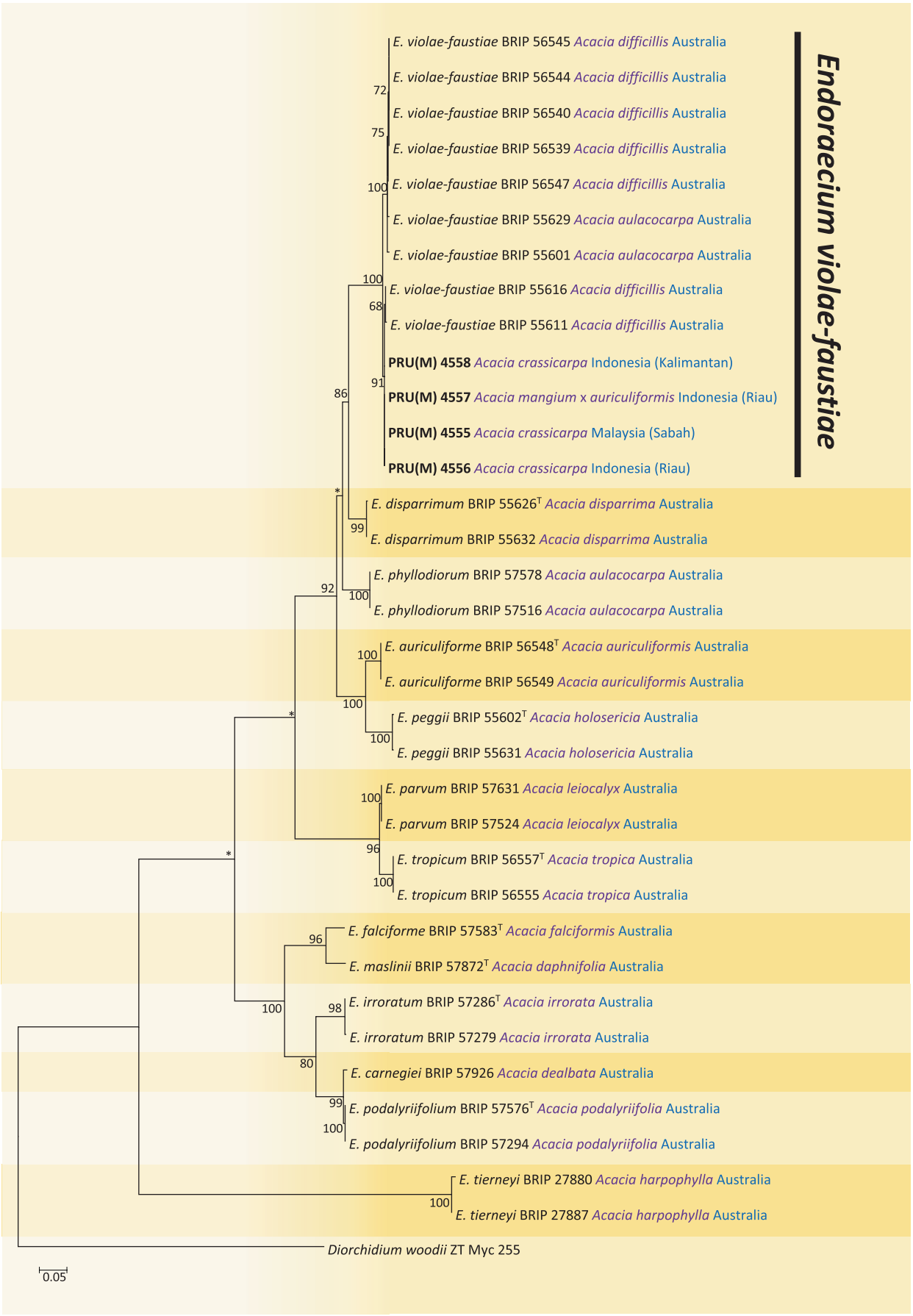
3.1 | Phylogenetic analysis

For all collected specimens, amplicons of approximately 780 bp were generated for the ITS region. The dataset used in the phylogenetic analysis included 34 ingroup taxa and contained 926 characters including alignment gaps. The four specimens from Indonesia (PRU(M) 4556, 4557, 4558) and Malaysia (PRU(M) 4555) had identical sequences and formed a well-supported (100%) monophyletic clade in the ML tree (Figure 2) that included all representative specimens of *Endoraecium violae-faustiae* from Australia. These specimens were thus identified as *E. violae-faustiae*.

3.2 | Morphology

Hyphae started to colonize under epidermis cells, occupied the palisade mesophyll cells and eventually grew towards the central parenchyma (Figure 3a). *Uredinia* were present on both the upper and underside of phyllodes, were ferruginous, scattered singly or in groups and became erumpent (Figure 3a,b,h). *Paraphyses* were present amongst urediniospores, hyaline to pale yellow, cylindrical, with apical knob-like structures similar to that of teliospores (Figure 3d,h). *Urediniospores* were yellowish brown, foveolate, sub-globose to oval or nearly cylindrical (length: width = 1.2–2.6:1), 33–48 × 17.5–30 (41 ± 4.09 × 24 ± 2.29) μm; walls were 3–5 (4 ± 0.43) μm thick at sides; apex acute or apiculate, 2–7 (5.4 ± 1.05) μm thick; bases were occasionally tapering to a point or truncated; 3–5 germ-pores were lined more or less equatorially; pedicels were mostly present, central or off-central, hyaline, tubular-shaped or trapezoid, and ranged in 1–20 (3.1 ± 2.7) μm long and 3–10.5 (7.6 ± 1.42) μm wide (Figure 3b,f–j). *Telia* were borne inside a uredinium or separately on the upper surface (Figure 3a,c). *Teliospores* were sub-hyaline to pale yellow, obovoid or cylindrical with the inflated upper part and 38–57 × 16–28 (21.8 ± 2.41) μm; walls were 1–2.5 (1.7 ± 0.38) μm thick at the sides;

FIGURE 2 Phylogenetic tree based on maximum likelihood (ML) analysis of ITS sequences for *Endoraecium* species. Specimens sequenced in this study are presented in bold. Aca*cia* host species are highlighted in purple, and the countries of origin of the specimens are in blue. Bootstrap values (≥60%) for ML analyses are indicated at the nodes. Specimen representing the holotypes are marked with a 'T'. Sequence for *Diorchidium woodii* (ZT Myc 255) represents the outgroup taxon.


Endoraecium violae-faustiae
E. violae-faustiae BRIP 56545 Acacia difficillis Australia
E. violae-faustiae BRIP 56544 Acacia difficillis Australia
E. violae-faustiae BRIP 56540 Acacia difficillis Australia
E. violae-faustiae BRIP 56539 Acacia difficillis Australia
E. violae-faustiae BRIP 56547 Acacia difficillis Australia
E. violae-faustiae BRIP 55629 Acacia aulacocarpa Australia
E. violae-faustiae BRIP 55601 Acacia aulacocarpa Australia
E. violae-faustiae BRIP 55616 Acacia difficillis Australia
E. violae-faustiae BRIP 55611 Acacia difficillis Australia
PRU(M) 4558 Acacia crassicarpa Indonesia (Kalimantan)
PRU(M) 4557 Acacia mangium x auriculiformis Indonesia (Riau)
PRU(M) 4555 Acacia crassicarpa Malaysia (Sabah)
PRU(M) 4556 Acacia crassicarpa Indonesia (Riau)
E. disparrimum BRIP 55626T Acacia disparrima Australia
E. disparrimum BRIP 55632 Acacia disparrima Australia
E. phyllodiorum BRIP 57578 Acacia aulacocarpa Australia
E. phyllodiorum BRIP 57516 Acacia aulacocarpa Australia
E. auriculiforme BRIP 56548T Acacia auriculiformis Australia
E. auriculiforme BRIP 56549 Acacia auriculiformis Australia
E. peggii BRIP 55602T Acacia holosericia Australia
E. peggii BRIP 55631 Acacia holosericia Australia
E. parvum BRIP 57631 Acacia leiocalyx Australia
E. parvum BRIP 57524 Acacia leiocalyx Australia
E. tropicum BRIP 56557T Acacia tropica Australia
E. tropicum BRIP 56555 Acacia tropica Australia
E. falciforme BRIP 57583T Acacia falciformis Australia
E. maslinii BRIP 57872T Acacia daphnifolia Australia
E. irroratum BRIP 57286T Acacia irrorata Australia
E. irroratum BRIP 57279 Acacia irrorata Australia
E. carnegiei BRIP 57926 Acacia dealbata Australia
E. podalyriifolium BRIP 57576T Acacia podalyriifolia Australia
E. podalyriifolium BRIP 57294 Acacia podalyriifolia Australia
E. tierneyi BRIP 27880 Acacia harpophylla Australia
E. tierneyi BRIP 27887 Acacia harpophylla Australia
Diorchidium woodii ZT Myc 255
0.05

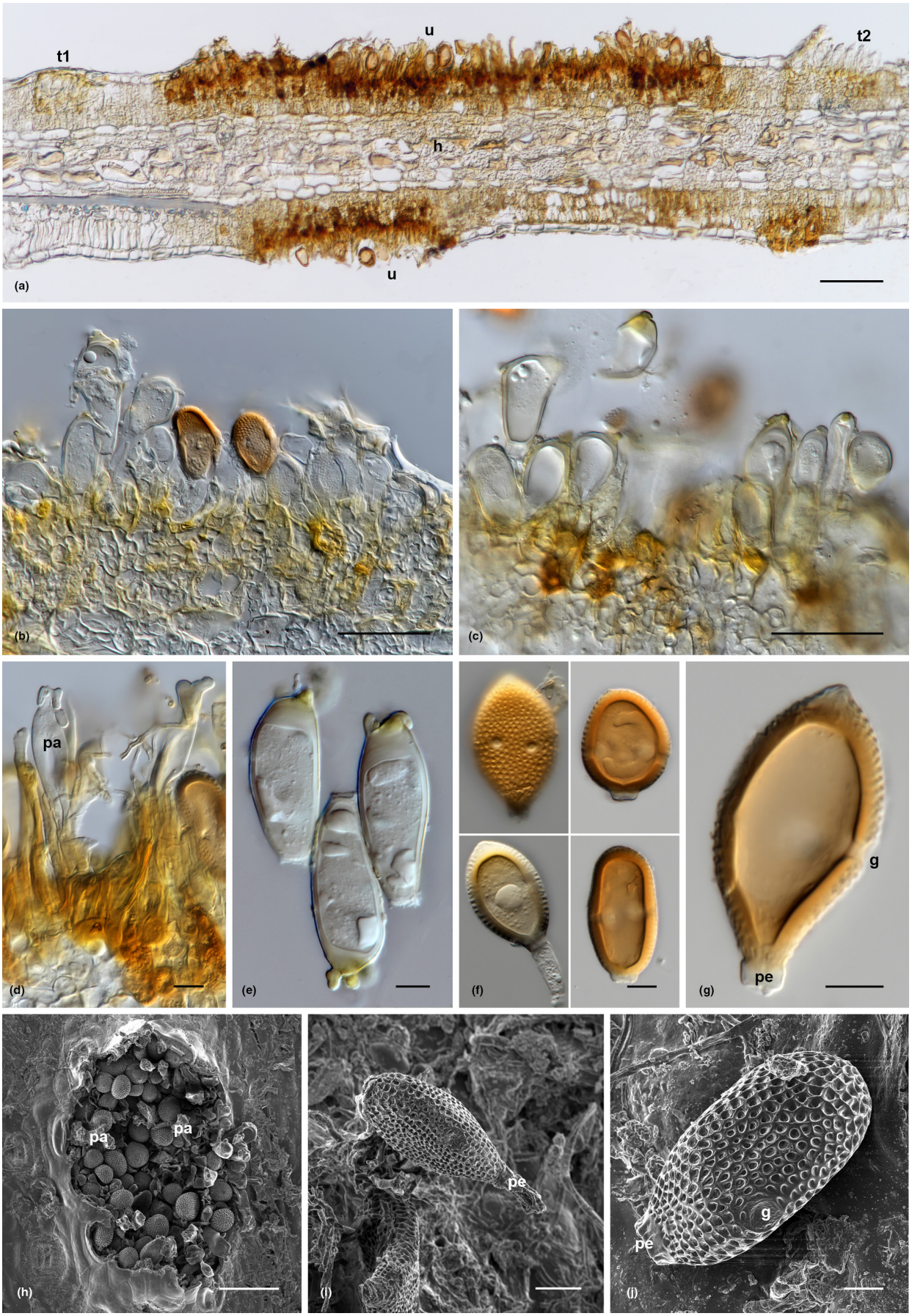

t1
u
t2
h
u
(a)
(b)
(c)
pa
(d)
(e)
(f)
g
pe
(g)
pa
pa
(h)
pe
(i)
g
pe
(j)

FIGURE 3 Micrographs of *Endoraecium violae-faustiae* infecting phyllodes of *Acacia crassicarpa*. (PRU(M) 4556: a–d, g; PRU(M) 4558 e, f, h–j). (a) Cross-section of a phyllode infected with *E. violae-faustiae* on both the upper and underside of phyllode (h: vegetative hyphae invading central parenchyma, t1: innate telium, t2: erumpent telium, u: uredinium). (b) Urediniospores and teliospores borne in a uredium. (c) Teliospores borne in a telium. (d) Paraphyses (pa) with apical ornaments similar to that of teliospores. (e) Teliospores with apical ornaments (knob-like) (f) Various shapes of urediniospores in optical section: young spore (left below) and focused on surface (left top). (g) Urediniospore in optical section showing germ-pore (g), pedicel (pe) and acute apex. (h) Erumpent uredinium producing urediniospores and paraphyses (pa). (i, j) Urediniospore. Scale bars: a = 100 μm; b, c, h = 50 μm: d–g, i = 10 μm; j = 5 μm.

apices were round, thickened with 1–4 knob-like structures; pedicels were hyaline, tubular-shaped, 3–15 (7.6 ± 5.06) μm long (Figure 3b,c,e).

4 | DISCUSSION

The results of this study have shown that the phyllode rust disease found on *A. crassicarpa* in Indonesia and Malaysia is caused by *Endoraecium violae-faustiae*. This is the first record of the rust on this tree species in plantations outside its area of origin. Although extensive surveys have not been carried out, collections included in this study were from relatively widely distributed locations. It is thus probable that *E. violae-faustiae* occurs more widely on *A. crassicarpa* in Southeast Asian plantations than is known from the present study.

Endoraecium violae-faustiae was described by Berndt (2011) in a detailed study of *Endoraecium digitatum*, which he separated into a number of discrete taxa. Prior to the present study, *E. violae-faustiae* was known only from the northern parts of Australia where it occurs on *A. crassicarpa*, *A. aulacocarpa* and *A. difficillis* (McTaggart, Doungsa-ard, Geering, et al., 2015). These species are well-adapted to tropical conditions, which accounts for the fact that *A. crassicarpa* has been extensively propagated in the humid tropics of Southeast Asia (Griffin et al., 2011; Midgley & Turnbull, 2003; Moran et al., 1989). Specifically, *A. aulacocarpa* is of interest for hybridization with *A. crassicarpa* (Griffin et al., 2011; Harwood et al., 2015) and future studies should consider the susceptibility of the former species and hybrids to infection by *E. violae-faustiae*. This could provide opportunities to avoid the disease in situations where it might cause significant defoliation.

Phyllode rust caused by *E. violae-faustiae* appears to be a relatively new disease on *A. crassicarpa* in Indonesia and Malaysia. This view is based on the fact that disease surveys have been conducted regularly in plantations of this tree since the early 1990s. It might, however, have occurred at sufficiently low levels so as not to be detected until relatively recently, when it first became obvious. Damage caused by the rust appears to be relatively mild at present, but it has become increasingly visible during the last year (Wingfield, personal observation).

The most likely pathway of entry of *E. violae-faustiae* into Southeast Asian plantations of *E. crassicarpa* would have been with infected plant material, accidentally introduced into some part of this large area, and after which air-borne dispersal would easily have occurred locally. Although various pathogens of trees widely utilized for plantation forestry, such as species of *Pinus* and *Eucalyptus*, are commonly seed-borne and have likely entered into new environments with such germplasm (Ioos et al., 2009; Jimu et al., 2015), rust pathogens would be unlikely to move in this way. The emergence of rust caused *by E. violae-faustiae* in plantations of *A. crassicarpa* emphasizes the need for adequate quarantine measures to prevent the accidental introduction of new pathogens into areas where non-native trees are being propagated for plantation forestry (Burgess & Wingfield, 2002, 2016; Healy et al., 2022).

It would be interesting to determine whether *E. violae-faustiae* in the present study, originated from a single introduction into one of the *A. crassicarpa* areas sampled, from which it then spread further. Rust fungi have powdery spores that are easily wind dispersed, which would have allowed for a single introduction of the pathogen to spread more widely. Alternatively, there could have been multiple introductions into different areas. Knowledge of this pathway of introduction and the diversity of the pathogen would require more extensive collections and a population genetic study, the value of which would lie in understanding patterns of resistance to the disease.

Acacia crassicarpa is one of the most important plantation-grown tree species in lowland areas of Indonesia, where it is extensively propagated. These trees have been relatively free of serious disease problems in the past. The appearance of a phyllode rust disease is thus of concern. Surveys should now be undertaken to assess the incidence and severity of the disease and to develop strategies to reduce its impact.

ACKNOWLEDGEMENTS

We acknowledge financial support linked to the RGE-FABI Tree Health Programme and the University of Pretoria that made this study possible. We are also most grateful to Dr. Alistair McTaggart who shared his extensive knowledge of rust pathogens with us and who kindly also provided advice on a pre-submission version of this paper.

CONFLICT OF INTEREST STATEMENT

The author declares no conflicts of interest.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

ORCID

Michael J. Wingfield https://orcid.org/0000-0001-9346-2009
Seonju Marincowitz https://orcid.org/0000-0002-4726-1211

Irene Barnes https://orcid.org/0000-0002-4349-3402
Marthin Tarigan https://orcid.org/0000-0002-9128-2650
Myriam Solís https://orcid.org/0000-0001-6468-135X
Alvaro Durán https://orcid.org/0000-0002-3035-9087
Nam Q. Pham https://orcid.org/0000-0002-4938-9067

REFERENCES

Barnes, I., Abdul Rauf, M. R., Fourie, A., Japarudin, Y., & Wingfield, M. J. (2023). *Ceratocystis manginecans* and not *C. Fimbriata* a threat to propagated *Acacia* spp. in Sabah, Malaysia. *Journal of Tropical Forest Science*, *35*, 16–26.

Beenken, L., & Wood, A. R. (2015). *Puccorchidium* and *Sphenorchidium*, two new genera of Pucciniales on Annonaceae related to *Puccinia psidii* and the genus *Dasyspora*. *Mycological Progress*, *14*, 49.

Beenken, L., Zoller, S., & Berndt, R. (2012). Rust fungi on Annonaceae II: The genus *Dasyspora* Berk. & M.A. Curtis. *Mycologia*, *104*, 659–681.

Beilharz, V. C., Pascoe, I. G., Wingfield, M. J., Tjahjono, B., & Crous, P. W. (2004). *Passalora perplexa*, an important pleoanamorphic leaf blight pathogen of *Acacia crassicarpa* in Australia and Indonesia. *Studies in Mycology*, *50*, 471–479.

Berndt, R. (2011). Taxonomic revision of *Endoraecium digitatum* (rust fungi, Uredinales) with description of four new species from Australia and Hawaii. *Mycological Progress*, *10*, 497–517.

Burgess, T., & Wingfield, M. J. (2002). Quarantine is important in restricting the spread of exotic seed-borne tree pathogens in the southern hemisphere. *The International Forestry Review*, *4*, 56–65.

Burgess, T., & Wingfield, M. J. (2016). Pathogens on the move: A 100-year global experiment with planted eucalypts. *Bioscience*, *67*, 14–25.

Coutinho, T. A., Wingfield, M. J., Alfenas, A. C., & Crous, P. W. (1998). Eucalyptus rust: A disease with the potential for serious international implications. *Plant Disease*, *82*, 819–825.

Doungsa-ard, C., McTaggart, A. R., Geering, A. D. W., & Shivas, R. G. (2018). Diversity of gall-forming rusts (Uromycladium, Pucciniales) on *Acacia* in Australia. *Persoonia*, *40*, 221–238.

Fraser, S., McTaggart, A. R., Roux, J., Nel, J., Potgieter, J., Shuey, L. S., Somchit, C., & Wingfield, M. J. (2021). The life cycle and field epidemiology of *Uromycladium acaciae* (Pucciniales) on *Acacia mearnsii* in South Africa. *Annals of Applied Biology*, *179*, 21–33.

Glen, M., Alfenas, A. C., Zauza, E. A. V., Wingfield, M. J., & Mohammed, C. (2007). *Puccinia psidii*: A threat to the Australian environment and economy—A review. *Australasian Plant Pathology*, *36*, 1–16.

Griffin, A. R., Midgley, S. J., Bush, D., Cunningham, P. J., & Rinaudo, A. T. (2011). Global uses of Australian acacias—Recent trends and future prospects. *Diversity and Distributions*, *17*, 837–847.

Harwood, C. E., Hardiyanto, E. B., & Yong, W. C. (2015). Genetic improvement of tropical acacias: Achievements and challenges. *Southern Forests: a Journal of Forest Science*, *77*, 11–18.

Healy, M. A., Warman, R., Mohammed, C., & Lawson, S. A. (2022). Acacia and eucalypt plantation biosecurity in Southeast Asia—A history, and directions for future research and engagement. *Australian Forestry*, *85*, 146–160.

Hurley, B. P., Barnes, I., & Wingfield, M. J. (2023). Diseases and insect pests of Australian *Acacia* species utilised in plantation forestry. In D. M. Richardson (Ed.), *Wattles: Australian Acacia species around the world*. CAB International (CABI), Wallingford.

Ioos, R., Fourrier, C., Iancu, G., & Gordon, T. R. (2009). Sensitive detection of *Fusarium circinatum* in pine seed by combining and enrichment procedure with real-time polymerase chain reaction using dual-labeled probe chemistry. *Phytopathology*, *99*, 582–590.

Jimu, L., Kemler, M., Wingfield, M. J., Mwenje, E., & Roux, J. (2015). The *Eucalyptus* stem canker pathogen *Teratosphaeria zuluensis* detected in seed samples. *Forestry*, *89*, 316–324.

Katoh, K., & Standley, D. M. (2013). MAFFT multiple sequence alignment software version 7: Improvements in performance and usability. *Molecular Biology and Evolution*, *30*, 772–780.

Kumar, S., Stecher, G., & Tamura, K. (2016). MEGA7: Molecular evolutionary genetics analysis version 7.0 for bigger datasets. *Molecular Biology and Evolution*, *33*, 1870–1874.

Laundon, G. F. (1967). Terminology in the rust fungi. *Transactions of the British Mycological Society*, *50*, 189–194.

Lynn, K. M. T., Wingfield, M. J., Durán, A., Marincowitz, S., Oliveira, L. S. S., de Beer, Z. W., & Barnes, I. (2020). *Euwallacea perbrevis* (Coleoptera: Curculionidae: Scolytinae), a confirmed pest on *Acacia crassicarpa* in Riau, Indonesia, and a new fungal symbiont; *Fusarium rekanum* sp. nov. *Antonie Van Leeuwenhoek*, *113*, 803–823.

Lynn, K. M. T., Wingfield, M. J., Durán, A., Oliveira, L. S. S., de Beer, Z. W., & Barnes, I. (2021). Novel *Fusarium* mutualists of two *Euwallacea* species infesting *Acacia crassicarpa* in Indonesia. *Mycologia*, *113*, 536–558.

McTaggart, A. R., Doungsa-ard, C., Geering, A. D. W., Aime, M. C., & Shivas, R. G. (2015). A co-evolutionary relationship exists between *Endoraecium* (Pucciniales) and its *Acacia* hosts in Australia. *Persoonia*, *35*, 50–62.

McTaggart, A. R., Doungsa-ard, C., Wingfield, M. J., & Roux, J. (2015). *Uromycladium acaciae*, the cause of a sudden, severe disease epidemic on *Acacia mearnsii* in South Africa. *Australasian Plant Pathology*, *44*, 637–645.

Midgley, S. J., & Turnbull, J. W. (2003). Domestication and use of Australian acacias: Case studies of five important species. *Australian Systematic Botany*, *16*, 89–102.

Mohd Farid, A., Terhem, R., Mohd Aswad, R., Augustini, L., Ho, W. M., Indrayadi, H., & Hidayati, N. (2023). Diseases of *Acacia* and control measures in the tropics. In *Forest Microbiology Vol. 3. Tree diseases and pests* (pp. 375–400). Academic Press.

Moran, G. F., Mouna, O., & Bell, J. C. (1989). Breeding systems and genetic diversity in *Acacia auriculiformis* and *A. crassicarpa*. *Biotropica*, *21*, 250–256.

Moreno Chan, J., Day, P., Feely, J., Thompson, R., Little, K. M., & Norris, C. H. (2015). *Acacia mearnsii* industry overview: Current status, key research and development issues. *Southern Forests: a Journal of Forest Science*, *77*, 19–30.

Moreno Chan, J., & Isik, F. (2021). Genetic variation in frost tolerance, *Uromycladium acaciae* rust resistance, and growth in an *Acacia mearnsii* population. *Forest Science*, *67*, 574–586.

Morris, M. J., Wingfield, M. J., & Walker, J. (1988). First record of a rust on *Acacia mearnsii* in southern Africa. *Transactions of the British Mycological Society*, *90*, 324–327.

Old, K. M., Lee, S. S., Sharma, J. K., & Yuan, Z. Q. (2000). *A manual of diseases of tropical acacias in Australia, South East Asia and India*. CIFOR.

Payn, T., Carnus, J.-M., Freer-Smith, P., Kimberley, M., Kollert, W., Liu, S., Orazio, C., Rodriguez, L., Silva, L. N., & Wingfield, M. J. (2015). Changes in planted forests and future global implications. *Forest Ecology and Management*, *352*, 57–67.

Pfunder, M., Schürch, S., & Roy, B. A. (2001). Sequence variation and geographic distribution of pseudoflower-forming rust fungi (*Uromyces pisi* s. lat.) on *Euphorbia cyparissias*. *Mycological Research*, *105*, 57–66.

Pham, N. Q., Wingfield, M. J., Marincowitz, S., Agena, A. T., Kumela, R. T., Weldesenbet, B. K., Hurley, B. P., Slippers, B., Germishuizen, I., Lawson, S. A., Healy, M. A., & Wondafrash, M. (2023). First report of the wattle rust pathogen, *Uromycladium acaciae* (Raveneliaceae, Pucciniales) in Ethiopia. *Forestry*, cpad040. https://doi.org/10.1093/forestry/cpad040

Richardson, D. M., Le Roux, J. J., & Wilson, J. R. (2015). Australian acacias as invasive species: Lessons to be learnt from regions with long

planting histories. *Southern Forests: a Journal of Forest Science*, *77*, 31–39.

Roux, J., & Wingfield, M. J. (2009). *Ceratocystis* species: Emerging pathogens of non-native plantation *Eucalyptus* and *Acacia* species. *Southern Forests: a Journal of Forest Science*, *71*, 115–120.

Stamatakis, A. (2014). RAxML version 8: A tool for phylogenetic analysis and post-analysis of large phylogenies. *Bioinformatics*, *30*, 1312–1313.

Tarigan, M., Roux, J., van Wyk, M., Tjahjono, B., & Wingfield, M. J. (2011). A new wilt and die-back disease of *Acacia mangium* associated with *Ceratocystis manginecans* and *C. acaciivora* sp. nov. in Indonesia. *South African Journal of Botany*, *77*, 292–304.

Tarigan, M., Wingfield, M. J., van Wyk, M., Tjahjono, B., & Roux, J. (2011). Pruning quality affects infection of *Acacia mangium* and *A. crassicarpa* by *Ceratocystis acaciivora* and *Lasiodiplodia theobromae*. *Southern Forests: a Journal of Forest Science*, *73*, 187–191.

Wingfield, M. J., Brokerhoff, E. G., Wingfield, B. D., & Slippers, B. (2015). Planted forest health: The need for a global strategy. *Science*, *349*, 832–836.

Wingfield, M. J., Roux, J., & Wingfield, B. D. (2011). Insect pests and pathogens of Australian acacias grown as non-natives—An experiment in biogeography with far-reaching consequences. *Diversity and Distributions*, *17*, 968–977.

Wingfield, M. J., Wingfield, B. D., Warburton, P., Japarudin, Y., Lapammu, M., Adul Rauf, M. R., Boden, D., & Barnes, I. (2023). Ceratocystis wilt of *Acacia mangium* in Sabah: Understanding the disease and reducing its impact. *Journal of Tropical Forest Science*, *35*, 51–66.

How to cite this article: Wingfield, M. J., Marincowitz, S., Barnes, I., Tarigan, M., Solís, M., Durán, A., & Pham, N. Q. (2023). First report of phyllode rust on *Acacia crassicarpa* outside its native range. *Forest Pathology*, *53*, e12839. https://doi.org/10.1111/efp.12839