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Identification and Diversity Analysis of *Uromycladium* on *Paraserianthes lophantha* subsp. *montana* Using a Multilocus Molecular Approach

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ABSTRACT

Gall rust caused by the *Uromycladium* complex is one of the most devastating diseases in forest plants. Gall rust affects several plant genera. *Paraserianthes lophantha* is one of the forest plants affected by gall rust disease. The disease also poses a serious threat to industrial forests such as *Falcataria moluccana*. However, to develop disease control strategies and risk assessments, no definitive species of *Uromycladium* associated with gall rust disease on *P. lophantha* subsp. *montana* is known in Indonesia. The research objective is to identify the existing *Uromycladium* species infecting *P. lophantha* in Indonesia using morphological identification and multilocus molecular analysis of the ITS and LSU regions. Morphological data for teliospore identification indicated that the causal agent of gall rust on *P. lophantha* was *Uromycladium tepperianum*, but molecular analysis identified it as *U. woodii.* Since the morphological features are more influenced by environmental conditions, we believed that molecular analysis was more accurate. Phylogenetic analysis showed that *U. woodii on P. lophantha* is distinctive from the *Acacia* group. These findings suggest that the *U. woodii* clade comprises several types of gall-forming rust fungi that differ by host species. Since *U. woodii* was also able to infect *F. moluccana,* an important industrial tree in Southeast Asia, monitoring and awareness of this rust fungus need to be improved.

Keywords: Gall rust, multi-gene, phylogeny, *Uromycladium woodii*

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INTRODUCTION

Paraserianthes lophantha (Willd.) I.C.Nielsen (syn. *Albizia lophantha*;

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Fabaceae) is a leguminous shrub or small tree with a disjunct distribution. The lophantha subspecies occurs in south-western Australia, whereas subsp. *montana* (Jungh.) I.C.Nielsen is native to Indonesia, where it is found at high elevations on Sumatra, Java, Bali and Flores (Nielsen et al., 1983). Molecular phylogenetic work has confirmed that *Paraserianthes*, in the strict sense, is now restricted to *P. lophantha* and that the genus is closely related to *Acacia sensu stricto* (Brown et al., 2011). In Indonesia, *P. lophantha* colonises elevations typically above 1,500m up to 3,000+ m asl and grows along crater edges and active volcanic and gravel beds, while in Australia, *P. lophantha* subsp. lophantha naturally inhabits coastal environments (Nielsen et al., 1983). In Indonesia, villagers considered this species a high-mountain pioneer, used it for firewood, and never cultivated it as a planted tree. According to Rahayu (2014), *P. lophantha* subsp. *montana* has also been infected by *Uromycladium tepperianum*, a rust-forming gall that has caused outbreaks of gall rust diseases on *Falcataria moluccana* (sengon) in the Philippines, Sabah, Malaysia, and Java Island, Indonesia.

Gall rust disease is one of the most destructive diseases in forest plants, including *Acacia, Paraserianthes*, *Falcataria*, and *Calliandra* (Fraser et al., 2021; Pham et al., 2024; Rahayu, 2014). The rust fungus *Uromycladium* spp., native to Australia, causes the disease. There are 28 species in the genus *Uromycladium*, including *U. tepperianum*, *U. falcatarium*, *U. acaciae*, and *U. woodii*. (Doungsa-ard et al., 2015; McTaggart et al., 2015; Ryley, 2024). These rust fungi have complex life cycles that can involve up to three fruiting body types and four spore types. With the possible exception of *U. bisporum*, all *Uromycladium* species are microcyclic. *U. tepperianum* has only two spore states: pycniospores (spermatia) and teliospores. Morris (1987) reported urediniospores in the life cycle of all other species except *U. bisporum*. *Uromycladium* fungi are autoecious and require only one host to complete their life cycle. Gall rust diseases can vary by site or remain similar across vast regions, depending on complex interactions among the disease, the tree, and site- or region-specific ecology. According to Rahayu et al. (2018), more closed sites, undulating topography, fog, lower age, and higher altitude were significant local site conditions that support gall rust disease incidence and severity. High relative humidity and low wind speed promoted gall rust disease development. The most severe gall rust disease on *F. moluccana* occurred at elevations above 900 m a.s.l. at SFI, Sabah (Anonymous, 1993), while in East Timor the disease was most severe at elevations of between 700 and 900 m a.s.l. in the mountains southwest of Dili (Old & Cristovao, 2003). However, they did not explain why the disease was more severe at that elevation. According to Braza (1997), high-elevation areas, where environmental conditions appear conducive to rust development, are not considered for planting *F. moluccana* in the Philippines. At high elevations, foggy conditions usually occur. According to Hoff (1986), foggy periods with

intermittent showers induced high spore production, favoring rust infection. Uromycladium infection led to cell hyperplasia, which eventually caused tree mortality (Rahayu et al., 2010). However, researchers have also used Uromycladium as a biological control agent for the invasive *Acacia saligna* in South Africa (Morris, 1991; Wood & Morris, 2007).

McAlpine (1906) first reported gall rust disease on *P. lophantha* in Indonesia, caused by *U. tepperianum*. However, Morris (1987) described the *Uromycladium* species as highly specific to *P. lophantha* subsp. *lophantha* was *U. woodii*. Researchers collected this rust from the Porongurup Ranges, Western Australia, and maintained it on living plants at CSIRO, Australia, and the Agricultural Research Council Plant Protection Research Institute, South Africa, where they studied it as a potential biological control agent for *P. lophantha* (Impson et al., 2011). Because more than one *Uromycladium* taxon has been associated with *P. lophantha*, namely *U. woodii* in Australia and *U. tepperianum sensu lato* in Java, the relationship among these rusts in Indonesia remains to be determined. Hence, it also raises the question of how many *Uromycladium* species exist in Indonesia.

According to Doungsa-ard et al. (2018), *U. tepperianum s. lat.* was divided into 16 species. Therefore, genetic analysis of rust fungi is necessary to support the morphological identification studies. Occasionally, taxonomic identification based on morphological data alone does not align with genetic analysis results, leading to ambiguity. One extreme example of incongruence between morphology-based taxonomic relationships and Small Subunit (SSU) data is the case of the two genera in the family Pileolariaceae (*Pileolaria* and *Uromycladium*) (Cummins & Hiratsuka, 1983). These two genera appear unrelated based on SSU data, and they do not co-occur in phylogenetic trees (Wingßeld et al., 2004). To obtain accurate molecular data, multilocus analysis is required. For rust fungi, multilocus analysis is commonly used to assess diversity (Corcoran et al., 2014; Doungsa-ard et al., 2018; Zhafarina et al., 2021).

Inconsistencies between morphological identification and molecular data can also occur in rust fungi, given their high diversity and host-jump events (McTaggart et al., 2016; van der Merwe et al., 2008). Therefore, morphological identification and multilocus molecular analysis must be conducted simultaneously. In addition, the only *P. lophantha* rust sequenced so far is *U. woodii* from subsp. *lophantha* in Western Australia. *U. tepperianum sensu lato* was recorded on subsp. *montana* in Java by Magnus (1892) and Boedijn (1959), but those records are now of uncertain identity.

It is unknown whether the Indonesian rust is *U. woodii*, *U. falcatariae,* or a distinct species. This research aimed to validate the existing *Uromycladium* species infecting *P. lophantha* in Indonesia using morphological identification and multilocus molecular analysis.

MATERIALS AND METHODS

Sample Collection

Freshly mature galled tissue with mature teliospores of *P. lophantha* subsp. *montana* was collected in February 2016 from two mountains on the island of Java, Indonesia: Mount Merbabu and Mount Lawu. We also collected fresh, mature teliospores of *F. moluccana* from Mount Merapi simultaneously (Figure 1). We sampled two sites on each mountain and, at each site, collected galls from at least 3 symptomatic trees spaced 10 m apart. For each sample, we recorded GPS coordinates, elevation, the infected plant part, and gall morphology, as shown in Table 1.

We placed galled tissue in paper bags, transported it to the laboratory, dried it in an oven at 60 °C for DNA extraction, and kept it fresh for morphological examination. We identified and confirmed the host plant as *F. lophantha* subsp. *montana* using material from the herbarium at the Faculty of Forestry, Universitas Gadjah Mada, and validated by an expert botanist.

Morphological Examination

We mounted freshly collected rust spores on glass slides in 100% lactic acid and gently heated them to boiling before microscopic examination. We used ImageJ 1.46 to measure teliospore size range, germ pore width, and number of striae. We expressed teliospore size ranges as (min-max) and germ pore width as (min-max). Means and standard deviations (SD) were calculated from at least 50 spores of each host. We captured images with an EOS Kiss X5 camera attached to a DMLB Fluorescent Microscope (Leica Microsystems, Wetzlar, Germany).

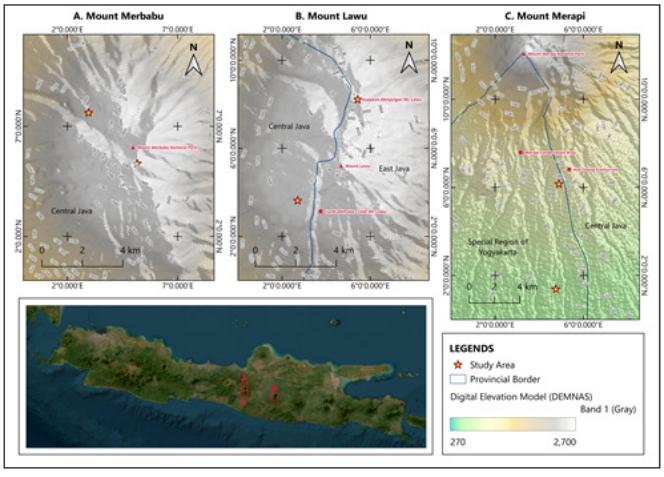

Figure 1. The study area for sample collection of (a) and (b) *Paraserianthes lophantha* subsp. *montana* forming galls infected by *Uromycladium* fungus at Mount Merbabu and Mount Lawu, (c) *Falcataria moluccana* at Mount Merapi infected by *U. falcatarium,* and (d) Their position in Java Island, Indonesia

Table 1
The location, host. elevation, infected plant part, and gall morphology of each sample

Sample	Mountain	Location	Host	Altitude m a.s.l.	Coordinates	Infected Plant Part	Gall Morphology
1	Merbabu	Boyolali Regency, Central Java	*Paraserianthes lophantha* subsp. *montana*	Pos 2, 2,507 m	-7.442683, 110.430005	branches and twigs, leaf petioles	globose, or woody convoluted masses
2	Merbabu	Boyolali Regency, Central Java	*Paraserianthes lophantha* subsp. *montana*	Kenteng Songo 3,100 m	-7.453925, 110.440491	branches and twigs, leaf petioles	globose, or woody convoluted masses
3	Lawu	Karanganyar Regency, Central Java	*Paraserianthes lophantha* subsp. *montana*	Telaga Menjangan 2,948 m	-7.612561, 111.198058	branches and twigs, leaf petioles	globose, or woody convoluted masses
4	Lawu	Karanganyar Regency, Central Java	*Paraserianthes lophantha* subsp. *montana*	Pos 3 Penggik 2,868 m	-7.635244, 111.184612	branches and twigs, leaf petioles	globose, or woody convoluted masses
5	Merapi	Cangkringan District, Yogyakarta Special Region	*Falcataria moluccana*	1,050 m	-7.589194, 110.458583	Stem and branch	as swollen, tumor-like, elongated
6	Merapi	Cangkringan District, Yogyakarta Special Region	*Falcataria moluccana*	650 m	-7.640667, 110.468639	Stem and branch	as swollen, tumor-like elongated

DNA Extraction, PCR Amplification, and Sequencing

DNA was extracted with a MagExtractor-Plant Genome Kit. Fungal nuclear rDNA was amplified by polymerase chain reaction (PCR) with LifePro Thermal Cycler version 2010-1.0 using the manufacturer-specified cycling and reaction conditions. The Large Subunit (LSU) region of rDNA was amplified with the primers Rust2inv/LR7 (Aime, 2006) and nested with the primers LR0R/LR6 (Vilgalys & Hester, 1990). The primers ITS1-F/ITS4-B were used for amplification of the Internal Transcribed Spacer (ITS) region of rDNA (Gardes & Bruns, 1993). The PCRs were performed with the following: initial denaturation at 94 ^{o}C for 2 min, followed by 40 cycles of denaturation at 94 ^{o}C for 30 s, annealing for 1 min, and extension at 72 ^{o}C for 1.5 min. The annealing temperatures were 56 ^{o}C for ITS amplification, 55 ^{o}C for initial LSU amplification, and 57 ^{o}C for nested LSU amplification. A final extension was performed at 72 ^{o}C for 7 min. PCR products were purified and subsequently sequenced by the Sanger direct sequencing method using an ABI PRISM 377 automated DNA sequencer.

Phylogenetic Analysis

Four samples in this study were sequenced, and the resulting sequences were aligned using BIOEDIT v. 7.0.9 (Hall et al., 2011). Additional ingroup sequences were selected based on BLAST searches (Altschul et al., 1990) and retrieved from GenBank based on their accession numbers listed in Table 2. A single outgroup sequence was included to root the phylogenetic trees. Maximum likelihood (ML) phylogenetic reconstruction was performed using MEGA 12 (Kumar et al., 2024). Prior to tree reconstruction, the best-fit nucleotide substitution model was determined using the model selection procedure, and the GTR+G+I model was selected for the analysis. Branch support was assessed using 1,000 bootstrap replicates (Felsenstein, 1985), and the resulting values were reported as maximum likelihood bootstrap percentages (MLBP). Bayesian inference (BI) was conducted using the BEAST software package (Bouckaert et al., 2019). The resulting phylogenetic trees were visualised using FIGTREE v. 1.4.3 (Rambaut, 2009), with Bayesian posterior probabilities (BPP) displayed as branch support values.

RESULTS AND DISCUSSION

Morphological Identification

Morphological examination showed that the rust fungi on *F. moluccana* and *P. lophantha* subsp. *montana* in Indonesia possess teliospores with converging striae and pedicellate teliospores (Figure 2). After comparing teliospore characteristics from *F. moluccana*, *Acacia* spp., and *P. lophantha* subsp. lophantha, the rust on *P. lophantha* subsp. *montana* showed striae similar to those of *Acacia* spp. (about 30-45). The striae range was broader than that of rust on *F. moluccana* (25-36). Teliospores from *P. lophantha* subsp. *montana* were

Table 2
List of rust specimens included in this study

No.	Taxon	Accession Number	Host	Genbank Accession	
				ITS Accession	LSU Accession
1	*Uromycladium brachycarpae*	BRIPa 58599	*Acacia brachycarpa*	KR994736	KR994685
2	*U. holosericeae*	BRIP 56538	*A. holosericea*	KR994740	KR994689
3	*U. farinosae*	BRIP 58154	*A. farinosa*	KR994737	KR994686
4	*U. implexae*	BRIP 57313	*A. implexa*	KR994743	KR994692
5	*U. leiocalycis*	BRIP 57285	*A. leiocalyx*	KR994746	KR994695
6	*U. ligustrinae*	BRIP 57708	*A. ligustrina*	KR994750	KR994699
7	*U. maslinii*	BRIP 57703	*A. latior*	KJ632999d	KJ632975d
8	*U. merrallii*	BRIP 58153	*A. merrallii*	KR994757	KR994711
9	*U. mitchellii*	BRIP 59355	*A. trudgeniana*	KR994836	KR994827
10	*U. morrisii*	BRIP 56963	*A. saligna*	KJ633022d	KJ632980d
11	*U. morrisii*	BRIP 57860	*A. saligna*	KJ633027d	KJ632988d
12	*U. murphyi*	BRIP 55674	*A. elata*	KR994837d	KR994828d
13	*U. murphyi*	BRIP 57629	*A. decurrens*	KR994838d	KR994829d
14	*U. murphyi*	BRIP 57929	*A. rubida*	KR994841d	KR994832d
15	*U. paradoxae*	BRIP 58152	*A. paradoxa*	KR994758	KR994712
16	*U. paradoxae*	BRIP 59221	*A. montana*	KR994761	KR994715
17	*U. paradoxae*	BRIP 59235	*A. verniciflua*	KR994762	KR994716
18	*U. tepperianum*	BRIP 57307	*A. ligulata*	KR994765	KR994719
19	*U. tepperianum*	BRIP 57707	*A. rostellifera*	KR994767	KR994721
20	*U. tepperianum*	BRIP 57742	*A. cupularis*	KR994769	KR994723
21	*U. tepperianum*	BRIP 59439	*A. xanthina*	KR994774	KR994728
22	*U. tepperianum*	BRIP 61265	*A. sclerosperma*	KR994777	KR994731

Table 2 (*continued*)

No.	Taxon	Accession Number	Host	Genbank Accession	
				ITS Accession	LSU Accession
23	*U. woodii*	BRIP 61600	*Paraserianthes lophantha*	KR994779	KR994733
24	*U. woodii*	BRIP 62249	*P. lophantha*	KR994780	KR994734
25	*Merbabu1 P. lophantha* subsp. *montana*	This study	*P. lophantha* subsp. *montana*	-	-
26	*Merbabu2 P. lophantha* subsp. *montana*	This study	*P. lophantha* subsp. *montana*	-	-
27	*Lawu1 P. lophantha* subsp. *montana*	This study	*P. lophantha* subsp. *montana*	-	-
28	*Lawu2 P. lophantha* subsp. *montana*	This study	*P. lophantha* subsp. *montana*	-	-
29	*Trachyspora intrusa*	BPI 843828	*Alchemilla vulgaris*	DQ354550	DQ354550

*Source: [d]Doungsa-ard et al. (2015)

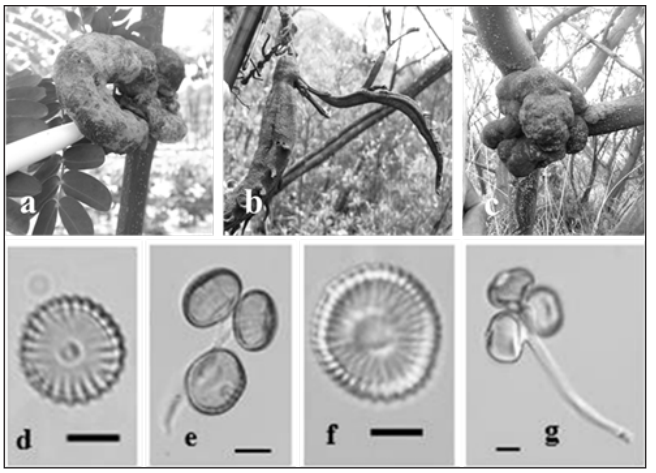

Figure 2. (a) The gall on the branch of *Falcataria moluccana*; (b) the gall on the twig; and (c) on the branch of *Paraserianthes lophantha* subsp. montana; (d) teliospores with converging striae and (e) pedicellate teliospores on *F. moluccana*; (f) teliospores with converging striae and (g) pedicellate teliospores on *Paraserianthes lophantha* subsp. montana Bar = 10 µm

bigger than those on *F. moluccana*, *Acacia* spp., and *P. lophantha* subsp. lophantha. The rust on *P. lophantha* subsp. *montana* also has larger germ pores (about 5-7 µm) than those of *F. moluccana* (2.5-4 µm), *P. lophantha* subsp. lophantha (3.5-5.5 µm), and *Acacia* spp. (3-5.5 µm) as well (Table 3).

Multi-Gene Molecular Analysis

Phylogenetic analysis based on the combined ITS and LSU rDNA sequences using Maximum Likelihood (ML) and Bayesian Inference (BI) approaches showed that the four *Uromycladium* samples obtained from *Paraserianthes lophantha* subsp. *montana* on Mount Merbabu and Mount Lawu clustered with two reference specimens of *U. woodii* (BRIP 61600 and BRIP 62249), receiving 100% ML bootstrap value (MLBP) and Bayesian posterior probability (BPP) of 1.00 (Figure 3). The two reference specimens of *U. woodii* formed a subclade supported by 90% MLBP and 1.00 BPP, while the internal relationships among the study samples showed varying levels of support. The five reference specimens of *U. tepperianum* associated with *Acacia* species formed a separate group, with support of 100% MLBP and 1.00 BPP. These results indicate that the sample in this study is more closely related to *U. woodii* than to *U. tepperianum*.

These results align with taxonomic revision by Doungsa-ard et al. (2018), who recognised *U. tepperianum sensu lato* as a species complex and described *U. woodii* as a distinct species. In that revision, *U. tepperianum sensu stricto* was restricted to hosts belonging to the *Acacia bevinosa* group, while *U. woodii* was described from *P. lophantha* subsp. *lophanta* in Western Australia (BRIP 61600 as the holotype and BRIP 62249 included

Table 3
Spore characteristics of Uromycladium *spp. on* F. moluccana, P. lophantha*, and* Acacia

Rust Species	Host	Size of Teliospores (µm)	Width of Germ Pore	Number of Striae	Colour and Shape of the Sori	Reference
U. tepperianum sensu Rahayu et al. 2010	*F. moluccana*	(18) 19-22 (24) × (17) 18-21 (22)	2-4	25-36	Cinnamon brown; depressed globose or subglobose	This study
U. tepperianum sensu Rahayu et al. 2010	*F. moluccana*	(17) 20-22 (24) × (17) 19-21 (22)	2-4	25-36	Cinnamon brown; depressed globose or subglobose	This study
U. falcatarium	*F. moluccana*	(17) 18-22 (24) × (13) 15-19 (21)	2.5-4	25-32	Cinnamon brown; depressed globose or subglobose	Doungsa-ard et al. 2015
Uromycladium sp.	*P. lophantha* subsp. *montana* sample 1	(22) 23-26 (28) × (20) 21-24 (25)	5-7	30-43	Cinnamon brown; depressed globose or subglobose	This study
Uromycladium sp.	*P. lophantha* subsp. *montana* sample 2	(22) 23-26 (29) × (21) 22-24 (25)	5-7	32-44	Cinnamon brown; depressed globose or subglobose	This study
Uromycladium sp.	*P. lophantha* subsp. *montana* sample 3	(23) 24-26 (27) × (21) 22-24 (25)	5-7	30-43	Cinnamon brown; depressed globose or subglobose	This study
Uromycladium sp.	*P. lophantha* subsp. *montana* sample 4	(21) 23-25 (28) × (20) 21-23 (25)	5-7	30-43	Cinnamon brown; depressed globose or subglobose	This study
U. tepperianum	*A. leiocalyx*[b]	(19) 20-22 (23) × (14) 14-20 (27)	4-4.5	31-38	Cinnamon brown; depressed globose or subglobose	Doungsa-ard et al. 2015
U. tepperianum	*A. implexa*	(16) 17-20 (21) × (12) 13-16 (17)	3-4.5	32-39	Cinnamon brown; depressed globose or subglobose	Doungsa-ard et al. 2015
U. tepperianum	*A. holosericea*	(18) 20-24 (24) × (16) 17-19 (20)	4-5.5	35-40	Cinnamon brown; depressed globose or subglobose	Doungsa-ard et al. 2015
U. tepperianum	*A. saligna*	(17) 18-22 (26) × (11) 14-18 (20)	4-5.5	33-40	Cinnamon brown; depressed globose or subglobose	Doungsa-ard et al. 2015
U. tepperianum	*A. latior*	(18) 20-22 (24) × (14) 16-18 (21)	4-5.5	30-44	Cinnamon brown; depressed globose or subglobose	Doungsa-ard et al. 2015

Table 3 (*continued*)

Rust Species	Host	Size of Teliospores (µm)	Width of Germ Pore	Number of Striae	Colour and Shape of the Sori	Reference
U. tepperianum	*A. coolgardiensis*	(19) 22-24 (26) × (15) 17-20 (22)	4-5.5	33-45	Cinnamon brown; depressed globose or subglobose	Doungsa-ard et al. 2015
U. tepperianum	*A. sibina*	(16) 19-22 (23) × (15) 16-18 (19)	4-5.5	33-44	Cinnamon brown; depressed globose or subglobose	Doungsa-ard et al. 2015
U. tepperianum	*A. acuminata*	(17) 19-23 (26) × (12) 15-18 (20)	4-5	33-44	Cinnamon brown; depressed globose or subglobose	Doungsa-ard et al. 2015
U. woodii	*P. lophantha* subsp. *lophantha*	(15.5)17-20 (21) × (21) 22-25(27)	3.5 - 5.5	22 - 35	Cinnamon brown; depressed globose or subglobose	Doungsa-ard et al. 2018

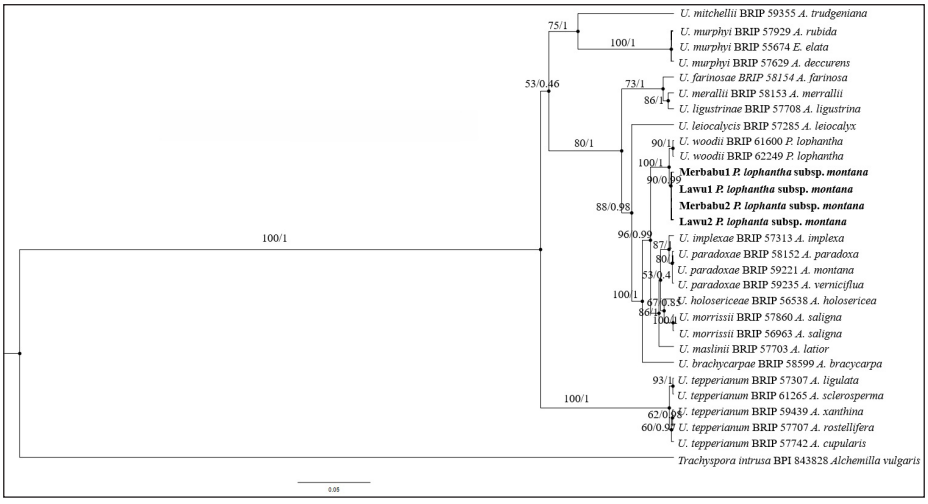

Figure 3. Phylogenetic tree using maximum-likelihood (ML) and Bayesian inference (BI) analysis. The number in each branch indicates the ML bootstrap value (MLBP) and Bayesian posterior probability values (BPP) based on ML analysis

as an additional specimen). Doungsa-ard et al. (2018) also noted that the taxonomic relationship between the rust fungus previously reported as *U. tepperianum sensu lato* on *P. lophantha* subsp. *montana* in Java and *U. woodii* remained unresolved. The present phylogenetic analysis provides a close relationship between the rust specimens collected from *P. lophantha* subsp. *montana* in Java and *U. woodii*.

We conducted phylogenetic analyses using a combined dataset of the internal transcribed spacer (ITS) region and the large subunit (LSU) rDNA, generated from an alignment of 28 ingroup and one outgroup sequences. We selected the ITS-LSU rDNA regions because they are widely used to infer phylogenetic relationships among closely related rust fungi (Aime et al., 2017; Doungsa-ard et al., 2018).

This research found that taxonomic identification varies between morphological data and multilocus molecular analysis. Based on the morphology of the *Uromycladium* rust fungus on *P. lophantha* subsp. *montana* was significantly different from those of *U. falcatarium* and *U. woodii*. The teliospores were larger, had a wider germ pore, and had a broader range of striae than those of *U. falcatarium* and *U. woodii*. The striae were similar to those of other *U. tepperianum*, except that the germ pore was larger and wider. Based on teliospore morphology, rust fungi on *P. lophantha* subsp. *montana* were identified as more closely related to *U. tepperianum*.

These size differences in *U. woodii* may be strongly related to environmental conditions. According to Loquez et al. (2025), Uromycladium teliospores increased significantly in length and width with elevation, with the largest dimensions at high elevations. Because the *U. woodii* collected by Doungsa-ard et al. (2018) came from a coastal area with higher

temperatures and lower relative humidity, whereas we collected the teliospore inoculum above 1,500 m a.s.l., where relative humidity is higher and temperatures lower, the teliospores tended to be larger. Teliospore size can vary with hydration state at the time of measurement, and lower temperatures often slow development but yield larger final cell sizes. Therefore, molecular identification based on LSU rDNA sequence data provided the most reliable species-level discrimination in this study.

Several studies have examined cross-inoculation of Uromycladium among hosts. Inoculation experiments on Acacia gall rust showed that Acacias were more susceptible to *U. tepperianum* isolates collected from the same *Acacia* species than to isolates from other species (Morris, 1987). Another study reported that teliospores collected from *F. moluccana* infected *P. lophantha* subsp. *montana* and vice versa (Rahayu, 2014). These findings suggest that *Uromycladium* diversity may include host-specific isolates that shape its genetic diversity. Thus, the occurrence of *U. woodii* in *P. lophantha* in Indonesia, which naturally grows in the highlands, poses a risk to other Fabaceae members. This study's findings illustrate Uromycladium diversity in Indonesia and raise awareness of gall rust disease, particularly at high elevations where *F. moluccana* grows near *P. lophantha*, which exhibits gall rust symptoms. Accordingly, at least three Uromycladium species have been recorded in Indonesia: *U. tepperianum*, *U. falcatarium*, and *U. woodii*.

CONCLUSION

The findings of this research showed that different identifications were based on morphological data and multilocus molecular analysis. However, molecular identification based on the combined ITS and LSU rDNA regions provided the most compelling evidence in this study. Based on multilocus analysis of the LSU and ITS regions, the causal agent of gall rust disease on *P. lophantha* subsp. *montana* was closely related to *U. woodii*. Therefore, at least three species of Uromycladium have been reported in Indonesia: *U. tepperianum*, *U. falcatarium*, and *U. woodii*.

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REFERENCES

Aime, M. C. (2006). Toward resolving family-level relationships in rust fungi (Uredinales). *Mycoscience*, *47*(3), 112-122. https://doi.org/10.1007/S10267-006-0281-0

Aime, M. C., McTaggart, A. R., Mondo, S. J., & Duplessis, S. (2017). Phylogenetics and phylogenomics of rust fungi. *Advances in Genetics, 100*, 267-307. https://doi.org/10.1016/bs.adgen.2017.09.011

Altschul, S. F., Gish, W., Miller, W., Myers, E. W., & Lipman, D. J. (1990). Basic local alignment search tool. *Journal of molecular biology*, *215*(3), 403-410. https://doi.org/10.1016/S0022-2836(05)80360-2

Anonymous. (1993). *Gall rust disease on F. moluccana plantation in Sabah, Malaysia* [Unpublished annual report]. SSB Company.

Boedijn, K. B. (1959). The Uredinales of Indonesia. *Nova Hedwigia, 1*, 463-496.

Bouckaert, R., Vaughan, T. G., Barido-Sottani, J., Duchêne, S., Fourment, M., Gavryushkina, A., Heled, J., Jones, G., Kühnert, D., De Maio, N., Matschiner, M., Mendes, F. K., Müller, N. F., Ogilvie, H. A., du Plessis, L., Popinga, A., Rambaut, A., Rasmussen, D., Siveroni, I., Suchard, M. A., Wu, C. -H., Xie, D., Zhang, C., Stadler, T., & Drummond, A. J. (2019). BEAST 2.5: An advanced software platform for Bayesian evolutionary analysis. *PLoS Computational Biology*, *15*(4), Article e1006650. https://doi.org/10.1371/journal.pcbi.1006650

Braza, R. D. (1997). Gall rust disease of Paraserianthes falcataria in the Philippines. *Forest, Farm, and Community Tree Research Reports, 2,* 61-62.

Brown, G. K., Murphy, D. J., & Ladiges, P. Y. (2011). Relationships of the Australo-Malesian genus *Paraserianthes* (Mimosoideae: Leguminosae) identifies the sister group of *Acacia sensu stricto* and two biogeographical tracks. *Cladistics*, *27*(4), 380-390. https://doi.org/10.1111/j.1096-0031.2011.00349.x

Corcoran, P., Dettman, J. R., Sun, Y., Luque, E. M., Corrochano, L. M., Taylor, J. W., Lascoux, M., & Johannesson, H. (2014). A global multilocus analysis of the model fungus Neurospora reveals a single recent origin of a novel genetic system. *Molecular Phylogenetics and Evolution*, *78*, 136-147. https://doi.org/10.1016/j.ympev.2014.05.007

Cummins, G. B., & Hiratsuka, Y. (1983). *Illustrated Genera of Rust Fungi* (Rev. ed.). The American Phytopathological Society Press.

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. https://doi.org/10.3767/persoonia.2018.40.09

Doungsa-ard, C., McTaggart, A. R., Geering, A. D. W., Dalisay, T. U., Ray, J., & Shivas, R. G. (2015). *Uromycladium falcatarium* sp. nov., the cause of gall rust on *Paraserianthes falcataria* in south-east Asia. *Australasian Plant Pathology*, *44*, 25-30. https://doi.org/10.1007/s13313-014-0301-z

Felsenstein, J. (1985). Confidence Limits on Phylogenies: An approach using the bootstrap. *Evolution*, *39*(4), 783-791. https://doi.org/10.2307/2408678

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*(1), 21-33. https://doi.org/10.1111/aab.12677

Gardes, M., & Bruns, T. D. (1993). ITS primers with enhanced specificity for basidiomycetes - application to the identification of mycorrhizae and rusts. *Molecular Ecology*, *2*(2), 113-118. https://doi.org/10.1111/j.1365-294X.1993.tb00005.x

Hall, T., Biosciences, I. and Carlsbad, C. (2011). BioEdit: An important software for molecular biology. GERF Bulletin of Biosciences, 2, 60-61.

Hoff, R. J. (1986). *Susceptibility of pine populations to western gall rust—central Idaho* (Research Note INT-354). U.S. Department of Agriculture, Forest Service, Intermountain Research Station.

Impson, F. A. C., Kleinjan, C. A., Hoffmann, J. H., Post, J. A., & Wood, A. R. (2011). Biological control of Australian *Acacia* species and *Paraserianthes lophantha* (Willd.) Nielsen (Mimosaceae) in South Africa. *African Entomology, 19*(2), 186-207. https://doi.org/10.4001/003.019.0210

Kumar, S., Stecher, G., Suleski, M., Sanderford, M., Sharma, S., & Tamura, K. (2024). MEGA12: Molecular evolutionary genetic analysis version 12 for adaptive and green computing. *Molecular Biology and Evolution*, *41*(12), Article msae263. https://doi.org/10.1093/molbev/msae263

Loquez, M. O., Amper, C. D., Tulod, A. M., & Gilbero, D. M. (2025). Teliospore morphology characterization of *Uromycladium falcatariae* in falcata plantations at different elevations in Mindanao Island, Philippines. *Biodiversitas Journal of Biological Diversity, 26*(1), 296-305. https://doi.org/10.13057/biodiv/d260129

Magnus, P. W. (1892). Zur Kenntniss der Verbreitung einiger pilze [On the knowledge of the distribution of some fungi]. *Bericht der Deutschen Botanischen Gesellschaft, 10*, 195-200. https://www.biodiversitylibrary.org/page/36160175

McAlpine, D. (1906). *The rusts of Australia: Their structure, nature, and classification.* Government Printer. https://doi.org/10.5962/bhl.title.31958

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. https://doi.org/10.1007/s13313-015-0381-4

McTaggart, A. R., Shivas, R. G., van der Nest, M. A., Roux, J., Wingfield, B. D., & Wingfield, M. J. (2016). Host jumps shaped the diversity of extant rust fungi (Pucciniales). *New Phytologist*, *209*(3), 1149-1158. https://doi.org/10.1111/nph.13686

Morris, M. J. (1987). Biology of the Acacia gall rust, *Uromycladium tepperianum*. *Plant Pathology, 36*, 100-106. https://doi.org/10.1111/j.1365-3059.1987.tb02183.x

Morris, M. J. (1991). The use of plant pathogens for biological weed control in South Africa. *Agriculture, Ecosystems & Environment*, *37*(1-3), 239-255. https://doi.org/10.1016/0167-8809(91)90153-O

Nielsen, I., Guinet, P., & Baretta-Kuipers, T. (1983). Studies in the Malesian, Australian and Pacific Ingeae (*Leguminosae-Mimosoideae*): The genera Archidendropsis, Wallaceodendron, Paraserianthes, Pararchidendron and Serianthes (Part I). *Bulletin du Muséum National d'Histoire Naturelle, Section B, Adansonia, 5*(3), 303-329. https://doi.org/10.5962/p.276287

Old, K. M., & Cristovao, C. D. S. (2003). A rust epidemic of the coffee shade tree (*Paraserianthes falcataria*) in East Timor. In H. da Costa, C. Piggin, C. J. da Cruz, & J. J. Fox (Eds.), *Agriculture: New directions for a new nation: East Timor (Timor-Leste)* (pp. 139-145). Australian Centre for International Agricultural Research.

Pham, N. Q., Wingfield, M. J., Marincowitz, S., Tanga, A. A., Tiki, K. R., Kassie, W. B., Hurley, B. P., Germishuizen, I., Lawson, S. A., Healey, M. A., & Wondafrash, M. (2024). First report of the wattle rust pathogen, *Uromycladium acaciae* (Raveneliaceae, Pucciniales) in Ethiopia. *Forestry: An International Journal of Forest Research*, *97*(2), 319-326. https://doi.org/10.1093/forestry/cpad040

Rahayu, S. (2014). *Strategi pengelolaan penyakit tanaman hutan di Indonesia: Penyakit karat tumor pada tanaman sengon (Falcataria moluccana)* [Forest plant disease management strategies in Indonesia: Gall rust disease on sengon (*Falcataria moluccana*)]. Gadjah Mada University Press.

Rahayu, S., Lee, S. S., & Shukor, N. A. A. (2010). *Uromycladium tepperianum*, the gall rust fungus from *Falcataria moluccana* in Malaysia and Indonesia. *Mycoscience*, *51*(2), 149-153. https://doi.org/10.1007/S10267-009-0022-2

Rahayu, S., See, L. S., Shukor, N. A. A., & Saleh, G. (2018). Environmental factors related to gall rust disease development on *Falcataria moluccana* (Miq.) Barneby & J. W. Grimes at Brumas Estate, Tawau, Sabah, Malaysia. *Applied Ecology and Environmental Research, 16*(6), 7485–7499. https://doi.org/10.15666/aeer/1606_74857499

Rambaut, A. (2009). FigTree v1.3.1: *Tree figure drawing tool*. Institute of Evolutionary Biology, University of Edinburgh.

Ryley, M. J. (2024). Wattle gall—the quintessential Australian plant disease. *Historical Records of Australian Science*, *35*(2), 116-129. https://doi.org/10.1071/HR230066

van der Merwe, M. M., Walker, J., Ericson, L., & Burdon, J. J. (2008). Coevolution with higher taxonomic host groups within the Puccinia/Uromyces rust lineage obscured by host jumps. *Mycological Research*, *112*(12), 1387-1408. https://doi.org/10.1016/j.mycres.2008.06.027

Vilgalys, R., & Hester, M. (1990). Rapid genetic identification and mapping of enzymatically amplified ribosomal DNA from several *Cryptococcus species*. *Journal of Bacteriology*, *172*(8), 4238-4246. https://doi.org/10.1128/jb.172.8.4238-4246.1990

Wingßeld, B. D., Ericson, L., Szaro, T., & Burdor, J. J. (2004). Phylogenetic patterns in the Uredinales. *Australasian Plant Pathology*, *33*, 327-335. https://doi.org/10.1071/AP04020

Wood, A. R., & Morris, M. J. (2007). Impact of the gall-forming rust fungus *Uromycladium tepperianum* on the invasive tree *Acacia saligna* in South Africa: 15 years of monitoring. *Biol. Control*, *41*(1), 68-77. https://doi.org/10.1016/j.biocontrol.2006.12.018

Zhafarina, S., Wibowo, A., & Widiastuti, A. (2021). Multi-genetic analysis of *Colletotrichum* spp. associated with postharvest disease of fruits anthracnose in Special Region of Yogyakarta, Indonesia. *Pakistan Journal of Biological Sciences*, *24*(1), 53-65. https://doi.org/10.3923/pjbs.2021.53.65