

Record of root endophytic fungi associated with Pteridophytes from Gunung Tukung Gede Nature Reserve, Indonesia

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Abstract

The root endophytic fungi coexist in a mutualistic relationship within their host plants. This study aimed to analyse the complex relationships between these plants and their fungi, revealing insight into the abundance of microbial communities living in the root systems of distinct Pteridophyte species inside this protected environment in the Gunung Tukung Gede Nature Reserve, Indonesia, and explore the potential functional roles of these fungal communities in the ecosystem. The research was carried out between July 2022 and January 2023. The roots of Pteridophyta were sampled randomly from three stations. The environmental variables examined were temperature, soil pH, moisture, and light intensity. The fungi were isolated and cultured in the PDA medium, which could then be identified. There were eight genera of root endophytic fungi: *Aspergillus* sp., *Trichoderma* sp., *Phoma* sp., *Phialophora* sp., *Penicillium* sp., *Paecilomyces* sp., *Humicola* sp., and *Fusarium* sp. In the designated conservation zones, this mutualistic connection contributes to the preservation of ecosystem health and biodiversity by being essential to Pteridophyta's resilience and intake of nutrients.

Keywords: conservation zone, diversity, ecosystem, mutualistic.

Introduction

Pteridophytes are essential to the ecology and growth of terrestrial ecosystems. The plants are seedless taxa having a vascular system, megaphylls, and spore dissemination as their reproductive mechanism (Aros-Mualin et al. 2021). Particular-

ly in areas with high humidity, like tropical rainforests, Pteridophytes are frequently seen in Indonesian woods. These ancient plants significantly impact biodiversity and ecological stability due to their unique reproductive strategies and widespread.

Gunung Tukung Gede Nature Reserve (GTGNR) is a forest area with an altitude

of 125–750 m with climate type A (tropical) and an average rainfall of $\pm 2151 \text{ mm} \cdot \text{yr}^{-1}$ (Sunarti 2011). Many Pteridophyte plants are found in the area. Information on biodiversity in the ecosystem at GTGNR that has been inventoried, such as flora diversity, as in the research of Djarwaningsih (2011), who managed to find 167 species of plants and fauna diversity, including mammals, birds, and reptilians. Khastini et al. (2021) reported on the diversity of entomopathogenic fungi from this area, but information on the diversity of root endophytic fungi is still lacking.

Various symbiotic microorganisms, known as root endophytic fungi, have attracted increasing attention in recent years as one of the many variables affecting the health and adaptation of Pteridophytes. These fungi serve essential ecological functions within forest ecosystems and act as a source of germplasm with potential applications in food and medicine (Suraida et al. 2013). Root endophytic fungi can colonise plant root tissues but do not inhibit the growth and development of their host plants (Sun et al. 2022). They can increase the growth of their host plants by optimising the absorption of nutrients through the structures they form or by producing growth-regulating hormones (Khastini 2018). They can produce secondary metabolite compounds that can be isolated into antimicrobial compounds and kill or inhibit pathogens (Harahap et al. 2017). Root endophytic fungi live in the roots and have the potential to influence plant communities (Rillig et al. 2014). Sporophytes and gametophytes of Pteridophytes form symbiotic associations with them (Muthukumar and Prabha 2012). This mutualistic symbiosis is essential for plant establishment, survival, and health. The host plant obtains nutrients from the fungi and provides a carbon

supply to them (Muthukumar et al. 2014). One of the advantages that fungal colonisation confers upon the host plant is the enzymatic degradation of organic nutrient sources in the soils and the subsequent transfer of the resulting products to plants. This is because root endophytic fungi are widely distributed in the rhizosphere or rhizoplane of plants growing in soils. They can increase drought tolerance resistance to soil pathogens, increase soil aggregate stability, and increase plant growth and yield so that it can be developed as a biological fertilizer (Watts et al 2023). Their role is quite important, so information on the diversity of microorganisms is needed.

Microhabitats play a critical role in creating the ecology of Pteridophyte communities in GTGNR. A complex interaction of variables, such as differences in the species mix of plants, the texture of the soil, and the presence of nearby flora, distinguishes these microenvironments. Pteridophytes flourish in these varied and dynamic environments, exhibiting extraordinary resilience and ecological value. Notably, they play a crucial role in preserving the stability of the ecosystem inside the reserve due to their capacity to reduce water loss, primarily through the activity of their sporophytes. Numerous adaptive traits have developed in these plants, including waxy cuticles on fronds that ward off water vapor, sensitive stomata that respond to variations in soil and air temperatures, and leaf water potentials that are influenced (Krieg and Chambers 2022). In contrast, gametophytes rely on their capacity to endure and recover from water fluctuation since they lack xylem, effective waxy cuticles, or stomata.

This study explores the variety of root endophytic fungi in Pteridophytes to uncover what is beneath the forest floor of

Gunung Tukung Gede Nature Reserve. It aims to analyse the complex relationships between these plants and their fungi, revealing insight into the abundance of microbial communities living in the root systems of distinct Pteridophyte species inside this protected environment. This study adds to the growing knowledge on plant-fungus interactions by concentrating on a particular and comparatively unexplored ecological niche of the Pteridophytes root endosphere in a biodiverse hotspot.

Object and Method

The object of the study

Root endophytic fungi were isolated from the Pteridophyta plant roots in GTGNR as illustrated in Figure 1. The sampling location was divided into three research stations, I and II in the eastern area while Station III was in the western area. It represents distinct conditions: open vegetation, closed vegetation, and near settlements condition. Samples were collected using random sampling in the plot. Plots were determined based on a plot size of 5×5 m.

Based on observation, Station I (Fig. 2A) is a typical open vegetation area, was dominated by *Pleocnemia irregularis* (C. Presl) Holttum with medium density. There are also three other species of Pteridophyta found in the station, e.g., *Davallia denticulate* (Burm. f.) Kuhn, *Cyclosorus terminans* (Wall. ex Hook.) Shing, *Christella* sp. The soil texture is loamy sand. *Gnetum gnemon* L. trees planted by residents surround this station but are not intertwined, so the Pteridophytes underneath get high sunlight.

Station II, with closed vegetation

(Fig. 2B), is dominated by *Stenochlaena palustris* (Burm. f.) Bedd. plants with a high density. *Pleocnemia irregularis* (C. Presl) Holtt. and *Adiantum tenerum* Sw. are two other species found in this station. The soil texture is clayey loam. This station is surrounded by *Pterospermum javanicum* Jungh., which is intertwined so that the Pteridophyte below are blocked from sunlight.

Station III is located near residential areas (Fig. 2C) dominated by *Christella dentata* (Forssk.) Brownsey & Jermy plants with moderate density. The soil texture is sandy loam. This station is surrounded by *Shorea* sp. that is not intertwined so that the Pteridophytes underneath are blocked from sunlight.

Environmental factors

Environmental parameters such as temperature, relative humidity and light intensity at the sampling sites are measured and documented using Fluke 971 Temperature Humidity Meter and Digital Lux Meter Photometer Smart Sensor AS823. The environmental parameters can be viewed in Table 1.

The average temperature of the coldest month in the GTGNR is typically around 15 °C, while the average temperature of the hottest month is around 25 °C. The absolute minimum temperature recorded in the area is 10 °C, and the absolute maximum temperature is 30 °C. The average annual temperature is approximately 20 °C. Regarding precipitation, the average monthly precipitation varies throughout the year. The highest monthly average precipitation is typically around 300 mm, while the lowest monthly average precipitation is around 50 mm. The total annual precipitation is approximately 1500 mm.

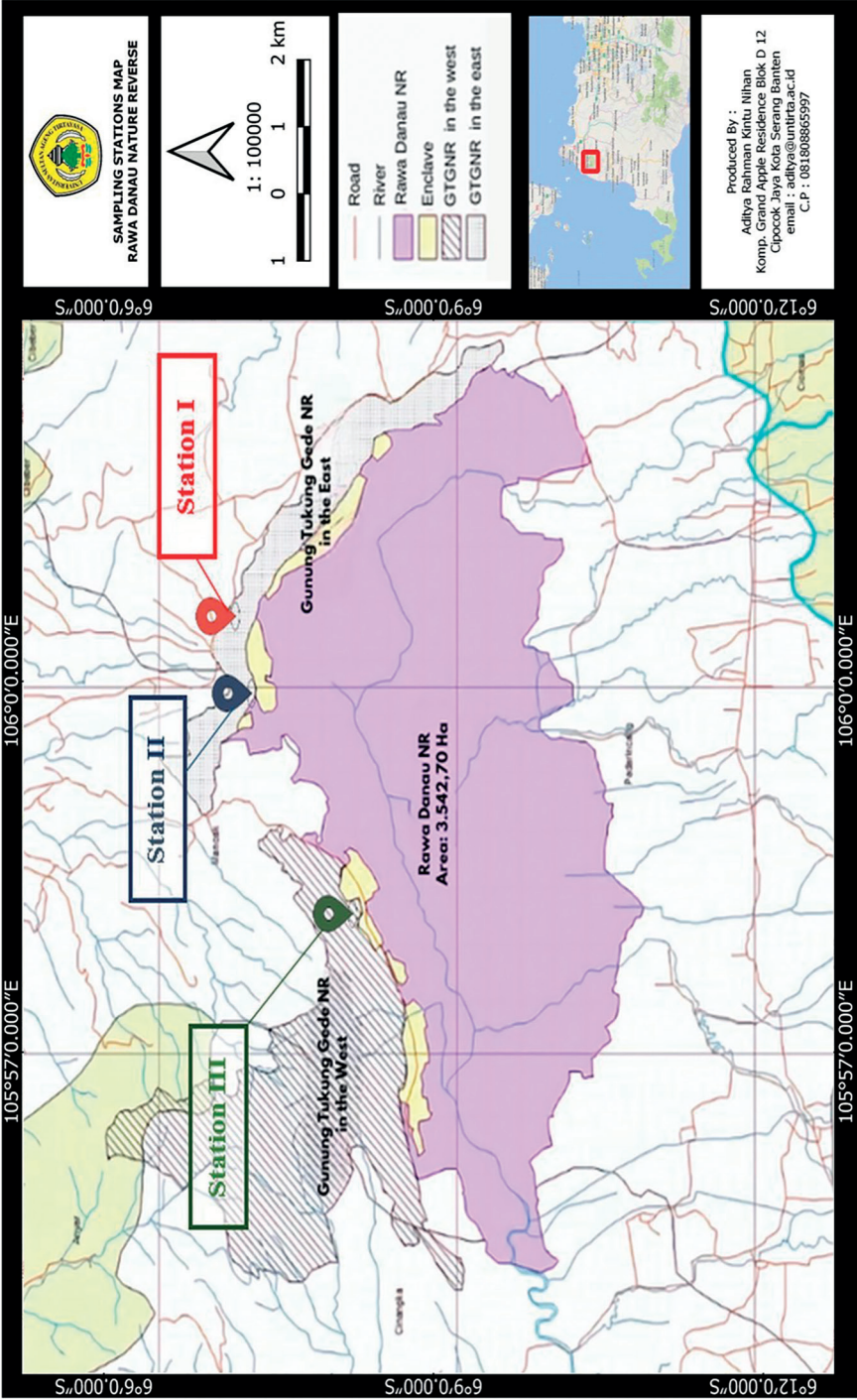


Fig. 1. Sampling location in the GTGNR.



Fig. 2. Sampling site condition: A – Station I, B – Station II, C – Station III.

Table 1. Condition of environmental factor of the sampling station.

Sta- tion	GPS coordinates	Tempera- ture, °C	Relative humidity, %	Light intensity, lux	Soil type	Soil pH
I	6°8'17.16" S, 106°0'18.84" E	28	51	809.6·10 ²	Ultisol	3.94
II	6°8'8.22" S, 106°0'36.48" E	26	58	10.6·10 ²	Ultisol	3.92
III	6°8'38.4" S, 105°58'52.32" E	27	48	29.8·10 ²	Latosol	6

Isolation and colonisation process of root endophytic fungi in Pteridophyta plant

Isolation of root endophytic fungi was carried out using a modification of the surface sterilisation method of Khastini et al. (2022). About 30 root samples of Pteridophyte were collected in each of the stations. They were washed with tap water to remove debris and then cut into approximately 1 cm. The segment root samples were washed with 70% alcohol for 1 min, followed by a solution of NaOCl 0.1% for 15 min, and rinsed three times using sterile water using a vortex mixer. The sterile root segments were air-dried overnight and then planted into PDA, pH 6.0. If mycelia growth was observed on PDA, they were then transferred to a new PDA. Single spore isolation is commonly performed to obtain a pure culture of the fungal isolate.

The pure isolates obtained were observed for their morphological characteristics macroscopically and microscopically using a compound microscope (Leica DM500). Macroscopic observations were

based on colony colour, colony dorsal colour, colony surface, fungal colony growth diameter, and concentric circles. Microscopic observations can be seen from the presence or absence of septa on hyphae, hyphal pigmentation, spore shape and ornamentation, and spore stalk shape and ornamentation (Khastini 2016). All isolates were identified based on their morphological characteristics (Barnett and Hunter 1998, Seifert and Gams 2011) as well as compared with well-identified living cultures from the collection of Biology Education Laboratory, UNTIRTA.

A methodical approach was used to investigate fungal growth within the root, followed Khastini et al. (2012). Pteridophyte's root system was carefully removed from the shoot. To get rid of any unnecessary items such as soil particles attached to it, it was first cleaned. After 20 min of clearing in 10% KOH at 80 °C, the roots were neutralised in 1% HCl for a further 20 min at the same temperature. The roots were dyed with a solution of 0.05% cotton blue in 50% acetic vinegar to aid in the visibility of fungal structures. To conduct further investigation, the roots were sectioned

by a freezing microtome to a thickness of 10–25 µm mounted in phenol glycerin jelly, carefully placed on slides and examined under a light microscope (Leica DM 2500) for microscopic details.

Result and Discussion

Isolation of root endophytic fungi associated with Pteridophyta plants

The results of the identification of root endophytic fungi obtained as many as 21 isolates with 8 different genera of root endophytic fungi, these are *Aspergillus*,

Trichoderma, *Phoma*, *Phialophora*, *Penicillium*, *Paecilomyces*, *Humicola*, and *Fusarium* (Table 2).

The composition of root endophytic fungi identified at each research station varies. *Penicillium* sp. and *Paecilomyces* sp. fungi were only found at Station I. *Phialophora* sp. and *Phoma* sp. fungi were only found at Station II. *Humicola* sp., *Trichoderma* sp. and *Aspergillus niger* Tiegh. were only found at Station III. Some of these fungi were only found at one station, while others were at more than one. *Aspergillus oryzae* was found at stations I and II, and *Aspergillus fumigatus* Fresenius at stations I and III. The difference in

Table 2. Identification results of root endophytic fungi species in Pteridophytes.

Station	Species Pteridophytes	Local name	Identification of root endophytic fungi	Isolate code	
I	<i>Pleocnemia irregularis</i>	Paku andam/ Paku kapal	<i>Aspergillus oryzae</i> (Ahlburg) E. Cohn	APi1	
			<i>Aspergillus fumigatus</i> Fresenius.	APi2	
			<i>Paecilomyces</i> sp.	APi3	
			<i>Fusarium</i> sp.	APi4	
	<i>Davalia denticulate</i> (Burm. f.) Kuhn	Paku tertutup	<i>Aspergillus oryzae</i>	ADd1	
			<i>Fusarium</i> sp.	ADd2	
	<i>Cyclosorus terminans</i>	-	<i>Penicillium</i> sp.	ACT1	
	<i>Christella</i> sp.	-	<i>Aspergillus oryzae</i>	AC1	
			<i>Fusarium</i> sp.	AC1	
	II	<i>Stenochlaena palustris</i>	Paku kalakai	<i>Fusarium</i> sp.	BSp1
<i>Aspergillus oryzae</i>				BSp2	
<i>Phialophora</i> sp.				BSp3	
<i>Phoma</i> sp.				BSp4	
<i>Pleocnemia irregularis</i>		Paku andam/ Paku kapal	<i>Fusarium</i> sp.	BPi1	
<i>Adiantum tenerum</i>		Paku suplir	<i>Fusarium</i> sp.	Bat1	
III		<i>Christella dentata</i>	-	<i>Aspergillus niger</i> Tiegh.	CCd1
				<i>Aspergillus fumigatus</i> Fresenius	CCd2
	<i>Christella</i> sp.	-	<i>Trichoderma</i> sp.	CC1	
	<i>Lygodium scandens</i> L. Sw.	Paku hata/Paku ribu-ribu	<i>Humicola</i> sp.	CLs1	
	<i>Pteris multifida</i> Poir.	-	<i>Aspergillus niger</i>	CPm1	

composition is due to differences in habitat and vegetation type. The noted variations in endophytic fungal composition are caused mainly by environmental variables. The interactions between soil fertility and the ecological environment shape endophytic fungal composition. According to studies by Min et al. (2016), this impact indirectly affects the structure and composition of the endophytic fungus population.

Environmental factors can influence differences in the composition of endophytic fungi. According to Table 2, Stations I and II have the highest number of isolates, but it is higher at Station I because it has a low pH of 3.94. Station III is stated to have low diversity in fungal isolates. The condition is due to two main factors, namely low humidity and high pH compared to other stations. Low humidity at Station III can affect the diversity of endophytic fungi that can grow in the area. Many fungi, including endophytic, have specific pH ranges within which they can grow optimally. When the pH level exceeds these optimal ranges, it can inhibit their growth, leading to a lower diversity and abundance of these species in the environment. This phenomenon is well-documented in Geisen et al. (2021), where it is observed that fungi with acidic growth preferences may be less abundant in environments with high pH levels.

Humid weather plays an important role in the growth of endophytic fungi. Its ability to facilitate the spread of fungal spores through rain splashes and wind affects the diversity of endophytic fungi that can grow. In addition, comparisons between Station III and other stations show that the low humidity and high pH at Station III contribute to the low diversity in the area. Manurung et al (2022) stated that humid weather influences the growth of endophytic fungi. Pteridophyte growth is significantly influenced by temperature. Different species of

these plants thrive temperature ranges of 26–28 °C because they are susceptible to temperature changes. Most Pteridophytes grow better in moderate temperatures, while some can adapt to colder climates, while others perform better in warmer climates. Temperature has a crucial role in the growth and spread of Pteridophytes because it influences the rate of photosynthesis, the activity of enzymes, and the total metabolic activities.

The pH of the soil and its surroundings significantly affects the development of Pteridophytes. Different species have varying pH requirements, with many preferring slightly acidic to neutral soils. This preference is due to the enzymatic activities of Pteridophytes, which are often optimised under these pH conditions. For example, enzymes involved in nutrient uptake and metabolic processes are more active in slightly acidic to neutral soils, facilitating the growth and development of Pteridophytes (Mishra et al 2021). It can be seen that the pH levels at each research station varied, with Station I having a low pH of 3.94, which is likely to support a higher diversity of fungi due to its optimal pH conditions for the growth of the isolated endophytes. In contrast, Station III, which is stated to have low diversity on fungal isolates, may have a higher pH level of 6, making it less favourable for the growth of certain endophytic fungi. This observation aligns with the general understanding that pH plays a crucial role in determining the growth and dominance of these fungi.

One important aspect controlling the growth of Pteridophytes is light intensity. Although variety may not be directly impacted, it has a major impact on plant development, with Station I showing the highest plant growth due to its highest light intensity $809.6 \cdot 10^2$ lux. According to

Behera and Biswal (1990), the Pteridophyta plant is a shade plant, Pteridophyta responds to various light intensities in a somewhat complicated way. These plants can tolerate various lighting situations; however, many prefer shaded or partially shaded settings. Desiccation may result from exposure to too much light, whereas photosynthesis and general development may be hampered by inadequate light. Pteridophytes frequently flourish in the woodland understory, where plants get filtered or dappled sunlight.

Soil moisture levels significantly influence Pteridophyte development and dispersion. A somewhat regular range of soil moisture content is revealed by the soil moisture levels at stations I, II, and III, which vary from around 48 % to 58 %. Since they lack the drought-resistant characteristics in certain higher plants, most Pteridophytes depend on regular soil moisture to survive. The amount of water in the soil impacts the plant's capacity to absorb nutrients, exert turgor pressure, and stay well hydrated. For the growth of fern sporophytes, which depend on water to distribute their reproductive spores, adequate soil moisture is essential. It is supported by Qian et al. (2022) that the Pteridophytes cannot effectively manage transpiratory water loss.

The study's findings should be understood in the context of the complex interactions between pH, temperature, and other environmental factors that influence the growth and distribution of fungi in GT-GNR. It is well recognised that environmental elements, including soil moisture, nutrient availability, and habitat heterogeneity, significantly impact fungal diversity in addition to pH and temperature. For instance, research conducted in cross coastal and inland Santa Barbara County, California by Mordecai et al. (2012)

discovered that soil moisture significantly impacted the fungus population's make-up. Similarly, Naranjo-Ortiz and Gabaldón (2019) discovered that in a temperate forest environment, the availability of nutrients and habitat heterogeneity had a significant role in determining the makeup of the fungal population. In order to fully understand the study's conclusions, it is essential to consider the holistic interactions between pH, temperature, and other environmental variables. Comprehending these intricate interplays is crucial in clarifying the factors that influence the fungal composition of the reserve and influencing conservation and management tactics meant to maintain its fungal biodiversity. We may obtain a more comprehensive picture of the ecological processes influencing fungal communities in GTGNR by incorporating a variety of environmental characteristics into our research.

Among the isolated endophytic fungi, a variety of colony morphologies have been reported (Fig. 3), ranging from *Aspergillus*'s fluffy, cotton-like texture to *Penicillium*'s unique powdery look. Initial insights into the possible functional variety and ecological functions of the detected endophytic fungus linked with the Pteridophyte can be gained from these apparent differences in colony formation. The physical variations point to adaptations to different ecological niches and substrate preferences, which raises the possibility of a variety of roles these fungi may play in the endosphere of the plant.

The isolation of endophytic fungus from Pteridophyta plant roots has revealed an interesting variety, particularly the existence of three different species of *Aspergillus*, e.g. *A. oryzae*, *A. niger*, *A. fumigatus* Fresenius (Fig. 3A-C). Comprehending the different characteristics of various *Aspergillus* species is crucial for

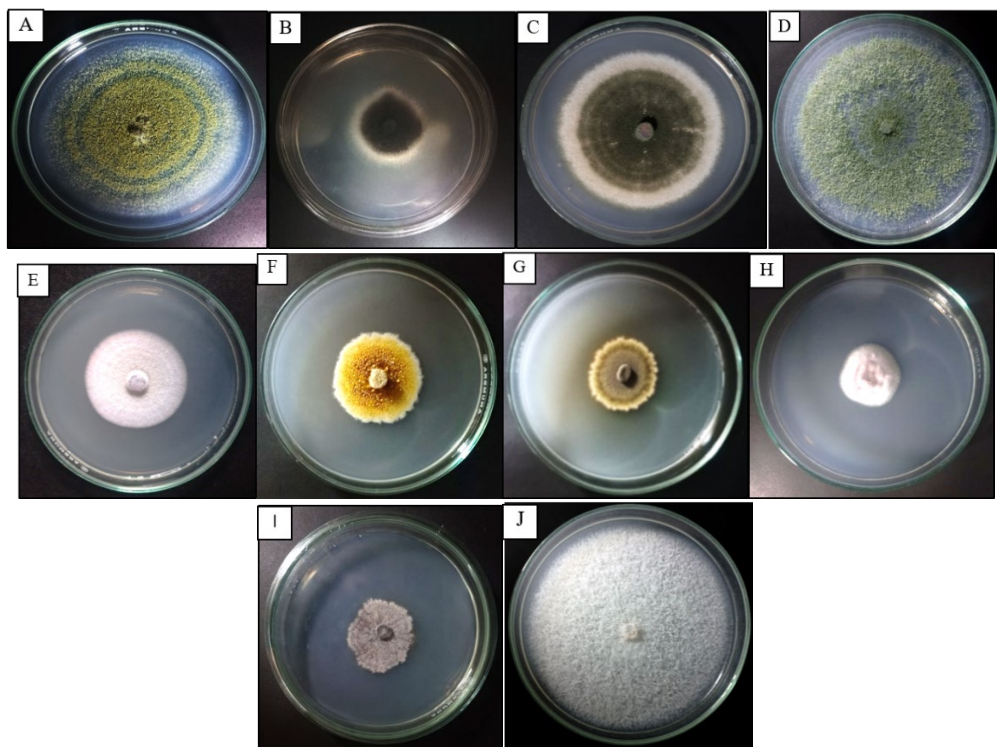


Fig. 3. Macroscopic picture of root endophytic fungi: **A** – *Aspergillus oryzae*, **B** – *A. niger*, **C** – *A. fumigatus*, **D** – *Trichoderma* sp., **E** – *Phoma* sp., **F** – *Paecilomyces* sp., **G** – *Penicillium* sp., **H** – *Phialophora* sp., **I** – *Humicola* sp., **J** – *Fusarium* sp.

the objective of biodiversity analysis and possible agricultural applications. *A. oryzae* is one strain of *Aspergillus* that has been shown to have beneficial impacts on plant growth and protection. This is consistent with the results of Wen et al. (2022), who found and effectively isolated *A. oryzae* from the *Paris polyphylla* Sm. rhizome in Dali. Furthermore, Galeano et al. (2021) revealed that the endophytic fungus *A. niger* 9-P showed activity associated with the development of plant growth-promoting (PGP) features when it was isolated from native fodder grass in the Pantanal of Nhecolândia area. Study conducted by Jiang et al. (2022) which revealed the presence of the endophytic fungus *A. fumigatus*, is another important

addition in providing evidence of possible symbiotic connections.

As for the *Trichoderma* (Fig. 3D) the green colonies with a granular surface, concentric circles, no exudate drops, and yellowish green colony back colour. These colonies grow rapidly, first white, and then greenish. According to Tseng et al. (2020), *Trichoderma* sp. endophytic strain enhances host growth and protects against pathogen attack. Our comprehensive research shows a noteworthy correlation between Pteridophyta plants and these fungi, adding to the growing information about endophytic fungi.

Furthermore, the complex relationship that exists between Pteridophyta plants and the fungal communities that are as-

sociated with them is strengthened by the presence of a variety of filamentous ascomycetes in the roots of these plants, such as *Paecilomyces* sp., *Penicillium* sp., *Phialophora* sp., *Humicola* sp., and different species of *Fusarium*. Tseng et al. (2020) highlight the dynamic character of interactions between fungal species and Pteridophyta plants, suggesting that the two may have a mutualistic connection. Nejad et al.'s (2022) finding of *Phoma* sp. in vascular tissue adds to our knowledge by illuminating other connections between *Phoma* and Pteridophyta species.

All of these study results make a substantial contribution to understanding the intricate connections that exist between endophytic fungus and Pteridophyta plants. There are developing practical uses for this information, which goes beyond taxonomic considerations. As Musa et al. (2023) have shown, *Paecilomyces lilacinus*, for example, may reduce heavy metal stress, giving host plants a useful benefit in harsh environmental settings. According to Toghueo and Boyom (2020), endophytic *Penicillium* sp. is highly adaptable, as evidenced by its capacity to flourish in a various ecological niche and

protect host plants from a range of stressors. Furthermore, as shown by Almeida et al. (2020), ecological activities of *Phialophora* sp. are increased by its potential as a bio-control agent against fungi linked to plant illnesses. Together, these many discoveries deepen our comprehension of the complex.

Colonisation process of root endophytic fungi associated with Pteridophyta plants

Based on the root section, it can be seen that the endophytic fungi will colonise the root cortex area and develop the fungal structure inside. Based on observation, several identified endophytic fungal structures, including conidiophores, vesicles, conidia, and microsclerotium, are presented in Figure 4. Colonising root endophytic fungi in the cortical area of plant roots involves several stages, including recognition, attraction, penetration, and the emergence of a mutualistic relationship. This symbiotic relationship benefits both the fungus and the plant; the fungus aids in nutrient uptake and stress tolerance, and the plant provides sugars and

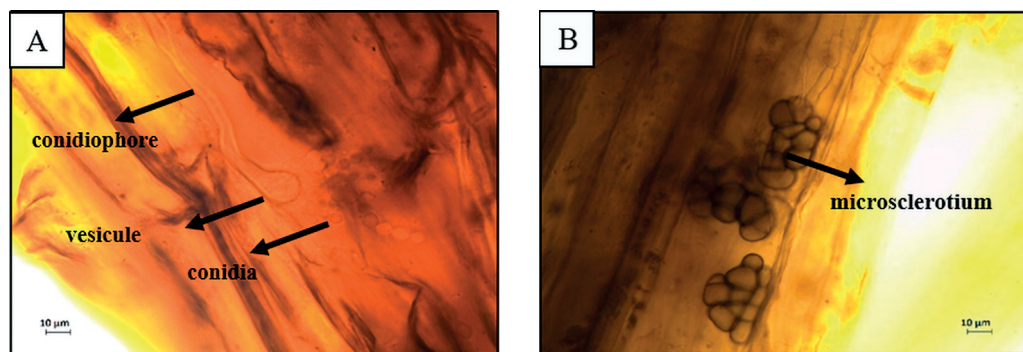


Fig. 4. The colonisation structure of endophytic fungi in the Pteridophyta root: **A** – asexual reproduction structure, **B** – microsclerotium as a specialised structure within the root cell (photo by F. I. Nurunnisa).

organic molecules (Alam et al. 2021). This intricate interaction is necessary for the environment's long-term viability and the health of plants.

Various fungi, such as arbuscular mycorrhizae, epiphytic lichens and microfungi, pathogens, saprotrophs, and pathogens are reported to be associated with the Pteridophyte (Del Olmo-Ruiz and Arnold 2014). The colonisation of Pteridophyte roots by endophytic fungi is closely related to the development of these organisms. The Pteridophytes and the root endophytic fungus have a favorable connection. The fungus colonised the plant's root tissues in this symbiotic relationship, expanding their hyphae into the surrounding soil. This colonisation significantly impacts Pteridophyte development in many ways.

Pteridophytes play a critical role in maintaining biodiversity and sustaining various ecosystems, as demonstrated by these environmentally relevant interactions between Pteridophyta plants and root-endophytic fungus, especially in protected areas. By assisting in nutrient absorption, particularly for phosphorus and micronutrients, which are frequently in short supply, the association helps Pteridophyte adapt to various environmental circumstances. By making it easier for Pteridophyta species to get nutrients, fungi aid in their recovery from disturbances and promote their growth in nutrient-poor soils. It becomes essential to maintain this mutualistic connection in conservation zones to preserve the general health and ecological balance of these crucial ecosystems and the survival and vigor of Pteridophyte populations. Conservation initiatives aimed at protecting Pteridophyta and their fungal partners are essential for these plants' well-being and preserving biodiversity in these unique environments.

Conclusion

We conclude that the important role of the relationship between Pteridophyta and root-endophytic fungi is very closely related to environmental factors in the Gunung Tukung Gede Nature Reserve. Pteridophytes serve an important role, highlighting ecological significance and adaptable features. In conclusion, research on endophytic fungi isolated from Pteridophyta plant roots has given us a thorough understanding of the complex interactions within these ecosystems. Different *Aspergillus* species have been identified, and the presence of filamentous ascomycetes such as *Paecilomyces* sp., *Penicillium* sp., *Phialophora* sp., *Humicola* sp., and different *Fusarium* species highlights how dynamic and intricate the relationships between fungus and Pteridophyta plants are. These discoveries deepen the knowledge of the complex relationships among endophytic fungal communities. Those endophytic fungi also provide insights into ecological dynamics and have potential implications in plant resilience and environmental management, extending beyond the field of taxonomy.

References

- ALAM B., LǏ J., GĚ Q., KHAN M.A., GÖNG J., MEHMOOD S., YUÁN Y., GÖNG W. 2021. Endophytic Fungi: From Symbiosis to Secondary Metabolite Communications or Vice Versa? *Frontiers in plant science* 12, 791033. 24 p. <https://doi.org/10.3389/fpls.2021.791033>
- ALMEIDA A.B., CONCAS J., CAMPOS M.D., MATERATSKI P., VARANDA C., PATANITA M., MUROLO S., ROMANAZZI G., FÉLIX M.D.R. 2020. Endophytic Fungi as Potential Biological Control Agents against Grapevine Trunk Diseases in Alentejo Region. *Biology*

- 9(12), 420. 23 p. <https://doi.org/10.3390/biology9120420>
- AROS-MUALIN D., NOBEN S., KARGER D.N., CARVAJAL-HERNÁNDEZ C.I., SALAZAR L., HERNÁNDEZ-ROJAS A., KLUGE J., SUNDUE M.A., LEHNERT M., QUANDT D., KESSLER M. 2021. Functional Diversity in Ferns is Driven by Species Richness Rather Than by Environmental Constraints. *Frontiers in plant science* 11, 615723. 11 p. <https://doi.org/10.3389/fpls.2020.615723>
- BARNETT H.L., HUNTER B.B. 1998. *Illustrated Genera of Imperfect Fungi*. 4th ed. USA: Prentice-Hall Inc. 218 p.
- BEHERA Y.N., BISWAL B. 1990. Leaf senescence in fern: effect of duration, intensity and quality of light. *Environmental and experimental botany* 30(2): 181–186. [https://doi.org/10.1016/0098-8472\(90\)90063-A](https://doi.org/10.1016/0098-8472(90)90063-A)
- DEL OLMO-RUIZ M., ARNOLD A.E. 2014. Interannual variation and host affiliations of endophytic fungi associated with ferns at La Selva, Costa Rica. *Mycologia* 106(1): 8–21. <https://doi.org/10.3852/13-098>
- DJARWANINGSIH T. 2011. Flora diversity in the Cikolelet village area – Tukung Gede mountain natural reserve, Serang – Banten [Keragaman flora di wilayah desa Cikolelet – Cagar Alam Gunung Tukung Gede, Serang – Banten]. Special Edition of the Biological Research Periodical [Berk. Penel. Hayati Edisi Khusus] 5A: 15–20 (in Indonesian). <https://berkalahayati.org/files/journals/1/articles/251/submission/251-785-1-SM.pdf>
- GALEANO R.M.S., FRANCO D.G., CHAVES P.O., GIANNESI G.C., MASUI D.C., RULLER R., CORRÊA B.O., BRASIL M.D.S., ZANOLO F.F. 2021. Plant growth promoting potential of endophytic *Aspergillus niger* 9-p isolated from native forage grass in Pantanal of Nhecolândia region, Brazil. *Rhizosphere* 18, 100332. <https://doi.org/10.1016/j.rhosph.2021.100332>
- GEISEN S., HOOVEN F.C., KOSTENKO O., SNOEK L.B., PUTTEN W.H. 2021. Fungal Root Endophytes Influence Plants in a Species-specific Manner that depends on Plant's Growth Stage. *Journal of Ecology* 109(4): 1618–1632. <https://doi.org/10.1111/1365-2745.13584>
- HARAHAP I., ELSIE, AULIA R. RAHMI R.A. 2017. Isolation of Endophytic Fungi from Vetiver Root Plants (*Vetiveria zizanioides* L.) and Antifungal Activity Test against *Candida albicans* [Isolasi Cendawan Endofit dari Tanaman Akar Wangi (*Vetiveria zizanioides* L.) dan Uji Aktivitas Antifungi terhadap *Candida albicans*]. Prosiding 2nd CELSci-Tech UNRI, vol. 2: 36–42 (in Indonesian). <https://ejurnal.umri.ac.id/index.php/PCST/article/view/265/166>
- JIANG Y., WU J., KAWAGISHI H., JIANG C., ZHOU Q., TONG Z., TONG Y., WANG P. 2022. Study on Secondary Metabolites of Endophytic Fungus, *Aspergillus fumigatus*, from *Crocus sativus* L. Guided by UHPLC-HRMS/MS-Based Molecular Network. *International Journal of Analytical Chemistry* 2022, 7067665. 11 p. <https://doi.org/10.1155/2022/7067665>
- KHASTINI R.O., OHTA H., NARISAWA K. 2012. The role of a dark septate endophytic fungus, *Veronaeopsis simplex* Y34, in *Fusarium* disease suppression in Chinese cabbage. *Journal of Microbiology* 50(4): 618–624. <https://doi.org/10.1007/s12275-012-2105-6>
- KHASTINI R.O. 2016. Dark Septate Root Endophytic Fungi in the Root System of *Avicennia* sp. Origin of the Pulau Dua Banten Nature Reserve and its Potential as a Producer of Antibacterial Compounds [Cendawan Endofit Akar Bersepta Gelap Pada Sistem Perakaran *Avicennia* sp. Asal Cagar Alam Pulau Dua Banten dan Potensinya sebagai Penghasil Senyawa Antibakteri]. Prosiding Seminar Nasional from Basic Science to Comprehensive Education Makassar, 26 Agustus 2016, 1992: 167–173 (in Indonesian).
- KHASTINI R.O. 2018. Isolation and Screening of Root Endophytic Fungi from the Rhizosphere of Talas Beneng [Isolasi, dan Penapisan Cendawan Endofit Akar Asal Rhizosfer Talas Beneng]. *Jurnal Biotek* 6(2): 25–36 (in Indonesian). <https://doi.org/10.24252/jb.v6i2.6823>
- KHASTINI R.O., MARYANI N., LESTARI I.D., RIFQIAWATI I., MILLAH N.U. 2021. Diver-

- sity of Entomopathogenic Fungi from Gunung Tukung Gede Nature Reserve. *Bioeduscience* 5(2): 116–121. <https://doi.org/10.22236/j.bes/526004>
- KHASTINI R.O., SUKARNO N., SUHARSONO U.W., HASHIDOKO Y. 2022. Isolation and Growth Response of Root Mutualistic Fungi on Several Agriculture and Forestry Plants [Isolasi dan Respons Tumbuh Cendawan Mutualistik Akar pada Beberapa Tanaman Pangan dan Kehutanan]. *Indonesian Journal of Agricultural Sciences [Jurnal Ilmu Pertanian Indonesia]* 27(1): 85–94 (in Indonesian). <https://doi.org/10.18343/jipi.27.1.85>
- KRIEG C.P., CHAMBERS S.M. 2022. The ecology and physiology of fern gametophytes: A methodological synthesis. *Applications in Plant Sciences* 10(2), e11464. 13 p. <https://doi.org/10.1002/aps3.11464>
- MANURUNG P.L., RAHMAWATI R., KURNIATUHADI R. 2022. Inventaritation of Endophyte Fungi from Avicennia Marina Leaves in Mempawah Mangrove Center, Pasir Village, Mempawah, West Borneo. *LenteraBio: Berkala Ilmiah Biologi* 11(3): 378–384 (in Indonesian). <https://doi.org/10.26740/lenterabio.v11n3.p378-384>
- MIN J., CHEN L., XIN H.-L., ZHENG C.-J., RAHMAN K., HAN T., QIN L.-P. 2016. A friendly relationship between endophytic fungi and medicinal plants: a systematic review. *Frontiers in Microbiology* 7, 906. 14 p. <https://doi.org/10.3389/fmicb.2016.00906>
- MISHRA A., GOND S.K., KUMAR A., SHARMA V.K., VERMA S.K., KHARWAR R.N. 2012. Sourcing the Fungal Endophytes: A Beneficial Transaction of Biodiversity, Bioactive Natural Products, Plant Protection and Nanotechnology. In: Satyanarayana T., Johri B. (Eds), *Microorganisms in Sustainable Agriculture and Biotechnology*. Springer, Dordrecht: 581–612. https://doi.org/10.1007/978-94-007-2214-9_26
- MORDECAI E.A. 2012. Soil moisture and fungi affect seed survival in California grassland annual plants. *PloS one* 7(6), e39083. 8 p. <https://doi.org/10.1371/journal.pone.0039083>
- MUSA M., JAN F.G., HAMAYUN M., JAN G., KHAN S.A., REHMAN G., ALI S., LEE I.J. 2023. An Endophytic Fungal Isolate *Paecilomyces lilacinus* Produces Bioactive Secondary Metabolites and Promotes Growth of *Solanum lycopersicum* under Heavy Metal Stress. *Agronomy* 13(3), 883. 17 p. <http://dx.doi.org/10.3390/agronomy13030883>
- MUTHUKUMAR T., PRABHA K. 2012. Fungal associations in gametophytes and young sporophytic roots of the fern *Nephrolepis exaltata*. *Acta Botanica Croatica* 71(1): 139–146. <https://doi.org/10.2478/v10184-011-0056-3>
- MUTHUKUMAR T., SATHIYARAJ G., PRIYADHARSINI P., UMA E., SATHIYADASH K. 2014. Arbuscular mycorrhizal and dark septate endophyte fungal associations in ferns and lycophytes of Palni Hills, Western Ghats, southern India. *Revista Brasileira de Botanica* 37(4): 561–581. <https://doi.org/10.1007/s40415-014-0085-y>
- NARANJO-ORTIZ M.A., GABALDÓN T. 2019. Fungal evolution: major ecological adaptations and evolutionary transitions. *Biological reviews of the Cambridge Philosophical Society* 94(4): 1443–1476. <https://doi.org/10.1111/brv.12510>
- NEJAD M.S., NAJAFABADI N.S., AGHIGHI S., PAKINA E., ZARGAR M. 2022. Evaluation of *Phoma* sp. Biomass as an Endophytic Fungus for Synthesis of Extracellular Gold Nanoparticles with Antibacterial and Antifungal Properties. *Molecules (Basel, Switzerland)* 27(4), 1181. 15 p. <https://doi.org/10.3390/molecules27041181>
- QIAN H., KESSLER M., VETAAS O.R. 2022. Pteridophyte species richness in the central Himalaya is limited by cold climate extremes at high elevations and rainfall seasonality at low elevations. *Ecology and Evolution* 12(5), e8958. 14 p. <https://doi.org/10.1002/ece3.8958>
- RILLIG M.C., WENDT S., ANTONOVICS J., HEMPEL S., KOHLER J., WEHNER J., CARUSO T. 2014. Interactive effects of root endophytes and arbuscular mycorrhizal fungi on an experimental plant community. *Oecologia* 174: 263–270. <https://doi.org/10.1007/s00442-013-2759-8>
- SEIFERT K.A., GAMS W. 2011. The Genera of

- Hyphomycetes, CBS biodiversity series No.9. Utrecht, The Netherlands: CBS-KNAW Fungal Biodiversity Center. Persoonia – Molecular Phylogeny and Evolution of Fungi 27(11): 119–129. 11 p. <https://doi.org/10.3767/003158511X617435>
- SUN R.-T., FENG X.-C., ZHANG Z.-Z., ZHOU N., FENG H.-D., LIU Y.-M., HASHEM A., AL-ARJANI A.-B.F., ABD ALLAH E.F., WU Q.-S. 2022. Root Endophytic Fungi Regulate Changes in Sugar and Medicinal Compositions of *Polygonum cuspidatum*. *Frontiers in Plant Science* 13, 818909. 11 p. <https://doi.org/10.3389/fpls.2022.818909>
- SUNARTI S. 2011. Research on Forest Vegetation in the Gantung Rock and Pagar Rock Areas, Mount Tukung Gede Nature Reserve, Serang [Penelitian Vegetasi Hutan di Wilayah Cadas Gantung dan Cadas Pagar, Cagar Alam Gunung Tukung Gede, Serang]. [Berk. Penel. Hayati Edisi Khusus] 5A: 109–119 (in Indonesian). <https://berkalahayati.org/files/journals/1/articles/266/submission/266-842-1-SM.pdf>
- SURAIDA S., SUSANTI T., AMRIYANTO R. 2013. Diversity of Ferns (Pteridophyta) in the Recognize Forest Park, Jambi City [Keane-karagaman Tumbuhan Paku (Pteridophyta) di Taman Hutan Kenali Kota Jambi]. *Prosiding Semirata FMIPA, Universitas Lampung*: 387–392 (in Indonesian). <https://jurnal.fmi-pa.unila.ac.id/semirata/article/view/640>
- TOGHUEO R.M.K., BOYOM F.F. 2020. Endophytic *Penicillium* species and their agricultural, biotechnological, and pharmaceutical applications. *Biotech* 10(3), 107. <https://doi.org/10.1007/s13205-020-2081-1>
- TSENG Y.-H., ROUINA H., GROTEN K., RAJANI P., FURCH A.C.U., REICHELT M., BALDWIN I.T., NATARAJA K.N., SHAANKER R.U., OELMÜLLER R. 2020. An Endophytic Trichoderma Strain Promotes Growth of Its Hosts and Defends Against Pathogen Attack. *Frontiers in plant science* 11, 573670. 16 p. <https://doi.org/10.3389/fpls.2020.573670>
- WATTS D., PALOMBO E.A., CASTILLO A.J., ZAFERANLOO B. 2023. Endophytes in Agriculture: Potential to Improve Yields and Tolerances of Agricultural Crops. *Microorganisms* 11(5), 1276. 29 p. <https://doi.org/10.3390/microorganisms11051276>
- WEN J., OKYERE S.K., WANG S., WANG J., XIE L., RAN Y., HU Y. 2022. Endophytic Fungi: An Effective Alternative Source of Plant-Derived Bioactive Compounds for Pharmacological Studies. *Journal of fungi* (Basel, Switzerland) 8(2), 205. 45 p. <https://doi.org/10.3390/jof8020205>