JURNAL BIOLOGICA SAMUDRA 7 (1): 9-17 (2025)
DOI. https://doi.org/10.33059/jbs.v7i1.10991

Keanekaragaman Jamur Endofit pada Ranting dan Akar *Baccaurea brevipes* Hook.f. di Hutan Lindung Kota Langsa, Aceh, Indonesia

Diversity of Endophytic Fungi in The Twigs and Roots of Baccaurea brevipes Hook.f. in Langsa Protected Forest, Aceh, Indonesia

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Received: 22 Mei 2025 **Accepted: 15 Juni 2025** **Published: 20 Juni 2025**

KATA KUNCI *Baccaurea, Keanekaragaman, isolasi, fungi endofit, Aceh*
KEYWORDS *Baccaurea, diversity, isolation, endophytic fungi, Aceh*

ABSTRAK

Tanaman *Baccaurea brevipes* Hook.f. telah dipelajari di masa lalu dan berpotensi menjadi ramuan terapeutik. Tanaman Tampoi (*B. brevipes*) telah digunakan sebagai obat oleh masyarakat Aceh untuk mengatur haid dan mempermudah buang air kecil. Di alam, jamur endofit tersebar luas dan dapat dengan mudah ditemukan di jaringan tumbuhan. Tanpa menginfeksi tanaman inang, endofit menjajah ruang antar dan intraseluler jaringan. Jamur endofit dapat membuat bahan kimia bioaktif yang mungkin mirip atau berbeda dari tanaman inangnya dan memiliki efek farmakologis. Namun, belum ada penelitian yang dilakukan pada jamur endofitnya sebagai sumber potensial untuk pengembangan pengobatan baru. Penelitian ini dilakukan untuk melihat jamur endofit pada akar dan ranting Baccaurea. Teknik yang digunakan dalam penelitian ini adalah pemurnian fungi endofit, identifikasi fungi endofit makroskopik dan mikroskopik, dan isolasi fungi endofit. Lima isolat diidentifikasi sebagai hasil isolasi dan pemurnian; hasil tersebut ditetapkan sebagai isolat BVA1, BVA2, BVA3, BVR1 dan BVR2. Genus *Neofusicocum* sp., *Colletotrichum* sp., *Mucor* sp., *Fusarium* sp. dan *Schizophyllum* sp. diasumsikan sebagai jenis isolat jamur endofit BVA1, BVA2, BVA3, BVR1 dan BVR2.

ABSTRACT

The Baccaurea brevipes Hook.F. plant has been studied in the past and has the potential to be a therapeutic herb. The Tampoi plant (B. brevipes) has been used as a medication by the Acehnese to regulate menstruation and make urinating easier. In nature, endophytic fungi are widespread and can be easily found in plant tissue. Without infecting the host plant, the endophytes colonize the inter and intracellular spaces of the tissue. Endophytic fungi can create bioactive chemicals that may be similar to or distinct from those of their host plants and have pharmacological effects. However, no studies have been done on its endophytic fungus as a potential source for novel medication development. This study was done to look into endophytic fungi in Baccaurea's twigs and roots. The techniques employed in this study were endophytic

fungi purification, identification of macroscopic and microscopic endophytic fungi, and endophytic fungi isolation. Five isolates were identified as a result of isolation and purification; they were designated as isolates BVA1, BVA2, BVA3, BVR1 and BVR2. The genera Neofusicocum sp., Colletotrichum sp., Mucor sp., Fusarium sp. and Schizophyllum sp. are assumed to be the kind of endophytic fungal isolates BVA1, BVA2, BVA3, BVR1 and BVR2.

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1. Introduction

The Leuser Ecosystem is close to the districts of Langsa City, Aceh (Navia *et al.*, 2019; Wibowo *et al.*, 2021). Additionally, there is a lot of biodiversity in the area that the local residents may utilize on a daily basis. Communities frequently use these plants as food and medicine, according to several research conducted at these sites (Lestari, 2014; Salusu *et al.*, 2020; Dwi *et al.*, 2017). People of Aceh province, on the other hand, have utilized a variety of plants, such as *Baccaurea* sp., as food and medicinal (Navia, *et al*., 2022). According to earlier research, the Aceh province has a number of *Baccaurea* species with great promise, including *Baccaurea brevipes*, but they have not yet been thoroughly investigated, especially the wild species. As the human population has increased, deforestation has accelerated, causing these plants to become extinct before their potential has been completely fulfilled. This work is essential for preserving the germplasm for long-term use, maintaining the wild *Baccaurea* sp. unique to Aceh, and advancing the improvement of healthcare and nutritional value.

Without producing any outward signs of illness, endophytic fungi live in plant tissue, including leaves, blossoms, twigs, and roots. It has been demonstrated that endophytes have a number of positive benefits on plants, such as enhanced growth and disease resistance (Murdiyah, 2017; Stone *et al.*, 2005; Strobel, 2003). They are commonly found in a variety of plant groupings, including grasses, trees, and vegetables (Zhai *et al.*, 2017; Mbilu *et al.*, 2018). Although it has been demonstrated that endophytic interactions with plants can protect them from infections, little is known about the complex interactions that endophytes have with pathogens and host plants. The creation of metabolite products derived from indigenous Acehnese raw materials may benefit from the potential secondary metabolites found in *Baccaurea* sp. wild plants. Several studies related to *Baccaurea* sp. has been carried out in Indonesia (Suciatmih, 2015; Hidayahti, 2017;Ladoh *et al.*, 2017), but this research still focuses on inventory activities and utilization of *Baccaurea* sp. certain types, especially types that have economic value. Research related to endophytic fungi from *Baccaurea* plants has never been reported before. Especially in Aceh province, it has never been found, so studies regarding the potential of *Baccaurea* sp. important to do. Several types of endophytic fungi associated with plants have potential in the agricultural, pharmaceutical and food and drug industries, because they contain secondary metabolite compounds as antibacterial, antimalarial and enzyme activation (Simamora *et al.*, 2021; Prihatiningtias and Wahyuningsih, 2005; Arnold and Lewis, 2005). The source of bioactive compounds currently being developed is from endophytic microbes which can produce bioactive compounds which have great potential to

be developed into new drugs. Endophytic microbes have great potential in the search for new sources of medicine. They can be processed using the fermentation method to produce large quantities of bioactive compounds. Consequently, the purpose of this study was to examine *B. brevipes* fungal endophytes.

Materials and Methods

2.1. Collection of the plant material

B. brevipes was collected from Langsa Protected Forest, Aceh. The specimen was deposited in a zip lock bag plastic, labelled, and put in a sample container for isolation at Biology Laboratory, Universitas Samudra.

2.2. Isolation and identification of endophytic fungi

Fresh *B. brevipes* twigs and roots were cleaned with tap water and sterilized using the technique suggested by Hallman *et al* (2006). After soaking in 70% alcohol and 5% NaOCL, the sterile materials were rinsed three times for five minutes each using distillate water. A tiny portion of the materials was dried, then inoculated onto different YGC plates and incubated at 27 ^{o}C. The growth of the endophytic fungi on the plates was checked daily for three to seven days. The growing colony was moved to new YGC plates for purification, and it was identified by its macroscopically and microscopically observed morphological properties, such as color, hypha, and reproductive structure. Literary sources and the Illustrated Genera of Imperfect Fungi Handbook were used in order to compare the traits (Barnett and Barry, 1972).

Result and Discussion

The species found in this investigation have never been documented before as *B. brevipes* endophytic fungus. A total of 5 isolates were isolated from the twigs and roots of *B. brevipes*. Of these, five isolates were successfully recognized by their morphology as *Neofusicoccum* sp. BVA-1, *Colletotrichum* sp. BVA-2, *Mucor* sp.BVA-3, *Fusarium* sp. BVR-1 and *Schizophyllum* sp. BVR-2. *Neofusicoccum* sp. BVA-1, *Colletotrichum* sp. BVA-2 and *Mucor* sp.BVA-3 were isolated from the roots of *B. brevipes*. *Fusarium* sp. BVR-1 and *Schizophyllum* sp. BVR-2 were found in the twigs. Based on the results of observations that have been made, endophytic fungi which has been successfully isolated from the twigs and roots of *B. brevipes*. Identified by looking at macroscopic and microscopic characteristics, with referring to the classification manual according to Barnett (1977) and were compared to other literary sources and the Illustrated Genera of Imperfect Fungi Handbook (Barnett and Barry, 1972) (Figure 1&2).

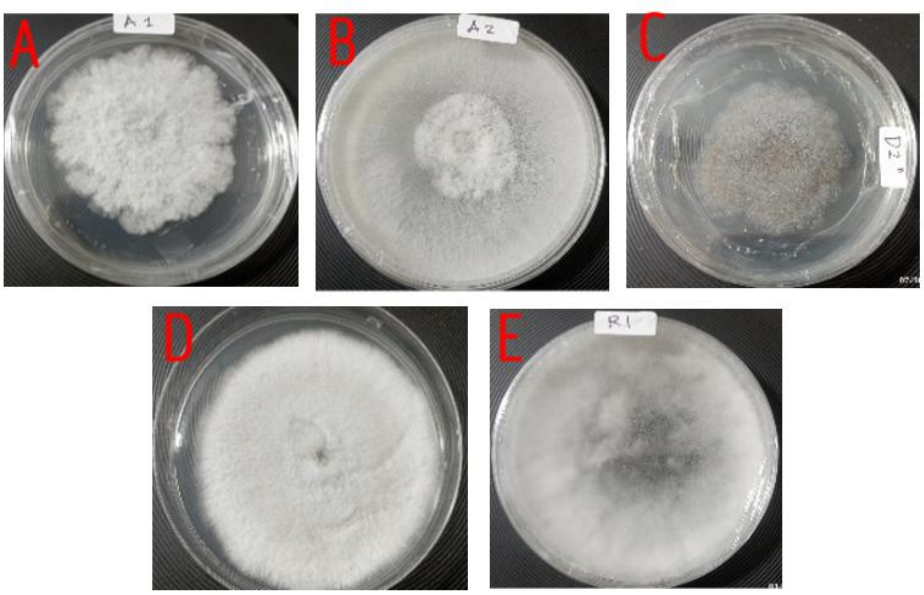


Figure 1. Photographs displaying the overall appearance of endophytic fungus isolated from *B. brevipes* that were cultivated on YGC media: (A) *Neofusicoccum* sp. BVA-1; (B) *Colletotrichum* sp. BVA-2; (C) *Mucor* sp. BVA-3; (D). *Fusarium* sp. BVR-1; (E) *Scizophyllum* sp. BVR-2.

A genus of fungi belonging to the Botryosphaeriaceae family is called *Neofusicoccum* (Figure 1A). *Neofusicoccum* is a genus of about 50 species that are found all over the world and are usually connected to plants. The diseases that *Neofusicoccum* infects on commercially and environmentally significant host plants are widely known. Members of this genus are specifically in charge of grapevine diseases include vascular darkening of the wood, fruit rots, leaf spots, stem dieback, bud necrosis, and perennial cankers. Numerous secondary metabolites were found in *Neofusicoccum* species, such as botryoisocoumarin A, cyclobotryoxide, botryosphaerones, and isosclerone. These compounds' structural variability and bioactivities may be linked to their involvement in fungal pathogenicity and virulence. The secondary metabolites of *Neofusicoccum* species are compiled in this review with an emphasis on their function in the fungus-host plant relationship (Salvatore *et al.,* 2021).

Figure 1B, *Colletotrichum* sp., on the other hand, came from twigs. Factors have been identified. *Colletotrichum* species inhabit a range of plant hosts as saprobes, endophytes, and diseases. In China and Northern Thailand, Ma *et al*. (2018) identified the endophytic *Colletotrichum* from *Dendrobium* spp. Similarly, in the Chinese regions of Guizhou and Hunan, two species of *Colletotrichum* : *C. jishouense* and *C. tongrenense* were isolated from *Nothapodytes pittosporoides* (Zhou *et al.,* 2019). Other citrus plants like *Citrus* sp., *C. medica* var. sarcodactylis, and *C. nobilis* var. microcarpa also have endophytic fungus (Suliati *et al.,* 2017; Puspita *et al.,* 2013; Ho *et al.,* 2012). Researchers have found that roots are a common plant part colonised by endophytic *Colletotrichum*, in the roots of *Arabidopsis thaliana* (Hiruma *et al.,* 2016) and *Dendrobium* sp. (Chen *et al.,* 2012). Endophytic *Colletotrichum* has been reported to control plant pathogenic fungi. For example, Casas *et al* (2021) reported that endophytic *C. siamense* reduced lesion caused by pathogenic *Colletotrichum* sp. in guarana plant (*Paullinia cupana* var. sorbilis) by producing phenylalanine ammonia lyase, chitinase, and peroxidase.

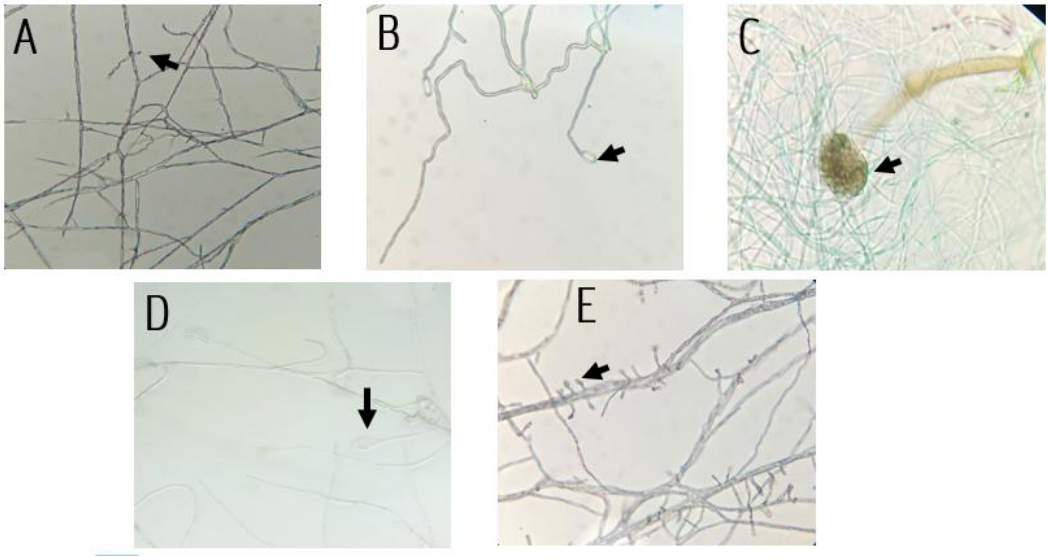


Figure 2. Photographs displaying the endophytic fungus reproductive structure that was isolated from *B. brevipes* as observed at x40: (A) *Neofusicoccum* sp. BVA-1; (B) *Colletotrichum* sp. BVA-2; (C) *Mucor* sp. BVA-3; (D). *Fusarium* sp. BVR-1; (E) *Scizophyllum* sp. BVR-2

The presence of *Mucor* in citrus plant tissues as endophytes has also been reported by Casas *et al* (2021); Puspita *et al* (2013); Ristiari *et al* (2019), that the ability of plants to adjust to metal pollution in the soil is improved by endophytic *Mucor* colonization (Domka *et al.,* 2019; Oliveira *et al.,* 2020). According to the authors, there isn't much proof that *Mucor* can biologically manage plant diseases.

Figure 1D, Isolates of *Fusarium* fungi are species of the Ascomycota class. According to Kuncoro (2011) *Fusarium* sp. can produce bioactive compounds that are antimicrobial, anticancer and antifungals. Research Jackson (1995), *Fusarium sambucinum* can produce secondary metabolites such as Fusacandin. Fusacandin, an antifungal agent of the papulacandin class. Endophytes and illnesses associated with the genus *Fusarium* have been found in a variety of vascular plant species and cultivars (Figure 1D). The majority of *Fusarium* endophytes form a variety of relationships with plants, such as commensals, mutualistic symbionts, and the most common, which are diseases in various crops. Furthermore, endophytic *Fusarium* bioactive secondary metabolites have demonstrated significant utility in agriculture as both phytostimulants and biocontrol agents by generating fungicidal and nematicidal chemicals and inducing defense responses. In order to address the problems of agriculture and climate change, endophytic *Fusarium* has become the best option for the creation of bioformulations, biopesticides, and biostimulants. By producing a variety of chemically varied and structurally unique secondary metabolites that have been shown to demonstrate an astounding range of biological activities, such as antimicrobial, anticancer, antiviral, antioxidants, antiparasitics, immunosuppressants, immunomodulatory, antithrombotic, and ability to biocontrol nematodes and plant diseases, In their host plants, these endophytic microorganisms may provide defense and survival mechanisms (Samal *et al.,* 2023; Toghueo, 2020).

Schizophyllum sp. (Figure 1E). Previous study by Maehara *et al* (2019), from the quina plant *Cinchona ledgeriana*, endophytic *Schizophyllum* was isolated, and the endophytic fungi showed the capacity to generate Cinchona alkaloids in synthetic liquid medium and potato dextrose agar. The morphological identification of certain fungal endophytes allowed for genus-level identification. However, because they were sterile and failed to develop any reproductive structures during the incubation period, some isolates are unidentifiable. Searching for and investigating prospective endophytes that can produce significant metabolites and bioactive chemicals began with a study on the isolation and identification of endophytic fungus. The main goal of endophyte research is to separate and to identify the bioactive substances that endophytic fungus produce (Xie *et al.,* 2023; , Wang *et al.,* 2025). To screen isolates for potential secondary metabolites and bioactive substances, more study on specific fungal endophytes is required. In the future, this study will keep screening for isolates that might produce antioxidant, immunomodulator, antiviral, anticancer, and antibiotic chemicals.

Conclusions

Five endophytic fungi were isolated from *B. Brevipes* Hook. f. and identified morphologically as *Neofusicocum* sp., *Colletotrichum* sp., *Mucor* sp., *Fusarium* sp. and *Schizophyllum* sp. Molecular identification will be used in the future to verify the species. More research is also necessary to fully comprehend how endophytic fungi can produce bioactive compounds, which are currently being developed. These compounds have a lot of potential for development and could be used to find new medical sources.

Acknowledgment

The authors thanks to Universitas Samudra's research institutions and community service (LPPM) for providing financing under the fiscal year 2023 "Penelitian Asisten Ahli" initiative with contract number 586/UN54.6/PG/2023.

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