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Identification of Endophytic Fungi of Balangeran (*Shorea balangeran* Korth.) by Morphological Characterization

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ABSTRACT

Endophytic fungi are the potential biological agent that could stimulate plant growth and inhibit plant disease. The existence of diverse and abundant endophytic fungi encourages continuous exploration. One of the plants that have potential as a source of endophytic fungi is balangeran (*Shorea balangeran* Korth.). The study aimed to identify the species of endophytic fungi from the leaf and twig of *S. balangeran* based on morphological characteristics. Fungi isolation was conducted by multiple sterilizations and purification. Furthermore, the macroscopic and microscopic morphological characteristics of the endophytic fungi isolates were also identified. The results showed nine colony characteristics of endophytic fungi. Endophytic fungi of *Colletotrichum* sp1. (SbD 1.1), *Phomopsis* sp. (SbD 1.3.1), *Colletotrichum* sp2. (SbD 1.3.2), and *Beauveria* sp. (SbD 3.1) were only found on the leaves, while *Aspergillus niger* (SbB 5.1), *Colletotrichum* sp3. (SbB 5.2), and *Nigrospora* sp. (SbB 5.3.2 and SbB 6.3) were only found on the twigs. Endophytic fungi *Phyllosticta* sp. (SbD 1.2) were found in the leaves and twigs. The growth rate of endophytic fungi showed that *Colletotrichum* sp3 (SbB 5.2) was the fastest, and *Phyllosticta* sp. (SbD 1.2) was the slowest among the nine isolates. Endophytic fungi that have been isolated will be analyzed for their benefits as a biological agent in future research.

1. Introduction

All plants in natural ecosystems appear to be symbiotic with fungal endophytes. Endophytic fungi are naturally associated with host plants and live-in plant tissue organs. This potential is supported by endophytic fungi that are very diverse and abundant in various plants. For example, endophytic fungi in the shoots, leaves, and stems of kemaitan (*Lunasia amara*) are very abundant consist of 37 different morphospecies (Istikorini and Hartoyo 2019). The role of endophytic fungi in plants is as a natural pest and disease control. Endophyte fungi from *Anredera cordifolia* can inhibit dieback disease severity 33.34 - 40.84% in jabon seedlings (Istikorini 2019). The endophytic fungi promote plant growth and resistance through the resulting bioactive compounds (Tejesvi et al. 2011). Endophytic fungi produce mycotoxins or other metabolites that cause changes in the host's physiology and biochemistry. The presence of endophytes in plant tissues can directly inhibit the development of pathogens in plants (Deka et al. 2017).

Each plant contains one or more endophytic microbes capable of producing antimicrobial secondary metabolites. Therefore, it is an excellent opportunity to explore endophytic fungi from various plants. One plant that can become a source of endophytic fungi is balangeran (*Shorea balangeran* Korth). Balangeran is a woody plant that grows in the habitat of peat swamp forests. This species is elong to *Dipterocarpaceae* family and the wood is classified into solid class II and durable class III (Suryanto et al. 2012). Balangeran wood is generally used as for construction purposes. The advantages of *S. balangeran* as a source of endophytic fungi are its resistant properties to wood-decaying fungi (Suryanto et al. 2012). Resistance to wood-decaying fungi attacks may be derived from metabolites produced by endophytic fungi in *S. balangeran*. Therefore, identifying the species of endophytic fungi in *S. balangeran* is the first step to researching other potentials of endophytic fungi.

Research of endophytic fungi derived from *S. balangeran* is very limited, even though *S. balangeran* has been known as wood species resistant to fungal attacks. Some endophytic fungi were found from *S. balangeran* twigs (Kumala and Fitri 2008). However, the endophytic fungi were not identified and only used the isolated code in subsequent tests. It shows that studies and information about the species of *S. balangeran* endophytic fungi are limited regarding the number of isolates and species identified. This plant's diverse and abundant endophytic can find other endophytes derived from leaves and twigs. In addition, the identification and morphological characteristics of fungi found need to be known to examine further the potential of bioactive compounds that are beneficial for improving plant growth and resistance. This study aimed to identify the species of endophytic fungi from *S. balangeran* based on macroscopic and microscopic morphological characteristics.

2. Materials and Methods

2.1. Time and Location

The study was conducted from November 2019 - March 2020. Samples of *S. balangeran* used were the healthy leaves and twigs obtained from IUPHHK PT. Dasa Intiga Kapuas Regency, Central Kalimantan (114° 39' East Longitude and 0° 04' North Latitude). The isolation of endophytic fungi *S. balangeran* was performed at the Pathology Laboratory, Department of Silviculture, Faculty of Forestry and Environment, IPB University.

2.2. Isolation and Identification of Endophytic Fungi

Samples of *S. balangeran* were obtained from IUPHHK PT. Dasa Intiga, Kapuas, Central Kalimantan, Indonesia. Samples were taken from as many as five plant seeds aged ± 9 months. Healthy samples with no plant disease symptoms were washed thoroughly using running water and air dry. Samples were separated between leaves and twigs. Leaf samples were cut into a ± 5 mm × 5 mm section, while the twig samples were cut with a length of ± 10 mm.

The method to isolate the endophytic fungi was done by modifying the procedure described by Photita et al. (2004). The slice cutting samples were sterilized by soaking in a solution of 96% ethanol for 30 seconds, natrium hypochlorite (NaOCl) 1% for 1 min; 70% ethanol for 1 min; 30% ethanol for 30 seconds; rinsed using sterile water for three times and dried in sterile filter paper. Samples were placed on PDA and incubated at room temperature for seven days. Each Petri dish

is planted with four segments of leaves or twigs. Mycelium fungi with different shapes and colors that grew on segment pieces were purified on PDA.

Subsequently, the isolates were observed on PDA and were identified based on macroscopic and microscopic morphological characteristics. Fungal isolates were characterized morphologically based on color and tint in colony surface and reverse, colony surface texture, colony margin, pattern, pigment exuded, and organ formed. Microscopic observations were conducted by observing the growth of hyphae, conidiophore, conidia, and other specific characteristics (Barnet and Hunter 1998; Watanabe 2002).

2.3. Growth Rate of Endophytic Fungi

Endophytic fungi obtained were then measured and observed in the growth rate of fungi colonies. Mycelial discs (diameter ± 6 mm) of each 7-day-old endophytic fungi were transferred to PDA. Colony diameter was measured daily for seven days, and growth rate was calculated as the 7-day average of mean daily growth (Prihastuti et al. 2009).

3. Results and Discussion

3.1. Number of Endophytic Fungi Isolates

The results of the isolation of endophytic fungi on the leaves and twigs of balangeran (*S. balangeran*) obtained nine morphological characteristics of the colony, which are coded isolate SbD 1.1, SbD 1.2, SbD 1.3.1, SbD 1.3.2, SbD 3.1, SbB 5.1, SbB 5.2, SbB 5.3.2, and SbB 6.3. Among the nine isolates, five isolates were found only in each part of the plant except the SbD 1.2 isolate found in both parts of the isolated plant (**Table 1**). Isolates SbD 1,1, SbD 1.3.1, SbD 1.3.2, and SbD 3.1 were only found in the *S. balangeran* leaves. In addition, isolates SbB 5.1, SbB 5.2, SbB 5.3.2, and SbB 6.3 were only found in *S. balangeran* twigs.

Table 1. Endophytic fungi isolate from balangeran (*S. balangeran*)

Isolate Code	Plant Section	
	Leaf	Twig
SbD 1.1	+	-
SbD 1.2	+	+
SbD 1.3.1	+	-
SbD 1.3.2	+	-
SbD 3.1	+	-
SbB 5.1	-	+
SbB 5.2	-	+
SbB 5.3.2	-	+
SbB 6.3	-	+
Total Isolate	5	5

Notes: + refer that fungi are present and – refer that fungi are absent in the specific plant section.

3.2. The Colony Growth Rate of Endophytic Fungi

The mycelium growth rate of endophytic fungi was observed for seven days. The growth rate of each isolate can be seen in **Fig. 1**. Fungi SbB 5.2 has a very fast growth rate because the SbB 5.2 colony has reached the edge of the Petri dish within seven days. Fungi SbD 1.1 and SbD 1.3.1 growth is not as fast as SbB 5.2, SbB 5.3.2, and SbB 6.3 with moderate growth. SbD 1.2, SbD

1.3.2, SbD 3.1 and SbB 5.1 are slow-growing isolates. The diameter of each isolate at the end of the observation can be seen in **Table 2**.

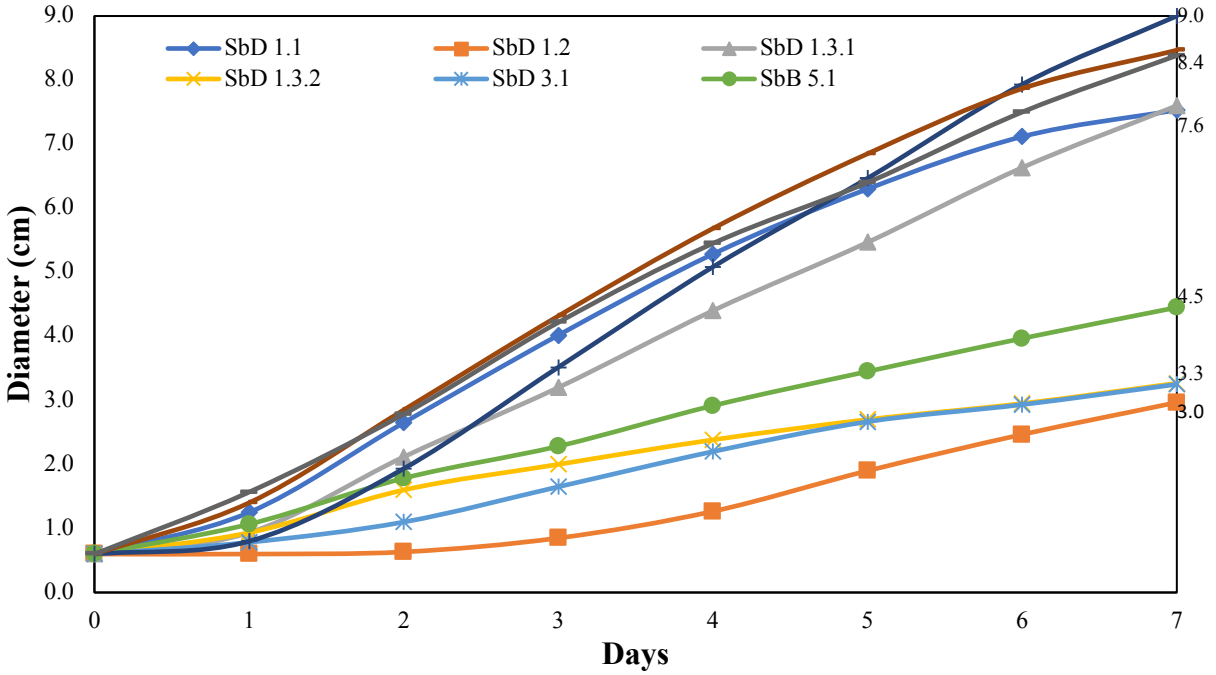

Fig. 1. The growth rate of endophytic fungi *S. balangeran* for seven days in PDA.

Table 2. Diameter of *S. balangeran* endophytic fungi colony on PDA 7 days after inoculation

Isolate Code	Colony Diameter (cm)
SbD 1.1	7.50
SbD 1.2	2.90
SbD 1.3.1	7.60
SbD 1.3.2	3.30
SbD 3.1	3.30
SbB 5.1	4.50
SbB 5.2	9.00
SbB 5.3.2	8.50
SbB 6.3	8.40

3.3. Morphological Characteristics of Isolate SbD 1.1

Isolate SbD 1.1 has the macroscopic features of white colonies with grayish-colored middles. Colonies on PDA at first white, becoming grey to dark grey at the center with age. The reverse color was white to grayish-green. Aerial mycelium is pale grey, dense, and cottony (**Fig. 2a**). Fungi SbD 1.1 has hyaline hyphae, stringed and hyaline conidiophores. The conidia are cylindrical and hyaline (**Fig. 2c**). Appressoria is dark gray to blackish brown and cylindrical (**Fig. 2d**). Based on these characteristics, isolate SbD 1.1 has the characteristics of the *Colletotrichum* sp.

3.4. Morphological Characteristics of Isolate SbD 1.2

The macroscopic feature of fungi SbD 1.2 shows dark-colored colonies. The colony color is gray at the beginning of development, then turns black. The edges of the colony are irregular, without aerial mycelium. The bottom appearance shows a blacker color (**Fig. 3b**). Fungi have macroscopic features of irregular colony edges, arachnoid patterns, smooth and flat colony

surfaces, and mycelium is dull grayish-green to blackness (**Fig. 3a**). Microscopic features of isolate SbD 1.2 are hyphae hyaline (**Fig. 3c**), hyaline conidia, and *ovoid* form (**Fig. 3d**).

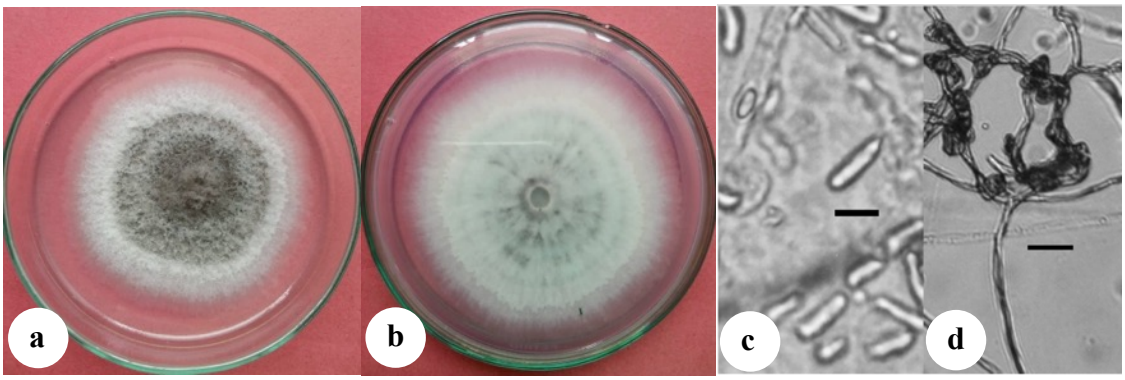

Fig. 2. The morphological characteristics of SbD 1.1: (a) upper surface colony, (b) lower surface colony, (c) conidia, and (d) appressoria. Scale bars = 10 µm.

Based on the macroscopic and microscopic features, the isolate SbD 1.2 is *Phyllosticta* sp. (teleomorph Guignardia). Fungi *Phyllosticta* sp has macroscopic features of irregular colony edges, smooth and flat colony surfaces, and dull grayish to blackish mycelium (Baayen et al. 2002). Conidia is ovoid, hyaline, aseptate, and surrounded by thick *mucoid* (Guarnaccia et al. 2017).

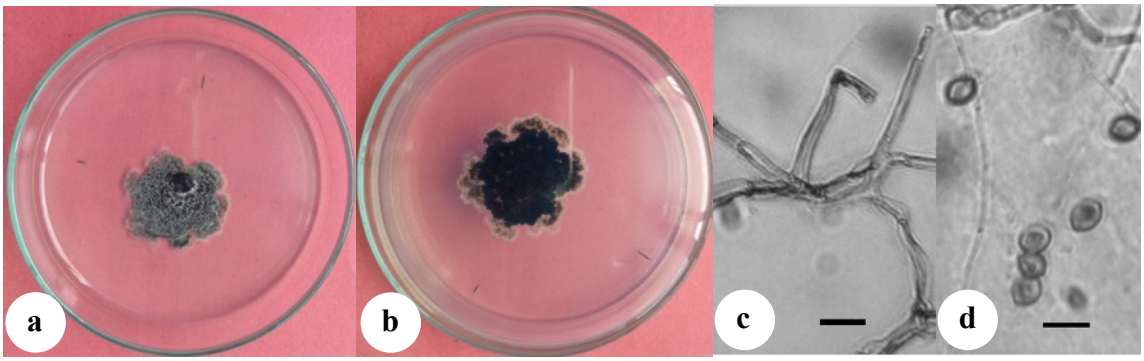

Fig. 3. The morphological characteristics of SbD 1.2: (a) upper surface colony, (b) lower surface colony, (c) hyphae, and (d) conidia. Scale bars = 10 µm.

3.5. Morphological Characteristics of Isolate SbD 1.3.1

Isolate SbD 1.3.1 has white colonies with layered circular growth with wavy edges. Its mycelium growth pattern is circularly wavy, forming a rose pattern (*flowery pattern*) with irregular mycelium *edges* (**Fig. 4a**). Mycelium has a *powdery* texture. Isolate SbD 1.3.1 has little air mycelium. Hyphae hyaline are pale brown and stringy. Conidia hyaline, 1-celled, dimorphic. Pycnidia is half-embedded in agar media (**Fig. 4c**). Based on the macroscopic and microscopic morphological characteristics, isolate SbD 1.3.1 has the characteristics of *Phomopsis* spp. The morphological characteristics *of Phomopsis* spp. has a bumpy colony appearance. The colony is white with wavy edges and the bottom of the grayish colony (Mahadevakumar et al. 2017). *Phomopsis* spp. has black conidiomata with cylindrical and elongated phialide containing non-agreed hyaline conidia, namely alpha and beta (Udayanga et al. 2011).

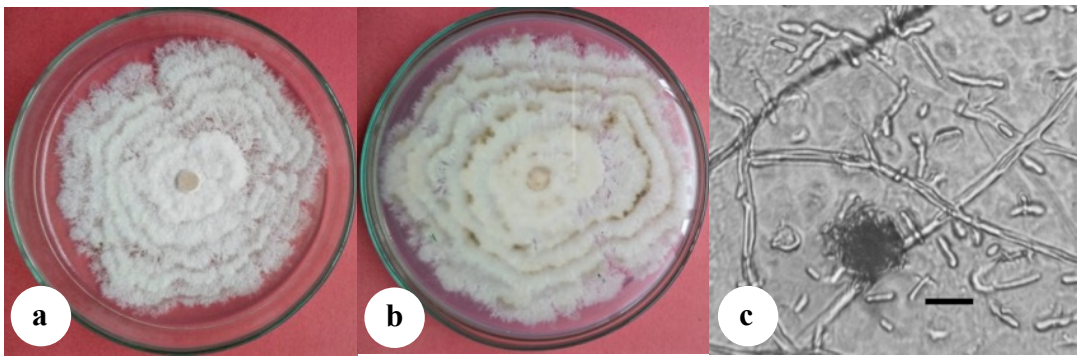

Fig. 4. The morphological characteristics of SbD 1.3.1: (a) upper surface colony, (b) lower surface colony, (c) conidia. Scale bars = 10 µm.

3.6. Morphological Characteristics of Isolate SbD 1.3.2

Isolate SbD 1.3.2 has macroscopic features of the colony in greenish-gray and blackish brown (**Fig. 5a**) with white edges at the bottom (**Fig. 5b**). The edges of the colony have growth that tends to be flat. Hyphae are hyaline and stringed. Conidia is falcate-shaped (**Fig. 5c**). Based on macroscopic and microscopic cyclical, isolate SbD 1.3.2 is identified as *Colletotrichum* sp.

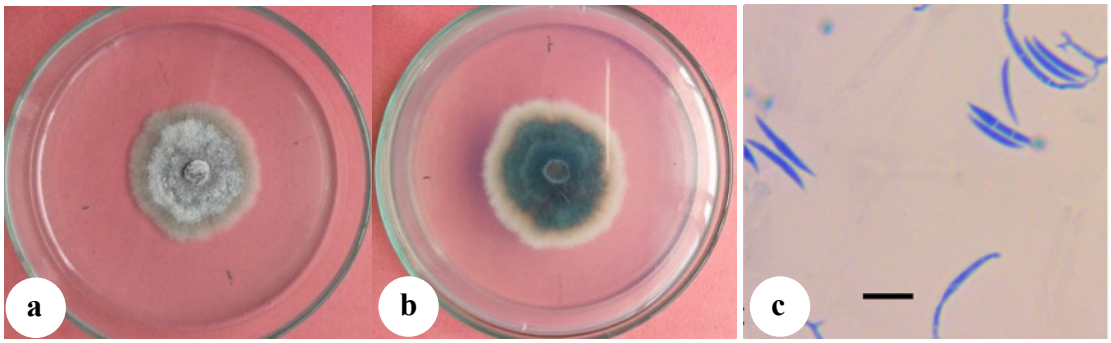

Fig. 5. The morphological characteristics of SbD 1.3.2: (a) upper surface colony, (b) lower surface colony, and (c) conidia. Scale bars = 10 µm.

3.7. Morphological Characteristics of Isolate SbD 3.1

Isolate SbD 3.1 has colonies of white macroscopic features on the upper and lower surfaces (**Fig. 6b**). The mycelia isolate misty like cotton (**Fig. 6a**). The edge of the isolate SbD 3.1 colony tends to be flat and has a colony growth of 3.25 cm at the end of the seven-day observation (**Table 2**). Isolate SbD 3.1 has hyphae attached to hyaline colors and thin hyphae walls (**Fig. 6c**). The conidia are round (*ovoid*) with hyaline color (**Fig. 6c**). Based on Oliveira et. al. (2010), the fungi are *Beauveria* sp.

3.8. Morphological Characteristics of Isolate SbB 5.1

The result of morphological observations of the colony showed that isolate SbB 5.1 has a yellow colony with a round black spore on the upper surface (**Fig. 7a**). In contrast, the lower surface of the colony is yellowish-white (**Fig. 7b**). Mycelium forms a concentric zone with irregular edges. The isolate has little air mycelium on the upper surface of the colony (**Fig. 7a**). SbB 5.1 has a light brown hyaline conidiophore, forms globular vesicles, and grows concentrically (**Fig. 6c**). At the end of the conidiophore, there is a phialide. Conidia is *globose*/round (**Fig. 6d**).

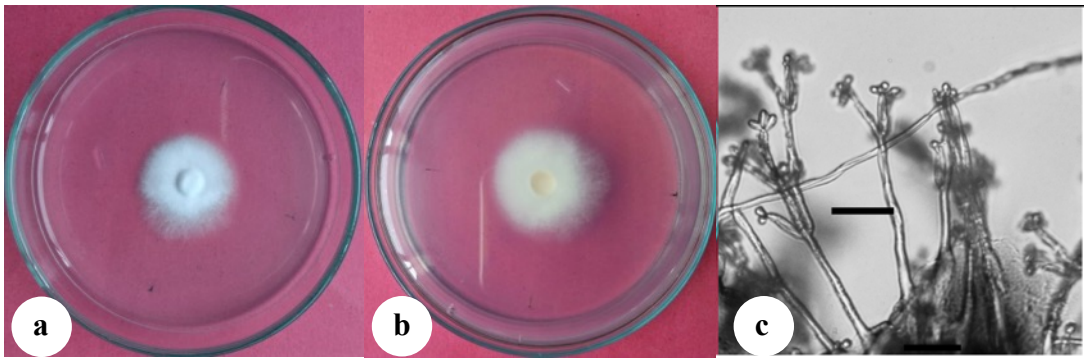

Fig. 6. The morphological characteristics of SbD 3.1: (a) upper surface colony, (b) lower surface colony, and (c) conidiophore and conidia. Scale bar*s* =10 µm.

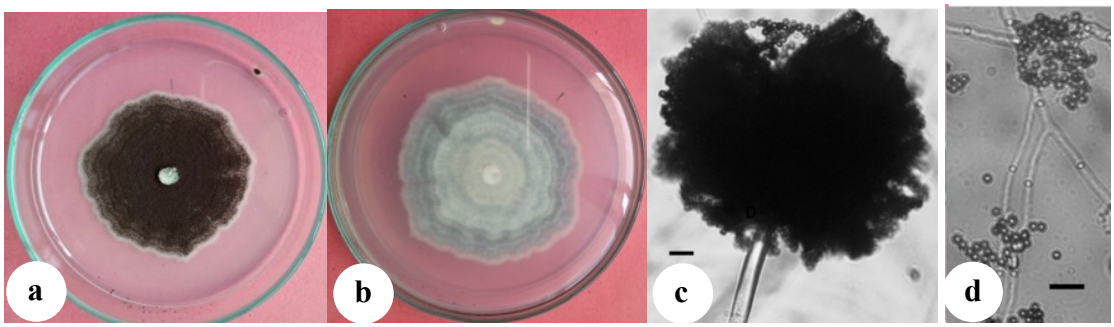

Fig. 7. The morphological characteristics of SbB 5.1: (a) upper surface colony, (b) lower surface colony, (c) conidiophore, and phialides (d) conidia. Scale bars =10 µm.

Isolate SbB 5.1 has the morphology characteristics of the fungi *Aspergillus niger*. Isolates have white colonies with round black spores on the upper surface, while the lower surface of the colony is yellowish-white (Silva et al. 2011). Konidiofor *A. niger* is fine-walled with a hyaline color that turns dark approaching vesicles (George and Ramteke 2019).

3.9. Morphological Characteristics of Isolate SbB 5.2

Isolate SbB 5.2 has a greenish-gray colony on the upper surface (**Fig. 8a**). The lower surface is blackish-brown gray (**Fig. 8b**). Isolate SbB 5.2 has a flat colony edge. Conidia hyaline, cylindrical conidia (**Fig. 8c**). Based on the macroscopic and microscopic characterization, sycopic of isolate SbB 5.2 is *Colletotrichum* sp. (Weir et al. 2012).

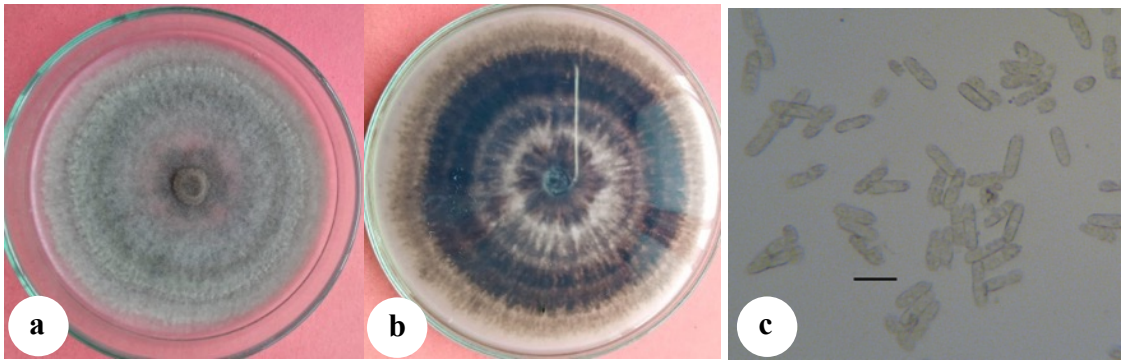

Fig. 8. The morphological characteristics of SbB 5.2: (a) upper surface colony, lower surface colony, and (c) conidia. Scale bars =10 µm.

3.10. Morphological Characteristics of Isolate SbB 5.3.2

Isolate SbB 5.3.2 has a cotton-like colony's features with corrugated colony edges (**Fig. 9b**). The upper surface has black spots that are spores (**Fig. 9a**). The isolate SbB 5.3.2 has microscopic features of hyphae hyaline and spherical (round) black conidia (**Fig. 9c**). This characteristic is *Nigrospora* sp. Based on Wang et al. (2017), *Nigrospora* sp. It has the characteristics of hyaline-colored hyphae and solitary conidia in a *spherical* (round) shape in black.

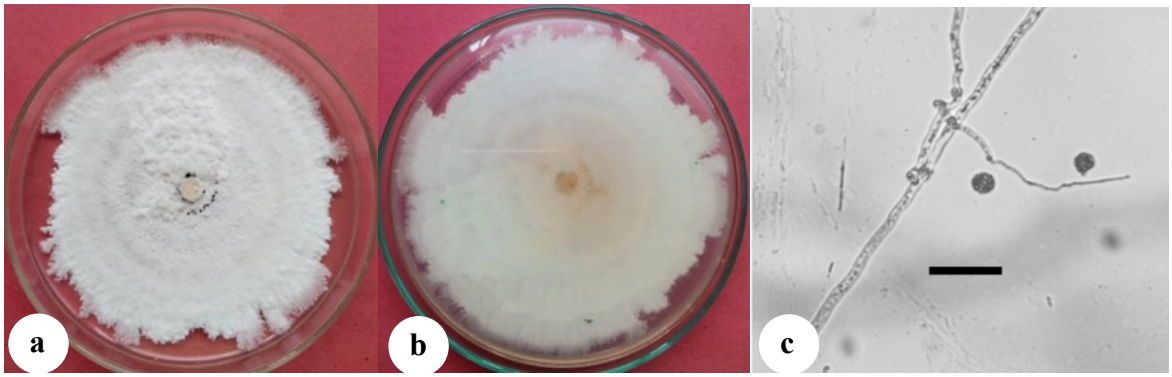

Fig. 9. The morphological characteristics of SbB 5.3.2: (a) upper surface colony, (b) lower surface colony, and (c) hyphae, and conidia. *Scale bars* =10 µm.

3.11. Morphological Characteristics of Isolate SbB 6.3

Isolate SbB 6.3 has the macroscopic features of a yellowish-white colony like cotton with corrugated colonies (**Fig. 10a**). Hifa hyalin and conidia are *spherical* (round) in black (**Fig. 10c).** The macroscopic and microscopic characteristics of SbB 6.3 is similar to isolate SbB 5.3, *Nigrospora* sp. (Wang et al. 2017). The growth of isolate SbB 6.3 colonies on the 7th day is 8.4 cm (**Table 2**).

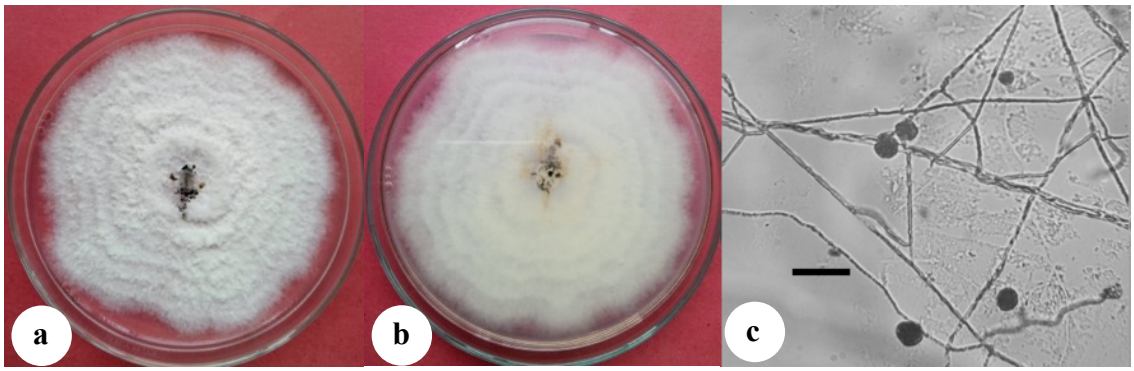

Fig. 10. The morphological characteristics of SbB 6.3: (a) upper surface colony, (b) lower surface colony, and (c) hyphae, and conidia. *Scale bars* =10 µm

The identification results show that the leaves and twigs of the *S. balangeran* contain some endophytic fungi, namely *Colletotrichum* sp1. (SbD 1.1), *Phylosticta* spp. (SbD 1.2). *Phomopsis* sp (SbD 1.3.1), *Colletotrichum* sp2. (SbD 1.3.2), *Beauveria* sp. (SbD 3.1), *Aspergillus niger* (SbB 5.1), *Colletotrichum* sp3. (SbB 5.2), *Nigrospora* sp. (SbB 5.3.2 and SbB 6.3). In this study, endophytic fungi found in the leaves and twigs of the *S. balangeran* were also seen as endophytic fungi in other plants. Three *Colletotrichum* species in this research have different macroscopic and microscopic morphological characteristics. Identification morphology has the challenge of

identifying at the species level. Such as the consistency and accuracy of identifications, molecular identification techniques are needed (Manoylove 2014).

Various endophytes were isolated from their host plants and then the secondary metabolites were produced by these endophytic microbes with isolated, purified and diluted their molecular structure. *Colletotrichum* sp. is an endophytic fungus from inside the stem of *Artemisia annua*, producing the highly potential metabolite artemisinin as an antimalarial (Lu et al. 2000). *Colletotrichum* sp1. (SbD 1.1), has characteristics like *C. fructicol*a. Fungi *C. fructicol*a has a white upper colony changing from gray to dark gray, and the lower colony is white and greenish. Conidiophores rare, septate, hyaline. Conidia ellipsoidal, smooth-walled, septate, hyaline. *C. fructicola* were identified as fungal endophytes of *Dendrobium* spp. (Hu et al. 2015; Ma et al. 2018).

Based on macroscopic and microscopic cyclical, *Colletotrichum* sp2. (SbD 1.3.2) has characteristics like *C. truncatum*. The characteristics of colony *C. truncatum* is a pale gray upper colony and a dark brown lower colony, and falcate-shaped conidia with blackish-brown appressoria (Liu et al. 2016). *Colletotrichum* sp3. (SbB 5.2) has characteristics like *C. queenslandicum* (Weir et al. 2012). Mukti et al. (2018) reported that eight species of endophytic fungi were successfully isolated and turned out to be from the same genus, Colletotrichum. Species of endophytic fungi successfully isolated from *Cordyline fruticosa* leaves include *C. coccodes, C. aotearoa, C. kahawae, C. fruticola, C. cordylinicola,* and *C. theobromicola.* The species of endophytic fungi that was successfully isolated from the *Cordyline fruticosa* stem is *C. queenslandicum*.

Phomopsis sp. produces antibacterial and antifungal metabolites Phomopsichalasin (Horn et al. 1995). Thirty strains of *Phyllosticta* have been isolated on healthy leaves in some plants (Wikee et al. 2013). Fungi *Beauveria* sp. is one of the endophytic entomopathogenic fungi in cocoa plants (Trizelia and Winarto 2016). Fungi *Nigrospora* sp. (SbB 5.3.2) has a very fast growth rate because it has reached the edge of a Petri dish within seven days. Fungi *Colletotrichum* sp1. (SbD 1.1) has a moderate growth rate. Fungi *Phylosticta* (SbD 1.2), *Beauveria* sp. (SbD 3.1), and *Colletotrichum* sp2. (SbD 1.3.2) are classified as slow-growth isolates. The resulting growth of fungi colonies has a vital role in its life cycle. In general, biological agents have rapid growth, forming large conidia to become an efficient saprophyte. The faster the growth of antagonistic fungi, the more effectively suppressing pathogens' growth.

Conidia are a means of asexual reproduction, spread, and the survival of fungi in their environment. The growth spurt indicates the competition mechanism of space and nutrition with pathogens. The faster the growth of antagonistic fungi, the more effectively suppressing pathogens' growth (Agrios 2008; Soesanto 2008). Endophytic fungi that have been isolated from the leaves and twigs of the *S. balangeran* need to be tested for their ability to produce beneficial secondary metabolites and tested for their potential as a biological agency.

4. Conclusions

The study obtained nine species of endophytic fungi based on the morphological characteristics of the leaves and twigs of *S. balangeran*. Fungi of *Colletotrichum* sp1. (SbD 1.1), *Phomopsis* sp (SbD 1.3.1), *Colletotrichum* sp2. (SbD 1.3.2), and *Beauveria* sp. (SbD 3.1) were found only on the leaves. Endofit fungi of *Aspergillus niger* (SbB 5.1), *Colletotrichum* sp3. (SbB 5.2), and *Nigrospora* sp. (SbB 5.3.2 and SbB 6.3) were only found on the twigs. Fungi *Phyllosticta*

sp. (SbD 1.2) were found on the leaves and twigs *of S. balangeran.* The morphological identification has the challenge of identifying at the species level; hence, molecular identification techniques are needed. Endophytic fungi from S. balangeran will be analyzed for their benefits as a biological agent in future research.

References

Agrios, N. G. 2008. Plant Pathology – Fifth Edition. Department of Plant Pathology. University of Florida. United States of America. 922p.

Baayen, R. P., Bonants, P. J. M., Verkley, G., Carroll, G .C., van der Aa, H. A., de Weerdt, M., van Brouwershaven, I. R., Schutte, G. C., Maccheroni Jr, W., Glienke de Blanco, C., and Azevedo, J. L. 2002. Nonpathogenic Isolates of the Citrus Black Spot Fungus, *Guignardia citricarpa*, Identified as a Cosmopolitan Endophytic of Woody Plants, *G. mangiferae* (*Phyllosticta capitalensis*). *Phytopathology* 92(5): 464-477. DOI: 10.1094/phyto.2002.92.5.464

Barnet, H. L., and Hunter, B. B. 1998. *Illustrated Genera of Imperfect Fungi 4ed. The American Phytopathological society*. APS Press. St. Paul Minnesota, US.

Deka, D., Tayung, K., and Jha, D. K. 2017. Harnessing Fungal Endophytes for Plant and Human Health. Endophytes: *Biology and Biotechnology* 59-98. DOI: 10.1007/978-3-319-66541-2_4

George, M., and Ramteke, P. W. 2019. Morphology, Molecular Identification, and Phylogenetic Analysis based on Internal Transcribed Spacer (ITS) of the Ribosomal Nuclear DNA (rDNA) Sequence of A Pathogenic Fungal Isolate *Aspergillus niger* LKO1. *Tropical Plant Research* 6(2): 166-170. DOI: 10.22271/tpr.2019.v6.i2.024

Guarnaccia, V., Groenewald, J. Z., Li, H., Glienke, C., Carstens, E., Hattingh, V., Fourie, P. H., and Crous, P. W. 2017. First Report of *Phyllosticta citricarpa* and Description of Two New Species, *P. paracapitalensis* and *P. paracitricarpa*, from Citrus in Europe. *Studies in Mycology* 87:161-185. DOI: 10.1016/j.simyco.2017.05.003

Horn, W. S., Simmonds, M. S. J., Schartz, R. E., and Blaney, W. M. 1995. Phomopsichalasin, a Novel Anti-Microbial Agent from an Endophytic *Phomopsis* spp. *Tetrahedron* 14: 3969-3978. DOI: 10.1016/0040-4020(95)00139-y

Hu, M. J., Grabke, A., and Schnabel, G. 2015. Investigation of the *Colletotrichum gloeosporioides* Species Complex Causing Peach Anthracnose in South Carolina. *Plant Disease* 99: 797-805. DOI: 10.1094/pdis-10-14-1076-re

Istikorini, Y. 2019. Potensi Cendawan Endofit pada Tanaman Binahong (*Anredera cordifolia* (Ten.) Steenis untuk Mengendalikan Botryodiplodia Theobromae Patogen. Penyebab Mati Pucuk pada Bibit Jabon (*Anthocepalus cadamba* (Roxb.) Miq. *Jurnal Silvikultur Tropika* 10 (02): 114-118.

Istikorini, Y., and Hartoyo, A. P. P. 2019. The Diversity of Endophytic Fungi in Kemaitan (*Lunasia amara* Blanco). *IOP Conf. Series: Earth and Environmental Science* 394:012016. DOI: 10.1088/1755-1315/394/1/012016

Kumala, S., and Fitri, N. A. 2008. Penapisan Kapang Endofit Ranting Kayu Meranti Merah (*Shorea balangeran* Korth.) sebagai Penghasil Enzim Xilanase. *Jurnal Ilmu Kefarmasian Indonesia* 6(1): 1-6.

Liu, F., Tang, G., Zheng, X., Li, Y., Sun, X., Qi, X., Zhou, Y., Xu, J., Chen, H., Chang, X., Zhang, S., and Gong, G. 2016. Molecular and Phenotypic Characterization of Colletotrichum

Species Associated with Anthracnose Disease in Peppers from Sichuan Province China. *Scientific Reports* 6:32761. DOI: 10.1038/srep32761

Lu, H., Zou, W. X., Meng, J. C., Hu, J., and Tan, R. X. 2000. New Bioactive Metabolites Produced by *Colletotrichum* sp., an Endophytic Fungus in *Artemisia annua*. *Plant Science* 151: 76-73. DOI: 10.1016/S0168-9452(99)00199-5

Ma, X., Nontachaiyapoom, S., Jayawardena, R. S., Hyde, K. D., Gentekaki, E., Zhou, S., Qian, Y., Wen, T., and Kang, J. 2018. Endophytic Colletotrichum Species from *Dendrobium* spp. in China and Northern Thailand. *MycoKeys* 43: 23-57. DOI: 10.3897/mycokeys.43.25081

Mahadevakumar, S., Amruthavalli, C., Sridhar, K. R., and Janardhana, G. R. 2017. Prevalence, Incidence, and Molecular Characterization of *Phomopsis vexans* (*Diaporthe vexans*) Causing Leaf Blight and Fruit Rot Disease of Brinjal in Karnataka (India). *Plant Pathology and Quarantine* 7(1): 41-58. DOI: 10.5943/ppq/7/1/5

Manoylov, K. M. 2014. Taxonomic Identification of Algae (Morphological and Molecular): Species Concepts, Methodologies, and Their Implications For Ecological Bioassessment. *Journal of Phycology* 50: 409-424. DOI: 10.1111/jpy.12183

Mukti, P. K., Hastuti, U. S, and Sulisetijono. 2018. Karakterisasi, Identifikasi, dan Observasi Histologik Letak Fungi Endofit yang Diisolasi dari Tanaman *Cordilyne fruticosa* (L.) A. Chev. *Proceeding Biology Education Conference* 15(1): 862-869.

Oliveira, I., Pereira, J. A., Bento, A., and Baptista, P. 2010. Viability of *Beauveria bassiana* Isolates After Storage under Several Preservation Method. *Annals of Microbiology* 63: 339-344. DOI: 10.1007/s13213-010-0147-8

Photita, W., Lumyong, S., Lumyong, P., McKenzie, E. H. C., and Hyde, K. D. 2004. Are Some Endophytes of Fungal Diversity Musa Acuminata Latent Pathogens? *Fungal Diversity* 16: 131-140.

Prihastuti, H., Cai, L., Chen, H., McKenzie, E. H. C., and Hyde, K. D. 2009. Characterization of Colletotrichum Species Associated with Coffee Berries in Northern Thailand. *Fungal Diversity* 39: 89-109.

Silva, D. M., Batista, L. R., Rezende, E. F., Fungaro, M. H. P., Sartori, D., and Alves, E. 2011. Identification of Fungi of the Genus Aspergillus Section Nigri Using Polyphasic Taxonomy. *Brazilian Journal of Microbiology* 42:761-773.

Soesanto, L. 2008. *Pengantar Pengendalian Hayati Penyakit Tanaman Suplemen ke Gulma dan Nematode*. Rajawali-Press. Jakarta, Indonesia. 573p.

Suryanto, Hadi, T. S., and Savitri, E. 2012. *Budidaya Shorea balangeran di Lahan Gambut. Banjarbaru*. Balai Penelitian Kehutanan Banjarbaru. Banjarbaru.

Tejesvi, M. V., Kajula, M., Mattila, S., and Pirttilä, A. M. 2011. Bioactivity and Genetic Diversity of Endophytic Fungi in *Rhododendron tomentosum* Harmaja. *Fungal Diversity* 47:97-107.

Trizelia dan Winarto. 2016. Keanekaragaman Jenis Cendawan Entomopatogen Endofit pada Tanaman Kakao (*Theobroma cacao*). *Pros Semnas Masy Biodiv Indon.* 2(2): 277-281.

Udayanga, D., Liu, X., McKenzie, E. H. C., Chukeatirote, E., Bahkali, A. H. A., and Hyde, K. D. 2011. The Genus Phomopsis: Biology, Applications, Species Concepts and Names of Common Phytopathogens. *Fungal Diversity* 50: 189-225.

Wang, M., Liu, F., Crous, P. W., and Cai, L. 2017. Phylogenetic Reassessment of Nigrospora: Ubiquitous Endophytes, Plant, and Human Pathogens. *Persoonia* 39: 118-142. DOI: 10.3767/persoonia.2017.39.06

Weir, B. S., Johnston, P. R., and Damm, U. 2012. The *Colletotrichum gloeosporioides* Species Complex. *Studies in Mycology* 73: 115-180. DOI: 10.3114/sim0011

Wikee, S., Lombard, L., Crous, P. W., Nakhasima, C., Motohashi, K., Chukeatirote, E., Alias, S. A., and McKenzie, E. H. C. 2013. *Phyllosticta capitalensia*, A Widespread Endophytic of Plants. *Fungal Diversity* 60:91-105.

Watanabe, T. 2002. *Pictorial Atlas of Soil and Seed Fungi: Morphologies of Cultured Fungi and Key to Species - Second Edition*. CRC Press LLC. London, UK. 486p.