

## Short Communication



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# Species Diversity of Genus *Aspergillus*, Endophytic Fungal Isolated From Mangrove *Ceriops tagal* and their Antibiotic Potential

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## ARTICLE INFO

## Article history:

Received February 20, 2025

Received in revised form April 9, 2025

Accepted April 15, 2025

## KEYWORDS:

Antibiotic,  
*Aspergillus*,  
*Ceriops tagal*,  
Diversity,  
Endophytic fungi



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## ABSTRACT

The rapid increase in antimicrobial resistance has encouraged development of new natural and urgent strategies to fight drug-resistant pathogens, one of which is using endophytic fungi. Endophytic fungi found in *Ceriops tagal* mangrove plant, which can adapt to extreme salinity environments of up to 60 ppt. Endophytic fungi are isolated from leaves, branch, and roots. Each isolate was identified morphologically and cultured in potato dextrose broth media for 21 days. After incubation, liquid culture was extracted with ethyl acetate and evaporated to obtain a thick extract. The Kirby Bauer method was used to evaluate the antibiotic potential of each endophytic fungal extract against pathogenic microbes *Staphylococcus aureus*, *Escherichia coli*, and *Candida albicans*. Concentration used was 5%, antibiotics used as positive controls were chloramphenicol (30 µg/disk), nystatin (100 IU/disk), as negative controls DMSO. Three endophytic fungi were isolated from roots (ECT7, ECT 8, ECT 10), two from leaves (ECT 26, ECT 28) and six from branches (ECT 31, ECT 47, ECT 48, ECT 51, ECT 55 and ECT 85). Fungal isolates ECT10 has been proven to effectively exhibit strong antibiotic activity against microorganisms *E. coli*, *S. aureus*, and *C. albicans*, with inhibition zones measuring 13.0 mm, 11.8 mm, and 11.6 mm, respectively.

## 1. Introduction

Mangrove ecosystems, as habitats located in coastal areas harbor extraordinary biodiversity, including endophytic microorganisms that live in plant tissue without damaging their hosts (Rivai *et al.* 2018). Endophytic fungi associated with mangroves have received significant attention in the last two decades (Sruthi *et al.* 2024). Mangrove ecosystems are an important source for the discovery of new endophytic fungal strains and are also known as the second-largest ecosystem (Cadamuro *et al.* 2021). The high

species diversity and environmental adaptability make endophytic fungi a potential source for the production of valuable secondary metabolites (Furtado *et al.* 2019). The environmental adaptability allows endophytic fungi to survive and thrive in a variety of extreme or unusual conditions (Sruthi *et al.* 2024). When faced with environmental changes, endophytic fungi often respond by producing secondary metabolites as a form of protection against stress (Yu *et al.* 2024). These metabolites often have protective properties, one of which is antibiotic activity (Wacira *et al.* 2024).

In addition, bioactive compounds from endophytic fungi have the advantage of a short life cycle (Bibi *et al.* 2020) and easier mass production of metabolites (Eshboev *et al.* 2024). Endophytic fungi isolated from

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mangrove (*Ceriops tagal* (Perr.) C.B Rob) are of particular interest because they can adapt to extreme salinity environments of up to 60 part per thousand (ppt) (Eshboev *et al.* 2024). *C. tagal* contains secondary metabolite compounds such as tannins, alkaloids, flavonoids, cardiac glycosides, saponins, and sterols. New compounds such as taglines, dolichol, cerioptins, tagalons, and cerotagalols have been identified (Manohar *et al.* 2023). *C. tagal* is particularly rich in dolabrance, a subtype of diterpenoid, with forty-five unique dolabranes, many of which possess new bioactive compounds (Istigomah *et al.* 2021). Endophytic fungi produce secondary metabolites that are identical or similar to those of their host plants and are present in relatively high quantities (Sopalun *et al.* 2021). One interesting group of endophytic fungi to study is the genus *Aspergillus*. This genus is a dominant group of fungi isolated from *C. tagal*. As an endophyte, *Aspergillus* can survive in harsh environmental conditions (Hagag *et al.* 2022). In addition, *Aspergillus* is known to produce secondary metabolites with various biological activities, including antibiotics (Revathy *et al.* 2024).

With the increasing global problem of antibiotic resistance, microorganisms are becoming resistant to drugs, making infections caused by bacteria and fungi increasingly difficult to treat and risking an uncontrollable rise in mortality rates (Tang *et al.* 2023). The rapid increase in antibiotic resistance is driving the development of new and natural strategies that are effective against drug resistant pathogenic microbes (Akinduti *et al.* 2022). The discovery of new

antibiotics is an urgent matter (Eshboev *et al.* 2024). To reduce dependence on conventional antibiotics which are increasingly losing their effectiveness (Olawade *et al.* 2024).

This research focused on the species diversity of genus *Aspergillus*, isolates of antibiotic-producing endophytic fungi from *C. tagal*. This study used systematic steps to identify endophytic fungal species of the genus *Aspergillus*. Identification of fungi was carried out based on three main characteristics, namely colonies, macroscopic characteristics, and microscopic characteristics. *C. tagal* is a mangrove plant that can adapt to extreme salinity environments of up to 60 ppt and is known to produce potential secondary metabolites. Therefore, it is hoped that this research can explore the potential of endophytic fungal isolates of the genus *Aspergillus* as producers of effective antibiotics, which can be used to treat infections caused by pathogens, especially those that are resistant to antibiotics.

## 2. Materials and Methods

### 2.1. Sample Collection

Sample collection was carried out in Sungai Pisang, Bungus, Padang City, West Sumatra, Indonesia. Samples were taken from 10 trees with a minimum diameter of 10 cm (Figure 1) (Reis *et al.* 2022). Sample isolation was carried out from roots, branches, and leaves of *C. tagal* that were healthy and did not show insect bite marks (Sophia *et al.* 2025). The leaves taken as samples were the third leaves from the tip of the branch (Agustien *et al.* 2022).

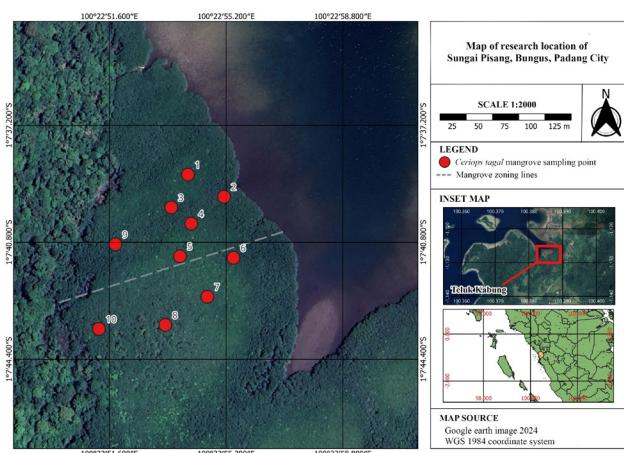


Figure 1. Distribution of sampling point in the Sungai Pisang, Bungus, Padang, West Sumatra (left) host plant of the isolate endophytic fungi and root (right)

al. 2018), while the branches selected had healthy leaves and relatively young roots, characterized by a brighter color and smoother texture (Cadamuro *et al.* 2021).

## 2.2. Isolation of Endophytic Fungi

Samples (leaves, branches, roots) were sterilized on the surface of the organs and planted directly in the growth medium (Sophia *et al.* 2025). Samplings were cleaned first with distilled (5 mins) to eradicate adhered soil particles and surface adhering microorganisms. The surface of the sample was sterilized by dipping it into 70% ethanol for 2 min and then soaking it in sodium hypochlorite for 1 min. Then, the sample was rinsed with sterile distilled water three times (2 min each time) and dried using sterile filter paper. The segments were cut into 2 cm long with a sterile scalpel blade and placed on a Potato Dextrose Agar (PDA) medium supplement with 50 mg/L chloramphenicol to prevent the growth of bacteria. In addition, incubated for 3-7 days at 27-30°C. The emerging endophytic fungi from the explants were isolated, purified, and maintained by continuous subculturing. The final rinse of sterile distilled water is considered a control measure. The media dish impregnated with 0,1 ml of lash-rinsed sterile distilled water with no growth of microbes determines the success ratio of the surface sterilization method (Aini *et al.* 2022).

## 2.3. Morphological Identification of Endophytic Fungi

The identification of fungi was conducted based on three main characteristics: colonies, microscopic characteristics and macroscopic characteristics. Macroscopic identification of endophytic fungi included observations on surface and reverse colonies, structure, elevation and pattern. Microscopic identification by observing hyphae, spore type, shape of spores and other specific characteristics by making culture slides to which one drop of lactophenol cotton blue reagent was added (Hapida *et al.* 2021). The characteristic data were compared to the fungal identification books (Watanabe 2010).

## 2.4. Cultivation of Endophytic Fungi

The pure isolate was cultivated in still culture in potato dextrose broth liquid medium in a 500 ml glass bottle. Cultivation is carried out for 21 days at a temperature of 20-25°C (Kiti *et al.* 2022).

## 2.5. Extraction of Endophytic Fungi

The culture of each isolate was macerated with ethyl acetate in a ratio of 1:1 and left for 24 hours (Handayani *et al.* 2019). The ethyl acetate extract was separated from the culture medium using filter paper. The extract obtained was then evaporated using a rotary evaporator to obtain a thick extract. The resulting extract was used to test antimicrobial activity (Neamul *et al.* 2022).

## 2.6. Screening for Antimicrobial Activity

Antimicrobial activity screening using the Kirby and Bauer (paper disc) method (Sundar *et al.* 2024). Antimicrobial activity was assessed against the bacteria *Staphylococcus aureus*, *Escherichia coli*, and the fungus *Candida albicans*. Nutrient agar and sabouraud dextrose agar containing a microbial suspension were added to petri dishes (0,5 and 0,7 Mc Farland for bacterial and fungus, respectively). A 6 mm sterile paper disc was dripped with 10 µL of extract, and placed on the agar surface. 5% DMSO was used as a negative control (Zhou *et al.* 2022) chloramphenicol (30µg/disk) and nystatin (100 UI/disk) as a positive control (Lestari *et al.* 2019). Petri dishes were incubated at 37°C for 18-24 hours for bacteria and 25-27°C for 3-7 days for fungi (Haryani *et al.* 2019). Measure the clear zone using a digital vernier caliper. The resulting extract is declared to have antimicrobial activity if a clear zone is found around the paper disc. Criteria inhibition zone categories (small 5 mm), weak (6-10 mm), strong (11-20 mm), and very strong (larger than 21 mm) (Lestari *et al.* 2019). Repetition was carried out three times (Sandrawati *et al.* 2023).

## 3. Results

### 3.1. Isolation of Fungal Endophytes

*C. tagal* were collected from the Sungai Pisang, Bungus, Padang City, West Sumatra, Indonesia from roots, branches, and leaves. Endophytic fungi have been isolated from plant organs, as shown in Figure 2.

To ensure that what grows is endophytic fungi, there was a control, namely sterile distilled water used for surface sterilization, taken 0.1 ml and incubated for 14 days, as seen in Figure 2 (A). In the control medium, no fungi grew, thus indicating that the fungi growing on the PDA medium are endophytic fungi from *C. tagal*.

Purification of the isolate was carried out by re-culturing on PDA media based on the color characteristics

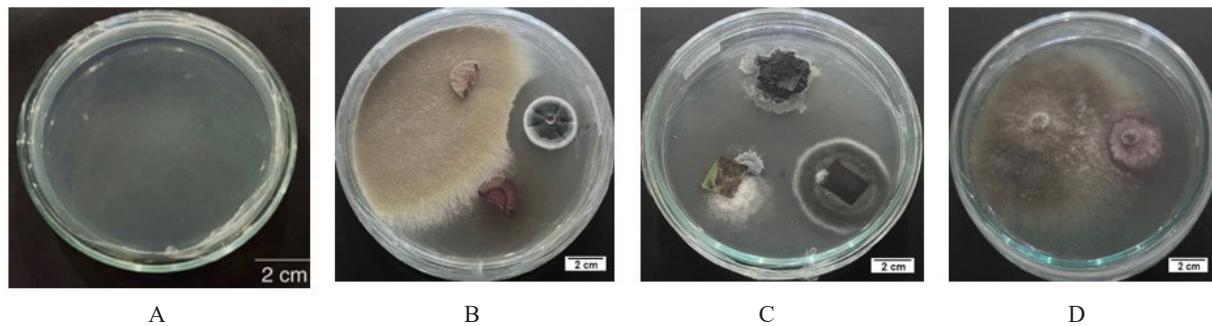


Figure 2. Isolation of endophytic fungi from mangrove *C. tagal* organ on PDA A: control, B: root, C: leave, D: branch

of the colony. Fungal isolates are identified based on three main characteristics: colony, macroscopic features, and microscopic features. The identification results are compared with the literature and keys used to determine the fungus. *Aspergillus* has distinctive colors such as green, yellow, or black, with a rough or hairy surface. Furthermore, microscopic analysis of the conidia and sporangium structure is conducted. *Aspergillus* has spherical or elliptical conidia located at the tips of conidiophores, as well as round or oval sporangia, which are characteristic of this genus. A total of eleven endophytic fungi of the genus *Aspergillus* were successfully isolated from different plant parts, namely roots, leaves, and branches of *C. tagal*. Three isolates from roots (ECT 7, ECT 8, ECT 10), two isolates from leaves (ECT 26, ECT 28), and six isolates from branches (ECT 31, ECT 47, ECT 48, ECT 51, ECT 55, ECT 85). Macroscopic characteristics, including front and back views, together with microscopic results of endophytic fungi isolated from roots, leaves, and branches are shown in Figure 3. Colonies formed by endophytic fungi of the genus *Aspergillus* grown on PDA media vary in color and texture. Morphological characteristics of fungal colonies isolated from sections for each isolate (Table 1 and Table 2).

The isolation and identification of endophytic fungi from three plant organs roots, branches, and leaves revealed that no fungal isolates were the same across all three organs. Macroscopic observations showed that the colonies of fungi growing from each organ exhibited different characteristics. The colonies from the roots were predominantly dark brown with a dense, hairy texture, the isolates from the leaves were lighter, with white and yellowish colors, while those from the branches formed colonies that were cream-colored, green, and had a smooth surface. Microscopic observations of the endophytic fungal isolates revealed that all isolates had conidia spores with a globose (round) shape. All isolates showed septate hyphal structures, meaning they had cross-

walls along the hyphal filaments. Additionally, all isolates were classified into the genus *Aspergillus* based on their distinctive microscopic characteristics.

Several specific characteristics were observed in each isolate. Isolates ECT 7 and ECT 10 exhibited globose conidia with septate hyphae that were hyaline and colorless. Isolates ECT 8, ECT 31, ECT 51, ECT 55, and ECT 85 showed the presence of long conidiophores with the characteristic columnar structure of *Aspergillus*. Isolate ECT 26 demonstrated the ability to produce abundant aerial hyphae. Isolate ECT 28 exhibited long conidiophores with rough walls, while ECT 47 had long conidiophores without additional structures. Isolate ECT 48 displayed a distinctive structure of phialides arranged in a single row, which is also characteristic of several *Aspergillus* species.

### 3.2. Species Diversity of Endophytic Fungi

The species diversity of endophytic fungi of the genus *Aspergillus* in each isolated part of the *C. tagal* plant showed significant variation (Figure 4).

The isolation and characterization of endophytic fungi from three plant organs roots, branches, and leaves resulted in 11 isolates with varying morphological characteristics. Three *Aspergillus* genus isolates were found in the roots, while two *Aspergillus* genus isolates were found in the leaves. Meanwhile, *Aspergillus* genus isolates were more commonly found in the branches. The endophytic fungi of the *Aspergillus* genus exhibited diverse distribution across the parts of *C. tagal*. Macroscopic and microscopic observations showed that no identical endophytic fungal isolates were found among the three organs. Each organ produced isolates with unique characteristics in terms of colony color, texture, growth patterns, and microscopic structures. These results indicate that although all isolates belong to the *Aspergillus* genus, the different morphological and microscopic characteristics between the organs suggest the presence of species or strain diversity, influenced

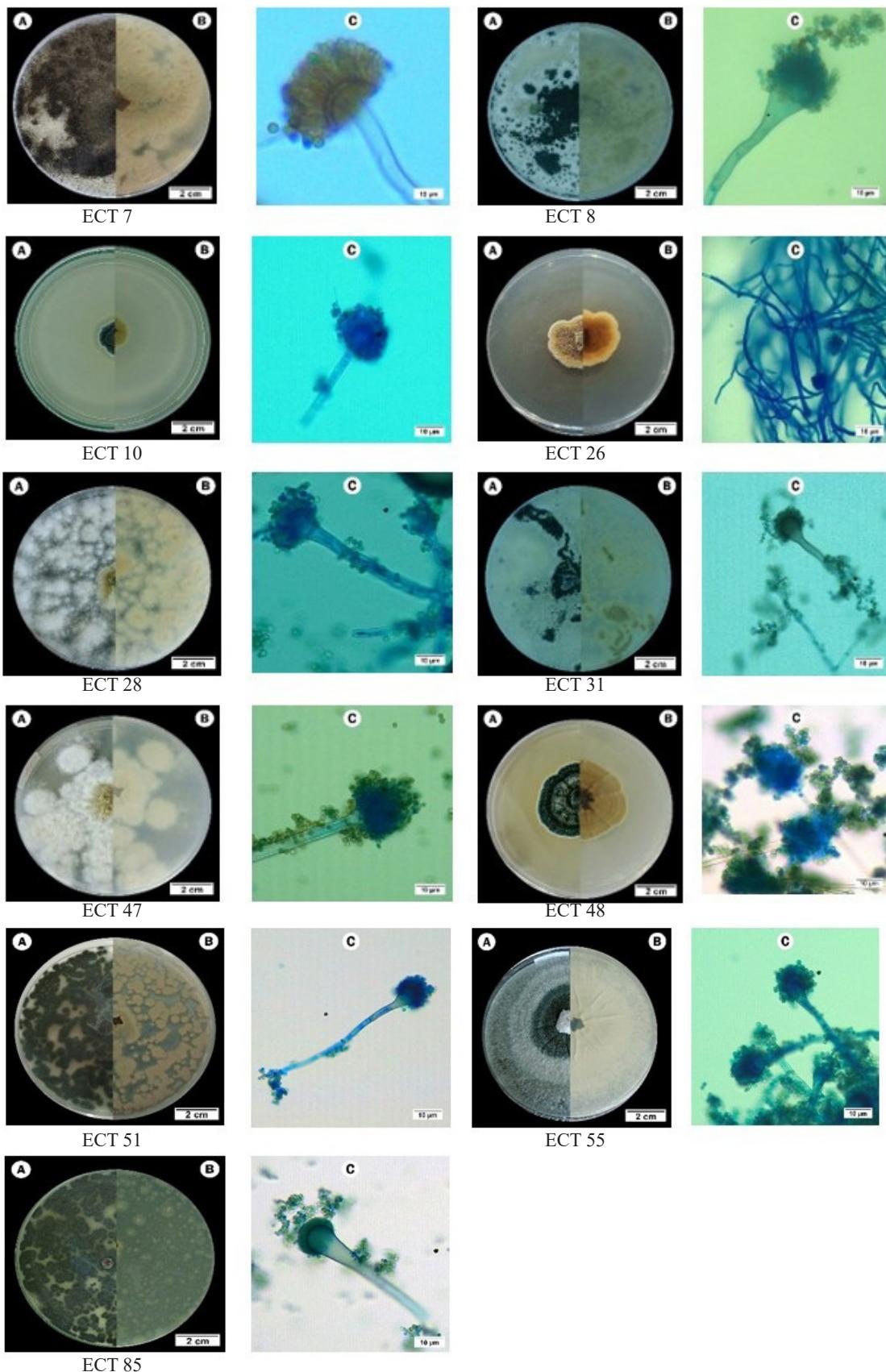


Figure 3. Macroscopic characteristics. A: front view, B:reverse view and C microscopic characteristics of endophytic isolated from *C. tagal*

Tabel 1. Colony characteristics of endophytic fungi from roots, leaves and branches

Code	Surface colony	Reserve colony	Structure	Elevation	Pattern
ECT 7	Black with white border	Cream	Powdery	Umbonate	Circular
ECT 8	Moss green with a white edge	Cream	Powdery	Umbonate	Irregular
ECT 10	Teal green with a white edge	Cream	Velvety	Umbonate	Circular
ECT 26	Yellowish brown with a white edge	Brown with yellow edges	Velvety	Flat	Irregular
ECT 28	White with a green center	Greenish white	Cottony	Flat	Irregular
ECT 31	Moss green with a white edge	Cream	Powdery	Flat	Irregular
ECT 47	Cottony white with a green center	Cream	Cottony	Raised	Circular
ECT 48	Blue with white edges	Cream	Velvety	Umbonate	Circular
ECT 51	Moss green	Thick cream	Powdery	Umbonate	Irregular
ECT 55	White with a green edge	Cream	Velvety	Umbonate	Circular
ECT 85	Moss green	Cream	Powdery	Umbonate	Irregular

ECT 7, ECT 8, ECT 10 endophytic fungal colonies isolated from the root; ECT 26, 28 endophytic fungal colonies isolated from the leaves; ECT 31, ECT 47, ECT 48, ECT 51, ECT 55, ECT 85 endophytic fungal colonies isolated from the branch

Tabel 2. Microscopic characteristics of the isolated endophytic fungi from roots, leaves and branches

Isolate	Spore	Shape	Hyphae	Characteristic	Genus
ECT 7	Conidia	Globose	Septate	Hyaline colorless	<i>Aspergillus</i>
ECT 8	Conidia	Globose	Septate	Long conidiophore with columnar structure	<i>Aspergillus</i>
ECT 10	Conidia	Globose	Septate	Hyaline colorless	<i>Aspergillus</i>
ECT 26	Conidia	Globose	Septate	Can produce abundant aerial hyphae	<i>Aspergillus</i>
ECT 28	Conidia	Globose	Septate	Long, rough walled conidiophore	<i>Aspergillus</i>
ECT 31	Conidia	Globose	Septate	Long conidiophore with columnar structure	<i>Aspergillus</i>
ECT 47	Conidia	Globose	Septate	The Conidiophore are long	<i>Aspergillus</i>
ECT 48	Conidia	Globose	Septate	Phialides single row	<i>Aspergillus</i>
ECT 51	Conidia	Globose	Septate	Long conidiophore with columnar structure	<i>Aspergillus</i>
ECT 55	Conidia	Globose	Septate	Long conidiophore with columnar structure	<i>Aspergillus</i>
ECT 85	Conidia	Globose	Septate	Long conidiophore with columnar structure	<i>Aspergillus</i>

ECT 7, ECT 8, ECT 10 endophytic fungal colonies isolated from the roots; ECT 26, ECT 28 endophytic fungal colonies isolated from the leaves; ECT 31, ECT 47, ECT 48, ECT 51, ECT 55, ECT 85 endophytic fungal colonies isolated from the branches

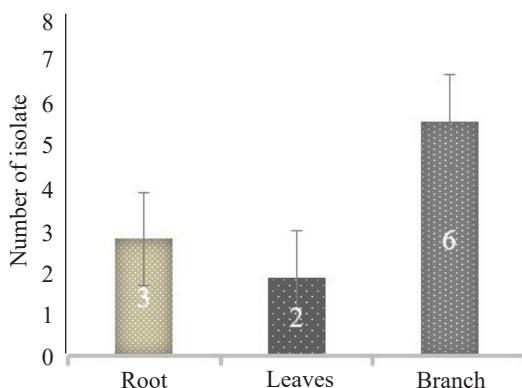


Figure 4. Number of isolate endophytic fungi genus *Aspergillus* isolated from *C. tagal*

by the microenvironmental conditions of each part of the plant.

The results of incubation of endophytic fungi of the genus *Aspergillus* in PDB media for 21 days under static conditions showed that the formed mycelium generally floated on the surface of the press or grew as clumps that spread in the liquid media. The color of the mycelium

varies, some are grayish-black, greenish-yellow, bluish-green, and cream. The cultivation results can be seen in Figure 5.

### 3.3. Antimicrobial Activity

Endophytic fungi of the genus *Aspergillus* isolated from *C. tagal* plants were extracted with ethyl acetate. Eleven isolates of the endophytic fungal genus were tested for antimicrobial activity using the agar well diffusion method (Figure 5). Antimicrobial activity against pathogenic microorganisms can be seen in Table 3.

The results showed that 9 isolates of endophytic fungi of the genus *Aspergillus* ECT 8, ECT 10, ECT 28, ECT 31, ECT 47, ECT 48, ECT 51, ECT 55, ECT 85 were able to inhibit microorganisms in the strong and weak categories. The ECT 8 isolate shows an inhibition zone against *E. coli* and *S. aureus* with a weak category but does not affect *C. albicans*. The ECT 10 isolate shows an inhibition zone against *E. coli*, *S. aureus*, and *C. albicans* with a strong category. The ECT 28 isolate shows an inhibition zone against *E. coli*, *S. aureus*, and

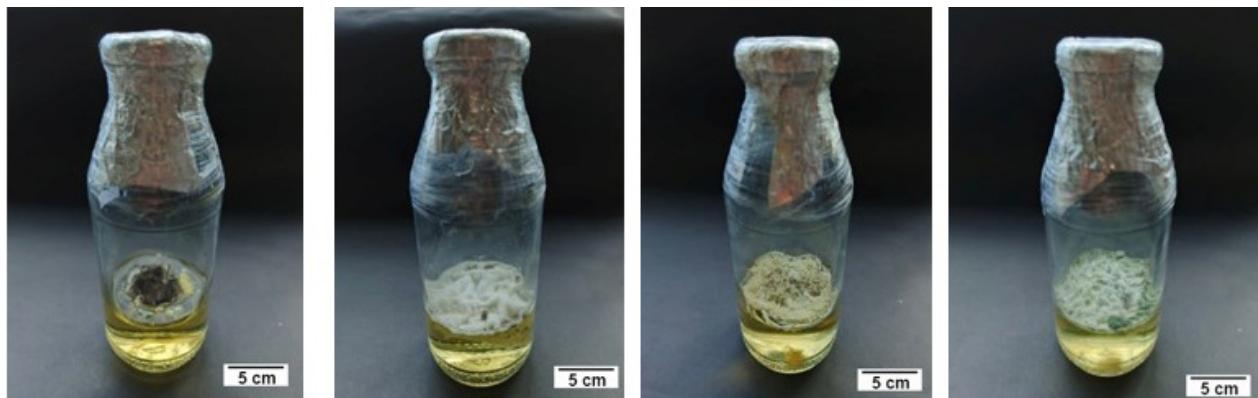


Figure 5. The endophytic fungus of the genus *Aspergillus* was incubated in PDB for 21 days

Table 3. Antimicrobial activity of endophytic fungal extract from the roots, leaves, and branches

Source of plant	Isolate	Zona of inhibition (mm)			Category		
		<i>E.Coli</i>	<i>S.aureus</i>	<i>C.albicans</i>	<i>E.Coli</i>	<i>S.aureus</i>	<i>C.albicans</i>
Root	ECT 7	-	-	-	-	-	-
	ECT 8	9.7	8.2	-	Weak	Weak	-
	ECT 10	13.0	11.8	11.6	Strong	Strong	Strong
Leaves	ECT 26	-	-	-	-	-	-
	ECT 28	7.2	7.4	8.2	Weak	Weak	Weak
Branch	ECT 31	8.6	-	-	Weak	-	-
	ECT 47	8.5	-	-	Weak	-	-
	ECT 48	-	-	6.6	-	-	Weak
	ECT 51	9.2	-	9.8	Weak	-	Weak
	ECT 55	8.2	10.5	8.8	Weak	Weak	Weak
	ECT 85	-	-	8.8	-	-	Weak
	Positive control	24	24	28	Very Strong	Very Strong	Very Strong

\* positive control *E. coli*, *S. aureus* :chrolamphenicol; positive control *C. albicans* :nystatin

*C. albicans* with a weak category. The ECT 31 and ECT 47 isolates show an inhibition zone against *E. coli* with a weak category, but no inhibition against *S. aureus* or *C. albicans*. The ECT 48 isolate shows an inhibition zone against *C. albicans* with a weak category but does not affect *E. coli* or *S. aureus*. The ECT 51 isolate shows an inhibition zone against *E. coli* and *C. albicans* with a weak category, but no inhibition against *S. aureus*. The ECT 55 isolate shows an inhibition zone against *E. coli*, *S. aureus*, and *C. albicans* with a weak category. The ECT 88 isolate shows only an inhibition zone against *C. albicans* with a weak category and does not affect *E. coli* or *S. aureus*. Stronger antibiotic activity was only found in ECT 10 (from the root), which showed larger inhibition zones and can be categorized as strong against all three tested microorganisms. This indicates a great potential for development as an antimicrobial agent.

Based on the research results, antimicrobial activity shows that the ECT 10 isolate has the potential to produce

antibiotics with strong activity against *Escherichia coli*, *Staphylococcus aureus*, and *Candida albicans*. The inhibition zone can be seen in Figure 6.

#### 4. Discussion

This research succeeded in analyzing the morphological characteristics of the species diversity of endophytic fungi of the genus *Aspergillus* isolated on *C. tagal* plants in the from the Sungai Pisang, Bungus, Padang City, West Sumatra, Indonesia. Eleven of species of the *Aspergillus* genus were successfully isolated from *C. tagal*. The diversity of endophytic fungi between species of the same host plant is strongly influenced by differences in host habitat conditions (Lee et al. 2019). The diversity of endophytic fungi found in different organs of the same plant indicates the specificity of endophytic communities to host tissue types (Wang et al. 2022). In this study, isolates obtained

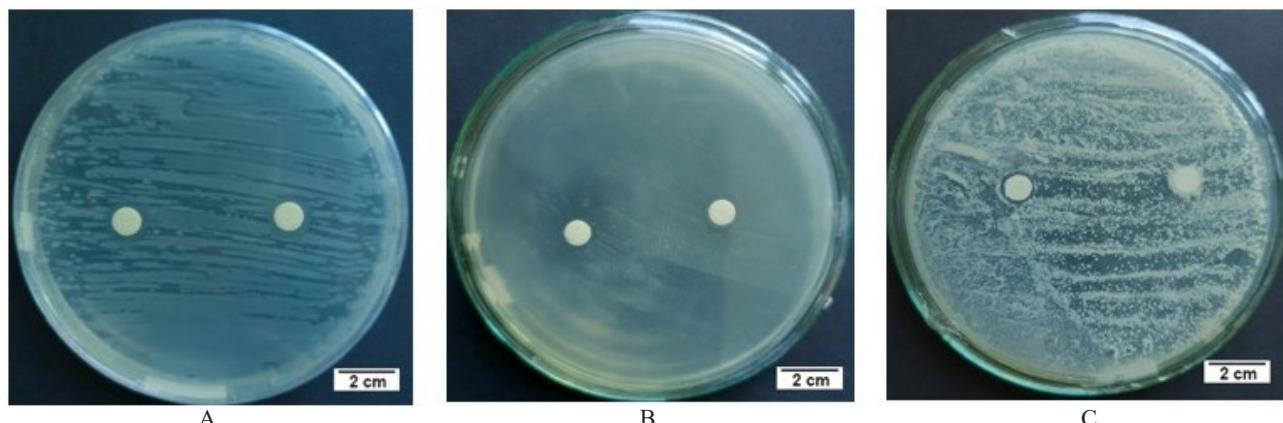


Figure 6. The agar plate picture of the inhibition zone of fungal isolate extract against the growth of (A) *Escherichia coli* (B) *Staphylococcus aureus* (C) *Candida albicans*

from the roots, branches, and leaves exhibited different macroscopic and microscopic characteristics, even though they all belong to the same genus *Aspergillus*. These differences reflect that each plant organ provides a unique microenvironment that is selective for fungi. The roots, as underground organs, have high moisture and nutrient access, as well as direct contact with the soil, which serves as a source of microbial inoculum. In contrast, the leaves and branches are located above ground, exposed to light, air, and temperature fluctuations, which can influence the types of fungi that can survive and thrive within them (Revathy *et al.* 2024). *Aspergillus* is the dominant genus of endophytic fungi isolated from Sungai Pisang, Bungus, Padang City, West Sumatra, Indonesia. This is in line with research conducted by (Revathy *et al.* 2024) that *Aspergillus* is the dominant genus of endophytic fungi isolated in *C. tagal* from Kollam, India. This result is also supported by (Suciatmih 2015) that *Aspergillus* is the dominant genus of endophytic fungi isolated in North Sulawesi.

*Aspergillus* fungi cultured on PDA media show colony growth with characteristics in the form of a fine texture such as powder, velvety, and hairy, as well as varying colors, such as green, yellow, black, and white. The edges of the colony are visible, often in a regular pattern. The *Aspergillus* genus has distinctive microscopic characteristics, which differentiate it from other fungal genera. Fungal hyphae are insular and branched, with a transparent or slightly pigmented structure, conidia are round or oval (Abdelgawad *et al.* 2022). This is in line with research (Qi *et al.* 2024) that the *Aspergillus* genus has a smooth, velvety texture on the surface of fungal colonies and if scratched, it

takes the form of powder. The shape of the fungal colony is irregular, the growth pattern is spread out, the elevation type of the fungal colony is flat, and the edges of the fungus are jagged. The *Aspergillus* genus has microscopic features in the form of clavate vesicles with fields/sterigmata attached directly to almost all parts or 3/4 of the vesicle (Makhlof *et al.* 2019). The transparent/clear conidiophores are long and the conidia are globose (round) (Jing *et al.* 2022).

The results obtained showed significant variations in the number and types of *Aspergillus* isolates found in various parts of the plant, namely roots, leaves, and branch. Overall, isolates of the endophytic fungus of the genus *Aspergillus* found on *C. tagal* showed quite high diversity, with 3 species found on roots, 2 species on leaves, and 6 species on branch. This shows that the endophytic fungi of the genus *Aspergillus* have different distributions in each part of the plant, with a higher prevalence of branch. The results of the research show that parts of *C. tagal* that are directly exposed to the external environment, such as branch, tend to have a higher diversity of endophytic fungi than other parts, such as roots and leaves. This is caused by differences in microhabitat conditions in each part of the plant (Revathy *et al.* 2024). The research results are in line with research conducted by (Zanudin *et al.* 2020) which reported that colonization of endophytic fungi on branches was higher than on leaves. The potential of endophytic fungi to colonize inter or intracellular plants depends on their ability to utilize different substrates synthesized in different parts of the plant host.

Isolate ECT 10 of the genus *Aspergillus* has strong potential to inhibit the growth of *Escherichia coli*, *Staphylococcus aureus*, and *Candida albicans*

microorganisms. The results of this study are similar to research conducted by (Mohamed *et al.* 2022). *Aspergillus* compound extract showed antibacterial activity against *Pseudomonas aeruginosa* and *Escherichia coli*. In addition, endophytic *Aspergillus* species are the main source of antimicrobial metabolites (Mohamed *et al.* 2022). *Aspergillus* strains are one of the most productive sources of secondary metabolites with diverse chemical classes and interesting biological activities (Padhi *et al.* 2017). The isolated metabolites vary chemically and show various biological activities such as antibacterial, anticancer, anti-plasmoidal, anti-inflammatory, and antibiotic (Hagag *et al.* 2022).

*Aspergillus* is an endophytic microbe that is productive in producing metabolite compounds. The results of research conducted (Dharmawati *et al.* 2024) show that the endophytic fungus *Aspergillus* originating from mangroves has the most antibacterial bioactive compounds, biphenyl derivatives, steroids, and isoprenoids, anthracene derivatives, biphenyl derivatives, alkaloids and polyketides (Wei *et al.* 2024). Endophytic fungi of the genus *Aspergillus* which originate from marine environments such as mangroves are capable of producing metabolite compounds with various chemical structures such as fatty acids, polyketides, sterols, alkaloids, terpenoids, peptides, butenolides, and others. The bioactivity of compounds has antimicrobial properties (Orfali *et al.* 2021). The research results are in line with (Bai *et al.* 2022). *Aspergillus niger* JX-5 isolated from *C. tagal* leaves contains a new piperazine-dione derivative bioactive compound, nigerpiperazine A.

Similarly, several bioactive compounds with antimicrobial and cytotoxic activities also have been reported from endophytic fungi isolated *C. tagal* in previous studies. For instance, it was discovered that *Talaromyces assiutensis* JTY2 produces alkaloids called pyranonigrins A and S, which have a wide range of antifungal activity against a variety of phytopathogenic fungus (Li *et al.* 2022). In addition, study conducted by (Deng *et al.* 2020) showed that fungal endophytes of *C. tagal* were also capable of producing steroids. He isolated and characterized ergosterols, sitosterol, and stigmasterone from *Cytospora* sp. cultures and found (22E,24R)5,8-epidioxy-5 $\alpha$ , 8 $\alpha$ -ergosta-6,22E-dien-3 $\beta$ -ol to be the most active against *Pseudomonas aeruginosa* and *Bacillus subtilis* with MIC values of 58.3  $\mu$ M. Moreover, (Putra *et al.* 2023) isolated *C. tagal* from endophytic fungus *Hypoxyylon mangrovei*,

exhibited strong inhibitory effects against *S. aureus* and *S. epidermidis*, with minimum inhibitory concentrations (MIC) of 125 and 250  $\mu$ g/ml-1, respectively. Overall, this study demonstrates the prospective antimicrobial properties of endophytic fungi from the *Aspergillus* genus species isolated from *C. tagal*.

## Acknowledgements

This research was funded by Andalas University with the Doctoral Dissertation Research Scheme for the 2024 fiscal year No. 24/UN16.19/PT.01.03/PDD/2024. The author also thanks all the teams who have supported this research.

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