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ANTIFUNGAL ACTIVITY FROM STEM AND LEAF CAPA EXTRACT (*Blumea Balsamifera* L.): TRADITIONAL MEDICINAL PLANT FROM SOUTH ACEH, INDONESIA

Masyudi[1,2,✉] , Khalisah[3], Saudah[4], Jailani[5], Irhamni[6], and S. Nurman[7]
[1]Department of Natural Resources and Environmental Management, Universitas Serambi Mekkah, Banda Aceh 23245, Indonesia
[2]Department of Public Health, Universitas Serambi Mekkah, Banda Aceh 23111, Indonesia
[3,4]Master of Biology Education, Universitas Serambi Mekkah, Banda Aceh 23245, Indonesia
[5]Department of Biology, Universitas Serambi Mekkah, Banda Aceh 23245, Indonesia
[6]Department of Environmental Engineering, Faculty of Infrastructure and Regional Technology, Institut Teknologi Sumatera, Lampung Selatan, 35365, Indonesia.
[7]Department of Ship Engineering, Politeknik Pelayaran Malahayati. Jl. Laksamana Malahayati No.Km.19, Durung, Mesjid Raya, Aceh Besar 23381, Aceh,
Corresponding author: masyudi@serambimekkah.ac.id

ABSTRACT
The use of synthetic chemicals for treating diabetic wounds has not yet demonstrated full effectiveness in promoting wound healing. Diabetic wounds generally heal slowly and are often referred to as gangrene. Fungal infections are commonly found in these wounds, further delaying the healing process. Therefore, alternative therapeutic agents derived from natural products are needed, as they are generally safer, have fewer side effects, and can accelerate wound healing in an environmentally friendly manner. The capa plant (*Blumea balsamifera* L.) has the potential to act as a treatment for diabetic wounds and to inhibit fungal growth. The urgency of this research lies in developing the potential of local medicinal plants as new resources to replace synthetic chemicals that may cause adverse effects. This study also aims to explore additional benefits of medicinal plants traditionally used by communities for various diseases. This research was conducted through laboratory experiments, including the extraction of capa plant materials, phytochemical analysis of capa leaves, and antifungal assays comparing leaf and stem extracts using different solvents. The results showed that both leaf and stem extracts of capa were capable of inhibiting the growth of *Candida albicans*. The largest inhibition zone was observed in the stem extract using ethyl solvent (10 mm). The stem extract exhibited stronger antifungal activity than the leaf extract, and both extracts demonstrated moderate inhibitory effects comparable to the positive control antifungal medicine. These findings indicate that capa leaves and stems contain antifungal compounds and have the potential to be developed as active ingredients for diabetic wounds.
Keywords: antifungal Activity, *Blumea bassamifera*, Traditional Medicinal Plant.
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INTRODUCTION

Capa plant (*Blumea balsamifera* L. DC), also known as Sembung, has shown promising antifungal activity against Candida albicans in recent studies. The essential oil extracted from *B. balsamifera* leaves has demonstrated significant antimicrobial properties, including antifungal effects against C. albicans with a minimum inhibitory concentration (MIC) of 1.2 mg mL^{-1}.[1] This activity is attributed to the plant's rich phytochemical composition, including compounds like (-)-borneol, trans-caryophyllene, and camphor.[2] Interestingly, the efficacy of *B. balsamifera* extracts varies depending on the extraction method and the plant's geographical origin. For instance, ethanolic extracts of *B. balsamifera* collected from a geothermal area in Indonesia contained proximadiol as the major compound, which may contribute to its potential therapeutic applications.[3] Additionally, icthyothereol acetate and cryptomeridiol isolated from *B. balsamifera* leaves have shown moderate to low antifungal activity against *C. albicans*.[4] In conclusion, while *B. Balsamifera* extracts have demonstrated promising antifungal activity against C. albicans. More research is needed to fully understand their mechanisms of action and optimise their potential as an

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antifungal agent. The plant's diverse phytochemical profile and its traditional use in wound healing suggest that it could be a valuable source for developing new antifungal treatments, particularly in the context of increasing drug resistance in *C. albicans* infections. Gangrene is a complication of wounds in diabetic patients that heals slowly and commonly affects the lower limbs, toes, and hands. The World Health Organisation (WHO) estimates that the number of diabetes mellitus patients in Indonesia will increase two- to threefold by 2030, from 8.4 million to 21.3 million people.[5] Diabetic wounds require special wound care, as diabetic patients have high blood glucose levels that inhibit wound healing. Therefore, a special formulation is needed to address this condition.[6,7] *B. balsamifera* L. has long been known among the Acehnese community and is used in traditional medicine for various diseases, including wound healing.[8] In South Aceh, people traditionally apply the leaves directly onto wounded skin. Capa plant extract has anti-inflammatory properties. The flavonoid content of the capa plant during the inflammatory phase increases the activity of the immune system, lymphocyte proliferation, and macrophage activity.[9,10] However, previous research has been limited to the use of the capa leaf extract or oil.[11] The researchers have previously applied capa leaf extract to incision wounds and found it to be more effective than the synthetic chemical products that are widely used. The use of synthetic chemical drugs has side effects and may prolong wound healing, particularly in diabetic patients with gangrene wounds. Therefore, it is necessary to conduct a study that produces innovation by utilizing natural resources traditionally used by the community, namely, capa leaves. The research question is how to formulate a capa leaf gel that is effective for the treatment of gangrene wounds requiring special care. This study aims to examine the potential of the Capa plant (*Blumea balsamifera* L.) as a wound-healing agent by the antifungal test. The results showed that extracts from the stems and leaves of capa were able to inhibit the growth of *Candida albicans* to a moderate degree.

EXPERIMENTAL

Study Location

The plants of the *B. balsamifera* plant were collected from the Geulumbuk village in the Kluet Utara subdistrict of the South Aceh-Indonesia Regency (Fig.-1, coordinates 03º07'13.7"N and 97o20'28.2" E). The location of the sampling was the same as it was in our earlier paper regarding the bioactive chemicals found in *B. balsamifera* leaf extracts from South Aceh, Indonesi.[11]

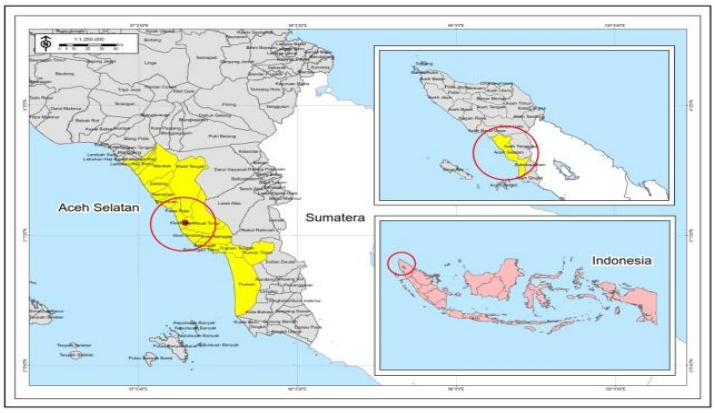

Fig.-1: Location Sampling

A separation of extracts from the stems and leaves of the capa plant (*Blumea balsamifera* L.) was carried out to compare their effectiveness in wound healing. The first stage involved collecting capa leaves and preparing the gel formulation according to the research protocol. The capa plants used in this study were obtained from Geulumbuk Village, North Kluet District, South Aceh, Indonesia. Plant materials were taken from specimens at least four months old. The parts collected were mature but not yellowing stems and leaves. Leaves close to the branches were selected rather than shoot tips, as flavonoids and other phytochemicals are among the main bioactive constituents in the aerial parts of *B. balsamifera*, and these compounds have been associated with antimicrobial and wound-healing activities in traditional and scientific studies. Flavonoids have been identified as key contributors to wound contraction, capillary

regeneration, collagen deposition, and enhanced expression of growth factors involved in skin repair in experimental models.[12–14]

Fig.-2: Capa Plant (*Blumea Balsamifera* L)

In this study, the extraction process of capa (Blumea balsamifera) began with the collection of leaves and stems, which were then washed under running water to remove adhering dirt and dust. The plant materials were subsequently dried at room temperature without direct exposure to sunlight. Drying was carried out by air-drying, in which the materials were placed on racks until completely dry. The dried capa leaf simplicia were then weighed to a total of 1.5 g,[7,15] as shown in Fig.-2 below:

Fig.-3: After Drying, the Capa Plant Materials were Separated into Leaves and Stems

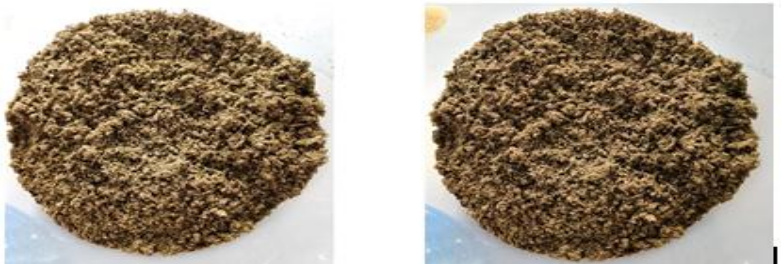

Fig.-4: The Dried Leaves and Stems of the Capa Plant Were Ground into a Fine Powder Using a Dry Blender

After being ground, the powdered leaves and stems of the capa plant were soaked in solvents for the extraction process using the maceration method. The solvents used were ethanol and ethyl acetate. The mixture was then filtered, as shown in the following Fig.-5:

Fig.-5: Maceration and Filtration Process

After filtration, the process was continued with the separation of the extract from the solvent using a rotary evaporator. This procedure was carried out at the Chemistry Laboratory, Faculty of Mathematics and Natural Sciences, Syiah Kuala University. The result of this separation was the leaf and stem extracts of the capa plant, as shown below:

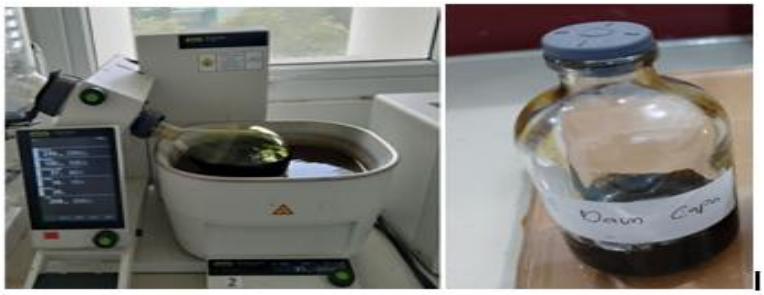

Fig.-6: Evaporation Process for Separating the Solvent from the Capa Leaf Extract

RESULTS AND DISCUSSION

Plant Identification

The plant determination conducted at the Herbarium Laboratory, Faculty of Mathematics and Natural Sciences, Syiah Kuala University, Banda Aceh, confirmed that the capa plant collected from Geulumbuk Village, South Aceh, and used as the research sample, belongs to the genus *Blumea* with the species *Blumea balsamifera* L. This identification was verified based on the official determination letter No. B/647/UN11.1.8.1/TA.00.01/2025. The determination process was essential to ensure the accuracy of the plant identity used as the research sample.

Characteristics of Simplicia

The results of the simplicia characterisation test for capa leaves are presented in Table-1. The moisture content and ash content of both the stem and leaf simplicia complied with the standard requirements for simplicia, which state that the moisture content should not exceed 10%. Simplicia with moisture content exceeding 10% are prone to microbial and fungal growth, which may negatively affect the research outcomes.[16]

Table-1: Results of Simplicia Characterisation Test

No.	Characterization	Capa leaf	Capa stem
1.	Water content	4.20%	4.05%
2.	Ash content	5.5%	4.3%

The ash content of the capa leaf simplicia was 5.5%, while that of the stem simplicia was 4.05%, both of which comply with the standard requirement for total ash content in simplicia, which should not exceed 11%. The determination of ash content in simplicia aims to identify the characteristics of the remaining inorganic components after incineration. The ash value should ideally be low, as this parameter indicates the presence of heavy metal contaminants that remain stable at high temperatures.[16]

Based on the results of the moisture and ash content tests, the capa leaves from Aceh used in this study met the required standards. High moisture content can promote the growth of microorganisms, whereas elevated ash content may interfere with the phytochemical analysis conducted. These findings determine whether the research can proceed to the next stage.

Extract Characteristics

The results of the extract characterisation test for capa leaves are presented in Table-2. The findings show that all three types of extracts produced had acceptable moisture levels, each below 10%. This condition is essential to ensure that the composition of the extract remains stable, as fungal and microbial growth can occur in moist extracts with high moisture content, potentially altering the chemical composition of the bioactive compounds.

Table-2 shows that the total ash content of all three extract types complies with the standard requirement for total ash in extracts, which must be below 11%. This confirms that the capa leaf extracts used in this study meet the established quality criteria.

Table-2: Results of Capa Leaf Extract Characterisation Test

No.	Extract Characteristics	Ethanol	n-heksan	Etil asetat
1.	Water content	2.1 %	2.8 %	3.15%
2.	Ash content	1.35%	1.44%	2.34%

Phytochemical Test

The results of the phytochemical test showed that the ethanol extract of capa leaves contained steroid compounds, indicated by the formation of a green colour after the addition of the Liebermann–Burchard reagent, as presented in Table-3.

Table-3: Phytochemical Test Results of Capa Leaf Extract

No.	Chemical content	Extract capa Etanol	Extract capa n-heksan	Extract capa Etil asetat
1.	Alkaloid	-	-	-
2.	Steroid	+	+	+
3.	Terpenoid	-	-	-
4.	Saponin	+	-	+
5.	Flavonoid	+	-	+
6.	Fenolik	+	+	+
7.	Tanin	+	+	+

Note: (+) indicates the presence of active compounds; (−) indicates the absence of active

Table-3 shows that the ethanol extract of capa leaves also contains flavonoid compounds, indicated by the formation of a green colour; tannins, indicated by a brownish-green colour; and saponins, indicated by the formation of foam. The phytochemical test results of the n-hexane extract revealed the presence of phenolic compounds, shown by the appearance of a green colour after the addition of $FeCl_3$, as well as tannins. Meanwhile, the ethyl acetate extract of capa leaves was found to contain steroids, saponins, flavonoids, phenolics, and tannins. The phytochemical results obtained in this study differ from those reported by Huang.[17], who identified alkaloids, steroids, tannins, and glycosides in *B. balsamifera* leaves. Another study by Isnawati A.[18] also reported that capa plants originating from Malang, Tawangmangu, and Bogor contained tannins and flavonoids. Other research has shown that capa leaf extracts contain alkaloids, flavonoids, terpenoids, and saponins.[19] The phytochemical test results of the capa leaf extracts in this study indicate several differences. The extracts were tested using different solvents: ethanol, n-hexane, and ethyl acetate. Variations in the active compounds detected among the extracts may be influenced by several factors, including the extraction method and the type of solvent used. Nevertheless, it can be concluded that the phytochemical constituents found in Aceh capa leaves are largely similar to those identified in capa leaves from other regions around the world. This demonstrates that Aceh capa leaves contain bioactive compounds relevant to the wound-healing process. *B. balsamifera* leaves from Aceh contain saponins, flavonoids, phenolics, tannins, and steroids, all of which are beneficial for wound healing. Among the three extract types produced, phytochemical identification showed that both the ethanol and ethyl acetate extracts contained similar classes of compounds. In contrast, the n-hexane extract of capa leaves contained only steroids, phenolics, and tannins.

Antifungal Activity

The method used in this antimicrobial assay was the disc diffusion method using Sabouraud Dextrose Agar (SDA) as the growth medium. The materials used in this test were as follows: ethyl extract of capa leaves, ethyl extract of capa stems, ethanol extract of capa leaves, ethanol extract of capa stems, 70% alcohol for sterilising the surface of the BSC table, tissue for cleaning the 70% alcohol, C. albicans isolate as the test microorganism, SDA as the growth medium for C. albicans, and distilled water (Aquadest) for media preparation. Ketoconazole discs were used as the positive control, sterile distilled water as the negative control, blank paper discs for soaking the extracts, and McFarland 0.5 as the turbidity standard.[12,20] The equipment used in this assay included: a Biological Safety Cabinet (BSC) as a sterile workspace during testing, a balance for weighing media and other materials, a spatula for handling media, Erlenmeyer flasks for media preparation, measuring cylinders for media preparation, an autoclave for

sterilizing media and materials, petri dishes for pouring media, growing microorganisms, and conducting the assay, an inoculating loop for collecting microbial colonies, syringes for collecting physiological NaCl, blue or yellow pipette tips for taking bacterial suspensions, a Bunsen burner for heating the inoculating loop, test tubes for preparing the C. albicans suspension, a vortex mixer for homogenizing solutions, micropipettes for taking small volumes of liquid, labels or permanent markers for marking the test plates, tweezers for handling paper discs, plastic wrap for sealing the edges of petri dishes, plastic bags for storing petri dishes during incubation, containers for incubating C. albicans at room temperature, a camera for documenting test results, a caliper for measuring the inhibition or killing zones, and an autoclave for sterilizing the test materials before washing the petri dishes and test tubes.

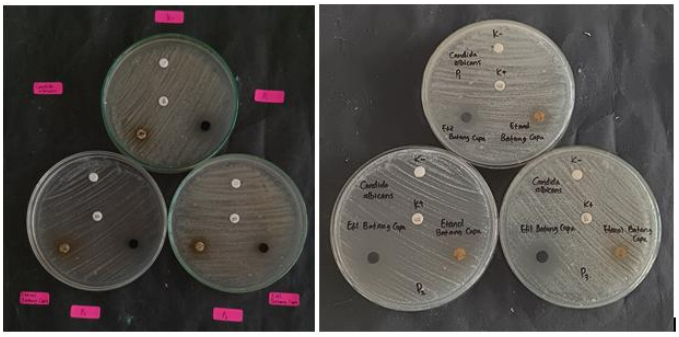

Fig.-7: Observation Results

The procedure for this antimicrobial assay was as follows: The sterilised medium was poured into petri dishes (±20 ml) while still hot and allowed to solidify. After the medium had solidified, it was streaked with *C. albicans* and incubated in a sterile container at room temperature for 24–48 hours. SDA medium was prepared at 20 ml per petri dish, and the test was conducted in three replications for the leaf extract and three replications for the stem extract, requiring a total of 120 ml of SDA. A total of 7.8 grams of SDA medium was weighed, placed into an Erlenmeyer flask, dissolved with distilled water up to 120 ml, covered with aluminium foil, and sealed with plastic wrap. Other materials, such as pipette tips, tweezers, petri dishes, and test tubes, were also sterilised. All items were placed in a plastic bag and sterilised using an autoclave for 15 minutes at 121 °C. After sterilisation, the media were poured and allowed to solidify. The extracts and sterile distilled water were soaked with blank paper discs for 30 minutes. A sufficient amount of physiological NaCl was placed into a test tube. A colony of *C. albicans* was taken using a heat-sterilised inoculating loop and transferred into the test tube. The suspension was homogenised using a vortex mixer, and its turbidity was adjusted to match the McFarland 0.5 standard.

Table-4: Antimicrobial Activity of Capa Leaf Extract Against *Candida albicans*

Replicate	Inhibition Zone Diameter (mm)			
	Positive control	Negative control	Capa extracts	
			Etil	Etanol
1	17.25	0	7.7	7.05
2	16.85	0	7.6	7.2
3	16.95	0	7.25	9.9
Average	17.02	0	7.52	8.05

Positive control: Miconazole zalp
Negative control: Aquades Sterile

For the leaf extract (Table-4), the average inhibition zones produced by the ethyl extract and the ethanol extract were 7.52 mm and 8.05 mm, respectively. This indicates that ethanol was slightly more effective than ethyl acetate in extracting antifungal compounds. The inhibition zones fall within the weak to moderate activity category. Meanwhile, the negative control showed no inhibition zone, confirming that the antifungal activity was derived from the plant's active compounds and not from the solvent or environmental factors. The results of the antimicrobial activity test of the leaf and stem extracts of *B.*

balsamifera against *C. albicans* showed that all extracts exhibited the ability to inhibit fungal growth, although the activity was not as strong as the positive control, miconazole ointment.

Table-5: Antimicrobial Activity of Capa Steam Extract Against *Candida albicans*

Replicate	Inhibition Zone Diameter (mm)			
	Positive control	Kontrol Negatif	Positive control	
			Etil	Etanol
1	17.55	0	10.35	6.3
2	18.3	0	9.9	0
3	18.15	0	9.75	0
Average	18	0	10	6.3

Positive control: Miconazole zalp
Negative control: Aquades Sterile

In the stem extract test (Table-5), the ethyl extract produced a higher inhibition zone (10 mm) compared with the ethanol extract (6.3 mm). This difference suggests that the bioactive compounds present in the stem are more efficiently extracted using ethyl acetate than ethanol. The 10 mm inhibition zone indicates that the ethyl stem extract possesses moderate antifungal activity against *Candida albicans*. When comparing the leaf and stem extracts, the stem extract, particularly with ethyl acetate, demonstrated stronger activity. This may be attributed to a higher concentration of secondary metabolites in the stem tissue, such as flavonoids, tannins, and terpenoids, which are known to possess antimicrobial properties. Overall, the results confirm that *B. balsamifera* has potential as a natural antifungal agent, especially when extracted with ethyl acetate from the stem. However, its activity remains lower than that of miconazole, indicating the need for further studies such as increasing extract concentration, purifying active compounds, or modifying formulations to enhance efficacy. This study reinforces the antifungal potential of *B. balsamifera*. Both the leaves and stems exhibited inhibitory activity against *C. albicans,* although not as strong as the positive control. The leaf extracts showed greater activity when extracted with ethanol, whereas stem extracts were more active when extracted with ethyl acetate. These differences may be related to the solvent selectivity toward secondary metabolites such as flavonoids, saponins, tannins, and terpenoids. The ethyl stem extract, which produced a 10 mm inhibition zone, falls within the moderate activity category, while the other extracts fall within the weak to moderate range. The presence of antifungal activity indicates that *B. balsamifera* has potential as a phytopharmaceutical agent. Nevertheless, since the activity is still lower than that of the positive control, further research is required, such as increasing extract concentration or purifying the active constituents. The present study demonstrated that *B. balsamifera* extracts exhibited antifungal activity against *C. albicans*, with notable differences depending on plant part and solvent used. The ethyl acetate stem extract produced a 10 mm inhibition zone, indicating moderate antifungal activity, whereas the ethanol stem extract showed a smaller inhibition zone (6.3 mm). This suggests that ethyl acetate more effectively extracts relevant bioactive compounds from stem tissue than ethanol, in line with other reports that solvent polarity influences the efficiency of secondary metabolite extraction and antimicrobial effects. Ethyl acetate, being intermediate in polarity, has been shown to extract a broader range of semi-polar compounds such as flavonoids and terpenoids, which contribute to antifungal effects against Candida species.[21] The stronger activity observed in stem extracts (with ethyl acetate) compared with leaf extracts supports the notion that the distribution of secondary metabolites varies among plant organs. Metabolites, including flavonoids, tannins, terpenoids, and saponins, are known to disrupt fungal cell membranes, interfere with ergosterol biosynthesis, and generate oxidative stress in fungal cells, which collectively contribute to growth inhibition. Such patterns are consistent with broader literature indicating that plant extracts rich in these compounds exert antifungal activity in vitro, particularly against Candida species. For example, comprehensive reviews on plant-derived antifungal agents highlight that a wide variety of herbal extracts demonstrate inhibitory activity against *C. albicans* and related species, and that the efficacy depends on phytochemical profiles and extract composition.[22] Solvent selectivity is further corroborated by other studies showing that crude extracts obtained with different solvents exhibit varied antifungal efficacy. In some medicinal plants, less polar solvents (like ethyl acetate) yield extracts with relatively stronger

antifungal effects compared to polar solvents (such as ethanol), reflecting differences in solubility of antifungal constituents such as flavonoids and terpenoids. These observations align with our finding that *B. balsamifera* stem ethyl acetate extract outperformed its ethanol counterpart.[21,23] Despite exhibiting moderate activity, all *B. balsamifera* extracts showed lower inhibitory effects than the positive control, miconazole. This outcome is consistent with general trends reported in the literature, where crude plant extracts often demonstrate less potency than standardised antifungal drugs due to the presence of a complex mixture of compounds and varying concentrations of the active constituents. Given this, optimisation of extraction conditions—such as increasing extract concentration, employing fractionation to enrich active compounds, or using synergistic combinations with conventional antifungals could enhance efficacy. Recent investigations into plant extracts have explored in vitro synergistic interactions with standard antifungal drugs, highlighting the potential for combination therapies to overcome resistance and improve antifungal performance.[23] In this context, the present results support that *B. balsamifera* possesses phytochemical components with inherent antifungal potential. The differing activities between leaf (ethanol) and stem (ethyl acetate) extracts underscore the importance of extraction strategy and plant part selection in evaluating antifungal plant agents. Furthermore, findings from broader studies indicate that plant extracts with diverse secondary metabolite profiles remain promising sources for the discovery of new antifungal compounds and could contribute to developing natural antifungal therapeutics or adjuvants to conventional treatments in the future.

CONCLUSION

The findings of this study demonstrate that *B. balsamifera* leaf and stem extracts possess antifungal activity against Candida albicans, although their effectiveness remains lower than that of the positive control, miconazole. Among the tested extracts, the ethyl acetate stem extract exhibited the strongest inhibition zone (10 mm), indicating moderate antifungal activity. In contrast, the leaf extracts showed weak to moderate activity, with ethanol being slightly more effective than ethyl acetate in extracting bioactive compounds. Differences in antifungal activity between leaf and stem extracts are likely influenced by variations in secondary metabolite content and solvent selectivity toward compounds such as flavonoids, tannins, saponins, phenolics, and terpenoids. These results confirm that *B. balsamifera* contains bioactive components that contribute to fungal growth inhibition and highlight its potential as a natural antifungal agent. However, because the observed activity is still lower than that of miconazole, further research is recommended, including increasing extract concentration, isolating and purifying active compounds, and optimising formulation for enhanced therapeutic effectiveness.

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CONFLICT OF INTERESTS

The authors declare that there is no conflict of interest.

AUTHOR CONTRIBUTIONS

The present work is related to *SDG-3: Good Health and Well-being*. All the authors contributed significantly to this manuscript, participated in reviewing/editing and approved the final draft for publication. The research profile of the authors can be verified from their ORCID ids, given below:

Masyudi https://orcid.org/0000-0002-6322-8745
Khalisah https://orcid.org/0009-0004-5423-9938
Jailani https://orcid.org/0000-0002-3426-5483
Irhamni https://orcid.org/0000-0002-2730-7980
Salfauqi Nurman https://orcid.org/0000-0002-2058-3288
Saudah https://orcid.org/0000-0002-5655-6820

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