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RESEARCH ARTICLE

Phytochemical Screening, Antioxidant and Antifungal Activity Test of Binahong Leaf Extract (*Anredera cordifolia (Ten.) Steenis*)

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ABSTRACT

Anredera cordifolia (Ten.) Steenis (Binahong) is a medicinal plant that has been used by the indigenous Indonesian community to treat various ailments including surgical wounds, stomach ulcers, and skin problems such as itching and sores. _Candida albicans_ is the most important species responsible for infections in patients with immunological problems associated with invasive fungal diseases. The resistance of Candida species to antifungal drugs has made scientists pay more attention to traditional herbal medicine. Binahong contains secondary metabolites such as alkaloids, flavonoids, polyphenols, saponins, steroids and triterpenoids which have antioxidative and antimicrobial activity. This research was conducted to determine the compounds contained in binahong leaf extract using the Harborne method, determine the antioxidant power of binahong leaf extract through DPPH assay, and its antifungal activity using the Kirby-Bauer disc diffusion method against _Candida albicans_. The results of the phytochemical screening confirmed that ethanol extract of binahong leaves contains flavonoids, tannins, saponins and steroid compounds. Meanwhile, methanol extract of binahong leaves contains saponin and steroid compounds. The antioxidant test using the DPPH method showed that ethanol and methanol binahong leaf extracts had weak antioxidant activity, with IC50 values of 370.26 ppm for ethanol extract and 318.85 ppm for methanol extract. Both the ethanol and methanol extracts displayed significant anfungal activity against _Candida albicans_, with extract concentration as low as 6.25%. With the results obtained, it is suggested that ethanol and methanol extracts can be used as antioxidants and antifungals.

Keywords: Antifungal, Antioxidant, Binahong (*Anredera cordifolia* (Ten.) Steenis), Phytochemical.

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1. Introduction

In developing nations, pathogenic microorganisms, including fungi and bacterial strains, represent the predominant etiological factors underlying life-threatening infections, leading to substantial rates of mortality and morbidity among individuals with compromised immune systems [1]. Many antimicrobials available in the market can kill microbes or inhibit the growth of pathogenic microorganisms. However, many microbes are becoming increasingly resistant to these drugs, in fact, many microbes were resistant to various drugs, even developing multidrug resistance that poses major threat to public health [2]. Herbal plants serve as a source of antimicrobials that are relatively safer, natural and inexpensive compared to other sources of antibiotics.

Binahong plant (*Anredera cordifolia* (Ten.) Steenis) with synonyms Boussingaultia gracilis, *Boussingaultia cordifolia*, *Boussingaultia cordata*, originates from China under the name *Dheng Shan Chi* which then spreads to Southeast Asia. In England it is known as Madeira vine or Mignonette vine [3], [4]. The binahong plant has several parts that can be used as alternative medicine, namely roots, stems, leaves, flowers and tubers. The benefits of binahong in the world of medicine are enormous. Binahong has high antioxidant, antifungal and antiviral content. Empirically binahong is used to cure several

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diseases such as swelling of the heart, normalizing blood pressure, kidney damage, diabetes, stroke, vomiting blood, hemorrhoids, constipation, ulcers, typhus, rheumatism, gout, recovery after surgery, shortness of breath, lowering high fever and increasing stamina [4].

Phytochemical compounds in plants function as protection from pests and from the environment, but they can also be used as drugs. Phytochemical tests can be carried out to determine the content of active compounds in plants. Plants generally contain active compounds such as alkaloids, flavonoids, triterpenoids, saponins, steroids, tannins, and quinones [5], [6]. Saponins, steroids, alkaloids and flavonoids have a potential to function as antimicrobial, and many flavonoids also have antioxidative properties [6]–[12]. The phytochemical contents of binahong plant has not been well elucidated.

Antioxidants are electron donor compounds capable of counteracting the formation of free radicals and overcome the negative effects of oxidants in the body such as damage to body cells. In the human body, antioxidants found naturally to balance free radicals that are formed [13]. Free radicals are chemical entities characterized by the presence of unpaired electrons, rendering them inherently unstable. In significant concentrations, these species pose considerable peril to the human organism as they can induce deleterious effects on vital cellular components, including nucleic acids, lipids, and cellular structures. By acting as potent inhibitors, antioxidants play a crucial role in neutralizing and eliminating the aforementioned free radicals, thereby mitigating the risk of various afflictions such as cardiovascular disorders, vascular complications, oncogenesis, and the ageing process within the human physiology [13].

The utilization of natural antioxidants demonstrates enhanced bioavailability, facilitating their absorption within the human body. These naturally occurring antioxidants originate from various botanical and zoological sources, encompassing plant components such as roots, stems, skins, fruits, flowers, and seeds. Primarily, these plant-derived entities comprise a repertoire of hydroxylated compounds, flavonoids, and tocopherols [13].

The 2,2-diphenyl-2-picrylhydrazyl (DPPH) assay is a straightforward and convenient technique that necessitates only a limited quantity of test samples. This method enables the evaluation of radical scavenging activity. Antioxidants participate in a chemical reaction with DPPH free radicals by electron donation, leading to a visible alteration in the initial purple hue of DPPH to a yellow tint. This change in coloration can be quantitatively assessed via a spectrophotometer set at a specific wavelength, such as 517 nm [14]. A reduction in color intensity serves as an indicative measure of enhanced antioxidative capacity, signifying an augmented ability of antioxidants to sequester free radicals. The DPPH assay is applicable for evaluating the antioxidant potential of compounds with both lipid and water solubility. The quantification of antioxidant activity is expressed in terms of IC50 (inhibitory concentration), denoting the concentration of the test extract required to inhibit DPPH activity by 50%. Consequently, a lower IC50 value corresponds to a heightened level of antioxidant activity, implying more effective radical scavenging capabilities [15].

Candida constitutes a prominent etiological factor in a substantial number of fungal infections worldwide [16]. Typically, members of the Candida genus reside as commensal organisms on the cutaneous and mucosal surfaces across the human body. Within the genus Candida, diverse species exhibit significant phylogenetic and phenotypic variability [17]. Concurrently, the surge in resistance against existing antifungal medications persists, necessitating urgent investigations into novel and potent antifungal agents that can address this pressing therapeutic challenge.

2. Method

The study undertaken was an experimental investigation employing samples of binahong leaves (*Anredera cordifolia* (Ten.) Steenis). The analytical procedure entailed a comprehensive phytochemical analysis conducted following the Harborne method [18], antioxidant activity was conducted utilizing the 2,2-diphenyl-2-picrylhydrazyl (DPPH) method [19], while the antifungal evaluation against Candida albicans was performed employing the Kirby-Bauer method [20].

2.1. Simplicial Preparation and Extraction

A total of 10 kg of freshly harvested binahong leaves (*Anredera cordifolia* (Ten.) Steenis) was gathered and subjected to a wet sorting process, aimed at eliminating extraneous substances such as soil, gravel, and vegetation, which might have been inadvertently collected along with the leaves during harvesting. Following this, the binahong leaves were subjected to a meticulous washing procedure using running water. Subsequently, the thoroughly washed leaves were drained and air-dried, employing a method that prevented direct exposure to sunlight, until the moisture content decreased to less than 10% or until a constant weight was attained. Once dried and in the form of simplicia, the samples were finely ground into a powder, which was further subjected to filtration through a 100-mesh sieve for homogenization and particle size uniformity [21].

The extraction procedure involved two different solvents, 70% ethanol and 70% methanol, in conjunction with 330 g of binahong powder. The simplicia was macerated at ambient temperature, employing 990 ml of each solvent, and the process was conducted for a duration of 24 hours, ensuring that the containers were adequately covered with parafilm to prevent contamination. Subsequently, after the 24-hour maceration period, the solutions were filtered using Whatman no. 40 filter paper in conjunction with a chemical funnel to separate the filtrate from the dregs. The filtrate, containing the extracted compounds, was collected and stored in suitable containers. Meanwhile, the remaining dregs were subjected to the maceration process again using a fresh batch of solvent. This maceration step was performed three times to maximize the extraction efficiency. Finally, to obtain the binahong leaf extract, the filtrate collected from the filtering process was subjected to evaporation using a rotary evaporator vacuum device, enabling the removal of the solvent and leaving behind the concentrated extract for further analysis and evaluation [19].

2.2. Phytochemical Screening

The Harborne method was used for the qualitative identification of phytochemical compounds present in the sample. This method relies on observing color changes that occur during specific chemical reactions with targeted phytochemical groups. Notably, the identified compounds encompassed alkaloids, various phenolic groups (including flavonoids, tannins, and saponins), triterpenoids, steroids, and quinones. These results provide valuable insights into the chemical composition of the sample and offer a foundation for further characterization and investigation of its potential bioactive properties [22].

2.3. Alkaloid Test

A sample consisting of 1 g of simplicia extract was introduced into a test tube, and then 3 drops of concentrated ammonia and 5 ml of chloroform were added to the mixture. Subsequently, the resulting solution underwent filtration using filter paper. The filtrate was further subjected to a reaction by adding 1 ml of sulfuric acid. The formation of an acid layer in the solution was carefully collected using a dropper pipette and subsequently divided into three equal portions, each placed in separate wells of a microplate. In the first portion, 3 drops of Dragendorff reagent were added. The appearance of an orange precipitate following this addition indicated a positive result, suggesting the presence of specific alkaloid compounds in the sample. Similarly, in the second portion, 3 drops of Mayer's reagent were added, and the formation of a white precipitate was regarded as a positive result, further confirming the presence of alkaloid compounds in the sample. Finally, in the third portion, 3 drops of Wagner's reagent were added, and the emergence of a brown precipitate confirmed the positive presence of alkaloid compounds in the tested material. These observations and outcomes provide valuable insights into the phytochemical constituents of the simplicia extract, specifically identifying the presence of alkaloids in the sample [18].

2.4. Phenolic Groups Test (Flavonoid, Tanin, Saponin)

A 5-gram quantity of binahong leaf extract was subjected to a heat treatment in distilled water for a duration of 5 minutes. The heated mixture was subsequently filtered through Whatman No. 40 filter paper to obtain the filtrate. The filtrate was then partitioned into three separate tubes for further qualitative analysis. In the first tube, intended for the assessment of flavonoid presence, the following reagents were added: 0.25 g of magnesium powder, 1 drop of amyl alcohol, HCl, and ethanol. The occurrence of a color change to yellowish orange or red in the solution indicated a positive result for the presence of flavonoids. In the second tube, employed to investigate the presence of tannin compounds, 3 drops of 10% ferric chloride were introduced. A color change to black-green in the solution indicated a positive result for the presence of tannins. The third tube was dedicated to the assessment of saponin compounds. Only the filtrate was placed in this tube, and vigorous shaking was performed. The presence of stable foam formation lasting approximately 10 minutes served as a positive indicator for the presence of saponins in the extract. These sequential tests provide valuable insights into the qualitative composition of the binahong leaf extract, specifically identifying the presence of flavonoids, tannins, and saponins. Such information aids in the characterization of the phytochemical constituents of the extract, which may have implications for its potential medicinal and therapeutic applications [18].

2.5. Triterpenoid and Steroid Test

A sample comprising 1 gram of binahong leaf extract was dissolved in hot ethanol and subsequently filtered using Whatman filter paper no. 40 to obtain the filtrate. The filtrate was then subjected to a drying process, resulting in a dry residue. This dry residue was further mixed with 1 ml of diethyl ether using a sonicator for homogenization. To this mixture, 1 drop of concentrated sulfuric acid and

1 drop of anhydrous acetic acid were added. Upon the addition of these reagents, the appearance of a red or purple coloration indicated a positive outcome in the triterpenoid test, signifying the presence of triterpenoid compounds in the sample. On the other hand, the formation of a green or blue coloration in the steroid test indicated a positive result, indicative of the presence of steroid compounds. These specific chemical reactions and color changes provide valuable insights into the presence of triterpenoids and steroids in the binahong leaf extract, contributing to the characterization of its phytochemical composition and potential biological activities [18].

2.6. *Quinone Test*

A test tube containing 1 g of binahong leaf extract was subjected to the addition of methanol and subsequent heating. The resulting mixture was then filtered using filter paper to obtain the filtrate. To this filtrate, 3 drops of 1 N NaOH solution were added. The appearance of a red coloration upon the addition of NaOH indicated a positive result [18].

2.7. *Antioxidant Activity Test*

The sample employed for antioxidant activity testing consisted of binahong leaf extract obtained using 70% ethanol and 70% methanol as the extraction solvents. The evaluation of antioxidant potential was carried out using the 2,2-diphenyl-2-picrylhidrazyl (DPPH) method. As a reference or positive control, vitamin C, was utilized to provide a standard baseline for comparison and validation of the assay results.

2.8. *Preparation of DPPH Solution*

A precisely measured amount of 2.5 mg of DPPH (2,2-diphenyl-2-picrylhydrazyl) powder was meticulously dissolved in high-purity ethanol (pro analysis grade) within a calibrated volumetric flask, subsequently filled up to the 50 ml mark to achieve the desired concentration. To protect the DPPH solution from light-induced degradation, the volumetric flask was securely covered with aluminum foil, shielding it from ambient light exposure.

2.9. *Sample Preparation and Vitamin C*

For the preparation of the samples, precisely 10 mg of binahong leaf extract and an equivalent amount of vitamin C were separately dissolved in 1 ml of dimethyl sulfoxide (DMSO). Both solutions were subjected to homogenization using a sonicator until complete dissolution was achieved.

2.10. *DPPH Test*

A total of 100 µL of each sample, appropriately diluted with high-purity ethanol (pro analysis grade), was dispensed into a microplate at varying concentrations, namely 2000 ppm, 1000 ppm, 500 ppm, 250 ppm, 125 ppm, 62.5 ppm, and 31.25 ppm. For replicates 1 and 2, 100 µL of the DPPH reagent was added to each sample well, while the negative control contained only 100 µL of high-purity ethanol (pro analysis grade). In the blank wells, positioned at the bottom row of the microplate, replicates 1 and 2 were filled with 100 µL of high-purity ethanol (pro analysis grade), and subsequently, 100 µL of the DPPH reagent was added. The negative control wells were filled solely with 200 µL of high-purity ethanol (pro analysis grade). Following the setup, the microplate was incubated at room temperature in a dark environment for 30 minutes. Subsequently, the absorbance of the samples was measured using an ELISA reader at a wavelength of 517 nm to quantify and assess the presence of antioxidant activity in the tested samples [21].

2.11. *Antimicrobial Activity Test (Antifungal)*

The antifungal activity test was performed by the disc diffusion method, also known as the Kirby-Bauer method. Firstly, a microbial suspension, standardized using McFarland 0.5, was uniformly distributed by pouring 1 mL into Petri dishes containing 10 mL of Mueller-Hinton Agar (MHA) medium at 30 °C. The dishes were then gently moved in a figure-eight motion to ensure even distribution and left undisturbed until solidification occurred. All procedures were carried out under sterile conditions to prevent contamination. Subsequently, paper discs were placed on the solidified media mixture, and each disc was saturated with different concentrations of the extraction solution, precisely 50 µL per disc. The concentration range of the extraction solution was prepared at 50%, 25%, 12.5%, 6.25%, and 3.125%. The Petri dishes containing the suspension and the extract-laden paper discs were then incubated at 37 °C for 24 hours. The experimental setup was replicated in duplicate to ensure reliability and accuracy. After incubation, the clear zones surrounding the paper discs were measured using a vernier caliper. The results indicated that the extract exhibited the ability to inhibit the growth of bacteria and fungi, as demonstrated by the presence of clear zones around the discs, signifying the potential antifungal activity of the tested extract [20].

2.12. *Data Analysis*

The collected data were subjected to statistical analysis using the Statistical Package for the Social Sciences 28.0 (SPSS) computer program. The analytical approach utilized was the one-way analysis of variance (ANOVA) test, specifically suitable for samples with a size of less than 30. The primary objective of this test was to examine the relationship between two variables, namely the dependent variable (fungal growth inhibition) and the independent variable (type and dosage of binahong leaf extract). This analysis aimed to assess whether there were any significant effects resulting from the application of different types and concentrations of binahong leaf extract on the inhibition of fungal growth. Prior to conducting the Anova test, the data underwent a normality assessment using the Kolmogorov-Smirnov test to verify if it adhered to a normal distribution. Additionally, the Homogeneity of Variance test was employed to assess the homogeneity of variances across the data.

The interpretation of the Anova analysis is as follows: If the p-value obtained from the Anova test is less than 0.05 ($p < 0.05$), it indicates that the binahong leaf extract had a significant effect on inhibiting fungal growth, and further, there are differences in the effectiveness of the extract among different treatment groups. To identify which treatment groups exhibited significant differences, the post hoc analysis using the Least Significant Difference (LSD) method was applied. Conversely, if $p > 0.05$, it suggests that binahong leaf extract did not have a significant effect on inhibiting fungal growth.

3. Result and Discussion

3.1. *Phytochemical Screening*

Phytochemical screening was conducted on the binahong leaf extract (*Anredera cordifolia* (Ten)) to qualitatively identify the presence of various secondary metabolites. The tests encompassed the detection of alkaloids, flavonoids, tannins, saponins, steroids, triterpenoids, and quinones. The outcomes of the phytochemical screening for the binahong leaf extract are presented in Table I revealing the presence or absence of these specific secondary metabolites in the extract under investigation. This comprehensive analysis serves to provide insights into the phytochemical profile of the binahong leaf extract, contributing to a better understanding of its potential biological activities and medicinal applications.

The results presented in Table I indicate that the ethanol extract contained flavonoids, tannins, saponins, and steroid compounds. On the other hand, the methanol extract was shown to contain saponins and steroids.

Flavonoids, a subgroup of the phenolic compounds, display characteristic color changes, such as the formation of an orange hue, when subjected to chemical reactions involving metal ions (e.g., Mg) and hydrochloric acid (HCl), leading to the reduction of the flavonoid compounds [23]. Flavonoids exert antimicrobial effects by disrupting cell walls, causing bacterial cell death. They also display robust antioxidant capacity, which empowers flavonoids to serve as protective agents against various diseases, conferring anti-atherosclerotic, anti-inflammatory, and antitumor effects. The multifaceted pharmacological attributes of flavonoids underscore their potential as promising therapeutic agents for combating a wide array of health conditions. However, further research and comprehensive investigations are warranted to unravel the full spectrum of their molecular interactions and therapeutic applications in human health and disease [6], [24].

TABLE I: The Phytochemical Test Results of the Binahong Leaf Extract Indicates that the Active Compound is Present (+) and Absent (−)

Active compound	Extract	
	Ethanol 70%	Methanol 70%
Alkaloid:		
-Wagner	–	–
-Mayer	–	–
-Dragendorf	–	–
Flavonoid	+	–
Tannin	+	–
Saponin	+	+
Steroid	+	+
Triterpenoid	–	–
Quinone	–	–

Tannins, belonging to the phenolic group, are characterized by their bitter taste. In plants, tannins serve as vital metabolic regulators and confer protection against herbivores [6]. In the presence of $FeCl_3$ during testing, the formation of complex compounds between Fe and tannins results in a distinctive greenish-black color [23]. Functionally, tannins exhibit a diverse range of biological activities, acting as anti-diarrheal agents, antibacterial agents, and antioxidants [25], [26].

Saponins, renowned as natural surfactants, demonstrate the ability to reduce surface tension in cells, leading to the disruption of bacterial membranes [7], [8]. In the saponin test, vigorous shaking leads to the formation of stable foam for approximately 10 minutes, signifying the presence of surface-active polar and non-polar groups that facilitate foaming in the test [23].

Steroids have been employed in therapeutic applications, including antibacterial, antifungal, anti-inflammatory, and anticancer treatments [10]. In the testing process, the addition of concentrated sulfuric acid and anhydrous acetic acid reacts with the binahong leaf extract, producing a distinct green color, providing evidence of the presence of steroids in the extract [23].

3.2. Antioxidant Activity Test

The antioxidant activity of the samples was assessed using the 2,2-diphenyl-2-picrylhydrazyl (DPPH) method, chosen for its simplicity, ease of implementation, and minimal sample requirement. This assay relies on the ability of potential antioxidant compounds to scavenge free radicals, causing a visible change in the color of the DPPH solution from purple to yellow. As this reaction occurs, there is a concurrent alteration in the maximum absorbance of the DPPH solution at a specific wavelength, signifying the scavenging activity against free radicals. The degree of antioxidant potency is quantitatively expressed as the Inhibitory Concentration (IC50) value, which indicates the concentration of the extract required to inhibit 50% of the DPPH radicals or its ability to reduce free radicals by 50%. A lower IC50 value indicates higher antioxidant activity, suggesting the extract's effectiveness in neutralizing free radicals and, consequently, its potential to mitigate oxidative stress and associated health benefits [27], [28]. Antioxidant activity can be categorized into distinct levels of potency, namely: very strong (IC50 $<$ 50 ppm), strong (IC50 between 50–100 ppm), moderate (IC50 between 100–150 ppm), weak (IC50 between 150–200 ppm), and very weak (IC50 > 200 ppm) [29].

The antioxidant activity of the binahong leaf extract was evaluated using the DPPH method and the results are presented in Table II.

In this study the antioxidant activity assay showed that the IC50 values for the ethanol and methanol extracts were 370.26 ppm and 318.85 ppm, respectively, classifying their antioxidant activity within the weak category. Conversely, vitamin C demonstrated a remarkably low IC50 value of 42.0, indicating a significantly potent antioxidant activity in the very strong category [28]. The observed weak antioxidant activity of the binahong leaf extracts could potentially be attributed to the extraction methodology used. It is plausible that the extraction process did not yield an optimal recovery of secondary metabolites with robust antioxidant properties. Further refinement of the extraction approach and a comprehensive evaluation of the phytochemical composition could potentially enhance the antioxidant potential of the binahong leaf extract [30].

In light of this, further refinements such as fractionation are warranted. This entails isolating specific compounds from the crude extract, with the objective of obtaining more purified fractions. It is plausible that a certain fraction contains a compound with markedly enhanced antioxidant potential compared to the impure extract. Consequently, fractionation holds promise as a strategic approach to attain higher antioxidant efficacy, potentially yielding IC50 values that signify stronger antioxidant activity.

Vitamin C, also known as ascorbic acid, was employed as a positive control in the study. Vitamin C is naturally present in a variety of fruits and vegetables and is acknowledged for its robust antioxidant capabilities. This attribute stems from vitamin C's capacity to counteract oxidative stress through its electron-donating properties, effectively mitigating the deleterious effects of free radicals [27]. The inherent antioxidative potency of vitamin C can be attributed to its ability to act as an electron donor, participating in redox reactions that neutralize free radicals. Consequently, this antioxidant mechanism supports the maintenance of cellular integrity and contributes to overall health by preventing oxidative damage [31].

TABLE II: Binahong Leaf Extract Antioxidant Activity Test Results

Test material	IC50 (ppm)	Antioxidant level
Ethanol extract	370.26	Very weak
Methanol extract	318.85	Very weak
Vitamin C	42.0	Very strong

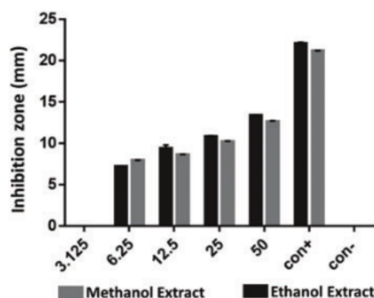

Fig. 1. Antifungal effect of Binahong leave extracts. Inhibition zone sizes of binahong leaf extract against Candida albicans was determined using disc diffusion method, various concentrations (50%, 25%, 12.5%, 6.25%, and 3.12%), positive control (Nystatin) and negative (no extract used).

3.3. Antifungal Candida albicans Test

The antifungal evaluation conducted on Candida albicans yielded affirmative outcomes, evident by the manifestation of distinct clear zones surrounding the paper discs impregnated with the two binahong leaf extracts. These zones of inhibition are indicative of the inhibitory influence exerted by the extracts on the growth and proliferation of Candida albicans. The discernible presence of these clear zones signifies the antifungal potency of the binahong leaf extracts against this fungal strain. The antifungal assay results, delineating the inhibitory effects of the binahong leaf extracts on the growth of Candida albicans, are visually represented in the provided Fig. 1.

The graph in Fig. 1 showed the inhibitory effects of the two binahong leaf extracts, spanning concentrations ranging from 6.25% to 50%, on the growth of Candida albicans. Evidently, an upward trend is observed as concentrations of the extracts increase, resulting in the augmentation of the inhibition zone diameters. Notably, concentrations of 6.25% for both the ethanol and methanol extracts elicited inhibition zone diameters of 7.17 mm and 7.93 mm, respectively. The dataset of inhibition zone diameters obtained from the two binahong leaf extracts underwent a comprehensive analysis, commencing with the Kolmogorov-Smirnov test to assess data normality. Notably, both the ethanol extract and the methanol showed normal distribution with p-values of 0.803 and 0.784, respectively. Homogeneity of Variances was also exhibited through p-value of 0.180. Employing the LSD post hoc test, significant mean differences were unveiled among various concentrations, with the inhibition zones of positive control > 50% > 25% > 12.5% > 6.25% > 3.125% extract treatment. The inhibition zone in the negative control was not significantly different from 3.125% extract treatment. Interestingly, the inhibitory effects of the two extracts were indistinguishable, suggesting that the solvent did not significantly affect the antifungal efficacy against Candida albicans.

Complementing these findings, literature elucidates that binahong leaves's active compounds, notably flavonoids, form intricate bonds with ergosterol proteins within the fungal cell wall, leading to protein denaturation, membrane disruption, and cell death. Tannins inhibit chitin synthesis, a core constituent of fungal cells, while also compromising the cytoplasmic membrane, resulting in plasmolysis. Saponins exert their antifungal activity by hindering fungal ergosterol protein biosynthesis, thereby undermining the integrity of cell membranes.

The amalgamation of empirical findings and established mechanisms provides a multifaceted understanding of the observed antifungal potential within binahong leaf extracts against Candida albicans, bolstering their potential utility as natural antifungal agents and inviting further research avenues in fungal infection management.

4. Conclusion

Phytochemical analysis of the ethanol extract of binahong leaves unveiled the presence of a composite profile encompassing flavonoids, tannins, saponins, and steroids. Meanwhile, the methanol extract showcased the occurrence of saponin and steroid compounds. The antioxidant potential, assessed using the DPPH method, demonstrated that both the ethanol and methanol binahong leaf extracts exhibited a low level of antioxidant activity. Both binahong extracts demonstrated a notable capability to restrain the proliferation of Candida albicans, ascertained across a concentration gradient spanning 6.25% to 50%, with both the ethanol and methanol extract showing a similar potency in hindering the growth of Candida albicans. This insight underscores the minimal influence of the choice of solvent on the antifungal activity of binahong leaf extracts against the fungal strain. In sum, the research findings underscore the multifaceted bioactivity profile of binahong leaf extracts,

encompassing a spectrum of phytochemical compounds, modest antioxidant capabilities, and notable antifungal potential against Candida albicans. The intricate interplay of these constituents and their biological effects culminate in a nuanced understanding of binahong leaf extracts' potential as natural agents with versatile applications in both antioxidant and antifungal contexts.

Conflict of Interest

Authors declare that they do not have any conflict of interest.

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