



Formulation of Antibacterial Solid Soap Containing Miana Leaf Extract Against *Staphylococcus aureus* Bacteria

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ABSTRACT

Miana leaf (*Coleus scutellarioides* (L.) Benth.) is known to contain antibacterial compounds such as flavonoids, tannins, saponins, and alkaloids. This study aimed to formulate solid soap with miana leaf extract at concentrations of 1%, 3%, and 5%, and to evaluate its antibacterial activity against *Staphylococcus aureus*. The soap formulations were evaluated for organoleptic properties, pH, foam height, skin moisture, and irritation potential. Antibacterial testing was conducted using the well diffusion method. Results showed that the solid soap met physical quality standards, with pH ranging from 9.39 to 10.6, foam height between 7.6 and 14.2 cm, and moisture levels increasing to 36.6–48.4%. Irritation tests revealed no adverse skin reactions. The antibacterial activity test confirmed that all formulations inhibited *S. aureus*, with the strongest inhibition observed at 5% extract concentration, producing an inhibition zone of 18.4 mm. In conclusion, solid soap containing miana leaf extract is safe for topical use and shows strong antibacterial activity, especially at higher extract concentrations, indicating its potential as a natural antiseptic formulation.



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ABSTRAK

Daun miana (*Coleus scutellarioides* (L.) Benth.) diketahui mengandung senyawa antibakteri seperti flavonoid, tanin, saponin, dan alkaloid. Penelitian ini bertujuan untuk merumuskan sediaan sabun padat dengan ekstrak daun miana pada konsentrasi 1%, 3%, dan 5%, serta mengevaluasi aktivitas antibakterinya terhadap *Staphylococcus aureus*. Sabun padat yang dihasilkan dievaluasi berdasarkan karakteristik organoleptik, pH, tinggi busa, kelembaban kulit, dan potensi iritasi. Uji antibakteri dilakukan dengan metode difusi sumuran. Hasil menunjukkan bahwa sabun padat memenuhi standar mutu fisik, dengan pH berkisar antara 9,39 hingga 10,6, tinggi busa antara 7,6–14,2 cm, dan kadar kelembaban kulit meningkat dari 36,6% menjadi 48,4%. Uji iritasi tidak menunjukkan reaksi negatif pada kulit. Uji aktivitas antibakteri menunjukkan bahwa semua formula mampu menghambat pertumbuhan *S. aureus*, dengan zona hambat terbesar diperoleh pada formula ekstrak 5% sebesar 18,4 mm. Kesimpulannya, sabun padat dengan ekstrak daun miana aman digunakan secara topikal dan memiliki aktivitas antibakteri yang kuat, khususnya pada konsentrasi ekstrak yang lebih tinggi, sehingga berpotensi dikembangkan sebagai sediaan antiseptik alami.

Kata Kunci: Sabun padat; Ekstrak daun miana; *Staphylococcus aureus*; Uji antibakteri; Humektan alami; Uji iritasi kulit

1. Introduction

Staphylococcus aureus is a pathogenic bacterium commonly found in the upper respiratory tract, mouth, urinary tract, nose, and skin. It is a gram-positive bacterium capable of causing various infections such as meningitis, boils, acne, and other skin-related diseases due to its ability to proliferate and spread within body tissues, producing harmful extracellular substances [1],[2].

One of the plants with antibacterial potential is the miana leaf (*Coleus scutellarioides* (L.) Benth.), which is characterized by its reddish-purple color. This plant contains various secondary metabolites such as flavonoids, polyphenols, steroids/terpenoids, tannins, essential oils, coumarins, saponins, alkaloids, minerals, and small amounts of mucilage [3]. These phytochemicals, especially secondary metabolites, are known to benefit not only the plant itself but also have pharmacological effects on humans [4].

Miana leaves exhibit antibacterial properties due to the presence of compounds such as flavonoids, tannins, and saponins [4]. Antibacterial soaps are highly preferred by consumers, as they are believed to cleanse the skin effectively and help prevent bacterial infections. Therefore, incorporating miana leaf extract into a solid soap formulation offers a practical and accessible form of antibacterial agent for topical use.

The skin, as the largest organ of the human body, serves as a primary barrier against external threats such as viruses and bacteria. Its health is essential to prevent disease, and compromised skin can result from factors such as environmental changes, climate, allergies, or weakened immunity. Under certain conditions, such as sun exposure or high humidity, bacterial or viral infections can easily penetrate and trigger systemic reactions [5].

Soap is a surfactant used with water for washing and cleaning purposes. Solid soap is one of the most widely used personal hygiene products due to its affordability, ease of storage, and longevity compared to liquid soaps. In addition to

general cleansing, solid soaps can be enhanced with antibacterial or moisturizing agents to increase their functionality [6].

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Although the antibacterial potential of miana leaf extract has been documented, there is limited research on its formulation into solid soap as a delivery system. Solid soap offers several advantages, including ease of use, storage stability, and economic value. Therefore, this study aims to formulate solid soap containing miana leaf extract and to evaluate its antibacterial activity against *S. aureus*.

2. Methods

Research Design

This study employed an experimental design to evaluate the antibacterial activity of solid soap formulations containing miana leaf extract (*Coleus scutellarioides* (L.) Benth.) against *S. aureus*.

Materials

The materials used included distilled water, miana leaf extract, 96% ethanol, nutrient agar (NA) medium, palm oil, olive oil, sodium hydroxide (NaOH), vanilla flavoring, and virgin coconut oil (VCO).

Preparation of Miana Leaf Extract

A total of 500 grams of fresh miana leaves were extracted using the maceration method with 96% ethanol as the solvent. The leaves were soaked in ethanol inside a glass container for 3×24 hours with occasional stirring. The macerate was collected daily and the solvent replaced. The filtrate was then aerated using a fan until a thick extract was obtained and weighed to a constant mass.

Phytochemical Screening

Phytochemical screening was conducted to identify the presence of secondary metabolites in the miana leaf extract. The following standard qualitative tests were performed:

Alkaloid Test: A total of 0.5 g of ethanolic extract of red miana leaves (*Coleus scutellarioides*) was dissolved in distilled water, followed by the addition of a few drops of concentrated hydrochloric acid (HCl) and 0.2 g of magnesium powder. The formation of a brick-red color confirmed the presence of flavonoids [9].

Flavonoid Test: A total of 0.5 g of ethanolic extract of red miana leaves (*Coleus scutellarioides*) was dissolved in distilled water, followed by the addition of a few drops of concentrated hydrochloric acid (HCl) and 0.2 g of magnesium powder. The formation of a brick-red color confirmed the presence of flavonoids [9].

Tannin Test: A total of 0.5 g of ethanolic extract of red miana leaves (*Coleus scutellarioides*) was dissolved in distilled water and then treated with a few drops of iron(III) chloride (FeCl₃). The appearance of a bluish-green coloration indicated the presence of tannins [9].

Saponin Test: A total of 0.5 g of ethanolic extract of red miana leaves (*Coleus scutellarioides*) was mixed with distilled water and shaken vigorously for approximately 1 minute. The presence of stable foam indicated a positive result for saponin compounds [9].

Solid Soap Preparation and Formulation

The formulation process followed the cold saponification method, modified from Supriyanta et al. (2021) [10]. First, NaOH was dissolved in distilled water and cooled. Oils (VCO, palm oil, and olive oil) were mixed and homogenized in a beaker. NaOH solution was then added to the oil mixture with continuous stirring until a thick soap paste formed. Miana leaf extract and vanilla fragrance were subsequently added. The mixture was poured into molds and allowed to harden for 24 hours. The complete composition of each formula, including active ingredients and excipients, is presented in Table 1.

Table 1. Solid Soap Formulation of miana leaf extract (*Coleus scutellarioides* L.) Benth.)

Material	F1	F2	F3	K(-)	K(+) (Thistle soap)	Function
Miana leaf extract	1%	3%	5%	-	-	Active ingredient
Virgin coconut oil (VCO)	20%	20%	20%	20%	-	Foaming agent
Palm oil	30%	30%	30%	30%	-	Foaming agent
Olive oil	10%	10%	10%	10%	-	Moisturizer
NaOH	8.9%	8.9%	8.9%	8.9%	-	Alkali (saponification)
Vanilla flavouring	q.s	q.s	q.s	q.s	-	Fragrance
Distilled water	ad 100 mL	ad 100 mL	ad 100 mL	ad 100 mL	-	Solvent

Note:

K(-): Negative control (soap base without extract);

K(+): Positive control (thistle soap);

q.s: quantum satis (as much as sufficient);

ad: up to volume

Evaluation of Soap Preparations

The solid soap formulations were evaluated through a series of tests to determine their physical characteristics, safety, and functional performance. The evaluations included organoleptic testing, pH, skin moisture, and irritation. The procedures conducted were as follows:

Organoleptic Test: Conducted using sensory evaluation of shape, color, and aroma before and after cycling tests to assess stability [11]

pH Test: Soap was dissolved in water, and pH was measured using a digital pH meter. Acceptable pH for solid soap is between 9–11 [12].

Moisture Test: The moisturizing effect of the soap formulations was assessed using a *Skin Moisture Analyzer* on 10 healthy volunteers. Skin moisture was measured on the back of the hand before and 15 minutes after applying the soap. Prior to testing, the

skin was washed and dried. Moisture levels were classified as follows: <40% = less humid, 40–60% = moist, >60% = very moist [13].

Irritation Test: To evaluate the safety of the soap on the skin, each formulation was applied to the back of each volunteer's ear and left for approximately 1 hour. The skin was monitored for any signs of redness, itching, or swelling [14].

Antibacterial Activity Test

Antibacterial activity was assessed using the well diffusion method [10], [15]. NA media was poured into Petri dishes and allowed to solidify. Wells were created, and bacterial suspension of *S. aureus* was introduced. Soap samples, including controls, were added into respective wells. Plates were incubated at 37°C for 24 hours. Inhibition zones were measured in millimeters using a caliper. Each test was conducted in triplicate to ensure reproducibility.

Statistical Analysis

All quantitative data obtained from the evaluation tests – namely pH values, skin moisture levels, and antibacterial inhibition zones – were subjected to statistical analysis to determine the significance of differences among soap formulations. The data were analyzed using one-way Analysis of Variance (ANOVA) followed by post hoc Tukey's test when applicable, with a confidence level of 95% ($\alpha = 0.05$). The analysis was conducted using SPSS version 25.0 (IBM Corp., Armonk, NY, USA). Results were expressed as mean \pm standard deviation (SD), and differences were considered statistically significant when $P < 0.05$.

Ethical Approval

This study was ethically approved by the Health Research Ethics Committee of Politeknik Kesehatan Kemenkes Makassar under letter No. 165/ML/KEPK-PTKMS/VII/2024. All procedures were conducted in accordance with the seven WHO 2011 ethical standards and the CIOMS 2016 guidelines, including considerations for scientific value, risks and benefits, confidentiality, and informed consent.

3. Results and Discussion

Phytochemical Screening

In this study, a phytochemical screening test was conducted on a sample of miana leaf extract (*Coleus scutellarioides* (L.) Benth.) to identify the presence of secondary metabolites, including alkaloids, flavonoids, saponins, and tannins. Phytochemical screening serves as a preliminary step in identifying the classes of bioactive compounds present in medicinal plants [9]. The results confirmed the presence of alkaloids, flavonoids, saponins, and tannins, as summarized in Table 2.

Table 2. Phytochemical Screening Results of Miana Leaf Extract

Compound Test	Reagents	Observed Result	Presence (+/-)
Alkaloids	Wagner's reagent	Brown precipitate	+
Flavonoids	Mg + HCl	Yellow coloration	+
Saponins	Distilled water	Stable foam formation	+
Tannins	FeCl ₃	Blackish green coloration	+

Note: + compound present; - compound not detected;

Organoleptic Test

Organoleptic testing was conducted to evaluate the physical stability of the miana leaf solid soap formulations before and after the cycling test. The parameters observed included shape, color, and smell, as summarized in Table 3. These sensory

evaluations are essential to assess the acceptability and physical changes in topical formulations during storage and usage [10].

The results indicated that all formulations maintained their physical integrity throughout the cycling test. There were no observable changes in shape, color, or smell in any of the samples. Formula K(-) remained milky white with a vanilla scent, while F1 to F3 showed a color transition from yellow to brownish yellow and chocolate, in line with the increasing concentration of miana leaf extract. The aroma was consistently described as "typical miana" across formulations F1-F3.

Table 3. Organoleptic Test Results of Miana Leaf Solid Soap

Formula	Shape (Before / After Cycling Test)	Color (Before / After Cycling Test)	Smell (Before / After Cycling Test)
K(-)	Solid / Solid	Milky white / Milky white	Vanilla / Vanilla
F1	Solid / Solid	Yellow / Brownish yellow	Typical miana / Typical miana
F2	Solid / Solid	Brownish yellow / Brownish yellow	Typical miana / Typical miana
F3	Solid / Solid	Chocolate / Chocolate	Typical miana / Typical miana

These findings suggest that the soap formulations have good physical stability and that the increasing concentration of miana extract influences the color intensity of the product. This is supported by Wiyono et al. (2023) [16], who reported that higher concentrations of plant-based actives tend to darken the color of soap formulations due to the accumulation of pigments and polyphenolic compounds. The absence of change in shape and smell also indicates resistance to physical degradation under stress conditions

pH test

pH is a critical parameter in solid soap formulations, as it affects the safety and compatibility of the product with human skin. A pH that is too low or too high can potentially increase skin permeability and cause irritation. According to the Indonesian National Standard (SNI), the acceptable pH range for solid soap is between 9 and 11 [9].

Table 4. pH Values (Mean \pm SD) Before and After Cycling Test of Miana Leaf Solid Soap

Formula	pH Before Cycling	pH After Cycling	Standard Range	Significance
K(-)	9.39 \pm 0.02	10.00 \pm 0.05		
F1	9.55 \pm 0.01	10.30 \pm 0.03	9-11 [8]	0.086 ($P > 0.05$)
F2	9.66 \pm 0.02	10.40 \pm 0.02		
F3	9.75 \pm 0.01	10.60 \pm 0.04		

Statistical analysis was conducted using one-way ANOVA. No significant differences were observed ($P > 0.05$)

In this study, the pH of miana leaf solid soap was measured before and after the cycling test to assess chemical stability. As shown in **Table 4**, all formulations experienced a slight increase in pH after cycling. However, the values remained within the standard acceptable range of 9-11, as defined by the Indonesian National Standard (SNI) [8].

Statistical analysis using one-way ANOVA revealed no significant difference between the formulations ($P = 0.086$), indicating that the increase in pH was not statistically significant. The observed increase is likely due to the presence of basic compounds such as alkaloids in the miana extract.

This observation is consistent with the findings of Zalfiatri et al. (2018) [12], who reported that formulations containing plant extracts rich in alkaloids tend to exhibit higher pH levels, as alkaloids are naturally basic in nature.

Moisture Test

The moisture test was conducted to evaluate the moisturizing effect of miana leaf extract (*Coleus scutellarioides* (L.) Benth.) in solid soap formulations. The test involved 10 healthy volunteers, where the skin moisture level was measured on the back of each participant's hand using a *Skin Moisture Analyzer* before and 15 minutes after applying the soap. Four groups were tested: negative control (K-, soap without extract), and three formulas containing miana leaf extract at 1% (F1), 3% (F2), and 5% (F3). Moisture levels were classified as follows: <40% = low moisture, 40–60% = moist, >60% = very moist [13].

Table 5. Skin Moisture Levels Before and After Using Miana Leaf Solid Soap Formulations

Respondent	Before Usage	K(-)	F1 (1%)	F2 (3%)	F3 (5%)
1	33.8	36.6	46.4	47.1	48.7
2	36.7	41.1	42.9	45.9	47.2
3	38.5	40.4	41.9	46.7	48.4
4	37.4	42.8	45.1	46.7	48.1
5	36.8	40.4	41.1	46.6	48.7
6	37.3	41.7	46.5	47.0	48.7
7	37.2	44.3	45.9	47.1	48.2
8	36.5	40.3	45.8	46.6	47.1
9	35.3	36.8	45.7	47.1	50.1
10	36.8	44.3	47.4	48.6	49.4
Average ± SD	36.6 ± 1.41	40.8 ± 2.52 ^a	44.8 ± 1.75 ^b	46.9 ± 0.67 ^c	48.4 ± 0.86 ^c

Note:

Skin moisture classification: <40% = low, 40–60% = moderate (moist), >60% = very moist [13].

Superscript letters (a, b, c) indicate statistically significant differences between groups ($P < 0.05$), based on one-way ANOVA followed by Tukey's post hoc test.

As shown in **Table 5**, the baseline average skin moisture before soap application was 36.6%, which falls under the "low moisture" classification. After applying the soap, the moisture levels increased in all groups. The negative control (K-) resulted in an average of 40.8%, classified as "moist." In contrast, F1, F2, and F3 formulations reached 44.8%, 46.9%, and 48.4%, respectively – surpassing the 40% threshold for "very moist" skin.

Statistical analysis using one-way ANOVA revealed a significant difference among the groups ($P < 0.001$). Tukey's post hoc test revealed that F1, F2, and F3 significantly differed from K(-), while F2 and F3 also showed significantly higher values than F1, though no significant difference was observed between F2 and F3 ($P > 0.05$), indicating a plateauing effect at higher extract concentrations.

These findings demonstrate that miana leaf extract enhances skin hydration in a concentration-dependent manner. The observed moisturizing effect is likely due to phytochemicals such as flavonoids and saponins, which possess humectant and skin-conditioning properties [13]. This aligns with the study by Wiyono et al. (2023) [16], which reported that increasing concentrations of plant-based active ingredients not only improve moisturization but also influence the product's appearance. The darker color

observed in F3 may be associated with a higher phytochemical content and enhanced bioactivity.

Irritation Test

The irritation test was conducted to assess the safety of the solid soap formulations containing miana leaf extract (*Coleus scutellarioides* (L.) Benth.) when applied to human skin. The purpose of this test was to observe any potential adverse reactions, such as redness, itching, or swelling. Ten volunteers participated in this test, where each formulation was applied to the back of the ear and left for approximately one hour. The skin was then observed for any signs of irritation.

As shown in **Table 6**, none of the volunteers experienced any skin irritation from any of the tested formulations, including the negative and positive controls. This indicates that the solid soap formulations are safe for topical use and do not cause skin irritation under the test conditions.

Table 6. Irritation Test Results

Respondents	Skin Reaction - K(-)	F1	F2	F3
1	-	-	-	-
2	-	-	-	-
3	-	-	-	-
4	-	-	-	-
5	-	-	-	-
6	-	-	-	-
7	-	-	-	-
8	-	-	-	-
9	-	-	-	-
10	-	-	-	-

Note: - indicates no visible signs of irritation (no redness, itching, or swelling)

These results are in agreement with findings by Tungadi et al. (2022) [14], who reported that solid soap formulations did not cause adverse reactions such as redness, itching, or swelling. This safety is attributed to the pH of the soap formulations remaining within the acceptable range of 9–11, which is considered non-irritating for normal skin conditions.

Antibacterial Activity Test

The antibacterial activity of solid soap formulations containing miana leaf extract (*Coleus scutellarioides* (L.) Benth.) was evaluated against *S. aureus* using the well diffusion method. The test was performed by placing a well tool into solidified Nutrient Agar (NA) that had been inoculated with a bacterial suspension. After creating the well, 1 mL of each soap formulation was added, and the plates were incubated at 37 °C for 24 hours. The antibacterial activity was assessed by measuring the diameter of the inhibition zone formed around each well.

As shown in **Table 7**, all soap formulations containing miana leaf extract demonstrated antibacterial activity against *S. aureus*, with inhibition zones ranging from 16.4 mm to 18.4 mm. Based on classification standards, all formulas fall within the “strong” category (10–20 mm).

Table 7. Antibacterial Activity of Miana Leaf Soap Formulations Against *S. aureus*

Formula	Inhibition Diameter (mm)	Average \pm SD	Category
K(+)	19.1, 19.2, 18.7	19.0 \pm 0.26	Strong
K(-)	0, 0, 0	0.0 \pm 0.00	None
F1 (1%)	16.3, 16.2, 16.6	16.4 \pm 0.21 ^b	Strong
F2 (3%)	17.2, 17.3, 17.6	17.3 \pm 0.21 ^b	Strong
F3 (5%)	18.4, 18.3, 18.6	18.4 \pm 0.15 ^b	Strong

Zone interpretation:

<5 mm = weak, 5–10 mm = moderate, 10–20 mm = strong, >20 mm = very strong [15]

Different superscript letters indicate no statistically significant differences ($P > 0.05$, one-way ANOVA).

The highest inhibition was observed in F3 (5% extract), followed by F2 (3%) and F1 (1%), indicating a dose-dependent response. The positive control (K+) produced an inhibition zone of 19.0 mm, slightly higher than F3, while the negative control (K-) showed no inhibition (0 mm), confirming that the antibacterial activity is directly attributable to the miana extract.

One-way ANOVA revealed no statistically significant differences in antibacterial activity among F1, F2, and F3 ($P > 0.05$). However, all three differed significantly from the negative control. This suggests that while increased extract concentration correlates with larger inhibition zones, the differences plateau beyond a certain concentration.

The antibacterial activity of miana extract may be attributed to its phytochemical constituents, particularly flavonoids, alkaloids, and tannins. These compounds are known to disrupt bacterial growth through mechanisms such as damage to cell membranes, enzyme inhibition, and interference with nucleic acid synthesis [16].

These findings are consistent with Nugraha et al. (2022) [7], who reported strong antibacterial activity of miana leaf extract against gram-positive bacteria, including *Staphylococcus epidermidis*. The ability of the soap formulation to inhibit *S. aureus* growth highlights its potential as a natural antiseptic product.

Although the results are promising, this study has several limitations. First, the antibacterial activity was assessed only through in vitro methods and not validated by in vivo or clinical trials. Second, the study focused solely on *S. aureus* and did not evaluate the spectrum of activity against other gram-positive or gram-negative pathogens. Lastly, the stability and efficacy of the soap formulations over long-term storage were not investigated. Future research should address these aspects to further validate the product's potential as a topical antimicrobial agent.

4. Conclusion

Based on the results of this study, it can be concluded that miana leaf extract (*Coleus scutellarioides* (L.) Benth.) exhibits antibacterial activity against *Staphylococcus aureus*. The optimum concentration was 5%, producing an inhibition zone of 18.4 mm, which indicates strong antibacterial activity. In addition to its antimicrobial effectiveness, the solid soap formulations demonstrated acceptable pH, good physical and organoleptic stability, moisturizing effects, and no observed skin irritation, suggesting that the product is safe and suitable for topical use. To support product development and enhance its applicability, it is recommended that future studies explore the in vivo antibacterial efficacy, assess long-term product stability, and evaluate the formulation against a broader spectrum of microorganisms.

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Conflicts of Interest:

The authors declare no conflict of interest regarding the publication of this article.

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