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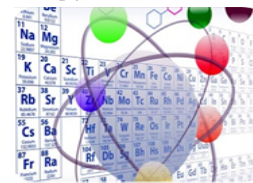
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Antibacterial Potential of Gompang Batu Plant Extract (*Lobelia nummularia* Lam) Against Skin Disorder-Causing Bacteria

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ABSTRACT

Skin disorders are common health problems that are often caused by bacterial infections such as *Staphylococcus epidermis* and *Propionibacterium acnes*. This study aims to analyze the antibacterial potential of n-hexane, ethyl acetate, and ethanol extracts of gompang batu (*Lobelia nummularia* Lam) plants against bacteria that cause skin disorders. Extraction was carried out by maceration with solvents of varying polarity. Antibacterial tests were carried out using the disc diffusion and microdilution methods with six treatments (chloramphenicol, DMSO, 1.25%; 2.5%; 5% and 10% extract). The results showed that the ethanol extract with a concentration of 10% had the most effective antibacterial activity with an inhibition zone of 14.0 mm (*P. acnes* bacteria) and 13.8 mm (*S. epidermis* bacteria). The MIC value of the extract ranged from 156.25 to 625 µg/mL and the MBC value of all extracts > 2500 µg/mL indicated bactericidal properties. Gompang batu has the potential as a natural antibacterial for skin infections. N-hexane extract contains steroids and tannins; ethyl acetate contains alkaloids, flavonoids, steroids and tannins; ethanol contains alkaloids, flavonoids, terpenoids and tannins.

Keywords: Skin disorders, *Lobelia nummularia* Lam, antibacterial, *Staphylococcus epidermis*, *Propionibacterium acnes*.

1. INTRODUCTION

Skin disorders are one of the health problems that are widely experienced by the community and can be caused by bacterial infections such as *Staphylococcus epidermis* and *Propionibacterium acnes*.¹⁰ In the Asian region, especially Indonesia, skin diseases such as acne dermatitis, psoriasis, and pruritus show a high prevalence rate. Indonesia is even ranked 29th out of 195 countries related to the transmission of skin diseases caused by several factors of poor hygiene

and limited access to health services.²² Treatment of skin infections generally involves the use of broad-spectrum antibiotics such as gentamicin, ampicillin, and chloramphenicol. However, inappropriate use of antibiotics often causes serious side effects and triggers antimicrobial resistance.²³ This condition encourages the need for safer alternative treatments, such as the use of natural ingredients that are antibacterial.

Medicinal plants have long been used in traditional medicine due to their pharmacologically active secondary metabolite content. One of the local plants that has this potential is *Lobelia nummularia* Lam or known as gompang batu. This plant is traditionally used by people in North Tapanuli to treat wounds and ulcers. The results of the determination of "Herbarium Medanense" No.1943/MEDA/2024, the gompang batu plant is a type of *Lobelia nummularia* Lam including the Campanulaceae family. The *Lobelia* genus is known to contain various bioactive compounds such as flavonoids, terpenoids, alkaloids and tannins which have antibacterial activity.²⁶

Previous studies have shown that several *Lobelia* species, including *L. chinensis*, show high effectiveness in inhibiting the growth of bacteria such as *Bacillus cereus*, with low MIC and MBC values.^{21,13} Based on this potential, this study aims to examine the antibacterial activity of *Lobelia nummularia* Lam extract against *S. epidermis* and *P. acnes* that cause skin disorders using the maceration extraction method with solvents of varying polarity (n-hexane, ethyl acetate, and ethanol) and the disc diffusion and microdilution methods. The active compounds obtained are expected to be able to inhibit or kill bacteria that cause skin disorders effectively, as well as being candidates for future natural antibacterial raw materials.

2. EXPERIMENTAL

2.1. Chemicals, Equipment and Instrumentation

The materials used in this study were fresh gompang batu leaves from Ruhut Bosi village, Pangaribuan District, North Tapanuli Regency, North Sumatra, n-hexane, ethyl acetate, 96% ethanol, distilled water, blank disc (Oxoid, UK), dimethyl sulfoxide (DMSO), 0.9% NaCl, chloramphenicol disc (Oxoid, UK), Mueller Hinton Agar (MHA), Mueller-Hinton Broth (MHB), magnesium powder, anhydrous acetic acid, concentrated sulfuric acid, dragendroff reagent, magnesium powder, *Staphylococcus epidermis* ATCC 6919, *Propionibacterium acnes* ATCC 6919, lieberman burchard reagent, concentrated hydrochloric acid. The equipment used in this study were grinder, analytical balance, rotary evaporator (Heidolph), incubator (Mettler), autoclave (TOMY ES-315), petri dish (Onemed), microplate (Nest), sprider, laminar (B-one V 915S), spatula, micropipette, vernier caliper, magnetic stirrer, buchner funnel, hotplate, erlenmeyer flask (Pyrex), test tube (Pyrex), vortex (SBS), vial, measuring cup (Pyrex), tweezers, 80 mesh sieve, glass stirring rod, ose, aluminum foil.

2.2. Research Procedure

Preparation and Extraction of Gompang Batu Plant (*Lobelia nummularia* Lam)

A total of 4 kg of fresh gompang batu (*Lobelia nummularia* Lam) plants were washed clean, drained and dried by airing without direct sunlight and ground into powder. The powdered simplicia was macerated using solvents with varying polarity (n-hexane, ethyl acetate and ethanol), each for 3 × 24 hours. The maceration results were filtered and the filtrate was concentrated using a rotary evaporator, obtaining a thick extract of gompang batu plants from each extract.¹⁴

Phytochemical Test

a) Testing of Flavonoid Compounds

Take 0.5 g of gompang batu leaf extract dissolved with solvent (n-hexane, ethyl acetate and ethanol), then pipette 1 mL and add enough Mg powder then drip with 10 drops of concentrated hydrochloric acid solution. Positive contains flavonoids if there is a color change then it is positive contains flavonoids.⁷

b) Testing of Alkaloid Compounds

Alkaloid compound test can be done by weighing 0.5 g of extract then dissolved with solvent (n-hexane, ethyl acetate and ethanol). Added 3 drops of Dragendroff reagent, the formation of brick red precipitate indicates the presence of alkaloids.¹⁸

c) Testing of Steroid/Terpenoid Compounds

Weighed as much as 0.5 g of extract dissolved with 10 mL of ether. As much as 0.5 mL of the solution is tested with Lieberman Burchard reagent. If a blue or green color is formed, it indicates the presence of steroids, a red or purple color indicates the presence of triterpenoid compounds.¹⁸

d) Testing of Tannin Group Compounds

Each extract of gompang batu plant was taken as much as 0.5 g and dissolved with solvents (n-hexane, ethyl acetate and ethanol). Then put in a test tube and added a few drops of 1% FeCl₃. The presence of tannin compounds is indicated by a blackish green color.²⁴

Antibacterial Activity Test

Sterilization and Media Preparation

The tools and supplies to be used in this study were sterilized, except for each extract and suspension of *Staphylococcus epidermis* and *Propionibacterium acnes* bacteria. The tools that had been washed and dried and the supplies were wrapped in paper then put into an autoclave and sterilized to a temperature of 121 ° C, left for 20 minutes at a consistent temperature, then removed and dried.¹⁶ MHA was weighed as much as 22.8 grams and dissolved in 600 ml of distilled water, while MHB as much as 1.05 grams was dissolved in 50 ml of distilled water. Then each was heated and stirred using a magnetic stirrer.

Preparation of Test Solution

The test solution was prepared by dissolving the extract in DMSO solvent of each gompang batu leaf extract dissolved until a test solution with a concentration of 1.25%; 2.5%; 5% and 10% was obtained. For 1.25% test solution, the extract was weighed as much as 0.0125 grams which was added with 1 mL of DMSO solvent then put into a microtube and vortexed until homogeneous. For 2.5%; 5% and 10% test solutions, each extract was weighed 0.025; 0.05 and 0.1 grams then dissolved in 1 mL of DMSO and put into a microtube and vortexed until homogeneous.

Preparation of Inoculum Suspension

Staphylococcus epidermis bacterial colonies were taken using a round loop, then slowly inserted into a test tube containing 0.9% NaCl and compared with the 0.5 Mc Farland standard. The same thing was done on *Propionibacterium acnes* bacteria.

Inhibitory Power Test Using Paper Disc Diffusion Method

100 µL of bacterial suspension was added onto the solidified media and scratched in one direction using a spreader, then left for approximately 5 minutes until the media suspension was slightly dry. Chloramphenicol disc was

placed on the positive control section, while the blank disc was placed on the negative control section and the concentration treatment of each extract. In the negative control section, 20 μL of DMSO was dropped. For the extract concentration treatment, each blank disc was dropped with each extract concentration of 1.25%, 2.5%, 5% and 10% as much as 20 μL . After that, the plate was closed and put into the incubator for 24 hours at a temperature of 37°C. After 24 hours, observations were made on each media plate and the diameter of the clear zone formed was measured, indicating the presence of an inhibition zone in each treatment.¹⁹ In this antibacterial test, the positive control used was a 30 μg chloramphenicol antibiotic disc, while the negative control was DMSO solvent.

Determination of Minimum Inhibitory Concentration (MIC) by Microdilution Method

The MIC test was carried out by taking 100 μL of bacterial suspension and then adding 4900 μL of MHB. Then 100 μL of MHB liquid media was inserted into the first hole as a negative control, while the second to twelfth holes were filled with 100 μL of MHB liquid media that had been suspended with bacteria. In the second hole, 100 μL of chloramphenicol solution was added as a positive control, while holes three to twelve were filled with test solutions using a concentration series starting from hole twelve which contained a test solution of 1000 $\mu\text{g/mL}$. The concentration series was carried out by taking 100 μL of test solution from hole twelve and inserting it into hole eleven. The same thing was done up to hole three with the amount of solution in each column being 100 μL . Furthermore, the microplate was incubated for 24 hours at a temperature of 37°C.¹⁸

Determination of Minimum Bactericidal Concentration (MBC) by Microdilution Method

MBC is the lowest concentration of the test solution that can kill bacteria. The MBC test is carried out by taking 10 μL of solution from each well in the microplate, then growing it on MHA agar media and incubating it for 24 hours at 37°C.¹⁸

Data analysis

The data obtained from the observation results are presented in table form. To determine the effect of extract administration on the growth of *Staphylococcus epidermis* and *Propionibacterium acnes* bacteria, an analysis was carried out using One Way ANOVA statistics.

3. RESULTS AND DISCUSSION

Preparation and Extraction of Gompang Batu Plant (*Lobelia nummularia* Lam)

The extraction carried out in this study was by the maceration method. Maceration is a simple and affordable extraction technique because it only requires a simple container as a place for extraction. However, this method takes quite a long time to complete the extraction process. This technique is one of the most common methods used to extract active compounds. The effectiveness of maceration is influenced by several factors, such as the amount of raw materials, the type of solvent used and the extraction time. In this process, maceration is carried out by soaking the sample in the solvent used for extraction.²⁵ The maceration process is carried out using solvents with varying polarity. The use of solvents with varying polarity in the maceration method aims to dissolve polar and non-polar compounds, so that it is very effective in extracting secondary metabolites from plants. Maceration was carried out by soaking 325 grams of gompang batu powder in a nonpolar solvent, namely n-hexane as much as 1300 mL for 3 $\times$ 24 hours and stirring and changing the solvent every 24 hours, in each solvent. The maceration results were filtered to separate the dregs and filtrate. The filtrate obtained was then collected and evaporated using a rotary evaporator to remove the solvent still contained in it. The n-hexane, ethyl acetate and ethanol extracts of gompang batu plants had yield values of 3.25%, 2.95% and 4.74% respectively. The ethanol extract of gompang batu plants had a higher yield value compared to the n-hexane and ethyl acetate extracts. The high yield of the extract produced was directly related to the solubility level

of the active compound in the solvent used. These results indicate that ethanol has a higher extraction capacity compared to n-hexane and ethyl acetate.²⁸

Phytochemical Test

The results of phytochemical tests of gompang batu plant extract (*Lobelia nummularia* Lam) show that gompang batu plants contain secondary metabolite compounds. The n-Hexane extract of gompang batu plants contains many steroid compounds; there are tannin compounds and no alkaloid, flavonoid and terpenoid compounds. The ethyl acetate extract contains many steroid compounds; there are alkaloids, flavonoids and tannins; there are no terpenoids. The ethanol extract contains many alkaloid, flavonoid, terpenoid and tannin compounds; there are no steroids. This shows that the research results obtained are in accordance with the research conducted by Vadeo (2023) which stated that the main compounds isolated in the *Lobelia* genus are flavonoids, terpenoids, alkaloids and so on.²⁶

The results of the alkaloid test using the dragendroff reagent showed that the extract contained alkaloid compounds, which were indicated by the formation of a brick red precipitate.²⁰ Positive results in the alkaloid test are thought to occur due to the formation of a complex between alkaloids and potassium with tetraiodobismuthate ions. Bi^{3+} ions will react with excess potassium iodide, forming potassium tetraiodobismuthate. The nitrogen group in the alkaloid will interact with the K^{+} ion from potassium tetraiodobismuthate through a coordinate covalent bond, which then forms a brick-red precipitate.¹

The results of the flavonoid test using Mg powder and concentrated HCl showed that the extract contained flavonoid compounds, which were indicated by the formation of red, orange or yellow colors.¹ The addition of Mg powder and concentrated HCl aimed to reduce the benzopyrone core contained in the flavonoid structure.⁹

The results of the steroid test using the Lieberman Burchard reagent showed that the n-hexane and ethyl acetate extracts of gompang batu leaves and fruits (*L. nummulari* Lam) were positive for containing steroid compounds and the ethanol extract was negative, meaning it did not contain steroid compounds. Positive results are indicated by the formation of a blue or green color. This is because the reaction between sterols and anhydrous acetic acid involving sulfuric acid produces a series of color changes. Sterols will undergo sulfonation and form conjugated dienes and trienes, which then cause a distinctive color change.¹²

The results of the terpenoid test using the Lieberman Burchard reagent showed that the n-hexane and ethyl acetate extracts of gompang batu leaves and fruits (*L. nummularia* Lam) were negative, meaning they did not contain steroid compounds, and the ethanol extract positively contained steroid compounds, which was indicated by the formation of a red or purple color. The color formation process occurs after anhydrous acetic acid is absorbed, followed by oxidation of the acid by sulfuric acid. Furthermore, the hydrogen group and its electrons are released, causing the compound to undergo conjugation extension, resulting in a red or purple color.⁶

The results of the tannin test using FeCl_3 showed that the gompang batu leaf extract (*Lobelia nummularia* Lam) positively contained tannin compounds, while in the fruit extract only the ethanol extract was positive for containing tannin compounds, which was indicated by the formation of a blackish green color.¹¹

Inhibitory Power Test Using Paper Disc Diffusion Method

The results of the inhibitory activity test of gompang batu plant extract (*Lobelia nummularia* Lam) against *Staphylococcus epidermis* bacteria can be seen in Table 2.

Table 2. Data on Inhibition Zone of Gompang Batu Plant Extract on the Growth of *Staphylococcus epidermis* Bacteria

Extract Type	Concentration	Repeat (mm)			Average (mm)
		1	2	3	
n-Hexane	Chloramphenicol	25,8	25,2	25,0	25,3
	DMSO	0	0	0	0
	1,25%	7,8	8,2	7,8	7,9
	2,5%	8,1	9,6	8,3	8,7
	5%	8,9	9,7	8,8	9,1
	10%	9,8	10,6	8,8	9,7
Ethyl Acetate	Chloramphenicol	25,1	23,4	25,3	24,6
	DMSO	0	0	0	0
	1,25%	8,2	8,5	7,9	8,2
	2,5%	9,0	8,7	8,4	8,7
	5%	9,2	8,8	8,9	9,0
	10%	12,4	9,6	11,6	11,2
Ethanol	Chloramphenicol	24,7	26,3	24,3	25,1
	DMSO	0	0	0	0
	1,25%	11,5	11,6	11,2	11,4
	2,5%	12,2	11,8	11,4	11,8
	5%	12,4	12,4	12,9	12,6
	10%	12,8	13,0	15,5	13,8

Based on Table 2, the extract of gompang batu plant (*Lobelia nummularia* Lam) showed antibacterial potential against *S. epidermis* bacteria. This potential is shown by the formation of a clear zone around the disc that has been dripped with n-Hexane, ethyl acetate and ethanol extracts at concentrations of 1.25%, 2.5%, 5%, and 10%.

n-Hexane extract with concentrations of 1.25%, 2.5%, 5% and 10% had an average inhibition diameter of 7.9 mm, 8.7 mm, 9.1 mm and 9.7 mm (moderate). Ethyl acetate extract with concentrations of 1.25%, 2.5%, 5% and 10% had an average inhibition diameter of 8.2 mm, 8.7 mm, 9.0 mm (moderate) and 11.2 mm (strong). Ethanol extract with concentrations of 1.25%, 2.5%, 5% and 10% had an average inhibition diameter of 11.4 mm, 11.8 mm, 12.6 mm and 13.8 mm (strong).

The results of the inhibitory activity test of gompang batu plant extract (*Lobelia nummularia* Lam) against *Propionibacterium acnes* bacteria can be seen in Table 3.

Table 3. Data on Inhibition Zone of Gompang Batu Plant Extract on the Growth of *Propionibacterium acnes* Bacteria

Extract Type	Concentration	Repeat (mm)			Average (mm)
		1	2	3	
n-Hexane	Chloramphenicol	25,4	24,2	24,9	24,8
	DMSO	0	0	0	0
	1,25%	9,3	9,7	9,0	9,3
	2,5%	9,7	10,6	10,3	10,2
	5%	10,2	9,9	11,6	10,6
	10%	10,7	14,4	12,6	12,6
Ethyl Acetate	Chloramphenicol	29,1	28,7	28,1	28,6
	DMSO	0	0	0	0
	1,25%	9,9	9,9	10,1	10,0
	2,5%	10,1	10,1	10,4	10,2
	5%	10,8	10,4	11,0	10,7
	10%	11,7	10,6	11,1	11,1
Ethanol	Chloramphenicol	28,6	28,6	29,0	28,7
	DMSO	0	0	0	0
	1,25%	10,8	11,0	12,8	11,5
	2,5%	11,3	12,3	12,9	12,2
	5%	12,4	13,3	13,9	13,9
	10%	13,7	14,1	14,2	14,0

Based on Table 3, the extract of gompang batu plant (*Lobelia nummularia* Lam) shows antibacterial potential against *P. acnes* bacteria. This potential is shown by the formation of a clear zone around the disc that has been dripped with n-Hexane, ethyl acetate and ethanol extracts at concentrations of 1.25%, 2.5%, 5%, and 10%. The use of these four concentrations aims to see the effectiveness of each extract from the lowest to the highest concentration.

The diameter of the inhibition of bacterial growth formed, measured and categorized based on its size. The criteria for antibacterial power based on the diameter of bacterial growth inhibition are <5 mm in the weak category, 5-10 mm in the medium category, 10-20 mm in the strong category and >20 mm in the very strong category.¹⁷

n-Hexane extract with concentrations of 1.25%, 2.5%, 5% and 10% had an average inhibition diameter of 9.3 mm (moderate), 10.2 mm (strong), 10.6 mm (strong) and 12.6 mm (strong). Ethyl acetate extract with concentrations of 1.25%, 2.5%, 5% and 10% had an average inhibition diameter of 10.0 mm (moderate), 10.2 mm (strong), 10.7 mm (strong) and 11.1 mm (strong). Ethanol extract with concentrations of 1.25%, 2.5%, 5% and 10% had an average inhibition diameter of 11.5 mm, 12.2 mm, 13.9 mm and 14.0 mm (strong).

The highest inhibition of bacterial growth was shown by the ethanol extract of the gompang batu plant compared to the n-hexane and ethyl acetate extracts against *P. acnes* and *S. epidermis* bacteria. A concentration of 10% ethanol extract of the gompang batu plant was declared the most effective in inhibiting the growth of *P. acnes* and *S. epidermis* bacteria, because a concentration of 10% ethanol extract gave antibacterial activity results with a larger diameter of bacterial growth inhibition. Inhibition zone test data from the three extracts on *P. acnes* and *S. epidermis* bacteria showed an increase and decrease along with increasing concentration. Many factors, including incubation temperature, extract concentration and bacterial suspension preparation procedures can affect this phenomenon.²⁴

The more inoculum used, the inhibition tends to decrease so that the inhibition zone formed becomes smaller. In addition, the concentration of the extract can affect the rate of diffusion of the active compound. The higher the concentration of the extract, the faster the diffusion process that occurs. Thus increasing the antibacterial effectiveness and expanding the diameter of the inhibition zone. This is in line with the results of the study which showed that extracts with a concentration of 100% produced a wider inhibition zone than extracts with concentrations of 25% and 50%.²⁷ Therefore, it can be concluded that increasing the concentration of the extract is directly proportional to the increase in antibacterial effectiveness indicated by the area of the inhibition zone.

Determination of Minimum Inhibitory Concentration (MIC) by Microdilution Method

The results of the MIC test of gompang batu plant extract (*Lobelia nummularia* Lam) against *Staphylococcus epidermis* bacteria are presented in Table 3.

Table 3. MIC Results of Gompang Batu Plant Extract against *Staphylococcus epidermis* Bacteria

No	Sample	MIC (µg/mL)
1	Chloramphenicol (5000 ppm)	4,88
2	n-Hexane (5000 ppm)	312,5
3	Ethyl Acetate (5000 ppm)	312,5
4	Ethanol (5000 ppm)	625

The MIC test was conducted using two test controls as a comparison, namely positive control and negative control. The positive control was in the form of Mueller Hinton Broth (MHB) media and bacteria, while the negative control was in the form of Mueller Hinton Broth (MHB) media. The positive control was used to ensure that the test bacteria remained alive after treatment which also indicated that the work procedure had been carried out correctly.

Negative controls are used to ensure that there is no contamination that can affect the test results.⁴ According to Dzoyem in (Juwitaningsih et al., 2021), the activity of an extract is categorized as strong when its MIC value is <100 µg/mL, considered moderate if 100<MIC<625 µg/mL and weak if >625 µg/mL.⁸

The MIC results of the gompang batu plant extract (*Lobelia nummularia* Lam) against *Staphylococcus epidermis* bacteria show that the minimum concentration values of n-hexane, ethyl acetate and ethanol extracts of the gompang batu plant are respectively 312.5 µg/mL, 312.5 µg/mL and 625 µg/mL. So it can be said that the MIC test of the gompang batu plant extract against *S. epidermis* bacteria is classified as moderate.

The results of the MIC test of gompang batu plant extract (*Lobelia nummularia* Lam) against *Propionibacterium acnes* bacteria are presented in Table 4.

Table 4. MIC results of Gompang Batu plant extract against *Propionibacterium acnes* bacteria

No	Sample	MIC (µg/mL)
1	Chloramphenicol (5000 ppm)	4,88
2	n-Hexane (5000 ppm)	156,25
3	Ethyl Acetate (5000 ppm)	312,5
4	Ethanol (5000 ppm)	625

The MIC results of gompang batu plant extract (*Lobelia nummularia* Lam) against *Propionibacterium acnes* bacteria showed that the minimum concentration values of n-hexane, ethyl acetate and ethanol extracts of gompang batu plants were respectively 156.25 µg/mL, 312.5 µg/mL and 625 µg/mL. So it can be said that the MIC test of gompang batu plant extract against *P. acnes* bacteria is classified as moderate. So it can be said that gompang batu plant extract (*Lobelia nummularia* Lam) against *Staphylococcus epidermis* and *Propionibacterium acnes* bacteria is bacteriostatic. Bacteriostatic is a substance that works by inhibiting bacterial growth.³

Determination of Minimum Bactericidal Concentration (MBC) by Microdilution Method

The results of the MBC test of gompang batu plant extract (*Lobelia nummularia* Lam) against *Staphylococcus epidermis* bacteria are presented in Table 5.

Table 5. MBC Results of Gompang Batu Plant Extract against *Staphylococcus epidermis* bacteria

No	Sample	MBC (µg/mL)
1	Chloramphenicol (5000 ppm)	4,88
2	n-Hexane (5000 ppm)	>2500
3	Ethyl Acetate (5000 ppm)	>2500
4	Ethanol (5000 ppm)	>2500

The MBC results showed that the extract of *L. nummularia* Lam against *S. epidermis* ATCC 6919 was bactericidal. Bactericidal is a substance that can kill bacteria. The working mechanism of antibacterial agents includes inhibition of bacterial cell wall formation, disruption of cytoplasmic membrane function, inhibition of protein synthesis and cessation of metabolic pathways.³

The results of the MBC test of gompang batu plant extract (*Lobelia nummularia* Lam) against *Propionibacterium acnes* bacteria are presented in Table 6.

Table 6. MBC Results of Gompang Batu Plant Extract against *Propionibacterium acnes* bacteria

No	Sample	MBC (µg/mL)
1	Chloramphenicol (5000 ppm)	19,53
2	n-Hexane (5000 ppm)	>2500
3	Ethyl Acetate (5000 ppm)	>2500
4	Ethanol (5000 ppm)	>2500

The MBC results showed that the extract of *L. nummularia* Lam against *P. acnes* ATCC 6919 was bactericidal. Chloramphenicol is bactericidal, because it has an MBC value of 19.53 µg/mL. The inhibitory ability is obtained from the secondary metabolite content found in the extract of gompang batu leaves (*L. nummularia* Lam). Each gompang batu extract contains secondary metabolite compounds such as alkaloids, flavonoids, phenolics, steroids, triterpenoids and tannins. The mechanism of action of alkaloids as antibacterials is by disrupting the activity of enzymes involved in bacterial protein synthesis. As a result, bacterial metabolism is disrupted so that bacterial cells are damaged.²⁹

Flavonoids function directly as antibiotics by interfering with the activity of microorganisms, including bacteria and viruses. The mechanism of flavonoid inhibition of bacterial growth is thought to be related to their ability to form complexes with extracellular proteins, activate enzymes and damage cell membranes. In general, flavonoids are effective in inhibiting the growth of gram-positive and gram-negative bacteria.⁵

Terpenoids found in gompang batu leaf extract likely interact with transmembrane proteins on the outer membrane of the bacterial cell wall and form strong polymer bonds, thereby damaging the transmembrane proteins. According to Alvarez-Martínez et al (2021), damage to transmembrane proteins, which function as entry and exit pathways for important compounds for bacterial cells, will reduce cell wall permeability. As a result, bacterial cells experience a lack of nutrients that inhibit growth and even death in bacteria.²

The way steroids work as antibacterials is related to the interaction between steroid compounds with the surface of the cell wall and bacterial cell membrane, which leads to changes in the primary structure of the bacterial cell wall and cell membrane. These changes can cause the formation of pores or holes and degradation of bacterial cell components. This compound can also interfere with the permeability of the bacterial cell membrane, which causes cell leakage due to seepage. Tannins act as antibacterials by inhibiting the activity of extracellular enzymes in bacteria and binding the substrates needed for their growth. In addition, tannins can also damage polypeptides in bacterial cell walls, which ultimately causes damage to the structure of the cell wall.¹⁷

Based on the One Way ANOVA test, the antibacterial activity of gompang batu plant extracts showed significant differences in each extract ($p < 0.05$). This shows that n-hexane, ethyl acetate and ethanol extracts of gompang batu plants have the potential to inhibit *Staphylococcus epidermis* and *Propionibacterium acnes* bacteria.

4. CONCLUSION

The n-hexane extract of *Lobelia nummularia* Lam contains steroid and tannin compounds, but does not contain alkaloids, flavonoids, and terpenoids. The ethyl acetate extract contains alkaloids, flavonoids, steroids, and tannins but does not contain terpenoids. Meanwhile, the ethanol extract contains alkaloids, flavonoids, terpenoids, and tannins, without steroid content. The most effective antibacterial activity was shown by the ethanol extract at a concentration of 10%, with an inhibition zone diameter of 14.0 mm against *Propionibacterium acnes* and 13.8 mm against *Staphylococcus epidermis*. The MIC values ranged from 156.25 to 625 µg/mL, while the MBC values of all extracts were >2500 µg/mL, indicating that the bactericidal properties were in the moderate category. Gompang batu has the potential as a natural antibacterial for skin infections.

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