



# Formulation and Antibacterial Activity Test Liquid Soap with Eugenol Variant from Clove Leaf Oil (*Eugenia aromaticum*) Against Bacteria *Staphylococcus aureus*

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## Abstract

**Background:** The skin, as the largest organ of the body, hosts various types of bacteria. Excessive sebum production combined with bacterial infections can lead to skin issues such as irritation. Although chemical-based antiseptic soaps are effective, they may negatively impact both the environment and skin health. This has increased public interest in natural alternatives, such as cloves (*Eugenia aromaticum*), which contain eugenol, an active compound with antiseptic, anti-inflammatory, and antibacterial properties. This study aimed to formulate a clove leaf oil-based liquid soap with varying concentrations of clove leaf oil: F1 (0%), F2 (4%), F3 (6%), and F4 (8%), and to evaluate its physical properties and antibacterial activity against *Staphylococcus aureus*. **Methods:** Antibacterial activity assay of the liquid soap was carried out using a disc diffusion method with commercial soap as the positive control (K+) and distilled water as the negative control (K-). The soap quality was evaluated for its organoleptic properties, foam level and stability, pH, spreadability, and viscosity. The disc diffusion method was employed to assess antibacterial activity. **Results:** The organoleptic test showed that the soap had a distinctive clove aroma, a yellow color, and a liquid to thick homogeneous consistency. The pH values ranged from 8.97 to 9.14, foam stability ranged between 84.37% and 88.18%, viscosity ranged from 2510.72 to 6190.57 cP, and spreadability varied from 3.39 to 6.42 cm. Antibacterial testing revealed inhibition zones of 2.87 mm (F1), 7 mm (F2), 10 mm (F3), and 15.25 mm (F4), indicating increasing antibacterial activity with higher clove oil concentrations. **Conclusions:** The F2 formula was identified as the most balanced, with acceptable pH, optimal foam stability, moderate viscosity, and a 7 mm inhibition zone classified in the medium category.

**Keywords:** Antibacterial; Eugenol; Liquid Soap; *Staphylococcus aureus*.



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## Introduction

The skin is one of the largest organs of the body. It contains various types of bacteria, germs, fungi, viruses, and also includes organisms that are harmless but can also be potential pathogens that can cause itching. According to the research by Hidayati et al. (2019), the itching appears due to bacterial infection on the surface of the skin. The bacteria produce enzymes that react directly with skin nerve cells, causing itching. Mochamad et al. (2017), noted that eczema or skin irritation occurs in around 20% of children and 10% of adults.

Skin infections are generally caused by *Staphylococcus aureus* bacteria, accompanied by increased sebum production on surfaces with hair, such as the scalp, hand skin, chest skin, and so on (Mellaratna & Ritonga, 2023). *Staphylococcus aureus* is a bacterium that commonly resides on the surface of the skin. It is found on skin that has been exposed to dry, heated, or cold air for too long. *Staphylococcus aureus* bacteria are found on the skin surface as normal flora, especially around the folds of the skin, nose, mouth, genitals, and

around the anus. This bacterium causes skin infections, usually in the form of a reddish rash accompanied by itching on the skin surface (Mapallawa & Surdam, 2022).

Allah SWT often tempts His servants with various diseases and conditions that have an impact on the body, such as skin irritation. As the outside layer of the body, the skin is susceptible to bacterial infections, germs, and various environmental factors such as hot weather, long-term exposure to sunshine, skin moisture, and air pollution. These factors can cause skin inflammation and other health problems (Kalangi, 2014). Allah SWT says in the Qur'an that Allah has given temptation to

Prophet Ayub AS by afflicting him with a skin disease for years in Surah Al-Anbiyaa' verse 83, which reads:

وَأَيُّوبَ إِذْ نَادَىٰ رَبَّهُ أَنِّي مَسَّنِيَ الضُّرُّ وَأَنْتَ أَرْحَمُ الرَّحِيمِينَ

And (remember the story of) Ayub, when he called out to his God: "O my God, indeed I have been afflicted with a disease, and You are the Most Merciful of all the Merciful".." (Q.S. Al-Anbiyaa': 83).

The skin disease suffered by Prophet Ayub AS was one of the conditions that made the skin injured and caused an excruciating itching, which was a severe test from Allah SWT. However, he remained patient and believed that healing would come by the will of Allah SWT (Hasanah, Agung, & Putra, 2022). Keeping our skin hygienic is essential to prevent skin irritation in our daily lives. Like Prophet Job AS, who kept trusting and surrendering to Allah, we should regularly take care of our health, including skin hygiene, by using antiseptic soap properly. Maintaining skin hygiene is a way to appreciate the health Allah has given us and prevent irritation, infection, and other skin diseases (Shakila et al., 2021).

Bacterial infections on the skin can be prevented by using pharmaceutical preparations that can reduce the growth of microbes in the body. Body wash is a primary alternative for many people to remove dirt or dust that accumulates on the body after activities, thereby protecting the body from disease-causing pathogens or infections. Soap is produced from the reaction of fatty acids and strong bases, which then produce soap and glycerol (Murtadho & Suryandari, 2023). Most of the community prefers liquid soap over solid soap due to its ease of use, portability, and hygienic storage in a tightly closed container (Dimpudus, Yamlean, & Yudistira, 2017).

The packaging used for liquid soap will become inorganic waste that is very difficult to decompose if it is just thrown away. The application of the 3R principle (Reduce, Reuse, Recycle) can be done to minimize community waste. Reduce is to reduce the overall amount of waste by avoiding single-use items. Using refillable liquid soap will reduce the amount of plastic bottle waste. Reuse can be practiced to extend the useful life of items before they go to the trash. Recycling is practiced to convert used items into more valuable objects, such as repurposing used plastic bottles into flower vases. Thus, the use of liquid soap can be the leading choice for the community (Junaidi & Alimun, 2023).

Soap in liquid form with natural components as active substances will be more attractive to the public because it has the advantage of being safer to use, easy to obtain, and, of course, environmentally friendly, so it doesn't cause damage to natural resources (Susilowati, Ariani, & Mulyani, 2022). The utilization of natural materials that have great potential as antimicrobials can reduce the negative impact of chemical use. One of the Indonesian plants that has properties in the health sector is Clove (Lestari, 2017).

Clove, with the Latin name *Eugenia aromaticum*, is an aromatic dried flower stalk with the main content of eugenol, which has antimicrobial activity (Juvensius, Andries, Paulina, Gunawan, & Supit, 2014). Dried clove leaves can be utilized by isolating the oil content using a steam distillation method to obtain clove leaf oil with an eugenol content of

± 80% which has properties such as antibacterial, antiseptic, anti-inflammatory, and antioxidant (Purnomo, 2018). Eugenol, the main constituent of clove leaf oil, is commonly used in health aspects to prevent skin problems, particularly skin infections caused by bacteria (Haryani & Hidayat, 2021). According to research by Fardan & Harimuti (2018), clove leaf essential oil with a 74.8% eugenol concentration can inhibit the growth of *Staphylococcus aureus* bacteria, resulting in an inhibition of 12.5mm, which is classified as

strong. The production of liquid soap using *Eugenia aromaticum* as an antiseptic has not yet been reported. This study aims to produce natural soaps with antiseptic properties from *Eugenia aromaticum*, a natural ingredient in skin care cosmetics.

## Methods

### Materials and Reagents

The tools used in this study include Beaker Cups, Measuring Cups, Erlenmeyer, Reaction Tubes, Reaction Tube Racks, porcelain cups, Petri dishes, Bunsen, Ose Needles, Cotton swabs, Cutter, Hallway, Hot Plate, stirrer, Analytical Scales, pH Meter, Brookfield Viscometer, Refrigerator, Incubator, Jars, Autoclave, Drop Pipettes, Tweezers, Vortex, LAF. The materials used in this study are clove leaf oil, eugenol, Virgin Coconut Oil (VCO), stearic acid, KOH, HPMC, glycerin, Cocamidopropyl betain, distilled water, NaCl, *Staphylococcus aureus* bacteria, and Mannitol Salt Agar (MSA).

### Clove Leaf Oil Certification

Certification of clove oil (*Eugenia aromaticum*) was carried out in this study at PT Selokaton Clove Plantation Banaran, Pesaren, Sukorejo, Kendal, Central Java. The purpose of certification is to obtain the results of the analysis of the content or compounds contained in clove leaf oil, used by researchers to avoid errors in the collection of the main ingredients of the study (Kevin, 2016).

### Procedure

#### *Formulation of Eugenol Clove Leaf Oil Liquid Soap (Eugenia aromaticum)*

The formulation in this study uses a formula that refers to the research of Muna et al. (2021) by making four formulas with varying concentrations of pure eugenol (0%, 4%, 6%, and 8%) as treatments. The preparation of liquid soap was carried out by mixing 30 grams of VCO and clove leaf oil eugenol with concentrations of 4 grams, 6 grams, and 8 grams and stirring using a magnetic stirrer with a heating temperature of 60-70 °C on a hotplate. A KOH solution of 6.2 g was added gradually, and 3.2 g of stearic acid was added to form a soap paste using a magnetic stirrer. Cocamidopropyl betaine was added in an amount of up to 20 g and stirred until homogeneous. 1.4 g HPMC, which has been developed with hot distilled water at 80-90 °C, was then added to the mixture and stirred until homogeneous. Added 6 g of glycerin and stirred until homogeneous. Distilled water is added to the mix until a homogeneous liquid soap preparation is obtained with a dosage volume of 100 ml.

### Evaluation of the physical quality of preparation

#### *Organoleptic tests*

Organoleptic tests were carried out by observing the shape, color, and aroma of the liquid soap formula with the addition of various concentrations of pure eugenol. The standard set by SNI for liquid soap is to have a distinctive odor and color, with a homogeneous liquid dosage form (Tajul et al, 2021).

#### *pH test*

A pH test was carried out by dissolving 1 gram of the preparation into 20 ml of distilled water. pH meters that had previously been calibrated with pH 4, pH 7, and pH 10 buffers were used to calibrate each liquid soap formula that had been dissolved with distilled water. According to SNI standards, the optimum pH of liquid soap is in the range of 8-11 (Lia et al, 2017).

#### *The foam height test*

The foam height test was conducted by dissolving each formula in a test tube to a volume of 1 mL, followed by the addition of 9 mL of water. The mixture was then stirred and shaken for approximately 20 seconds to produce foam. Measured the height of the foam formed and allowed it to stand for 5 minutes. Liquid bath soap has a foam height requirement of 13-220 mm (Tajul et al, 2021).

### ***The homogeneity test***

The homogeneity test was conducted by applying a liquid soap preparation to a glass plate, covering it with another glass plate, and then observing its homogeneity. The preparation is declared homogeneous if there are no undissolved particles when applied on a glass plate (Hutauruk et al, 2020).

### ***The viscosity test.***

The viscosity test was carried out using a Brookfield Viscometer with spindle number 4 at a speed of 30 rpm. The standard value of liquid soap viscosity is 400-4,000 cp (Maydhi, 2021).

### ***The spreadability test.***

The spreadability test was conducted by applying 0.5 g of clove leaf oil eugenol liquid soap preparation to a glass object. A watch glass was then placed over the preparation, followed by a 150 g weight, and the mixture was allowed to stand for 1 minute. The diameter of the spread area was then recorded. Good spreadability values range from 5-7 cm (Rakmadhan et al, 2022)

### **Antibacterial Activity Test: Sterilization of Tools**

Test tubes, Erlenmeyer flasks, measuring cups, and drop pipettes were wrapped with gauze at the mouth and then wrapped in paper. They were sterilized in an autoclave at 121 °C with a pressure of 15 lbs for 15 minutes. Cleaned Laminar Air Flow (LAF) from dust and sprayed using 70% ethanol, left for 15 minutes, and sterilized again with a UV lamp flame for 5 minutes before use (Napitupulu, et al., 2019).

### ***Mannitol Salt Agar (MSA) Media Preparation***

MSA powder was weighed 21.6 g and then suspended in 200 mL of distilled water by heating to form a clear solution. The MSA suspension was then placed in an Erlenmeyer. The mouth of the tube was closed with cotton and gauze, and the setup was sterilized using an autoclave at 121 °C and 15 lbs of pressure for 15 minutes. The sterilized MSA suspension was then poured into sterile Petri dishes of 15 ml each and then cooled and allowed to solidify in the refrigerator (Napitupulu et al., 2019).

### ***Pure Culture of Staphylococcus aureus***

A pure culture of *Staphylococcus aureus* was inoculated onto sloping solid media in test tubes by scraping aseptically, then incubated for 24 hours at 37 °C (Nengah, 2017).

### ***Preparation of Staphylococcus aureus Bacterial Suspension***

Enter 1-2 ounces of *Staphylococcus aureus* culture into a test tube containing 9 mL of 0.9% NaCl. The bacterial suspension was homogenized using a vortex until it reached a turbidity like the McFarland standard (Retnaningsih A., & Dayanti, R., 2017).

### ***Bacterial Isolation***

The method used in bacterial isolation is the continuous scratch inoculation method, which involves scraping the bacterial suspension with a sterile cotton swab onto solid MSA media in a zigzag pattern, both vertically and horizontally, and then allowing it to stand closed for 5-10 minutes (Handayani et al., 2020).

### ***Disc Diffusion Method***

Each blank paper disk was saturated with the test solution for 15 minutes in a sterile petri dish before being applied to the test media. After the sample is absorbed on the paper disk, it is placed on the surface of the media that has been inoculated with bacteria and then incubated for 24 hours at 37 °C. The zone that looks clear around the disc paper is then measured using a caliper (Lukluatun, 2024).

### Measurement of the Zone of Inhibition

Measurement of the inhibition zone on the media can be done after 24-48 hours of incubation. The clear zone formed around the disc paper indicates the effectiveness of the inhibition of clove oil liquid soap preparation against *Staphylococcus aureus* bacteria. The size of the clear zone is measured using a caliper in millimeters vertically and horizontally (Husnul et al, 2023).

### Data Analysis

The data collected were both qualitative and quantitative. The results of the organoleptical test, which include color, aroma, texture, and homogeneity, are presented in descriptive qualitative form. Quantitative data in the form of pH test results, foam height test, viscosity test, and spreadability test. Antibacterial test data against the growth of *Staphylococcus aureus* using the One Way Anova statistical analysis normality test to see differences in antibacterial activity of each sample containing positive control, negative control, and treatment control of clove oil (*Eugenia aromaticum*) liquid soap preparations with a concentration variation of 0%, 4%, 6%, and 8% in inhibiting the growth of *Staphylococcus aureus* bacteria. The statistical program used was SPSS 26 with a significance level of 95%.

## Result

### Organoleptic test.

The organoleptic was evaluated by observing the aroma, colour, and texture. The liquid soap organoleptic test in this study is presented in Table 1.

**Table 1.** Organoleptic Test of Clove Leaf Oil Eugenol Liquid Soap

| Formulations | Parameters            |        |                 |
|--------------|-----------------------|--------|-----------------|
|              | Textures              | Colour | Aroma           |
| F1           | Liquid slightly thick | White  | VCO             |
| F2           | Liquid slightly thick | Yellow | Clove specialty |
| F3           | Thick                 | Yellow | Clove specialty |
| F4           | Very thick            | Yellow | Clove specialty |

Table 1 shows the results of organoleptic tests on various soap formulations. The parameters observed include texture, color, and aroma. Formulation F1 has a slightly thick liquid texture with a white color and a distinctive aroma of VCO. Meanwhile, formulations F2, F3, and F4 exhibit a texture that thickens as the formulation increases, displaying a yellow color and a distinctive aroma of clove oil.

### pH value

pH value determines the soap's suitability as a liquid soap. The pH value in soap indicates the absence of free caustic alkali. The pH value of Clove Leaf Oil Eugenol Liquid Soap in this study is presented in Table 2.

**Table 2.** pH value of Clove Leaf Oil Eugenol Liquid Soap

| Parameters | Formulations | Result            | Standard |
|------------|--------------|-------------------|----------|
| pH         | F1           | 9,14 <sup>b</sup> | 8-11     |
|            | F2           | 9,13 <sup>b</sup> |          |
|            | F3           | 9,09 <sup>b</sup> |          |
|            | F4           | 8,97 <sup>a</sup> |          |

Table 2 shows the pH value of eugenol clove leaf oil and eugenol liquid soap. The pH value of the soap ranges from 9.13 to 8.97, with a tendency to decrease pH as the concentration of clove leaf oil eugenol increases.

### Foam High and Stability Test

Foam is defined as a dispersion system consisting of gas bubbles covered with a layer of liquid. The fatty acid content of the material affects the height of liquid soap foam, as well as its stability and high test results. The results of this study are shown in Table 3.

**Table 3.** Height and Foam Stability Test Results of Eugenia Clove Leaf Oil Liquid Soap

| Parameters                | Formulations | Initial foam (mm) | Final foam (mm) | Result (%) |
|---------------------------|--------------|-------------------|-----------------|------------|
| Foam Height and Stability | F1           | 50                | 43              | 86         |
|                           | F2           | 55                | 48,5            | 88,18      |
|                           | F3           | 56,75             | 47,75           | 84,37      |
|                           | F4           | 53                | 45,5            | 85,84      |

Table 3 shows the results of the foam height and stability test on eugenol clove leaf oil liquid soap preparations. Formulation F3 had the highest initial foam height (56.75 mm), while formulation F1 had the lowest initial foam height (50 mm). After stability testing, the final foam height ranged from 43 mm to 48.5 mm, with the highest percentage of foam stability in formulation F2 (88.18%) and the lowest in formulation F3 (84.37%).

### Homogeneity test

Homogeneity can be achieved by applying the preparation to glass objects or transparent materials and observing that there are no visible particle specks. If there are no details, the preparation can be considered homogeneous based on the research conducted on Liquid soap. If preparations do not form granules, they are considered homogeneous. The results are presented in Table 4.

**Table 4.** Homogeneity Test Results of Clove Leaf Oil Eugenol Liquid Soap

| Parameters  | Formulations | Result  | Standard                |
|-------------|--------------|---------|-------------------------|
| Homogeneity | F1           | Homogen | Homogeneous Preparation |
|             | F2           | Homogen |                         |
|             | F3           | Homogen |                         |
|             | F4           | Homogen |                         |

Table 4 shows the results of the homogeneity test on clove leaf oil eugenol liquid soap preparations. All formulations (F1, F2, F3, and F4) show homogeneous results, in accordance with the standards of homogeneous preparations.

### Viscosity test.

Viscosity is the resistance of a fluid to flow. The liquid soap Viscosity in this study is shown in Table 5.

**Table 5.** Viscosity Test Results of Clove Leaf Oil Eugenol Liquid Soap

| Parameters | Formulations | Result               | Standard    |
|------------|--------------|----------------------|-------------|
| Viscosity  | F1           | 2510,72 <sup>a</sup> | 400-4000 cp |
|            | F2           | 3353,85 <sup>b</sup> |             |
|            | F3           | 5131,62 <sup>c</sup> |             |
|            | F4           | 6190,57 <sup>d</sup> |             |

Table 5 shows the results of the viscosity test on clove leaf oil eugenol liquid soap preparations. The viscosity values of the soap formulations ranged from 2510.72 cp to 6190.57 cp. Formulation F1 had the lowest viscosity (2510.72 cp), while formulation F4 had the highest viscosity (6190.57 cp). Formulations F3 and F4 had viscosity values above the set standard (400-4000 cp).

### Spreadability test

The spreadability test is a method used to assess how easily a formulation. The liquid soap spreadability test in this study is shown in Table 6.

**Table 6.** Spreadability Test of Clove Leaf Oil Eugenol Liquid Soap (*Eugenia aromaticum*)

| Parameters    | Formulations | Result            | Standard |
|---------------|--------------|-------------------|----------|
| Spreadability | F1           | 6,43 <sup>d</sup> | 5-7 cm   |
|               | F2           | 5,52 <sup>c</sup> |          |
|               | F3           | 3,78 <sup>b</sup> |          |
|               | F4           | 3,39 <sup>a</sup> |          |

**Table 6** shows the results of the spreadability test on clove leaf oil eugenol liquid soap preparations. The spreadability values of the soap formulations ranged from 3.39 cm to 6.43 cm. Formulation F1 has the highest spreadability (6.43 cm), while formulation F4 has the lowest spreadability (3.39 cm). Formulations F1 and F2 have a spreadability that complies with the standard (5-7 cm), while formulations F3 and F4 have a spreadability below the standard.

### Antibacterial test

Effective antibacterial activity results can be assessed based on the diameter of the clear zone produced. Results of antibacterial activity test of Oil Eugenol Liquid Soap (*Eugenia aromaticum*) can be seen in **Table 7**.

**Table 7.** Antibacterial Activity Test Results of Clove Leaf Oil Eugenol Liquid Soap (*Eugenia aromaticum*)

| Formulations | Inhibition (mm)    | Categories |
|--------------|--------------------|------------|
| F1           | 2,87 <sup>a</sup>  | Low        |
| F2           | 7 <sup>b</sup>     | Medium     |
| F3           | 10 <sup>c</sup>    | Strong     |
| F4           | 15,25 <sup>d</sup> | Strong     |
| K+           | 18,8               | Strong     |
| K-           | 0                  | Low        |

Note:

F1: liquid soap formulation of clove leaf oil, Eugenol 0%

F2: liquid soap formulation of clove leaf oil, Eugenol 4%

F3: liquid soap formulation of clove leaf oil, Eugenol 6%

F4: liquid soap formulation of clove leaf oil, Eugenol 8%

K+: commercial soap

K-: aquadest

**Table 7** shows the results of the antibacterial activity test on clove leaf oil eugenol liquid soap preparations. The diameter of the inhibition zone ranged from 2.87 mm to 15.25 mm. Formulation F1 has the lowest antibacterial activity with an inhibition zone of 2.87 mm (low category), while formulation F4 has the highest inhibition zone of 15.25 mm (strong category). The positive control (K+) had an inhibition zone of 18.8 mm (strong category), while the negative control (K-) showed no antibacterial activity (inhibition zone 0 mm, low category).

### Discussion

The process of making liquid soap involves mixing VCO and clove leaf oil, then gradually adding KOH until a saponification reaction occurs, which produces glycerol and soap. Melted stearic acid was added and stirred to form a soap paste. Cocamidopropyl betaine is incorporated to create a homogeneous mixture, followed by HPMC, which has been developed with hot water to increase the viscosity of the soap. Glycerin was then added as a humectant to maintain the moisture and homogeneity of the soap, resulting in a softer and more stable preparation.

Physical quality tests were carried out, which included organoleptic test, pH test, homogeneity test, foam height and stability test, viscosity test, and spreadability test.

### Evaluation of Liquid Soap Preparation

Organoleptic tests assessed the shape, color, and aroma of clove leaf oil eugenol liquid soap. Results showed that F1 and F2 were slightly viscous liquids, while F3 and F4 were

thicker to very thick. F1 is white in color with a typical VCO aroma due to no clove leaf oil, while F2, F3, and F4 are yellow in color with a typical clove aroma due to the addition of eugenol. The clove aroma comes from the phenol (-OH) group that binds to the benzene ring in the eugenol structure. These results show that eugenol affects the characteristics of liquid soap (Widodo, Adhani, Solihatun, Prastya, & Annisa, 2020).

The pH test is conducted to control the quality and safety of liquid soap, which tends to be alkaline to remove oil-based dirt effectively. The results show that the pH value of liquid soap is in the range of 9.14-8.97, with F1 having the highest pH (9.14) and F4 the lowest (8.97). This difference is due to variations in the concentration of clove leaf oil eugenol, which is acidic (pH 4-6) due to its phenolic and eugenol content (Haryani & Hidayat, 2022). Nevertheless, all formulations are still in accordance with SNI 06-4085-1996 standards (pH 8-11). The alkaline nature of the soap is influenced by KOH, as well as the addition of stearic acid and eugenol, which help neutralize the pH, so that it remains safe for the skin (Indriyani, 2020).

Foam height and stability tests were conducted to control the quality of clove leaf oil eugenol liquid soap and ensure its safety for the skin. Foam is formed through the saponification reaction between VCO and KOH, and the addition of Cocamidopropyl betain (CAPB) as an amphoteric surfactant that produces soft and environmentally friendly foam (Putri, Hadi, Nuryani, & Ambarwati, 2024). Test results showed foam height in the range of 50-56.75 mm, meeting safety standards (13-220 mm), with F3 having the highest foam (56.75 mm) due to the optimal role of clove leaf oil as a natural surfactant. Foam stability ranged from 84.37-88.18%, meeting the standard (60-90%), with F2 showing the best stability at 4% eugenol concentration, providing a balance between foam formation and stability. Too high a concentration of eugenol may lead to foam breakdown due to its lipophilic nature (Lomboan & Yamlean, 2021).

The homogeneity test aims to ensure that the molecules in a preparation are evenly mixed and stable over a long period of time. The test results show that clove leaf oil eugenol liquid soap has a homogeneous structure, characterized by the absence of coarse particles or components that are not thoroughly mixed. This indicates that the preparation meets the SNI 06-4085-1996 standard, which requires that all ingredients in the product must be evenly mixed without leaving coarse particles or non-uniform colors. Thus, this liquid soap has good quality in terms of homogeneity and formulation stability (Rasyadi & Putri, 2019).

The viscosity test measures the viscosity level of a preparation to ensure the convenience of use and quality. The test results of clove leaf oil eugenol liquid soap showed that F1 had the lowest viscosity (2510.72 cp) and F4 the highest (6190.57 cp), with an increase in viscosity as the concentration of clove leaf oil increased. Formulas 1 and 2 met the viscosity standard (400-4000 cp) (Rosmainar, 2021), while formulas 3 and 4 exceeded the maximum limit. The 99% eugenol content in clove leaf oil, which is hydrophobic, interacts with HPMC as a thickening agent, strengthening the molecular structure and increasing viscosity (Putri, Hidayat, & Rahmah, 2014). These results are in line with the research of Khatimah et al. (2023), which states that the addition of clove flower extract increases the viscosity of the preparation.

The spreadability test measures the ability of liquid soap to spread on the skin surface when used. The test results showed that F1 had the highest spreadability (6.32 cm), while F4 had the lowest (3.39 cm), with values decreasing as the concentration of clove leaf oil eugenol increased. Formulas 1 and 2 (0% and 4% eugenol) met the standard of good spreadability (5-7 cm), providing comfort when applied. In contrast, formulas 3 and 4 did not meet the standard, making it more difficult to spread on the skin (Dhrik & Sawiji, 2023). These results are in line with the viscosity test, which shows that the thicker the preparation, the smaller the resulting spreadability. The viscosity increases due to the hydrophobic nature of eugenol interacting with HPMC as a thickening agent, strengthening the molecular network structure in the liquid and reducing its spreadability (Husnul, Siti, & Destik, 2023).

### Antibacterial Activity Test of Liquid Soap Preparations

Testing the antibacterial activity of clove leaf oil eugenol liquid soap against *Staphylococcus aureus* was carried out by the disc diffusion method on MSA media to assess the potential of the preparation in inhibiting bacteria that cause skin irritation. MSA media is used because it can distinguish *Staphylococcus aureus* from other bacteria, where the pink color of the media will turn yellow due to mannitol fermentation by lipochrome pigments produced by these bacteria (Hayati, Tyasningsih, Praja, Chusniati, & Yunita, 2019). The test results showed that the lowest inhibition was found in F1 with an average of 2.87 mm (weak category), while F4 had the highest inhibition with an average of 15.25 mm (strong category). Statistical analysis with the Post Hoc (Duncan) test showed a significant difference between formulas, confirming that the increase in clove leaf oil eugenol concentration was directly proportional to the rise in bacterial inhibition (Najiah, Hernawati, & Meylani, 2021).

Eugenol affects the growth of *Staphylococcus aureus* by damaging the bacterial cell membrane through lipophilic interactions with phospholipids, leading to increased membrane permeability and leakage of ions and essential metabolites. This disruption inhibits the function of membrane proteins and the DNA replication process, thus inhibiting bacterial growth effectively. The higher the concentration of clove leaf oil in the liquid soap preparation, the greater the inhibition produced, making it a potential antibacterial agent in skin cleansing products (Hasanuddin, 2020).

### Conclusions

Based on the study's results, clove leaf oil eugenol liquid soap meets the quality evaluation standards for preparation in the organoleptic test, pH, foam height and stability, and homogeneity. However, it does not meet the standards in the viscosity and spreadability test. This preparation effectively inhibits *Staphylococcus aureus*, with the most significant inhibition in F4 (8%) of 15.25 mm (strong category). The best formula is F2 with 4% clove leaf oil, which meets SNI 06-4085-1996 standards with homogeneous shape, pH 9.13, foam height 55 mm, foam stability 88.18%, viscosity 3353.85 cp, and spreadability 5.52 cm. This study highlights the potential of clove leaf oil as a natural antibacterial agent in liquid soap formulations.

### Declaration statement

The authors report no potential conflict of interest.

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