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# FORMULATION AND ANTIBACTERIAL ACTIVITY OF LIQUID SOAP FROM KECOMBRANG STEM EXTRACT (*Etlingera elatior*) AGAINST *Staphylococcus aureus*

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## ABSTRACT

Kecombrang (Etlingera elatior) is a plant used by the Baduy ethnic group as a substitute for soap to cleanse the skin and exhibit antibacterial activity. Staphylococcus aureus is the cause of skin infections. Liquid soap is a type of soap that can protect the skin against microorganisms, generally caused by S. aureus. This study aims to develop an antibacterial liquid soap containing kecombrang stem extract, in accordance with the quality requirements of liquid soap, and to determine its antibacterial activity against S. aureus. The extract is formulated into four liquid soap formulas, namely F0, F1, F2, and F3. Physical evaluations include organoleptic properties, homogeneity, pH, liquid soap foam height, viscosity, free alkali content, specific gravity, and antibacterial tests using the well diffusion method. In the organoleptic test, the liquid preparation is brown and has a distinctive odor, with a pH value of around 10.61 to 10.85. The foam height ranges from 33 to 93 mm, viscosity is 412 to 489 cP, specific gravity is 1.02 to 1.08, and free alkali content is 0.05 to 0.16%. Liquid soap with ethanol extract from kecombrang stem inhibits the growth of S. aureus bacteria, with inhibition zones of 9.2 mm for F1, 14.8 mm for F2, and 16 mm for F3, all with p-values <0.05. All formulas met the quality requirements for liquid soap, and F3 (17% extract) demonstrated the highest antibacterial activity.

**Keywords**: Antibacterial, Kecombrang Stem, Liquid Soap, Staphylococcus aureus, Kecombrang

## INTRODUCTION

Indonesia has substantial potential for natural resources. According to various sources, at least 400 ethnic groups in Indonesia are closely tied to nature daily (Badrunasar & Santoso, 2016). One of them is the Baduy ethnic group. Baduy is one of the ethnic groups in Indonesia that reside on the slopes of the Kendeng mountains in Lebak Regency, Banten Province. The Baduy ethnic group utilizes kecombrang (*Etlingera elatior*) to maintain body cleanliness; specifically, the stem is crushed to serve as a soap for cleansing the body (Agustina et al., 2017).

Kecombrang (*E. elatior*) belongs to the Zingiberaceae family, a plant type with antibacterial activity (Effendi et al., 2019). The potential antibacterial activity of kecombrang stems is shown by an essential oil content of 0.0029% and flavonoids (Adini et al., 2025; Lingga et al., 2016). Flavonoid

compounds exert antibacterial effects by disrupting the cytoplasmic membrane, inhibiting energy generation, and blocking nucleic acid synthesis (Tan et al., 2022). At a concentration of 100 ppm, the stem extract of kecombrang stem exhibits antibacterial activity against *S. aureus* with an inhibition zone of 1 mm (Sahidin et al., 2019). Furthermore, at concentrations of 40% and 20%, inhibition zones of 20.56 mm and 15.13 mm, respectively, are observed. Additionally, at a concentration of 78.125 ppm, the extract effectively inhibits the growth of *S. aureus* by up to 33.74% (Adini et al., 2025).

Soap can be a skin cleanser to remove bacteria and dirt (Darusman et al., 2023). One type of soap is liquid soap. Liquid soap has advantages: it is practical, easy to use, hygienic, and stored in tightly closed packaging. Liquid soap that can protect the skin from bacteria is known as antiseptic liquid soap. Antiseptic liquid soap protects the skin from microorganisms that cause infections, generally caused by *S. aureus* (Sari et al., 2024). Liquid soap is generally required to comply with key quality parameters described in previous studies, such as organoleptic properties, homogeneity, appropriate pH, foam height, viscosity, minimal free alkali content, and specific gravity.

Research on kecombrang stem extract has thus far primarily focused on identifying its active compounds and evaluating its antibacterial activity in vitro. However, no studies have developed this extract into a liquid soap formulation and evaluated its antibacterial effectiveness against *S. aureus*. Addressing this gap, the present study aims to formulate a liquid soap containing kecombrang stem extract that meets quality requirements for liquid soap and to evaluate its antibacterial activity against *S. aureus*, a common causative agent of skin infections.

## METHODS

### Materials

pH meter (AMT20), glassware (Pyrex), analytical balance (Fujitsu), disposable Petri dishes (one med), autoclave (Hirayama HVE-50), oven (DHG9053A), rotary evaporator (R-1001 VN), water bath (YNC-WBE-8L), Brookfiled viscometer (NDJ8S), pycnometer (IWAKI), disposable loop (one med), tweezers, vernier calliper (Mitutoyo), Laminar Air Flow (LAF) (MYCO7), micropipette (JoanLab), test tube (Pyrex), Magnetic Stirrer (IKA C-MAG HS 7) and Incubator (Memmert). Kecombrang stem (*Etlingera elatior*), *S. aureus* culture strain ATCC 25923, olive oil (Organic Essential Oil), potassium hydroxide (Brataco), carboxyl methyl cellulose

(Brataco), stearic acid, butyl hydroxy anisole (BHA), phenolphthalein, 70% alcohol, Nutrient agar (Merck), Dettol liquid soap, 0.9% NaCl (B Braun), 0.1 N HCl, distilled water, and Mc. Farland solution 0.5.

**Research Path**

**1. Plant determination**

The determination of kecombrang plants (*Etlingera elatior*) was carried out at the Biology Laboratory of the Faculty of Science and Technology, Ahmad Dahlan University, Yogyakarta.

**2. Kecombrang stem extraction**

Extraction was carried out using the kinetic maceration method. 1 kg of kecombrang simplicia was extracted using 70% ethanol at a ratio of 1:20 for 2 hours (Adini et al., 2023).

**3. Kecombrang stem extract liquid soap formulation**

All the ingredients are weighed first according to the formula. Add olive oil and potassium hydroxide to a beaker and heat using a magnetic stirrer at 60°C at a stirring speed of 400 rpm for 45 minutes. Add enough distilled water to the liquid soap paste, then stir the CMC dissolved in hot distilled water until homogeneous. Then, add stearic acid and SLS, kecombrang stem extract, and stir until the mixture is homogeneous. Add distilled water until the volume reaches 100 ml (Hasibuan et al., 2019). The formulation was prepared in three independent batches (biological replicates) to account for variability in plant extract–based formulations.

**Table 1.** Formulation of liquid soap from kecombrang stem extract (% w/v)

| Ingredients | Uses | F0 | F1 | F2 | F3 |
|---|---|---|---|---|---|
| Kecombrang stem extract | Active ingredient | 0 | 7 | 12 | 17 |
| Olive oil | Fatty acid | 15 | 15 | 15 | 15 |
| Potassium hydroxide (KOH) | Alkali | 8 | 8 | 8 | 8 |
| Carboxymethyl Cellulosa (CMC) | Thickener | 0.5 | 0.5 | 0.5 | 0.5 |
| Sodium Lauryl Sulfate (SLS) | Surfactant | 0.5 | 0.5 | 0.5 | 0.5 |
| Stearic acid | Foam stabilizer | 0.25 | 0.25 | 0.25 | 0.25 |
| Green tea | Fragrance | 1 | 1 | 1 | 1 |
| Distilled water | Solvent | ad 100 | ad 100 | ad 100 | ad 100 |

**4. Liquid soap evaluation**

All evaluations of the liquid soap formulas were performed using biological triplicate batches.

**4.1 Organoleptic test**

Organoleptic testing involves observing the colour, odour, and shape of liquid soap.

### 4.2 Homogeneity test

A homogeneity test is performed by applying the preparation to an object glass and then observing the surface's appearance and whether any separate parts are present. The preparation of a piece of clear glass, observed under a microscope, will reveal a homogeneous composition; no particle spots should be visible. The preparation can be homogeneous if no grains exist (Rinaldi et al., 2021).

### 4.3 pH tes

First, the pH meter will be calibrated using a standard pH meter buffer (pH 7.01) and an acid buffer solution (pH 4.01) until the device displays the correct pH value. Then, the electrode is washed with distilled water and dried with tissue. The sample is made with a concentration of 1%. Then, the electrode is dipped in the solution. Leave it until the device shows a constant pH number. The number displayed on the pH meter is the pH of the liquid soa (Leny et al., 2022).

### 4.4 Foam height test

The foam height test is done by measuring the height of the foam in a measuring cup. A sample of 1 gram of liquid soap in 50 mL of distilled water is placed into a 100 mL closed measuring cup and shaken evenly for 20 seconds. Let it stand for 5 minutes, and measure the height of the foam (Sari & Ferdinan, 2017).

### 4.5 Viscosity test

The foam height test is done by measuring the height of the foam in a measuring cup. A sample of 1 gram of liquid soap in 50 mL of distilled water is placed into a 100 mL closed measuring cup and shaken evenly for 20 seconds. Let it stand for 5 minutes, and measure the height of the foam (Sari et al., 2022).

### 4.6 Alkali free test

Weigh 5 g of liquid soap and place it in neutral alcohol. Heat until almost boiling, then add a phenolphthalein indicator. If the solution is not alkaline (i.e., not red), cool to 70 °C and titrate with a 0.1 N KOH solution in alcohol until a red color lasts for 15 seconds (Aras & Lestari, 2024).

### 4.7 Specific gravity test

The pycnometer is dried and weighed. Water is then put into the pycnometer, left at 25°C for 10 minutes, and weighed. To repeat the test, a liquid soap sample is used in place of water (Leny et al., 2022).

### 4.8 Antibacterial activity test

The antibacterial test uses the well diffusion method. Each plate was inoculated with 100 µL of *a S. aureus* suspension standardized to 0.5 McFarland, followed by the addition of 20 mL of nutrient agar (NA), and then allowed to become homogeneous and solidified. Wells with a diameter of 6 mm were created in each agar plate using a cork

borer. Put liquid soap test samples F0 (negative control), F1, F2, F3, and Dettol liquid soap (positive control) into the well for 15 µl. The plates were incubated aerobically at 37 °C for 18–24 hours. After incubation, the diameter of the inhibition zone was measured using a vernier caliper with a precision of 0.01 mm (Peters et al., 2018). All tests were conducted in triplicate.

**Data Analysis**

All statistical analyses were conducted using SPSS. Before testing the hypotheses, we assessed the normality of the data with the Shapiro–Wilk test and evaluated the homogeneity of variance using Levene's test. Data that met both assumptions ($p > 0.05$) were analyzed with a parametric one-way ANOVA. For datasets that did not satisfy these assumptions ($p < 0.05$), we applied a non-parametric Kruskal–Wallis test.

## RESULTS AND DISCUSSION

### 1. Plant determination

Plant determination was conducted at the Biology Laboratory, Faculty of Science and Technology, Ahmad Dahlan University, Yogyakarta. The results were based on certificate No. 163/Lab.Bio/B/III/2024 states that the plant used was indeed the kecombrang plant, *Etlingera elatior*.

### 2. Kecombrang stem extraction

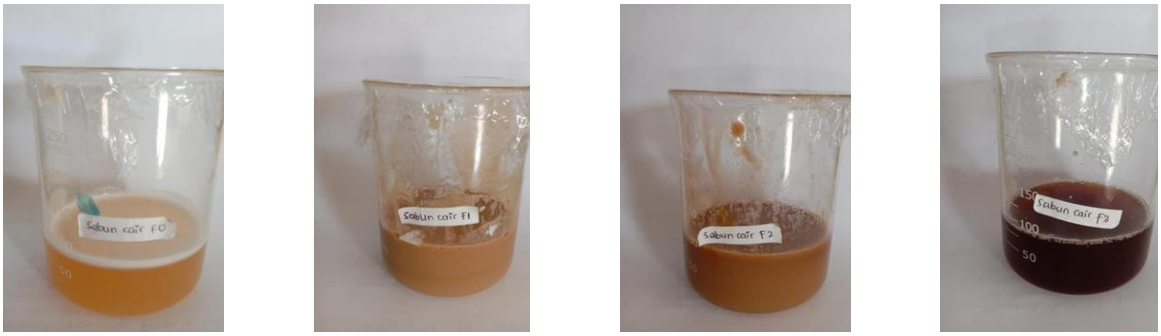
The contracted extract was obtained from the kinetic maceration process, using a 1:10 ratio, specifically 1 kg of kecombrang stem powder with 20 litres of 70% ethanol solvent. The filtrate was concentrated using a rotary evaporator at 50°C to concentrate the extract and separate the solvent from the active compound.

**Table 2**. Extract the yield of kecombrang stem

| weight of simplicia powder (g) | Weight contracted extract (g) | Extract yield (%) |
|---|---|---|
| 1000 | 75.6 | 7.56 |

Table 2 shows that the yield of kecombrang stem extract was 7.56%. This result is higher than that of Dimpudus et al. (2017), which produced a yield of kecombrang stem methanol extract of 4.06%. This difference in yield is attributed to the use of different solvents, as the solvent can significantly impact the extract's yield (Kadek Widhiana Putra et al., 2020).

### 3. Liquid soap evaluation




(a) (b) (c) (d)

**Figure 1.** Liquid soap with kecombrang stem extract; (a) Formula 1 without extract; (b) Formula 2 with 7% extract; (c) Formula 3 with 12% extract; (d) Formula 3 with 17% extract

### 3.1 Organoleptic test

**Table 3.** Organoleptic test results

| Formula | Colour | Odour | Shape |
|---|---|---|---|
| F0 | White | Distinctive liquid soap odour | Liquid |
| F1 | Brown | Distinctive kecombrang aromatic | Liquid |
| F2 | Brown | Distinctive kecombrang aromatic | Liquid |
| F3 | Dark Brown | Distinctive kecombrang aromatic | Liquid |

Note:
F0: Liquid soap formula without kecombrang stem extract
F1: Liquid soap formula with 7% kecombrang stem extract
F2: Liquid soap formula with 12% kecombrang stem extract
F3: Liquid soap formula with 17% kecombrang stem extract

Organoleptic tests were conducted by observing changes in the liquid soap preparation, including color, odor, and shape. Organoleptic tests of liquid soap preparations with kecombrang stem extract revealed differences in color and odor between F0, F1, F2, and F3. The difference is because F0 does not contain extracts, F1 and F2 have almost similar colours, while F3 has a more concentrated colour. The odour of F0 is typical of liquid soap due to the aroma of the fragrance used. In contrast, F1, F2, and F3 retain the typical odour of kecombrang stem extract, and although they have been treated with a fragrance, they do not eliminate the characteristic odour of kecombrang extract. The greater the concentration of liquid soap in the formula, the more concentrated the colour and the usual odour of the kecombrang stem extract.

### 3.2 Homogeneity test

**Table 4.** Homogeneity test results

| Formula | Homogeneity | Qualification |
|---|---|---|
| F0 | Homogeneous | Qualify |
| F1 | Homogeneous | Qualify |
| F2 | Homogeneous | Qualify |
| F3 | Homogeneous | Qualify |

The homogeneity test utilizes a microscope to verify that all particles are uniform in size and composition. The test results on all liquid soap formulas showed no coarse grains, indicating that the material particles were mixed homogeneously and qualified.

### 3.3 pH tes

**Table 5.** pH test results

| Formula | pH | | | Average ± SD | Qualification |
|---|---|---|---|---|---|
| | 1 | 2 | 3 | | |
| F0 | 10.6 | 10.62 | 10.57 | 10.6±0.03 | Qualify |
| F1 | 10.65 | 10.63 | 10.63 | 10.64±0.01 | Qualify |
| F2 | 10.83 | 10.82 | 10.82 | 10.82±0.01 | Qualify |
| F3 | 10.86 | 10.85 | 10.84 | 10.85±0.01 | Qualify |

The pH test is an essential parameter because liquid soap is in direct contact with the skin and can cause problems if the pH value does not match the skin's pH. A pH value that is too low can increase the absorption of liquid soap on the skin, potentially causing skin irritation (Untari & Robiyanto, 2018). However, if the pH is too alkaline, it can cause swelling of the stratum corneum protein, damage the lipid lamellar structure through the ionization of fatty acids, destabilize the skin barrier function, and disrupt the normal microflora, allowing the growth of opportunistic bacteria such as *S. aureus* (Mijaljica et al., 2022). When the pH of the formulation is excessively alkaline, neutralization is required to achieve an acceptable pH range. The addition of citric acid is widely applied in liquid soap formulations because it effectively reduces free alkali and adjusts the pH (Dianursanti et al., 2020). Based on Table 5, all liquid soaps with kecombrang stem extract formulas meet the pH value requirement according to SNI 06-4085-1996, which is 8-11 (Dewan Standarisasi Nasional, 1996). The Kruskal–Wallis analysis demonstrated that variations in kecombrang stem extract concentration significantly affected the pH value, with a p-value of 0.017 (< 0.05).

### 3.4 Foam height test

**Table 6.** Foam height test results

| Formula | Foam Height (mm) | | | Average ± SD (mm) | Qualification |
|---|---|---|---|---|---|
| | 1 | 2 | 3 | | |
| F0 | 30 | 36 | 35 | 33.67±3.21 | Qualify |
| F1 | 60 | 62 | 67 | 63±3.61 | Qualify |
| F2 | 80 | 85 | 88 | 84.33±4.04 | Qualify |
| F3 | 90 | 93 | 66 | 93±3 | Qualify |

The foam height test aims to evaluate the amount of foam produced by the liquid soap formula. Based on Table 6, all liquid soap formulations containing kecombrang stem extract met the acceptable foam height range of 13–220 mm (Dimpudus et al., 2017). One-way ANOVA analysis demonstrated that variations in the concentration of kecombrang stem extract had a significant effect on foam height, with a p-value of 0.000 (< 0.05).

### 3.5 Viscosity test

**Table 7.** Viscosity test results

| Formula | Viscosity (cps) | | | Average ± SD (cps) | Qualification |
|---|---|---|---|---|---|
| | 1 | 2 | 3 | | |
| F0 | 417.5 | 413 | 405.5 | 412±6.06 | Qualify |
| F1 | 450 | 460 | 450 | 453.33±5.77 | Qualify |
| F2 | 460 | 470 | 475 | 468.33±7.64 | Qualify |
| F3 | 489 | 480 | 499.5 | 489.5±9.76 | Qualify |

Based on Table 7, all liquid soap formulations of kecombrang stem extract meet the viscosity requirements according to SNI 06-4085-1996, which is a range of 400-4000 cps. The higher the kecombrang stem extract concentration, the higher the viscosity obtained. Increasing the concentration of extract in formulas F1, F2, and F3 resulted in a corresponding rise in viscosity. This observation aligns with the typical characteristics of plant extracts, which are rich in secondary metabolites and polysaccharides. These constituents can increase the density of the formulation, resulting in higher viscosity as the extract concentration increases (Xie et al., 2023). The one-way ANOVA data analysis results show that differences in kecombrang stem extract concentration can significantly affect the viscosity of liquid soap, with a p-value of 0.000 (< 0.05).

### 3.6 Alkali free test

**Table 8.** Alkali free test results

| Formula | Alkali free (%) | | | Average ± SD (%) | Qualification |
|---|---|---|---|---|---|
| | 1 | 2 | 3 | | |
| F0 | 0.167 | 0.145 | 0.156 | 0.16±0.01 | No qualify |
| F1 | 0.112 | 0.089 | 0.089 | 0.1±0.01 | Qualify |
| F2 | 0.1 | 0.078 | 0.067 | 0.08±0.02 | Qualify |
| F3 | 0.067 | 0.044 | 0.044 | 0.05±0.01 | Qualify |

The requirement for the free alkali value of liquid soap, according to SNI 06-4085-1996, is ≤0.14% (Dewan Standarisasi Nasional, 1996). Based on Table 8, the free alkali value in F0 does not meet the requirements because it is >0.14%. This elevated value suggests that the saponification process in the base formula may not have been entirely complete (Sukeksi et al., 2021). In contrast, the free alkali values in F1, F2, and F3 were within the acceptable range (<0.14%). The observed reduction in free alkali values in F1–F3 may be related to the addition of kecombrang stem extract. Plant extracts are commonly rich in phenolic and flavonoid compounds, which exhibit characteristic behavior under alkaline conditions (phenolate formation and other alkali-catalyzed reactions), and previous formulation studies have reported changes in measured free alkali following incorporation of herbal extracts (Ji et al., 2019; Pasquet et al., 2024). The results of data analysis using one-way ANOVA showed that differences in the concentration of kecombrang stem extract could significantly affect the free alkali value, with a p-value of 0.000 (< 0.05).

### 3.7 Specific gravity test

**Table 9.** Specific gravity test results

| Formula | Specific Gravity (g/mL) | | | Average ± SD (g/mL) | Qualification |
|---|---|---|---|---|---|
| | 1 | 2 | 3 | | |
| F0 | 1.02 | 1.01 | 1.03 | 1.02±0.01 | Qualify |
| F1 | 1.03 | 1.02 | 1.05 | 1.03±0.02 | Qualify |
| F2 | 1.02 | 1.1 | 1.08 | 1.07±0.04 | Qualify |
| F3 | 1.08 | 1.06 | 1.09 | 1.08±0.02 | Qualify |

Based on Table 9, all liquid soap formulas have a specific gravity that meets the requirements according to SNI 06-4085-1996, which falls within the 1.01-1.1 g/mL range (Dewan Standarisasi Nasional, 1996). The results of data analysis using the

Kruskal-Wallis test showed that differences in extract concentrations had no significant effect on specific gravity in liquid soap, with a p-value of 0.081 ($p > 0.05$).

### 3.8 Antibacterial activity test

The well-diffusion method was chosen because the liquid soap formulation contains surfactants and has a viscosity that prevents uniform absorption into the paper disc, making disc diffusion less suitable for this formulation. Furthermore, the well-diffusion technique enables the accurate delivery of sample volume, ensuring direct contact between the liquid formulation and the agar surface (Bubonja-Šonje et al., 2020; Hossain, 2024). Although the agar well diffusion method is widely used for preliminary screening of antibacterial activity, the clinical interpretation of inhibition zones requires caution. The measured zone diameter largely reflects the diffusion characteristics of the tested compounds in the agar matrix, which are governed by molecular weight, solubility, and penetration kinetics, rather than solely their intrinsic antimicrobial potency (Hossain, 2024).

**Table 10.** Antibacterial activity results

| Sample | Zone of Inhibition (mm) | | | Average ± SD (mm) |
|---|---|---|---|---|
| | 1 | 2 | 4 | |
| K+ (Dettol liquid soap) | 20 | 18.7 | 17.6 | 18.77±1.20 |
| F0 | 0 | 0 | 0 | 0.00±0.00 |
| F1 | 7.7 | 9.4 | 10.5 | 9.20±1.41 |
| F2 | 16 | 14 | 14.6 | 14.87±1.03 |
| F3 | 17.1 | 15.5 | 15.4 | 16.00±0.95 |

Based on Table 10, F3 exhibited the highest antibacterial activity among all test formulas, with an inhibition zone of 16.00 ± 0.95 mm against *S. aureus*. The greater activity of F3 was associated with its higher kecombrang stem extract concentration compared to F1 and F2, demonstrating a dose-response relationship where antibacterial activity increased with extract concentration. Compared to the positive control (Dettol liquid soap), which produced an inhibition zone of 18.77 ± 1.20 mm, F3 showed slightly lower activity. The antibacterial effect of Dettol is primarily due to chloroxylenol, which disrupts microbial cell membranes and inhibits vital intracellular enzymatic processes (Han et al., 2024). In F0, the addition of green tea fragrance did not produce any inhibition zone, confirming that the fragrance does not contribute to antibacterial activity. Therefore, the observed antibacterial effects

in the test formulas can be attributed solely to the kecombrang extract. One-way ANOVA analysis showed a significant difference among the inhibition zones of all samples (K+, F0, F1, F2, and F3) on *S. aureus* growth, with a p-value of 0.000 (<0.05).

All liquid soap formulas developed in this study met the physical quality requirements. However, in terms of sensory characteristics, there were still shortcomings, such as the soap's unattractive color and the distinctive aroma of kecombrang stems that could not be completely masked by adding fragrances. Therefore, future studies should explore safe natural colorants and more effective combinations of fragrance agents to better mask the distinctive aroma of active ingredients without compromising their antibacterial efficacy.

## CONCLUSIONS

The ethanol extract of kecombrang stem (Etlingera elatior) can be successfully formulated into a liquid soap that meets the criteria for homogeneity, pH, foam height, viscosity, free alkali content, and specific gravity. Among the tested formulations, F3 demonstrated the best performance, producing an inhibition zone of 16.00 ± 0.95 mm.

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