

## Formulation, Stability Test and Antibacterial Activity of Banana Leaf Extract Liquid Soap (*Musa sp.*) against the Growth of *Escherichia coli*

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**Article Info****Article history:**

Received May 7, 2024

Revised July 29, 2024

Accepted October 3, 2024

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**Keywords:**

Antibacterial activity

Banana leaf extract

*Escherichia coli*

Liquid soap

Stability tests

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**ABSTRACT**

Liquid soap is an important component in maintaining skin cleanliness. Good liquid soap is soap that contains natural antioxidants because it can protect the body from free radicals and prevent cell damage and is also antibacterial because it can prevent and kill pathogens. One source of easily available phytochemical compounds with antioxidant and antibacterial activity is banana leaves (*Musa sp.*). The aim of this research was to formulate physically and chemically stable banana leaf extract liquid soap and test the antibacterial effectiveness of banana leaf extract liquid soap with formulations of 0%, 1%, 3%, 5% and 7% against the growth of *Escherichia coli*. This research was carried out using Completely Randomized Design which included making extracts, phytochemical screening tests, organoleptic tests, hedonic tests, pH tests, homogeneity tests, foam stability tests, and antibacterial tests on *Escherichia coli* using the diffusion method (paper disc). Then statistical analysis of variance (ANOVA) was carried out to determine the differences in the five formulas. Integrating banana leaf phytochemical compounds into soap formulas produces products that are antioxidant and antibacterial. The best formula is at an extract concentration of 7% because it provides optimal antibacterial test results and meets SNI standards for foam stability.

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**1. INTRODUCTION**

One health problem that can arise, especially due to the growth of *Escherichia coli*, is diarrhea. Diarrhea is the most common cause of death in infants and toddlers. According to the Central Statistics Agency (BPS) in 2020, the death rate for toddlers (under 5 years) in Indonesia reached 28,158 people in 2020. From this data, 20,266 toddlers (71.97%) died at the age of 0-28 days (neonatal). A total of 5,389 toddlers (19.13%) died at the age of 29 days – 11 months (post neonatal), 2,506 toddlers (8.9%) died at the age of 12-59 months. Meanwhile, 9.8% of post-neonatal toddlers die from diarrhea (Milindasari, 2024). Symptoms involve passing stools with a soft or liquid consistency, often three or more times in one day. Diarrhea can cause poor nutritional status, growth failure, and permanent weight loss due to fluid loss and dehydration (Arda *et al.*, 2020 in Hutasoit, 2020). Factors that cause diarrhea include microbes such as *Escherichia coli* and contaminated food (Amaliah, 2010 in Hutasoit, 2020) and environmental, individual and behavioural factors such as water quality, malnutrition, sanitation and food hygiene. Diarrhea can cause serious complications such as loss of body fluids, hypovolemic shock, organ damage, and even coma. Factors such as uncleanliness, crowded environment and lack of clean water also contribute to diarrhea (Utami, 2016 in Hutasoit, 2020).

Liquid soap is an integral component and basic human need in maintaining skin hygiene. Several variations of liquid bath soap are produced through factory production processes, with the productive distribution regarding variations in aroma, colour, type and benefits. The safe and marketable quality of liquid bath soap is determined through the saponification process, where fat or oil reacts with an alkali metal base (Potassium hydroxide or Sodium hydroxide). Heating fat with potassium hydroxide causes saponification, changing the fat into fatty acids and glycerol in an alkaline environment, producing soap. With population growth increasing every year, the use of liquid soap is expected to increase, opening up significant opportunities for Indonesia in the liquid bath soap production industry (Karina *et al.*, 2022). Currently, there are cosmetic preparations circulating on the market, both chemical and natural, but it is known that the use of cosmetics made from chemicals is still very high even

though they can cause skin damage and cancer. This condition encourages innovation in the development of cosmetics made from natural ingredients to improve skin safety and reduce side effects (Ekayanti et al., 2023). One potential natural raw material that can be utilized is abundant local plants, for example banana plants.

The banana plant (*Musa sp.*), known as the "plant of a thousand uses" in Indonesia, offers diverse benefits, especially in the cosmetic realm. Cosmetics, as skin care products, are vital in treating various skin problems, from moisturizing to preventing premature aging. Amid concerns about the negative impact of cosmetics made from chemicals, the development of cosmetic formulations made from natural ingredients is increasingly receiving attention, driven by safety and the lack of side effects. Banana plants are rich in flavonoids, alkaloids, and tannins. Flavonoids, as natural antioxidants, protect the body from free radicals and prevent cell damage. Alkaloids, which are distributed in various parts of the plant, have physiological effects and antibacterial activity. Tannin has a bitter and chelating taste and is generally used as an anti-inflammatory and anti-diarrhea. Banana leaves are known to contain flavonoids, phenolics and tannins which are useful as anti-inflammatories. Using banana plants in cosmetics offers a potential natural alternative, reducing chemical concerns. These ingredients provide a scientific basis for developing natural cosmetic products that are effective and safe for the skin (Ekayanti et al., 2023).

Based on this description, apart from reducing dependence on synthetic ingredients in cosmetic products, especially soap, the formulation of banana leaf extract into soap can increase the safety of natural products for the skin and minimize the side effects they cause. This research aims to determine whether banana leaf extract (*Musa sp.*) can be formulated into physically and chemically stable liquid soap and to test the antibacterial activity of banana leaf extract liquid soap with concentrations of 0%, 1%, 3%, 5% and 7% against *Escherichia coli*.

## 2. RESEARCH METHOD

This research is an experimental research using Completely Randomized Design which includes making extracts, phytochemical screening tests, organoleptic tests, hedonic tests, pH tests, homogeneity tests, foam stability tests, and antibacterial tests. The antibacterial effectiveness test on the growth of *Escherichia coli* was carried out using the diffusion method (paper disc). Then, statistical analysis was carried out using analysis of variance (ANOVA) to determine the differences in the five formulas.

The main focus of this research involves the stages of formulation development, stability testing, and evaluation of the antibacterial activity of liquid soap. The liquid soap that will be formulated contains banana leaf extract (*Musa sp.*). In addition, this research will evaluate the potential of liquid soap to inhibit the growth of *Escherichia coli*. These steps can contribute to the development of cosmetics made from natural and local ingredients and support efforts to maintain holistic skin health.

The first thing to do is to prepare the banana leaf extract. Preparation of banana leaf extract includes washing, drying and making the extract. Washing banana leaves is done by washing them thoroughly under running water and drying them in the oven for 24 hours at 40°C until the water content is reduced from 10%. Next, the dried banana leaves are crushed using a blender and sieved using a sieve (Widarta & Wiadnyani, 2019). Next, banana leaf extract is made using 70% ethanol (Jaya Mulyo Kimia & Parfum, Special Region of Yogyakarta, Indonesia). Banana leaf powder is given ethanol in a ratio of 1:5 (44 g banana leaf powder: 220 ml ethanol). Then, the simplicial powder is poured into a jar with ethanol, soaked for 2 x 24 hours (2 days), and filtered to obtain 200 ml of simplicia. After obtaining the liquid extract, it was thickened in a water bath at 60°C for 45 minutes (Nisa et al., 2017).

Next is the screening of phytochemical compounds. Next is the screening of phytochemical compounds which include alkaloid test, flavonoid test, saponin test, and tannin test. Alkaloid test is done by putting 1 mL of extract into a test tube, then adding 3 drops of dragendorff reagent (Molecular Laboratory of Universitas 'Aisyiyah Yogyakarta, Special Region of Yogyakarta, Indonesia). If an orange or brick red precipitate is formed, the extract is positive for containing alkaloids. Flavonoid test is done by putting 1 mL of extract into a test tube, then heating 0.2 mg of magnesium powder (Molecular Laboratory of Universitas 'Aisyiyah Yogyakarta, Special Region of Yogyakarta, Indonesia) and 3 drops of concentrated hydrochloric acid (Molecular Laboratory of Universitas 'Aisyiyah Yogyakarta, Special Region of Yogyakarta, Indonesia) at a temperature of 105°C. If a red, yellow, or orange solution is formed, the extract is positive for containing flavonoids. Saponin test is done by putting a total of 1 mL of extract into a tube. The reaction was then added with 1 mL of warm distilled water (Jaya Mulyo Kimia & Parfum, Special Region of Yogyakarta, Indonesia) and shaken vigorously for ± 1 minute. If stable foam is formed, the extract is positive for containing saponin. Tannin test is done by putting a total of 1 mL of extract into a test tube, and then adding 1 mL of 1% ferric chloride (FeCl<sub>3</sub>) (Molecular Laboratory of Universitas 'Aisyiyah Yogyakarta, Special Region of Yogyakarta, Indonesia). If a blackish-green solution is formed, the extract is positive for containing tannin (Adjeng *et al.*, 2019).

After knowing the phytochemical compound content in banana leaf extract, the next step is to formulate the banana leaf extract into a banana leaf extract liquid soap formulation as shown in the following table.

Table 1. Banana Leaf Extract Liquid Soap Formulation

Materials	Liquid Soap Formulation					Units (%w/v)	Function
	F0	F1	F2	F3	F4		
Banana Leaf Extract	0	1	3	5	7	%	Active substance
Coconut oil	30	30	30	30	30	%	Fatty acids
Potassium hydroxide	16	16	16	16	16	%	Basa
CMC-Na	1	1	1	1	1	%	Fillers and thickeners
Sodium Lauryl Sulfate	1	1	1	1	1	%	Surfactant
Stearic acid	0.5	0.5	0.5	0.5	0.5	%	Stabilizer
Essential oils	2	2	2	2	2	%	Fragrance
Distilled water	Add	Add	Add	Add	Add	%	Solvent
	100	100	100	100	100		

(Legi *et al.*, 2021)

Based on the formula above, liquid soap from banana leaf extract is made in 5 formulations with different concentrations, namely 0%, 1%, 3%, 5% and 7%. All ingredients are weighed first according to the formula that has been made. After that, 30 mL of coconut oil (Boga Mas) is put into a beaker, and then 16 mL of Potassium hydroxide (Jaya Mulyo Kimia & Parfum, Special Region of Yogyakarta, Indonesia) is added little by little while heating at 50°C. Stir using a magnetic stirrer until a soap paste is formed. 30 mL distilled water was added to the soap paste, then CMC-Na (Jaya Mulyo Kimia & Parfum, Special Region of Yogyakarta, Indonesia) was developed in hot distilled water and stirred until smooth. Stearic acid (Jaya Mulyo Kimia & Parfum, Special Region of Yogyakarta, Indonesia) is added and stirred until smooth. Then, add SLS (Jaya Mulyo Kimia & Parfum, Special Region of Yogyakarta, Indonesia) and stir until smooth. Add vanilla aroma essential oil (Jaya Mulyo Kimia & Parfum, Special Region of Yogyakarta, Indonesia) and stir until smooth. Banana leaf extract is added according to the formula and then stirred again until smooth. Finally, distilled water is added until the volume is 100 mL, and the soap is put into a clean container that has been prepared, and the soap can be tested (Legi *et al.*, 2021).

After the process of making liquid soap from banana leaf extract is complete, several tests are carried out. The first test carried out is the organoleptic test. Organoleptic testing is carried out by looking at the naked eye to see a preparation's physical appearance, including shape, color, and smell (Syamsu *et al.*, 2022). The second test is the hedonic test. The hedonic test was carried out using the organoleptic test method for a total of 10 people where each panellist was given a sample, namely banana leaf extract liquid soap with the formula F0, F1, F2, F3, and F4, who would fill out a questionnaire regarding the soap (aroma, texture and color). The technique used to determine the panelists in this study was the non-probability sampling technique (Accidental Sampling) with the criteria for panelists in this study: male/female at least 20 years old, not currently experiencing skin disorders such as tinea versicolor, ringworm, ringworm and so on, no history of skin disease, and not currently experiencing respiratory problems (Haque *et al.*, 2022).

This is due to the level of skin stability they have. At the age of 20 years and over, skin conditions are generally stable and will not experience drastic changes that usually occur in adolescence. This makes the evaluation of liquid soap more consistent and reliable. On the other hand, skin thickness will also increase during the first 20 years of life with a stable number of cells in each layer (Yusharyahya, 2021). Skin aging at the age of 20 is irreversible which is a complex biological process resulting from intrinsic (from within the body such as genetics) and changes that develop over time and extrinsic impacts caused by environmental factors (Mackiewicz & Rimkevicius, 2008 in Runtuwene *et al.*, 2019).

The third test is the pH test. PH paper is used for pH testing. A good pH for liquid bath soap is 8-11 (Syamsu *et al.*, 2022). The fourth test is the homogeneity test of liquid soap. The homogeneity test was done by using a glass object, taking 0.5 mL of the preparation, attaching it to the glass object, and then glass object for observation (Pardosi, 2018 in Shamsu *et al.*, 2022). Testing can be done to see if there is particle rough or material that is not evenly mixed in a way, as well as a formed lump in preparation so stated homogeneous (Kharisma & Safitri, 2020). The fifth test is the height and stability test of liquid soap foam. Test the height and stability of the foam using a 1g sample of liquid soap, then put it in a tube containing 10 mL of distilled water, cover it and shake it for approximately 20 seconds, then measure the height of the foam. Then, leave it for 5 minutes and measure the height of the foam reduction (Syamsu *et al.*, 2022). Then, the calculation is carried out:

$$\text{Foam Stability \%} = (\text{final foam height})/(\text{initial foam height}) \times 100\%$$

The sixth test is an antibacterial test against *Escherichia coli*. First of all, it begins with preparation *Escherichia coli* inoculum. Bacteria were obtained from the Molecular Biology Laboratory of Universitas 'Aisyiyah Yogyakarta. *Escherichia coli* colonies were taken from bacterial culture stocks that had grown on slanted Nutrient Agar (NA) media (Merck) using a sterile loop needle. Next, it is suspended in sterile Nutrient Broth (NB) media (Merck) and shaker for 48 hours (Oktiana *et al.*, 2021).

The antibacterial activity test of liquid soap was carried out using the agar diffusion method with the paper disk technique. Next, a positive control was created using antibiotics (Ciprofloxacin) and a negative control using distilled water (Bawondes *et al.*, 2021). First, 1 mL of bacterial suspension was taken and distributed in 10 mL

NA media. The paper disk was inserted into each sample, and positive and negative control solutions were placed on the media, kept at 37°C upside down, and waited for 18, 24, 42 and 48 hours. The diameter of the clear zone between the paper disks was measured as a sample inhibition zone against bacteria (Syamsu *et al.*, 2015 in Syamsu *et al.*, 2022). Activity bacteria said weak if the diameter of the inhibition zone is <5 mm, medium is between 5-10 mm, the strong category is between 10-20 mm, and very strong if it is > 20 mm (Devi & Mulyani, 2017).

According to Toy *et al.* (2015), the diameter of the inhibition zone can be measured using the formula:

$$\text{Inhibition zone: } ((DV - DC) + (DH - DC))/2$$

Description: Zone of inhibition

DV: Vertical diameter

DH: Horizontal diameter

DC: Disc diameter

Is known that the diameter of the paper disc = 6 mm





Lastly, data processing and analysis. The parameters observed included color, aroma, texture, soap sample, positive control and negative control. Then, proceed with statistical analysis using variance analysis (ANOVA). If the ANOVA results show a real difference between treatments, a further test is carried out, namely the Duncan test. This research data was processed with the help of IBM SPSS Statistics 25 software. Then, the data that was not analyzed using statistical tests were the results of organoleptic tests, pH tests, homogeneity tests and foam stability tests.

### 3. RESULT AND DISCUSSION

#### Phytochemical Screening

Screening phytochemicals aims to identify group compounds in medium plants analyzed. Screening phytochemicals with the use of a method involving tube observation change color moment do the test with a certain reactor colour (Adjeng *et al.*, 2019). The results of screening phytochemicals in extracts of banana leaves are shown in Table 1.

Table 1. Phytochemical screening test results

Reagent	Change	Conclusion	Picture
Extract + reagent dragendorff	Orange/brickred precipitate	Alkaloid positive	
Extract + Mg powder + concentrated HCl	Colour red	Positive for flavonoids	
Extract + distilled water warm then shaken	A stable foam is formed	Saponin positive	
Extract + FeCl <sub>3</sub> 1%	Blackish green in color	Positive tannin	








Based on the results of phytochemical screening tests, it can be seen that banana leaf extract contains alkaloid, flavonoid, saponin and tannin phytochemical compounds. Alkaloids have an inhibitory mechanism that disrupts the peptidoglycan component in bacterial cells, disrupting the formation of cell wall layers and ultimately causing cell death (Juliantina, 2008 in Syinta, 2018). The base groups containing nitrogen in alkaloid compounds react with amino acid compounds, which are components of bacterial cell walls and DNA. This process can stimulate bacterial cell lysis, which results in cell death in the bacteria (Gunawan, 2009 in Syinta, 2018). In soap making, flavonoids are known to have benefits as antioxidants. It can also maintain healthy skin when applied in a soap formula (Dellima & Rosita, 2023).

Furthermore, there are also saponin compounds (foam producers), which are secondary metabolic compounds that function as antiseptics, so they have antibacterial capabilities, and the presence of these antibacterial substances can block the formation or transport of each component to the cell wall, which can later result in bacterial death (Effendi, 2018 in Yani, 2022). Tannin functions to inhibit bacterial growth. Alkaloid, flavonoid, saponin and tannin compounds can inhibit bacterial growth, so they can be used to make soap (Rahmawati *et al.*, 2023). So, if these phytochemical compounds are included in the soap formula, it will produce soap that contains antioxidant compounds and antibacterial compounds.

#### Organoleptic test

The organoleptic test aims to determine whether the quality of the liquid soap preparation is in accordance with the soap quality standards determined by SNI (Haque *et al.*, 2022).

Table 2. Organoleptic test results

Formulas	Test parameters	Repetition			Picture
		1	2	3	
<b>F0</b> <b>(0%</b> <b>Extract)</b>	Color	White	White	White	
	Aroma	Soap aroma	Soap aroma	Soap aroma	
	Texture	Liquid	Liquid	Liquid	
<b>F1</b> <b>(1%</b> <b>Extract)</b>	Color	Beige	Beige	Beige	
	Aroma	Rather strong essential oil	Rather strong essential oil	Rather strong essential oil	
	Texture	Liquid	Liquid	Liquid	
<b>F2</b> <b>(3%</b> <b>Extract)</b>	Color	Soft yellow	Soft yellow	Soft yellow	
	Aroma	Combination of extract and essential oil	Combination of extract and essential oil	Combination of extract and essential oil	
	Texture	Liquid	Liquid	Liquid	
<b>F3</b> <b>(5%</b> <b>Extract)</b>	Color	Yellow	Yellow	Yellow	
	Aroma	Rather strong extract aroma	Rather strong extract aroma	Rather strong extract aroma	
	Texture	Liquid	Liquid	Liquid	
<b>F4</b> <b>(7%</b> <b>Extract)</b>	Color	Deep yellow	Deep yellow	Deep yellow	
	Aroma	Strong extract aroma	Strong extract aroma	Strong extract aroma	
	Texture	Liquid	Liquid	Liquid	

Based on the research results, it was found that formula 3 was the best formula in terms of color, formula 1 was the best formula in terms of aroma and formulas 1 and 2 were the best formulas in terms of texture. This is because the higher the concentration of the extract given, the more intense the color and the stronger the aroma produced (Syarifah *et al.*, 2021). Based on the results obtained, the results of this organoleptic test have met the

standards set by the Indonesian National Standard (SNI), namely having a liquid texture, having a distinctive aroma and color (Anggraini *et al.*, 2021).

### Hedonic Test

The hedonic test is a favourite test for panellists, which is carried out using the organoleptic test method with a total of 10 panellists; each panellist is given a sample, namely banana leaf extract liquid soap, with the five formulas, namely F0, F1, F2, F3 and F4, three replications each. Panellists will be asked to fill out a questionnaire form regarding their preference for the color, aroma, and texture produced by banana leaf extract liquid soap (Haque *et al.*, 2022).

### Liquid soap color

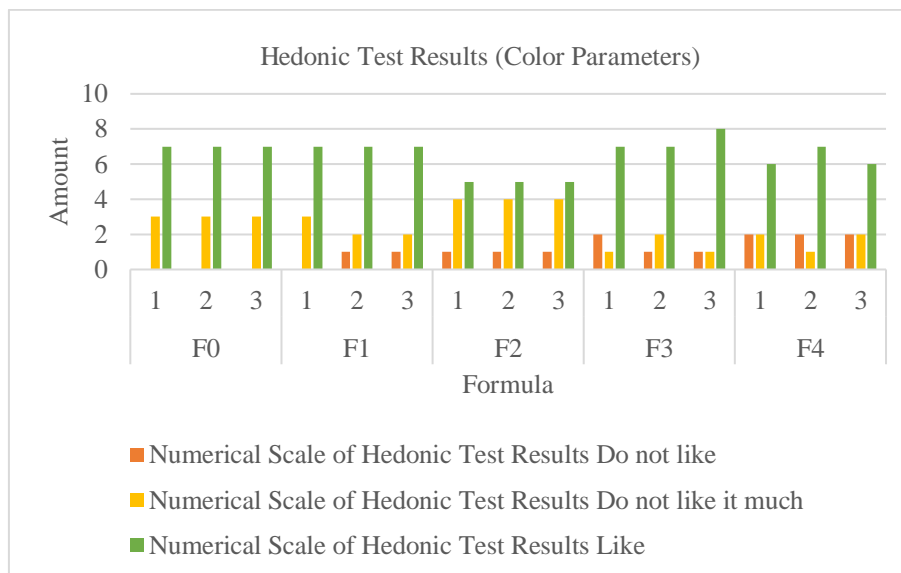


Figure 1. Hedonic test results (Color parameters)

Based on Figure 1, the results of the hedonic test for liquid soap color parameters, panellists preferred formula 3 because the color produced was not too light and also not too old or thick compared to formula 0, formula 1 and formula 2, which had a lighter color concentration level compared to formula 4 which has a deeper color concentration level. The addition of different extract concentrations causes this, the lower the extract concentration given, the lighter the color produced and the higher the extract concentration given, the darker or deeper the color produced by the statement of Handayani *et al.* (2023) states that the colour density difference at each concentration is caused by the number of active substances added to each formulation. The color change is due to the addition of the extract. The higher the concentration of the active substance, the more intense the color produced in the formula.

Next, to see the real differences between the five formulas, an ANOVA test was carried out. The following ANOVA test results are presented in Table 3.

Table 3. ANOVA table for numerical scale hedonic test color parameters

Frequency of respondents preferences for color parameters					
	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	277,333	14	19,810	127,347	0,000
Within Groups	4,667	30	0.156		
Total	282,000	44			

The results of statistical analysis using the ANOVA test obtained a significance value (p-value) of 0.000, which means  $< 0.05$ , this shows that the data above has a significant difference. Because there were quite significant differences, a post hoc test was carried out with Duncan's test to find out which treatments were different.

Table 4. DUNCAN hedonic test table for color parameters

Formulas	Numerical Scale			Average
	Do not like	Do not like it much	Like	
0	0.00 h	3.00 e	7.00 ab	3.33
1	0.67 i	2.33 f	7.00 ab	3.33
2	1.00 hi	4.00 d	5.00 c	3.33
3	1.33 ghi	1.33 ghi	7.33 a	3.33
4	2.00 fg	1.67 fgh	6.33 b	3.33
Average	1.00	2.47	6.53	(+)

From Duncan's test, it is known that there is a (+) sign, which means there is a real interaction between the five formulas (F0, F1, F2, F3, and F4) in the hedonic color parameter test. So, there is a real difference in the influence of the formula on the color produced by soap between treatments F0, F1, F2, F3, and F4. This means that the effect of the formula on the color produced by the soap between treatments F0, F1, F2, F3, and F4 has different values. Based on the results obtained, it can be said that the different formulas influence the colors produced in the five formulas (F0, F1, F2, F3, and F4).

**Liquid soap aroma**

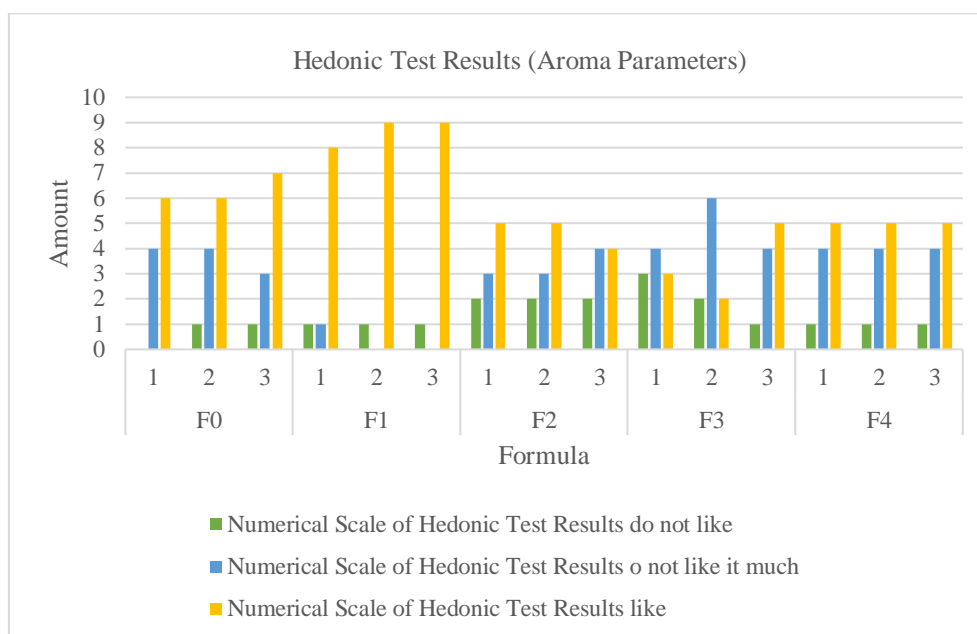


Figure 2. Hedonic test results (Aroma parameters)

Based on Figure 2. Hedonic test results for liquid soap aroma parameters, panellists prefer formula 1. This is because the aroma produced is not too strong because the extract added is not too much. The more extract given, the more pungent the resulting aroma will be (Syarifah *et al.*, 2021), so this causes the panellists to like it less. Next, to see significant differences between the five formulas, an ANOVA test was carried out. The following ANOVA test results are presented in Table 5.

Table 5. ANOVA Table of Numerical Scale for Hedonic Test of Aroma Parameters

Frequency of respondents preferences for aroma parameters					
	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	224,578	14	16,041	34,374	0,000
Within Groups	14,000	30	0,467		
Total	238,578	44			

The results of statistical analysis using the ANOVA test obtained a significance value (p-value) of 0.000, which means  $< 0.05$ , this shows that the data above has a significant difference. Because there were quite significant differences, a post hoc test was carried out with Duncan's test to find out which treatments were different.

Table 6. DUNCAN table for hedonic test of aroma parameters

Formulas	Numerical Scale			Average
	Do not like	Do not like it much	Like	
0	0.67 i	3.67 e	6.33 ab	3.56
1	1.00 hi	0.33 h	8.67 a	3.33
2	3.00 ghi	3.33 fg	4.67 c	3.33
3	2.00 fgh	4.67 b	3.33 f	3.33
4	1.00 hi	4.00 d	5.00 ab	3.33
Average	1.33	3.20	5.60	(+)

From Duncan's test, it is known that there is a (+) sign, which means there is a real interaction between the five formulas (F0, F1, F2, F3, and F4) in the hedonic aroma parameter test. So, there is a real difference in the influence of the formula on the aroma produced by soap between treatments F0, F1, F2, F3, and F4. This means that the effect of the formula on the aroma produced by the soap between treatments F0, F1, F2, F3, and F4 has different values. Based on the results obtained, it can be said that the different formulas influence the aroma produced in the five formulas (F0, F1, F2, F3, and F4).

**Liquid soap texture**

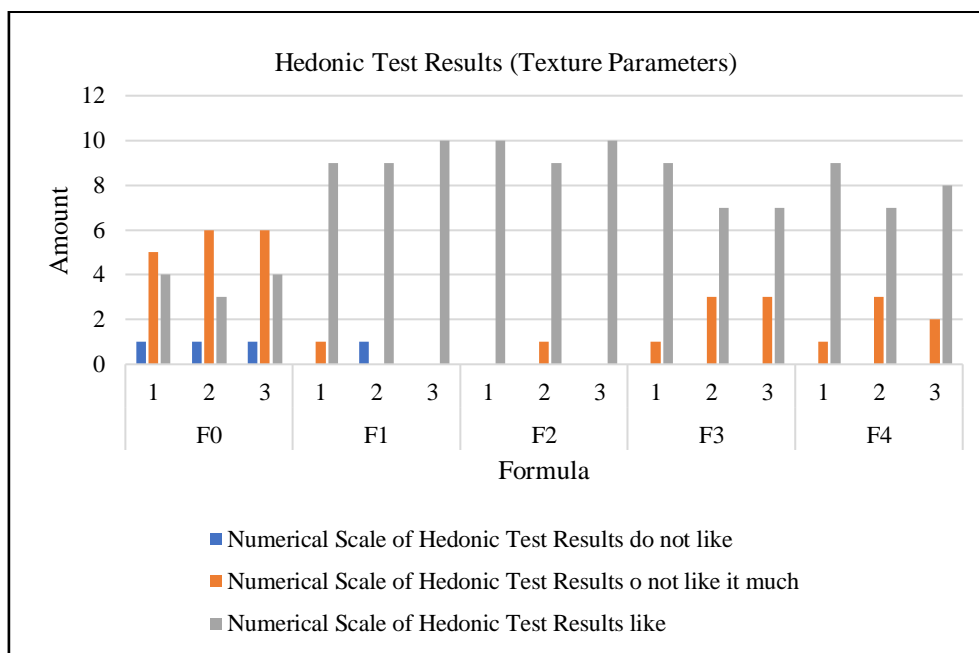


Figure 3. Hedonic test results (Texture parameters)

Based on Figure 3. Hedonic test results for liquid soap texture parameters, panellists preferred formula 1 and formula 2. This was influenced by the water content in the formula. The water content in the soap formula will affect the viscosity level of the soap, and the viscosity will decrease if the water/soap ratio increases because the viscosity is influenced by the water content in the soap. The less water content in the soap, the higher the viscosity of the soap, and the more water content in the soap, the lower the viscosity of the soap (Wijana *et al.*, 2009 in Nauli *et al.*, 2015).

Next, to see significant differences between the five formulas, an ANOVA test was carried out. The following ANOVA test results are presented in Table 7.



Table 7. ANOVA Table for Numerical Scale Test of Hedonic Texture Parameters

Frequency of respondent's preferences for aroma parameters					
	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	572,311	14	40,879	87,599	0,000
Within Groups	14,000	30	0.467		
Total	586,311	44			

The results of statistical analysis using the ANOVA test obtained a significance value (p-value) of 0.000, which means  $< 0.05$ , this shows that the data above has a significant difference. Because there were quite significant differences, a post hoc test was carried out with Duncan's test to find out which treatments were different.

Table 8. DUNCAN table for texture parameter hedonic test

Formulas	Numerical Scale			Average
	Do not like	Do not like it much	Like	
0	1.00 fg	5.67 c	3.67 d	3.44
1	0.33 ghi	0.33 ghi	9.33 ab	3.33
2	0.00 h	0.33 fgh	9.67 a	3.33
3	0.00 i	2.33 e	7.67 b	3.33
4	0.00 hi	2.00 f	8.00 ab	3.33
Average	0.27	2.13	7.67	(+)

From Duncan's test, it is known that there is a (+) sign, which means there is a real interaction between the five formulas (F0, F1, F2, F3, and F4) in the hedonic test of texture parameters. So, there is a real difference in the influence of the formula on the texture produced by soap between treatments F0, F1, F2, F3, and F4. This means that the effect of the formula on the texture produced by soap between treatments F0, F1, F2, F3, and F4 has different values. Based on the results obtained, it can be said that the different formulas influence the texture produced in the five formulas (F0, F1, F2, F3, and F4).

#### PH Test

PH is a parameter that describes the total concentration of hydrogen ions ( $H^+$ ) in a solution, indicating the level of acidity or alkalinity of a solution (Wibisono & Cahyono, 2022). The purpose of pH testing is to determine whether a formulation has a pH value that matches the skin's tolerance (Kharisma & Safitri, 2020).

Table 9. Results of measuring the pH value of the extract

Results of measuring the pH value of the extract			
Sample	Average	Result	pH standards
Extract	6.3	Acidic	Fulfil

Based on Table 9, it is known that the pH value of banana leaf extract is 6.3, which indicates that the pH of the extract is acidic. This is in accordance with the statement by Wibisono & Cahyono (2022) which states that the pH value ranges from 0 to 14. A solution that has a pH value of 0-6 is acidic while a pH of 8-14 is alkaline. Quantitative information regarding the degree of acidity or alkaline is reflected in the pH value, which is closely related to hydrogen ion activity. If the concentration of hydrogen ions ( $H^+$ ) exceeds the concentration of hydroxide ions ( $OH^-$ ), then it can be said that the solution is in the acidic category with a pH value below 7. On the other hand, if the concentration of ( $H^+$ ) is lower than ( $OH^-$ ), then the solution is classified as alkaline in nature with a pH value above 7.

PH testing is important in maintaining the quality of liquid soap. This is because liquid soap interacts directly with the skin and can cause problems if the pH does not match the skin's natural pH. The skin has the ability to tolerate and adapt quickly to products that have a pH range of 8.0 to 10.8 (Frost *et al.*, 1982 in Korompis *et al.*, 2020). The results of the soap pH test can be seen in Table 10 below.

Table 10. Soap pH test

Formulas	Soap pH Measurement Results				Result	PH standards
	Repetition			Average		
	1	2	3			
F0	13.5	13.5	13.5	13.5	Alkaline	Does not fulfil
F1	10	10	10	10	Alkaline	Fulfil
F2	9.9	9.9	9.9	9.9	Alkaline	Fulfil
F3	13.3	13.3	13.3	13.3	Alkaline	Does not fulfil
F4	13.3	13.3	13.3	13.3	Alkaline	Does not fulfil

The Indonesian National Standard (SNI) allows a pH range for liquid soap between 8 and 11 (Korompis *et al.*, 2020). The pH value of the soap solution depends on the type of fat used. Soap made from coconut oil usually has a pH value between 9 and 10 (Bidilah *et al.*, 2017). The research results showed that the pH value of liquid soap complies with SNI standards, namely F1 and F2. Meanwhile, at F0, F3 and F4 the resulting pH values do not meet SNI standards. The research results stated that the pH of the soap produced by the five formulas, namely F0, F1, F2, F3 and F4, was alkaline. This is because the pH value of the soap produced is in the range 8-14, which means the pH value is alkaline (Wibisono & Cahyono, 2022). The pH value tends to decrease with increasing water/soap ratio. This happens because the alkali used (Potassium hydroxide) reacts more completely with the fatty acids contained in the oil, so that the kalium hydroxide residue becomes lower and the soap no longer becomes too alkaline. The pH value of soap is influenced by the alkali content, the pH value will increase as alkalinity increases and will decrease as acidity increases, besides that the pH also decreases over time (Wijana, 2009 dalam Bidilah *et al.*, 2017).

#### Homogeneity test

Evaluation of liquid soap formulations aims to determine adequate physical stability in accordance with the standards set for liquid soap (Maharani *et al.*, 2021). The homogeneity test results can be seen in Table 11.

Table 11. Homogeneity test results

Formulas	Repetition	Homogeneity
F0	1	Homogeneous
	2	Homogeneous
	3	Homogeneous
F1	1	Homogeneous
	2	Homogeneous
	3	Homogeneous
F2	1	Homogeneous
	2	Homogeneous
	3	Homogeneous
F3	1	Homogeneous
	2	Homogeneous
	3	Homogeneous
F4	1	Homogeneous
	2	Homogeneous
	3	Homogeneous

Based on Table 11, it can be seen that the homogeneity test results of the five formulas, namely F0, F1, F2, F3, and F4, have homogeneous results, which are indicated by the absence of coarse particles or unmixed materials. This is to the statement by Kharisma & Safitri (2020), which states that testing can be carried out by seeing whether some coarse particles or materials are not mixed evenly and that no lumps form in the preparation so that it is declared homogeneous.

This means the five formulas, namely F0, F1, F2, F3, and F4 have homogeneous results. This is caused by factors that influence the soap making process, including the concentration of the alkali solution, temperature, stirring and time. Alkali that is too concentrated will cause the emulsion to break up in the solution so that the phase is not homogeneous, whereas if the alkali used is too dilute, it will take even longer. Therefore, the alkali concentration must be calculated based on reaction stoichiometry, where the addition of oil must be slightly excessive so that the resulting soap does not have an excessive free alkali value. Next is temperature; if viewed from a thermodynamic perspective, an increase in temperature will reduce the yield because the saponification reaction is exothermic, so an increase in temperature will reduce the balance constantly; however, if viewed from a kinetics perspective, an increase in temperature will cause an increase in the speed of the chemical reaction. Then, it is also influenced by the stirring speed because the stirring process aims to increase the probability of

interaction of the reactant molecules that react. Lastly, there is the effect of time; the longer the reaction time, the more oil will be saponified, so the results will also be higher (Hasibuan *et al.*, 2019).

#### Foam Stability Test

Data from the test results for the foam stability test of banana leaf extract liquid soap on the five formulas, namely F0, F1, F2, F3, and F4 in three are presented in Table 12 below.

Table 12. Foam stability test results

Formula	Repetition	Foam height initial (mm)	Foam height end (mm)	Stability (%)	Average value of Foam Stability	Results based on SNI requirements
F0	1	44	3	6.8	7.16	Does not fulfil
	2	38	3	7.8		Does not fulfil
	3	43	3	6.9		Does not fulfil
F1	1	45	40	88	81.7	Fulfil
	2	42	30	71.4		Fulfil
	3	42	36	85.7		Fulfil
F2	1	46	35	76	71.46	Fulfil
	2	52	31	59.6		Fulfil
	3	52	41	78.8		Fulfil
F3	1	58	44	75.8	74.7	Fulfil
	2	59	38	64.4		Fulfil
	3	50	42	84		Fulfil
F4	1	57	54	94.7	83.5	Fulfil
	2	52	45	86.5		Fulfil
	3	62	43	69.3		Fulfil

Based on the test results listed in Table 12, the test results that meet SNI are F1, F2, F3, and F4. This is because the stability of the foam produced by the four formulas is in the range of 13-220 mm. Meanwhile, at F0 the foam stability does not reach that figure, so it can be said that at F0 the foam stability value does not meet SNI standards. This is by the opinion of Haque (2022), who states that one of the advantages of liquid soap is its foam content. Based on SNI, the requirement for the height of foam from liquid soap is 13-220 mm and the stability of the foam for 5 minutes must remain 60-70%. Based on the test results show that the high stability of the foam increases along with increasing extract concentration. Increasing the extract concentration in liquid soap preparations correlates with increasing the foam stability of banana leaf extract. This is caused by the saponin content in banana leaves.

#### Antibacterial Test

Data on the results of antibacterial testing of banana leaf extract in the five formulas, namely F0, F1, F2, F3, and F4 in three replications are presented in Table 13 below.

Table 13. Antibacterial Test Results Banana Leaf Extract

Hourly observations	Inhibition zone diameter		
	Sample	Control (+)	Control (-)
0	0mm	0mm	0mm
18	1mm	51mm	0mm
24	1mm	51mm	0mm
42	2mm	51mm	0mm
48	2mm	51mm	0mm

Based on Table 13, the research results state that the diameter of the inhibition zone begins to form at the 28<sup>th</sup> hour, namely 1 mm and increases at the 42<sup>nd</sup> hour to 2 mm, which means that the extract has a weak ability to inhibit the growth of *Escherichia coli* (Devi & Mulyani, 2017). According to research conducted by Bell in 1984, a material is said to have antibacterial activity if the diameter of the barrier formed is greater than or equal to 6 mm (Toy *et al.*, 2015). However, banana leaves contain phytochemical compounds, including alkaloids, flavonoids, saponins and tannins, which are believed to be able to inhibit the growth of bacteria so they can be used as ingredients for making soap (Rahmawati *et al.*, 2023).

Next, an antibacterial test was carried out on the liquid soap formula on the five formulas, namely F0, F1, F2, F3, and F4, with 3 replications each; the data is presented in Table 14 below.

Table 14. Liquid Soap Antibacterial Test Results

Formulas	Repetition	1		2			3			
		Hour	Sample	(+)	(-)	Sample	(+)	(-)	Sample	(+)
F0	0	0	0	0	0	0	0	0	0	0
	18	11	33,5	0	14,5	38,5	0	19	37	0
	24	12	42	0	16	36	0	19,5	38	0
	42	13	38	0	18,5	38	0	20	39	0
	48	14	39	0	20	40	0	21	40	0
F1	0	0	0	0	0	0	0	0	0	0
	18	12	38.5	0	15	58	0	19.5	34.5	0
	24	12.5	39.5	0	16.5	58	0	21	36	0
	42	13.5	41.5	0	19	58.5	0	21.5	38	0
	48	14.5	39.5	0	20.5	59	0	22	38.5	0
F2	0	0	0	0	0	0	0	0	0	0
	18	7	36	0	14.5	44.5	0	51.5	39	0
	24	9	39	0	14.5	46	0	52	42	0
	42	10	40	0	16	48	0	52.5	46	0
	48	11	41	0	17	49	0	53	49	0
F3	0	0	0	0	0	0	0	0	0	0
	18	18	52.5	0	26	39.5	0	19.5	34.5	0
	24	19	53.5	0	27	40.5	0	21	36	0
	42	28	54.5	0	33	44	0	21.5	36	0
	48	30.5	55	0	35	45.5	0	22	36	0
F4	0	0	0	0	0	0	0	0	0	0
	18	19.5	43	0	43	31.5	0	38.5	33.5	0
	24	20	45.5	0	44	33	0	39	35	0
	42	22	51.5	0	45	39	0	43	48.5	0
	48	23	53	0	45	41	0	45.5	50.5	0

Based on Table 14, the research results state that F4 has the best inhibition zone value compared to other formulas. Although the research results state that banana leaf extract has a weak ability to inhibit *Escherichia coli* growth when formulated into soap, it will produce soap with a medium to very strong ability to inhibit the growth of *Escherichia coli*, as shown in Table 14. The higher the formulation, the higher the ability to inhibit the growth of *Escherichia coli*. This means that the more extracts added to the soap formulation, the stronger the soap will inhibit the growth of *Escherichia coli*. This is the statement of Suharyanisa et al. (2021), which states that a clear zone is formed around the disc because it contains secondary metabolite compounds that play an important role in antibacterial activity. The inhibition zone formed in the five formulas shows an increase in the clear zone of each formula. The greater the concentration of the extract added, the greater the inhibition zone of the active substance formed.

Next, an ANOVA test was carried out to see significant differences between the five formulas. The following ANOVA test results are presented in Table 15.

Table 15. ANOVA test result

Dependent Variable: Diameter of the Inhibition Zone					
Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Model	37756.500a	25	1.510.260	12.425	.000
Formula	2.300.413	4	575.103	4.731	.003
Hour	7.111.780	4	1.777.945	14.627	.000

Dependent Variable: Diameter of the Inhibition Zone					
Formula * Hour	619.453	16	38.716	.319	.993
Error	6.077.500	50	121.550		
Total	43834,000	75			

Based on Table 15, the results of statistical analysis using the ANOVA test obtained a significance value (p-value) of 0.993, which means  $> 0.05$ , this shows that the data above does not have a significant difference. Because there is no significant difference, there is no need to carry out further tests. This means that the results obtained from the five formulas (F0, F1, F2, F3, and F4) do not have significant differences because the diameter of the inhibition zone does not increase significantly from each observation time, namely hours 0, 18, 24, 42, and 48.

#### 4. CONCLUSION

Based on research results show that banana leaf extract is rich in useful phytochemical compounds, such as alkaloids, flavonoids, saponins and tannins. Integration of these compounds into soap formulas produces products that have antioxidant and antibacterial properties. Although the ability of banana leaf extract to inhibit *Escherichia coli* is considered weak, soap formulations with this extract show a significant increase in antibacterial ability, especially in formula 4 with an extract concentration of 7%. Therefore, formula 4 can be considered the best choice for making liquid soap with banana leaf extract. Because it not only provides optimal results in inhibition zone testing but also meets SNI standards for foam stability.

#### 5. ACKNOWLEDGEMENT

The authors to Biotechnology Study Program, Universitas Aisyiyah Yogyakarta.

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