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Formulation of babadotan (*Ageratum conyzoides*) leaf extract natural liquid soap

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Abstract. Babadotan leaf has an antibacterial agent which able to protect the skin from types of infectious diseases. The antibacterial agent is potential to be developed as an antibacterial agent in soap preparation also potential replaced the SLS soap. The purpose of this study is to determine the best concentration extract of babadotan on liquid soap based SNI 06-4085- 1996. The concentration treatment of babadotan leaf extract consisted of 9 levels (0-40%). Observations made included the degree of acidity (pH), specific gravity, foam stability, total plate count, sensory test consisting of a scoring test and a hedonic test. Then the data were analyzed using the Least Significant Difference (LSD) test at the 5% level. The result is the best babadotan extract liquid bath soap at an extract concentration of 25%. Babadotan extract liquid soap produces the best characteristics with a score color 3.32 (typical babadotan), aroma 3.67 (fresh), texture 2.95 (less viscous), foam 3.32 (like), and overall acceptability 3.52 (like). The 25% extract treatment resulted in a pH value of 6.59, a specific gravity is 1.03 and a total plate number of 3.4×10^4 . The stability of the resulting foam is at 94.35%, antibacterial inhibition formed was 18.41 mm.

1. Introduction

Antibacterial soaps on the market usually use synthetic surfactants such as Sodium Lauryl Sulfate (SLS) and triclosan which have negative effects on human skin and the environment. Using SLS surfactants in their daily activities produce waste containing SLS surfactants then go to the environment. The presence of SLS in aquatic environments can be disturbing ecosystems such as the resulting foam can reduce dissolved oxygen concentrations and can interfere [1].

Using the synthetic surfactants has a negative impact for humans is irritating to humans' skin. Also, SLS can attract more than just dirt. Natural oils on the skin will also be absorbed and thrown away. This is one of the main causes of irritation or dryness of the skin. Usage Surfactants and antibacterial in soap preparations can be replaced with natural ingredients so they are safe for humans and the environment. Babadotan is one of the plants that can use as antibacterial agent. Babadotan leaves have active ingredients such as alkaloids, saponins, polyphenols, essential oils, and flavonoids. Babadotan leaf has bioactivity as an antibacterial and can be used as an inhibitor microorganism [2]. Babadotan extract has antibacterial activity against *Staphylococcus aureus*, *Bacillus subtilis*, *E. coli*, and *Pseudomonas aerogenase*.



Babadotan leaves have a very potential function to improve the quality of liquid body soap. This soap can replace the SLS soap and finally the used of SLS can be reduced. However, there is no research that discusses the proper formulation for making liquid soap babadotan extract. Soap making in this study did not use Sodium Lauryl Sulfate but using antibacterial agents in the form of natural extracts so that this soap product is expected to be able to minimize skin irritation, provides added value to babadotan weeds and is friendly environment. Therefore, it is necessary to carry out further research and observations related to soap formulations from babadotan extract which is safe and conforms to SNI for liquid bath soap, namely SNI 06-4085-1996.

2. Materials and methods

The materials used in this study were babadotan leaf extract, decyl glucoside (natural surfactant), cocamidopropyl betaine (surfactant), lelgard (preservative), vegetable glycerin (humectant), cosmetics grade fragrance, guar gum (gelling agent), xanthan gum (gelling agent), citric acid, aquades, acetone, diethyl ether, cool water, alcohol 70%, media PCA, media NA, mueller hinton agar, filter paper, and *Staphylococcus aureus*. The tools used in this study were Erlenmeyer, beaker glasses, food dehydrator, digital scales, micropipette, dropper pipette, pH meter, leaf color chart (BWD), petri dish, finn tip, hotplate, pycnometer.

The sampling technique is making babadotan leaf extract, making liquid soap from babadotan leaf extract was prepared using a single factor in a Completely Randomized Block Design (RAKL) with two repetitions. The factor to be used is the concentration of babadotan leaf extract. Data were analyzed using ANOVA with the least significant difference (LSD) at the 5% level of significance. The concentration treatment of the babadotan leaf extract is 0%, 5%, 10%, 15%, 20%, 25%, 30%, 35% and 40%.

2.1. Production of babadotan leaf extract liquid soap

The formulation of the babadotan leaf extract liquid soap was carried out with 9 variations of the concentration of the babadotan leaf extract. The formulation for making liquid soap from babadotan leaf extract can be presented in Table 1.

Table 1. Formulation of babadotan leaf extract liquid soap.

Material	Unit	Concentration								
		P0	P1	P2	P3	P4	P5	P6	P7	P8
Babadotan leaf extract	ml	0	5	10	15	20	25	30	35	40
Lexgard	g	1	1	1	1	1	1	1	1	1
Fragrance	ml	0.35	0.35	0.35	0.35	0.35	0.35	0.35	0.35	0.35
Vegetable Glycerin	ml	5	5	5	5	5	5	5	5	5
Xanthan Gum	g	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3
Guar Gum	g	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15
Decyl Glucoside	ml	15	15	15	15	15	15	15	15	15
Cocamidopropyl Betaine	ml	8	8	8	8	8	8	8	8	8
Citric Acid	g	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3
		Aquades (mL) added up to 100 mL								

The first step in making this soap is starting with all the ingredients being weighed according to Table 1 above. Next, mix babadotan leaf extract with lelgard and fragrance. After that, mix vegetable glycerin, xanthan gum and guar gum. Mix until homogenous. Then, add decyl glucoside, cocamidopropyl. Mix it again. After that, add distillate water until the volume is 100 ml and add citric acid as pH regulator. Next, the sample would be tested such as sensory test, pH test, specific gravity test, total plate count (TPC) and antibacterial activity test.

3. Results and discussion

3.1. Sensory test

3.1.1. *Scoring color*. The Table 2 descript scoring color test between babadotan leaf extract liquid soap with different extract concentration.

Table 2. LSD test result (0.05) scoring color.

Babadotan Leaf Extract Concentration	Average Color Scoring
P1 (0 %)	1,3250 ^a
P2 (5%)	1,7000 ^{ab}
P3 (10%)	2,0250 ^b
P4 (15%)	2,6500 ^c
P5 (20%)	2,9250 ^c
P6 (25%)	3,3250 ^d
P7 (30%)	3,6250 ^d
P8 (35%)	4,1750 ^e
P9 (40%)	4,4500 ^e

Color is the first sensory property observed when consumers see industrial products. The liquid soap preparation of babadotan leaf extract has a dark brown, light brown to brownish yellow color. This color comes from the activity of the polyphenol oxidase enzyme due to heating of the leaves before extraction. In addition, during the extraction process, the tannins contained in the babadotan leaves are released. Tannins are known as brown or brown coloring agents [3].

3.1.2 *Scoring scent*. The Table 3 descript scoring scent test between babadotan leaf extract liquid soap with different extract concentration.

Table 3. LSD test result (0.05) scoring scent.

Babadotan Leaf Extract Concentration	Average Scent Scoring
P1 (0%)	2.9000 ^a
P4 (15%)	2.9000 ^a
P2 (5%)	2.9750 ^a
P3 (10%)	2.9750 ^a
P5 (20%)	3.3000 ^{ab}
P7 (30%)	3.6500 ^{bc}
P6 (25%)	3.6750 ^{bc}
P8 (35%)	4.0500 ^{bcd}
P9 (40%)	4.1750 ^d

The concentration of 0% babadotan leaf extract makes the fragrance smell less strong. This is because the aroma of the fragrance fades slightly and is covered by the aroma of the surfactant reaction. The surfactant used is a natural surfactant derived from the synthesis of natural ingredients, namely coconut oil. Therefore, surfactants have an unpleasant odor. Mixing the 2 types of ionic and non-ionic surfactants causes a slight unpleasant odor. The fatty acid content in nonionic surfactants (decyl glucoside) makes the green tea fragrance fade and the preparation smells a little rancid and less fresh.

As the concentration of the extract increases, the aroma of the green tea fragrance gets stronger. This is due to the fresh aroma in green tea fragrance due to the content of phenolic and tannin compounds in it [4]. The addition of babadotan leaf extract increased the content of phenolics and tannins in the preparation. Even though the aroma produced was slightly unpleasant, the panelists preferred the aroma of soap added with babadotan leaf extract. Choosing the right fragrance also makes the extract and fragrance harmonize and produce a fresh scent. The distinctive fresh aroma of the leaves in the green tea fragrance disguises the unpleasant aroma of the extract.

3.1.3. *Scoring texture.* The Table 4 descript scoring texture test between babadotan leaf extract liquid soap with different extract concentration.

Table 4. LSD test result (0.05) scoring texture.

Babadotan Leaf Extract Concentration	Average texture Scoring
P2 (5%)	2.5250 ^a
P3 (10%)	2.7000 ^{ab}
P4 (15%)	2.7000 ^{ab}
P5 (20%)	2.7500 ^{abc}
P1 (0%)	2.8000 ^{abc}
P6 (25%)	2.9500 ^{bcd}
P7 (30%)	3.0500 ^{cde}
P8 (35%)	3.2750 ^{de}
P9 (40%)	3.3750 ^e

Babadotan liquid extract is thicker than distilled water. This is what causes the more the extract, the thicker the resulting texture. The water content that is bound by the gelling agent is less, so that the soap preparation with a higher extract content will be slightly thicker. In addition, the addition of extracts will produce a more concentrated preparation so that it looks as if it is thicker than preparations without added extracts.

3.1.4. *Hedonic foam.* The Table 5 descript hedonic foam test between babadotan leaf extract liquid soap with different extract concentration.

Table 5. LSD test result (0.05) hedonic foam.

Babadotan Leaf Extract Concentration	Average Foam Hedonic
P1 (0%)	2.8750 ^a
P2 (5%)	2.9250 ^{ab}
P3 (10%)	2.9500 ^{ab}
P4 (15%)	3.1250 ^{ab}
P5 (20%)	3.1750 ^{bc}
P6 (25%)	3.3250 ^{cd}
P7 (30%)	3.5000 ^d
P8 (35%)	3.5000 ^d
P9 (40%)	3.7750 ^e

Decyl glucoside is one of the surfactants used in the preparation of babadotan leaf extract liquid soap. The surfactant is a type of natural surfactant and is hydrophilic. This surfactant has less foam than other surfactants. To overcome this problem, in this study another type of surfactant was added, namely cocamidopropyl betaine and the addition of an extract containing saponins. This is so that more foam is

produced. According to [5] saponins are natural foaming agents and have the potential to become natural surfactants.

Cocoamidopropyl betaine used in this study is known to be compatible with other anionic, cationic and nonionic surfactants. In addition, this surfactant has a very low potential for eye and skin irritation in animal safety tests. Cocoamidopropyl betaine is an amphoteric surfactant with good foaming properties and can make the skin feel soft [6] Based on this formulation, the resulting soap gives a lot of foam and is smooth in the hands. The more addition of extract concentration, the more foam produced.

3.1.5. Hedonic overall acceptance. The Table 6 descript hedonic overall acceptance test between babadotan leaf extract liquid soap with different extract concentration.

Table 6. LSD test result (0.05) hedonic overall acceptance.

Babadotan Leaf Extract Concentration	Average Overall Acceptance Hedonic
P1 (0%)	2.8250 ^a
P2 (5%)	2.9750 ^{ab}
P3 (10%)	3.0000 ^{ab}
P4 (15%)	3.1750 ^{abc}
P5 (20%)	3.3000 ^{bcd}
P6 (25%)	3.5250 ^{cde}
P7 (30%)	3.5250 ^{cde}
P8 (35%)	3.5500 ^e
P9 (40%)	3.7500 ^e

The overall acceptance score ranges from 2.8250 – 3.7500 (less likes and likes). The panelists did not like the P1 treatment because the scoring test for the P1 treatment (control) gave a white color and less fresh aroma. Panelists preferred P9 soap with an extract concentration of 40% with a blackish brown color, fresh aroma and thicker texture. In terms of color, soap with an extract concentration of 40% has a blackish brown color, which makes the soap look thick and thick. The refreshing aroma of the soap and the more foam also made the panelists prefer babadotan extract liquid soap with an extract concentration of 40%.

3.2. pH test

The results of the analysis of variance showed that the concentration of babadotan leaf extract at ($\alpha = 0.05$) was not gives a significant effect on the pH value of liquid body soap. Based on the results of observations that have been the pH of babadotan leaf extract liquid soap ranged from 6.11 to 6.92. Leaf extract concentration babadotan had no significant effect on the resulting pH value. The lowest pH was in the P2 treatment (concentration of 5% babadotan leaf extract) with a pH value of 6.11, while the highest pH value was in P9 treatment (40% concentration of babaotan leaf extract) with a pH value of 6.92. pH increases after adding the extract because the babadotan leaf extract has a fairly high pH, which is equal to 7.18. This is because the extract contains pheophytin obtained from the degradation process of chlorophyll [7].

3.3. Specific gravity

The results of the analysis of variance showed that the concentration of babadotan leaf extract at ($\alpha = 0.05$) was not gives a significant effect on the value of the specific gravity of liquid body soap. Based on the results of the observations has been carried out, the results obtained that the specific gravity of babadotan leaf extract liquid soap ranged from 1.0253 – 1.0352. The concentration of babadotan leaf extract had no significant effect on the resulting specific gravity. Most specific gravity low was in treatment P1 (concentration of babadotan leaf extract 0%) with a specific gravity value of 1.0253, while the highest specific gravity was in treatment P6 (25% concentration of babadotan leaf extract) with a

specific gravity of 1.0352. The addition of extract concentration with the specific gravity of liquid soap. The addition of babadotan leaf extract will make the value of the specific gravity of liquid soap even greater.

3.4. Foam stability

The results of the analysis of variance showed that the concentration of babadotan leaf extract at ($\alpha = 0.05$) was not gives a significant effect on the value of the foam stability of liquid body soap. Based on the results of the observations has been carried out, the stability of babadotan leaf extract liquid soap foam ranges from 93.0250 - 94.6550%. The concentration of babadotan leaf extract had no significant effect on the foam stability. The lowest foam stability was in treatment P1 (0% concentration of babadotan leaf extract) with foam stability of 93.0250%, while the highest foam stability was in treatment P3 (10% concentration of babadotan leaf extract) with foam stability of 94.6550%. . However, all concentrations had very stable foam stability percentages. According to [8] good foam stability should remain between 60 – 70%.

Foam stability be affected because of Saponin. Saponin's ability as a natural foaming agent cannot be separated from its hydrophilic and hydrophobic groups. The properties of saponin are similar to the properties of surfactants and are able to substitute for the use of surfactants. The combination of structures that make up saponin, in the form of nonpolar sapogenin fragments and polar side chains that are soluble in water [9].

The combination 2 natural surfactants also causes the stability of foam is good. Decyl glucoside is one of the non ionic surfactant and *cocamidopropyl betaine* is amphoteric surfactant. Mixing 2 types of surfactant can boost the foam and made the foam is more stable.

3.5. Total Plate Count (TPC)

The LSD test results showed that the concentration of the babadotan leaf extract had a significant effect on total plate count liquid soap. The results of the 5% LSD follow-up test at the concentration of babadotan leaf extract can be seen in Table 7.

Table 7. LSD test results (0.05) Total Plate Count test.

Babadotan Leaf Extract Concentration	Total Microbe (log x) (cfu/ml)
P3 (10%)	3.6700 ^a
P1 (0%)	4.0529 ^b
P2 (5%)	4.1689 ^{bc}
P7 (30%)	4.2960 ^c
P9 (40%)	4.3471 ^c
P4 (15%)	4.3701 ^c
P8 (35%)	4.4618 ^c
P6 (25%)	4.5194 ^c
P5 (20%)	4.5273 ^c

Based on the results of observations that have been made, the babadotan extract liquid soap preparation has total Microbial contamination is below the threshold set by SNI 06-4085-1996. That limit stipulated by SNI is a maximum of 1.0×10^5 . The presence of active ingredients such as saponins, tannins, and flavonoid in babadotan leaf extract is able to fight microorganisms that grow. In addition to the use of preservatives natural lesgard or preservatives also help reduce microbial growth in liquid soap. Lexgard Natural is an anti-microbial substance that contains two components, namely Glyceryl Caprylate and Glyceryl Undecylenate and works by damaging membranes and destroying microbial cells [10].

3.6. Best concentration

Determination of the best concentration in this study was based on the results of testing all observed parameters, namely pH value, specific gravity, foam stability, total plate count, and sensory testing (color, aroma, texture, foam and overall acceptance). The expected values are pH between 6 – 8, foam stability is more than 70%, specific gravity and total plate count are in accordance with SNI 06-4085-1996 and sensory data with the highest score. The Table 8 is a table for determining the best concentration for babadotan leaf extract liquid bath soap.

Table 8. Determination best concentration of babadotan leaf extract.

Parameter	Concentration									SNI Requirement
	P1	P2	P3	P4	P5	P6	P7	P8	P9	
Sensory										
Color	1.32	1.70	2.02	2.65	2.92	3.32*	3.62	4.17*	4.45	Homogenous
Scent	2.90	2.97	2.97	2.90	3.30*	3.67	3.65	4.05*	4.17	
Texture	2.80	2.52	2.70	2.70	2.75	2.95	3.05*	3.27	3.37	
Foam	2.87	2.92	2.95	3.12	3.17*	3.32*	3.50	3.50	3.77*	
Overall Acceptance	2.82	2.97	3.00*	3.17	3.30	3.52*	3.52	3.55	3.75	
pH	6.41*	6.11*	6.31*	6.43*	6.65*	6.59*	6.44*	6.66*	6.91*	6 – 8
Specific Gravity	1.03*	1.03*	1.03*	1.03*	1.03*	1.03*	1.03*	1.03*	1.03*	1.01 – 1.10
Foam Stability	93.0*	93.5*	94.6*	93.7*	94.5*	94.3*	93.3*	93.3*	93.8*	-
Total Plate Count	4.05*	4.17*	3.67*	4.35*	4.53*	4.52*	4.30*	4.47*	4.37*	Max 1.0×10^5
Best Concentration	P6 (Concentration of babadotan leaf extract 25%)									

3.7. Antibacterial activity

Based on the results of observations of the inhibition zone formed by 18.41 mm (very strong) around the paper disc describing the antibacterial sensitivity of babadotan leaf extract in inhibiting growth of *Staphylococcus aureus*. The antibacterial inhibition zone formed is due to the content Chemical compounds of the flavonoid group which are secondary metabolite compounds that have antibacterial properties [11] The way phenol works in killing microorganisms is by: denatures cell proteins. The liquid bath soap inhibition zone against *Staphylococcus aureus* bacteria can be seen in Figure 1.

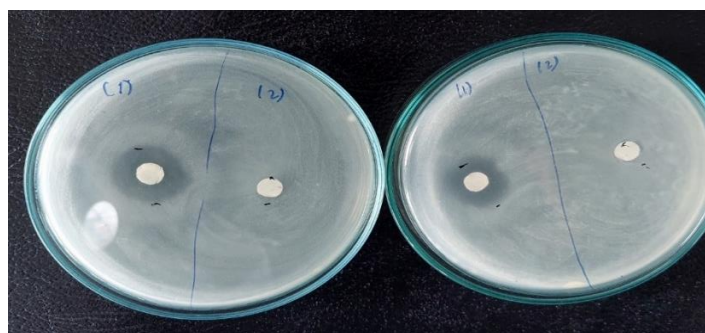


Figure 1. *Staphylococcus aureus* inhibition zone on Mueller Hinton Agar (MHA) media.

Terpenoid compounds react with porins on the outer membrane of the bacterial cell wall, forming polymeric bonds strong enough to damage the porin. Porin damage which is the entrance and exit the compound will reduce the permeability of the bacterial cell wall resulting in a deficiency of bacterial cells nutrients, so that the growth of bacteria is inhibited or killed. Saponins have an aglycone group that plays a role as an antibacterial. The mechanism of action of saponins can change cell permeability and interfere permeability of the bacterial cell membrane. Gram positive bacteria such as *S. aureus* do not have such an outer membrane possessed by most gram-negative bacteria. Besides that, *S. aureus* has a cell wall that is hydrophobic on the outside, so that low concentrations of the extract will be able to kill all bacteria *S. aureus* bacteria [12].

Alkaloid compounds have a mechanism of action as an antibacterial, namely by interfering peptidoglycan constituent components in bacterial cells so that the cell wall layer is not formed completely and causes cell death. *S. aureus* has a thicker peptidoglycan layer without presence of an outer membrane. The activity of phenolic compounds will easily cause damage to cell walls and membranes thereby interfering with the active transport system. Next is the compound will denature and inactivate proteins such as enzymes thereby affecting cell metabolism [12].

Flavonoid compounds act as an antibacterial agent by inhibiting synthesis of nucleic acid, also inhibiting the function of the cytoplasmic membrane and inhibiting the energy of metabolism of bacteria. Because of a lot of ingredients which can inhibit the bacteria, the babadotan liquid extract soap has a strong inhibitory.

4. Conclusion

The conclusion of this study was that the best concentration of babadotan liquid soap was selected extract concentration of 25%. The best babadotan extract liquid soap has the following characteristics: color score 3.3250 (typical babadotan), aroma score 3.6750 (fresh), texture score 2.950 (less thick), foam score 3.3200 (likes) and overall acceptance of 3.5250 (likes). Treatment of 25% extract resulted in a pH value of 6.5950, weight type 1.0352 and total plate number 3.4×10^4 which is in accordance with SNI 06-4085-1996. foam stability produced is at 94.35% which means the foam is stable. Antibacterial inhibition formed is equal to 18.41 mm which belongs to the very strong category. The result shows that the babadotan leaf extract potential for replaced the SLS soap and can be a product which safe for human and environment.

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