FORMULATION AND ANTIBACTERIAL TEST TRAVELING PAPER SOAP MADE FROM VIRGIN COCONUT OIL AND DURIAN SEEDS

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Abstract: Soap as a cleaning product has been the primary needs since the spread of Covid-19. Government regulations require people to wash their hands. Hence the demand for soap production is increasing. This study aimed to obtain a Traveling Paper Soap formulation from VCO and durian seeds for antibacterial activity on the skin. The experimental laboratory method uses quantitative and qualitative descriptive analysis. The treatment in this study was the production of Durian Seed Bioethanol with a bioethanol capacity of 19.95% from the hydrolysis of 486 g of Durian Seed flour. The treatment in this study was the manufacture of durian seed bioethanol with a bioethanol content of 19.95% from the hydrolysis of 486 g of durian seed flour. The parameters of the bioethanol viscosity test were 0.7137 cP, and the bioethanol density was 0.9669 g/mL. Based on our research, the best traveling paper soap results are formulation 2 with a water content of 7%, pH value of 10, free alkali content of 0.21%, and antibacterial activity against Staphylococcus Epidermis of 22.3 mm. According to the Indonesian National Standard Mandatory Solid Soap, SNI 3532:1994, it shows that soap complies with the SNI 3532:1994 standard.

Keywords: Soap, VCO, Durian Seeds, Antibacterial, S. aureus, S. epidermis

INTRODUCTION

Soap is one of the main needs used as a cleaning product with water media [1]. The increase in demand for soap production has been increasing since the spread of a new outbreak, namely Covid-19, which was first discovered in the city of Wuhan, China, at the end of December 2019 with a very fast transmission rate [2]. To avoid spreading the virus, the government implements health protocols that must be obeyed by the public, namely using masks, using hand sanitizer, washing hands, and cleaning the body using soap when traveling or leaving the house [3]. Washing hands with soap is more effective at killing germs and viruses than using a hand sanitizer [4]. Regular use of soap can make the skin texture rougher and drier because the soap's working system attracts natural fats on the skin's surface in excess [5]. Therefore, adding a natural additive formula containing vitamin E, probiotics, lauric acid (HC12H23O2), and others are necessary to improve skin health and protect skin from irritation. One of the natural ingredients that can overcome this problem is Virgin Coconut Oil (VCO) [6].

VCO is made from fresh coconut milk by extracting oil or kernels through a fermentation method without heating without chemicals (7). VCO contains protein, lauric acid, enzymes, omega-3, probiotics Lactobacillus fermentum, and Saccharomyces cerevisiae as antimicrobials against pathogenic bacteria Staphylococcus aureus and Escherichia coli, and vitamin E as anti-inflammatory [8]. Based on Armita's research, 2014 VCO contains monolaurin, which has an antibacterial effect against epidermal staphylococcus. The manufacture of antibacterial and antiviral soaps also requires natural ingredients like bioethanol, the main ingredient—ethanol extract against epidermal staphylococcus [9]. The manufacture of antibacterial and antiviral soaps also requires natural ingredients in the form of the main bioethanol ingredients. Methanol extract of Montong durian fruit seeds (Durio zibethinus murray) can be used as an antibacterial against Staphylococcus aureus [9]. Bioethanol is made from starch or cellulose biomass, the main food ingredient [10], so if used on a large scale, it can interfere with human food needs. Therefore, we need a starch biomass source not used as food, one of which is durian seeds. The abundance of durian seed waste has not been widely used [11], containing 54.14% starch. It has succeeded in making bioethanol from durian seeds with optimum conditions of fermentation pH = 4, producing bioethanol levels of 40%-47.02% and IC50 vitamin C values of 3.76 μg/mL [12].

Using VCO with a combination of Durian Seeds can provide innovations in manufacturing antibacterial and antioxidant soap. Therefore, using the Saponification method, it is necessary to research the formulation and effectiveness of soap made from VCO and durian seeds with the SNI standard. The soap formula test for antibacterial activity was carried out using the cram diffusion method using Staphylococcus aureus and Staphylococcus epidermis bacteria. This research innovates to print.

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paper soap to make it more practical and attractive. Therefore, this study focused on determining the formulation and activity test of Traveling Paper Soap antibacterial made from VCO and Durian Seed Bioethanol following Indonesian National Standard.

RESEARCH METHODS
Equipment and Materials
This research uses supporting equipment: distillation set; petri dish, silicon mold, Erlenmeyer (pyrex), watch glass, beaker, measuring cup, hot plate, paper Disk, crucible, dropper, stainless steel blade, shoeelace, pH meter, swab, thermometer 100°C, weighing scale analytical, mortar and pestle.

The materials used in this study were distilled water, acetone, tissue, sarin paper, sulfuric acid (H2SO4) 2.5%, sulfate (H2SO4) 0.3 M, stearic acid, amoxicillin, seeds durian, fengling A and B, glycerin, standard glucose, sodium hydroxide (NaOH) 30%, Nutrient agar, KH2PO4, Saccharomyces cerevisiae, Virgin Coconut Oil (VCO), and urea (CO(NH2)2). The test bacteria used were Staphylococcus aureus and Staphylococcus epidermis.

Making Durian Seed Bioethanol
The first step in making durian seed bioethanol is washing the durian seeds thoroughly and peeling the skin of the durian seeds. The samples were cut into smaller sizes and air-dried to reduce the moisture content. Durian seed flour was obtained after mashing dry samples of durian seeds using a blender. The hydrolysis process was carried out by mixing durian seed flour and 0.3 M H2SO4 (1:10). The mixture was stirred for 30 minutes, heated at 100°C using a water bath, and refluxed for 1-1.5 hours. The test of glucose levels on the hydrolysis results was carried out by the Fehling [13] and Molisch [14] methods.

The result of hydrolysis is then fermented. Fermentation was carried out by adding 9 grams of tape yeast into 54 mL of the hydrolyzed solution by adjusting the pH using NaOH in the range of 4.5-5. Aquades (1 L) were added after entering the nutrients in the form of 3 grams of NPK and urea. The fermentation bottle is tightly closed and connected to a plastic hose which is inserted into a bottle filled with water to accommodate the CO2 gas produced and allowed to stand for 3×24 hours. The fermented solution was distilled at 78°C for 3 hours to obtain pure ethanol (bioethanol). Bioethanol was tested by testing the viscosity and density [15].

\[
\text{Bioethanol content} = \frac{\text{bioethanol weight}}{\text{weight of durian seed flour}} \times 100\%
\]

Making Traveling Paper Soap Made from Virgin Coconut Oil (VCO) and Bioethanol Durian Seed
Making Traveling Paper Soap begins with weighing all ingredients according to the formula (Table 1). The first mixture was made by adding 30% NaOH into the VC. Then it was heated to a temperature of 60°C and stirred until homogeneous at a temperature of 50°C. The second mixture was made by diluting stearic acid and adding glycerin (F1 did not use glycerin), durian seed ethanol, and distilled water with stirring for each addition of ingredients. The first mixture is poured into the second mixture, adding essential oils, and heat for 30-60 minutes with constant stirring and not too fast. The mixture is poured into molds and left for 24 hours to harden in the form of paper, then cut into 3x3 cm sizes and characterized [16]. The characterization of Traveling Paper Soap includes pH test, organoleptic test, water content test, free alkali test, and antibacterial activity Traveling Paper Soap with Disk Diffusion method.

Table 1. Traveling Paper Soap and Virgin Coconut Oil (VCO) Durian Seed Bioethanol

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>F1 (%)</th>
<th>F2 (%)</th>
<th>F3 (%)</th>
<th>F4 (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Seed Bioethanol</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Durian VCO</td>
<td>16.3</td>
<td>16.3</td>
<td>16.3</td>
<td>16.3</td>
</tr>
<tr>
<td>NaOH</td>
<td>17.80</td>
<td>17.80</td>
<td>17.80</td>
<td>17.80</td>
</tr>
<tr>
<td>30% Stearic acid</td>
<td>7.83</td>
<td>7.83</td>
<td>7.83</td>
<td>7.83</td>
</tr>
<tr>
<td>Glycerin</td>
<td>0</td>
<td>7.12</td>
<td>10.7</td>
<td>14.23</td>
</tr>
<tr>
<td>Essential Oil</td>
<td>0.71</td>
<td>0.71</td>
<td>0.71</td>
<td>0.71</td>
</tr>
<tr>
<td>Aquades</td>
<td>44.48</td>
<td>37.81</td>
<td>33.80</td>
<td>30.30</td>
</tr>
</tbody>
</table>

Characterization of Traveling Paper Soap pH Test
The pH test was carried out by dissolving each sample of soap formula using distilled water for 24 hours, then the pH meter was inserted into the sample and observed [17].

Water Content Test
The water content test was carried out using the SNI 06-3532-1994 method. The sample of soap formula was weighed as much as 2 g and put into an aluminum foil container, heated in an oven at 105°C for 2 hours until the weight was constant, then weighed and counted.

\[
\text{Water Content} = \frac{B - C}{B - A} \times 100\%
\]

Information:
A = weight of empty container (g)
B = weight of container + sample (g)
C = weight of container + dry sample (g)

Free Alkali Test
Test The free alkali test is carried out by the method SNI 06-3532-1994. Each soap formula was weighed about 1.25 g, then put into a neutral alcohol solution, set in the reflux condenser, and simmered for 30 minutes until the soap dissolved completely.
The free alkali test was carried out by titrating the sample, which had been heated using 0.1 N HCl in alcohol from the burette until the red color disappeared. Then the free alkali content was calculated.

Free Alkali Content = \( \frac{V \times N \times 0.04}{W} \times 100\% \)

Information:
- \( V \) = volume of HCl titration (mL)
- \( N \) = normality of HCl (N)
- \( W \) = sample weight (g)
- 0.04 = equivalent weight of NaOH

**Organoleptic Test**

Test soap was carried out with 15 respondents with measurement indicators in each formulation, including color, aroma, texture, foam, solubility, ability to clean dirt, and skin texture after use. The rating scale ranges from 1-5. The data obtained were processed using the Statistical Package For The Social Science (SPSS) [18].

**Antibacterial Activity Test**

Antibacterial activity test was carried out in vitro Disk Diffusion Method. Filter paper disk that serves as a place to accommodate antimicrobial substances (19). The media used is Nutrient Agar and made into solid media. Media Nutrient agar (NA) cooled and solidified, then planted with bacteria. Bacteria were evenly planted on the surface of nutrient agar (NA) using a spreader, and then Dish has placed in nutrient agar (NA) media that had been planted with bacteria. The next step is that each Traveling Paper Soap is inserted into a Disk for comparison using an antibiotic from Amoxilin, carried out by incubation for 24 hours at 37°C. The greatest antibacterial activity was indicated by the diameter of the largest clear zone formed from this concentration. The smallest sample concentration that can inhibit the inoculated bacteria with the formation of a clear zone is the value of the Minimum Inhibitory Concentration of the sample [20].

**RESULTS AND DISCUSSION**

**Making Durian Seed Bioethanol**

The yield of bioethanol obtained was 97 mL with a bioethanol content of 19.95% from the hydrolysis of 486 grams of durian seed flour. In the glucose level test, the results of hydrolysis obtained positive results. The formation of a purple ring indicates the glucose level test with the Molisch test with a positive result, and the Fehling test with a positive result is indicated by the formation of a red brick precipitate after heating. Characterization of durian seed bioethanol was carried out by testing specific gravity and viscosity. In this study, the specific gravity of durian seed bioethanol was 0.9669 g/mL, and the viscosity was 0.7137 cP (temperature 28°C). The value of specific gravity and viscosity of the study's results was higher than that of Amiran et al. (2019), which obtained a specific gravity of 0.8654 g/mL and a viscosity of 0.693 cP in the bioethanol of bidara fruit [13]. This difference in value can be caused by differences in the type of sample used. It is suspected that there are aquades that also evaporate in the distillation process of durian seed bioethanol.

**Formulation Traveling Paper Soap**

The results of making Traveling Paper Soap based on four formulations can be seen in Figure 1.

**Characterization of Traveling Paper Soap**

**Formulation Traveling Paper Soap**

**pH Test**

pH test is a benchmark for quality and requirements for commercialization. According to SNI standard, the pH of transparent solid soap is between 9-11. The results of pH testing on each formulation of Traveling Paper Soap durian seeds have met SNI standards, namely data F1, F2, F3, and F4 respectively of 10.4, 10.4, 10.5, and 10.5.

**Water Content Test**

The formulation of Traveling Paper Soap according to calculations ranging from 8-17% (see Table 4). According to SNI 06-3532-1994, a good water content for solid soap is no more than 15%, so the Traveling Paper Soap has met the requirements for the water content of SNI. The data from the water content test (Table 4) shows that formulation 2 is the best in the water content test because it produces the lowest percentage of water content, which is 7%. Formulation 4 is a formulation that violates the SNI standard 06-3532-1994 because the water content obtained is 17%.
Test of Free Alkali Levels

Test results of free alkali on the formulation of Traveling Paper Soap ranged from 0.032-0.048% (see Table 5). These results indicate that the soap has met the required free alkali of SNI 06-3532-1994, namely no more than 0.1% for NaOH and 0.14% for KOH. The data from the water content test (Table 4) shows that formulation 2 is the best in the water content test because it produces the lowest percentage of free alkali, which is 0.032%.

Table 4. Data of The Result for Water Content of Traveling Paper Soap

<table>
<thead>
<tr>
<th>Sample</th>
<th>Water Content</th>
</tr>
</thead>
<tbody>
<tr>
<td>F1</td>
<td>8%</td>
</tr>
<tr>
<td>F2</td>
<td>7%</td>
</tr>
<tr>
<td>F3</td>
<td>12%</td>
</tr>
<tr>
<td>F4</td>
<td>17%</td>
</tr>
</tbody>
</table>

Table 5. Data of Free Alkali Level Test Results of Traveling Paper Soap

<table>
<thead>
<tr>
<th>Sample</th>
<th>Free Alkali Levels</th>
</tr>
</thead>
<tbody>
<tr>
<td>F1</td>
<td>0.048%</td>
</tr>
<tr>
<td>F2</td>
<td>0.032%</td>
</tr>
<tr>
<td>F3</td>
<td>0.048%</td>
</tr>
<tr>
<td>F4</td>
<td>0.048%</td>
</tr>
</tbody>
</table>

Organoleptic Test

Physical characterization of Traveling Paper Soap bioethanol durian seeds using an organoleptic test with testing on 15 respondents. The formulation with the best respondents was formulation 4, with the best respondents on the indicators of aroma, texture, foam, solubility, ability to clean dirt, and skin texture after use. Formulation 3 is the best formulation after formulation 4, superior in indicators of texture, foam, solubility, and skin texture after use. Formulation 2 is the best third-order formulation because it has the same advantages in aroma indicators and the ability to clean dirt. Formulation 1 became the formulation with the lowest advantage because no glycerin was added.

Antibacterial Activity Test

The bacteria used for the Traveling Paper Soap are gram-positive Staphylococcus aureus and Staphylococcus epidermis which can attack the skin. The antibacterial activity test was carried out using the Disk method, while the Disk had a diameter of ±6 mm. Antibacterial test analysis paper soap against Staphylococcus aureus and Staphylococcus epidermis can see in Table 6 and Table 7. In this antibacterial activity test, a control was used with the test sample using amoxicillin. Table 6 and Figure 7 show that the control treatment had the clearest zone or the lowest inhibition diameter (DDH) against Staphylococcus aureus bacteria, namely in formula 1 of 11.6 mm, while the highest was in F3 and F4 of 14.6 mm. In the control treatment of Staphylococcus epidermis, the lowest diameter was at F1 of 15 mm, while the highest was at F2 of 22.3 mm. It proves that the antibacterial activity of the traveling Paper Soap that has been made is more effective against the Staphylococcus epidermis, especially in Formula 2 (F2).

Table 6. Data of Antibacterial Test Traveling Paper Soap Against Staphylococcus epidermis

<table>
<thead>
<tr>
<th>Formula</th>
<th>Experiment (mm)</th>
<th>Mean (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>F1</td>
<td>14</td>
<td>16</td>
</tr>
<tr>
<td>F2</td>
<td>23</td>
<td>21</td>
</tr>
<tr>
<td>F3</td>
<td>22</td>
<td>21</td>
</tr>
<tr>
<td>F4</td>
<td>20</td>
<td>23</td>
</tr>
<tr>
<td>Control</td>
<td>25</td>
<td>28</td>
</tr>
</tbody>
</table>

Table 7. Data of Antibacterial Test Traveling Paper Soap Against Staphylococcus aureus

<table>
<thead>
<tr>
<th>Formula</th>
<th>Experiment (mm)</th>
<th>Mean (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>F1</td>
<td>9</td>
<td>15</td>
</tr>
<tr>
<td>F2</td>
<td>10</td>
<td>15</td>
</tr>
<tr>
<td>F3</td>
<td>14</td>
<td>16</td>
</tr>
<tr>
<td>F4</td>
<td>14</td>
<td>15</td>
</tr>
<tr>
<td>Control</td>
<td>34</td>
<td>40</td>
</tr>
</tbody>
</table>

Figure 2. Graph of Organoleptic Test Against 15 Respondents
CONCLUSION
Based on the research that has been done, it can be concluded that the formulation of making traveling paper soap can be done using Virgin Coconut Oil (VCO) and Durian Seed Bioethanol as raw materials. The produced soap meets the Indonesian national Standard SNI 3532:1994 standard.

REFERENCES