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Soap Made from Kesum Leaf Squeezedto Inhibit Growth *Staphylococcus aureus.*

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Abstract. Biodiversity, especially in West Kalimantan, is very diverse, one of which is the kesum plant (Polygonum minus husks). The kesum plant is a plant that can be used as herbal medicine. The utilization of kesum plants is usually done as part of the leaves, namely as local processed cooking spices. Based on Fitofarmaka studies, kesum plants are believed to have antibacterial, antifungal, anti-viral, antioxidant, anti-cancer, and antiulcer activities. The study's aim was to find out how well different concentrations of kesum leaf juice-based solid soap formulations stopped the growth of Staphylococcus aureus bacteria. This study used kesum leaves (Polygonum minus husks), which were squeezed, and then made solid soap preparations with concentrations of 3.93%, 5.25%, and 6.56%. Nine repetitions were carried out in each treatment, so the number of samples used was 27. The results of the antibacterial inhibition test showed that the soap preparation samples had the ability to inhibit the growth of Staphylococcus aureus bacteria. The result of the statistical analysis value in the Friedman test is P = 0.000, which states that there is a difference in each concentration of solid soap prepared from kesum leaf juice. Dilution concentrations of 5.25% and 6.56% are more effective in inhibiting the growth of Staphylococcus aureus bacteria than 3.93%.

Keywords: Soap, kesum leaf, Staphylococcus aureus

1. Introduction

The search for substitute antibacterial medicines is imperative due to the growing threat that antibiotic-resistant bacteria represent to world health. The gram-positive bacteria *Staphylococcus aureus* is known to cause a variety of diseases and is commonly found on human skin and nasal surfaces (Syari *et al.*, 2022). The need for innovative alternatives is emphasized by the declining effectiveness of conventional antibiotics. Kesum leaves (*Polygonum minus Huds*) have been shown in studies to have possible antibacterial effects against Staphylococcus aureus (Lumbantoruan, 2013; Qader *et al.*, 2011). To close the existing knowledge gap, nevertheless, a thorough grasp of its effectiveness, particular antibacterial mechanisms, and possible uses in contemporary medicine is required (Lumbantoruan, 2013; Tong *et al.*, 2015)

Natural antibacterial substances have been studied in the past using a variety of techniques, such as agar well diffusion tests, broth microdilution, and disc diffusion assays. These approaches have benefits like ease of use and affordability, but they also have drawbacks including environmental fluctuation and a limited capacity to identify certain antibacterial pathways. Our comprehension of the effectiveness of kesum leaves may be improved by molecular techniques that clarify the genetic basis of antibacterial action (Vikram *et al.*, 2014; Howden *et al.*, 2023). To overcome current methodological restrictions, however, there is still a need for uniform methodologies across research and the investigation of cutting-edge approaches.

There is a study void on the precise antibacterial processes and standardized assessment techniques for kesum leaves against *Staphylococcus aureus*, despite the increased interest in

natural antibacterial agents. Although studies such as (Lumbantoruan, 2013). demonstrate encouraging antibacterial activity, there is a deficiency in a thorough synthesis of current research. It is still difficult to identify the precise bioactive substances with antibacterial qualities and how they work. To fully utilize kesum leaves and include them into contemporary antibacterial techniques, these gaps must be filled (Vikram *et al.*, 2014; Sukmayadi, Lestari and Dinda Ayu, 2023).

By methodically examining the antibacterial properties of kesum leaves against *Staphylococcus aureus*, this study seeks to close the current research gap. Both sophisticated molecular methods and conventional antibacterial testing will be used to offer a thorough grasp of the underlying processes. By doing this, the research hopes to further the creation of all-natural antibacterial medicines and open the door for kesum leaves to be used in conventional medical procedures. It is anticipated that the results will confirm conventional usage and provide new perspectives on how to combat the worldwide issue of antibiotic resistance (Lumbantoruan, 2013; Vikram *et al.*, 2014).

2. Methods

2.1 Research design

The research design used was a quasi-experiment (quasi-experiment) using the disc diffusion method to measure the inhibition zone of solid soap preparations from the juice of kesum leaves (Polygonum Minus Huds) concentrations of 15%, 20%, and 25%, and dilution was carried out at concentrations of 3.93%, 5 .25%, and 6.56% against the growth of Escherichia coli bacteria. The principle of the examination is that a paper disc is soaked in a soap preparation that has been added with juice from kesum leaves (Polygonum Minus Huds) in various concentrations, then placed on agar media that has been planted with Staphylococcus aureus bacteria, incubated for 24 hours, and the diameter of the inhibition zone is measured. The population in this study was kesum leaves (Polygonum Minus Huds. The samples in this study were squeezed and then made into solid soap preparations with concentrations of 15%, 20%, and 25% and diluted with concentrations of 3.93%, 5.25%, and 6.56%.

2.2 Making Kesum Leaf Juice

The kesum leaves that have been sorted are washed thoroughly with running water, dried, and then crushed until smooth. The crushed leaves are squeezed with sterile gauze. The resulting squeezed juice is placed in a sterile Erlenmeyer flask and closed tightly. Then it is formulated to make solid soap preparations with various concentrations.

2.3 Making Preparation Soap Congested

The stearate acid is melted by heating it to 70°C. NaOH is then diluted with aquadest and cooled to the NaOH solution's temperature. A solution of 4 grams of palm oil and 2 grams of coconut oil (VCO) is then added, mixed with a mixer rod, and the oil is gradually thickened by adding the NaOH solution. Once the oil is homogeneous and well-mixed, pumpkin leaves are added. The mixture is then poured into a sterile soap mold and allowed to solidify for one to two days (Table 1).

Table 1. Method making Preparation Soap Congested Squeeze Leaf Nasty

Material	15% Formulation	Formulation 20%	25% Formulation	Function
Squeeze the kesum leaves	3 ml	4 ml	5 ml	Active ingredients
NaOH 18%	3.6 gr	3.6 gr	3.6 gr	Alkali
Palm oil	4 gr	4 gr	4 gr	Fatty acid
VCO coconut oil	2 gr	2 gr	2 gr	Fatty acid
Stearic acid 17%	3.4 gr	3.4 gr	3.4 gr	Foam stabilizer
Aquadest	4 ml	3 ml	2 ml	Solvent

2.4 Evaluation Preparation Soap Congested

Organoleptic Test . Organoleptic tests carried out is a physical test from soap covering liquid, color , smell and shape (Maulana, 2022) .

Test pH. pH testing is carried out using Universal pH with the method of 1 gram of sample soap liquid dissolved in 10 ml of distilled water, then homogenize . Then dip universal pH. The results are read on the pH scale and recorded (Maulana, 2022)**.**

Foam Height Test. Foam height testing is carried out by dissolving a 5 gram sample of solid soap with 10 ml of water, placing it in a test tube, closing it using your thumb, and shaking for 1 minute, then measuring the height of the foam formed. The height and stability of the foam were observed 5 minutes after shaking (Maulana, 2022) **.**

2.5 Power Test Resistor Preparation Soap Congested Squeeze Leaf Nasty Method Diffusion Disc

The paper disc is dipped in a soap solution at each concentration for 10 minutes . The sterile swab is inserted into the Staphylococcus aureus bacterial suspension. and inoculated onto the entire surface of the MHA media evenly, then left for 3-5 minutes. The paper discs that have been soaked in each concentration are drained for 5 minutes, and the paper discs are attached to the surface of the inoculated media. then incubated at a temperature of 37 for 18–24 hours in an incubator . The results were observed by looking at the size of the zone of inhibition.

3. Results

Table 2. Phytochemical Screening Results Juice of Kesum Leaves (*Polygonum Minus Huds*)

Phytochemical Test	Test results
Alkaloids	Positive
Flavonoids	Positive
Saponins	Positive
Tannin	Positive
Phenol	Positive

Table 2 indicates the presence of many chemical components, including flavonoids, saponins, tannins, alkaloids, and phenols, in the leaves of the positive pineapple.

Table 3. Organoleptic Test Results Preparation Solid Soap from Kesum Leaf Juice (*Polygonum Minus Huds*)

Concentration	Color	Smell	Form
15%	Green	Typical notice	congested
20%	Green	Typical notice	congested
25%	Green	Typical notice	congested
Base control	White	No smell	congested

Table 3 presents the findings of the organoleptic tests conducted on the chickpea leaves during the manufacture of solid soap. (*Huds Polygonum Minus*). An odorless, solid, white preparation is produced by the control base. The preparation takes on a solid shape, a pronounced leaf scent, and a green tint at concentrations of 15%, 20%, and 25%. The observation of leaf polygonum minus huds is the source of the soap preparation's green hue.

Table 4 . Test Results for the Degree of Acidity (pH) of Solid Soap Preparation from Kesum Leaf Juice (*Polygonum Minus Huds*)

Concentration	Degree of Acidity (pH)
15%	11
20%	11
25%	11
Control soap base	10

Table 4 shows that a sample of solid soap preparation has an average pH of 15%, 20%, 25%, or 11.

Table 5. Foam Height Test Results Preparation of Solid Soap from Squeezed Kesum Leaves (*Polygonum Minus Huds*)

Concentration	Foam Height Test
15 %	6.9 cm
20%	6.4 cm
25%	8.5 cm

Table 5 shows that the average foam height created during the manufacturing of thick soap at concentrations of 15%, 20%, and 25% is 6.9 cm, 6.4 cm, and 8.5 cm, respectively.

Table 6. Power Test Results Resistor Preparation of Solid Soap from Squeezed Kesum Leaves (*Polygonum Minus Huds*)

Replication	Inhibition zone (mm)			
	Concentration 3.93 %	Concentration 5.25 %	Concentration 6.56 %	Negative control
R1	0mm	7.5mm	9mm	There are no inhibition zones
R2	0mm	8.5mm	9.5mm	There are no inhibition zones
R3	7mm	7.5mm	9.5mm	There are no inhibition zones
R4	6.5mm	8.5mm	10mm	
R5	7mm	7.5mm	10.5mm	
R6	7mm	8.5mm	11mm	
	7.5mm	9mm	9mm	
	6.5mm	8.5mm	10.5mm	
	0mm	9mm	10mm	
erage	4,611mm	8.111mm	9,889mm	

Table 6 above indicated that the average concentration of the barrier zone was 3.93% at 4.6 mm, the average barrier area was 8.1 mm with a 5.25% concentration, and the average concentration was 9.8 mm with a 6.56% concentration.

4. Discussion

The research that was carried out used samples of solid soap from the juice of kesum leaves (Polygonum minus husks) with concentrations of 15%, 20%, and 25% to determine the differences in the inhibitory power of each concentration in inhibiting the growth of Staphylococcus aureus bacteria using the diffusion method. Then dilute the soap with concentrations of 3.93%, 5.25%, and 6.56%, dilute using 5 ml of distilled water, and carry out 9 repetitions. Kesum leaf juice was subjected to a phytochemical screening test, including testing for flavonoid compounds, saponins, tannins, alkaloids, and phenols, which contain antibacterial compounds. The positive phytochemical screening test results obtained can be seen in Table 3. Based on research conducted by(Kartikasari, Ristia Rahman and Ridha, 2022) (Dewi *et al.*, 2019) It can be concluded that the results of the phytochemical screening test of kesum leaves contain phenols, terpenoids, alkaloids, flavonoids, saponins, and tannins. So the research that has been carried out is in accordance with the results of previous research. Phytochemical testing is an important step in efforts to reveal the potential of plant resources. The purpose of phytochemical screening is to find out whether there are compounds that the researcher wants in leaf juice (Habibi, Firmansyah and Setyawati, 2018). Bacterial identification tests were carried out by carrying out microscopic tests and biochemical tests, namely catalase and coagulase. The results of microscopic observations using gram staining show the characteristics of gram-positive bacteria with a round cluster arrangement. The results of the biochemical test on catalase showed positive results with the formation of air bubbles. The catalase test gives positive results because the peroxidase enzyme plays a role in microbial survival. Catalase hydrates hydrogen peroxide (H2O2) in bacterial cells before it damages the cells. The coagulase test showed positive results for the formation of coagulation. Coagulase is a protein that resembles an enzyme that can coagulate plasma oxalate or citrate with the help of a factor found in serum. The results of the research that has been carried out are in accordance with the research that has been carried out by(Lasmini and Sitorus, 2018) identified Staphylococcus aureus bacteria using the catalase test to differentiate bacterial genera. The coagulase test is used to determine whether or not the coagulase enzyme is produced by Staphylococcus bacteria. The microscopic test is used to see the characteristics and shape of Staphylococcus bacteria using a microscope. Making solid soap through a saponification process involves heating fats and oils as triglycerides and reacting them with NaOH as an alkali that has been dissolved first to produce a soap preparation. The results of organoleptic tests on soap preparations include color, odor, and shape. The soap preparation is green in color, solid in shape, and has a characteristic smell of leaf juice. The green color in the soap preparation comes from the juice of the leaves of kesum (Polygonum Minus Huds). According to research conducted by (Maulana, 2022), Organoleptic tests, smell tests, shape tests, and color tests were carried out to determine the physical properties of soap preparations. The results of the pH check using universal pH paper showed that the results for soap preparations with concentrations of 15%, 20%, and 25% were 11, and the base control pH was 10. The pH value is an important parameter in making soap because pH determines the suitability of the soap. The pH requirement for safe soap is 9–11 (Setiawati and Ariani, 2021), and if the pH value is very high, it can cause irritation to the skin. The research soap preparation meets the standards and is safe to use because it is in the pH range that meets the requirements. The results of the examination of the foam height were around 6–8 cm, carried out by shaking the test tube, then observing and measuring the height of the foam formed after leaving it for 5 minutes. In terms of foam height, the solid soap produced meets the requirements.

The results of the antibacterial inhibition test of soap preparation samples that had been diluted with concentrations of 3.93%, 5.25%, and 6.56% had the ability to inhibit the growth of Staphylococcus aureus bacteria using the diffusion method, as indicated by the formation of a clear zone on the MHA media (*Muller Hinton Agar*). According (Hidayah, Mustikaningtyas and Harnina, 2017) antibacterial activity is influenced by several factors, namely the compound content and concentration of antibacterial compounds. The higher the concentration of antibacterial compounds in juice, the larger the diameter of the inhibition zone, thereby slowing bacterial growth. The amount of antibacterial compounds released increases, making it easier for these compounds to enter bacterial cells. Soap preparations that have been diluted to a concentration of 3.93% produce an average inhibitory zone diameter of 4.6 mm; soap dilution preparations with a concentration of 5.25% produce an average inhibitory zone diameter of 8.1 mm; and soap dilutions with a concentration of 6.56 percent produce an average inhibition zone diameter of 9.8 mm. The diameter of the inhibition zone produced at each concentration shows that there are differences. Based on the results of this examination, the strength of the inhibitory power can be categorized. According to (Surjowardojo et al., 2016), inhibitory strength is divided into 4 categories: weak inhibitory strength (≤5mm), medium inhibitory strength (6–10 mm), strong inhibitory strength (11–20 mm), and very strong inhibitory strength (≥21mm). The results of the inhibition zone for the preparation of kesum leaf juice soap (Polygonum Minus Huds) which had been diluted with a concentration of 3.93%, were in the weak category, and concentrations of 5.25% and 6.56% were in the medium category.

5. Conclusion

This study aimed to evaluate the antimicrobial properties of solid soap formulations derived from kesum leaf juice (Polygonum minus husks) at varying dilution concentrations (3.93%, 5.25%, and 6.56%). The main findings of the research revealed distinct differences in the inhibitory zone diameters, with average values of 4.6 mm, 8.1 mm, and 9.8 mm, respectively. The statistical analysis, conducted through the Friedman test, demonstrated a significant disparity in the average inhibition zones among the different soap formulations, with a p-value of 0.000. This highlights the importance of the kesum leaf juice concentration in influencing the antimicrobial efficacy of the solid soap. Looking ahead, future research should delve into optimizing the kesum leaf juice concentration for maximal effectiveness in soap formulations. More research into things like how kesum leaf juice might work with other ingredients or different ways of making soap could make it even more useful for keeping things clean and stopping the growth of microbes.These endeavors would contribute to a more comprehensive understanding of the potential benefits of kesum leaf juice in soap preparations and its broader implications for public health.

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