

Anti Acne Cream Formulation Ethanol Extract of Katuk Leaves (*Sauropus androgynus* (L.) Merr against *Staphylococcus Epidermis* and *Staphylococcus Aureus*

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ARTICLE INFO

Article history:

Received: 5th October 2022

Revised: 20th November 2022

Accepted: 3rd December 2022

Keywords:

Anti-acne

Cream

Katuk leaf extract

Staphylococcus epidermis

Staphylococcus aureus

ABSTRACT

Acne is a skin problem that generally occurs during puberty. Under normal conditions, during puberty, there are *Staphylococcus epidermal* bacteria and *Propionibacterium acnes* that proliferate so that inflammation occurs, causing acne on the skin. Since ancient times, our ancestors believed in the use of natural ingredients to treat various diseases, one of which was katuk leaves. Katuk plants have secondary metabolic compounds such as alkaloids, saponins, flavonoids and tannins. The potential of Katuk leaves has been tested for many pharmacological activities, such as antibacterial, antihypertensive, anti-infective, antihyperlipidemic, antianemic and also increases milk production. Katuk leaves are often used in the form of extracts as antibacterial, so it needs to be developed into a pharmaceutical preparation to increase its use. One of the pharmaceutical preparations is cream preparations to treat the causes of acne. This study aims to determine the ability of katuk leaf extract cream to act as an antibacterial against *Staphylococcus Epidermis* and *Staphylococcus Aureus* bacteria. The design of this study is an experimental method, making katuk leaf extract by maceration method by immersing in 70% ethanol solution for 5 days. After 5 days, the pulp and filtrate were separated and then evaporated using a Vacuum Rotary Evaporator to obtain a thick extract. The preparation of katuk leaf extract cream was carried out with a concentration of 10%, 15%, 20% and an antibacterial test was carried out using the pitting method. The antibacterial test results of the katuk leaf cream extract showed an inhibition zone marked with a clear area around the well. Katuk leaf extract cream was able to inhibit the growth of *S. aureus* and *S. epidermis* bacteria with the best concentration in formula 3, namely the concentration of katuk leaf extract of 20%.

I. Introduction

Phytochemical compounds contained in the katuk plant are saponins, flavonoids, tannins and isoflavonoids (Susanti et al., 2014). Saponins are proven to be efficacious as anticancer, antimicrobial and improve the immune system. The mechanism of action of saponins as antimicrobials is to damage the membrane by disrupting its permeability (Canell, 1998). The antimicrobial activity of katuk leaf infusion showed a zone of inhibition against *Candida albican*. Clinical features of acne include excess oil production, non-inflammatory lesions such as open comedones and closed comedones, inflammatory lesions such as papules and pustules. The prevalence of acne sufferers in Indonesia ranges from 80-85% in adolescents with a peak incidence aged 15-18 years, 12% in women > 25 years and 3% at the age of 35-44 years (Hendra et al., 2019).

Extraction using the maceration method has the advantage that it guarantees that the extracted active substance will not be damaged. During the immersion process, cell walls and cell membranes break down so that secondary metabolites are broken and dissolved in organic



solvents. Cream is a cosmetic preparation that is easy and practical to use and is defined as a semi-solid preparation containing one or more dissolved drug ingredients (Febria, 2012). Generally, it is formed from oil to water in the oil and humectant phases which looks almost the same as lotion (Lestari et al., 2020). Antibacterial power testing is one way to measure the ability of antibacterial substances to inhibit bacterial growth in vitro. Well diffusion is one of the methods in determining antibacterial potential by measuring the diameter of the transparent zone, so that the media that has been overgrown with bacteria is made wells. The well was filled with antibacterial test preparations and then incubated 24 hours at 37°C and then observed.

II. Methods

Materials

Katuk leaves, 70% alcohol, cream base ingredients are stearic acid, glycerin, TEA, propyl paraben, methyl paraben, aquadest. Nutrient Agar (NA) media, Nutrient broth Agar (NB), Staphylococcus Epidermis (S.epidermis) and Staphylococcus Aureus (s.aureus) bacteria obtained from the Microbiology Laboratory of IIK STRADA Indonesia Kediri, Garamycin cream, H₂SO₄ 0.36 N, BaCl₂ .2H₂O 1.175%, NaCl 0.9%, filter paper, label paper, aluminum foil.

Tools

The equipment used are: water bath, electric stove, simple grinding machine, 65 mesh sieve, analytical balance, Rotary Evaporator, erlemeyer, measuring cup, test tube, stirring rod, incubator, oven, borehole, ose needle, petri dish, tweezers , autoclave, bunsen, vernier calipers.

Preparation Sample

This study used all shoots of katuk leaves taken in Ds. Ceweng, Diwek District, Jombang Regency. The collected leaf shoots are separated from impurities, then washed with running water until clean then drained and dried by aerating. The dried shoots of katuk leaves were then pollinated using a simple grinding machine, the resulting powder was sieved using a 65 mesh until a uniform powder was obtained and then put into a closed glass container.

Extract Making

Making katuk leaf extract using the maceration method by soaking it using 70% alcohol solvent, where the amount of simplicia used is 200 grams then put into a dark bottle and left for 5 days while stirring occasionally. After 5 days, it was filtered using a flannel cloth, then filtered again using filter paper to produce filtrate 1 and the rest of the dregs 1. The dregs 1 was then added with 70% alcohol solvent until completely submerged and left for 2 days, stirring occasionally. After 2 days it was filtered again and the results of filtrate 2 were mixed with filtrate 1, then concentrated using a rotary evaporator until a thick extract was obtained.

Making Katuk Leaf Extract Cream

Katuk leaf extract cream was made with 3 formulas with a cream weight of 100g. Formulas I, II, and III used stearic acid as an emulsifier with various extract concentrations of 10%, 15%, and 20%. The formula in this study can be seen in table 1.

Table 1. Formula of Katuk Leaf Extract Cream

Material Name	Quantity of ingredients (%)		
	F1	F2	F3
Katuk leaf extract	10	15	20
Stearat Acid	14	14	14
Gliserin	10	10	10
Trietanolamin	1	1	1
Propil Paraben	0,02	0,02	0,02
Metil Paraben	0,075	0,075	0,075
Aquadest (ad)	Ad 100	Ad 100	Ad 100

The manufacture of oil phase cream, namely stearic acid, propyl paraben, was heated at a temperature of 70°C-80°C. The aqueous phase, namely katuk leaf extract, glycerin, triethanolamine, methyl paraben and distilled water was heated to a temperature of 70°C-80°C. The melted oil phase is put into a preheated mortar and then the water phase is added little by little until while stirring and a homogeneous cream is formed.

Sterilization

The equipment used in the antibacterial test study was sterilized first. Glass utensils were sterilized in an oven at 170°C for 24 hours, ose needles and tweezers were sterilized by burning directly using busen burner, while the media were sterilized using an autoclave at 121°C for 15 minutes.

Bacterial growth media

NA as much as 15 ml which has been liquefied is poured into a sterile test tube and then allowed to solidify at a slope of 30° and the media is ready to be used for bacterial inoculation.

Bacterial Inoculation

The test bacteria were taken using a sterile ose needle, then implanted on an inclined medium by scraping and then incubated in an incubator at 37°C for 24 hours.

Preparation of Bacterial Suspension

Preparation of bacterial suspension by taking 1 sterile ose of bacterial colonies mixed with NaCL solution in a test tube and homogenized with a vortex mixer then compared with a 0.5 McFarland solution which is equal to 1.5×10^8 bacteria.

Antibacterial Activity

The antibacterial activity was tested by inserting 15 ml of NA in a petri dish and then adding 0.5 ml of suspension of *S. aureus* and *S. epidermis* bacteria inside and waiting until solid. After the solid media, bore holes according to the number of test preparations, then the test preparations are placed according to the marks on the wells and incubated at 37°C for 24 hours and observed.

Observation and Measurement

Observations were made after 1x24 hours of bacterial incubation, the results of antibacterial activity were based on the measurement of the Diameter of the Inhibitory Area (DDH) of bacterial growth formed around the hole or well, the measurement was made from the bottom of the petri dish with a caliper in millimeters (mm).

Data analysis

The results of the antibacterial test of katuk leaf extract cream (*Sauropus androgynus* (L.) Merr) on the diameter of the inhibition zone for the growth of *S. aureus* and *S. epidermis* bacteria were analyzed using the One way ANOVA method with Statistical Services Solution (SPSS 24). carried out by the Tukey method. The test aims to determine the difference between the gel formula (F1, F2, F3) and the positive control. The existence of a significant difference in the test was marked by the value of $p < 0.05$.

III. Results and Discussion

The study used katuk leaf extract cream to determine the antibacterial activity against *S. aureus* and *S. epidermis*. The sample used in this study was previously identified in Materia Medica Batu City. Based on the identification, it was found that the sample used was katuk leaf (*Sauropus androgynus* (L.) Merr).

The samples used were the shoots of the katuk leaf plant which were light green in color, the shoots of the katuk leaves were then dried to prevent the growth of fungi so that they could be stored in the long term. Prior to extraction using the maceration method, the dried leaf shoots were ground to speed up the penetration of the solvent into the plant cells and also to dissolve the compounds contained in the sample.

Maceration uses 70% alcohol solvent, where 70% alcohol contains water to wet the dry sample so that the plant cells will expand making it easier for the solvent to penetrate to bind the compounds contained therein. The results of the macerate were separated and fed with a rotary evaporator until a thick extract of 67.80 grams was obtained. The results of organoleptic examination of the thick extract of katuk leaves were greenish brown. After conducting preliminary tests of secondary metabolites from katuk leaf extract, it was shown that the katuk leaf plant contains flavonoid, phenolic, alkaloid and saponin compounds.

The results of bacterial identification showed that the bacteria were *S. aureus* and *S. epidermis*. This is evidenced by the use of gram staining, namely *S. aureus* and *S. epidermis* bacteria showing blue color because they retain crystal violet dye with a round, clustered shape.

Antibacterial testing used a negative control using a cream base without active substances and as a comparison or positive control, namely Garamicin cream, namely the antibiotic gentamicin. Selection of cream base as a negative control because it does not affect the results of the antibacterial activity test. The selection of comparisons using Digenta cream is an antibiotic that has guaranteed levels and activities, making it easier for researchers to get maximum observations. Antibacterial compounds do not kill but inhibit the growth of bacteria, while antibiotics in small concentrations can inhibit and kill microorganisms according to their mechanism of action. The results of measuring the diameter of the inhibition zone of of katuk leaf extract cream against *S. aureus* and *S. epidermis* bacteria can be seen in table 2.

Table 2. Inhibitory Zone Leaf Extract Katuk Cream against *S. aureus* and *S. epidermis*

No	Concentration	Diameter of Bacterial Growth Inhibitory Zone (mm)							
		<i>Stapylococcus aureus</i>				<i>Stapylococcus epidermis</i>			
		I	II	III	average	I	II	III	average
1.	10%	2,12	2,08	2,16	2,12	2,34	2,42	2,4	2,39
2.	15%	3,11	3,07	3,08	3,09	3,15	3,05	3,2	3,1
3.	20%	4,06	4,14	4,09	4,10	4,34	4,32	4,3	4,3
4.	Control (+)	6,87	6,91	6,8	6,86	7,06	7,12	7,09	7,09
5.	Control (-)	0	0	0	0	0	0	0	0

Based on the results in table 3. shows that there is antibacterial activity of katuk leaf extract cream against *S. aureus* and *S. epidermis* bacteria. The results of the diameter of the inhibition zone in the 3 formulas compared to the positive control were smaller, while the negative control did not have a growth inhibition zone, this indicates that the cream base is an inert material and does not act as an antibacterial. The results of this study showed that the positive control and the three formulas with concentrations of 10%, 15% and 20% gave significant differences in antibacterial activity. While the negative control and positive control showed a significant difference, between the negative control and the three formulas there was also a significant difference. From the data above for formula 3 with a concentration of 20% katuk leaf extract, it shows that the sample has antibacterial activity against both bacteria and the result is a larger diameter than formula 1 and formula 2.

Based on the results of the statistical test analysis for the inhibition zone area, the result was ($p < 0.05$), which means that the administration of katuk leaf extract had an effect on the

inhibition zone area on *S. aureus* and *S. epidermis* bacteria, although the diameter of the inhibition zone was larger in the positive control.

IV. Conclusion

Based on the research results of the Anti-Acne Cream Formulation of the Ethanol Extract of Katuk Leaves (*Sauropus androgynus* (L.) Merr against *Staphylococcus Epidermis* and *Staphylococcus Aureus*, it can be concluded that:

1. Katuk leaf extract cream shows an inhibitory effect on bacteria.
2. Katuk leaf extract cream was able to inhibit the growth of *S. aureus* and *S. epidermis* bacteria with the best concentration in formula 3, namely the concentration of 20% katuk leaf extract.

V. References

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