Jurnal Kesehatan
Vol. 15, No. 1, April 2024
ISSN 2086-7751 (*Print*), ISSN 2548-5695 (*Online*)
http://ejurnal.poltekkes-tjk.ac.id/index.php/JK

Development of an Anti-Acne Facial Wash Gel with *Cassia alata* L. Leaf Extract

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ARTICLE INFO

Article history

Received date
06 Feb 2024

Revised date
27 Feb 2024

Accepted date
27 Mar 2024

Keywords:

Antibacterial;
Ketepeng Plant (*Cassia alata* L.);
Skin disease.

ABSTRACT

Acne is a skin disease that is often encountered. Acne can be caused by various factors, including bacterial infection. To prevent acne, keep facial skin clean using facial wash. Using facial wash made from synthetic substances can cause more significant side effects than natural ingredients, so research is needed on natural ingredients that can be used as anti-acne agents. One natural ingredient that has antibacterial activity is ketepeng leaf (*Cassia alata* L.). This research aims to formulate a facial wash gel preparation from ketepeng leaf extract and test its antibacterial activity against *Staphylococcus aureus* bacteria. This research is experimental research starting from making extracts, dosage formulation, evaluating the physical properties of the dosage (stability test, organoleptic test, homogeneity test, pH test, viscosity test, spreadability test, and foamability test), skin irritation test, hedonic test and antibacterial activity against *Staphylococcus aureus* bacteria. The research results showed that the inhibition zone produced by formula 1 was 9.73mm, formula 2 was 10.33mm, and formula 3 was 13.18mm. This research concludes that Ketepeng leaf can be used as an active ingredient in anti-acne facial wash gel preparations because the Ketepeng leaf extract facial wash gel preparation meets the requirements for evaluating good physical properties and has antibacterial activity against *Staphylococcus aureus* bacteria in each formula.

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INTRODUCTION

Acne or acne vulgaris is a skin disease that often occurs in adolescents aged 16-19 to adults aged 30. The incidence rate in men is higher than in women, ranging from 95%-100% in men and 83%-85% in women. Acne is not a life-threatening skin disease, but acne can have a psychological effect that will reduce a person's confidence level and affect their quality of life. Acne can also lead to skin scarring, resulting in uneven and pitted skin surfaces (Wardani, 2020).

Skin is the largest organ of the human body, where 15% of the total body weight of adults is formed by skin (Singh et al., 2015). One of the most common skin diseases is acne. Acne is a skin problem that attacks the face (Gunarti, 2018). Acne can be caused by various factors such as genetics, air pollution, diet, skin type, cosmetic use, stress, and bacterial infections (Putri et al., 2022). *Staphylococcus aureus* is a normal flora that can cause skin infections (Juariah et al., 2020). *Staphylococcus aureus* causes acne by clogging the skin's pores (Sarlina et al., 2017). One of the efforts to prevent acne is to maintain facial hygiene by using facial wash.

Facial wash is a facial cleansing soap with a soft and light texture and functions to maintain the cleanliness of facial skin. Facial wash is an alternative used as an anti-acne where its use is more practical, economical, and widely recognized by the public (Nirmala et al., 2021).

The growth of *Staphylococcus aureus* bacteria can be inhibited using synthetic and natural materials. One. One of the natural materials that have antibacterial activity is ketepeng leaf (*Cassia alata* L.). The content of secondary metabolites in ketepeng leaf that have potential as antibacterials are alkaloids, flavonoids, saponins, tannins, and anthraquinones (Sayuti, 2015). Ketepeng leaf are used as anti-acne because they can inhibit the growth of *Propionibacterium acnes, Staphylococcus aureus* and *Staphylococcus epidermidis bacteria*.

The inhibition of ketepeng leaf against *Staphylococcus aureus* bacteria is 28.6mm

(Rezali et al., 2018). These results indicate that the inhibition produced is in the strong category. Some studies also show the results of the inhibition zone produced by ketepeng leaf in the medium category, namely 5.21mm (Hamni et al., 2022) and 7mm (Hastuty et al., 2018). Based on research by Fitriani & Nuryanti (2023), ketepeng leaf extract at a concentration of 5% is able to inhibit *Propionibacterium acnes* and *Staphylococcus epidermidis* bacteria with the resulting inhibition zone of 12.66 mm and 19.66 mm, these results indicate that the inhibition zone produced is in the strong category.

Currently, people are starting to switch to using herbal ingredients rather than using synthetic drugs or ingredients because the side effects caused by synthetic drugs are greater than herbal ingredients (Nirmala et al., 2021). Most acne medications contain synthetic antibiotics such as clindamycin and erythromycin that bind to cell receptors or inhibit enzymes. Acne medications containing these synthetic antibiotics can cause unwanted effects such as irritation, resistance, organ damage, and even immune hypersensitivity (Wardani, 2020). Therefore, the use of natural ingredients as active substances is an alternative to replace synthetic ingredients used to make facial wash preparations.

One of the herbal ingredients that can be used as an anti-acne alternative is ketepeng leaves (*Cassia alata* L.). Therefore, a study was conducted to create an anti-acne facial wash gel formula from ketepeng leaf extract with safe, effective, and stable characteristics that are able to inhibit staphylococcus aureus bacteria.

METHOD

This laboratory experimental study was carried out across various specialized facilities within Sekolah Tinggi Ilmu Kesehatan Harapan Ibu Jambi, including the Pharmaceutical Technology Laboratory, Chemistry Laboratory, Research Laboratory, and Microbiology Laboratory from March to July 2023. The identification of plant material was performed at the Faculty of Pharmacy, Gadjah Mada University, Yogyakarta, and ethical clearance was obtained from Poltekkes Kemenkes Jambi.

This study has been confirmed that the plants used are Senna alata (L.) Roxb or Cassia alata L. Based on a certificate from the Ethics Committee of the Jambi Ministry of Health Polytechnic Health Research Number LB.02.06/2/524/2023, this research is ethically feasible for human testing.

Tools and materials

The study utilized a range of equipment, including a digital scale, blender, sieves, maceration bottles, a rotary evaporator, and other standard laboratory instruments. The materials used comprised ketepeng leaf, various chemical reagents, and *Staphylococcus aureus* bacterial cultures.

Plant extraction

Ketepeng leaf that have been collected are sorted to remove dirt attached to the leaf, then dried in the air for seven days until the leaf are dry which is characterized by constant leaf weight and then mashed with a dry blender (Linda et al., 2011). The dry powder was extracted using the maceration method with 96% ethanol solvent; the sample was stirred occasionally and soaked for 3x24 hours at room temperature. After that, it was filtered so that the filtrate and pulp were obtained (Rezali et al., 2018). The filtrate was concentrated with a rotary evaporator with a temperature of 40-50 ℃ and evaporated using a water bath to obtain a thick extract (Linda et al., 2011).

Formulation of gel facial wash

Table 1. Gel facial wash formula of ketepeng leaf extract

Materials	F1 (%)	F2 (%)	F3 (%)
Ketepeng Leaf Extract	2	4	8
Carbopol	1	1	1
Propilen glikol	15	15	15
Trietanolamine	2	2	2
SLS	2	2	2
DMDM hydantoin	0,1	0,1	0,1
Oleum rosae	0,1	0,1	0,1
Aquadest	ad 100	ad 100	ad 100

All ingredients used were weighed according to the formula. Carbopol was dispersed using hot distilled water while stirring evenly in a mortar. Ketepeng leaf extract was dissolved in distilled water. DMDM hydantoin preservative was dissolved using distilled water. The gel base was mixed with dissolved ketepeng leaf extract, and then dissolved DMDM hydantoin, propylene glycol, and oleum rosae were added. Triethanolamine as a pH controller was added to the mixture and mixed homogeneously. Sodium lauryl sulfate was added with slow stirring in a glass beaker.

Stability evaluation of physical properties of preparations

The stability test was carried out using the freeze-thaw cycling method for three cycles or six days. The gel preparation was stored at 4±2℃ for 24 hours and then removed and stored at 40±2℃ for 24 hours for one cycle. The preparation was observed after each cycle (Dwiastuti & Ardiyati, 2020). The stability test parameters of the physical properties of the preparation include an organoleptic test, homogeneity test, pH test, viscosity test, spreadability test, and foamability test.

1. Organoleptic Test
   Organoleptic evaluation includes color, odor, and form, which are analyzed manually using the eyes and nose (Eugresya et al., 2017).
2. Homogeneity Test
   A 0,1gram sample was placed on an object glass, then observed (Afita et al., 2021). The homogeneity test was carried out to see the uniformity of the preparation. Homogeneous preparations are characterized by the absence of coarse grains on the object glass (Putri, 2021).
3. pH test
   A sample of 1 gram was diluted with 10 ml of distilled water in a beaker glass (Afita et al., 2021). The pH value measurement is carried out using a pH meter, which is dipped into the diluted solution. Record the pH indicated on the device (Bayti et al., 2021).
4. Viscosity Test
   100 grams of gel preparation in a beaker glass was measured using a Brookfield viscometer by setting the speed and spindle to be used (Utami et al., 2019).
5. Spreadability Test
   A sample of 0,5grams is placed on a clear glass, and then a cover glass is placed on the preparation; the first one is not given a load. Next, 50grams, 100grams, and 150grams were given a load, respectively, alternating with an interval of 1 minute, and the diameter of its horizontal and vertical spreadability was measured (Irianto et al., 2020).
6. Foamability Test
   A sample of 1gram in the reaction tube is added to 10ml of distilled water. Then shaken, by flipping the test tube, then the height of the foam formed is measured. Then, the foam height is measured again after 5 minutes (Yuniarsih et al., 2020).

Irritation test

The irritation test method used is the closed patch method, which involves applying 1 gram of facial wash preparation on the inner arm, then covering it with gauze and plaster and leaving it for 30 minutes. Then, observe the conditions that arise on the skin (Wahyuni et al., 2021). Assessment of the degree of irritation is done by giving a score of 0-4, as seen from the severity of erythema and edema on the skin.

Score 0 if no erythema occurs; if erythema occurs very little (diameter <25mm), then score 1; if erythema is clearly visible (diameter 25.1-30mm), then score 2; if moderate erythema (diameter 30.1-35mm) score 3, and if severe erythema occurs (red skin with a diameter >35mm) then score 4. For the assessment of edema reaction, if no edema occurs, it is given a score of 0; if the edema is very little or almost invisible, it is given a score of 1; if there is a clear bordered edge edema with a thickness of <1mm, it is given a score of 2, if the edema that occurs is moderate with an edge rise of ±1mm, it is given a score of 3, if the edema is severe which is marked by an edge rise of >1mm and extends outside the area of application of the preparation on the skin, it is given a score of 4 (Laras et al., 2014).

Hedonic test

The hedonic test was conducted by giving the facial wash formulas (F1, F2, F3) to 9 volunteers. Volunteers gave an assessment on a numerical scale on the prepared assessment sheet. If the volunteer really likes the preparation, it is given a value of 5; if they like the preparation, they are given a value of 4; if they somewhat like it, they are given a value of 3; if they don't like it, they are given a value of 2, and if the volunteer really dislikes the formula, they are given a value of 1 (Kurniasari, 2016).

Antibacterial activity test of preparations

1. Sterilization of tools and materials
   Tools and materials were sterilized using an autoclave for 15 minutes at 12˚C. Ose and tweezers were sterilized by bunsen flame (Khaerati & Ihwan, 2011).
2. Preparation of Nutrient Agar (NA)
   Media NA weighed 2.8g and dissolved in 100ml of distilled water, then heated on a

hotplate. The media was put into several test tubes of 5ml each, then sterilized using an autoclave for 15 minutes at 121℃ (Juariah et al., 2020). The test tube is tilted so that the NA media freezes in an oblique shape (Utomo et al., 2018).

3. Rejuvenation of Test Bacteria

   Staphylococcus aureus bacteria were taken one ose and then rejuvenated by the scratch method on an inclined NA medium, then incubated for 24 hours at 37˚C (Yusriana et al., 2014). Colonies formed indicate bacterial growth and are ready to be used for further tests (Utomo et al., 2018).
4. Preparation of Muller Hinton Agar (MHA)

   Media MHA of as much as 38grams was dissolved with 1 liter of distilled water and then heated. Then, the media was sterilized using an autoclave for 15 minutes at 121℃. After sterilization, the media was poured into sterile Petri dishes and allowed to stand at room temperature until the media solidified (Utomo et al., 2018).
5. Preparation of Test Bacteria Suspension

   Bacterial colonies of 1 ose of NA media were diluted using a sterile 0.9% NaCl solution (Utomo et al., 2018). Then, the turbidity of the bacterial suspension was measured using UV-Vis spectrophotometry with a wavelength of 580nm until a transmittance value of 25% was obtained (Handayani, 2016).
6. Antibacterial Activity Test

   A sterile cotton swab is dipped into the bacterial suspension that has been standardized for turbidity until it sinks into the cotton swab. The cotton swab was removed by pressing it against the tube wall. The cotton swab is streaked on solid MHA media by rotating the petri dish until it is evenly distributed over the surface of the media. Allow 5-15 minutes for the suspension to dry or soak into the media (Juariah et al., 2020).

   Paper discs (6mm diameter) that have been soaked in facial wash gel preparations in each formula marketed facial wash preparations as positive controls and facial wash gel bases as negative controls for 15 minutes are aseptically placed on the surface of MHA media containing test bacteria, then incubated for 24 hours at 37℃. Furthermore, the diameter of the growth inhibition of the test bacteria was measured, which was indicated by the formation of a clear zone around the disc paper (Utomo et al., 2018).

RESULTS

Plant extraction

Fresh ketepeng (*Cassia alata* L.) leaves that have been cleaned as much as 4 kg are dried and then mashed so that dry simplisia is obtained in as much as 1,2 kg, dry simplisia is extracted by maceration method using 96% ethanol solvent to obtain liquid extract with a distinctive odor and blackish green color, then the liquid extract is concentrated using a rotary evaporator and water bath so that a thick extract of 161,28g is obtained.

Table 2. Extract yield results

Sample	Weight (g)	Yield of extract (%)
Fresh ketepeng leaves	4,000	
Dried symplisia	1,200	13.44
Thickened extract	161.28	

The maceration results were then concentrated, and a thick extract of ketepeng leaf was obtained with a yield value of 13.44%.

Formulation of gel facial wash preparation

Gel facial wash preparation of ketepeng leaf extract was made in 3 formulas, namely formula 1 (2% ketepeng leaf extract facial wash), formula 2 (4% ketepeng leaf extract facial wash), and formula 3 (8% ketepeng leaf extract facial wash).

Figure 1. Ketepeng leaf extract gel facial wash

Table 3. Stability evaluation of physical properties of preparations

Evaluation	Parameter	Formula 1	Formula 2	Formula 3
Organoleptic	Cycle 0	Form: viscous Color: brownish yellow Odor: oleum rosae	Form: viscous Colour: Brown Odor: oleum rosae	Form: viscous Color: blackish brown Odor: oleum rosae & extract
	Cycle 1	Form: viscous Color: brownish yellow Odor: oleum rosae	Form: viscous Colour: Brown Odor: oleum rosae	Form: viscous Color: blackish brown Odor: oleum rosae & extract
	Cycle 2	Form: viscous Color: brownish yellow Odor: oleum rosae	Form: viscous Colour: Brown Odor: oleum rosae	Form: viscous Color: blackish brown Odor: oleum rosae & extract
	Cycle 3	Form: viscous Color: brownish yellow Odor: oleum rosae	Form: viscous Colour: Brown Odor: oleum rosae	Form: viscous Color: blackish brown Odor: oleum rosae & extract
Homogeneity	Cycle 0	Homogen	Homogen	Homogen
	Cycle 1	Homogen	Homogen	Homogen
	Cycle 2	Homogen	Homogen	Homogen
	Cycle 3	Homogen	Homogen	Homogen
pH	Cycle 0	5,9	5,77	5,47
	Cycle 1	5,9	5,83	5,53
	Cycle 2	5,87	5,83	5,43
	Cycle 3	5,87	5,8	5,43
Viscosity	Cycle 0	3470,67 cp	3343,00 cp	3233,00 cp
	Cycle 1	3427,00 cp	3223,00 cp	2489,67 cp
	Cycle 2	3424,33 cp	3117,00 cp	2476,33 cp
	Cycle 3	3393,00 cp	3109,00 cp	2453,00 cp
Spreadability	Cycle 0	5,67cm	5,73cm	5,83cm
	Cycle 1	5,70cm	5,87cm	6,08cm
	Cycle 2	5,73cm	5,88cm	6,10cm
	Cycle 3	5,88cm	5,90cm	6,15cm
Foam stability	Cycle 0	90,70%	91,22%	92,24%
	Cycle 1	89,08%	90,51%	91,91%
	Cycle 2	88,13%	89,99%	90,97%
	Cycle 3	87,92%	88,76%	89,84%

Table 4. Irritation test results

Respondents	Reaction	F1	F2	F3
1	Erythema	0	0	0
	Edema	0	0	0
2	Erythema	0	0	0
	Edema	0	0	0
3	Erythema	0	0	0
	Edema	0	0	0
4	Erythema	0	0	0
	Edema	0	0	0
5	Erythema	0	0	0
	Edema	0	0	0
6	Erythema	0	0	0
	Edema	0	0	0
7	Erythema	0	0	0
	Edema	0	0	0
8	Erythema	0	0	0
	Edema	0	0	0
9	Erythema	0	0	0
	Edema	0	0	0

Assessment of the degree of irritation is done by giving a score of 0-4, as seen from the severity of erythema and edema on the skin. The results of the irritation test conducted on nine students of STIKES Harapan Ibu Jambi showed that there was no erythema response (skin redness and itching) and edema on the skin of volunteers for all formulas (F1, F2, and F3). Testing was carried out on the inner arm using the closed patch method to avoid contamination from microorganisms that risk interfering with the results of the irritation test.

Table 5. Hedonic test results

F	Parameters	Number of respondents				
		Sts	Ts	As	S	Ss
F1	Color	0	1	2	6	0
	Odor	0	0	0	4	5
	Form	0	0	1	3	5
F2	Color	0	2	5	2	0
	odor	0	0	1	4	4
	Form	0	0	4	2	3
F3	Color	0	2	5	2	0
	odor	0	0	2	3	4
	Form	0	0	4	2	3

Description:
Sts: Strongly Dislike; Ts: Dislike; As: Somewhat Like; S: Like; Ss: Liked Very Much

From the results of the hedonic test that was carried out on formula 1, formula 2, and formula 3, it was found that the formula that had the highest value in the hedonic test was formula 1.

Antibacterial activity test of preparations

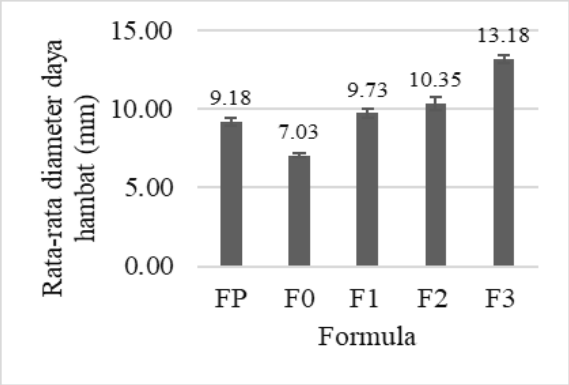

Figure 2. Antibacterial activity test results of facial wash gel preparations

The results of the antibacterial activity test of ketepeng leaf extract facial wash preparations increased in each preparation group, between formula 1 (2% ketepeng leaf extract facial wash), formula 2 (4% ketepeng leaf extract facial wash), and formula 3 (8% ketepeng leaf extract facial wash).

In the antibacterial activity test, the facial wash preparation of ketepeng leaf extract showed the inhibition zone of FP (market facial wash) of 9.18±0.27mm, F0 (Base) of 7.03±0.12mm, F1 (facial wash 2% extract) of 9.73±0.29mm, F2 (facial wash 4% extract) of 10.35±0.39mm and F3 (facial wash 8% extract) of 13.18±0.27mm.

DISCUSSION

This study used ketepeng plants, where ketepeng plants are obtained from Mandiangin District, Sarolangun Regency, Jambi. Ketepeng leaves are used because they have various benefits, one of which is as an antibacterial (Nurlansi & Jahidin, 2018). Ketepeng plant samples were determined which aims to ensure that the plants used are true species of *Cassia alata* L. Based on the determination results, it was found that the sample was *Senna alata* (L) Roxb. or *Cassia alata* L. which belongs to the Fabaceae family.

Ketepeng leaf symplisia was extracted using maceration method with 96% ethanol solvent. The maceration method was chosen because it is simple, fast, relatively low cost, and can avoid damage to secondary metabolite compounds that are not heat resistant (Sayuti, 2015). The use of ethanol solvent is because ethanol is a polar solvent with a high level of polarity, more volatile, can prevent the growth of mold and bacteria, and the temperature required for concentration is lower so as to minimize the risk of shrinkage of active compounds. Flavonoids, saponins, tannins, and some alkaloids are polar compounds that can be extracted using polar solvents. Therefore, ethanol solvent is very effective in the extraction process of ketepeng leaf because the compounds contained in ketepeng leaf that have antibacterial activity are polar (Sumiati et al., 2021).

The maceration results were then concentrated and obtained a thick extract of ketepeng leaf with a yield value of 13,44%, which shows that this value is in accordance with the Herbal Pharmacopoeia Indonesia Edition II standard, which is for plants of the Fabaceae family to have a yield of not less than 8.1% (Ministry of Health RI, 2017).

Ketepeng leaf extract is then formulated for facial wash preparation. Facial wash is a mild and gentle facial cleansing soap that maintains skin cleanliness. Facial wash is an anti-acne alternative that is practical and economical and has been widely recognized by the public (Nirmala et al., 2021). The ketepeng leaf extract facial wash was then evaluated for the stability of the physical properties of the preparation, irritation test, hedonic test and antibacterial activity test.

The stability test aims to determine whether the preparation remains stable at a certain temperature. The results of the stability test for organoleptic and homogeneity tests showed that there were no significant changes in all formulas, both changes in color, odor, form, and homogeneity. For the pH stability test, there was a decrease in pH in each formula. The decrease in pH of the preparation is due to extreme temperature changes that occur and because the extracts used are acidic (Falahi et al., 2020). The viscosity stability test showed a decrease in the viscosity of the preparation in each cycle. This is in line with the research of Falahi et al. (2020); the results of the viscosity test after freeze-thaw decreased; this is due to the influence of high temperatures, which can make the decrease in viscosity permanent so that it makes the viscosity decrease. In addition, the decrease in viscosity during storage can be caused by packaging that is less airtight so that the gel absorbs water vapor from outside and increases the amount of water in the gel (Sayuti, 2015).

Interestingly, the decreased viscosity positively influenced spreadability, while foam stability diminished, likely due to pH-dependent soap-lathering characteristics (Setiawati & Ariani, 2020). Despite these changes, the physical stability within the tested parameters remained within acceptable ranges, suggesting a robust formulation capable of withstanding variable storage conditions.

Based on the observations of the irritation test that has been carried out, it can be concluded that the ketepeng leaf extract facial wash preparation does not cause irritation to the skin. This is in line with the research of Rifkowaty & Fitriarni (2020), which states that soap preparations with active ingredients of keeping leaf extract do not cause irritation.

The hedonic test conducted on respondents aims to see which formula is good and accepted by consumers, while the parameters assessed from the hedonic test include the color, odor, and form of the preparation. From the results of the hedonic test conducted on formula 1, formula 2, and formula 3, it was found that the formula that had the highest value in the hedonic test was formula 1. This is because formula two and formula 3 have a more intense color that is less preferred by respondents.

Notably, the antibacterial tests delineated a dose-dependent increase in efficacy, with Formula 3 exhibiting the most substantial inhibition zone. This trend affirms the hypothesis that higher extract concentrations yield greater antibacterial action. The base formula's inherent antibacterial properties, attributed to DMDM hydantoin and propylene glycol, provide a foundational antimicrobial effect, which is enhanced by the addition of ketepeng extract (Tsabitah et al., 2020).

The secondary metabolites in ketepeng leaves, such as alkaloids, flavonoids, saponins, and triterpenoids/steroids, have been implicated in the disruption of bacterial cell walls and membranes, inhibition of cell wall synthesis, and porin interaction (Egra et al., 2019). These modes of action reinforce the potent antibacterial nature of the ketepeng leaf extract, validating its use in higher concentrations for more pronounced effects.

The ketepeng leaf extract's integration into facial wash formulations not only meets the physical stability requirements but also exhibits a promising antibacterial capacity.

CONCLUSION

Ketepeng (Cassia alata L.) leaf extract can be utilised as an active ingredient in anti-acne facial wash gel preparation. The three formulas showed good physical properties evaluation results and fulfilled the requirements. Of the three formulas, F3 has the greatest antibacterial activity against Staphylococcus aureus bacteria, which is 13.18±0.27mm, which shows a strong category inhibition response.

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