Antibacterial Activity Of Clay Mask From *Houttuynia Cordata* Leaves Extract Against *Staphylococcus Aureus*

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Abstract

Acne, a common skin problem, often causes discomfort and concern for many people. *Staphylococcus aureus* is a bacterium responsible for causing acne. A clay mask is composed of clay minerals, specifically bentonite and kaolin. Clay masks have anti-acne properties as they absorb excess oil from the skin. Excessive oil production is the primary cause of acne. This study aimed to determine the antibacterial activity of clay masks from *Houttuynia cordata* (HC) against *Staphylococcus aureus*. The HC extract of the clay mask was obtained using maceration 96% ethanol solvent. The pour plate diffusion technique evaluated the antibacterial activity at concentrations of 10%, 20%, and 30%. The antibacterial activity will be assessed by measuring the diameter of the clean zone around the well using a caliper. The antibacterial activity of clay mask HC at concentrations of 10%, 20%, and 30% are 1,612±0,040 cm, 1,706±0,046 cm, and 1,806±0,039 cm, respectively. The physical parameters of the clay mask have successfully satisfied the specified criteria. The study determined that the clay mask from HC has antibacterial properties and is the most effective clay mask HC at a 30% concentration.

Keywords: h*outtuynia cordata*, clay mask, s*taphylococcus aureus,* antibacterial

1. INTRODUCTION

The skin is a protective barrier against external physical and chemical factors and significantly affects a person's appearance (Retnaningsih et al., 2019). Acne or acne vulgaris is a skin disease due to chronic pilosebaceous gland inflammation characterized by blackheads, papules, pustules, and nodules (Agustini et al., 2021).

This study aims to determine the antibacterial activity of *Houttuynia cordata* Thunb (HC) extract clay mask against *Staphylococcus aureus* and the physical characteristics of the clay mask. *Staphylococcus aureus* is the bacteria responsible for causing acne. Medicinal plants are selected as an option to reduce the adverse effects of synthetic medication use (Abna, 2017). Natural products from plants have the potential to be developed as traditional medicine is HC. HC in Indonesia is known as amis-amisan leaf. HC leaves contain secondary metabolites such as flavonoids, alkaloids, and essential oils (Miyata et al., 2010). According to (Fu et al., 2013), HC of ethanol extract has antibacterial activity against *Staphylococcus aureus*.

A face mask is made from clay, such as kaolin or bentonite. It is suitable for those with oily faces because this clay mask can absorb excess oil levels on the face (Dian et al., 2022). This research is essential because it is a new study that uses the clay mask from HC for anti-acne therapy against *Staphylococcus aureus*.

2. METHODS

The sample used in this study was HC obtained from Gunungpati, Semarang. The sampling technique used in this study is simple random sampling.

MATERIALS

The materials used in the research are Houttuynia cordata, glassware, spectrophotometers UV-Vis, ethanol, chloroform, methanol, water, *Staphylococcus aureus*, Mannitol Salt Agar, Nutrient Broth, and Nutrient Agar.

EXTRACTION

HC extract was made by re-maceration using a 96% ethanol solvent. Two hundred grams of HC powder are put into a glass jar with 96% ethanol solvent as much as 2000 mL (1:10). Extraction is carried out for 3x24 hours, and the filtrate is separated. Filtrate is collected and evaporated using a water bath until a viscous extract is obtained.

ANTIBACTERIAL ACTIVITY

They are planting *Staphylococcus aureus* on Nutrient Agar (NA), incubated for 24 hours at 37°C in an incubator. They suspended *Staphylococcus aureus*, which is put into Nutrient Broth (NB) media at 37°C for 24 hours (Rosmania and Yanti, 2020). The antibacterial activity is done by diluting sterile Mannitol Salt Agar (MSA) and then measuring 10 mL in a sterile petri dish. After solidifying, insert the cylinder cup into the substrate. Measured 15 mL of sterile MSA media, then mixed with 0.5 mL suspension of homogenized *Staphylococcus aureus*. The mixture is put into a petri dish as a second layer and waited for it to solidify. After solidifying, the cylinder cup is taken.

Each sample with a predetermined concentration was inserted into a well. glycerin (phase 1). there is a different container. Sodium lauryl sulfate was poured and crushed slowly until a homogeneous paste was formed (phase 2). Phases 1 and 2 were combined and homogenized until a clay mask base paste

Positive and negative controls are inserted with the same amount, then incubated for 1x24 hours at 37ºC. Observe bacterial growth and measure the diameter of the clear zone formed using a caliper (Ngajow et al., 2013).

Table 1. Formula of Clay Mask HC extract

Material	F 0 (0%)	F I (10%)	F II (20%)	F III (30%)
HC extract		10 %	20 %	30 %
Bentonite	8 %	8 %	8 %	8 %
Xanthan Gum	1 %	1 %	1 %	1 %
Kaolin	30 %	30 %	30 %	30 %
Glycerine	5 %	5 %	5 %	5 %
Sodium Lauryl Sulphate	0,5 %	0,5 %	0,5 %	0,5 %
Titanium hydroxide	2 %	2 %	2 %	2 %
Water	Added 50 grams	Added 50 grams	Added 50 grams	Added 50 grams

F 0: Base clay mask
F I: Formula clay mask 10% HC leaf extract
F II: Formula clay mask 20% HC leaf extract
F III: Formula clay mask 30% HC leaf extract

A 50g clay mask as much as 27 ml of water was poured into a mortar, and bentonite was added. Bentonite is allowed to moisten, added to xanthan gum, and crushed quickly until the whole gum dissolves. Kaolin was added gradually into the mortar while homogenizing and adding TiO2 and

formed. HC extract concentration (10%, as much as 5 grams; 20%, as much as 10 grams; and 30%, as much as 15 grams) is homogeneous.

Table 2. Phytochemical Screening of HC extract

Compound	Identification	Reference	Result		Conclusion	
			Powder	Extract	Powder	Extract
Flavonoids	Powder/extract+ powder Mg+ HCl 2N + amyl alcohol	Red/yellow/orange color formed on the amyl alcohol layer	An orange color formed on the amyl alcohol layer	An orange color forms on the amyl alcohol layer	+	+
Alkaloids	Powder/extract + Mayer Reagent	Formed yellowish-white precipitate	The white precipitate is formed	The white precipitate is formed	+	+
	Powders/extracts+ dragendorf reagents	Brick-red deposits are formed	Brick-red deposits are formed	Brick-red deposits are formed	+	+
	Bouchardat powder/extract + reagent	Blackish-brown deposits are formed	Blackish-brown deposits are formed	Blackish-brown deposits are formed	+	+
Saponins	Powder/extract + water shaken vigorously until it arises foam. Then plus HCl 2N	Formed stable foam after dripping HCl 2N	Formed stable foam after dripping HCl 2N	Formed stable foam after dripping HCl 2N	+	+
Tannins	Powder/extract+ $FeCl_3$ 1%	A blackish-green or blue-black color is formed	Formed blackish-green color	Formed blackish-green color	+	+
	Extract + Gelatin solution 10% + NaCl 1%	A white precipitate is formed	A white precipitate is formed	A white precipitate is formed	+	+
Triterpenoid	Powder/extract + n-hexane + acetic acid + H2SO4	Formed green color (steroids), formed red color (triterpenoids)	Formed red color	Formed red color	+	+

3. RESULTS AND DISCUSSION

The study experiment was conducted to observe and determine the inhibitory activity of clay masks from HC extract against *Staphylococcus aureus*. Viscous extract of HC btained 57.069 grams, obtaining a yield of 28.534%. The results of phytochemical screening and TLC can be seen in Tables 2 and 3.

Phytochemical screening results indicate that HC contains active chemicals such as flavonoids, alkaloids, saponins, tannins, and steroids/triterpenoids. The confirmation through TLC found that HC extract contains these compounds.

Table 3. TLC of HC extract

Compound	Reagent	Reference	Result	Conclusion	Rf value
Flavonoids	Ammonia vapors	Yellow or brownish-yellow	Yellow	+	0,912 0,537
Alkaloids	Dragendorf	Brown	Brown	+	0,6
Saponins	Anisaldehid-heated sulfuric acid	Yellow, green, red, dark blue, purple, and brownish-yellow	Brownish-yellow	+	0,362 0,675
Tannins	FeCl3 5%	Blackish green	Blackish green	+	0,187 0,350 0,437 0,612 0,850 0,937
Steroids/Triterpenoids	Anisaldehid-heated sulfuric acid	Red	Red	+	0,275 0,537

Table 4. Antibacterial Activity of Clay Mask HC extract

Replication	Antibacterial activity (cm)				
	Concentration				
	10%	20%	30%	Control (+)	Control(-)
1	1,655	1,743	1,848	0,000	0,000
2	1,581	1,667	1,775	0,000	0,000
3	1,567	1,667	1,765	0,000	0,000
4	1,590	1,673	1,783	0,000	0,000
5	1,667	1,780	1,860	0,000	0,000
Average ± SD	1.612 ± 0.040	1.706 ± 0.046	1.806 ± 0.039	0,000	0,000
Category	Strong	Strong	Strong	inhibit	inhibit

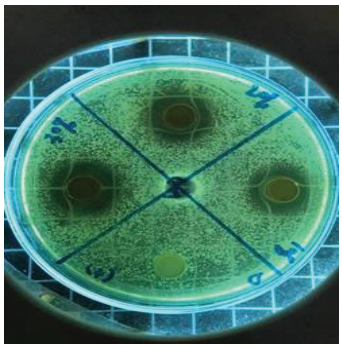

Figure 1. Antibacterial activity of Clay Mask HC extract

Antibacterial activity used the diffusion and pour plate techniques, HC extract concentrations of 10%, 20%, and 30%. The positive control used was the clay mask published in the market, and the negative control was the clay mask dosage base, and five times replication was carried out. The presence of a clear zone indicates antibacterial inhibitory activity.

The results of formulas, positive and negative control, have different inhibition categories. Formula 1 is the category of strong with 1,612 cm. Formula 2 is a category of strong inhibition with 1,706 cm. Formula 3 has a strong inhibitory activity of 1,806 cm, while the positive control and negative are in the non-inhibiting category, which has a value of 0.000 cm. The positive control does not have antibacterial activity or ingredients that have antibacterial compounds because the clay mask is used for care, not treatment.

Extract with a concentration of 30% producedthe most excellent antibacterial activity. The higher the concentration of the extract HC, the greater the inhibitory activity produced. The results of formula 10%, formula 20%, formula 30%, and positive control show significant antibacterial activity. The greater the concentration of the extract, the greater the antibacterial activity it has.

Table 5. Characteristics of Clay Mask H

No	Activity	F 1	F II	F III	Conclusion
1.	Organoleptic	Semi-solid	Semi-solid	Semi-solid	√
	Color	Green	Green	Green	
	Smell	Typical Extracts	Typical Extracts	Typical Extracts	
2.	Homogeneity	Homogeneous	Homogeneous	Homogeneous	√
3.	pH	6,668	5,408	4,706	√
4.	Dispersion	5,210	5,342	5,508	√
5.	Adhesion	5 seconds	6 seconds	7 seconds	√
6.	Irritation	No symptoms of irritation	No symptoms of irritation	No symptoms of irritation	√
7.	Viscosity	11.576	10.806	9.154	√

C extract

The physical characteristics of all formulas meet the requirements of homogeneity, pH, dispersion, adhesion, irritation, and viscosity. Suggestions for further research on the antibacterial activity of HC extract on different bacteria or fungi, extraction solvents, and clay mask HC made in other dosage forms.

4. CONCLUSION

The clay mask extract HC has antibacterial activity against *Staphylococcus aureus*. Clay mask of HC has antibacterial activity and is the best formula at a concentration of 30%.

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