

# Antibacterial activity of *Allium sativum* L. cream formulation against *Propionibacterium acnes* and *Staphylococcus epidermidis*

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## ABSTRACT

*Allium sativum* L. is one of the plants that has been widely known by the public for a long time and is used as a traditional medicine, one of them as antiacne causes *Propionibacterium acnes* and *Staphylococcus epidermidis* <sup>[1,2]</sup>. *Allium sativum* L. contains *Allicin* that may help in inhibits gram-negative or gram-positive bacteria. This study aimed to determine the activity of *Allium sativum* L. cream formulation with various concentration (25%,35%,45%) against *Propionibacterium acnes* and *Staphylococcus epidermidis*. Antibacterial activity was determined by the diameter of the zone of inhibition. Diameter of the zone of inhibition calculated as the difference in diameter of the observed zones and the paper discs.

The data of diameter of the zone of inhibition was analyzed with one-way Anova Test. The results showed that *Allium sativum* L. with all extract concentration had the same effect as negative control in inhibits *Propionibacterium acnes* and *Staphylococcus epidermidis* ( $p>0,05$ ).

**Keywords:** *Allium sativum*, acnes, epidermidis, cream

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## INTRODUCTION

Healthy facial skin is identical to facial skin free from skin problems like acne. This acne generally occurs at the age of 14-17 years in women, and 16-19 years in men. About 85% of teenagers are known to have experienced acne problems <sup>[3]</sup>. Factors that cause acne include internal factors (race, genetic, hormonal), as well as external factors (season, food, stress). In addition to these factors, bacteria also become one of the causes of *Propionibacterium acnes* and *Staphylococcus epidermidis* <sup>[4]</sup>. One that can cope with acne is garlic (*Allium sativum* L). *Allium sativum* L has active compounds such as sativine, allicin, allyl sulfide, allyl propyl disulphide, allyl vinyl sulphoxide, allistatin, garlicin, and alkyl thiodulphinate <sup>[5]</sup>. Niyomkam (2010) states that *Allium sativum* L. extract is able to inhibit the growth of *Propionibacterium acnes* bacteria with a minimum inhibitory concentration of 312.0 µg / mL <sup>[1]</sup>. Furthermore, Hindi (2013) mentions *Allium sativum* L. extract can inhibit the growth of *Staphylococcus epidermidis* bacteria by 28 mm with a concentration of 50% <sup>[2]</sup>. Traditional materials that have been proven activity, for easy use, it is necessary to develop dosage. Cream preparations are transdermal preparations that are widely used for the treatment of the skin, because it can deliver active substances properly. This cream preparation needs to be tested for the activity of bacteria *Staphylococcus epidermidis* and *Propionibacterium acnes* to prove that this preparation can serve as anti acne.

## MATERIALS AND METHODS

### Materials

Bawang putih, *S.epidermidis*, *P.acnes*, *Muller Hinton Agar*, aquades, asam stearate, vaseline album, trietanolamin, propilenglikol, cera album, metil paraben, pewangi lemon, etanol 70%, viscometer Brookfield LV, *rotary evaporator*, pH *stick universal*, *Laminar Air Flow* (Mascotte Model LV-S) dan autoklaf (UL Model 25x2).

### Determination of Plants

Plants was obtained in Mersi Village, East Purwokerto, Banyumas are determined to find out the true identity of the plant being tested. Determination was done at the Laboratory of Plant Taxonomy, Faculty of Biology, University of Jenderal Soedirman.

### Extract Preparation

*Allium sativum* L are cut into pieces and dried to dry in a room that is not exposed to sunlight. The dried samples were then weighed again and blended. 100 grams of simplicia powder was soaked using 70% ethanol for 24 hours with occasional stirring. The residue and filtrate are separated by using filter paper. Conducted remaseration by adding 1000 ml ethanol with occasional stirring. The overall filtrate obtained was evaporated using an evaporator, then evaporated using oven at a temperature of 40°C to obtain a concentrated extract <sup>[6]</sup>.

## Ointment Formulation and Evaluation

Table 1. Formula of Ointment [7]

Material	Formulation (g/ml)			
	FI 5%	FII 10%	FIII 20%	NC (-)
Extract	12,5	17,5	22,5	-
Cera Alba	1	1	1	1
Stearic Acid	10	10	10	10
Vaselin Album	4	4	4	4
Triethanolamine	0.75	0.75	0.75	0.75
Propylene Glycol	4	4	4	4
Methyl Paraben	0.1	0.1	0.1	0.1
Aquades	ad 50	ad 50	ad 50	ad 50
Fragrance	q.s	q.s	q.s	q.s

Note : X brand ointment as a positive control

Ointment formulation was done, then it was evaluated such as organoleptic, viscosity, pH, disperse, and adhesiveness test. Organoleptic test were conducted by observing ointments from its form, smell, and colour [8]. And then, viscosity was measured using viscometer Brookfield, pH test uses pH stick in order to know the acidity of ointment. Furthermore, disperse test was done by method of 0.5 g ointment is placed on a round glass (d: 15 cm), other glass is placed on it and left for 1 minute. The diameter of the ointment was measured, after which 100g of load was added and then measured a constant diameter. While for the adhesiveness test was done by method of 0.5 g ointment on top of object glass and other object glass placed on the ointment. Then pressed with a load of 1 kg for 5 minutes. Mounted object glass on the test equipment, load as much as 80 g then released and recorded the time until the second glass of this object is released [9].

### Antibacterial Activity Test

The bacterial suspension was taken as much as 0.1 ml and then spread evenly on the surface of Muller Hinton Agar (MHA) medium. The disc paper is immersed in each cream formulation for 10-15 minutes. After that, the disc paper was placed in the middle of the MHA medium and incubated for 24 hours at 37 °C. Observed and measured its inhibit zone.

### Data Analysis

The results of this study will be analyzed descriptively and statistically. Descriptive analysis was used on organoleptic and pH, while the data of viscosity, disperse and adhesiveness was analyzed statistically using Anova if it fulfilled normality and homogeneity of data requirement.

## RESULT AND DISCUSSION

### Organoleptic Tests

Organoleptic tests were conducted by observing ointments from its form, smell, and colour.

Table 2. Organoleptic Test Result

Formulation	Form	Smell	Color
FI	semi-solid	spesific	brown
FII	semi-solid	spesific	brown
FIII	semi-solid	spesific	brown
NC	semi-solid	none	white

Notes :

FI : ointment with 25% extract concentration  
 FII : ointment with 35% extract concentration  
 FIII : ointment with 45% extract concentration  
 NC : ointment base as a negative control

### Viscosity Test

The research result showed that viscosity value is around 2000-11600 cps. This indicates that it qualifies a good cream (2000-50000 cps) [10]. Viscosity value in this research is influenced by concentration of extract and storage time ( $p < 0,05$ ). The addition of the extract can decrease the viscosity, while the longer the cream is stored then the viscosity increases.

### pH Test

The ointment's pH in this research (Table 3) has met the requirement, i.e. having pH corresponding to skin's physiological pH of about 4.5-6.5 [11]. Excessively low pH values can cause irritation, while too high pH values can cause scaly skin [9].

Table 3. pH Test Result

Formulation	pH	
	Day 0	Day 30
FI	6	6
FII	6	6
FIII	6	6
NC	6,5	6

Notes :

FI : ointment with 25% extract concentration  
 FII : ointment with 35% extract concentration  
 FIII : ointment with 45% extract concentration  
 NC : ointment base as a negative control

### Disperse Test

Disperse test was conducted to determine the ability of ointment's diffusion on the skin. The easier the ointment is flattened on the skin, the greater the absorption of its active substances. Based on the table 4, showed that FI, FII, and FIII have not met the requirement a good dispersing power (5.6 - 6.4 cm) [12]. The addition of the extract can decrease the disperse of cream ( $p < 0,05$ ).

Table 4. Disperse Tests' Average Result

Formulation	Dispersing power's average (cm)± SD
FI	4,6 ± 0,23
FII	4,1 ± 0,1
FIII	3,9 ± 0,1
NC	5,8 ± 0,23

Notes :

FI : ointment with 25% extract concentration  
 FII : ointment with 35% extract concentration  
 FIII : ointment with 45% extract concentration  
 NC : ointment base as a negative control

### Adhesiveness Test

Adhesiveness test was to determine the ointment's ability to persist longer on the skin. A good

adhesiveness is not less than 4 seconds<sup>[13]</sup>. Adhesiveness tests result showed that the adhesiveness of FI, FII and negative control was poor because it was less than 4 seconds. The addition of the extract can increase the adhesiveness of cream ( $p < 0,05$ ).

Table 5. Adhesiveness's Average Result

Formulation	Adhesiveness (seconds)
FI	2,71 ± 0,25
FII	3,32 ± 0,29
FIII	4,88 ± 0,63
NC	1,82 ± 0,27

Notes :

FI : ointment with 25% extract concentration  
 FII : ointment with 35% extract concentration  
 FIII : ointment with 45% extract concentration  
 NC : ointment base as a negative control

### Antibacterial Activity Test

Table 6. Result of Antibacterial Activity

Formulation	Inhibition Zone Diameter (mm)	
	<i>S epidermidis</i>	<i>P acnes</i>
NC	0	0
PC	33,83 ± 2,09	22,16 ± 2,08
FI	0	0
FII	0	0
FIII	0	0

Notes :

FI : ointment with 25% extract concentration  
 FII : ointment with 35% extract concentration  
 FIII : ointment with 45% extract concentration  
 NC : ointment base as a negative control  
 PC : X brand ointment available on the market

Based on the table 6 showed that positive control have antibacterial activity against the *Staphylococcus epidermidis* and *Propionibacterium acnes* with each inhibition zone diameter are 33,83 mm and 22,16 mm. While, FI, FII and FIII does not have antibacterial activity against the *Staphylococcus epidermidis* and *Propionibacterium acnes*. Allicin has properties that are less stable at room temperature so it can turn into other compounds that have a smaller potential or even no potential at all<sup>[14]</sup>.

### CONCLUSION

*Allium sativum* L. cream does not have antibacterial activity against the *Staphylococcus epidermidis* and *Propionibacterium acnes*.

### REFERENCES

[1] Niyomkam, P., et al. Antibacterial Activity of Thai Herbal Extract on acne involved Microorganism. *Pharmaceutical Biology*, 2010, vol 48. No 4, h.375-380.  
 [2] Hindi, Nada Khazalkadhim. In Vitro Antibacterial Activity of Aquatic Garlic Extract, Apple Vinegar and Apple Vinegar-Garlic Extract Combination. *America Journal of Phytomedicine and Clinical Therapeutics*, 2013, Vol.1, No.1.  
 [3] Dlugosz, 2013, Quick Referral Drugs For Practical Use, BPharm, EGC, Jakarta  
 [4] Djuanda. A., 2011, Skin Diseases and Sex, Publisher Faculty of Medicine University of Indonesia, Jakarta

[5] Puspitasari, I., 2008, Activity of Garlic Extract (*Allium sativum* L) Against *Staphylococcus aureus* In Vitro [Thesis]  
 [6] Mercy et al, 2014. *Anti-dermatophytic Activity of Garlic (Allium sativum L) Extract on some Dermatophytic Fungi*. Switzerland, Vol.24  
 [7] Kusumawardah, A., 2012, Cream of Rosella Flower Extract (*Hibiscus sabdariffa* Linn) : Test of Physical Properties and Antibacterial Activity of *Staphylococcus epidermidis* [Thesis].  
 [8] Budiman, M.H., 2008, Physical Stability Test of Antioxidant Cream of Tomato Powder Extract [Thesis], FMIPA UI, Jakarta  
 [9] Izzati, U., 2015, *Effectiveness of Burn Wound Healing of Ointment of Senggani Leaf (Melastoma malabathricum L) Ethanol Extract On Male Rats (Rattus norvegicus) of Wistar Strain*, Pontianak, Fakultas Kedokteran, Universitas Tanjungpura.  
 [10] [SNI] Indonesian National Standard, 1996, *Sunscreen*, National Standardization Agency, Jakarta  
 [11] Depkes RI, 1995, *Pharmacopoeia of Indonesia, Edition IV*, Departemen Kesehatan Republik Indonesia, Jakarta  
 [12] Rajalakshmi, G.NI., 2009, *Formulation and Evaluation of Clotrimazole and Ictammo ointment*, International Journal Of Pharma and Bioscience 4 : 10-12.  
 [13] Ulaen, S. P. J., Banne, Y., and Suatan, R. A., 2012, *Making Antiacne Ointment from Temulawak Rhizomes Extract ( Curcuma Xanthorrhiza Roxb.)*, Journal of Pharmaceutical Sciences Volume 3 (2), Page 47-48  
 [14] Salima, J., 2015, *Antibacterial Activity of Garlic (Allium sativum L.)*, J Majority Vol 4 (2), Page 30-39