Available online on 15.08.2020 at http://ajprd.com

Asian Journal of Pharmaceutical Research and Development

Open Access to Pharmaceutical and Medical Research

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Research Article

The Comparison of Antibacterial Activities of Soursop Leaf (*Annona muricata* L.) and Basil Leaf (*Ocimum americanum* L.) Ethanolic Extracts on Gel Formulated Against *Staphilococcus aureus* and *Propionibacterium acnes*

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ABSTRACT

Objectives: The purpose of this study was to determine antibacterial activity of Soursop leaf (*Annona muricata* L.) and Basil leaf (*Ocimum americanum* L.) ethanolic extract on gel formulated againts *Staphilococcus aureus* and *Propionibacterium acnes*.

Design: This study uses an experimental laboratory design. This research uses difusion method, the inhibitory comparison between positive and negative control performed. This study compared the antibacterial activity of two gels that have different active ingredients to the growth of skin bacteria, namely *Staphilococcus aureus* and *Propionibacterium acnes*.

Interventions: The sample used was two types of gels each containing Soursop leaf and basil leaf ethanol extract that in various concentration of 5, 10 and 15%. As a comparison, Clindamycin gel concentration 1% was used. The distilled water was used as a negative control.

Main outcome measure: The results showed a significant difference in inhibition between the samples groups against negative controls. The inhiibitory zone diameter of soursop leaf in 5, 10, and 15% concentrations were 9.30±0.17, 10.37±0.63, and 12.27±0.27 mm in inhibiting the growth of *Staphilococcus aureus*, and 9.00±0.00, 10.40±0.20, and 12.07±0.13 mm in inhibiting *Propionibacterium acne*. The result of the basil gel were 10.37±0.82, 11.50±0.50 and 12.43±0.29 against *Staphilococcus aureus* and 10.90±0.38, 12.00±0.00 and 13.37±0.20 against *Propionibacterium acnes*.

Conclusion: The both gels containing soursop and basil leaves ethanol extract have good antibacterial activity.

Keywords: antibacterial activity, *Annona muricata* L.,*Ocimum americanum* L., *Propionibacterium acnes, Staphilococcus aureus*

ARTICLE INFO: Received03 May -2020; Review Completed27 June 2020; Accepted 15 July 2020; Available online 15 August. 2020

Cite this article as:

Rumanti R M, Rimala M, Efendy I, Ginting I, Simarmata YBC, Diana V E, The Comparison of Antibacterial Activities of Soursop Leaf (*Annona muricata* L.) and Basil Leaf (*Ocimum americanum* L.) Ethanolic Extracts on Gel Formulated Against *Staphilococcus aureus* and *Propionibacterium acnes*, Asian Journal of Pharmaceutical Research and Development. 2020; 8(4):01-04. DOI: http://dx.doi.org/10.22270/ajprd.v8i4.770

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INTRODUCTION

Acne can occur at a young or old age with a percentage of incidence in women as much as 27% and 43% in men. Although it does not include serious diseases that can cause death, acne if left untreated can cause depression and a crisis of confidence in the sufferer[1]. Soursop leaf (*Annona muricata* L.) contains several secondary metabolite compounds including: alkaloids, flavonoids, terpenoids, coumarin and lactone, anthraquinone, tannins, glycosides, phenols, phytosterols, and saponins[2,3]. According to research results soursop leaf extract contains secondary metabolites of flavonoid compounds that can function as antibacterial[4].

Basil leaves (*Ocimum americanum* L.) have active compounds such as essential oils, alkaloids, saponins, flavonoids, triterpenoids, steroids, tannins and phenols.

Some of these chemical constituents can inhibit the growth of *Escherichia coli*, *Staphylococcus aureus* and *Klebsiella pneumonia* bacteria such as alkaloids, essential oils and phenols. This property of inhibitors is called bacteriostatic or bacteriocide[5-7].

Soursop leaves and basil leaves are often used by Indonesian people as traditional medicine. In the treatment of skin, both plants are believed to prevent the growth of acne and are often formulated in a traditional powder preparation[8-10].

In this study, a comparison of the activity of the two plants has been tested in suppressing the growth of acne-causing bacteria, *Staphilococcus aureus* and *Propionibacterium acnes*.

MATERIALS AND METHODS

Plants and Chemical Materials

Samples used in this study were soursop leaves (*Annona muricata* L.) and basil leaves (*Ocimum Americanum* L.) each as much as 2 kg obtained from the Helvetia District of Medan, carried out species identification at the Herbarium Bogoriense Indonesian Institute of Science.

The tools used in this study are glassware (pyrex), parchment paper, dropper pipettes (pyrex), spatulas, glass objects, analytical scales, blenders, rotary evaporators, petri dishes, porcelain plates, mortars and containers.
The chemicals used in this study were ethanol extracts of soursop leaves and basil leaves, concentrations of 5%, 10%, 15%, CMC-Na, glycerin, propylenglycol, Aquadest, Propionibacterium acnes and Staphylococcus aureus, NA (Sodium Agar) media, NaCl 0.9%, H2SO4, BaCL2H2O 1.175%, Medi-Klin® (Clindamycin) Gel by Surya Dermato Medica Laboratories®.

Plant Extraction

In the manufacture of soursop leaf extract and basil leaves it uses maceration method. Each of 2 kg of soursop leaves and fresh basil cleaned, chopped and then dried and obtained a dry sample. The extraction process was carried out for 5 days, where each 350 g of simplicia soursop and basil leaves were put into a container then immersed using a 70% ethanol 70% solvent covered with aluminum foil for 3 days (every day stirring) then filtered using filter paper and obtained filtrate 1 and pulp 1.

The waste was re-immersed using 875 ml of 70% ethanol solvent for 2 days (stirring daily), then filtered using filter paper and obtained filtrate 2 and pulp. Furthermore, filtrate 1 and 2 are combined into one, then concentrated using a rotary evaporator until a thick extract is obtained[11,12].

Formulation of Gel

In this research, gel preparations with different concentration of extracts will be made, namely Soursop leaf ethanol extract gel formula with a concentration of 5%, 10%, 15%, basil leaf ethanol extract gel formula with a concentration of 5%, 10%, 15%. The draft formula can be seen in table 1 and 2 .

Tabel: 1 The Gel Formula of Soursop (SG)

Materials	SG 5%	SG 10%	SG 15%
Soursop Leaf Extract	0.25 g	0.5 g	0.75 g
CMC-Na	0.25 g	0.25 g	0.25 g
Glycerin Propylen glykol	0.5 g 0.25 g	0.5 g 0.25 g	0.5 g 0.25 g
Distilled water (up to)	5 ml	5 ml	5 ml

Tabel: 2 The Gel Formula of Basil (BG)

Bahan	BG 5%	BG 10%	BG 15%
Basil Leaf Extract	0.25 g	0.5 g	0.75 g
CMC-Na	0.25 g	0.25 g	0.25 g
Glycerin Propylen glykol	0.5 g 0.25 g	0.5 g 0.25 g	0.5 g 0.25 g
Distilled water (up to)	5 ml	5 ml	5 ml

Measurement of Antibacterial Activity

To make a bacterial suspension of Propionibacterium acne and Staphylococcus aureus that is by means of the culture of Propionibacterium acne and Staphylococcus aureus taken with a sterile wire, then suspended into a test tube containing 10 ml NaCl 0.9% to obtain the same turbidity with the standard turbidity of Mc Farland's solution[13].
The base layer is made by pouring each of 10 ml of nutrient agar into a petri dish, then allowed to solidify, after solidifying the surface the base layer is planted with 5 steel parts that are spaced so that the observation area is not concentrated. The bacterial suspension is mixed into the media. Next 25 ml of media was poured on each petri dish which was placed as a second layer of metal scrap. After the second layer solidifies, the metal scrap is removed aseptically using tweezers from each petri dish, so wells are formed to be used in bacterial testing[14,15].
Microbiological test to determine the antibacterial activity of ethanol extracts of soursop leaves and basil leaves gel carried out by agar diffusion, by measuring the diameter of bacterial growth inhibition against *Propionibacterium acnes* bacteria and *Staphylococcus aureus*. The test method is that the wells that have been made on the test media are dripped with 50 µl of test solution using a micropipette, then incubated in an incubator at 35 ± 2 ° C for 24 hours, after which the diameter of the inhibition zone (clear zone) is measured[14, 15].The data obtained are presented in tabular form which is the result of inhibition zone measurements. Furthermore, the data obtained from the results of the study are processed with statistics, namely Analysis of variance (ANOVA).

RESULT AND DISCUSSION

The results of the antibacterial activity test can be seen in table 3.

Table: 3 Antibacterial Activity Test Result

Sample	Diameter of Inhibition Zone (mm)	
	***Staphylococcus aureus* ± Std. Error**	***Propionibacterium acne* ± Std. Error**
Positive Control	36.40 ± 0.00*	35.00 ± 0.00*
Negative Control	0.00 ± 0.00	0.00 ± 0.00
SG 5%	9.30 ± 0.17*	9.00 ± 0.00*
SG 10%	10.37 ± 0.63*	10.40 ± 0.20*
SG 15%	12.27 ± 0.27*	12.07 ± 0.13*
BG 5%	10.37 ± 0.82*	10.90 ± 0.38*
BG 10 %	11.50 ± 0.50*	12.00 ± 0.00*
BG 15%	12.43 ± 0.29*	13.37 ± 0.20*

Explanation : * Significantly different from negative controls $p \leq 0.05$

Based on the test results in the table the results obtained on the SG gel against Staphylococcus aureus and Propionibacterium acne bacteria showed a smaller inhibitory zone compared to BG gel, but both of them had good activity as evidenced by significant results in negative control.

The content of secondary metabolites from soursop and basil leaves which act as antibacterial so that when combined can increase the antibacterial activity and produce a greater inhibition zone than a single extract. The main ingredients of basil leaves are essential oils, flavonoids and other content such as flavone apigenin, luteolin, O-glucosidoid flavonapigenin 7-O glucuronide, luteolin 7-O glucoronide, flavone C-glucoside orientin, and ursolic acid which functions as antibacterial. While the soursop leaf active compound which has the potential to be an antibacterial compound is flavonoids. The results of identification of the flavonoid group showed soursop leaf extract containing falvonoid, flavone, dihydroflavonol, flavonol and flavonon groups. The synergistic effect of active ingredients is a condition when the effects produced by the active compounds together are greater than the single effect of each active compound[2-5].

Clindamycin works by inhibiting protein synthesis from bacteria by inhibiting ribosome translocation. Clindamycin binds to 50S from bacteria, specifically it binds mainly to the 23S RNA subunit. The use of positive control functions as a control of the test substance, by comparing the diameter of the inhibition zone formed. With this comparison we can see whether the natural gel preparations made have better results compared to the preparations with active chemicals that are circulating on the market as anti-acne[16-18].

CONCLUSSION

Both gels which have active ingredients of soursop and basil leaf extract have good activity against Staphilococcus aureus and Propionibacterium acnes bacteria.

CONFLICT OF INTEREST

All author have no to declare.

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