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by N M Saptarini

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Application of Fig (*Ficus carica* L.) Leaf Extract as A Herbal Antibacterial Body Scrub

Nyi M. Saptarini[1]*, Irma E. Herawati[2], Gendhis M. Amdasari[3]

[1]Department of Pharmaceutical Analysis and Medicinal Chemistry, Faculty of Pharmacy, Universitas Padjadjaran, Bandung-45363 West Java, Indonesia

[2]Indonesian School of Pharmacy, Bandung-40266, West Java, Indonesia

[3]Department of Pharmacy, Faculty of Mathematics and Natural Sciences, Al Ghifari University, Bandung-40293, West Java, Indonesia

*Corresponding author: Nyi Mekar Saptarini; *Email: nyi.mekar@unpad.ac.id

Jl. Ir. Soekarno km 21 Jatinangor, West Java, Indonesia, 45363

+62-8156078248

ABSTRACT

Fig (*Ficus carica* L.) leaves contain alkaloids, saponins, steroids, terpenoids, tannins, flavonoids, and phenolic compounds. Fig leaf extract has been proven to have antibacterial activity on *Propionibacterium acnes* and *Staphylococcus epidermidis*, which causes acne. Acne occurs when the skin pores are blocked with dead skin, oil, or bacteria. The study aimed to formulate and evaluate fig leaf extract body scrub to *P. acnes* and *S. epidermidis* as acne-causing bacteria. The evaluation was carried out for 56 days to observe the body scrub stability during storage at ambient temperature. The methods include body scrub formulation with 3% fig leaf extract as active ingredients and variations in silica sand concentration (1-3%) as scrubbing agents. Furthermore, body scrub evaluations include organoleptic, pH, viscosity, spreadability, antibacterial activity to *P. acnes* and *S. epidermidis*, irritation, and hedonic test. The results of fig leaf extract body scrub with variation of 1-3% silica sand has light green color with specific scent and rough texture, which comparable to extract concentration, the range of viscosity of 7750-9550 cps, pH of 6.8-7.3, spreadability of 4.9-6.2 cm, inhibition zone of 14.56-14.68 mm to *P. acnes* and 15.67-15.69 mm to *S. epidermidis*, non-irritant on back of volunteer's hand, and stable for 56 days at ambient temperature. Silica sand concentration affected viscosity and roughness. The formula with 3% silica sand was the most preferred preparation based on color, scent, and texture in 30 volunteers. It concluded that fig leaf extract can be formulated into a body scrub with good physical characteristics and is safe.

Keywords: silica sand, safe, non-irritant, stable

Introduction

Ficus carica L. or fig, belonging to the family Moraceae, is one of the largest genera of Angiospermae with more than 800 species of trees, epiphytes, and shrubs in subtropical and tropical regions worldwide. It is a native plant of south-western part of Asia Minor, which is now distributed throughout the world.[1] Botanical classification for fig is: Kingdom: Plantae Division: Magnoliophyta Class: Magnoliopsida Order: Urticales Family: Moraceae Genus: Ficus Species: carica.[2] The fruit, leaf, and root of fig were used as traditional medicines as an

anti-inflammatory and antispasmodic remedy and to treat digestive, such as appetite loss, colic, and diarrhea; cardiovascular; respiratory, such as cough, sore throat, and bronchial problems.[3]

Fig can grow in Indonesia. One of the places for fig cultivation is Ciwidey District in West Java Province, Indonesia.[4] The fig leaf are broad ovate or nearly orbicular, more or less deeply lobed, blade 10-20, petiole 5-7.5 cm, rough above and pubescent below.[2] The fig leaf from Ciwidey District have been proven to contain alkaloids, saponins, steroids, terpenoids, tannins, flavonoids, and phenolic compounds. The ethanolic fig leaf extract contains a total phenolic content of 2.52 ± 0.24 mg GAE/g simplicia and total flavonoid content of 2.03 ± 0.01 mg RE/g simplicia.[4] The identified phenolic compounds in fig leaf are 3-CQA(3-O-caffeoylquinic acid), 5-CQA (5-O-caffeoylquinic acid), Q-3-Glu(quercetin3-O-glucoside), Q-3-rutin (quercetin3-O-rutinoside), ferulic acid, psoralen, bergapten, pyrogallic, phenol,3–5-dimethoxy, coumaric, phenolpthethin, pinocembrin, chrysin, galangin, proto catechol, vinallin, cinnamic, quercetin and pinostrobin.[5] While, the identified flavonoid in fig leaf is luteolin, luteolin-6C-hexose-8C pentose, quercetin, quercetin rutinoside, quercetin glucoside, quercetin acetyl glucoside, biochanin-A, apigenin rutinoside, and kaempferol rutinoside.[6]

The 30% to 60% ethanolic fig leaf extract showed antibacterial activity against *Propionibacterium acnes* and *Staphylococcus epidermidis*.[7] Increasing the extract concentration from 30% to 60% showed an increase in the inhibition zone, which is proportional to the increasing antibacterial activities. *P. acnes* is localized in the follicle,[8] while *S. epidermidis* is a human skin microbiota.[9] Both bacteria are involved in acne pathogenesis.[8,9] Long-term antibiotic use causes antibiotic resistance, so to overcome this, herbal remedies have been developed.[10]

Our previous study showed that fig leaf extract was successfully formulated into a cream with antibacterial activity on *P. acnes* and *S. epidermidis*.[11] It has been proven that there is a relationship between the concentration of flavonoids and phenolic compounds with antibacterial activity. There is no standardized topical formulation with good efficacy, safety, and stability for extract,[12] including fig leaf extract. The study aimed to formulate and evaluate fig leaf extract body scrub to *P. acnes* and *S. epidermidis* as acne-causing bacteria. The evaluation was carried out for 56 days to observe the body scrub stability during storage at ambient temperature. The methods include body scrub formulation with 3% fig leaf extract as active ingredients and variations in colloidal silicon dioxide (silica sand, SiO_2) concentration (1-3%) as scrubbing agents. Furthermore, body scrub evaluations include organoleptic, pH, viscosity, spreadability, antibacterial activity to *P. acnes* and *S. epidermidis*, irritation, and hedonic test. This study is important to scientifically evaluate the efficacy, safety, and quality of herbal preparations.[13] The novelty of this study was the utilization of fig leaf waste from Ciwidey District, Indonesia, so it can increase the economic value of fig leaf and reduce plant waste. Currently, generally only fig fruits have economic value, whether consumed fresh or processed into food or cosmetic products.

Materials and Methods

Materials

The ethanolic fig leaf extract was obtained from previous studies,[4] while fig leaf were collected from Ciwidey District (GPS coordinates of 7° 4' 59.0232" S and 107° 26' 52.7100" E), West Bandung Regency, West Java Province, Indonesia. Silica sand, stearic acid, cera alba, vaseline album, methyl paraben, propyl paraben, propylene glycol, and triethanolamine

(TEA) was cosmetic grade and purchased from TTK Science Co. (Thailand). *P. acnes* ATCC 1223 and *S. epidermidis* ATCC 12228 obtained from the Microbiology Laboratory, School of Pharmacy, Bandung Institute of Technology. Sulfuric acid, barium chloride, and dimethyl sulfoxide (DMSO) was analytical grade and purchased from Sigma Aldrich (Germany). Mueller Hinton Agar (MHA) was bacteriology grade and purchased from Oxoid (UK).

Formulation of body scrub

This body scrub formula based on modified formula of Chasanah.[14] The oil phase consisted of 12.5% stearic acid, 0.5% cera alba, and 10% vaseline album which were heated at 70^{0}C in a water bath, then 0.02% propyl paraben was added. The water phase consisted of distilled water which was heated at 70^{0}C in a water bath, then 0.18% methyl paraben, 15% propylene glycol, and 1.5% TEA were added. The water phase was mixed for 15 min into the oil phase in the mixing tank (Cosmo Machinery, China). Fig leaf extract (3%) and silica sand (1, 2, and 3%) were added in the body scrub base and homogenized in the mixing tank (Cosmo Machinery, China).

Evaluation of body scrub

All body scrubs were observed for 56 days at ambient temperature. Evaluations were carried out every week for 8 weeks with three repetitions. The evaluation methods as described in Saptarini et al.[11]

1. The organoleptic properties, i.e. color, scent, and texture, of the body scrubs were assessed.
2. Homogeneity was analyzed from visual inspection for the appearance and clog existence.
3. pH of the 30% body scrub was measured using a calibrated pH meter (Beckman, Germany).
4. Viscosity was determined to 100 g of body scrub, using a CAP-2000 Brookfield viscometer with spindle no. 64 at 60 rpm and 25 °C. The results were recorded after a stable value.
5. Spreadability was determined to 1 g of the body scrub was placed in the center between two watch-glasses. The upper watch-glass was weighted by placing a weight until it reached 150 g, and left for 60 seconds. Evaluations were made by measuring the diameter of spreadable body scrub.

Antibacterial activity assay

Antibacterial activity assay was described in Saptarini et al.[15] The 0.5 Mc Farland solution was used as a turbidity standard for *P. acnes* and *S. epidermidis* suspensions prepared from a mixture of 1% sulfuric acid solution and 1.175% barium chloride solution (9.95:0.05). Petri dishes containing 20 mL of MHA and 20 µL of bacterial suspension were allowed to solidify. Then, the agar was perforated with a perforator and filled with 50 µL of 1% DMSO or body scrub, which was prepared by dispersing 1 g of body scrub in 1 mL of 1% DMSO. Each inhibition zone was measured using a caliper.[11,15]

Irritancy and preference test

Ethical clearance for irritancy and preference test was obtained and approved by the Health Research Ethics Committee of the Dr. Hasan Sadikin Hospital, West Java, Indonesia. Ethical approval letter no. 476/UN6.KEP/EC/2023. The methods were described in Saptarini et al.[11]

1. Irritation test using a patch test technique (patch test) on 10 volunteers. The body scrub was applied on the dorsal left-hand surface of the volunteers with an area of 2.5x2.5 cm^2, then observed for 24 hours. A positive irritation reaction was indicated by redness, itching, or swelling on the skin of the treated hand.
2. Preference test (hedonic) was conducted on 30 volunteers consisting of men and women aged 20-25 years old using a questionnaire. The volunteers were asked for their responses regarding color, scent, and texture on the skin with a rating of very like (4), like (3), dislike (2), and very dislike (1).

Data analysis

Data was processed using SPSS 21 statistical analysis, one way ANOVA and repeated measures ANOVA as parametric analysis, then the Kruskal Wallis test and Friedman test as nonparametric analysis ($p < 0.05$).

Results and Discussion

Body scrub was made, because it contains scrubbing agents, which can exfoliate dead skin and provide a relaxing effect. This preparation also increases the absorption of extracts, so it can increase its antibacterial effect.[16] Fig leaf extract has a thick consistency, dark brownish green with a distinctive fig scent. The extract was proven to contain phenolic and flavonoid compounds that are comparable to antibacterial activity in *P. acnes* and *S. epidermidis*.[11] This underlined the formulation of antibacterial body scrub from fig leaf extract. The three body scrub formulas were successfully made with variations in silica sand concentration, i.e. 1-3%. The three formulas have a similar appearance, i.e. light brown semisolid preparation with a distinctive fig scent (Figure 1). Silica sand effectively removes dead skin cells without causing irritation or damage to the skin.[17] The purpose of various concentration of silica sand was to increase the mild chemical exfoliator, which functions as a mechanical exfoliation that helps to remove dead skin to produce a smooth and rejuvenated skin surface, dead skin can clog skin pores and cause acne. In addition, the body scrub provides physical stimulation to the skin through a massage effect.[18,19] The choice of exfoliator affected scrub performance, in this study, we have chosen silica sand, because of the character of water-insoluble abrasives.[18] In this study, TEA was used to neutralizes fatty acids, i.e. stearic acid, adjusts and buffers the pH to neutral, and solubilizes oil in the formula that was not completely soluble in water.[20] Stearic acid is used as a surfactant and softening agent in cosmetics and as a thickening agent and an emulsifier in soap and detergents. It can be used as skin-smoothing abilities.[21] Stearic acid and TEA as emulsifiers will reduce surface tension and prevent separation of the oil and aqueous phases. The advantage of oil-in-water cream is that it is easily washed off with water.[22]

Organoleptic observation showed that the body scrub was a semisolid form, light brown in color, with a distinctive fig leaf scent. Organoleptic observation showed that there was no alteration in form, color, and scent after being stored for 56 days. This was because of methyl and propyl paraben, as the preservative, which prevent the growth of microorganisms.[23] An air-tight and light-resistant container was important in maintaining a stable scent.

Homogeneity observation showed that there was no clog of extract or silica sand, due to the right mixing process, so the oil and water phase can completely mix. In topical preparations, such as body scrub, the function of stearic acid was as an emulsifying and solubilizing agent.[21] The combination with TEA will cause stearic acid to be partially neutralized to form a creamy base with the appearance and plasticity depending on the TEA concentration.[24] This was in accordance with the combination of TEA with stearic acid forms fine-grained and stable oil-in-water emulsions.[22] The concentration of 1.5% TEA in the formula was in accordance with the function and safety of TEA for skin and hair preparations, i.e. 1-3%. At higher concentrations of TEA, TEA can cause irritation and redness.[25]

All three formulas have a neutral pH, i.e. 7.1 ± 0.06 (Figure 2), so it is in accordance with the requirements of topical preparations.[21] The pH of the three formulas was not statistically significant (p value = 0.07). This was because the difference was only in the concentration of silica sand, while silica sand is neutral, so it does not affect the pH of the body scrub.[17] After 56 days of storage, there was a significant change in pH (p value = 0.04). The decrease in pH by 0.2 to 0.4 caused by the presence of carbon dioxide from the air, which reacted with the water phase and created addition of hydrogen ions, then decreased the pH. However, decreased pH was still within the pH requirements of topical preparations.[22]

The viscosity is attributed to the excipients and the formulation process. The viscosity was assessed to ensure the body scrub sensation and behavior on the skin.[26] TEA also functions as a thickening agent and formula stabilization.[24] Body scrub viscosity was ranging from 9200 to 9500 cPs (Figure 3). Increasing the concentration of the silica sand was inversely proportional to body scrub viscosity, but there was no significant difference in the three formulas (p value = 0.56). The viscosity of the three formulas were in accordance with SNI 16-4399-1996 about cosmetic products, i.e. 2000-50000 cPs.[27] However, the viscosity in the three formulas decreased by 1400 to 1620 cPS and significantly different during 56 days of storage (p value = 4.7×10^{-9}). After 56 days, the viscosity of the three formulas became 7800 to 7850 cPs (Figure 3). This value still fulfilled the requirements of cosmetic viscosity, but decreased viscosity was undesirable. Decreased viscosity in each formula probably due to the mixing time was not optimal, so the function of TEA as a thickening agent was not optimal. Other studies show that a mixing time of 25 min and a temperature of 70 °C affect TEA effectiveness as an emulsifier.[28] Therefore, for body scrub formulas, it is recommended to increase the mixing time to 25 min.

The spreadability of the three formulas ranged from 5.00 to 5.26 cm (Figure 4), and met the requirements for good spreadability of 5-7 cm.[28] Increasing the concentration of silica sand was inversely proportional to the diameter of the spreadability, although statistically, the spreadability of the three formulas was not significantly different (p value = 0.39). However, spreadability increased in all three formulas and was significantly different during 56 days of storage (p value = 3.99×10^{-8}). This was related to viscosity, if the viscosity was decreased, so the diameter of spreadability was increased.[29] The spreadability was increased of 5.96 to 6.10 cm (Figure 4), due to decreased viscosity, caused by the TEA mixing time was not optimal.

Body scrub formula on various silica sand concentration with 3% fig leaf extract gave significantly different results with 1% DMSO as a solvent, but no significantly different with 1% clindamycin as a standard, namely p-value = 0.089 for *P. acnes* and p-value = 0.134 for *S. epidermidis.* This proves that there is no effect of the excipients to the diffusion ability of the fig leaf extract. The inhibition zone of body scrub on *S. epidermidis* was bigger than that of *P. acnes* and showed a significantly different (p value = 3.47×10^{-12}). The compatibility of the

body scrub base with fig leaf extract determines the distribution of secondary metabolite compounds in the base and the diffusion ability of secondary metabolites through agar containing *P. acnes* or *S. epidermidis*. Table 1 showed that *S. epidermidis* is more sensitive to secondary metabolites of fig leaf extract compared to *P. acnes*. This was accordance with the results of previous studies.[7,15]

The results on 10 volunteers showed that all volunteers gave negative results for the irritation reaction parameters, i.e. red skin, itching, or swelling. It was concluded that the scrub preparation was safe for use. This is due to the use of raw materials with cosmetic grade, in addition, the composition of the ingredients in the formula is adjusted to the Limits for the use of these materials in body scrub preparations. Stearic acid is regarded as a nontoxic and non-irritant material.[21] There are no adverse reactions to parabens have been reported, although associated with hypersensitivity reactions.[23] Silica sand is regarded as the nontoxic and nonirritant excipient.[17] TEA is safe for skin and hair at concentrations of 1-3%. Higher than this concentration can cause irritation and redness.[25] In this study, only 1.5% TEA was used, below the maximum concentration, which was 3%.[25]

Preference test was conducted on 30 volunteers, who evaluated the three formulas regarding color, scent, and texture on the skin. Figure 5 showed that volunteers liked the color and texture of all formulas, while scent was less preferred. The volunteers did not like the scent, because of the low volatile compounds in fig leaf extract, so the scent was too weak to be detected by olfactory sensory neurons. The concentration of fig leaf extract was the same for all three formulas, so the color and scent gave almost the same value and was not statistically significant (p value = 0.91). The volunteers preferred formula 3 with the highest silica sand concentration, based on assessment on the sensory test on the skin.[30]

Conclusion

Fig leaf extract contains secondary metabolites that have antibacterial activity, so it can be formulated into a body scrub. Silica sand is intended to remove dead skin that is a place for the growth of acne-causing bacteria, such as *P. acnes* and *S. epidermidis*. Fig leaf extract can be formulated into a body scrub with good and safe physical characteristics after being stored for 56 days. This is a prospect to be developed into an effective and safe herbal antibacterial body scrub.

Conflict of Interest

The authors declare no conflict of interest.

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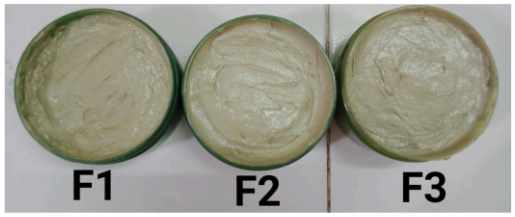

Figure 1: Body scrub with 1% (F1), 2% (F2), and 3% (F3) silica sand.

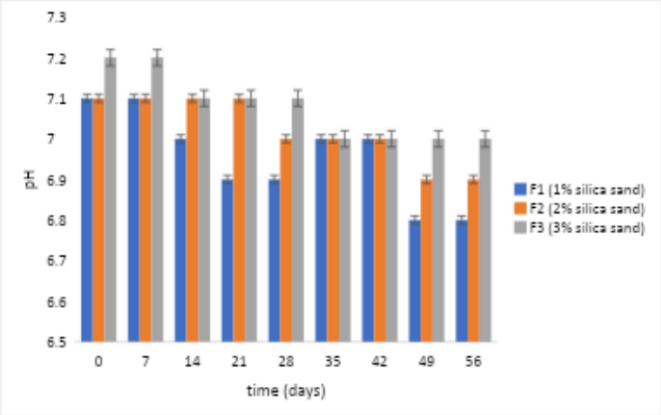

Figure 2: Observation of pH of body scrub of fig leaf extract during 56 days (n=3)

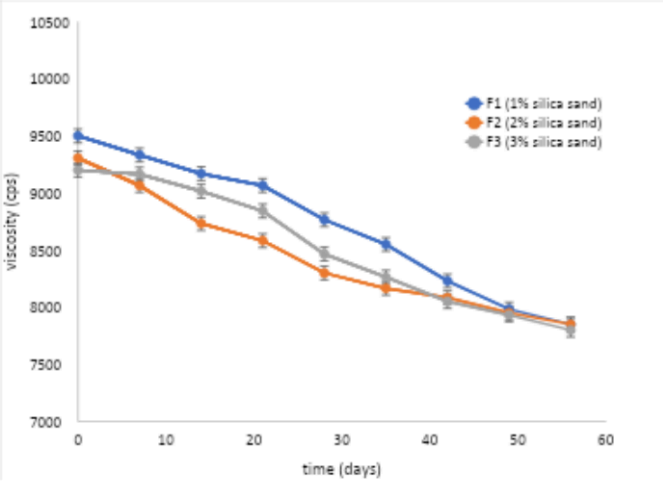

Figure 3: Observation of the viscosity of the body scrub during 56 days (n = 3)

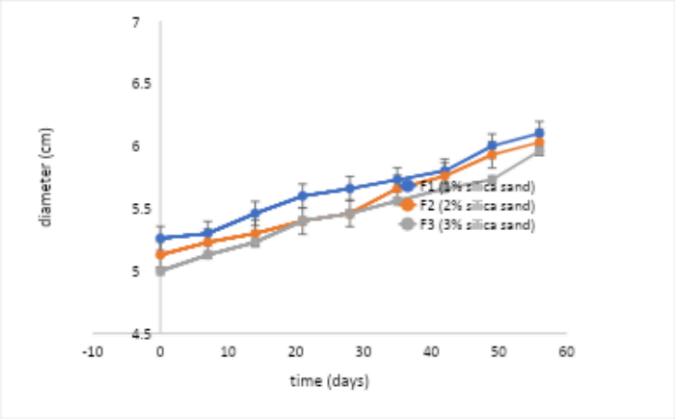

Figure 4: Observation of the spreadability of body scrub during 56 days (n = 3)

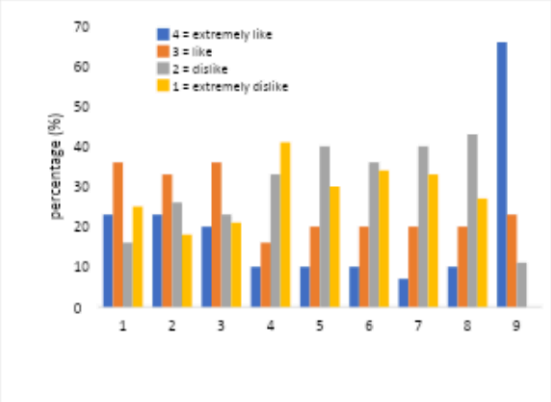

Figure 5: Observation of the hedonic test on the fig leaf extract body scrub (n = 30)

Table 1: Antibacterial activity result of fig leaf extract body scrub

Sample	Inhibition zone (mm)	
	P. acnes	*S. epidermidis*
1% DMSO	0	0
1% clindamycin	14.72 ± 0.56	15.76 ± 0.47
F1 (1% silica sand)	14.56 ± 0.42	15.67 ± 0.53

F2 (2% silica sand)	14.62 ± 0.43	15.68 ± 0.23
F3 (3% silica sand)	14.68 ± 0.45	15.69 ± 0.36

Note: Perforator diameter was 6 mm

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