



FORMULATION AND TEST ACTIVITIES OF ANTIDANDRUFF SAMPLE OF LEAVES ETHANOL EXTRACT (*Acanthus ilicifolius*) OF *Pityrosporum ovale*

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Abstract

Dandruff is an anomaly condition on the scalp and one of the causal factor is *Pityrosporum ovale*. *Acanthus ilicifolius* leaf a natural substance which is containing of anti-fungi compound, namely alkaloid, flavonoid, tanin, saponin, and steroid. The purpose of this research was to make the formulation of anti-dandruff shampoo preparation of *Acanthus ilicifolius* Leaf Ethanol Extract with four variations of concentration and to examine the increasing concentration effect of *Acanthus ilicifolius* Leaf Ethanol Extract in anti-dandruff shampoo preparation to anti-fungi activity. In this research shampoo preparation with four formulas was made. Each formula contains a different concentration of *Acanthus ilicifolius* Leaf Ethanol Extract ranging from 5,5-10%. After that the physical properties of the shampoo preparation are evaluated, then an antifungal activity was tested using the well diffusion method. The result of this research proved that *Acanthus ilicifolius* leaf could be formulated as an anti-dandruff shampoo preparation which was eligible in organoleptic, pH, foam height, water level, viscosity, and specific gravity. *One Way Anova* test showed that there were the significant difference in the diameter of inhibitory zone and was continued with *Tukey HSD* test of confidence level showed that the highest inhibitory zone diameter was in concentration 10% and control (+) *Head and Shoulder* shampoo while the lowest inhibitory zone diameter was at 5,5% and control (-) aquadest. Based on these data it can be concluded that the higher the concentration of the ethanol extract of the *Acanthus ilicifolius* leaves, the greater the inhibitory zone formed.

Keywords: *Leaves Acanthus ilicifolius, Sampo Antidandruff, Pityrosporum ovale*

Introduction

Jeruju (*Acanthus ilicifolius*) is one of the plants that is found and grown on the coast of Indonesia, including in Jambi Province, Tanjung Jabur Barat and Tanjung Jabung Timur. *Acanthus ilicifolius* is one type of plant that is widely found in mangrove forests. *Acanthus ilicifolius* contains chemical compounds in the form of alkaloids, flavonoids, tannins, saponins, steroids, lignans, phenol and terpenoid components (Purnobasuki, 2004). The Jeruju (*Acanthus ilicifolius*) plant is widely used by the people of Tanjung Jabung Timur Regency for herbal treatments. Jeruju leaves have been

used by the people of East Tanjung Jabung Regency as rubbing drugs and removing toxins due to arrow wounds (Sukardjo, 1986). Jeruju leaves also have antifungal activity.

The results of the study (Bose, 2008a and Bose, 2008b), showed that alcoholic extract, butanol and chloroform from *Acanthus ilicifolius* leaves showed strong inhibitory action against the fungi *Candida albicans*, *Aspergillus niger* and *Aspergillus fumigatus*. In *Acanthus ilicifolius* 10 mg / mL or 1% alcoholic extract, the inhibition of *Candida albicans* is 22 mm, *Aspergillus niger* is 21 mm and *Aspergillus fumigatus* is 19 mm.

Research that has demonstrated the ability of alcoholic extracts and butanol and chloroform from the leaves of Jeruju *Acanthus ilicifolius* in inhibiting fungi *Candida albicans*, *Aspergillus niger* and *Aspergillus fumigatus* showed the presence of antifungal activity in the leaves of jeruju. Of all the results of the study, no testing of the fungus *Pityrosporum ovale* has been carried out.

Pityrosporum ovale is a yeast or single-celled fungus that is a member of the genus *Malassezia* sp, and is the cause of superficial mycoses that affect the stratum corneum in the epidermal layer. *Pityrosporum ovale* is a microorganism that is thought to be the main cause of dandruff, this fungus is actually a normal flora on the scalp, but in conditions of hair with excess oil glands, this fungus can thrive (Oktaviani, 2012).

One of the cosmetic products that can be used to inhibit the growth of fungi, *Pityrosporum ovale*, causes dandruff, namely shampoo. Shampoo is a liquid, gel, emulsion or surfactant (surface active ingredient) cosmetics preparation, so it has detergency, humectant and foam properties. Shampoo is a cosmetic preparation that is used to clean the hair, so that the hair and scalp are clean and wherever possible soft, easily regulated and shiny (Faizatun and Lilyana, 2008).

Based on the preliminary results of the inhibitory test, the extract of ethanol extract of jeruju leaves (*Acanthus ilicifolius*) on *Pityrosporum ovale* obtained a dispersion of 20 mm at a concentration of 7% indicating that the extract of the jeruju leaves had a strong antifungal activity category, the researchers were interested in making a cosmetic product namely shampoo with various concentrations of jeruju leaf extract (*Acanthus ilicifolius*) and determine which extract is the best in inhibiting the *Pityrosporum ovale* fungus.

Material and Methods

Material

The ingredients used were ethanol extract of jeruju leaves, sodium lauryl sulfate, TEA, NaCMC, citric acid, methyl paraben, menthol, ethanol 96%, SDA media, 0.9% NaCl, *Pityrosporum ovale* and aquadest.

Instrument

The tools used are measuring cup, cup glass, test tube, petri dish, drop pipette, hot plate, porcelain cup, micropipette, scale ruler, autoclave, Laminar Air Flow (LAF), ovens, bunsen

lamps, incubator, digital scales, ovens, blenders, aluminum foil, furnaces, filter paper, support rods, shampoo containers, pH meters, picnometers, funnels, watch glass, water baths, spatulas, and Viscometer Brookfield.

METHODS

Plant Determination

The study of the ethanol extract of the leaves of Jeruju (*Acanthus ilicifolius*) was initiated by determining the Jeruju plant in the Laboratory of Biotechnology and Genetic Engineering at the Jambi University Faculty of Science and Technology. The aim is to prove the validity of the identity of the plants used, whether these plants are truly desirable plants in the study.

Sample Setup

Jeruju leaf samples (*Acanthus ilicifolius* L.) were taken from the Tanjung Jabung Timur Mangrove Forest, in the coastal area of the Sabak Sea Village. Taking samples is done during the day, the sample is taken by cutting the shoots (young leaves) from the jeruju (*Acanthus ilicifolius*) using a knife or other cutting tool.

Simplicia Making

Samples that have been collected are sorted dry, followed by sorting wet and washed with running water to remove impurities that are still attached to the leaves. Then chopped into small pieces to speed up the drying process. Then the sample was dried in an oven at 50°C for 3x24 hours. The dry sample is weighed to a constant weight and powdered using a grinder. The weight of the formula is calculated by the yield.

Determination of Drying Shrinkage and Content of Jeruju Leaves Simplicia ash

Drying shrinkage

10 grams of powder is put into a petri dish that has been weighed previously. The cup is put into the oven and dried at a temper

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ature of 105°C for 2 hours until the weight remains. Drying losses are calculated in percent.

Ash content

2 grams of powder is weighed carefully into the porcelain crucible which has been incandescent and ground, then flattened. Crushes are glowed slowly until the charcoal is exhausted, the spawning is carried out at 6000C for 3 hours then cooled and weighed until a fixed weight is obtained. The total ash content is calculated in percent (Ministry of Health, 2008).

Making Ethanol Extract of Jeruju Leaves (*Acanthus ilicifolius* L.)

Simplicia powder was put into a maceration bottle and ethanol 96% was added, soaked for 2 days, then filtered and remacerated. The resulting macerate is then concentrated with a rotary evaporator until thick extracts are obtained (Jamal, 2017).

Making Shampoo Preparation

The prepared shampoo formula is presented in Table 1. Na CMC was developed with hot water (M1). Methyl paraben is dissolved with a few drops of ethanol to dissolve (M2). Some aquadest are heated on the hot plate and put in sodium lauryl sulfate, stirring until homogeneous. TEA added to it while continuing to stir until homogeneous. M1 and M2 are mixed into it and stirred until the liquid thickens (M3). Ethanol extract of jeruju leaves (*Acanthus ilicifolius*) is mixed into M3, stirring until homogeneous. The shampoo solution added citric acid. M3 shampoo solution was cooled and menthol was added. Enough with distilled water to 100 ml and stir until homogeneous. The shampoo is put in the container.

Table 1. Modification of the Shampoo Formula ethanol extract of jeruju leaves (*Acanthus ilicifolius*)

No	Bahan	Konsentrasi (%)			
		F1	F2	F3	F4
1.	Ekstrak	5.5	7	8.5	10
2.	Natrium Lauril Sulfat	10	10	10	10
3.	Triethanolamine (TEA)	4	4	4	4
4.	Na CMC	3	3		3
5.	Asam Sitrat	0.25	0.25	0.25	0.25
6.	Metil Paraben	0.15	0.15	0.15	0.15
7.	Methanol	0.5	0.5	0.5	0.5
8.	Aquades	ad 100	ad 100	ad 100	ad 100

Evaluation of Shampoo Preparation Organoleptis

The organoleptic examination performed refers to the Ministry of Health of the Republic of Indonesia (1995) which includes visual inspection of shape, color and odor.

pH Measurement

1 gram of shampoo is dissolved into 10 ml of water and measured by pH using a digital pH meter (Anonim, 1992). Measuring pH of the shampoo preparation is done once a week for one month.

Foam Height Measurement

A 0.1 g shampoo is dissolved in 10 mL of water. Then put into a test tube, closed and shaken for 20 seconds by reversing the reac-

tion tube in a regular manner. Then the height of the foam formed (Ratnawulan, 2009) was measured.

Test Stability Test

The shampoo preparation of ethanol extract of jeruju leaves is stored at low temperatures for 24 hours, then transfer to high temperature for 24 hours (1 cycle). Do it for 6 cycles (12 days) (Djajadisastra, 2004). Then observed the physical changes in the shampoo before and after the Cy-cling Test.

Moisture Measurement

1 g of the sample was weighed in a petri dish which was initially known to the masses.

Samples and petri dishes were heated in an oven at an oven temperature of 103-105 ° C for 24 hours then cooled in a desiccator and weighed. After cold, the sample is heated for 2 hours and weighed again. This step is carried out until a constant weight is obtained (Anonymous, 1992).

Viscosity Testing

Viscosity test by placing an antidandruff shampoo preparation to be examined in a becker glass (\pm 25 mL), then placed under a Brookfield viscometer with a suitable spindle. Spindles are inserted into the preparation until submerged (Mahataranti et al, 2012).

Type Weight Measurement

A blank and dry blank pycnometer is measured for weight, then the weight of the aquadest picnometer is measured. Furthermore, the picnometer containing the dosage of shampoo was measured for its weight, measured closed (Ministry of Health, 1995).

Hedonic Test

The hedonic test was carried out by collecting 10 panelists and given a questionnaire containing an assessment of the preferences of the organoleptic tests performed visually on shampoo preparations, including color, smell and taste and given a rating scale of 1-4 as follows: 1 (no likes), 2 (less like it), 3 (like), 4 (like it).

Antifungal Activity Test

Sterilization Tools

The tools and media were sterilized using an autoclave at 1210C for 15 minutes, the needle and tweezers were sterilized with fixation.

Making SDA Media (Sabouraud Dextrose Agar)

6.5 grams of Sabouraud Dextrose Agar (SDA) dissolved in aquadest 100 mL. Heated on the hot plate while stirring until the solution becomes homogeneous. The homogeneous medium is sterilized in an autoclave at 1210C, a pressure of 2 atm for 15 minutes.

Rejuvenation of Mushrooms

Standard mushrooms are taken by using ose which has been spawned with fire by scraping. Then the mushrooms that have been taken with the ose are planted by zigzagging

them on the surface of the media. Mushrooms are incubated at 370C for 3-4 days.

Manufacture of Test Mushroom Suspensions

1 ose of mushroom is suspended in 0.9% NaCl liquid 10 mL. The mushroom suspension was vortexed for 15 seconds, then poured into cuvet for 7 mL. Cuvet was inserted into a spectrophotometer with a wavelength of 530 nm and an absorbance rate of 0.5 - 0.6, which was equivalent to the standard Mc Farland 0.5.

Antifungal Activity Testing

The test fungus suspension was taken as much as 50 μ l using a micropipette and then dropped on the center of the agar surface which had solidified. The drop of the mushroom suspension was then flattened. Solid media that has been mixed with test fungi is made well by using a cork (perforator) hole with a diameter of 5 mm. In the wells, various tests were carried out with various concentrations of shampoo, the ethanol extract of the leaves of jeruju. Each test solution with various concentrations of shampoo is put into a hold, then incubated for 2x24 hours at 370C in an incubator. Triple repetitions are performed for each formulation.

Data Analysis Test

Data on simplicia yield, shrinkage of simplicia drying, simplicia ash content, extract yield, organoleptic extract, extract water content, organoleptic properties of shampoo, homogeneity, and hedonic test were analyzed descriptively. Data on inhibition zone diameter, pH, foam height, moisture content, viscosity, pH stability of shampoo preparation, and shampoo specific gravity were analyzed statistically with variance analysis using SPSS software.

Results and Discussion

Determination of Jeruju Plants and Sample Preparation

Plant determination aims to prove the truth of the plant identity used, whether the plant is really the desired plant in the study. Thus errors in the use of plants used can be avoided. The parts of the plant that are determined are the stems, leaves, and fruit of the jeruju plant. Based on the results of plant de-

terminations carried out in the laboratory of Biotechnology and Genetic Engineering, Faculty of Science and Technology, University of Jambi, it was found that the plants used were true jeruju (*Acanthus ilicifolius*).

Sampling is done during the day because the plants undergo photosynthesis and also make it easier when taking because of low tide. Samples were taken by cutting the shoots (young leaves) from the *Acanthus ilicifolius* plant using a knife and scissors. By taking the shoots of the shoots, new shoots will emerge after a few months. So that it does not damage the Jeruju plant and preserve the Jeruju plant.

Simplicia Making

Making the simplicia of the required Jeruju leaves is washed with running water until all the dirt is gone. Next it is chopped so that the surface of the jeruju leaves becomes wider so that it speeds up the drying process. The weight of the initial sample before drying is 5.2 kg. Then dried in the oven at 50°C. Drying aims to reduce the water content, so it can prevent the occurrence of enzymatic reactions and prevent a decrease in the quality of the leaves of jeruju. After drying, 1 kg of dried simplicia was obtained and obtained a yield value of 19.23%. After getting dry simplicia, the simplicia is pollinated. Pollination aims to expand the surface of the simplicia of the leaves of jeruju so that contact with large solvents so that the active compounds can be carried by the solvent.

Determination of Drying Shrinkage and Content of Jeruju Leaves Simplicia ash

Determination of drying shrinkage aims to provide minimum limits or ranges of the amount of evaporating water content or components lost in the drying process. The drying rate of the simplicia of the jeruju leaves is 9.3% indicating that the number of components of the compound is lost during the drying process.

Determination of ash content aims to provide an overview of internal and external mineral content from the initial process to the formation of extracts (MOH). Ash content is useful to determine the purity level of a medicinal raw material against an impurity in the form of foreign inorganic material found in a

plant sample. The level of jeruju leaves simplicia is 2.45%.

Extract Making

Extraction is the chemical or physical separation of a number of ingredients from simplicia using solvents. The purpose of extraction is to attract all the chemical components contained in simplicia. The extraction process was carried out by maceration method using 96% ethanol solvent. The reason for choosing 96% ethanol solvents is due to the universal nature of ethanol, so that secondary metabolites such as flavonoids (polar) and saponins (non-polar) can be completely detected. In addition, 96% ethanol will be easier to penetrate into simplicia cells than lower concentrations of ethanol solvents such as 70% ethanol, so that the resulting extract will be concentrated.

The results of maceration evaporated with a rotary evaporator at 50°C obtained a total thick extract of 66.1 grams with thick form, blackish green extract color, distinctive smell of jeruju leaves, bitter taste and obtained a yield of 14.028%.

Antifungal Activity Test of Ethanol Extract of Jeruju Leaves (*Acanthus ilicifolius*)

Determination of the Minimum Inhibitory Concentration (MIC) was carried out on some extract concentrations. The test results of the antifungal activity of the ethanol extract of the jeruju leaf showed the largest inhibition zone at a concentration of 7% ie 20 mm and a small inhibition zone at a concentration of 5% ie 9 mm. Which means that the ethanol extract of the leaves of Jeruju (*Acanthus ilicifolius*) has a strong antifungal activity in the category of *Pityrosporum ovale* at a concentration of 7% and a weak category at a concentration of 5% as a KHM value.

Positive controls in this test are used by candistatin. Candistatin was chosen as a positive control because candistatin was fungistatic and fungicidal, in vitro killing various molds and fungi. The mechanism of action of candistatin by binding to sterols in the fungal cell membrane, causing changes in membrane permeability and leakage of intracellular components that cause fungal death. From the results obtained in this test, the positive control

of candistatin shows that the diameter of the largest inhibition zone is 25 mm.

Aquadest is used as a negative control where negative controls are used to determine whether there is an effect of solvents on the growth of test fungi, so that it can be seen that the activity indicated by the extract is not the substance used in the sample. From the results of the aquadest test in this study did not show the existence of a zone of inhibition so that the inhibitory zone produced from the extract in this test originated from the activity of the substances contained in it.

Evaluation of the Physical Properties of Ethanol Extract Shampoo Jeruju leaves

Organoleptis

Organoleptic tests were carried out to determine the shape, color and odor of preparations made which were visually observed for four weeks at room temperature storage (28-300C). Organoleptic observations for four weeks at room temperature (28-300C) showed

that all shampoo formulas were stable with thick and dark green (F1), concentrated dark green (F2 and F3) and dark green (F4), this was due to concentration extract on the four different formulas the higher the concentration of extract, the more concentrated the color of the shampoo preparation. The results for the fourth odor of the F1, F2, F3 and F4 shampoo formulas for four weeks smelled of menthol, which was caused by the addition of menthol to the four formulas. The thick form of the shampoo preparation is influenced by the presence of additional ingredients in the form of Na-CMC.

PH test

pH is a parameter that can affect the absorptive power of the preparation into the skin. The pH test aims to see the acidity of the shampoo preparation and determine the safety of the preparation at the time of use. The results of testing the pH value are presented in Table 2.

Table 2. Data on pH measurements of shampoo preparations

No	Formulation	Week				pH Rata-rata ± SEM
		I	II	III	IV	
1.	F1	7.66	7.53	7.5	7.46	7.54a ± 0.043
2.	F2	7.63	7.5	7.43	7.36	7.48a ± 0.057
3.	F3	7.5	7.43	7.4	7.36	7.42a ± 0.029
4.	F4	7.46	7.43	7.36	7.3	7.39a ± 0.035

Description: The same Superskip in the same column shows nothing significant difference ($P > 0.05$) F1: Jeruju extract 5.5%, F2: Extract Jeruju 7%, F3: Jeruju Extract 8.5%, F4: Jeruju Extract 10%.

The results of pH measurements using a pH meter, in table 2 shows that during the storage process the shampoo decreases pH. The decrease in pH that occurs is due to the decomposition of the phenol group on polyphenol compounds contained in the ethanol extract of the leaves of jeruju. This decomposition causes an increase in the amount of H + so that the pH of the shampoo decreases. The higher the concentration of jeruju leaf extract, the lower the pH of shampoo. Addition of menthol to a concentration of 0.5% also causes a decrease in pH because menthol is a weak acidic phenol group, so the shampoo added with menthol causes pH to be low for four weeks of storage.

From the results of statistical tests showed that the pH of the shampoo prepara-

tion was normally distributed and homogeneous with a value of sig > 0.05. Based on statistical tests using One-Way Anova shows that the pH value of the shampoo preparation has no significant effect between F1, F2, F3 and F4, seen from the sig value > 0.05. This shows that the varied concentrations of ethanol extract of jeruju leaves have no significant effect ($P > 0.05$) on the pH value of the shampoo preparation.

Foam High Test

Foam height test aims to show the ability of surfactants to form foam. Foam from shampoo is very important. This is because foam keeps the shampoo in place on the hair, makes the hair easy to wash, and prevents hair sticks from fusing so that it causes tangles

(Mitsui, 1997). The foam height test results are presented in Table 3.

Table 3. Data on Foam Height Measurement

No	Formulation	Week				Rata-rata ± SEM
		I	II	III	IV	
1.	F1	9.5	9.16	9.1	8.3	9.01a ± 0.16
2.	F2	10.16	9.26	9.13	9.23	9.44a,b ± 0.2
3.	F3	10.5	10.16	10.03	10.26	10.23b,c ± 0.13
4.	F4	11.33	10.96	10.7	10.33	10.83c ± 0.21

Description: Different Superskip in the same column shows there significant difference (P <0.05) F1: Jeruju extract 5.5%, F2: Extract Jeruju 7%, F3: Jeruju Extract 8.5%, F4: Jeruju Extract 10%.

Based on the results of statistical tests using One-Way Anova, it was shown that the concentration of ethanol extract of the jeruju leaves that was used had a significant effect (P <0.05) on the high value of the shampoo preparation. The results of the Tukey HSD test showed that all formulations of shampoo preparations made had a foam height that was significantly different (P <0.05), where the high value of F1 foam was significantly lower than the high value of foam F2, F3 and F4. Increasing the concentration of the ethanol extract of the jeruju leaves can increase the high value of the shampoo preparation foam. This is because the ethanol extract of the jeruju leaves contains saponins. According to Harbone (1996) saponins are soapy so that the foam obtained will be more if the concentration of extract of jeruju leaves is in high shampoo.

During the four-week storage high value of foam has decreased, but a decrease in the high value of foam is not very significant. The

foam height produced from the four shampoo formulas ranged from 8 to 12 cm fulfilled the requirements for foam height according to Wilkinson (1982), namely 1.3-22 cm. The foam height does not show the ability to clean. This is more related to the aesthetic and psychological values of consumers, who like the emergence of excess foam.

Test Stability Test

Stability is defined as the ability of a product to survive within the specified specifications throughout the period of storage and use to guarantee the identity, strength, quality and purity of the product. Stable preparations are preparations that are still within acceptable limits during storage and use, where the properties and characteristics are the same as those they have when made. The Cycling test results are presented in Table 4.

Table 4. Shampoo Stability with Cycling test

Observation	Cycle To	Formulation			
		F1	F2	F3	F4
Organoleptis	I	-	-	-	-
	VI	-	-	-	-
Separation	I	x	x	x	x
	VI	x	x	x	x
pH ± SEM	I	7,73a ± 0,03	7,6a ± 0,11	7,8a ± 0,10	7,76a ± 0,08
	VI	7,5a ± 0,10	7,33a ± 0,03	7,43a ± 0,08	7,5a ± 0,10

Description: The same Superskip in the same column shows nothing significant difference (P > 0.05) F1: Jeruju extract 5.5%, F2: Extract Jeruju 7%, F3: Jeruju Extract 8.5%, F4: Jeruju Extract 10%.

The organoleptic test results of the cycling test method did not show differences between before and after the test. Before the cycling test stability test was conducted, the

organoleptic properties produced were dark green (F1), thick dark green (F2 and F3) and dark green (F4) with a menthol smell and thick form. The resulting preparation shows a ho-

mogeneous preparation. The same organoleptic properties are also shown after the stability test of the cycling test method. This shows that the organoleptic preparations produced are stable during storage.

From the results of the stability test of the shampoo, the cycling test method in terms of pH after cycling test, all formulas experienced a decrease in pH (Graph 7). Where before the pH stability test the shampoo preparation ranged from 7.4-7.9, while the pH value of the shampoo preparation after the stability test ranged from 7.3-7.7. Changes in pH value will be affected by media decomposed by the temperature at storage which produces acids or bases. The decrease in pH is affected by high temperatures, due to an increase in water in the shampoo. Despite the decline, the pH of the shampoo preparation after the cycling test still fulfills the requirements of the shampoo

preparation according to SNI (1992), namely 5.0-9.0.

Based on the results of statistical tests using One-Way Anova, it was shown that the pH value of the shampoo preparation before and after cycling test did not significantly affect the pH value of the shampoo preparation. This is seen from the sig value > 0.05 which is 0.460 for the pH value before cycling test and 0.500 for pH value after cycling test.

Moisture Test

Test the value of water content is very important to do in a shampoo product, because the water content is related to the physical shampoo and affects the shelf life of a shampoo product. The results of testing the moisture content of shampoo are presented in Table 5.

Table 5. Shampoo Water Measurement Data

No	Formulation	Replication			Rata-rata ± SEM
		I	II	III	
1.	F1	81.12	82.27	80.23	81.20a ± 0.59
2.	F2	71.69	74.06	71.23	72.32b ± 0.87
3.	F3	72.07	75.41	72.05	73.17b ± 1.11
4.	F4	72.62	75.12	71.85	73.19b ± 0.98

Description: Different Superskip in the same column shows there significant difference (P <0.05) F1: Jeruju extract 5.5%, F2: Extract Jeruju 7%, F3: Jeruju Extract 8.5%, F4: Jeruju Extract 10%.

Based on the results obtained by the water content produced, the greater the concentration of the extract added, the smaller the percentage of water content obtained. Based on the results of statistical tests using One-Way Anova showed that the concentration of ethanol extract of jeruju leaves used was significantly (P <0.05) on the value of the water content of the shampoo. The results of the Tukey HSD test showed that all the formulations of shampoo preparations made had significantly different water content (P <0.05), where F1 water content values were significantly higher

than the values of moisture content F2, F3 and F4. This states that the higher the concentration of the ethanol extract of jeruju leaves in the shampoo preparation the lower the value of the water content of the preparation.

Viscosity Test

The viscosity test aims to determine the thickness of the preparations produced. The higher the viscosity, the use of preparations on the skin will feel uncomfortable and vice versa (Osborne and Amann, 1990). The results of measuring the amount of viscosity of shampoo can be seen in table 6.

Table 6. Shampoo Viscosity Test Data

No	Formulation	Replication			Rata-rata ± SEM
		I	II	III	
1.	F1	2448.235	950.145	715.585	1371.321a ± 542.69
2.	F2	2492.26	3024.48	1694.78	3605.760a ± 386.38
3.	F3	2542.6	2797.035	2793.45	2711.028a ± 84.22
4.	F4	1556.445	1731.325	2697.115	1994.961a ± 354.68

Description: The same Superskip in the same column shows nothing significant difference (P > 0.05) F1: Jeruju extract 5.5%, F2: Extract Jeruju 7%, F3: Jeruju Extract 8.5%, F4: Jeruju Extract 10%.

In this study the viscosity of the preparation of shampoo decreased with increasing concentration of jeruju leaf extract, this is

probably because the jeruju leaf extract has a high water content of 9.95%. But this is inversely proportional to the water content of the

shampoo, the higher the ethanol extract of the jeruju leaves, the lower the value of the shampoo water content because the less water is added to the shampoo preparation.

According to Schimit and William (2006) the viscosity of shampoo preparations should be in the range of 400-4000 cps. In this study the dosage viscosity produced ranged from 1371,321 - 3605,760 cps. This means that the viscosity of the preparation of formulas 1, 2, 3 and 4 shampoo meets the desired viscosity criteria. The higher the concentration of jeruju leaf extract is added, the lower the value viscosity obtained. This is because the water content of the jeruju leaf extract is high so that more water is contained in the shampoo preparation.

Judging from the results of the analysis of the viscosity data, the shampoo is normally

distributed and homogeneous with a sig value > 0.05, which is 0.115. The statistical test using One-Way Annova shows that the value of the shampoo's viscosity does not significantly affect F1, F2, F3 and F4, this is seen from the sig value > 0.05, which is 0.151. It means that the various concentrations of the ethanol extract of the jeruju leaves in the shampoo preparation do not affect the value of viscosity.

Type Weight Test

Specific gravity is one of the physical analyzes performed to determine the stability of a preparation during the storage period, with known density, it can also be seen the purity value of a preparation, especially the preparation in the form of a solution (Ansel, 1989).

Table 7. Measurement Data for Weight Types of Ethanol Extract of Jeruju Leaves

No	Formulation	Replication			Rata-rata ± SEM
		I	II	III	
1.	F1	1.048	1.059	1.052	1.053a ± 0.003
2.	F2	1.019	1.029	1.050	1.032a ± 0.009
3.	F3	1.012	1.014	1.050	1.025a ± 0.012
4.	F4	1.062	1.056	1.052	1.056a ± 0.002

Description: The same Superskip in the same column shows nothing significant difference ($P > 0.05$) F1: Jeruju extract 5.5%, F2: Extract Jeruju 7%, F3: Jeruju Extract 8.5%, F4: Jeruju Extract 10%.

The results of the measurement of the specific gravity of the antidandruff shampoo in the ethanol extract of the jeruju leaf can be seen in Table 7. The measurement of the fourth type weight of the shampoo preparation formula is between 1.012-1.062 gram / ml, which means the weight of the preparation of the shampoo ethanol extract is still within the permissible limits. There is an increase or decrease in the value of the type of weight of the shampoo preparation for jeruju leaf extract because the picnometer used during the evaluation is not equipped with a thermometer as a temperature regulator, as stated in Indonesian Pharmacopoeia Edition IV.

Judging from the data analysis, the specific gravity of the shampoo is distributed normally and homogeneously with a sig value > 0.05. Based on statistical tests using One-Way Annova shows that the value of the type of shampoo preparation does not significantly

affect F1, F2, F3 and F4, this is seen from the sig value ($P > 0.05$). It means that the various concentrations of the ethanol extract of the jeruju leaves in the shampoo preparation did not affect the value of specific gravity. Species weight is influenced by the components in the preparation. The more components there are, the higher the weight fraction. So that the density will also be higher (Martin *et al.*, 1993).

Hedonic Test

The hedonic test was carried out on 15 panelists which included an assessment of the characteristics of the shampoo preparation namely appearance, aroma and color. The hedonic test results obtained are tabulated and determined the interval of quality values using the formula by looking for the average for the panelists with a confidence level of 95% (Tranggono and Latifah, 2007). The hedonic test results can be seen in Figure 1.

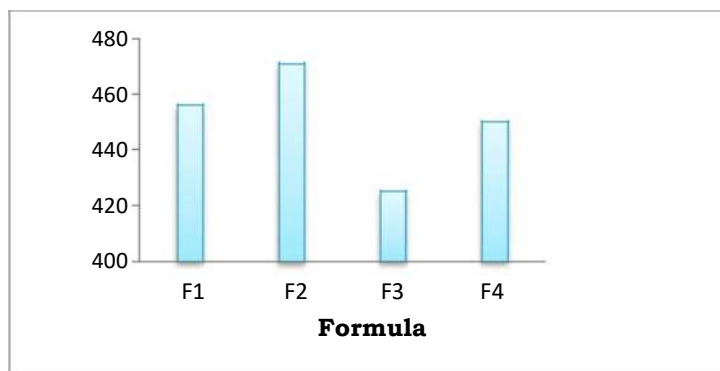


Figure 1. Hedonic Test Chart of 15 Panelists

In the hedonic test conducted on 15 panelists, it was found that the total score of the questionnaire (Appendix 6) shampoo with a concentration of 7% (F2) was more in demand than shampoo with a concentration of 5.5% (F1), 8.5% (F3), and 10% (F4).

Antidandruff Activity Test of Ethanol Extract of Jeruju Leaf

The shampoo formula for the ethanol extract of the jeruju leaves that was made was tested for its activity against the fungus *Pityrosporum ovale* using the well diffusion method and using SDA as a growth medium. The well method is used because it is easier to measure the extent of the inhibitory zones that are formed because isolates are active not only on the upper surface of the agar but also down (Saputro, 2006). Also because the results of the well method will be easier to see and show more tangible results (Warsa, 1994).

Positive control in this test is used head and shoulder shampoo. From the results obtained in this test positive controls showed the largest inhibition zone diameter with an average of 25.33 mm compared to the shampoo extract of the jeruju leaves.

Aquadest is used as a negative control where negative controls are used to determine whether there is an effect of solvents on the growth of test fungi, so that it can be seen that the activity shown by the extract is the substance contained in the sample not from the solvent used. From the results of the aquadest test in this study did not show the existence of a zone of inhibition so that the inhibitory zone produced from the extract in this test originated from the activity of the substances contained in it. The results of the testing of the anti-dandruff activity of the jeruju leaves ethanol extract can be seen in table 8.

Table 8. Antifungal Test Results for Ethanol Extract of Jeruju Leaves

No	Formulation	Replication			Rata-rata ± SEM
		I	II	III	
1.	F1	15	15	14	14.66a ± 0.33
2.	F2	19.5	19.5	20	19.66b ± 0.16
3.	F3	21	20	20	20.33b,c ± 0.33
4.	F4	20.5	22.5	22.5	21.83c ± 0.66
5.	K+	25	25	26	25.33d ± 0.33
6.	K-	0	0	0	0

Description: Different Superskip in the same column shows there significant difference (P <0.05) F1: Jeruju extract 5.5%, F2: Extract Jeruju 7%, F3: Jeruju Extract 8.5%, F4: Jeruju Extract 10%.

Based on the results obtained in the anti-dandruff shampoo formula, the largest inhibition zone was found in antidandruff shampoo containing *Acanthus ilicifolius* extract with a concentration of 10% (F4), while the lowest inhibition zone was found in

Antidandruff shampoo containing *Acanthus ilicifolius* extract with a concentration of 5.5% (F1). This shows that the higher the concentration of ethanol extract duan jeruju, the greater the active substance contained in it

which can inhibit the growth of *Pityrosporum ovale* fungi and is characterized by the greater diameter of the inhibitory zone formed.

The results of the statistical analysis One-way Annova showed that the antidandruff shampoo of *Acanthus ilicifolius* extract had antifungal activity against *Pityrosporum ovale*. Based on the One-way Annova analysis data, the sig value was obtained (P <0.05), which means that there were significant differences in the diameters of the inhibi-

tory zone in each treatment. Tukey's advanced test results show control (-), control (+) and F1 provide different antifungal activities. The highest inhibitory zone diameter was obtained in the positive control (+) and the lowest inhibition zone diameter in the negative (-) control which showed significantly different from *Acanthus ilicifolius* extract shampoo with a concentration of 7%, 8.5% and 10%.

CONCLUSIONS

The ethanol extract of the leaves of Jeruju (*Acanthus ilicifolius*) has anticorruption activity against *Pityrosporum ovale*. The best formulation is formula 4 (10%) which can inhibit the fungus *Pityrosporum ovale*. The higher the concentration of ethanol extract of the leaves of jeruju, the better the anti-dandruff activity in inhibiting the *Pityrosporum ovale* fungi.

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