

Phytochemical Screening and Evaluation of Transparent Soap with Addition of Wedi Snakefruit (*Salacca zalacca*) Rind Extract

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Abstract. Wedi snakefruit (Salacca Zalacca) was a native fruit from Bojonegoro District. Snakefruit contains secondary metabolites that act as antioxidants, this allows the rind of snakefruit to also contain the same secondary metabolites. Antioxidants in snakefruit rind can be used as a source of natural antioxidants for the transparent soap. Transparent soap with natural antioxidant is expected to prevent damage caused by free radicals in the skin. This research aims to evaluating the secondary metabolites from wedi snakefruit (Salacca zalacca) rind extract and to knowing the effect of wedi snakefruit (Salacca zalacca) rind extract addition toward transparent soap quality. The research samples were collected at Wedi Village, Bojonegoro. The extract of wedi snakefruit (Salacca zalacca) rind was prepared by maceration method using ethanol 96% as a solvent and followed by evaporating using rotary evaporator. The secondary metabolites of crude extract was analyzed by phytochemical screening with standard methods. Phytochemical screening performed to determine the pharmacological active ingredient such as flavonoids, saponins, alkaloids, steroids and triterpenoid, especially those with antioxidant activity. The analysis on quality of transparent soap were sensory evaluation test, irritation test, foam height and stability, pH and water content. The result showed that the chemical compounds of wedi snakefruit (Salacca zalacca) rind extract positive for flavonoids, saponins, alkaloids, steroid and triterpenoids. Transparent soap with addition of wedi snakefruit (Salacca zalacca) rind extract have a yellow, transparent and distinctive smell of fragrance oil, irritation value 0, foam height between 1.3 and 5.2 cm that were no significantly different and stable after 5 min of testing, pH between 9.5 and 10. Wedi snakefruit (Salacca zalacca) rind extract compounds the secondary metabolites that act as antioxidant. All the results of the evaluation of transparent soap according to the parameters Indonesian National Standard (SNI).

Keywords: phytochemical screening · evaluation of transparent soap · wedi snakefruit (Salacca zalacca) rind extract

1 Introduction

Wedi snakefruit (Salacca zalacca) was a native fruit from Bojonegoro District. Its a hereditary heritage, growing in almost every yard belonging to the community of Wedi village and Tanjungharjo Village. There is available snakefruit plantation area for 75 ha and production of snakefruit in 2013 reaching 19,511 quintals [5]. Wedi snakefruit has its own characteristics and has different taste from the other snakefruit variety. Wedi snakefruit (Salacca zalacca) has a potential role in the economic and social for surrounding community [16]. Snakefruit contains secondary metabolites that act as antioxidants, this fact also allows the rind of snakefruit also contain the same secondary metabolites as natural antioxidant. Previous study showed that phytochemical test of snakefruit rind contain flavonoid, tannins and a little alkaloid. Based on the value of antioxidant capacity and IC50, snakefruit rind extract has high potential to be marketed as products with high antioxidants content, it can also increase the economic value of snakefruit, especially Wedi snakefruit [8].

The antioxidants are chemical substances that inhibit or block the process of formation of free radicals. The use of antioxidants can also be considered important or maintaining the stability of the formulations. The topical application of antioxidants may represent an attractive strategy for skin protection against oxidative stress, prevents skin aging and other damage caused by free radical [6]. Antioxidants in snakefruit rind can be used as a source of natural antioxidants for soap. Natural antioxidants in the form of plant extracts are typically added as additives to suppress the oxidation of polyunsaturated fatty acids in soaps [14].

Soap is the product of saponification of fats with a base, which is used as a body cleanser, in the form of solid, foamy, with or addition of other ingredients and do not cause skin irritation [11]. Soap is a cleaning agent that have a various types of soap, such as bar soap, liquid soap, opaque soap (non-transparent solid soap), translucent (transparent but not see-through soap), and transparent soap. Transparent soap is a type of soap for the face and body to cleanse and protect the skin from free radicals [7]. Transparent soap is one of the innovative soap products that makes soap more interesting, more aesthetic and more expensive than ordinary bar shapes. This research aims to evaluating the secondary metabolites from Wedi snakefruit (Salacca zalacca) rind extract and to evaluating characteristics of transparent soap with addition of Wedi snakefruit (*Salacca zalacca*).

2 Methods

Research design was used to analyze the research data that was completely randomized design (CRD) with 4 treatment and 5 repetitions. The observations were consist of the observation of snake fruit rind extract and observations transparent soap added snake fruit rind extract. Observation of snake fruit rind extract was consist of a phytochemical screening of an antioxidant activity, while the observation of transparent soap were sensory evaluation test, irritation test, foam height and stability, pH and water content.

The extract of Wedi snakefruit (Salacca zalacca) rind was prepared by maceration method using ethanol 96% as a solvent. Maceration was carried out for 3 day with

stirring occasionally and changing the solvent every 1×24 hours. Maserat that obtained then filtered and concentrated using a rotary evaporator at a temperature of 550C [1]. The secondary metabolites of crude extract was analyzed by phytochemical screening. Screening for flavonoid by shinoda test (HC test), few fragments of Mg and dropwise HCl were added to 1 ml plant extract, which gives pink reddish/brownish pink or green or blue colour in few minutes. Screening of saponin by foam test, a small amount of extract was shaken with water and observed for the presence of foam. Screening of alkaloids was carried out by Mayer and Dragendorf test. Mayer test, 1ml of sample was added to a few drops of Mayer's reagent. Formation of white or pale yellow precipitate indicates the presence of alkaloids in the sample. Dragendorf test, 5ml of distilled water was added to the 2 ml of sample, then 2N HCl and 1 ml of Dragondrof's reagent was added. Orange/orange red precipitate indicates the presence of alkaloids. Test for steroids by salvoski test, 1 ml of test sample was dissolved in 1 ml of chloroform and equal amount of concentrated H2SO4. Formation of Bluish red to cherry colour in chloroform layer shows the presence of steroids. Test for triterpenoids by 1ml of plant extract, 2ml of chloroform and 3ml of conc H2SO4 was added. A reddish brown precipitate at the interface, confirmed the presence of triterpenoids [21]. Evaluation of antioxidant activity was carried out quantitatively using the DPPH method, 2 ml of 40 ppm DPPH solution was pipetted and put into a test tube. Added 2 ml of test solution of each concentration (20; 40; 60; 80; and 100 ppm) into a test tube, then vortexed and allowed to stand for 30 min. Measurement were performed by using a spectrophotometer with absorbance readings on wavelength 518 nm. The IC50 value will be obtained by calculation by linear regression where the sample concentration (ppm) is the abscissa (x-axis) and the percentage value of attenuation is the ordinate (y-axis) [18].

The process of making soap using the hot method with a water bath as the medium. Coconut oil and olive oil that had been placed in a glass beaker were heated in a water bath. Stearic acid was added and stirred until homogeneous. Then, 30% NaOH solution was added. Other supporting ingredients including 96% ethanol, glycerin, sucrose, coco-DEA, NaCl, and fragrance oil were added and stirred until the entire mixture was completely mixed. For the addition of rind extract, the temperature of the soap mixture was lowered to ± 50 °C. Stirring step was re-carried out until the extract was completely mixed and then the solution was poured into a silicone mold. This was then allowed to stand for 24 h at room temperature. After 24 h, the curing process was carried out for ± 3 weeks [24].

The analysis on quality of transparent soap were sensory evaluation test, irritation test, foam height and stability, pH and water content. Sensory evaluation was determined using 10 untrained panellists in the color, aroma and shape of the soap. Irritation test measured by observed the symptoms irritation on skin that occurred after 1 h soaked soap applied to the skin. Foam height and the foam stability measured by the height of the formed foam. 1 mL of liquid soap was diluted with 9 ml of distilled water, then homogenized using vortex for 20 s. The homogenized soap was then precipitated for 10 min before the height of the formed foam was measured. The pH test was carried out by measuring the pH using a pH meter. Calibrated pH meter using standard which was pH 7 buffer solution. Electrodes were washed with distilled water, then dried with paper towels. Measurement of pH soap done by diluting 1 g of soap with 10 ml of distilled

water in a cup, then the electrode was dipped into the solution and let it move up to the position of a constant number determined pH of soap. Water content measured by ± 4 g of sample which has been prepared which has been known cup its weight. Preheat to oven at 105°C for 2 h until the weight remains. Water contain is calculated using the standard formula [3].

3 Results and Discussion

The process of making soap applied the hot method using a water bath as a heat source. The soap was made with four formulations by differentiating in the amount of extract and the type of oil used in each formulation. The formulation was developed from the transparent solid soap formulation that had been carried out in previous studies, but was modified in the use of oil. The original formulation used palm oil while the research formulation used a combination of coconut oil and olive oil. Coconut oil was chosen because it contains lauric acid between 44 and 52%. The lauric acid content affects the characteristics of the soap foam, while olive oil contains oleic acid which can moisturize the skin [9]. Soap formulations are presented in Table 1.

Phytochemical Screening of Wedi Snakefruit (Salacca zalacca) Rind Extract

Phytochemical screening test was carried out to determine the secondary metabolites contained in a sample. Phytochemical screening carried out included examination of flavonoids, saponins, alkaloids, steroid and triterpenoids. The results of the phytochemical screening showed that the Wedi snakefruit rind extract was positive for alkaloids, flavonoids, saponins and triterpenoids. The results of phytochemical screening tests for Wedi snakefruit rind extracts are presented in Table 2.

Material	Formula 1	Formula 2	Formula 3	Formula 4
Coconut Oil	30	30	30	30
Olive Oil	30	30	30	30
Stearic Acid	21	21	21	21
NaCl 30%	0.6	0.6	0.6	0.6
Ethanol 96%	45	45	45	45
Sugar syrup	45	45	45	45
Aquadest	23.7	23.7	20.7	20.7
Glycerin	39	39	39	39
Coco-DEA	3	3	3	3
Fragnance oil	0.3	0.3	0.3	0.3
Snakefruit rind extract	0	1.5	3	4.5

Table 1. Formulation Of Transparent Soap

Test	Procedure	Observations	Conclusions	
Flavonoids	Extract + Mg + HCl will gives pink reddish/brownish pink or green or blue colour	A green colour	Positive flavonoids	
Saponins	Extract was shaken with water and observed for the presence of foam	Persistent foam	Positive saponin	
Alkaloids	Extract + mayer reagent formation of white or pale yellow precipitate Extract + dragendorf reagent gives orange/orange red precipitate	Formation of white preciptate An orange colour	Positive alkaloid	
Steroid	Extract + chloroform +	brown precipitate at thr	Positive triterpenoid	
Triterpenoids	H_2SO_4	interface,		

Table 2.	The Phytocher	nical Screening	of Wedi	Snakefruit Extract

Transparent Soap Antioxidant Test Results

Antioxidant test of transparent soap was carried out using the DPPH method. A 2 ml of 40 ppm DPPH solution was pipetted, put into a test tube, added with 2 ml of test solution of each concentration (20; 40; 60; 80; and 100 ppm), vortexed and allowed to stand for 30 min. After that, the absorbance was at the maximum wavelength with a UV-Vis Spectrophotometer alternately at the five concentrations. Each absorbance is recorded and then the percentage of attenuation is calculated by the formula. From the value of the percentage attenuation at each concentration, a regression curve was made. From the linear regression curve IC50 of transparent soap was obtained by the calculation of the 50% damping percent. The IC50 value of transparent soap showed that the transparent soap with addition of wedi snakefruit (Salacca zalacca) rind extract has antioxidant activity in the weak category.

The secondary metabolites that have an antioxidant activity such as flavonoid, saponin, alkaloids and triterpenoid. The antioxidant activity of natural flavonoids is primarily exerted by phenolic hydroxyl groups. Flavonoid have antioxidant properties as high as typical phenolic compounds such as quercetin, trans-resveratrol, trolox, and ascorbic acid. Mechanism of antioxidant activity of flavonoid is not only scavenge free radicals but also reduce free radical formation [22]. Saponins have potential as antioxidants through their ability to reduce superoxide thereby preventing biomolecular damage by free radicals [20]. Wedi snakefruit rind positively contain flavonoid, saponin, alkaloids and triterpenoid. This shows that Wedi snakefruit rind has antioxidant activity. Many cosmetics that are marketed nowadays often contain antioxidants as the active ingredients. Topical antioxidants could terminate the chain reactions by removing the

free radical intermediates and inhibit other oxidation reactions by being oxidized themselves; this could defend the skin against the environmental stress caused by free radicals [12]. Wedi snakefruit rind can produce natural antioxidant compounds that could control the oxidative stress caused by sunlight and oxygen on skin.

The color of the transparent solid soap comes from the oil used. Coconut oil is white in color and olive oil is yellow in color, so formulations that use a combination of coconut oil and olive oil cause the color of the transparent solid soap to turn yellow color, due to color of olive oil. The difference in the amount of snakefruit rind extract of Wedi variety did not affect the color because the percentage of extract was 1.5-4.5% w/v. The smell of the soap produced was fragrant, but not the typical smell of snakefruit rind extract. The smell of snakefruit rind extract produced was quite distinctive, but when it was formulated into a transparent soap, the smell of the extract was eroded by the smell of fragnance oil, so that the end result of the soap had a distinctive aroma from the used fragnance oil. The difference in the type of oil used as a transparent solid soap base and the difference in the amount of snakefruit rind extract of the Wedi variety did not affect the odor because the oil used was generally odorless, and the percentage of extract was only 1.5-4.5% of the total formulation. The organoleptic test results on transparent solid soap stored at room temperature for 4 weeks showed that all preparations did not change. Therefore, transparent solid soap preparations in all formulations during storage at room temperature (25-30°C) were organoleptically stable. Organoleptic testing also has a goal to measure the level of fulfillment of aspects that are acceptable (acceptability) by the community as consumers [26]. The results of organoleptic tests that are stable on storage can increase the level of acceptance (acceptability) of transparent soap with snakefruit rind extract of wedi variety by the community.

Irritation test results of all formula was 0, it showed that transparent soaps added with snake fruit rind extract were safety, no irritation detected on skin. Surfactants and other irritants initially interact with the stratum corneum. These responses can take different forms, including sensory irritation, damage to the surface corneocytes and super-hydration of the stratum corneum. There are many mechanisms by which such sensations are produced. Some personal care products. Sensory irritation can frequently be detected before clinical signs can be observed. The epidemiological study reported that many people experience sensory irritation in the absence of visible signs and discontinue use of that product before visible irritation appears [23]. Even though the irritation value of transparent soap with addition of snakefruit rind extract is 0, it could be that the soap still causes irritation to the skin.

Foam is a colloidal system with a gas dispersed phase and a liquid dispersion medium [15]. The foam height test was carried out to see the stability of the resulting soap foam. The height of the soap foam in all formulations ranged from 1.3 to 5.2 cm and met the requirements for good soap foam (1.3–22 cm) on SNI standard. The foam on transparent soap is influenced by the substance in the oil used, the speed and time of stirring and the use of surfactants [2]. The resulting transparent solid soap has a foam height met the SNI requirement because it uses coconut oil in all formulations. Coconut oil contains high lauric acid which functions to harden, clean and produce soft foam in soap formulations and uses coco-DEA which acts as a surfactant to increase the stability of the resulting transparent soap foam [4].

The pH of the transparent solid soap of snakefruit rind ranges from 9.5 to 10. These results indicate that soap has a good pH because it is on accordance to SNI, which is a good soap must has a pH in the range of 9–11 [19]. Soaps with pH of 9–10 do not need the addition of antibacterial agents because the pH range are not favored by bacteria and microbes. Solid soap, including transparent solid soap, is a product of the saponification reaction requires a certain amount of alkali for a complete reaction to occur. The high and low pH of soap is influenced by the saponification process during soap making [10]. The pH value of the transparent solid soap produced indicates that the formulation used is proper, especially in the amount of oil and the amount of alkali used because it produces soap in the pH range that is safe for the skin and free from bacteria and microbes. The in the percentage of Wedi snakefruit rind extract did not affect the pH although there are different values in each formulation.

Water content can affect soap characteristics such as hardness. Various soap hardness textures influenced by the amount of water contained in different soaps. There is the water content in SNI has a maximum quality requirement of 15% fraction mass [10]. This research showed that water content of transparent soap between 8.47 and 14.01 in accordance to SNI standard.

4 Conclusions

Wedi snakefruit (Salacca zalacca) rind extract compunds the secondary metabolites that have an antioxidant activity such as flavonoids, saponins, alkaloids and triterpenoids. All the results of the evaluation of transparent soap in accordance with Indonesian national standards (SNI).

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