FORMULATION OF THE BODY SCRUB CREAM CONTAINING MORINGA SEED POWDER (*Moringa oleifera*) AND ITS EXAMINATION DERMAL ACUTE IRRITATION

Hendrawati1, Annita Karunia Savitri1, Nina Fitriyati1, Aulia Andi Mustika2

1Departement of Chemistry Faculty of Science and Technology State Islamic University Syarif Hidayatullah Jakarta, Indonesia
2Faculty of Veterinary Medicine, Bogor Agricultural University, IPB, Indonesia

*Corresponding Author, Received: 15 Jan. 2018, Revised: 30 April 2019, Accepted: 25 May 2019

ABSTRACT: Moringa seeds (*Moringa oleifera*) have many substances such as antioxidants, minerals and vitamins that are useful for skin health. Moringa seed could be used as additional material of cosmetic. One of the misscrub cream by using moringa seed as abrasive material (abrasiver). The purpose of this research is to know the optimal formulation of scrub cream moringa seed and characterization based on the requirement of quality skin moisturizers according to SNI 16-4399-1996 and the safety of its use by conducting dermal acute irritation test. The dermal acute irritation test was performed by the Draize test method on white rats (*Rattus norvegicus*) male sprague-dawley strain. The results of the study show that the most optimum scrub cream products based on panelist preferences level are scrub cream with the addition of 4.5% moringa seed with a score of 5.67. Formulation scrub cream with the addition of 3.5; 4.5; and 5.5% moringa seed qualify skin moisturizers according to SNI 16-4399-1996 with pH value of 6.88-7.31; density 1 g/mL; emulsion stability of 95.19-96.61%; and negative microbial contamination. The three formulations of moringa seed scrub cream also do not give skin irritation effect to test animals at 24 and 48 hours with primary dermal irritation index or PDII=0.

Keywords: Body scrub cream, Bioseed, Draize test, Moringa oleifera, Cosmetics

1. INTRODUCTION

Moringa seeds produce oil commercially known as the behen oil. The oil is widely used in beauty products because it has a high antioxidant content with an IC₅₀ value of 91.13 μg/mL [1] – [2]. Moringa seeds are also known to have nutrients such as vitamins and minerals that are beneficial to skin health. Compahore et al [3] state that moringa contains high enough minerals such as calcium, copper, phosphorus, zinc, magnesium, manganese, potassium and sodium. The mineral content of calcium, sulfur, magnesium, and potassium can lift dead skin cells (exfoliator) and nourish the skin so the skin feels soft and smooth [4]. Moringa seeds are rich in vitamins B1, B2, B3, C, and E [5]. Vitamin B1, B2, B3 are known to have a role in maintaining skin moisture and brightening the skin [4].

Burlando et al. [6] state that moringa seeds have potential as raw materials in cosmetic because they have a high nutrient content that is beneficial to the skin. Cosmetics has now become a necessity that is considered important for some people. Various types of cosmetic products are used for skin care in order to appear more attractive. Ojiako and Okeke [7] utilize high antioxidant content in moringa seed oil in body lotion, while Duraivel et al. [8] utilize moringa seed oil in an anti-wrinkle cream. Cosmetic product used for other skin care is scrub cream.

Based on the content of vitamins, minerals, and high antioxidant content in moringa seeds, researchers have an interest in using moringa seed powder as an active component of skin abrasive and also as a nutritional addition to cream scrub. The effort to add moringa seed powder in scrub cream has never been done before. Scrub cream is made by varying the concentration of moringa seed powder 3,5; 4,5; and 5.5%, followed by organoleptic test by 30 panelists and characterization test (pH analysis, emulsion stability, species weight, and total microbial contamination) in accordance with the quality standard of National Standardization Body 1996. Body scrub cream fulfills the quality requirement of Agency National Standardization 1996 followed by dermal acute irritation test.

The content of the substances used for the production of scrub cream has the possibility to cause irritation to the skin, therefore to know the safety of the use of cream scrubs on the skin, then this study conducted dermal acute irritation test [9]. This test uses the white rat rats (*Rattus norvegicus*) Sprague-Dawley strain with the method Draize [10]. The Draize method is done by observing the reaction of erythema and edema in the skin that occurs after cream scrub products applied to the skin of test animals for 24 hours and 48 hours.
2. METHOD

2.1 Time and Place of Implementation

The research was conducted in January until September 2017 in Chemical Laboratory, Central Integrated Laboratory (PLT), Syarif Hidayatullah State Islamic University Jakarta and Laboratory Animal Laboratory Unit (UPHL), Faculty of Veterinary Medicine, Bogor Agricultural University.

2.2 Tools and Materials

The tool used in this research is oven (Memmert), analytical scale (Ohaus), pH meter (Martini MI 150), incubator (Lequeux), bath (Heidolph MR 3001 K), micro tube (Eppendorf), petri dish (Pyrex), vortex mixer (Thermolyne M 16700 Maxi), micropipet (Socorex), filter paper 0.45 μm (Whatman), thermometer (Boeco), magnetic stirrer, spirits, stopwatch, glassware, organoleptic test equipment, sterile gauze (DRC) hypoallergenic plaster (Mikropore), and hair shaver (Wahl).

The materials used in this study were Moringa oleifera samples obtained from Pamulang, Ciputat, South Tangerang, stearic acid, cetyl alcohol, triethanolamine, glycerin, methyl paraben, propyl paraben, isopropyl mercate (BratacoChemika), fragrance, aquadest, commercial cream scrubs for comparison, sterile NaCl, plate count agar, and white rat test animals (Rattus norvegicus) Sprague-Dawley strains of male sex with age 2-3 months and weight 200-250 grams (UPHL FKH IPB). Approval of ethical review for experimental animals (rats) in this study was obtained from Animal Ethics Committee, Faculty of Veterinary Medicine, Bogor Agricultural University, with number 076 / KEH / SKE / XI / 2017.

2.3 Procedures

2.3.1 Preparation of moringa seed [11]

Moringa selected fruit that is dark brown then taken the seeds. Seeds selected moringa good quality (has a round and whole shape dry) then the seeds moringa peeled skin. The contents of moringa seeds are dried with the oven at 60 °C for 1 hour. The content of moringa seeds was then smoothed using a blender and sieved with a particle size filter of 595-420 μm according to Yuliati and Binarjo research [12].

2.3.2 Formula scrub cream [13]

The preparation of a cream base is carried out according to the composition of the formula shown in Table 1. F0; F1; F2; F3 is a cream scrub formulation with the addition of moringa seed powder respectively 0; 3.5; 4.5; and 5.5% (w / w) of the dough weight.

<table>
<thead>
<tr>
<th>Material</th>
<th>Weight</th>
</tr>
</thead>
<tbody>
<tr>
<td>Glycerin</td>
<td>15</td>
</tr>
<tr>
<td>Stearic acid</td>
<td>12</td>
</tr>
<tr>
<td>Cetyl alcohol</td>
<td>4</td>
</tr>
<tr>
<td>Triethanolamine</td>
<td>3</td>
</tr>
<tr>
<td>Isopropyl misle</td>
<td>2</td>
</tr>
<tr>
<td>Propylparabene</td>
<td>0,02</td>
</tr>
<tr>
<td>Fragrance</td>
<td>0,2</td>
</tr>
<tr>
<td>Distilled water, up to</td>
<td>100</td>
</tr>
</tbody>
</table>

The method of making moringa seed cream scrub is as follows stearic acid and cetyl alcohol which is the oil phase mixed and melted in porcelain cup until it reaches 70 °C above water bath, after a perfect melt the temperature is lowered to 65 °C then fed into a cup of trophies and then added propyl paraben and isopropyl impregnate into the oil phase mixture while stirring until homogeneous (Dough 1). Glycerin and water which is a water phase mixed and heated to temperature 80°C in different containers and then cooled to a temperature of 65 °C while slowly incorporating triethanolamine (Dough 2). Dough 1 and 2 are mixed while stirring until a fine cream emulsion (dough 3) is formed. Dough 3 is allowed until the temperature drops to 40 °C. Moringa flower dust and powder with concentration variation (b/b) is added while continuous stirring. A cold cream scrub is inserted into a plastic bottle.

2.3.3 Organoleptic test scrub cream [14]

Organoleptic test conducted on herbal cream products includes color, aroma, consistency, texture, and overall product acceptance. Sample used include Moringa seed cream scrub and cream scrubs on the market (commercial). Panelists who conducted the organoleptic test were 30 untrained panelists. The result of the organoleptic test was processed by a statistical method using SPSS application with one-way Anova test method.

2.3.4 Characterization scrub cream

Analysis of the scrub cream produced includes analysis of pH [15], the stability of the emulsion...
[15], the specific gravity [16] and total microbial contamination [17]. As a comparison used commercial cream scrubs. Scrub cream characterization test results processed with statistical methods using SPSS with one way ANOVA test method.

2.3.5 Acute irritation dermal scrub cream [10] – [18]

The number of rats required for each group is determined by Federer's (n-1) (t-1) ≥15, where t represents the number of treatments and n is the number of replications. This research has 6 treatments to get the number of rats each treatment 4 mouse (value n≥4). Samples tested were cream base, 3 moringa cream seed scrub; 4.5; 5.5%. The normal control treatment is mice without treatment.

The acclimatized mice for 14 days had their heads shaved with an electric razor on the back with an area of 3x3 cm until cleaned up before the sample application. The shaving is done carefully so as not to injure the rats' backs. The test material is applied to the back of the mice that have been shaved hair of 0.3 grams per head, then covered with sterile gauze and glued with hypoallergenic plaster. The rats were then placed and left in individual cages. The plaster and sterile gauze are opened and the test area rinsed with water after 24 hours. Observation on mouse skin was done after 40 minutes of plaster and the sterile gauze was opened and then scoring of erythema and edema formed based on Draize Method. Observations continued at 48 hours after treatment (mice smeared test material).

3. RESULT AND DISCUSSION

3.1 Organoleptic Test Scurb Cream

Here is the result of organoleptic test scrub cream produced in this study.

![Fig. 1 Result of organoleptic test scrub cream](image)

The results of the color organoleptic test showed that the highest panelist's favorite level was on the cream formula without the addition of Moringa seed powder (K0%) of 5.9. Formula cream without Moringa seed dust has a typical white cream scrub color. This indicates that the panelists prefer a creamy white scrub. Cream formula with the addition of moringa seed powder 3.5; 4.5; 5.5% (w/ w) respectively decreased favor rate by 5.20; 4.50; and 4.03. This indicates that the more concentrated the color of herbal cream scrubs the panelist's favorite level decreases.

Table 2. Organoleptic test results of cream scrub

<table>
<thead>
<tr>
<th>Parameter</th>
<th>F0</th>
<th>F1</th>
<th>F2</th>
<th>F3</th>
<th>KK</th>
</tr>
</thead>
<tbody>
<tr>
<td>Color</td>
<td>5.9</td>
<td>5.2</td>
<td>4.0</td>
<td>4.0</td>
<td>5.1</td>
</tr>
<tr>
<td>Aroma</td>
<td>5.1</td>
<td>5.4</td>
<td>5.5</td>
<td>5.6</td>
<td>5.2</td>
</tr>
<tr>
<td>Viscosity</td>
<td>4.1</td>
<td>4.4</td>
<td>5.7</td>
<td>4.3</td>
<td>4.6</td>
</tr>
<tr>
<td>Texture</td>
<td>3.6</td>
<td>4.7</td>
<td>5.3</td>
<td>4.9</td>
<td>5.67</td>
</tr>
<tr>
<td>General</td>
<td>2.60</td>
<td>4.9</td>
<td>5.7</td>
<td>4.3</td>
<td>6.0</td>
</tr>
</tbody>
</table>

Note: F0 = formulation scrub cream without addition of moringa seed scrub, F1 = formulation scrub cream with addition of 3.5% (w/w) moringa seed scrub, F2 = formulation scrub cream with addition of 4.5% (b/w) moringa seed scrub, F3 = formulation scrub cream with addition of 5.5% (w/w) Moringa seed scrub, KK = commercial scrub cream.

3.1.1 Color

Based on statistical test results using one-way Anova, there are significant differences in the color preferences level of each treatment is indicated by a probability value of 0.000 (P <0.05). According to Diana and Thaman[19] color formed on the product is affected by the color of its constituent materials. Moringa seed powder that is added to the formula was instrumental in giving color to the product. Moringa seed powder has a brownish yellow color. The more moringa seed powder added then the color of cream scrubs formed will be more brown.

3.1.2 Aroma

The smell of cream scrub products is influenced by the ingredients. The most influential ingredients give the flavor of Moringa seed powder, so it is added fragrance to the formula to reduce the aroma from the sample of moringa. The scent produced by cream scrub depends on the concentration of the moringa seed powder. The more moringa seed powder added to the formula the stronger the aroma. Based on the organoleptic test, the highest scores were obtained in F3 cream scrub with a score of 5.63 followed by cream scrub F2; F1; LK; and F0 with a score of 5.53; 5.43; 5.20; and 5.10.

Based on statistical test results using one-way Anova, there is no significant difference in aroma preferences level of each treatment which is shown with a probability value of 0.434 (P> 0.05). This suggests that the addition of moringa seed powder in the cream formula does not affect the panelist's preference level. This is because the panelists who conduct the assessment are panelists who are not trained so as not to be sensitive to the aroma of the products produced.
3.1.3 Viscosity

Based on the results of the organoleptic test, the most preferred scrub cream by the panelists is the formula scrub cream with the addition of 4.5 mole seed (F2) with an average score of 5.67. The result of the statistical test using one-way anova shows that there is a significant (mean) difference between the viscosity favorite level of each treatment. It is shown with the probability value of 0.001 (P <0.05).

Generally, panelists prefer a product scrub cream that is not too thick. The more moringa seed powder added to the formula will make the more thickness of the scrub cream. Likewise, when the less the Moringa seed powder, will be shaped less scrub and look watery. This is because Moringa seeds have stearic acid content of 12.62% [20]. Stearic acid is an emulsion stabilizer and a thickening agent which is useful in forming a thickened or semi-solid (cream) [21].

3.1.4 Texture

Based on organoleptic tests, scrub cream F2 with a score of 5.3 is more preferable to scrub cream F3 and F1 with a score of 4.9 and 4.73. Based on statistical test results using one-way Anova, there is a significant difference in texture preferences level of each treatment is indicated by the probability value of 0.000 (P <0.05). This suggests that the addition of moringa seed powder in the cream formula affects the panelist’s preference level. This is due to the increasing percentage of scrub increase the friction power contained in the product with the skin so it is easier to remove dead skin cells and impurities in the skin. The scrub grinder also serves as a massaging effect on the body. The percentage of good scrubs ranges from 34% of the cream scrub weights because the amount is ideal for stimulating exfoliation of skin epidermis. Percentage of scrub above 5% will damage skin tissue and make skin dry [22].

3.2 Results of Scrubs Cream Characteristics

The following is the result of a characteristic test of scrub cream produced in this research.

3.2.1 pH value

The result of pH analysis of scrub cream product each treatment has pH value which is still in the range of quality requirement according to SNI 16-4399-1996, that is with pH value range 6,88-7,45. Based on statistical test results with one-way anova showed a significant difference in the four samples. The pH value on the Moringa seed cream scrub product increased when compared with the scrub cream product without the addition of moringa seeds. This proves that moringa seeds capable of affecting the acidity of the cream product. Cream formula with the addition of 3.5; 4,5; 5.5% (b / b) of moringa seeds increased pH value compared with a cream formula without moringa seed powder due to moringa seed powder is base with pH value of 7.5-5.5.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>F0</td>
</tr>
<tr>
<td>pH</td>
<td>6,88</td>
</tr>
<tr>
<td>Specific gravity (g/mL)</td>
<td>1,00</td>
</tr>
<tr>
<td>Stability of emulsion (%)</td>
<td>95,1</td>
</tr>
<tr>
<td>Total microbial contamination (colony/ g)</td>
<td>Nol</td>
</tr>
</tbody>
</table>

3.2.2 Specific gravity

The test results scrub cream characteristics indicate that the value of specific gravity is still within the range of quality requirements SNI 16-4399-1996 with the average value of specific gravity of 1 g/ mL. This means that the resulting scrub cream product has a good level of stability. The result of statistical test with one-way Anova showed that scrub cream F0 with F1; F2; F3; and LK did not show any significant difference. This proves that with the addition of moringa seed powder does not affect the specific gravity of a cream product.

3.2.3 Stability of emulsion

The test results of cream scrub characteristics showed that the average emulsion stability value reached 95.19 - 96.01%. These results indicate that the sample hardly indicates a phase separation or a very small phase separation indicating that the cream product has a stable emulsion. The result of the statistical test with one-way Anova showed no significant difference, probability value 0.453 (P> 0.05). This indicates that moringa seed powder tends
not to affect the stability of the emulsion on a scrub cream products.

3.2.4 Total microbial contamination

The result of the characteristic test of total microbial contamination to cream scrub product showed that microorganisms contained in the four negative products. This is due to the addition of propyl paraben and methyl paraben formula scrub cream products. Propyl and methyl paraben is a preservative that can prevent the growth of bacteria and fungi. Moringa seed is also able to optimize the prevention of microbial contamination in cream product samples in addition. This is because Moringa seeds contain active compounds of seeds that are antimicrobial, including saponins, tannins, flavonoids, and alkaloids. Moringa seed active compounds work by inhibiting protein synthesis and destroying bacterial cell membranes so as to be antimicrobial [23].

3.3 Test Result Acute Dermal Scrub Cream

Based on the irritation test results (Table 3), no erythema and edema reaction in the skin of test animals of any formula with the value of the primary skin irritation index (PDII) was 0. If seen from the category of skin irritation index response then the PDII value goes into the range 0-0.4 so that it can be categorized as a material that does not irritate the skin. These results indicate that the scrub cream with the addition of a moringa seed scrub 3.5; 4.5; 5.5% and the base is safe to use. Test animal skin does not show an erythema reaction and edema is probably caused by the resulting moringa seed cream scrub having a pH still in the range of SNI 16-4399-1996 quality requirements so that its use does not cause irritation. According to Tranggono and Fatmah [24], pH is one of the things that can trigger the occurrence of side effects on the skin such as erythema and edema.

Table 4. Primary irritation test results of moringa seed scrub cream on Sprague-Dawley's white rat's skin

<table>
<thead>
<tr>
<th>Treat ment</th>
<th>24th hours</th>
<th>48th Hours</th>
<th>PDII</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Erythema</td>
<td>Edema</td>
<td>Erythema</td>
</tr>
<tr>
<td>F0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>F1</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>F2</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>F3</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>KK</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>KN</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

Note: F0 = rat smeared cream formula without addition of moringa seed scrub, F1 = rat smeared cream formula with 3.5% (b/w) moringa seed scrub, F2 = rat smeared cream formula with 4.5% (w/w) moringa seed scrub, F3 = rat smeared cream formula with 5.5% (b/w) moringa seed scrub, KK = rat smeared cream of commercial cream, KN = untreated rat.

Some components in cosmetics can potentially irritate the skin such as preservatives (antimicrobial substances), surfactants, fragrances, and dyes [25]. Scrub cream products in this study using commercial preservatives are methyl and propyl paraben. The JECFA (Joint FAO/WHO Expert Committee on Food Additives) [26] in 1990 stated that methyl and propyl paraben had an ADI (Acceptable Daily Intake) value of 0-10 mg/kg body weight, while methyl and propyl paraben toxicity data in mice orally is 0.5-2 g/250 grams of body weight. This study used methyl paraben as much as 0.2 gram and propyl paraben as much as 0.02 gram which is still in safe range according to JECFA guideline. The study also used cetyl alcohol, stearic acid, and triethanolamine.

4. CONCLUSION

Based on the research that has been done can be concluded: The most optimum scrub cream formulation is scrub cream with the addition of 4.5% moringa seed which has the highest general favorite level with a score of 5.67. Characteristic of scrub cream with the addition of moringa seed 3.5; 4.5; and 5.5% fulfill the standard that has been determined SNI 16-4399-1996, that is with pH value 6.88-7.31; the weight of type 1 g/mL; emulsion stability of 95.19-96.61%; and negative microbial contamination.

The irritation test results showed that the scrub cream with the addition of moringa seed 3.5; 4.5; 5.5% and the base do not cause irritation effect on the skin of white rat test animals (Rattus norvegicus) with the value of primary skin irritation index (PDII) equal to 0 that indicates scrub cream moringa seed safe to use.

5. REFERENCES


