Formulation and Physical Quality Test of Body Lotion of *Averrhoa bilimbi* L. Extract as Anti-Mosquito Agent for *Aedes aegypti*

Farica Eryani¹, Nanda Riztina Miryati¹, Ike Afni Faulatun¹, M. Arya Gifari¹, Fajri Ramadhan², Sri Seno Handayani^{1*}

¹Department of Chemistry, Faculty of Mathematics and Natural Science, University of Mataram, Mataram, Indonesia.

²Department of Pharmacy, Faculty of Medicine, University of Mataram, Mataram, Indonesia

*E-mail: srihandayani@unram.ac.id

Received: January 10, 2024. Accepted: May 25, 2024. Published: May 26, 2024

Abstract: Mosquitoes are one of the vectors with a high disease spread compared to several other types of insects. Making a body lotion from *Averrhoa bilimbi* L. leaf extract is an effort to reduce the spread of *Aedes aegypti* mosquitoes. *Averrhoa bilimbi* L. leaves contain secondary metabolite compounds that can be utilized as natural repellants. This study aimed to test the phytochemical compounds, physical quality and repellant power of the body lotion formula of the *Averrhoa bilimbi* L. leaf extract. The test results showed that the *lotion* preparation was stable in organoleptic observation and homogeneity test. Repellant activity against *Aedes aegypti* mosquitoes was conducted for 5 hours. The highest protection power of *lotion with Averrhoa bilimbi* L. leaf extract was obtained at 76.92% at 4% extract concentration from three formulas.

Keywords: Averrhoa Bilimbi L.; Repellent Body Lotion; Star Fruit Leaves.

Introduction

Dengue fever (DF) is a disease transmitted by the *Aedes aegypti* mosquito. This fever has become one of the most frightening and troubling diseases because of its rapid spread. According to Gianjar (2008), the factors that cause DF are climatic factors, including temperature, humidity and heavy rainfall. The population density can also affect the increasing number of DF cases. DF can affect infants, children, and adults and can be characterized by flu-like symptoms for 2 to 7 days [1].

Dengue cases are also increasing in West Nusa Tenggara Province. Mataram City is the area with the highest exposure to DF, with 536 cases and two deaths each year [2]. The high number of dengue cases must be handled immediately so that they do not increase, which can disturb the community. Some methods that can be done to control *Aedes aegypti* include sowing larvicides using insecticides and repellents [3]. Based on research by [4], the most effective type of mosquito repellent is *lotion*. Commercial repellent lotions generally contain the chemical *N-diethyl-m-toluamide* (DEET), which can increase immunity to mosquito bites but is very harmful to the skin [5]. Innovations are needed, such as creating lotions from natural ingredients safe for all skin types.

The research results [6] state that Averrhoa bilimbi L. leaf extract has anti-mosquito activity. This activity is due to the chemical content of tannins, flavonoids and saponins. According to [7], the compounds contained in plants that function as natural insecticides are cyanide groups, saponins, tannins, flavonoids, steroids and essential oils. Therefore, Averrhoa bilimbi L. leaves are suitable natural additives in anti-mosquito lotions because they contain many active compounds.

The active compounds in *Averrhoa bilimbi* L. leaf extract can act as repellants by entering the larval body through the respiratory system, which will cause nervous exhaustion and damage to the respiratory system, then cause the larvae to be unable to breathe and eventually die [8]. Tannins are contact toxins that can break the active cell due to proteolytic enzymes in mosquito body cells [9]. Saponins act as a stomach poison that can attack the digestive system in larvae that enter through the larval mouth and irritate the mucosa of the digestive tract, which can cause death in larvae [6].

Based on the compound content of *Averrhoa bilimbi* L. leaf extract that has been described, this study will formulate and test the physical quality of body lotion of *Averrhoa bilimbi* L. leaf extract as an anti-mosquito agent for *Aedes aegypti* that is safe for use by all skin types and all groups of children, adolescents and adults.

Research Methods

Materials

The tools used in this research are a 60 mesh sieve, stirring rod, blender, chamber, glass funnel, 500 mL beaker, measuring cylinder, mortar and pestle, magnetic stirrer, oven, water bath, drop pipette, rotary evaporator, pH meter and analytical balance.

The materials used were fresh leaves of *Averrhoa bilimbi* L., stearic acid, cetyl alcohol, glycerin, TEA, liquid paraffin, methylparaben, distilled water, and Whatman filter paper.

Research Procedure

Preparation

The sample is fresh *Averrhoa bilimbi* L. leaves that have been sorted. The sorting process was done by selecting and sorting the intact and undamaged *Averrhoa bilimbi* L. leaves. The leaves were then washed thoroughly to avoid damage and decreased quality of the active compound content.

Extraction

The preparation of *Averrhoa bilimbi* L. leaf extract was carried out using the maceration method, referring to the process of [10]. 1 kg of the leaves was dried using an oven (60°C) and then grinded. *Averrhoa bilimbi* L. leaves grinded as much as 500 g were added into a closed container and soaked using 70% ethanol for 72 hours. The extraction results were concentrated using a rotary evaporator at 60°C.

Phytochemical Screening

Phytochemical screening of *Averrhoa bilimbi* L. leaf extract was carried out to determine the presence of flavonoid, tannin, and saponin compounds.

Flavonoid Test

Averrhoa bilimbi L. leaf extract was added to 70% ethanol (1:3), then shaken, heated and shaken again. The filtrate obtained was then added to 1 g of Mg powder and two drops of concentrated HCl. The appearance of red in the ethanol layer indicates the positive presence of flavonoid compounds.

Tannin Test

Averrhoa bilimbi L leaf extract was added to distilled water (1:20) and then heated. Two to three drops of 1% FeCl₃ solution were added to the filtrate. The appearance of a greenish-brown or dark blue colour indicates the positive presence of tannin compounds.

Saponin Test

Averrhoa bilimbi L. leaf extract was added with distilled water (1:10). The filtrate obtained was shaken until homogeneous and then heated for 2-3 minutes. The filtrate is cooled, shaken for 10 minutes, and let stand again for 15 minutes. The presence of foam indicates the positive presence of saponin compounds.

Preparation of Body Lotion

The manufacture of body lotion in this study was based on the method of [14] by modifying the formulation of *Averrhoa bilimbi* L leaf extract with 2 mL, 4 mL, and 6 mL. Materials used in the oil phase (stearic acid, liquid paraffin and cetyl alcohol) and the water phase (glycerin, TEA, distilled water). Each material was heated at 70°C while stirring until homogeneous. The dissolved oil phase is added to the water phase while stirring until a thick and homogeneous mass is formed. Finally, Averrhoa bilimbi L. leaf extract, phenoxyethanol and perfume were added and stirred until homogeneous.

Table 1. Star Fruit Leaf Extract Lotion Formulation

Material name	Formulation		
	F1	F2	F3
Extract	2 mL	4 mL	6 mL
Stearic acid	2.5 g	2.5 g	2.5 g
Cetyl alcohol	5 g	5 g	5 g
Glycerin	0.5 mL	0.5 mL	0.5 mL
TEA	2 mL	2 mL	2 mL
Liquid paraffin	10 mL	10 mL	10 mL
Phenoxyethanol	0.2 mL	0.2 mL	0.2 mL
Perfume	0.8 mL	0.8 mL	0.8 mL
Distilled water	77 mL	75 mL	73 mL

Physical Quality Stability Test of Formula Organoleptic Test

This test is carried out by observing the shape, colour, texture and aroma of the body lotion preparation of *Averrhoa bilimbi* L. leaf extract [11].

Homogeneity Test

This test is done by placing 1 gram of body lotion of *Averrhoa bilimbi* L. leaf extract in the centre of the glass object and then covered with another glass object and observing the presence or absence of coarse grains. The sample is homogeneous if no coarse grains are in the formulation [16].

Acidity (pH) Test

This test uses a pH meter by dissolving the *Averrhoa bilimbi* L. leaf extract with 10 mL of water, stirring until dissolved and then measuring using a digital pH meter calibrated with a buffer solution. The resulting pH is in the range of 4.5-8 [11].

Spread Ability Test

This test was carried out by placing 0.5 grams of body lotion of *Averrhoa bilimbi* L. leaf extract in the middle of the spread ability glass and then giving a consecutive load of 0, 25, 75 and 125 grams in the centre of the glass with an observation period of every 1 minute. Then, the average diameter of the formula is spread from the average length of several sides [11].

Repellency Test

This test was carried out by applying body lotion of *Averrhoa bilimbi* L. leaf extract to the hand and then putting the hand into the mosquito cage. Testing was conducted for 5 hours, with an observation period of every 15 minutes [5].

Results and Discussion

Extraction of Averrhoa bilimbi L. Leaves

Extraction is separating active compounds from plant tissues using certain solvents. The selection of methods and solvents must be appropriate to get maximum results [13]. The maceration extraction method was chosen because the method and equipment are simple, without a heating process to prevent the decomposition of active substances contained in the sample due to the influence of temperature and compounds that cannot withstand heating [14].

The extraction process of *Averrhoa bilimbi* L. leaves uses the maceration method with 70% ethanol solvent as

much as 3.75 L for 72 hours. The maceration results obtained were then filtered with filter paper. The extract obtained as much as 1.85 L was then concentrated on a rotary evaporator to obtain a blackish-green pure extract of 145 mL with a yield of 3.867%.

Phytochemical Screening of The Extracts

Flavonoid Test

Flavonoid testing uses magnesium powder, and concentrated HCl added to the Averrhoa bilimbi L. leaf extract. The addition of magnesium powder and concentrated HCl can cause the reduction of flavonoid compounds in the extract by marking the colour change to brick red [15]. Flavonoid compounds are believed to damage bacterial cells by forming complex compounds with extracellular proteins, causing a decrease in oxygen function with the appearance of nerve and spiral disorders in bacteria.

Flavonoids work by denaturing proteins. This process also disrupts cell formation, thus changing the composition of protein components. Disrupted cell membrane function can lead to increased cell permeability, followed by bacterial cell damage. The damage causes bacterial cell death.

Tannin Test

Tannin testing was carried out using 1% FeCl₃ reagent added to the Averrhoa bilimbi L. leaves extract. Adding 1% FeCl₃ reagent aims to identify the presence of phenol groups in the extract of Averrhoa bilimbi L. leaves by marking it with a colour change to greenish brown or dark blue [15-16].

Tannin compounds are one of the macromolecular compounds of polyphenolic compounds that are polar with bitter taste characteristics. The bitter taste of this tannin compound can prevent insects from eating it [17-18]. This occurs due to tannin compounds that react with proteins to form insoluble copolymers in water, making proteins difficult for animal digestive fluids to reach.

Saponin Test

Saponin compounds have benefits that can damage the skin mucosa of insects if absorbed, causing hemolysis of blood cells, and, as a result, breathing becomes obstructed and causes death. Testing for saponin compounds shows a positive outcome if foam formation after shaking lasts a long time. Saponin compounds have glycosyl as a polar group and steroid or triterpenoid groups as nonpolar groups so that they form micelles when shaken with distilled water. The structure of the micelle polar groups will face outward, and the nonpolar groups will face inward, so this situation looks like foam.

Table 2. Phytochemical screening results

No.	Test	Description	Image	Results
1	Tannins	The		
		greenish-		
		brown colour		+
		appears		
2	Flavonoids	Red color		
		appears		+
3	Saponins	Froth		
		appears		+

The phytochemical test results showed quite significant differences compared to the results of research [17], which stated that *Averrhoa bilimbi* L. leaf extract only contained steroids.

Physical Quality Stability of Formula

Organoleptic and Homogeneity Test

The results of organoleptic test observations for four weeks in F1 (first formula) have a brownish yellow colour, F2 is light brown, and F3 is brown, while for other characteristics, it has the same characteristics in each formula, like smooth texture, the distinctive odour of extract and semi-solid shape. Based on Table 2, the body lotion has characteristics that do not change from week 0 to week 4. Body lotion has a different colour in each formula due to adding extracts. The concentration of extracts added to the body lotion affects the colour intensity of the body lotion.



Figure 1. Averrhoa bilimbi L. leaf body lotion

Based on the homogeneity test for four weeks, it shows that the three formulas are homogeneous by not forming coarse particles on the object glass, and the formula is stable at room temperature from week 0 to week four observations. Homogeneity testing is done so that the active ingredients in a lotion preparation can be evenly distributed and do not irritate the skin when used.

Spread Ability Test

The lotion spread ability test was conducted at the beginning of manufacture and lasted up to 4 weeks. Observations were made weekly from week 0 to week 4. The spread ability test aims to determine the spread ability of a lotion preparation. Each formulation has a great spread ability of around 5-9 cm. The concentration of each formula influences the spread ability of body lotion. The higher the concentration of Averrhoa bilimbi L. leaf extract, the more dilute the body lotion. The lotion diameter set is in the range of 5cm-7cm. If the spread ability is too small or below the standard, it will cause the lotion preparation to be complex and absorbed by the skin, irritating skin. The lotion cannot be adsorbed normally if the spread ability is too high or above the standard [11].

Acidity (pH) test

A good lotion does not irritate the skin, with a pH standard that meets the quality of moisturizing preparations for the skin is 4.5-8. If the pH is too low or below, the skin can be scaly, itchy, and irritated. If the pH is too high, it can make the skin dry, affecting the skin's elasticity (Figure 2).

Repellency Test

In the repellant power test, all lotion formulas of Averrhoa bilimbi L. leaf extract (F1, F2, F3) showed

mosquito-repellent activity. Formula 2 (F2) obtained the highest repellant power of 76.92% (Table 3).

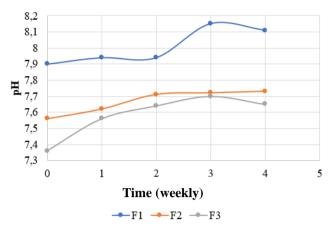


Figure 2. Acidity graph

Table 3. Repellency results

Formula	Repellency (%)
F1	30.77
F2	76.92
F3	50

Conclusion

Based on the research that has been done, it is concluded that the best formulation of Averrhoa bilimbi L. leaf extract lotion based on the results of physical quality tests is F2 (4% extract concentration) with physical properties of light brown, semi-solid texture, distinctive aroma of Averrhoa bilimbi L. leaf extract, spread ability range 5.5-9.46 and more stable pH range 7.56-7.73 and with a protective power of 76.92%.

References

- [1] Binsasi, E., Bano, E. N., dan Salsinha, C. N., (2021). Analisis Model Penyebaran Penyakit Demam Berdarah Dengue di Kota Kefamenanu, *Jurnal Statistika dan Matematika*, 3(1): 1-11.
- [2] Rahmawati, C., Nopitasari, B. L., Wardani, A. K., Nurbaety, B., Anjani, B. L. P., Hati, M. P., Furqani, N., Wahid, A. R., Safwan, Hendriyani, I., Pradiningsih, A., Fitriana, Y., dan Ittiqo, D. H., (2022). Edukasi Mencegah Penyakit Demam Berdarah Dengue (DBD) pada Masyarakat Lingkungan Dasan Sari Ampenan, Jurnal Pengabdian Masyarakat Berkemajuan, 6(4): 3038-3042.
- [3] Sukokhar, A., (2014). Demam Berdarah Dengue (DBD), *Medical Jorunal of Lampung University*, 2(2): 1-15.
- [4] Wahyono, T. Y. M. dan Oktarinda, M. W., (2016). Penggunaan Obat Nyamuk dan Pencegahan Demam Berdarah di DKI Jakarta dan Depok, *Jurnal Epidemiologi Kesehatan Indonesia*, 1(1): 35-40.
- [5] Handayani, S. S., Gunawan, E. R., Suhendra, D., Murniati, dan Bali, R. D. K., (2021). Daya Repelan Lotion Minyak Ketapang pada Berbagai Variasi Konsentrasi Atsiri Kulit Jeruk Purut, *Jurnal Pijar MIPA*, 16(2): 262-266.

- [6] Feftyanto, H. R., Syuhriatin, Sulastri, M. P., dan Rahayu, S. M., (2019). Efektivitas Filtrat Daun Belimbing Wuluh (Averrhoa bilimbi) Sebagai Larvasida Nyamuk Aedes sp, Lombok Journal of Science, 1(1): 20-23.
- [7] Kardinan, A., (2002), *Pestisida Nabati: Ramuan dan Aplikasi*, Jakarta: Penebar Swadaya.
- [8] Cania, E., & Setyanimgrum, E. (2013). Uji efektivitas larvasida ekstrak daun legundi (Vitex trifolia) terhadap larva Aedes aegypti. *Medical Journal of Lampung University*, 2(4).
- [9] Ramayanti, I., Layal, K., Pratiwi, P. U., UMP, K., Banten, J. A. Y. T., & Plaju, U. (2017). Efektivitas Ekstrak Daun Kemangi (Ocimum basilicum) sebagai Bioinsektisida Sediaan Antinyamuk Bakar Terhadap Kematian Nyamuk Aides aegypti. *Journal of Agromedicine and Medical Sciences*, 3(2), 6-10.
- [10] Hasim, H., Arifin, Y. Y., Andrianto, D., & Faridah, D. N. (2019). Ekstrak etanol daun belimbing wuluh (Averrhoa bilimbi) sebagai antioksidan dan antiinflamasi. *Jurnal Aplikasi Teknologi Pangan*, 8(3), 86-93.
- [11] Jubaidah, L. (2019). Formulasi dan Uji Mutu Fisik Sediaan Lotion Ekstrak Kulit Buah Jagung (*Zea mays* L.): Formulasi Dan Uji Mutu Fisik Sediaan Lotion Ekstrak Kulit Buah Jagung (Zea mays L.). *Jurnal Insan Farmasi Indonesia*, 2(2), 175-184.
- [12] Arisanty, A., & Anita, A. (2018). Uji Mutu Fisik Sediaan Krim Ekstrak Etanol Buah Belimbing Wuluh (Averrhoa bilimbi L.) Dengan Variasi Konsentrasi Na. Lauril Sulfat. *Media Farmasi*, 14(1), 22-27.
- [13] Kurniawati, A. (2017). Pengaruh jenis pelarut pada proses ekstraksi bunga mawar dengan metode maserasi sebagai aroma parfum. *Journal of Creativity Student*, 2(2), 74-83.
- [14] Wendersteyt, N. V., Wewengkang, D. S., Abdullah, S. S., & Stout, D. (2021). Antimicrobial Activity Test Of Exstracts And Fractions Of Ascidian Herdmania Momus From Bangka Island Waters Likupang Against The Growth of Staphylococcus Aureus, Salmonella Typhimurium, And Candida Albicans Uji Aktivitas Antimikroba Dari Ekstrak Dan FR. Journal Of Pharmacy, 10-17.
- [15] Simaremare, E. S. (2014). Skrining fitokimia ekstrak etanol daun gatal (Laportea decumana (Roxb.) Wedd). *PHARMACY: Jurnal Farmasi Indonesia* (*Pharmaceutical Journal of Indonesia*), 11(1).
- [16] Roanisca, O., Rani, R., & Mahardika, R. G. (2021). Phytochemical Screening and Antibacterial Potency of Jeruk Kunci Fruit Waste (Citrus x microcarpa Bunge) Extract Against Propionibacterium acnes. *Jurnal Pijar Mipa*, 16(3), 387-392.
- [17] Damayanti, I. A. M., Antari, N. W. S., & Megayanti, S. D. (2022). Antioxidant potential of gotu kola leaf extract (Centella asiatica (L.) Urban) as an alternative to antihyperglycemic herbal drinks. *Jurnal Pijar Mipa*, 17(6), 782-786.
- [18] Hartini, E., Yulianto, Y., Sudartini, T., & Pitriani, E. (2022). Efikasi Ekstrak Daun Kipahit (Tithonia diversifolia) terhadap Mortalitas Ulat Bawang (Spodoptera exigua Hubn.). *Media Pertanian*, 7(1), 23-33.

- [19] Febriyanti, R. T., & Rudi, L. (2022). Uji Fitokimia Ekstrak n-Heksana Daun Belimbing Wuluh (Averrhoa bilimbi L.). *Sains: Jurnal Kimia dan Pendidikan Kimia*, 11(2), 133-143.
- [20] Widia, W., Ependi, E., Amelia, A., Candra, C., & Roanisca, O. (2023). Cytotoxicity activity of ethanol extract of mentangor stem skin (Calophyllum rufigemmatum) against breast cancer cells MCF-7 in vitro: Chemistry. *Jurnal Pijar Mipa*, *18*(1), 72-76.