١MA 100808

Jurnal Farmasi Sains dan Praktis

(JFSP)

http://journal.unimma.ac.id/index.php/pharmacy



PHYSICAL STABILITY TEST HAIR TONIC COMBINATION Cinnamon (Cinnamomum burmanii) ESSENTIAL OIL AND VCO (VIRGIN COCONUT OIL) WITH CYCLING TEST METHOD AND **IRRITATION TEST ON THE RABBIT**

Junvidya Heroweti^{1,3}, Danang Novianto Wibowo^{2,3} Arini Najikhatal Ulya³, Desmita Linda³

¹Department of Pharmacology and Clinical Pharmacy, Universitas Wahid Hasyim, Semarang 50236, Indonesia

²Department of Pharmaceuticals and Pharmaceutical Technology, Universitas Wahid Hasyim, Semarang 50236, Indonesia

³Pharmacy Study Program, Faculty of Pharmacy, Universitas Wahid Hasyim, Semarang 50236, Indonesia

junvidyaheroweti@gmail.com

https://doi.org/10.31603/pharmacy.v9i1.7903

Article info:	ABSTRACT
Submitted : 17-09-2022	Cinnamon (Cinnamomum burmanii) and VCO (Virgin Coconut Oil) essential
Revised : 03-04-2023	oils contain cinnamaldehyde and lauric acid which have potential for hair growth, they can affect the physical stability of hair tonic preparations. This study aims
Accepted : 01-05-2023	to determine the physical stability and determine the effect of the irritation effect on hair tonic preparations of a combination of cinnamon essential oil and VCO.
	Cinnamon essential oil hair tonic preparations were made with a concentration of
	1% (F1), 3% (F2), and 5% (F3) and then tested for physical characteristics
BY NC	including organoleptic tests, homogeneity, pH, density test, and viscosity.
This work is licensed under	Evaluation of physical stability was carried out using the cycling test method.
a Creative Commons	irritation test were analyzed descriptively using the primary irritation index
Attribution-NonCommercial	obtained from the appearance of erythema and edema on the rabbit's skin.
4.0 International License	Observation of the physical stability test on organoleptic and homogeneity
Deck 1's harm	showed that formula I had a cloudy white color, a characteristic cinnamon aroma,
Publisher:	a slightly thick liquid, and was homogeneous; formula 2 and 3 has a clear color,
	characteristic of cinnamon aroma, slightly viscous and homogeneous liquid;
Universitas Muhammadiyah	stable pH in formula 2; stable density in formulas 1, 2 and 3; and stable viscosity
Magelang	primary irritation index for F1, F2, and F3 was 0.0; 0.3; 0.3 and it can be
	concluded that F2 and F3 can irritate with very mild categories.
	Keywords: Cycle test; Erythema & edema; Irritation index; Physical characteristics

1. INTRODUCTION

Hair has an important role in supporting appearance. Hair loss is a condition where more than normal. Excessive hair loss can lead to baldness. Hair loss can be prevented by external or internal treatment. Internal treatment is by taking nutrition or drugs, give an injection, and use hair fertilizer. External treatment can use a solution for hair care, one of which is hair tonic (Aini, 2017). Hair tonic is a cosmetic preparation used to treat hair growth (BPOM, 2013).

One of the herbal ingredients used to maintain hair care are cinnamon essential oil (Cinnamomum burmanii) and VCO (Virgin Coconut Oil). Cinnamon essential oil contains a compound cinnamaldehyde 67.94%. Cinnamaldehyde in cinnamon essential oil can function to prevent hair loss, increase hair length and hair follicle diameter. Coconut oil has a triglyceride component of lauric acid which is a good nutrient and is able to penetrate the hair cavity easily and give the effect of hair growth (Muhammud et al., 2014). Pure coconut oil or VCO can also be

a stable hair fertilizer (Usman & Yuliana, 2020). Cinnamaldehyde in cinnamon essential oil can increase blood flow caused by vasodilation of blood vessels so that nutrients and oxygen can be filled from the hair roots, and hair loss can be prevented with accelerates hair growth (Indriana et al., 2018). While VCO is composed of organic compounds, a mixture of esters and gelisols, acids fat. According to the research, unsaturated fatty acids are elements essential for skin care, including the scalp because the mechanism as the emollient. Coconut oil can reduce protein at loss hair or at damaged and undamaged hair. VCO contains are lauric acid, vitamin K, vitamins E, and iron which functions as good nutrition and vitamins for nourish hair (Setiaji &Prayugo, 2006). Virgin coconut oil can stably maintain healthy hair due to the content of lauric acid and capric acid. Physical stability test on hair tonic needs to be done, because the presence of cinnamon essential oil and VCO can affect the physical stability of the preparation.

Hair tonic preparations with active ingredients of essential oils are physically stable. Previous research has shown that the results of the characteristic test with eucalyptus oil ingredient concentrations 2.5%, 5% and 7.5% showed a hair tonic formulation was clear, slightly cloudy, white and cloudy white, and have weak, medium and strong aroma of eucalyptus oil. The preparation of the physical stability using the cycling test method for 6 cycles and showed that the hair tonic was stable both in terms of color, smell and homogeneity (Sugesti et al., 2018).

Hair tonic is used topically so it is necessary to do an acute skin irritation test to determine the effect of irritation that occurs on the skin. Irritation test was carried out using animals by assessing the safety of the preparation when exposed to the skin with erythema and edema score (BPOM, 2014). Erythema is a reddish reaction that occurs due to an increase in the amount of blood in the dermis which causes vasodilation of blood vessels due to inflammation (Harlim, 2017). While edema is swelling that occurs due to the accumulation of fluid in the intercellular tissue resulting from abnormal expansion of the interstitial fluid volume (Trayes et al., 2013).

Based on this background, this study aims to determine the physical stability of the hair tonic formulation of a combination of cinnamon essential oil (*Cinnamomum burmanii*) and VCO (Virgin Coconut Oil) for six storage cycles and test for acute dermal irritation on animal test before product is used by consumers.

2. METHODS

2.1. Research Material

The sample materials used in this study were cinnamon essential oil (*Cinnamomum burmanii*) produced by Nusaroma Essential Oil and VCO (Virgin Coconut Oil) from CV Coco Indo Jaya. The hair tonic formulation consisted of 96% alcohol, methyl paraben, menthol, dexpanthenol, polyoxyethylene 40 hydrogenated castor oil, propylene glycol, tween 80 and aquadest. Animal experiment use three rabbits (Oryctolagus cuniculus) with a body weight of 2-3 kg from Kendal rabbit farm.

2.2. Research Tools

The tools used in this research are a set of glassware (Iwaki), pH meter (handylab ph11/set), analytical balance (Ohaous), Mixer (Maspion), pycnometer, viscometer (Otswald), climatic chamber (HWS-70BX), refrigerators, and packaging bottles. The tools used in the skin irritation test are manual razors, hair clippers, rulers and urgical pens (Tondaus), sterile gauze (one med), duct tape (handsaplast) and non-irritant plasters (hypafix) to cover the treated skin.

2.3. Formula for Hair Tonic Cinnamon Essential Oil and VCO

Hair tonic preparations are made with varying concentrations of cinnamon essential oil (*Cinnamomum burmanii*) combined with VCO (Virgin Coconut Oil). Cinnamon oil hair tonic preparations were made based on the reference formulation from Sugesti (Sugesti et al., 2018). Based on the reference formula, the modified cinnamon essential oil (*Cinnamomum burmanii*)

hair tonic formulation was combined with VCO. Cinnamon essential oil was made with various concentrations including 1%, 3%, and 5%, the formulations can be seen in **Table 1**.

Matarial	Material Requirements			
Material	F1 (1%)	F2 (3%)	F3 (5%)	
Cinnamon essential oil	1 g	3 g	5 g	
VCO	2.5 g	2.5 g	2.5 g	
Alcohol 96%	24 g	24 g	24 g	
Methyl paraben	0.3 g	0.3 g	0.3 g	
Menthol	0.2 g	0.2 g	0.2 g	
Dexpanthenol	1 g	1 g	1 g	
Tween 80	20 g	20 g	20 g	
Polyoxyethylene 40 hidrogenated castor oil	1 g	1 g	1 g	
Propilen glikol	5 g	5 g	5 g	
Aquadest ad	100 ml	100 ml	100 ml	

Table 1. Cinnamon Essential Oil and VCO Hair Tonic Modified Formula

2.4. Preparation Hair Tonic Combination of Cinnamon Essential Oil and VCO

Preparation of hair tonic is done by heating aquadest at a 75-80 °C temperature. Then tween 80 and Polyoxyethylene 40 hidrogenated castor oil (PEG 40) were dissolved using hot aquadest while stirring with a mixer until homogeneous. Propylene glycol was dissolved with aquadest then put into a mixture of tween 80 and PEG 40. The next step is adding methyl paraben, menthol which has been dissolved in a small amount of alcohol and dexpanthenol added slowly, then added alcohol gradually. Cinnamon essential oil that has been mixed with VCO is added slowly and stirred until homogeneous. Mixing is carried out using a level 1 mixer speed.

2.5. Physical Evaluation of Hair Tonic Preparations

2.5.1. Organoleptic test

The organoleptic test of the hair tonic preparation was observed with the five senses. The organoleptic test of the hair tonic preparation includes the shape, smell, and color of the hair tonic preparation (Ambari et al., 2020).

2.5.2. Homogeneity test

The homogeneity test of the hair tonic formulation was carried out by applying the hair tonic on a glass plate, then leveled and observed whether there were particles that were not homogeneously distributed (Indriyani, F., & Susi, 2021).

2.5.3. pH test

The pH test was measured with a pH meter that had been calibrated by dipping the electrode in two solutions until the pH meter showed a constant number. Commonly used solutions are pH 4 and pH 7 (Apriani et al., 2021).

2.5.4. Viscosity and Rheological Test

The viscosity test of the hair tonic preparation was carried out using an Otswald viscometer. 10 ml of hair tonic formulation is put into tube A and then withdrawn until it passes the "b" limit and the "a" limit. The hair tonic preparation is allowed to flow from the "a" limit to the "b" limit. The time for the hair tonic preparation to flow was calculated using a stopwatch. Viscosity calculation can be calculated by the equation 1:

$$\frac{\eta_1}{\eta_2} = \frac{\rho_1 t_1}{\rho_2 t_2}$$
(1)
Where: η_1 = unknown fluid viscosity; η_2 = standard fluid viscosity; ρ_1 and ρ_2 = density liquid;

 t_1 and t_2 = flow time (in seconds)

Rheology test was carried out on the three formulas. Rheological measurements were carried out using a cone and plate viscometer. Samples were prepared and placed under the viscometer display panel. The cone was lowered until it was immersed in the sample, then the cone was rotated with an initial speed of 10 rpm to a constant speed of 60 rpm. The viscometer will show the value on display.

2.5.5. Density Test

Density test is carried out using clean and dry pycnometer and has been calibrated by determining the weight of the pycnometer and the weight of water boiled at 25 °C. The temperature of the hair tonic preparation was measured to 20 °C, then put into a pycnometer and the temperature was measured to 25 °C.

2.5.6. Physical Stability Test with Cycling Test Method

The three-hair tonic formulation F1, F2 and F3 were tested for physical stability using the cycling test method. The cycling test is carried out by storing the hair formulation in the refrigerator at 4 °C for 24 hours. The next step is removing the preparation and placing it in a climatic chamber at 40 °C for 24 hours. This process is calculated in 1 cycle. In each cycle, the hair tonic formulation was tested for physical properties including organoleptic test, homogeneity, pH, viscosity tests and density test. Cycling test was carried out for 6 cycle.

2.5.7. Procedure of Irritation Test

Irritation test was conducted with a rabbit. The treatment procedure is by dividing the rabbit's back with an area of approximately 10x15 cm or not less than 10% of the body surface. The first step is by shaving the rabbit's hair. Then were allowed to stand for 24 hours before being used. The area was divided into five parts, namely area 1 as an area without treatment, area 2 for control without cinnamon and areas 3,4, and 5 respectively to be given a hair tonic formulation with a concentration of 1%, 3% and 5% with an area of 2x3 cm. The irritation test was carried out using a patch test. The dose of hair tonic formulation given is 0.5 mL. Exposure to the test formulation is carried out on the desired skin area, then sterile gauze and a non-irritating plaster are applied to the exposure site (BPOM, 2014).

Hair tonic formulation was administered for four hours at the rabbit skin, then the residue was removed and cleaned. The emergence of toxic symptoms that need to be observed is the presence of primary irritation in the form of erythema and edema during the 1st, 24th, 48th and 72nd hours at the same time and hours (BPOM, 2014). Assessment of erythema and edema scores can be carried out after observation at Table 2.

Tuble 2. Evaluation of Skill Reaction (DI ONI, 2014)					
Erythema and crust formation	Score				
No erythema	0				
Very slight erythema (barely indistinguishable)	1				
Erythema is clearly visible	2				
Moderate to severe erythema	3				
Severe erythema (flesh-red) to eschar formation which hinders the assessment of erythema	4				
Formation of edema	Score				
No edema	0				
Very small edema (almost indistinguishable)	1				
Minor edema (demarcated area clearly visible)	2				
Moderate edema (increase in area by about 1 mm)	3				
Severe edema (increased area of more than 1 mm and extends beyond the area of exposure of the test preparation)	4				

Table 2. Evaluation of Skin Reaction	(BPOM,	2014)
--------------------------------------	--------	-------

The results of the skin reaction evaluation were carried out by calculating the primary irritation index and to determine the irritation response category based on Table 3. Obtained data

was evaluated to conclude primary irritation index in equation 2 and determined by category of irritation and presented at Table 3.

$$Primary\ irritation\ index = \frac{A-B}{C}$$
(2)

Where:

- A: The total score of edema and erythema of all sample observation points at 24,48 and 72 hours divided by the number of observations
- B: Total score of edema and erythema of all control observation points at 24,48 and 72 hours divided by the number of observations
- C: Number of Animals

Table 3. Category Irritation Response In Rabbits (BPOM, 2014)	
Evaluation	Score
Very Light (negligible)	0.0-0.4
Mild Irritant (slight)	0.5-1.9
Moderate Irritant (moderate)	2.0-4.9
Strong Irritant (severe)	5.0-8.0

2.5.8. Data analysis

Organoleptic and homogeneity data were analyzed descriptively. One-way anova were used to determine the statistical significance (p<0.05) of the differences between the values of various group of the preparation pH, viscosity, density test. The stability of the hair tonic formulation combination of cinnamon essential oil and VCO stated that there was no significant difference during 6 cycles of storage (12 days) if the significance value was (p>0.05). Irritation test was analyzed descriptively using the primary irritation index score obtained from the appearance of erythema and edema on the rabbit skin.

3. RESULTS AND DISCUSSION

3.1. Physical Stability

The preparation of hair tonic combination of cinnamon (*Cinnamonum burmanii*) essential oil with VCO (Virgin Coconut Oil) was tested for physical stability. The stability test in this study used the cycling test method. This stability test is a study designed by applying pressure to product storage at high temperatures used to cause product damage. This is done so that a product is degraded (Bajaj et al., 2012). The physical stability test in this study was stored at low temperature (4 °C) and high temperature (40 °C). Parameters for stability test include organoleptic test, homogeneity, pH, viscosity and density test. This study used three formulas with varying concentrations of cinnamon essential oil. The result of the organoleptic and homogeneity showed at Table 4.

		-		
Cycle/ Formula		Organolepti	s	Homogonoity
0-6	Color	Aroma	Texture	nomogeneity
F1 (1%)	Cloudy white	Cinnamon	Slightly thick liquid	Homogeneous
F2 (3%)	Clear	Cinnamon	Slightly thick liquid	Homogeneous
F3 (5%)	Clear	Cinnamon	Slightly thick liquid	Homogeneous

Table 4. Physical stability test results FI, F2, F3 from cycle 0-6 of Hair Tonic Cinnamon Essential Oil

Organoleptic testing aims to determine consumer acceptance for the product. Organoleptic testing by observing the color, aroma, texture of a formula (Ambari et al., 2020). Physical stability test result of hair tonic formula 1, 2 and 3 which were stored for 6 cycle result no color, aroma and texture change occurs. While the formulation results of each preparation show that hair tonic formula 1 has a cloudy white color, with a distinct cinnamon aroma and have the slightly viscous

liquid texture. Formulation 2 and 3 have clear color, cinnamon aroma and slightly viscous (**Figure 1**).



Figure 1. Organoleptic test in the sixth cycle at: (a) formula 1; (b) formula 2; and (c) formula 3

Homogeneity testing carried out in the presence of granules or particles on preparation of hair tonic formula 1, 2 and 3 on glass slides. The homogeneity test results of three formulas in each cycle show results homogeneous (Figure 2).



Figure 2. Homogeneity test in the sixth cycle at: (a) formula 1; (b) formula 2; and (c) formula 3

Data from the results (**Table 5**) of the pH test formula I were analyzed using the Kruskal Wallis test statistic and showed that there was a difference (p<0.05) so it should be continued with the Mann-Whitney test and showed a significant difference in pH (p<0.05) during storage at cycles 1, 3, 4 and 5. This indicates that the pH in formula 1 is not stable during storage. Data from the results of the pH test formula II were analyzed using One Way Anova test statistics and showed that there was no significant difference in pH during storage (p>0.05). This indicates that the pH in formula 2 is stable during storage. While data from the results of the pH test formula III showed a significant difference in pH (p<0.05) during storage at cycles 0, 2, 4, 5 and 6. This indicated that the pH in formula 3 was not stable during storage. Changes in pH indicate a reaction or damage to the constituent components in the hair tonic preparation, causing an increase and decrease in the pH value of the hair tonic preparation (Puluh et al., 2019).

Cable 5. Average pH test at formul	a 1, 2,	, 3 of hair	tonic	formulation
---	---------	-------------	-------	-------------

Formula Average pH ±SD							
Formula	0	1	2	3	4	5	6
F1	5.973	6.363	6.123	6.340	6.483	6.420	6.150
	±0.186	±0.041	±0.040	±0.050	±0.020	±0.020	± 0.060
F2	5.807	6.863	5.470	5.756	5.860	5.863	5.703
	±0.371	±0.189	±0.153	±0.193	±0.190	±0.196	± 0.188
F3	5.683 ± 0.076	5.580 ± 0.105	5.350 ± 0.125	5.550 ± 0.049	5.623 ± 0.115	5.733 ± 0.056	5.353 ± 0.250

Changes in pH are also influenced by the active substance content of volatile cinnamon essential oil, namely Cinnamaldehyde and trans cinnamil acetate (Verdini et al.,2022). Changes in the pH value of the preparation during several storages indicate that the preparation is less stable during storage (Putra et al., 2014).

Formula	Average Viscosity ±SD						
rormula	0	1	2	3	4	5	6
F1	2.404	2.520	2.593	2.497	2.617	2.592	2.599
	±0.173	±0.047	±0.019	±0.028	±0.012	±0.012	± 0.084
F2	1.869	2.520	2.544	2.568	2.601	2.536	2.553
	±0.394	±0.021	± 0.086	±0.055	±0.039	±0.009	±0.034
F3	2.432	2.498	2.519	2.474	2.594	2.541	2.598
	±0.061	±0.126	±0.093	±0.184	±0.017	±0.050	±0.070

Table 6. Viscosity test results at formula 1, 2, 3 of hair tonic formulation

The results of the viscosity test (**Table 6**) of formula 1 were analyzed using the Mann-Whitney test and showed a significant difference in viscosity (p<0.05) during storage in cycle 4 This indicates that the viscosity of formula is not stable during storage. While The results of the viscosity test of formula 2 and 3 result that the viscosity of formula is stable during storage. The increased viscosity after the cycling test was carried out due to additional ingredients from hair tonic preparations that could increase the moisture in the preparation due to storage at low temperatures (4 °C) so as to increase the viscosity value of the preparation. Meanwhile, the decreased viscosity after the cycling test was carried out because the high temperature (40 °C) could increase the distance between the particles so that the forces between the particles would be reduced.

Table 7. Density test results at formula 1, 2, 3 of hair tonic formulation

Formula		Average Density ±SD						
r ormula	0	1	2	3	4	5	6	
F1	1.016	1.000	1.008	1.010	1.006	1.008	1.012	
	± 0.022	± 0.002	± 0.002	± 0.006	± 0.007	±0.003	±0.025	
F2	1.013	1.001	0.986	1.010	1.002	1.002	0.997	
	± 0.005	±0.003	±0.033	± 0.006	±0.003	± 0.006	±0.009	
F3	1.007	0.994	0.978	1.008	1.004	0.992	1.009	
	± 0.007	±0.017	±0.027	± 0.005	±0.003	±0.014	±0.023	

Data from the results of the density test (Table 7) of formula 1,2 and 3 were analyzed using the Kruskal Wallis test statistic and showed that there was no significant difference in density (p>0.05) during storage. This indicates that the density of all three formulas is stable during storage. The results of this study found that formula 2 had the most stable result with organoleptic, homogeneity, pH, viscosity, density that stable during storage.

3.2. Viscosity and Rheological Test

Viscosity test was conducted to determine the hair tonic resistance of the combination of cinnamon essential oil with VCO when flowing. The higher the value of the viscosity of the solution, the more viscous the resulting solution. Thick hair tonic will cause crust on the scalp so it can cause dandruff. A good hair tonic has a low viscosity so that it produces a liquid preparation.



Figure 3. Rheological test results formula 2

The hair tonic viscosity test at formula 2 is carried out using an ostwald viscometer because the liquid hair tonic dosage form is slightly thick. Hair tonic has a newtonian flow type (Figure 3), because hair tonic has a constant characteristic as the shear rate increases (Allen et al, 2011).

Liquid preparation generally has a newton flow type. The influence of the flow properties will affect the pouring power of the preparation. The viscosity of a Newtonian fluid is directly proportional to the force and the shear rate. The higher the viscosity of a dosage form, the larger the force per unit area required to produce a shear rate. From the **Figure 3**, results of the flow properties of viscosity measurements showed that hair tonic preparations on each formula has a Newtonian flow because it has characteristics constant viscosity with increasing shear rate.

3.3. Irritation Test

The acute dermal irritation test has the principle that the hair tonic preparation in a single dose is exposed to the skin of the test animal with the control in the form of a skin area that is not given the preparation. The benefits testing for acute dermal irritation test animals are able to provide information on the prediction of the dangers that will arise when the preparation is exposed to human skin (Lestari et al., 2020). The three males New Zealand white rabbit (Oryctolagus cuniculus) was chosen because it did not a reproductive process that during the irritation test process there were no significant changes in the rabbits. Parameters in the selection of test animals on this study are albino rabbit with healthy skin (BPOM, 2014). The rabbits were acclimatized for 5 days with the aim that the rabbits were able to adapt environment. Shearing rabbit hair need carefully to avoid abrasions caused by the shaver because the injured skin will absorb more of preparation than intact skin. Scratches on the rabbit skin cause the stratum cornemum layer to be damaged and cause the preparation to penetrate the underlying layer faster and irritation (Rahman et al., 2013). Before applying the preparation to the rabbit's skin, it is necessary to check the pH of the preparation, because it will affect the occurrence of skin irritation. The result of a pH that is too acidic will affect the skin condition become scaly, itchy and irritated. Meanwhile, if the pH is too alkaline, it will make slippery, dry skin and will affect skin elasticity (Safitri & Luluk, 2019). The pH requirements for hair tonic preparation that are suitable are pH 3-7 (Indriyani & Susi, 2021). The result of primary irritation index showed at Table 8.

Formulation	Primary Index Irritation	Evaluation	Category
F0	0.0	Not irritating	Very light
F1	0.0	Not irritating	Very light
F2	0.3	Very slightly irritating	Very light
F3	0.3	Very slightly irritating	Very light

Table 8. Primary Irritation Index hair tonic combination of cinnamon essential oil with VCO

Where:

F0 : treatment without cinnamon essential oil

F1: Formula hair tonic oil combination Cinnamon with VCO 1%

F2: Formula hair tonic oil combination Cinnamon with VCO 3%

F3: Formula hair tonic oil combination Cinnamon with VCO 5%

The content of cinnamaldehyde in cinnamon essential oil can cause skin irritation by increasing the cold pain threshold, increasing temperature and perfusion of the skin disruption of the structure and function of the cell membrane caused by swelling of the cell membrane and changes in the permeability of the bacterial cell membrane (Andersen et al., 2016). In this study, the F0 treatment was also used as a control without essential oil. In addition to cinnamaldehyde, the content of terpenoid compounds in essential oils can also cause irritation to the skin because these compounds can accumulate in the lipid network resulting in disruption of the structure and function of cell membranes (Sikkema, J., et al., 1994). Natural terpene has high activity, little

skin reaction, and low toxicity and can safely and effectively promote transdermal absorbtion of drugs (Yang, W, et al., 2020). However, its topical application could cause skin irritation (Stanojević et al., 2019). The irritation test showed at Figure 4.



Figure 4. Irritation observation results: (A) erythema condition; (B) edema condition

The results of the hair tonic irritation test of the combination of cinnamon essential oil with VCO in this study were in accordance with (Lestari et al., 2020) research which showed that the anti-dandruff shampoo preparation of cinnamon essential oil at a concentration of 3% was slightly irritating with a score of 0.78. Based on the results of this evaluation, it can be concluded that the hair tonic combination of cinnamon essential oil with VCO causes erythema and edema in the very mild category (negligible) on rabbit skin, so it is still considered safe if applied to humans because rabbit skin sensitivity is higher than human skin (Kurniawidjaja et al., 2021). The high sensitivity of rabbit skin causes the irritation index in humans cannot be ascertained if the irritation value in test animals has a mild index (Zulfa et al., 2018).

4. CONCLUSION

Based on the physical stability test of the hair tonic preparation of the combination of cinnamon essential oil with VCO, it can be concluded that formula I (1%) has a cloudy white color, a characteristic aroma of cinnamon, homogeneous, and physically unstable because of the pH and viscosity, the results show that there are significant differences. during storage. Formula II (3%) has a clear color, characteristic cinnamon aroma, homogeneous, and physically stable during storage, while formula III (5%) has a clear color, characteristic cinnamon aroma, homogeneous, physically unstable due to pH, and showed that there was a significant difference during storage. Hair tonic has a newtonian flow type.

Hair tonic combination of cinnamon essential oil (Cinnamomum burmanii) with VCO (virgin coconut oil) as hair growth causes irritation in the form of erythema and edema on rabbit skin with very mild category (negligible). The resulting primary irritation index value is 0.0 in formula 1 and 0.3 in formula 2 and 3. It is necessary to test the chemical stability of hair tonic combination between cinnamon essential oil and VCO to determine the levels of active substance during storage using gas chromatography mass spectrometry (GC-MS).

5. REFERENCES

- Aini, Q. (2017). Uji Aktivitas Pertumbuhan Rambut Kelinci Jantan Dari Sediaan Hair Tonic Yang Mengandung Ekstrak Etanol Daun Mangkokan (Nothopanax Scutellarium L.). Jurnal Farmasi Lampung, vol. 6, no. 2.
- Alamsyah, A.N. (2005). Virgin Coconut Oil Minyak Penakluk Aneka Penyakit. Jakarta: PT. Agro Media
- Allen, L.V., Popovich, N.G., & Ansel, H.C. (2011). Ansel, s pharmaceutical dosage forms and drug delivery system, 9th edition, Lippincott, USA, 389-394.

- Andersen, H., Gazerani, P., & Arendt-Nielsen, L. (2016). High-concentration L-menthol Exhibits Counter-irritancy to Neurogenic Inflammation, Thermal, and Mechanical Hyperalgesia Caused by Trans-cinnamaldehyde, *Journal of Pain*, 17, 10-12.
- Ambari, Y., Fitra, N., Arista, W., Iif, H., & Butet, S. (2020). Studi Formulasi Sediaan Lip Balm Ekstrak Kayu Secang (Caesalpinia sappan L.) dengan Variasi Beeswax. *Islamic Pharm*, 5(2), 36–45.
- Apriani, E.S, Ahmadi, A., & Noviani, V. (2021) Formulation and Evaluation of Water Fraction Hair Tonic Containing Flavonoids from Ethanolic Extract of Green Tea Leaves (Camellia sinensis L.), *Majalah obat tradisional*, Vol. 26(2), p 77-83, ISSN-p: 1410-5918 ISSN-e: 2406-9086.
- Bajaj, S., Singla, D., & Sakhuja., N. (2012). Stability testing of pharmaceutical products, *Journal of applied pharmaceutical applied*, 02 (03); 2012:129-138.
- BPOM. (2013). Peraturan Kepala Badan Pengawas Obat dan Makanan Republik Indonesia Nomor 34 Tahun 2013 Tentang Kriteria dan tata cara pengajuan notifikasi Kosmetika.Depkes RI.Jakarta.
- BPOM. (2014). Pedoman Uji Toksisitas Nonklinik Secara In Vitro. Depkes RI. Jakarta, 66-70.
- Harlim, A. (2017). Buku Ajar Ilmu Kesehatan Kulit Dan Kelamin Dasar Diagnosis Dermatologi. FK UKI, Jakarta, 100.
- Indriana, L., Wimpie, P & I Gusti, M. (2018). Minyak Atsiri Kayu Manis (Cinnamoum burmanii) Topikal Memiliki Efektivitas Yang Sama Dengan Minoxidil Dalam Menambah Panjang Rambut Dan Ukuran Diameter Folikel Rambut Pada Tikus Putih Jantan Galur Wistar (Rattus norvegicus). *Indonesian Journal of Anti Aging Medicine*, 2(1), 14–17.
- Indriyani, F., & Susi, E. (2021). Formulasi dan Uji Stabilitas Hair Tonic Ekstrak Lidah Buaya (Aloe vera L.) dan Seledri (Apium graviolens L.). *Indonesian Journal On Medical Science*, 8(1), 19.
- Kurniawidjaja, L., Fatma, L., Mila, T & Doni, H. (2021). Konsep Dasar Toksikologi Industri, Fakultas Kesehatan Masyarakat Universitas Indonesia, Depok, 124&164.
- Lestari, IS., Darusman, F., & Dewi, L, M. (2020). Formulasi Sediaan Sampo Antiketombe Dari Minyak Atsiri Kulit Batang Kayu Manis (*Cinnamomum burmani* Nees ex BI.) dan Uji Aktivitas Antijamur *Candica albicans* Secara In Vitro. *Prosiding Farmasi*, 6, 61.
- Muhammud, A., Amin, A.R., Bakar, R.A., &Jaafar, R. (2014). The Effectiveness of Coconut Oil Mixed with Herbs to Promote Hair Growth. *International Journal of Ethics in Engineering* & *Management Education*, 1(3), 27–30. SSN: 2348-4748, Volume 1.
- Putra, M.M., Dewantara, I.G.N.A., & Swastini, D. (2014). Pengaruh Lama Penyimpanan Terhadap Nilai pH Sediaan Cold Cream Kombinasi Ekstrak Kulit Buah Manggis (Garcinia mangostana L.), Herba Pegagan (Centella asiatica) dan Daun Gaharu (Gyrinops versteegii (gilg) Domke). Jurnal farmasi Udayana, Vol.3, No1.
- Puluh, E.A., Hosea, J.E., & Jainer, P.S. (2019). Formulasi dan Uji Antibakteri Sediaan Masker Gel Peel-Off Ekstrak Etanol Daun Alpukat (Persea americana Mill.) Terhadap Bakteri Staphylococcus epidermidis Sebagai Antijerawat, *Pharmacon*, 8 (4), 866.
- Rahman, A., Ika, Y., & Binar, A. (2013) Formulasi Lotion Ekstrak Rimpang Bangle (*Zingiber purpureum* Roxb) Dengan Variasi Konsentrasi Trietanolamin Sebagai Emulgator dan Uji Iritasinya. *Pharmacy*, 10, 52.
- Safitri, C & Luluk, J. (2019). Formulasi dan Uji Mutu Fisik Sediaan Lotion Ekstrak Kulit Buah Jagung (Zea mays L.), Jurnal Insan Farmasi Indonesia, 2, 181.
- Setiaji & Prayugo, S. (2006), Membuat VCO Berkualitas Tinggi, Jakarta: Penebar Swadaya.
- Sikkema, J., Debont, J & Poolman, B., 1994, Interaction of Cyclic Hydrocarbon with Biological Membranes, *The Journal of Biological Chemistry*, 269 (11), 8024.
- Stanojević, L., Stanojević, J., Savić, V., Cvetkovic, D., Kolarevic, A., Marjanović-Balaban, Ž., & Nikolic, L. (2019), Peppermint and Basil Essential Oils: Chemical Composition, in vitro Antioxidant Activity and in vivo Estimation of Skin Irritation. *Journal of Essential Oil-Bearing Plants* JEOP, 22(4):1-5, DOI-10.1080/0972060X.2019.1661793.
- Sugesti, R., Febriana, T.W., & Dwi, S.R. (2018). Karakteristik Hair Tonik Menggunakan Minyak Kayu Putih (Melaleuca sp) Sebagai Bahan Aktif, Program Studi Kehutanan, Universitas Mataram, 3, 8.
- Trayes, KP., Studdiford, JS., Pickle, S., & Tully, AS. (2013) Edema: diagnosis and management. Am Fam Physician. *PMID*: 23939641, 88(2):102-10.

- Usman, R.S, & Yuliana. (2019). Pengaruh Penggunaan Buah Alpukat dan Virgin Coconut Oil (VCO) Terhadap Perawatan Rambut Rontok. *Jurnal Pendidikan dan keluarga*, [S.l.], v. 11, n. 02, p. 74-85, ISSN 2549-9823.
- Verdini, L., Budi, S., Tiurma, T., Ahmad, S., & I, W.T.W. (2022), Komponen Bioaktif Ekstrak Akuades Kayu Manis (*Cinnamomum burmanii Blume*) Asal Sumatera, Indonesia, *Jurnal Teknologi dan Industri Hasil Pertanian*, 27 (1), 28.
- Yang W, Chen X, Li Y, Guo S, Wang Z, & Yu X. Advances in Pharmacological Activities of Terpenoids. Natural Product Communications. 2020;15(3). doi:10.1177/1934578X20903555
- Zulfa, E., Liya, L & Mimik, M. (2018). Formulasi Sediaan Krim Daun Binahong (Anredera cordifolia (Ten.) Steenis): Kajian Karakteristik Fisika Kimia Dan Uji Iritasi Kulit. *Inovasi Teknik Kimia*, *3*(1), 51