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Activity test cream anti aging extract ethanol 70% star fruits leaves (*Averrhoa bilimbi l.*) on skin new zealand rabbit backs exposed to UV-A light

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ABSTRACT

Skin is an organ that protects humans from external influences. Damage to the skin can interfere with human health and appearance. One of the most feared things is premature aging, the main cause of skin aging and malignant melanoma is the entry of ultraviolet (UV) rays from the sun. One of the plants, namely starfruit leaves, has antioxidant content that can change or reduce free radicals and also as an anti-free radical such as flavonoids, saponins, alkaloids, and tannins. Cream can be used by applying it to the skin of the face to prevent anti aging. Objective: To determine the activity of anti-aging cream of 70% ethanol extract of starfruit leaves (Averrhoa bilimbi L.) on the back skin of New Zealand rabbits exposed to UV-A light. Methodology: Extract starfruit leaves using the maceration method with 70% solvent and make a cream preparation to test anti-aging activity with concentrations of 3%, 5%, and 7%. Results: Formulation 3 with a concentration of 7 % of starfruit leaf extract cream can provide optimal effects as an anti-aging agent when exposed to UV-A rays, seen from the average value which is almost the same as the positive control.

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1. Introduction

Starfruit (Averrhoa bilimbi L.) also called sour starfruit is a type of tree that is thought to originate from the Maluku Islands. Starfruit is a type of fruit and traditional medicine plant. Starfruit leaves contain flavonoids, phenols, alkaloids, tannins and coumarins [1]. The

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ability of flavonoids as antioxidants has been widely studied, where flavonoids have the ability to change or reduce free radicals and also as anti-free radicals. Antioxidants are substances that can provide endogenous protection and exogenous oxidative stress by capturing free radicals [2].

Active antioxidant ingredients are very abundant in nature, one of which is starfruit leaves. Starfruit leaf extract contains saponins, tannins, flavonoids and alkaloids. From the content of the starfruit leaf extract, the one that has antioxidant activity is flavonoids. The flavonoid content in starfruit leaf extract is 97.28 μ g/mL. It is reported that 70% ethanol extract of starfruit leaves has an IC50 of 0.118ppm [3]. This shows that the starfruit leaf extract has strong antioxidant activity because it has an IC50 value of 50 - 100 μ g/mL, because the smaller the IC50 value, the lower the dose needed to reduce 50% of free radicals [4].

The use of cosmetics that have antioxidant activity is starting to be noticed because of concerns about the impact of cell oxidation, one of which is premature aging. Premature aging is an aging process that is faster than it should be. This can happen if someone is often exposed to ultraviolet rays [5]. Starfruit (*Averrhoa bilimbi L*) also called sour starfruit is a type of tree that is thought to originate from the Maluku Islands. Active antioxidant ingredients are very abundant in nature, one of which is starfruit leaves. Starfruit leaves contain flavonoids, phenols, alkaloids, tannins and coumarins [1].

Cream is a cosmetic that is often used by women. The definition of cream is a semi-solid preparation in the form of a thick emulsion containing about 60% water and is used for external use. Cream preparations have advantages including having a level of comfort in use and having a fairly high aesthetic value. Anti-aging cream is a cosmetic that has bioactivity that can prevent or improve signs of premature aging [6]. In accordance with the background above, the researcher wants to conduct research on starfruit leaves extracted with 70% ethanol as an anti-aging cream and then tested on the back skin of New Zealand rabbits exposed to UV-A light.

2. Method

2.1. Materials

Pyrex measuring cup, digital scale, 60 mesh sieve, rotary vacuum evaporator ika rv10, pyrex test tube, pyrex porcelain cup, measuring pipette, waterbath, spirit lamp, adhesiveness tester, spreading power tester, moisture balance, viscometer rior vt-04 rotor number 2, pH meter, analytical balance, mortar, stamper, glass object, cream container, measuring pipette, refrigerator, aluminum foil, UV-A light lamp, skin analyzer, 70% ethanol extract of starfruit leaves (*Averrhoa bilimbi L.*), stearic acid, glycerin, cetyl alcohol, distilled water, TEA, liquid paraffin, methyl paraben, concentrated HCl, 2N HCl, New Zealand rabbit.

2.2. Cream Formulation

Table 1. Cream Formulation

Material		Formulation			Function Material
	F0	F1	F2	F3	-
Starfruit Leaf Extract	-	3 g	5 g	7 g	Material Active
Stearate Acid	16 g	16 g	16 g	16 g	Emulsifier
Setil Alcohol	2 g	2 g	2 g	2 g	Thickener
Methyl Paraben	0.2 g	0.2 g	0.2 g	0.2 g	Preservative
Glycerin	8.5 g	8.5 g	8.5 g	8.5 g	Moisturizer
TEA	0.35 g	0.35 g	0.35 g	0.35 g	Emulsifier
Paraffin Liquid	10 ml	10 ml	10 ml	10 ml	Lubricants
Aquadest	Ad 100 g	Ad 100 g	Ad 100 g	Ad 100 g	Solvent

2.3. Physical Quality Testing of Starfruit Leaf Extract Cream

2.3.1. Organoleptic Test

Direct visual and post-sensory observation of the color, shape, and odor of the preparation that has been made [7].

2.3.2. Cream Homogeneity Test

The substance to be tested on a piece of glass or other suitable material must show a homogeneous composition and not show coarse grains [8].

2.3.3. pH Measurement Test

The cream is soaked in a pH meter and then waited until the pH meter number stops stably. A skin pH of around 4.5-6.5 meets the standards for skin preparation [7].

2.3.4. Spreadability Test

Spreadability or scattering test is carried out using a glass object and weights. A sample of 0.5 grams is placed on a glass object then the sample is given a 50-250 gr load using a weight, after which the diameter of the spread is measured. A good cream spreadability is between 5-7 cm [9].

2.3.5. Viscosity Test

Testing of the preparation was carried out using spindle number 1 at a speed of 60 rpm. The cream preparation was put into a beacker glass, then the spindle needle that had been installed was lowered until it reached the immersion limit of the preparation, the viscometer was run with a rotation limit of 60 rpm. Furthermore, the scale was read and recorded when the numbers shown were stable.

2.3.6. Adhesive Power

The adhesive power test was carried out by applying 0.5 grams of cream to a glass plate. The two glass plates were attached until the plates were united, given a load of 250 grams for 5 minutes after which it was released, then given a release load of 80 grams. Recording the time required for the glass objects to separate from each other [7].

2.3.7. Cream Type Test

A number of preparations are placed on a glass object, 1 drop of sudan three is added, stirred with a stirring rod. If sudan three is evenly distributed, it means the preparation is type O/A, but if there are only spots, it means the preparation is type A/M [10].

2.4. Anti Aging Cream Activity Testing

The anti-aging activity test began by irradiating the back skin of New Zealand rabbits with UV-A rays for 14 days. The smooth skin condition will become dry and wrinkled due to UV-A rays, this indicates that the rabbit's back skin is experiencing photoaging. Furthermore, the rabbit's back skin was given cream for 30 days and observed.

2.4.1. Collagen

Water content measurement is done using a collagen checker tool found in the skin analyzer device. The method is to press the power button and attach it to the surface of the skin. The number displayed on the tool is the percentage of collagen in the skin being measured [11].

2.4.2. Elasticity

Elasticity measurement is done with a tool found in the skin analyzer device. The method is to press the power button and attach it to the surface of the skin. The number displayed on the device is the percentage of elasticity in the skin being measured [11].

2.4.3. Moisture

Moisture measurement is done with a tool found in the skin analyzer device. The method is to press the power button and attach it to the surface of the skin. The number displayed on the tool is the percentage of moisture in the skin that is measured [11].

3. Results and Discussion

3.1. Cream Making Results



Figure 1. Starfruit Leaf Extract Cream

3.2. Physical Quality Test of Cream

3.2.1. Organoleptic.

Organoleptic testing was carried out with 3 replications of the formulation, there was no change in color, odor, and shape of the preparation. So that the starfruit leaf extract cream meets the standards.

3.2.2. Homogeneity.

The results of observations on cream homogeneity show that in formulation 0 to formulation 3 the cream produces a homogeneous preparation, because there are no coarse grains and lumps after the cream sample is applied to a piece of glass or other suitable transparent material.

3.2.3. pH

Table 2. pH

Formula		рН		Average ±SD
Repli	cation I	Replication II	Replication III	_
F0	3.16	3.16	3.16	3.16±0
FI	3.14	3.14	3.12	3.13±0.00
FII	3.09	3.07	3.07	3.77±0.09
FIII	3.02	3.02	3.02	3.20±4.40

Test was carried out with 3 formulation replications where during the process there was an increase, in formulation 0 the pH value was stable at 3.16, in formulation I the pH value was stable at the value 3.14, in formulation II the pH is stable at 3.07, while in formulation III the pH is stable at 3.02. According to Edy, 2016 the pH of a good preparation is in accordance with the skin pH, which is 4.5-6.5. However, the results of this study did not meet the pH standards for topical preparations, because the preparation was too thick and there was a high influence of the formulation ingredients.

3.2.4. Test Adhesion

Table 3. Test Adhesion

Formula	Test Adhesion (Second)				
	Replication I	Replication II	Replication III	Average ±SD	
F0	65	65	65	65±0.00	
FI	95	95	101	97±2.82	
FII	118	118	119	118.33±0.47	
FIII	122	122	124	122.66±0.94	

The results of the adhesive power test observations obtained showed that formulation 3, namely 70% ethanol extract cream, had high adhesive power, while formulations 0 to 2 had mark the adhesive power small. Increase The adhesive power occurs as the concentration of starfruit leaf extract increases, causing the consistency of the cream to become thicker so that its ability to adhere also increases. This has a significant effect on the preparation when applied to the skin of the back rabbit and influential when the tool that used will be washed. The requirement for good adhesion is more than 1 second.

3.2.5. Power Spread

Table 4. Power Spread

Formula	Power Spread (cm)			Average ±SD
	Replication I	Replication II	Replication III	
F0	2.5	2.25	2.25	2.33±0.11
FI	3.25	3.25	3.25	3.25±0
FII	3.3	3.3	3.3	3.3±0
FIII	3.22	3.22	3.22	3.22±0

The results of observations of the cream's spreading power test obtained showed that that formula 0 with average 2.33 own the smaller spread value compared to formula 1 is 3.25, 2 is 3.3 and 3 is 3.22. The wider the spread area produced by the cream, the greater the spread ability. better when smeared. Cream easier spread because the presence of glycerin functions as a humectant, namely maintaining the water content level in the cream by reducing water evaporation so that the cream is easier to spread and still awake humidity. According to Helen, 2016 [9] the requirement for good spreading power is 5-7 cm, but the results of this study showed that the spreading power was less than the requirement due to the thick cream preparation, so it did not have a good level of spreading.

3.2.6. Viscosity

Table 5. Viscosity

Formula	Viscosity (dPa.s)			Average ±SD
	Replication I	Replication II	Replication III	•
F0	500	500	500	500±0
FI	610	600	610	606.66±4.71
FII	900	890	900	870±42.42
FIII	940	940	940	940±0

The results of the observation of the cream viscosity showed that formula 0 to formula 3 increased along with the increase in extract concentration. The increase in viscosity is possible because the extract added to the preparation has a thick consistency and is hygroscopic, so that with the increase in the concentration of starfruit leaf extract, it can increase the viscosity value of the cream. The viscosity value requirement is >50 dPa.s, so that from this study it has mark viscosity Which Good And fulfil condition Because >50 dPa.s.

3.3. Type Cream

Table 6. Cream Type

Formula	Test Type Cream		
	I	II	III
F0	M/A	M/A	M/A
FI	M/A	M/A	M/A
FII	M/A	M/A	M/A
FIII	M/A	M/A	M/A

This test is done by adding a water-soluble dye, namely sudan tiga (red). The red dye will dissolve in the water phase, so that the water phase will be blue. From the results of the 4 cream tests, it was found that the starfruit leaf extract cream has an oil-in-water cream type. This is in accordance with previous research from Fieby et al., 2020 stating that the type of starfruit leaf extract cream is the same as this study.

3.4. Anti Aging Cream Testing On New Zealand Rabbits

3.4.1. Collagen

Table 7. Parameter Percentage Collagen Decreased

Collagen % Parameter				
Cream	Before Induction (%)	After Induction (%)	Decrease (%)	
0	35	25	10	
I	70.56	25	45.56	
II	68.7	25	43.7	
III	60.33	25	35.33	
K+	25	25	0	

Table 8. Parameter Percentage Ascension Collagen

	Collagen % parameter				
Cream	After Radiation (%)	After Induction (%)	Increase (%)		
0	25	25	0		
I	71.33	25	46.33		
II	69.52	25	44.52		
III	57.33	25	32.33		
K+	40.66	25	15.66		

The results of observations in the collagen test table showed a decrease in the percentage of reduction in formulation 0. 10%, formulation I 45.56%, formulation II 43.7%, formulation III 35.33% and positive control 0% this is seen from before induction and after induction. While the percentage increase experienced an increase in formulation 0 0%, formulation I 46.33%, formulation II 44.52%, formulation III 32.33% and positive control 15.66% this can be seen from after irradiation and after induction.

3.4.2. Elasticity

Table 9. Parameter Percentage Decrease in Elasticity

	Elasticity % Parameter				
Cream	Before induction (%)	After Induction (%)	Decrease (%)		
0	49	38.33	10.67		
I	66.32	19.33	46.99		
II	68.33	20.66	47.67		
III	52.4	21	31.4		
K+	22.33	19.33	3		

Table 10. Parameter Increase in Elasticity

	Elasticity % Parameter				
Cream	After Radiation (%)	After Induction (%)	Increase (%)		
0	9.89	38.33	2.61		
I	68.52	19.33	49.19		
II	68.45	20.66	47.79		
III	59.3	21	38.3		
K+	48.66	22.33	26.33		

The results of observations in the elasticity test table on the percentage of decrease experienced a decrease in formulation 0 10.67%, formulation I 46.99%, formulation II 47.67%, formulation III 31.4% and positive control 3% this is seen from before induction and after induction. While the percentage of increase experienced an increase in formulation 0 -28.44%, formulation I 49.13%, formulation II 47.79%, formulation III 38.3% and positive control 26.33% this can be seen from after irradiation and after induction.

3.4.3. Moisture

Table 11. Parameter Percentage Decrease Moisture

	Moisture Parameter %				
Cream	Before Induction (%)	After Induction (%)	Decrease (%)		
0	17	3	14		
I	59.36	11	48.36		
II	52.66	7	45.66		
III	36.25	3	33.25		
K+	40.77	3	21.88		

Table 12. Parameter Percentage Ascension Moisture

	Moisture Parameter %				
Cream	After Radiation (%)	After Induction (%)	Increase (%)		
0	15	3	12		
I	56.24	11	49.24		
II	49.67	7	42.67		
III	36.55	3	33.55		
<u>K</u> +	26.33	3	23.33		

The results of observations in the moisture table test showed a decrease in percentage in formulation 0 14%, formulation I 48.36%, formulation II 45.66%, formulation III 33.25% and control this is 61% positive seen from before induction and after induction. While the percentage increase experienced an increase in the 0-12% formulation, the formulation I 0%, formulation II 42.67%, formulation III 33.55% and positive control 23.33% can be seen after irradiation and after induction.

4. Conclusion

Based on the results of the research that has been conducted, the following conclusions can be drawn: Starfruit leaf extract contains flavonoid, saponin, alkaloid and tannin compounds. Starfruit leaf extract cream has anti-aging activity on the back skin of New Zealand rabbits by testing the parameters of collagen, elasticity, and moistur. Formulation 3 with a concentration of 7 grams of starfruit leaf extract cream can provide optimal effects

as an anti-aging agent exposed to UV-A rays seen from the average value which is almost the same as the positive control.

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