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Formulation Of Combined Antioxidant Serum Preparation Patikala (Etlingera Elatior) Leaves Extract And Black Cumin Seeds Extract (Nigella Sativa) By Dpph Method

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Abstract

Patikala leaves (Etlingera elatior) and black cumin seeds (Nigella sativa) are plants that are efficacious as antioxidants, so they can be used in the manufacture of drugs and cosmetics. One preparation that is widely used in beauty products is facial serum. The purpose of this study is to formulate these natural ingredients to determine the most optimum formula and preparation as an antioxidant by making F0 in the form of formulations without extracts, F1 with patikala leaf extracts (Etlingera elatior) and black cumin seeds (Nigella sativa) respectively (15%: 1.5%), F2 (20%: 2%), and F3 (25%: 2.5%). This research is an experimental research. Physical properties test with an average replication in the form of organoleptic test, homogeneity, pH, spreadability, adhesiveness, viscosity, and protective ability test. Serum preparations with both active substances can affect the physical properties of the preparation with results that meet the requirements or not in the preparation of facial serum. The preparation is also not irritating in all formulas, and the most preferred preparation is F0. The IC50 value for the best antioxidant test was found in F2 at 7,125189 µg/mL (very strong).

Keywords: Antioxidants; black cumin; patikala; serum.

1. Introduction

Free radicals are chemical compounds that are unstable and highly reactive, this is because they contain one or more unpaired electrons. Free radicals can be formed during the body's metabolism, such as substances that can trigger free radicals in the form of cigarette smoke, and in food and other pollutants. In addition, free radicals can also be exposed to radiation from electronic products such as cell phones, televisions, and computers. Free radicals produced in the body can be neutralized by compounds that are natural antioxidants for the body under normal conditions, but if the amount of free radicals is too high, then it cannot be neutralized, so it requires antioxidants from outside the body (Kurniasih, 2019; Ozil, 2014 in Herliningsih & Sholihah, 2022).

Antioxidants are compounds that can inhibit the free radical process by binding free radicals, reactive molecules, and suppressing cell damage. Antioxidant compounds can be obtained in sufficient quantities, this can improve immunology and can inhibit the development of degenerative diseases (Fahleny et al., 2014 in Herliningsih & Sholihah, 2022). UV exposure causes facial skin to become dull and unkempt. The presence of antioxidant compounds can help bind free radicals caused by excessive exposure to sunlight.

Indonesia has abundant biodiversity. The wealth of natural materials that Indonesia has has a very potential medicinal plant potential of around 9600 plant species. Currently, there are many natural ingredients developing and attracting great market interest. This means that these medicinal plants can be processed into cosmetic formulations, one of which is a serum

formulation. Serum. Serum is a cosmetic that can provide a comfortable effect, and is commonly used because it can be easily absorbed by the skin and produces a stronger effect, and is effective for skin care (Herliningsih & Sholihah, 2022).

Serum is a preparation that has a low viscosity, therefore serum is classified as an emulsion. Serum preparations have advantages, namely the active ingredients are contained in high concentrations, so the effect is stronger to absorb quickly into the skin for a more comfortable and easy effect, the viscosity is not too high so that it spreads on the surface of the skin (Farhamzah & Indrayati, 2019). Serum preparations on the market still use harmful chemicals so that serum preparations are needed to be combined with natural ingredients to reduce the harmful effects that arise from the use of cosmetic preparations such as serum.

Patikala (Etlingera elatior) is a plant endemic to the people of South Sulawesi, which is precisely found in the city of Palopo and its surroundings. Patikala plants ranging from leaves, stems, flowers, and rhizomes can be used as natural medicines. According to Handayani et al., (2014) patikala leaves (Etlingera elatior) contain bioactive compounds such as polyphenols, flavonoids, alkaloids, saponins, steroids, and essential oils that can be used as a source of antioxidants that can capture free radicals. With the content contained in patikala leaves (Etlingera elatior) as an antioxidant, it can be used as a natural raw material for making antioxidant preparations such as serum preparations.

Not only that, the content contained in black cumin seeds (Nigella sativa) is known to have very high antioxidant activity. According to Widyatmoko et al., 2016; Pratiwi, 2018; Putri, 2017; Thamrin, 2006; in Hendyana & Rahmiati (2022) black cumin (Nigella sativa) contains vitamin C, vitamin B1, vitamin B2, vitamin B6, vitamin A, zinc, and calcium which are efficacious as antioxidants.

Based on this background, the author is interested in combining the two natural ingredients to make an antioxidant preparation that meets the requirements of the physical quality test of the preparation, irritation, hedonic, and antioxidant tests.

2. Methodology

Tools and materials

The tools used in this research are analytical scales, digital scales, stirring rods, dropper pipettes, Brookfield viscometer, beaker glass, object glass, round glass, universal ph, white cloth, blender, glass jar, measuring flask, measuring pipette, horn spoon, watch glass, petri dish, UV-Vis spectrophotometer.

The materials used in this study are patikala leaves (Etlingera elatior) black cumin seeds (Nigella sativa), xanthan gum, distilled water, glycerin, potassium sorbate, sodium benzoate, ethanol 96%, ethanol 70%, methanol PA, NaOH, paraffin, phenolphthalein (pp) solution, scent (green tea), aluminum foil, filter paper, DPPH, vitamin c.

Preparation of Patikala Leaf Simplisia (Etlingera elatior)

The simplisia used in this study were patikala leaves (Etlingera elatior). Making patikala leaf simplisia (Etlingera elatior) is by first picking the leaves that have been picked with the criteria that the patikala leaves taken are still fresh and the leaves used are old leaves, choosing old leaves makes it easier for the leaf crushing process because it is easier to crush than younger leaves. Patikala (Etlingera elatior) leaves are picked at 09.00-12.00 when the photosynthetic reaction has been completed, then clean by washing with water and then

separated between dirt and leaves, then wet sorting is carried out. After that, it is aerated at room temperature, not exposed to direct sunlight for 3-5 days until dry. This is in line with the literature of the Indonesian Ministry of Health (1985) which says that drying simplisia containing essential oils uses a black cloth cover in order to avoid evaporation too quickly which can reduce the quality or evaporate the compound content in the simplisia. Then dry sorting is carried out to ensure that the simplisia is free from impurities after which the patikala leaves are made simplisia by chopping them until they are coarse. After that, the sample is then changed in shape by blending the dried simplisia and then stored in a closed container (Fauziah et al., 2020; Tohomi et al., 2014).

Making Simplisia of Black Cumin Seeds (Nigella sativa)

Making black cumin seed simplisia (Nigella sativa) is done by washing it thoroughly with water, then drying it in the open air, namely without direct sunlight to avoid damage to the active ingredients. Drying is done using a black cloth cover to avoid evaporation too quickly which can reduce the quality or evaporate the compound content in the simplisia. Then, black cumin seeds (Nigella sativa) are made into a fine powder by sifting using a fine sieve to get simplisia (Linianti et al., 2017); Depkes RI, 1985).

Simplisia Quality Standardization

Drying Shrinkage

Drying shrinkage is done by determining the constant weight of the weighing bottle by heating at 1050 for 30 minutes, then tare. Then, weigh 1-2 grams of simplisia powder and put it in a porcelain cup. Next, dry in an oven at 1050 for 30 minutes, then weigh and determine the constant weight (Depkes RI, 1995).

Moisture content

Water content is done by determining the constant weight of the weighing bottle by heating at 1050 for 30 minutes, then tare. Then, weigh 10 grams of simplisia powder and put it in a porcelain cup. Next, dry in an oven at 1050 for 5 hours, then weigh and determine the constant weight (Depkes RI, 1995).

Patikala Leaf Extraction (Etlingera elatior)

The extraction process of patikala leaves (Etlingera elatior) is by putting 50 grams of simplisia powder into a maceration container, then adding 750 mL of 96% ethanol solvent, then left for 3 days and stirring occasionally. Then, after the maceration process is complete, the liquid extract obtained is then evaporated by aerating it using a fan until a thick extract is obtained (Syarif et al., 2016 in Pramiastuti et al., 2018; Pratiwi et al., 2021).

Black Cumin Seed Extract (Nigella sativa)

Black cumin seeds (Nigella sativa) were washed, dried and pureed with a blender. The material was 340 grams then put into a maceration container and soaked using 70% ethanol solvent as much as 1 liter for 5 days, the sample was occasionally stirred, then filtered the filtrate. Soaking is done once, after obtaining the liquid extract, it is concentrated by aerating it using a fan until a thick extract is obtained (Hidayat et al., 2022; Subaidah, 2020; Pratiwi et al., 2021).

Preparation of Antioxidant Serum of Patikala Leaf Extract (Etlingera elatior) and Black Cumin Seed Extract (Nigella sativa)

Preparation of facial serum from patikala leaf extract (Etlingera elatior) and black cumin seed extract (Nigella sativa), namely by adding xanthan gum and then adding 20 times distilled water and stirring until an emulsion is formed. Then glycerin is added little by little while stirring continuously. Next, sodium benzoate and potassium sorbate that have been refined are then added to the two ingredients, then patikala leaf extract (Etlingera elatior) and black cumin seeds (Nigella sativa) are added and stirred until homogeneous. After that, 50 mL of distilled water is added, stirred until homogeneous, then stored in a container (Aryani et al., 2015, in Kurniawati & Wijayanti, 2018). Then green tea essences were added 2-3 drops and then homogenized using a magnetic stirrer and then put into a container for further testing.

Tabel 2.1 Antioxidant face serum formula

Nama Bahan	Fungsi	Konsentrasi (% b/v)			
		Formulasi 0 (*Control*)	**Formulasi I**	**Formulasi II**	**Formulasi III**
Ekstrak daun patikala (*Etlingera elator*)	Bahan Aktif	-	15	20	25
Ekstrak biji jintan hitam (*Nigella sativa*)	Bahan Aktif	-	1,5	2	2,5
Xanthan gum	Pengental	0,5	0,5	0,5	0,5
Gliserin	Humektan	10	10	10	10
Potassium sorbat	Pengawet	0,1	0,1	0,1	0,1
Sodium benzoat	Pengawet	0,1	0,1	0,1	0,1
Essens green tea	Pengaroma	q.s	q.s	q.s	q.s
Aquadest	Pelarut	Ad 50 ml	Ad 50 ml	Ad 50 ml	Ad 50 ml

Serum Antioxidant Testing of Patikala Leaf Extract (Etlingera elatior) and Black Cumin Seed Extract (Nigella sativa)

Physical Quality Testing of Preparations

Organoleptical observations are made by looking at the color, aroma, and texture of the preparation (Kurniawati & Wijayanti, 2018).

The homogeneity test is carried out using two glass objects, in which the sample is placed evenly on one of the glass objects. A good preparation must be homogeneous and free of particles that are still clumped (Kusumawati et al., 2022).

The pH test of the preparation is carried out using a universal pH. The pH paper is dipped into the preparation which is matched with the color listed on the appropriate ph paper container and the pH obtained is recorded (Rahmatullah et al., 2020). Serum pH standards are 4-6 (Hairunnisa et al., 2022) and skin pH is 4.5-6.5 (Kusumawati et al., 2022).

The spreadability test is carried out by weighing the serum preparation as much as 0.5 grams, then a round glass is taken and then placed the preparation on it, then place another glass on it and leave it for 1 minute. Then, place a 150 gram load on it and let it stand for 1 minute and finally measure the constant diameter (Hairunnisa et al., 2022). A good spreadability test range for serum preparations with a diameter between 4-7.5 cm (Sasmiyandri et al., 2019).

The adhesion test is carried out by placing the serum preparation between 2 glass objects of 0.5 grams, then for 5 minutes it is pressed using a load weighing 1 kg, then record

the time until the two glass objects are released. Good adhesion standards for serum preparations are >1 second (Hairunnisa et al., 2022; Kharisma & Safitri, 2020).

The viscosity test is carried out using a lamy rheology viscometer by inserting the preparation into a 50 mL beaker glass then spindle 4 is lowered at a speed of 50 rpm (Kusumawati et al., 2022). Good viscosity standards in serum preparations are in the range of 230-1150 cp (Hairunnisa et al., 2022).

The protective ability test was carried out by cutting the filter paper with a size of 5cm × 5cm and then dried after being moistened using phenolphthalein (PP) solution. Next, the serum preparation was smeared on the paper on the surface side. The preparation was smeared as people usually use serum (first paper). Then, filter paper of size 2.5cm×2.5cm was cut and paraffin was applied on the edge of the filter paper area (second paper). Next, stick the second paper on top of the first paper, then the NaOH solution is dripped on the paper, finally observing the test causes a reddish stain or not (Hairunnisa et al., 2022).

Hedonic Test

Hedonic test is conducted to observe the organoleptic and level of liking of serum preparations by giving questionnaires to panelists (Kurniawati & Wijayanti, 2018).

Irritation Test

The irritation test was carried out by applying the serum preparation openly on human skin. The serum preparation is applied to the forearm with a diameter of 2 cm and the results can be seen for 5 hours. Symptoms that arise are observed in the form of flushing, itching, and swelling of the skin as a sign of irritation to the skin (Liandhajani et al., 2022).

Antioxidant Test

Preparation of DPPH mother solution

Preparation of parent solution was made by weighing DPPH powder as much as 5 mg which was dissolved into a 50 ml flask until the limit mark. Preparation of DPPH mother solution is put into a 50 ml volumetric flask wrapped in aluminum foil, to avoid light dissolved using methanol PA (Ifaya, 2023).

Preparation of Sample Solution

Sample solution with a concentration of 1000 ppm as the parent sample, made by weighing 5 mg of sample and dissolving it into a 5 mL flask using methanol PA. Then make different concentrations, in this case made concentrations of 100 ppm, 120 ppm, 140 ppm, 160 ppm, and 180 ppm. Each concentration is pipetted from the sample parent solution according to the calculation into a 5 mL flask which is sufficient, namely 100 ppm pipetted 0.5 mL, 120 ppm as much as 0.6 mL, 140 ppm as much as 0.7 mL, 160 ppm as much as 0.8 mL, and 180 ppm as much as 0.9 mL (Ifaya, 2023).

Preparation of Comparison Solution

Comparison solution was made by making vitamin C mother solution. Vitamin C was weighed as much as 1 mg which was then dissolved with methanol as much as 10 ml and shaken until homogeneous. After that, the test tube is covered with aluminum foil until there are no parts exposed to light. Furthermore, vitamin C concentrations were made with various concentrations, namely 0.6 ppm, 1 ppm, 1.3 ppm, 2 ppm, and 2.6 ppm. Making vitamin C series solution is done by pipetting the vitamin C mother solution and then adding methanol to a volume of 2250 µl and then adding 750 µl DPPH (0.02%) (Dzaky, 2018).

Preparation of Blank Solution

Making a comparison solution is by using DPPH parent solution. DPPH parent solution was pipetted as much as 1 ml, then added with 3 mL methanol PA into a test tube that has been closed using aluminum foil. Homogenized and then incubated for 5 minutes, and then measured the absorbance using a UV-Vis spectrophotometer (Ifaya, 2023).

Measurement of Antioxidant Activity with DPPH Method

Testing antioxidant activity with DPPH method is done by pipetting 1 ml of DPPH solution, 2 ml of PA methanol, and 1 ml of sample ocean into a recitation tube that has been closed using aluminum foil. This test was carried out for each of the concentrations that had been made. Then incubate for 0-5 minutes. The absorbance was then measured using a UV-Vis Spectrophotometer at a wavelength of 517 nm (Ifaya, 2023).

From the absorbance data obtained, antioxidant power can be calculated by calculating % inhibition using the formula (Rahmayani et al., 2013).

$$\% \text{ Inhibition} = = \frac{\text{A blank} - \text{A sample}}{\text{A blank}} \times 100\%$$

The IC50 value is determined by making a linear curve between the concentration of the test solution (x-axis) and the % inhibition result (y-axis) obtained by the equation y = a + bx which in this case can be calculated IC50 value using the following formula (Handayani et al., 2020):

$$\text{IC50} = \frac{(50 - a)}{b}$$

Description: y = % inhibition
a = Intercept (line intersection on y-axis)
b = Skype (slope)
x = Concentration

3. Result and Discussion

3.1. Result

Test of Physical Properties of Preparations

Organoleptic test

Table 1. Organoleptic test observation data on serum preparations

Formulatian	Replicasi	Colour	Form	Aroma
F0	1	Colorless	Semi thick	Tipical base smell
	2	Colorless	Semi thick	Tipical base smell
	3	Colorless	Semi thick	Tipical base smell
F1	1	Green	Semi thick	Green tea
	2	Hi green brownish	Semi thick	Green tea
	3	Hi green brownish	Semi thick	Green tea
F2	1	Green	Semi thick	Green tea
	2	Hi green brownish	Semi thick	Green tea
	3	Hi green brownish	Semi thick	Green tea
F3	1	Green	Semi thick	Green tea
	2	Hi green brownish	Semi thick	Green tea

	3	Hi green brownish	Semi thick	Green tea

Homogeneity Test

Table 2. Observation data of homogeneity test on serum preparations

Formulation	Replication	Homogeneity
F0	1	Homogeneous
	2	Homogeneous
	3	Homogeneous
F1	1	Homogeneous (but still contains extract details)
	2	Homogeneous (but still contains extract details)
	3	Homogeneous (but still contains extract details)
F2	1	Homogeneous (but still contains extract details)
	2	Homogeneous (but still contains extract details)
	3	Homogeneous (but still contains extract details)
F3	1	Homogeneous (but still contains extract details)
	2	Homogeneous (but still contains extract details)
	3	Homogeneous (but still contains extract details)

pH Test

Table 3. Observation data of pH test on serum preparations

Formulation	pH Test			Averange	Range
	Replication 1	Replication 2	Replication 3		
F0	5	5	5	5	pH serum 4-6 (Hairunnisa *et al*., 2022) dan pH Skin 4,5-6,5 (Kusumawati *et al*., 2022)
F1	5	4	4	4,3	
F2	5	4	4	4,3	
F3	4	4	4	4	

Spreadability test

Table 4. Observation data of spreadability test on serum preparations

Formulatin	Spreadability test			Rata-rata	Range
	Replication 1	Replication 2	Replication 3		
F0	8,6 cm	8,75 cm	8,65	8,66 cm	5-7 cm (Sayuti, 2015)
F1	6,05 cm	6 cm	7,35 cm	6,46 cm	
F2	5,6 cm	6,25 cm	5,8 cm	5,88 cm	
F3	6,75 cm	7,6 cm	7,1 cm	7,15 cm	

Adhesion test

Table 5. Observation data of adhesion test on serum preparations

Formulation	Adhesion test			Averange	Range
	Replikasi1	Replikasi 2	Replikasi 3		
F0	3 detik	2 detik	2 detik	2 detik	>1 detik (Hairunnisa *et*
F1	2 detik	1 detik	1 detik	1 detik	

F2	2 detik	2 detik	1 detik	1 detik	*al*., 2022)
F3	3 detik	1 detik	1 detik	1 detik	

Viscosity test

Table 6. Observation data of viscosity test on serum preparations

Formulation	Uji viskositas			Averange	Range
	Replication 1	Replication 2	Replication 3		
F0	600 cp	440 cp	696 cp	579 cp	230-1150 cp (Hairunnisa *et al*., 2022)
F1	300 cp	504 cp	540 cp	448 cp	
F2	468 cp	516 cp	432 cp	472 cp	
F3	636 cp	372 cp	576 cp	528 cp	

Protection ability test

Table 7. Observation data of protective ability test on serum preparations

Formulation	Replication	Protection ability test
F0	1	There are red stains
	2	There are red stains
	3	There are red stains
F1	1	There are red stains
	2	There are red stains
	3	There are red stains
F2	1	There are red stains
	2	There are red stains
	3	There are red stains
F3	1	There are red stains
	2	The red stain disappeared
	3	The red stain disappeared

Irritation Test

Table 8. Observation data of irritation test on serum preparations

Respondent	Reaction to skin		
	F1	F2	F3
1	No irritation occurs	No irritation occurs	No irritation occurs
2	No irritation occurs	No irritation occurs	No irritation occurs
3	No irritation occurs	No irritation occurs	No irritation occurs
4	No irritation occurs	No irritation occurs	No irritation occurs
5	No irritation occurs	No irritation occurs	No irritation occurs
6	No irritation occurs	No irritation occurs	No irritation occurs
7	No irritation occurs	No irritation occurs	No irritation occurs
8	No irritation occurs	No irritation occurs	No irritation occurs
9	No irritation occurs	No irritation occurs	No irritation occurs
10	No irritation occurs	No irritation occurs	No irritation occurs

11	No irritation occurs	No irritation occurs	No irritation occurs
12	No irritation occurs	No irritation occurs	No irritation occurs
13	No irritation occurs	No irritation occurs	No irritation occurs
14	No irritation occurs	No irritation occurs	No irritation occurs

Hedonic Test

Table 9. Observation data of antioxidant test on serum preparations

Formulation	Indicator	Panelist			
		SS	S	AS	TS
F0	Colour	8	6	–	–
	Aroma	–	6	6	2
	Texture	3	11	–	–
F1	Colour	2	9	1	2
	Aroma	8	1	5	–
	Texture	3	6	3	2
F2	Colour	2	8	2	2
	Aroma	6	5	3	–
	Texture	3	6	5	–
F3	Colour	3	8	2	1
	Aroma	7	6	1	–
	Texture	5	4	5	–

Antioxidant Test

Table 10. Observation data of antioxidant test on serum preparations

Samples &Comparisons	**Concentration**	**Absorbanse Sample**	**Blank Absorbance**	**IC50**
F1	100 ppm	-0,05767	0,7433	278,406 µg /mL weak)
	120 ppm	-0,05167	0,7433	
	140 ppm	-0,092	0,7433	
	160 ppm	-0,08733	0,7433	
	180 ppm	0,109	0,343	
F2	100 ppm	0,067667	0,489667	7,125189 µg /mL (very strong)
	120 ppm	0,068333	0,489667	
	140 ppm	0,065333	0,489667	
	160 ppm	0,009667	0,489667	
	180 ppm	-0,06533	0,489667	
F3	100 ppm	0,024	0,343	156,0946 µg /mL (medium)
	120 ppm	0,142	0,343	
	140 ppm	0,08	0,343	
	160 ppm	0,2165	0,343	
	180 ppm	0,213	0,343	
Vitamin C	0,66 ppm	0,643	0,653	5,28 µg /mL (sangat kuat) (Dzaky, 2018)
	1 ppm	0,621	0,653	
	1,33 ppm	0,602	0,653	
	2 ppm	0,567	0,653	
	2,66 ppm	0,506	0,653	

3.2. Discussion

Test of Physical Properties of Preparations

Testing the physical properties of preparations in preparations F0, F1, F2, and F3 was carried out with 3 replications, namely every 3 cycles in 2-3 months, this is in line with the BPOM literature (2010) in Gunarti et al (2021) that testing the stability of the preparation lasts for 3 months, which means that it can be carried out during the 2-3 month testing range. In addition, testing every 3 cycles is carried out in accordance with the literature of Hidayati et al (2020) which tests 3 replications or repetitions for 3 cycles.

Organoleptic test

Organoleptic testing in the form of color, aroma, and shape. The purpose of this test is to see the physical properties of the preparation by looking at the color, aroma, and shape of the preparation during the storage process (Erwiyani et al., 2018).

The results of the F0 test from replication 1 to replication 3 are colorless, semi-viscous, and odorless. This is because there is no active substance content in the form of extracts, only bases and additives contained therein. For F1, F2, F3, the results of replication 1 were obtained green, semi-viscous, and a distinctive odor of essences (green tea), while for replicates 2 and 3 brownish green, semi-viscous, and a distinctive odor of essences (gren tea). Based on these results, it is known that each treatment for each cycle in the range of 2-3 months in the FI, F2, and F3 preparations obtained color results which in replication 1 were green but in replications 2 and 3 turned brownish green. The shape and odor of the preparation did not change significantly, which is still semi-viscous and smells typical of essences. The factor that affects the color change in the preparation according to Astra et al (2020) in Sugiastawa et al (2021) is the pH value. The presence of extract content in F1, F2, and F3 can affect the color of the preparation.

Homogeneity Test

The homogeneity test aims to see the preparation with the parameters of the level of smoothness and uniformity of texture of the preparation (Erwiyani et al., 2018). The homogeneity test results of the F0 preparation were homogeneous. For preparations F1, F2, and F3 are homogeneous but there are still clumpy extract granules, this is due to the characteristics of the extract, this is in line with the literature according to Binugraheni & Larasati (2020) the extract obtained from the maceration of kecombrang leaves or patikala leaves obtained the results of concentrated and clumpy extract consistency. The homogeneity test requirement according to (Hairunnisa et al., 2022) is homogeneous. It can be seen that the preparations F0, F1, F2, and F3, have met the requirements even though there are clumpy extract granules.

pH Test

The pH test uses a universal pH. The purpose of measuring the pH of the preparation is to determine the suitability of the serum so that irritation does not occur (Hairunnisa et al., 2022). The results of the pH test on the F0 preparation with the final result of 5. It can be known that the F0 preparation without extract has a stable pH value from each test. As for F1, the final result is 4.3, the F2 preparation has a final result of 4.3, and F3 has a final result of 4, namely the preparation from replication 1 to replication 3 has a decrease in pH value. It can

be seen that the higher the concentration value of the extract, the lower the pH value, and the longer the storage on the extract, the lower the pH value of the preparation, this is in line with the literature of Farhan et al (2023) that the pH test results are unstable because the higher the concentration of the preparation made, the lower or lower the pH value produced. The requirements of the pH test for serum preparations are 4-6, while for skin pH is 4.5-6.5 (Hairunnisa et al., 2022) Kusumawati et al., 2022). According to Farhan et al (2023) a pH that is too alkaline will cause the skin to become dry, and if the pH is too acidic it will cause skin irritation.

Spreadability test

The spreadability test aims to determine the ability of the speed of the preparation to spread when applied to the skin. The value of the spreadability of the preparation is inversely proportional to the viscosity. The greater the spreadability produced, the wider the active substance to spread on the skin (Tungadi et al., 2023). From the table above, it can be seen that preparations F0, F1, F2, and F3 have different results for each replication. The final results obtained by F0 were 8.66 cm, F1 6.46 cm, F2 5.88 cm, and F3 7.15 cm. The requirements of a good spreadability test according to Sayuti (2015) are in the range of 5-7 cm. It can be seen that F1 and F2 have met the requirements of good spreadability, while F0 and F3 have spreadability results that are too high. The spreadability that is too high (too spread) will reduce the level of user comfort (Agustiani et al., 2022). The high dispersion value of the preparation is influenced by the mixing temperature of the preparation ingredients. This is because the lower the temperature used during mixing, the higher the water content contained in the preparation so that it can produce a wide spread, besides that the length of stirring is inversely proportional to the particle size, namely the longer the stirring will cause a narrower spread so that it can more easily cause a wider spread (Baskara et al., 2020).

Adhesion test

The adhesion test aims to describe the preparation adhering well to the skin, besides that good adhesion can allow the drug to not easily escape and the longer it sticks to the skin, so that in this case it can produce the desired therapy or effect (Hairunnisa et al., 2022). Adhesion is directly proportional to viscosity, namely the greater the viscosity or high viscosity of a preparation, the longer the ability of the preparation to adhere to the skin (Hasriyani et al., 2022). The results of adhesion can be seen in the table above, with the final result of F0 for 2 seconds, F1, F2, and F3 for 1 second. It can be seen that the formula without extract (F0) has met the requirements, the formula with extract (F1, F2, F3) does not meet the requirements, this is in accordance with the requirement that good adhesion is> 1 second (Hairunnisa et al., 2022). Factors that can affect adhesion do not meet, namely the influence of the consistency of the preparation, changes in the value of adhesion that fluctuate with each replication, it can be seen that there is a relationship with viscosity because adhesion is directly proportional to viscosity, that is, if the thicker the preparation, the higher the value of adhesion, while if the more dilute there will be a decrease in adhesion, this is in line with the literature of Hasriyani et al (20 This is in accordance with the literature of Farhamzah & Indrayati, (2019) which states that serum preparations have advantages including having a concentration of active ingredients and a concentration of active ingredients. In addition, the adhesion that does not meet the requirements with no duration of the preparation can be

attached to the skin, this is in accordance with the literature of Farhamzah & Indrayati, (2019) which states that serum preparations have advantages including having a high concentration of active ingredients, so that the effect is greater quickly absorbed into the skin. It can be seen that serum preparations are quickly absorbed by the skin so that small adhesion can make serum preparations can be absorbed quickly by the skin so that the desired antioxidant effect can be achieved.22)

Viscosity test

The viscosity test uses a Brookfield viscometer with a 64 spindle at a speed of 50 rpm. According to Tungadi et al (2023) the viscosity test of the preparation aims to determine the viscosity of the preparation which is expected to be easily applied. The final results of F0 579 cp, F1 448 cp, F2 472 cp, and F3 528 cp. It can be seen that all dosage formulas have met the requirements of a good serum preparation viscosity test, namely in the range of 230-1150 cp (Hairunnisa et al., 2022). The viscosity value of F0 (without extract) is higher than F1, F2, and F3. According to Liandhajani et al (2022), the higher the concentration of extracts in serum preparations, the higher the viscosity value. In this case, it can be seen that the F0 preparation (without extract) is not in line with the literature, this is due to the influence of temperature. Temperature can cause the polymer of the preparation base to undergo changes which result in a tighter preparation. Of course, this change can make the serum preparation thicker than the initial preparation in each formula. As for other factors that can affect the increase and decrease in viscosity values according to Lumbantoruan & Yulianti (2016), namely factors that can affect viscosity, namely temperature, solution concentration, molecular weight, and pressure. So viscosity will be inversely proportional to temperature, if the temperature rises, the viscosity will decrease, and vice versa.

Protection ability test

The protective power test aims to find out which preparations can provide protective effects from heat, mechanical and chemical irritation. This is expected to achieve the criteria for a good preparation to achieve the desired therapy (Hairunnisa et al., 2022). The results of testing the protection power of F0, F1, F2 show red stains. For F3 in replication 1, the results showed that there were red stains, but in replications 2 and 3, the results showed that the red stains had disappeared with a maximum observation time limited to 5 minutes (Margisuci et al., 2015).

The serum preparation tested when dripped with NaOH which is a strong base can cause a red stain, in this case indicating that the preparation does not provide protection against external influences such as mechanics, heat and chemistry. A good preparation should be able to provide protection against external influences as indicated by the absence of red stains on the filter paper. Serum preparations are generally used at night so that when using them there is no concern about external influences such as mechanical, heat and chemical irritation. It can be seen that F0, F1, F2 do not meet the requirements for a good protective power test, while the F3 preparation has met the requirements, the requirements for good protective power, namely that it does not form a red stain or the red stain disappears (Hairunnisa et al., 2022)[2]

Irritation Test

The irritation test was carried out on 14 respondents which was obtained from the Slovin formula using an error of 20%. This is because the population used is small

(Patarianto, 2015). The results of the irritation test were obtained from 14 respondents for the three formulations, namely that there was no irritation, by looking at the symptoms that appeared on the respondents' arms in the form of redness, itching and swelling of the skin (Liandhajani et al., 2022). It can be seen that the three formulas do not cause irritation. The preparation (F0) was not tested for irritation because it did not contain extracts.

Hedonic Test

The hedonic test (liking test) was carried out on 14 respondents using the Slovin formula using an error of 20%. This is because the population used is small (Patarianto, 2015). The results of the hedonic test were obtained from 14 respondents, namely the preferred preparation of all formulas, namely F0 compared to F1, F2, and F3. This is because the F0 preparation produced has a color and texture that is more attractive than the other three preparations, even though F0 does not have an aroma like the other preparations.

Antioxidant Test

Antioxidant tests are carried out to find out how strong or high the serum preparation can be useful as an antioxidant, namely to ward off free radicals, which is one of the causes of aging on facial skin. The results of the antioxidant test can be seen in the table above, namely F1 has an IC50 value of 278.406 µg / mL, F2 has an IC50 value of 7.125189 µg / mL, and F3 has an IC50 value of 156.0946 µg / mL, while for the comparison sample vitamin C is used according to Dzaky (2018) the IC50 value of vitamin C is 5.28 µg / mL. The IC50 value has a very strong level of antioxidant strength using the DPPH method <50 µg/mL, strong 50-100 µg/mL, moderate 100-250 µg/mL, weak 250-500 µg/mL, and inactive >500 µg/mL (Susilowati & Wulandari, 2019 in Rahmadhani, 2022). In this case, it can be seen that F1 has weak antioxidants, F2 has very strong antioxidants, and F3 has moderate antioxidants, while vitamin C according to the Dzaky (2018) literature has very strong antioxidants.

The antioxidant values in FI, F2, and F3 preparations have antioxidants that fluctuate depending on the IC50 value. One of the factors that influence this is temperature and storage which can cause an increase or decrease in phenolic compounds. For example, the increase is caused by the degradation of complex compounds turning into simple phenolic compounds, the release of phenolic compounds from the cell walls of the material, the polymerization of other compounds into phenolic compounds. The decrease is due to the destruction of phenolic compounds due to high temperatures, such as the degradation of phenolic compounds which can become smaller molecules (Mahardani & Yuanita, 2021). The length of storage can also affect the antioxidant value, according to Juwitaningtyas & Khairi (2018) the longer the storage time, the more antioxidant content will increase, so it can be seen that the IC50 value of the sample rises and falls due to temperature or storage or long storage time which can cause the value antioxidants may decrease. In this study, antioxidant testing was less than optimal due to limited time and costs required for antioxidant testing research on these samples.

Apart from that, the blank and the length of incubation time affect the absorbance values and readings on the UV-Vis spectrophotometer. In this case, the blank has a big influence on the calculation of the % inhibition of a measured sample. The length of incubation time is also very influential, namely an incubation time of 5 minutes or even more than 30 minutes can change the color of a purple sample to clear or change to a yellowish color. This means that the sample being tested contains antioxidants. In line with the literature

(Wulansari, 2018) that initially DPPH is a deep purple color which provides absorption at a wavelength of 517 nm, but after undergoing reduction, DPPH will change to a diphenyl picryl hydrazine compound whose color will gradually fade to yellow and the absorption value will be proportional to the number of electrons received. In this case, the incubation was carried out for less than 30 minutes, this was because the sample absorbance could not be read on the UV-Vis spectrophotometer, this was because incubation for 30 minutes would change the DPPH compound from purple and gradually faded to clear or even yellow, and when measured the absorbance cannot be read by a UV-Vis spectrophotometer. In the research, an incubation time of 0-5 minutes was used, this is in line with the literature of Wulandari et al (2020), the length of incubation time used in determining the maximum wavelength is at 5 minute intervals until a stable absorbance is obtained. In this case, the length of time used can affect the absorbance in determining the wavelength of the sample.

4. Conclusion

Based on the results of research and data analysis of extracts and serum formulas from patikal leaf extract (Etlingera elatior) and black cumin seed extract (Nigella sativa) it can be concluded that:

a. The optimal serum preparation formula for patikala leaf extract (Etlingera elatior) with black cumin seed extract (Nigella sativa) as an antioxidant based on this research is F2 with an IC50 value of 7.125189 µg/mL.
b. Test results evaluating the physical, irritation and hedonic characteristics of serum preparations from patikala leaf extract (Etlingera elatior) with black cumin seed extract (Nigella sativa), namely that all preparations have different characteristics starting from F0, F1, F2 and F3. The best evaluation test for the physical characteristics of the preparation was formula 3 with 25% patikala (Etlingera elatior) leaf extract and 2.5% black cumin seed (Nigella sativa) extract. The irritation test resulted in no irritation in 14 respondents. The preferred hedonic test preparation is F0.
c. Antioxidant test results for serum preparations of patikala leaf extract (Etlingera elatior) with black cumin seed extract (Nigella sativa) using the DPPH method in this study, namely F1 has an antioxidant value of 278.406 µg / mL (weak), F2 is 7.125189 µg / mL (very strong), and F3 of 156.0946 µg / mL (moderate).

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6. Reference

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