International Journal Of Health, Engineering And Technology (IJHET) *E-ISSN 2829 - 8683*
Volume 4, Number 6, March 2026, *Page. 2041 - 2054*
Email : editorijhess@gmail.com

The Effect Of Variations In The Concentration Of Ethanol Extract Of Green Okra Fruit (*Abelmoschus Esculentus L. Moench*) On The Antioxidant Activity Of Face Mist Preparations Using The DPPH Method

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Abstract

Skin aging due to oxidative stress from free radicals from UV exposure and pollution requires natural antioxidants in topical products. Ethanol extract from green okra fruit (Abelmoschus esculentus L. Moench) rich in phenolics has strong potential to capture DPPH. This quantitative preclinical experimental study aims to test the effect of extract concentration (3%, 6%, 9%) on antioxidant activity and physical quality of face mist preparations. The population of face mist preparations; purposive samples include controls and three formulas with three replications. UV-Vis spectrophotometer instruments for IC50 DPPH at 516 nm (operating time 15 minutes), pH meter, Brookfield viscometer. Descriptive and inferential analysis One-Way ANOVA and Tukey HSD (SPSS, p <0.05). The results showed IC50: control 71.92 ppm (strong), Formula I 146.75 ppm (moderate), II 80.63 ppm (strong), III 78.72 ppm (strong); pH 4.57-5.15 (skin compatible); viscosity 1.22-3.75 mPa.s (meets <5 mPa.s); homogeneous. High concentrations significantly increase antioxidant activity (p<0.05). In conclusion, the 9% formula of optimized strong antioxidant face mist meets cosmetic standards, with anti-aging potential.

Keywords: Antioxidant Activity, DppH, Face Mist, Green Okra, Ic50.

INTRODUCTION

Skin aging is becoming an increasingly prominent phenomenon as the global population ages, particularly on facial skin, which is most exposed to the environment. This process is characterized by a gradual decline in skin function and capacity, affecting its structure and appearance.

Intrinsic factors such as genetics, cellular metabolism, and hormonal changes contribute to natural aging, while extrinsic factors, including exposure to ultraviolet and infrared radiation, and air pollution, accelerate damage. Continuous exposure to these elements triggers the production of excess free radicals, which damages skin cells and accelerates degeneration. The resulting free radicals cause oxidative stress, impair skin cell regeneration, and trigger inflammation, which exacerbates premature aging. The use of external antioxidants through topical products is an important strategy to neutralize free radicals and protect skin health.

Green okra (*Abelmoschus esculentus L. Moench*) fruit stands out as a natural antioxidant source thanks to its high total phenolic content (921.21 mg GAE/g), flavonoids (2.79 mg/g), and strong activity with an IC50 of 27.15 ppm via the DPPH method. However, the integration of okra ethanol extract into face mist preparations faces challenges, such as the effect of concentration on physicochemical stability and retention of antioxidant activity.

Variations in extract concentration often alter the homogeneity, clarity, and viscosity of facial mists, which can reduce the effectiveness of topical application to facial skin. Previous research has shown the need for formula optimization to maintain a low IC50 while meeting cosmetic standards.

This study aims to test the effect of varying concentrations of green okra fruit ethanol extract (3%, 6%, 9%) on antioxidant activity (DPPH method) and the physical quality of face mist. The urgency lies in the need for practical natural products to combat skin aging amidst increasing pollution exposure, while the novelty of this study is a comprehensive evaluation of the optimal okra face mist formula with the lowest IC50 and superior stability compared to previous studies.

RESEARCH METHODS

This study is a preclinical experimental study that aims to test the effect of variations in the concentration of ethanol extract of green okra fruit (Abelmoschus esculentus L. Moench) on the antioxidant activity of face mist preparations using the DPPH method. This type of experimental research was chosen because it involves manipulation of independent variables in the form of extract concentrations (3%, 6%, and 9%) to observe their effects on dependent variables such as IC50 values and physical quality of the preparation, in accordance with the principles of experimental research design that emphasize causality testing. A quantitative approach was applied through numerical measurements and statistical analysis to ensure the objectivity and generalizability of the results, as described in the quantitative research methodology.

The main instruments include a UV-Vis spectrophotometer for absorbance measurement in the DPPH test, a Brookfield viscometer for viscosity, a pH meter for acidity, and supporting tools such as a rotary evaporator, moisture balance, and analytical balance for standardization of simplicia and extracts. Data analysis techniques include descriptive analysis for organoleptic parameters, homogeneity, pH, and viscosity, as well as inferential analysis using One-Way ANOVA and post-hoc Tukey HSD in SPSS to compare differences between formulas with a significance level of $p < 0.05$. This approach ensures data validity through repeated replications and fulfillment of normality and homogeneity assumptions, according to experimental data analysis standards.

The study population consisted of face mist preparations with varying concentrations of green okra fruit ethanol extract, including a positive control formula (commercial face mist) and formulas I (3%), II (6%), and III (9%). Samples were taken purposively with three replications per formula for physical and antioxidant quality tests, meeting the representative criteria in laboratory experimental design. This sample selection followed the principle of non-probability sampling appropriate for preclinical research with a limited population.

The procedure begins with plant determination at the Traditional Health Service Unit of Dr. Sardjito General Hospital, preparation of simplicia through washing, drying, and grinding, and standardization (organoleptic, yield, drying loss, water content, ash content). Extraction is carried out by 70% ethanol maceration, followed by extract standardization, phytochemical screening, face mist formulation, physical quality evaluation (organoleptic, homogeneity, pH, viscosity), and DPPH antioxidant test through preparation of stock solution, determination of maximum λ (516 nm), operating time (15 minutes), and IC50 calculation. All stages are carried out at the Pharmacy Laboratory of Duta Bangsa University Surakarta from November 2025 to January 2026, with a systematic sequence to maintain the quality control chain.

RESULTS AND DISCUSSION

Plant Determination

Plant determination was conducted in the laboratory of the Tawangmangu Traditional Health Services Functional Implementation Unit of Dr. Sardjito General Hospital. The determination results showed that the simplicia used was the green okra plant (Abelmoschus esculentus L. Moench) from the Malvaceae family.

Standardization of Simple Drugs

To ensure the quality of plant simplicia, it is necessary to establish specific and non-specific quality standards so that standardized simplicia can be used with consistent and accountable active compound content (Wibowo et al., 2024)

Specific Parameters

1. Organoleptic

Organoleptic observations of green okra fruit simplicia include color, odor, and texture parameters shown in the following table:

Table 1. Results of Organoleptic Observations of Green Okra Fruit Simplicia

Sample	Observation		
	Color	Smell	Texture
Green okra fruit simplicia (Abelmoschus esculentus L. Moench)	Green	Typical okra	Fine powder

Non-Specific Parameters

1. Simplicia Yield

Calculation of the yield of simple drugs is used to determine the levels of secondary metabolites carried by the solvent (Rahadyana et al., 2024)

Table 2. Results of Green Okra Fruit Simplex Yield

Wet weight (g)	Dry weight (g)	Percentage (%)
10.455 g	1.079 g	10.29%

Based on Table 2, the yield value of green okra fruit simplicia was 10.29%. This indicates that the yield of green okra fruit simplicia has met the quality requirements, namely >10%.

2. Drying Loss

Table 3. Results of Drying Shrinkage of Green Okra Fruit Simples

Replication	Drying Loss (%)
1	5.55%
2	6.15%
3	6.85%
Average	6.18%

Based on Table 3, the average drying loss value of green okra fruit simplicia was 6.18%. This drying loss data indicates that the green okra fruit simplicia has met the requirements for good drying loss quality, which is no more than 10%.

3. Water content

The water content aims to maintain the quality of the simple ingredients which aims to prevent the rapid growth of fungi in the simple ingredients (Wandira et al., 2023).

Table 4. Results of Water Content of Green Okra Fruit Simplex

Replication	Water content (%)
1	5.60%
2	6.15%
3	6.85%
Average	6.20%

Based on Table 4, the average water content of green okra fruit simplicia is 6.20%. This water content data indicates that the green okra fruit simplicia meets the water content quality requirements of no more than 10%.

4. Ash Content

Determining total ash content aims to provide an overview of the total amount of ash obtained and the material remaining after the high-temperature annealing process. This residue is primarily mineral, consisting of physiological ash, which originates from the plant tissue itself, and non-physiological ash, which is residual dirt such as sand and soil attached to the plant surface (Wibowo et al., 2024).

Table 5. Results of Ash Content of Green Okra Fruit Simplicia

Replication	Ash Content (%)
1	10.90%
2	10.95%
3	11.25%
Average	11.03%

Based on Table 5, the average ash content of green okra fruit simplicia was 11.03%. This ash content data indicates that the green okra fruit simplicia meets the quality requirements for good ash content, which is no more than 15%.(Narsa et al., 2022).

Extract Standardization

Standardization is carried out by establishing specific and non-specific quality standards to maintain stability and safety, while maintaining the consistency of the active compound content in both extracts (Mewar and As'ad, 2023).

Specific Parameters

1. Organoleptic

Green okra fruit extract has a brown color, a distinctive okra odor, and a thick, creamy texture.

Table 6. Results of Organoleptic Observations of Green Okra Fruit Extract

Sample	Observation		
	Color	Smell	Texture
Green okra fruit extract (Abelmoschus esculentus L. Moench)	Chocolate	Typical okra	Thick liquid

Non-Specific Parameters

1. Extract Yield

The calculation of extract yield is used to determine the levels of secondary metabolites carried by the solvent (Rahadyana et al., 2024)

Table 7. Results of Green Okra Fruit Extract Yield

Weight of simple substance (g)	Extract weight (g)	Percentage %
1,000 g	337 g	33.7%

Based on Table 7, the yield of green okra fruit extract was 33.7%. This indicates that the yield of green okra fruit extract has met the quality requirements, namely >10%.

2. Drying Loss

Drying loss aims to provide an overview of the maximum limits of compounds lost during the drying process (Pine et al., 2023).

Table 8. Results of Drying Loss of Green Okra Fruit Extract

Replication	Drying Loss (%)
1	6.55%
2	8.90%
3	6.55%
Average	7.33%

Based on Table 8, the average drying loss value of green okra fruit extract was 7.33%. This drying loss data indicates that the green okra fruit extract has met the requirements for good drying loss quality, which is no more than 10%.

3. Water content

The purpose of moisture content testing is to determine the minimum water content in the extract. Increasing moisture content can trigger the growth of mold and mildew, resulting in decreased biological activity during storage (Azizah et al., 2022).

Table 9. Results of Water Content of Green Okra Fruit Extract

Replication	Water content (%)
1	6.55%
2	8.99%
3	6.64%
Average	7.39%

Based on Table 9, the average water content of green okra fruit extract was 7.39%. This water content data indicates that the green okra fruit extract meets the requirements for good drying loss, which is no more than 10%.

4. Ash Content

Ash content is used to determine the non-volatile components that remain during combustion and heating of organic compounds. The higher the ash content, the higher the mineral content of the extract (Utami et al., 2020).

Table 10. Results of Ash Content of Green Okra Fruit Extract (Abelmoschus esculentus L. Moench)

Replication	Ash Content (%)
1	12.05%
2	12.05%
3	12.20%
Average	12.10%

Based on Table 10, the average ash content of green okra fruit extract was 12.10%. This ash content data indicates that the green okra fruit extract meets the quality requirements for a good ash content, which is no more than 15%.(Narsa et al., 2022).

5. Ethanol Free

The ethanol-free test is conducted to determine whether or not there is any remaining ethanol in the extract. This test aims to ensure that the resulting extract is free of ethanol solvents, making it safe to use and meeting established quality requirements (Muadifah et al., 2025).

Table 11. Ethanol-Free Results of Green Okra Fruit Extract

Replication	Ethanol Content
1	No color change occurs
2	No color change occurs
3	No color change occurs

Based on table 11, it shows that the green okra fruit extract is free from ethanol, as seen from the absence of a change in the color of the extract when reacted with H2SO4 and potassium dichromate.

Phytochemical Screening

Phytochemical screening is a way to find out what secondary metabolite compounds are contained in plant extracts.

Table 12. Screening Results of Green Okra Fruit Extract

Compound	Reagent	Results	Information
Phenolic	FeCl3	+	A blackish green color is formed
Flavonoid	n-hexane+ ethanol 96% + concentrated HCl + magnesium	+	A red-orange color is formed
Alkaloid	Mayer	+	There is white sediment
	Dragendorff	+	A brownish orange precipitate forms
Saponin	Aquadest	+	Foam is formed
Tannin	Ethanol 70%+ FeCl3	+	A blue-green color forms and there is sediment.

The phenolic test results showed a positive result, indicated by the formation of a blackish-green color. This color change occurs due to the reaction between the $FeCl_3$ reagent and the aromatic hydroxyl groups, indicating that the test sample contains phenolic compounds (Tunny et al., 2023).

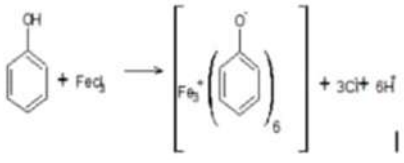


Figure 1. Identification Reaction of Phenolic Compounds with FeCl3 (Wulandari and Qodri, 2023)

Testing of flavonoid compounds showed a positive result, namely the formation of a red color. The resulting color is the result of a reaction between concentrated HCl and magnesium metal. Flavonoid compounds are oxidized by Mg2+, forming a complex with magnesium ions. The

polyhydroxyl group of flavones is reduced by magnesium metal in concentrated HCl, forming a flavylium salt, which is used to detect the presence of flavonoid compounds (Nurshazidah et al., 2023).

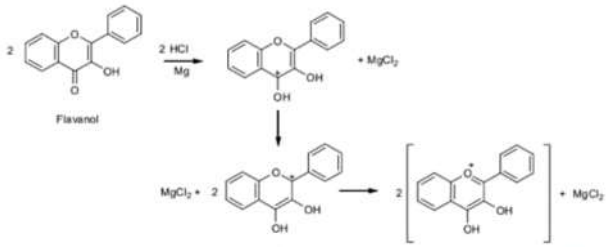


Figure 2. Flavonoid Identification Reaction with Concentrated HCl and Mg Powder (Syamsudin et al., 2022)

The test sample showed positive results for alkaloids. The test was conducted using Mayer and Dragendroff reagents. In Mayer's reagent, the formation of a white precipitate can be caused by the reaction between nitrogen in the alkaloid and the K+ metal ion from potassium tetraiodomercurate (II) to form a precipitated K-alkaloid complex (Wowor et al., 2022). The formation of a brownish-orange precipitate in the test sample with Dragendroff's reagent occurs because the precipitate is a potassium-alkaloid complex. This is produced by the formation of a coordinate covalent bond between the K+ metal ion and the alkaloid (Febry and Usman, 2024).

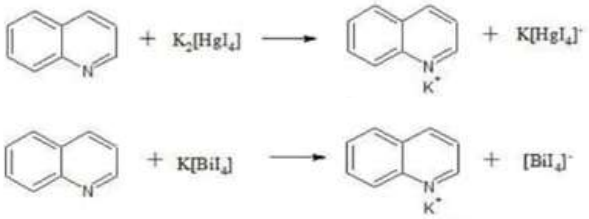


Figure 3. Alkaloid Identification Reaction with (a) Mayer, (b) Dragendroff Reagents (Hanifah et al., 2021)

The saponin test sample showed a positive result, indicated by the formation of stable foam. Foam formation is caused by the saponin compound's glycoside structure, which contains nonpolar sapogenin compounds and water-soluble polar side chains. When shaken with water, saponins can form micelles due to the presence of surface-active polar and nonpolar groups. The polar groups face outward, while the nonpolar groups face inward. This condition is what creates foam (Hanifa et al., 2021).

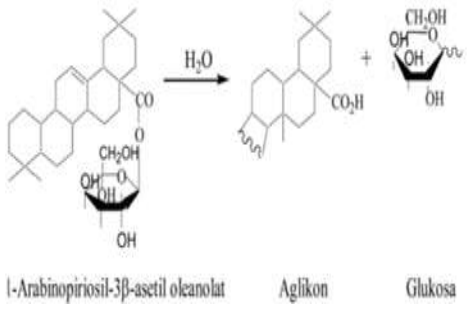


Figure 4. Identification Reaction of Saponin Compounds by Aquadest (Hanifah et al., 2021)

A positive tannin test result is indicated by the formation of a blue-green color with a precipitate. Tannin bind compounds with Fe3+ to form a complex that causes a color change (Hanifa et al., 2021).

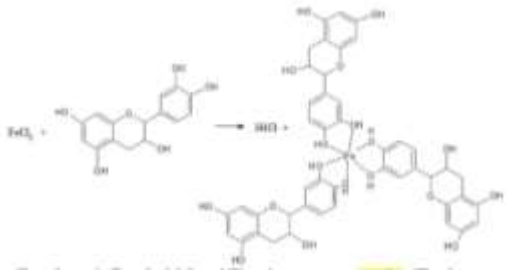


Figure 5. Identification Reaction of Tannin Compounds by FeCl3 (Hanifah et al., 2021)

Antioxidant Activity of the Preparation

The DPPH method's mechanism of action is characterized by a decrease in absorbance due to a change in the solution's color. DPPH reacts with hydrogen atoms from free radical-scavenging compounds to form the more stable DPPH hydrazine. The reaction between the DPPH reagent and the antioxidant causes a color change from purple to yellow, with the color intensity depending on the antioxidant's ability (Aryanti et al., 2021).

1. Preparation of 40 ppm DPPH Stock Solution

   This DPPH stock solution is used as a free radical indicator in antioxidant activity tests. DPPH solutions are susceptible to light and oxygen exposure, which can cause degradation. To reduce this risk, the solution should be protected with aluminum foil and stored in the dark (Ansyori et al., 2024).

2. Wavelength Determination

   Maximum wavelength measurements are performed to determine the absorbance value at the point where a substance exhibits the highest absorption. The purpose of measuring at maximum wavelength is to increase analytical sensitivity, allowing for better detection of small changes in concentration, and to reduce the possibility of measurement errors and improve the precision and accuracy of analytical results (Agustiarini and Wijaya et al., 2022).

   The maximum absorbance of a 40 ppm DPPH solution was obtained at a wavelength of 516 nm with an absorbance value of 0.767. Theoretically, DPPH is known to have a maximum absorbance at a wavelength of 517 nm. This difference in wavelength is still considered reasonable and acceptable, because according to theory, experimental observation results can experience a wavelength shift compared to the theoretical value, with a shift range that generally ranges from 0 to 4 nm (Pranata et al., 2020).

3. Determining Operating Time

   Determination of operating time is carried out to determine the length of measurement time required for a compound to react completely with another compound to form a stable reaction product. The stability of the reaction can be determined by observing changes in absorbance values from the beginning of the reaction until it reaches a relatively constant absorption condition (Maulidya et al., 2023). Based on the results of determining the operating time using a UV-Vis spectrophotometer with a measurement interval of every 1 minute for 60 minutes, it was obtained that the absorbance value began to show stability in a time span of 15 minutes with an absorbance value of 0.864 at a wavelength of 516 nm. Based on the results of the operating time obtained, the test sample that had been reacted with a 40 ppm DPPH solution was left for 15 minutes before its absorbance was measured.

4. Measurement of DPPH Comparative Activity with Face Mist

   A commercial antioxidant face mist solution and green okra fruit extract were made into a 100 ppm stock solution of 100 mL. From the stock solution, 2 mL, 4 mL, 6 mL, 8 mL, and 10 mL were taken and then dissolved with methanol to the mark in a 10 mL volumetric flask to obtain solutions with concentrations of 20 ppm, 40 ppm, 60 ppm, 80 ppm, and 100 ppm. The purpose of making concentration variations was to find the IC50 value using a mathematical equation obtained through the correlation between inhibition and sample concentration (Rusli et al., 2023). 2 mL of sample was taken from each solution and then mixed with 2 mL of 40 ppm DPPH solution and left for 15 minutes.

The absorbance of samples of each concentration was observed using a UV-Vis spectrophotometer at a maximum wavelength of 516 nm. The results of measuring the antioxidant activity of Wardah antioxidant face mist can be seen in the following table.

Table 13. Face Mist Antioxidant Test Results

Sample	Concentration	Absorbance			Average	% Inhibition	IC50
Blank DPPH	40 ppm	Rep 1	Rep 2	Rep 3			
			0.767				
*Face Mist*Commercial Antioxidants	20 ppm	0.521	0.525	0.522	0.523	31,812	71,924
	40 ppm	0.497	0.499	0.498	0.498	35,071	
	60 ppm	0.415	0.410	0.416	0.416	46,023	
	80 ppm	0.356	0.355	0.355	0.355	53,715	
	100 ppm	0.302	0.302	0.301	0.302	60,625	
Formula I	20 ppm	0.662	0.665	0.664	0.664	13,428	146.75
	40 ppm	0.581	0.561	0.595	0.579	24,511	
	60 ppm	0.547	0.542	0.544	0.544	29,074	
	80 ppm	0.531	0.531	0.531	0.531	30,760	
	100 ppm	0.496	0.483	0.474	0.484	36,897	
Formula II	20 ppm	0.605	0.604	0.605	0.605	21,121	80.63
	40 ppm	0.532	0.536	0.534	0.534	30,378	
	60 ppm	0.443	0.441	0.436	0.440	42,633	
	80 ppm	0.374	0.443	0.359	0.392	48,891	
	100 ppm	0.311	0.322	0.319	0.317	58,670	
Formula III	20 ppm	0.561	0.564	0.562	0.562	26,727	78.72
	40 ppm	0.465	0.457	0.459	0.460	40,026	
	60 ppm	0.402	0.405	0.403	0.403	47,457	
	80 ppm	0.379	0.372	0.363	0.371	51,629	
	100 ppm	0.352	0.352	0.365	0.356	53,585	

The absorbance readings were performed three times to minimize the possibility of errors during the analysis process and ensure more accurate data. The absorbance values of both commercial antioxidant face mist and green octa fruit extract face mist obtained met the good absorbance range, meeting Lambert-Beer requirements between 0.2-0.8 (Suharyanto and Prima, 2020). The resulting measurements showed that the higher the concentration (ppm) used, the lower the absorbance value due to the sample's ability to reduce free radicals in the form of DPPH (Medica et al., 2024).

The inhibition percentage (%) is used as a parameter to evaluate the ability of an antioxidant compound to inhibit free radical activity. This inhibition percentage value describes the level of antioxidant effectiveness in neutralizing free radicals, where the higher the inhibition percentage produced, the greater the compound's ability to inhibit free radicals. Thus, a high inhibition percentage value indicates stronger antioxidant activity (Kolompoy et al., 2024). The results of calculating the inhibition percentage of commercial antioxidant face mists showed 31.812% (20 ppm), 35.071% (40 ppm), 46.023% (60 ppm), 53.715% (80 ppm), and 60.625% (100 ppm). For the results of the green okra fruit extract face mist, Formula I recorded an inhibition percentage of 13.423% (20 ppm), 24.511% (40 ppm), 29.074% (60 ppm), 30.760% (80 ppm), and 36.897% (100 ppm). Formula II showed values of 21.121% (20 ppm), 30.378% (40 ppm), 42.633% (60 ppm), 48.891% (80 ppm), and 58.670% (100 ppm), while Formula III had an inhibition percentage of 26.727% (20 ppm), 40.026% (40 ppm), 47.457% (60 ppm), 51.629% (80 ppm), and 53.585% (100 ppm). The calculation results show that the lower the absorbance, the higher the inhibition percentage. This indicates that increasing the concentration of a compound is directly proportional to its activity as a free radical scavenger. Therefore, the higher the concentration used, the greater the compound's ability to scavenge free radicals (Medica et al., 2024).

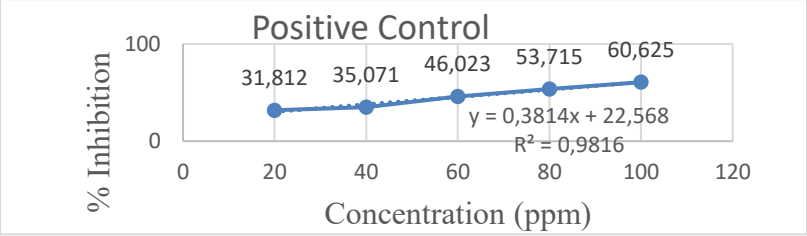
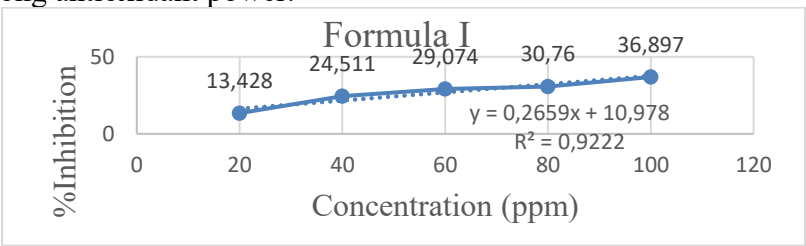


Figure 6. Graph of % Inhibition of Commercial Antioxidant Face Mist

From the calculation of the inhibition percentage (%) a standard calibration curve was produced to calculate the IC50 value of the face mist. The IC50 value is the concentration of antioxidants capable of inhibiting 50% of free radicals (Sylvia et al., 2020). The results of the linear regression equation obtained were y = 0.3814x + 22.568 and obtained an r value of 0.9908. From the results of the linear regression equation, the IC50 of the commercial antioxidant face mist preparation was 71.924 ppm, thus indicating that the commercial antioxidant face mist preparation had an IC50 value that indicated strong antioxidant power.



Picture7. Graph of % Inhibition of Face Mist Formula I

From the results of the linear regression equation, the equation y = 0.2659x + 10.978 was obtained with a correlation coefficient of r = 0.9603. Based on the linear regression equation, the IC50 value of the green okra fruit extract face mist in formula I was calculated at 146.75 ppm. This result indicates that the face mist preparation has moderate antioxidant activity of 146.75 ppm.

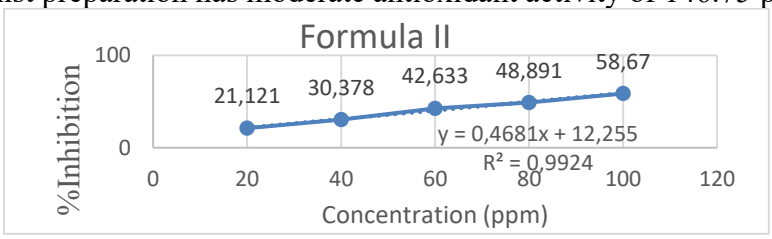
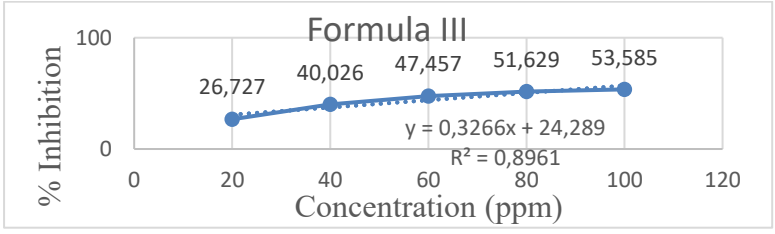


Figure 8. Graph of % Inhibition of Face Mist Formula II

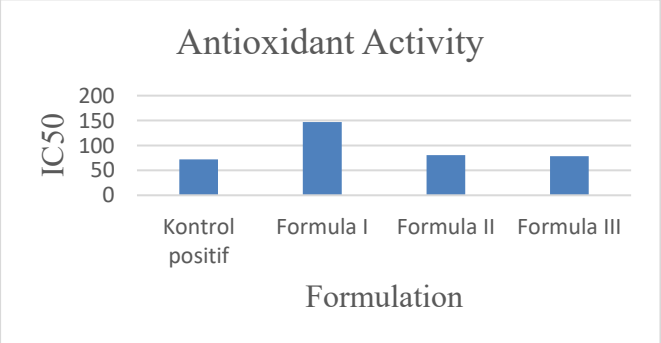
From the linear regression analysis, the equation y = 0.4681x + 12.255 was obtained with a correlation coefficient of r = 0.9962. Based on the regression equation, the IC50 value of face mist formula II was obtained at 80.63 ppm, which indicates that this preparation has relatively strong antioxidant capabilities.



Picture9. Graph of % Inhibition for Face Mist Formula III

The results of the linear regression equation obtained y = 0.3266x + 24.289 and get a value of r = 0.9466. From the results of the linear regression equation Formula III obtained IC50 face mist preparation of green okra fruit extract (Abelmoschus esculentus L. Moench) of 78.72 ppm, thus indicating that the Wardah antioxidant face mist preparation has an IC50 value that indicates strong antioxidant power.

The results of measuring the antioxidant activity between Wardah antioxidant face mist as a positive control with Formula I (3% extract), Formula II (6% extract), and Formula III (9% extract) face mist preparations showed that with increasing concentration, the resulting absorbance value decreased. This shows that the higher the concentration, the greater the IC50 value produced.



Picture10. IC50 graph.

Physical Evaluation of Preparations

1. Organoleptic

Organoleptic tests were carried out to describe the color, odor, and texture of the face mist preparation.

Table 14. Organoleptic Test Results of Face Mist Preparations

Formula	Organoleptic		
	Color	Smell	Texture
Positive control	Clear	Typical	Liquid
Formula I	Light brown	Typical okra	Liquid
Formula II	Dark brown	Typical okra	A bit thick
Formula III	Dark chocolate	Typical okra	A bit thick

Based on the results obtained, the higher the concentration of green okra fruit extract used, the more concentrated the color of the preparation will be.

2. Homogeneity

The homogeneity test aims to determine the mixing of ingredients in the green okra fruit face mist preparation.

Table 15. Face Mist Homogeneity Test Results

Formula	Homogeneity
Positive control	Homogeneous
Formula I	Homogeneous
Formula II	Homogeneous
Formula III	Homogeneous

The results of the homogeneity test on the green okra fruit face mist preparation and the commercial antioxidant face mist control preparation showed that all formulas were homogeneous with no visible coarse particles so that the face mist preparation was able to be dispersed well on the skin (Nurfita et al., 2021).

3. pH

The pH test of the preparation is carried out to determine the degree of acidity of a preparation so that the resulting preparation does not cause skin irritation.

Table 16. Results of pH Test of Face Mist Preparations

Formula	pH
Positive control	4.57
Formula I	5.15
Formula II	5.08
Formula III	5.02

The results of the pH test on the green okra fruit face mist preparation for Formula I had a pH of 5.15, Formula II had a pH of 5.08, and Formula III had a pH of 5.02. Meanwhile, the commercial antioxidant face mist control preparation has a pH of 4.57. These results indicate that the face mist

preparation meets the requirements for a good pH value for a face mist preparation because it is still within the skin's pH range, which is 4.5-6.5.(Herliningsih and Anggraini, 2021).

The higher the extract concentration used in the formulation, the lower the pH value of the preparation. This is because green okra fruit extract contains the largest group of flavonoid compounds, a phenolic compound, which is slightly acidic (Thomas et al., 2022).

Based on the results of the Shapiro-Wilk normality test, a significant value of p-value > 0.05 was obtained in all treatment groups, so it can be concluded that the pH data of the preparation is normally distributed. Levene's homogeneity of variance test obtained a significance value of 0.250 (p-value > 0.05) so it can be concluded that the pH data of the preparation has a homogeneous variance. The pH data meets the requirements for the One Way ANOVA test. The results of the ANOVA pH value show a significant difference between the concentration variations p-value < 0.05.

Table17. Results of the Tukey HSD Post Hoc Test on the pH of Face Mist Preparations

Treatment group	Treatment group	Sig	Information
Positive control	Formula I	0,000	There is a difference
	Formula II	0,000	There is a difference
	Formula III	0,000	There is a difference
Formula I	Positive control	0,000	There is a difference
	Formula II	0.022	There is a difference
	Formula III	0.001	There is a difference
Formula II	Positive control	0,000	There is a difference
	Formula I	0.022	There is a difference
	Formula III	0.077	There is no difference
Formula III	Positive control	0,000	There is a difference
	Formula I	0,000	There is a difference
	Formula II	0.077	There is no difference

Based on the results of the Tukey HSD post hoc test on the pH parameters of the face mist preparation with varying extract concentrations, it was found that the positive control was significantly different from Formula I, Formula II, and Formula III (0.000). This indicates that the addition of extracts at various concentrations significantly affected the pH value of the preparation compared to the positive control. Comparison between formulas showed that Formula I was significantly different from Formula II and Formula III (p-value <0.05). This difference indicates that increasing the extract concentration has an effect on changes in the pH of the preparation. Meanwhile, Formula II and Formula III did not show a significant difference (0.077), which indicates that increasing the extract concentration in the two formulas did not provide significant changes in pH. Overall, the results of this test indicate that the extract concentration affects the pH of the face mist preparation, but at a certain concentration the pH change tends to be stable. The pH value obtained is still within the appropriate range for topical preparations, so it is safe for use on the skin.

4. Viscosity

Viscosity testing is carried out to determine the level of viscosity in a preparation.

Table 18. Viscosity Test Results of Face Mist Preparations

Formula	Viscosity (mPa.s)
Positive control	1.22
Formula I	3.09
Formula II	3.43
Formula III	3.75

The results of viscosity testing on the green okra fruit face mist preparation for Formula I have a viscosity of 3.09, Formula II has a viscosity of 3.43, and Formula III has a viscosity of 3.75. Meanwhile, the commercial antioxidant face mist control preparation has a viscosity of 1.22. These results indicate that the face mist preparation meets the requirements for a good viscosity value for a face mist preparation because it meets the viscosity requirements for a face mist preparation, which is <5 mPa.S (Noor et al., 2023).

The higher the extract concentration used in the formulation, the higher the viscosity of the preparation. This is because the higher the extract concentration, the less water is added (Ikhsan et al., 2023).

Based on the results of the Shapiro-Wilk normality test, a significance value of more than 0.05 was obtained for all groups, so that the viscosity data for all preparations were normally distributed. Levene's homogeneity of variance test obtained a significance value of 0.551 ($p > 0.05$), so it can be concluded that the viscosity data of the preparation has a homogeneous variance.

The viscosity data meets the requirements for One Way ANOVA testing. The ANOVA results showed a significant difference in viscosity values between concentration variations ($p < 0.05$).

Table19. Tukey HSD Post Hoc Test Results for Viscosity of Face Mist Preparations

Treatment group	Treatment group	Sig	Information
Positive control	Formula I	0,000	There is a difference
	Formula II	0,000	There is a difference
	Formula III	0,000	There is a difference
Formula I	Positive control	0,000	There is a difference
	Formula II	0.001	There is a difference
	Formula III	0,000	There is a difference
Formula II	Positive control	0,000	There is a difference
	Formula I	0.001	There is a difference
	Formula III	0.002	There is a difference
Formula III	Positive control	0,000	There is a difference
	Formula I	0,000	There is a difference
	Formula II	0.002	There is a difference

A post hoc Tukey HSD test showed that the positive control, Formula I, Formula II, and Formula III differed significantly from each other ($p < 0.05$). This indicates that increasing the extract concentration significantly affected the viscosity parameters, so that each formulation had different characteristics.

The viscosity value of the preparation increased with increasing extract concentration, namely the positive control (1.22), Formula I (3.09), Formula II (3.43), and Formula III (3.75). All formulations were still below the maximum requirement, which was less than 5 mPa.S (Noor et al., 202)

CONCLUSION

This study shows that variations in the concentration of green okra fruit ethanol extract significantly affect the antioxidant activity and physical quality of the face mist preparation, with Formula III (9%) producing optimal performance in the form of IC50 78.72 ppm which is included in the strong category, pH 5.02, viscosity 3.75 mPa.s, and good homogeneity and organoleptic, all of which meet cosmetic standards (pH 4.5-6.5 and viscosity <5 mPa.s). One-Way ANOVA analysis confirmed significant differences between formulas ($p<0.05$), where increasing the extract concentration increased free radical inhibition while maintaining physical stability, confirming the potential of green okra as a natural anti-aging skin ingredient through the DPPH mechanism. These findings support the integration of herbs in practical topical formulations to combat oxidative stress from extrinsic factors such as pollution and UV. The research focused on preclinical research without in vivo testing on human skin, potential degradation of bioactive compounds during long-term storage, and a lack of microbiological and skin irritation evaluation. Suggestions for further research include accelerated stability testing, clinical studies, and combinations with other hydrating agents to enhance efficacy. Practically, this optimal formula has implications for the development of safe, effective, and commercially ready okra-based natural cosmetic products for the pharmaceutical and beauty industries, supporting the clean beauty trend in Indonesia.

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