Medical Sains : Jurnal Ilmiah Kefarmasian
Vol. 9 No. 4, October - December 2024
https:/ojs.ummada.ac.id/index.php/iojs

ANTIOXIDANT ACTIVITY OF 96% ETANOL EXTRACT OF KROKOT PLANT (*Portulaca oleraceae* L.) IN FACE SPRAY PREPARATION BY DPPH METHOD

Dina Pratiwi[1*], Abdul Aziz Setiawan[1], Dede Anggraeni[1]
[1]*Department of Pharmaceutical, Universitas Muhammadiyah A.R. Fachruddin, Tangerang, Indonesia*
**Email Corresponding: dinapratiwi@unimar.ac.id*

Submitted: July 2, 2024 ***Revised: August 27, 2024*** ***Accepted: December 18, 2024***

ABSTRACT

Krokot plants (*Portulaca oleracea* L.) are weeds that are commonly found in rice fields, plantations, and home yards. Krokot plants have many benefits, including their use as antirheumatic, antibacterial, and antioxidant drugs. The secondary metabolites contained in krokot plants, namely flavonoids, act as the main bioactive compounds. This study aimed to determine the physical properties of face spray preparations and the antioxidant activity of extracts and *face spray* preparations of 96% ethanol extract of krokot plant (*Portulaca oleracea* L.) to prevent free radicals. The research method used was Descriptive and inferential analyses were conducted in this study. Krokot plant extract was prepared by maceration using 96% ethanol solvent at a ratio of 1:10 and then used as the active ingredient of formulation (F) at various concentrations of *face spray* formulations. F1 as negative control, F2 at 0.7%, F3 at 1.4%, and F4 at 2.8%. The results of the physical evaluation of face spray preparations for all formulas met the requirements (organoleptic, pH, homogeneity, dry time test, viscosity, and spray spreadability test). The antioxidant activities of face spray preparations in F1, F2, F3, and F4 had IC_{50} values of 110.914, 106.581, 85.261, and 64.936 ppm, respectively. The conclusion of this study is that the extract and face spray preparation formula of 96% ethanol extract of krokot plant (*Portulaca oleracea* L.) have antioxidant activity. Formula 4 showed the best physical properties of face spray preparation.

Keywords: Formulation, krokot plant extract, *face spray*, antioxidant.

INTRODUCTION

Indonesia is a country rich in various types of plants, and thousands of plants have been recognized by the public as having benefits and are often used to treat various diseases, including krokot. Several decades ago, the global community began to shift its attention back to the use of natural medicines, a trend known as the "*back to nature*" movement (Widaryanto and Azizah, 2018).

Krokot is one type of plant that has medicinal properties. Krokot is commonly found in rice fields and homeyards. The krokot plant has many benefits, including its use as an antirheumatic, antibacterial, and antioxidant drug (Yuniarsih et al., 2022). However, limited information about the benefits of krokot plants results in a lack of utilization and use by the community.

Flavonoids, which are secondary metabolites found in krokot plants, play major roles as bioactive substances (Sicari, 2018). Flavonoid compounds are strong antioxidants that prevent the formation of free radicals. Free radicals are atoms or molecules that have one or more unpaired electrons in their outer orbit, making them highly reactive. At high concentrations, free radicals can cause oxidative stress, which damages cell structures. This

can accelerate the process of premature aging of the skin and increase the risk of cancer, heart disease, and various other diseases (Yuslianti, 2018).

Premature aging of the skin should be avoided because the skin is a protective layer that covers the entire surface of living organisms and is responsible for protecting the body from various external threats. Therefore, the skin must always be healthy (Sari, 2015).

Face spray is a cosmetic that provides freshness to the skin, reduces excess oil on the skin, and helps narrow the skin pores. In addition, face spray has a higher level of practicality and is safer because the risk of microorganism contamination is lower. This is due to its use by spraying without requiring direct contact with the hands, in contrast to topical preparations (Aprtitasari et al., 2018).

RESEARCH METHODS

Tools and Materials

The tools used in this study consisted of glass jars, filter paper, flannel cloth, measuring cup, *beaker glass*, glass funnel, test tube, dropper pipette, spray bottle, analytical balance (*Pyrex*), oven (*Memmert*, germany), *rotary evaporator* (IKA, germany), *waterbath* (*Grant*, UK), *volumetric* flask, stirring rod, pH meter (*Ohaus*, united states), spectrophotometer UV-Vis (*Shimadu UV-1601*, japan), pycnometer, Viscometer (*Lamyrheology Ostwald*, china). The materials used in this study were krokot plants from Singabraja Village, Babakan Village, Tenjo District, Bogor Regency, 96% ethanol (*teknik grade*), methanol (*semiconductor grade*), Phenoxyethanol, glycerin, *fragnance*, distilled water (*laboratory ragen*), 2N hydrochloric acid, Steasny reagent, Mayer reagent, 10% HCl, magnesium powder, amyl alcohol, 70% ethanol, iron (III) chloride reagent, vitamin C and DPPH powder.

Research Procedure

Material Collection

This study used krokot (*Portulaca oleracea* L.) plant extracts from Singabraja Village, Babakan Village, Tenjo District, Bogor Regency. A total of 5 kg of krokot plants were manually harvested. Stems and leaves of each plant were used.

Plant Determination

Plant determination aims to verify the authenticity of the plants that will be used during research. Determination was performed at the Biology Learning Laboratory, Faculty of Applied Science and Technology, Ahmad Dahlan University, Jl. Ringroad Selatan, Tamanan, Banguntapan, Bantul.

Simplisia Preparation

The process of making simplisia is first washing and wet sorting krokot plants as much as 5 kg, then chopping the krokot plants into small parts to facilitate the drying process. Subsequently, krokot plants were dried in an oven at 50^{o} C for approximately 5 days. The dried krokot plants were pulverized using a blender and then filtered through a sieve with a mesh size of 40.

Extract Preparation

Preparation of extracts was carried out by the maceration method; 355 grams of simplisia was soaked in 96% ethanol solvent at a ratio of 1:10 for 5 days while stirring occasionally every day, and then filtered using filter paper. Subsequently, remaceration was performed until the solution became transparent. All filtrates collected were concentrated into a thick extract using a Rotary Evaporator at 40^{o} C in a water bath.

Extract Parameter Testing

The test of extract quality parameters included specific parameters, namely extract identity and organoleptic parameters. In addition to non-specific parameters such as extract yield, water content, and ash content.

Phytochemical Screening

In this study, phytochemical screening was carried out to identify the secondary metabolites present in the krokot plant extract (*Portulaca oleracea* L.). Phytochemical screening includes flavonoid, tannin/polyphenol, saponin, and alkaloid tests.

Face Spray Dosage Formulation

The formulations were prepared using various concentrations of krokot plant extract (*Portulaca oleracea* L.).

Table I. Face Spray Formulation

Material	Concentration %				Function
	F1	F2	F3	F4	
Krokot plant extract	0 %	0,7%	1,4%	2,8%	Active substance
Glycerin	20	20	20	20	Moisturizers and emollients
Phenoxyethanol	1	1	1	1	Preservatives
Fragrance	0,16	0,16	0,16	0,16	Fragrance
aquadest	Ad 100	Ad 100	Ad 100	Ad 100	Solvent

Preparation of Face Spray

Krokot plant extract is used as an active substance in face spray preparation. The first step was to weigh all the ingredients (krokot plant extract, glycerin, phenoxyethanol, fragrance, and distilled water). Glycerin was poured into a *beaker glass and heated* in a water bath at 80^{o} C until it expanded completely (mass 1). Prepare a mortar and put the krokot plant extract, dissolve it with phenoxyethanol until dissolved, and homogeneous (Mass 2). Mixing masses 1 and 2 with hot water slowly until homogeneous (Mass 3). Add *fragrance* to Mass 3 and stir until homogeneous. Finally, the mixture was poured into a spray bottle and distilled water was added until it reached a volume of 100 mL.

Physical Evaluation of Face Spray

Organolipetic Test

Organoleptic testing was carried out to visually observe the preparation that has been prepared, including the assessment of shape, color, and aroma.

pH test

The pH of the face spray preparations was measured using a pH meter that was calibrated using a standard pH 4 and pH 7 solution or universal pH paper. Face spray preparations must meet the skin pH criteria, which are in the interval of 4.5-6.5 (Anggriani, 2022).

Homogeneity Test

Homogeneity was tested by spraying the face spray preparation on a glass or transparent surface, where the face spray preparation was expected to show a uniform distribution and no visible coarse particles.

Test Dry time

In the dry time test, the preparation was sprayed on the inside of the volunteer's forearm. Next, the time required for the sprayed liquid to dry was calculated (Anggriani, 2022).

Viscosity Test

Viscosity was measured using a *Lamy Rheology* Viscometer with spindle number R-5 at 50 rpm for 25 seconds. The viscosity range considered good is between 38-396 cp (Shavira *et al.*, 2021).

Spray Spreadability Test

The face spray was sprayed on a mica plastic sheet at a distance of 5 cm, and the spreadability of the preparation was measured using a ruler. The greater the spreadability, the larger the area of skin affected by the face spray, thus accelerating the absorption of substances into the skin.

Antioxidant Testing

Preparation of 0.05 mM DPPH Solution

In the preparation of the 0.05 mM DPPH solution, it was carefully weighed to 2 mg DPPH (BM 394.32), which was then dissolved with methanol pro analysis (p.a). The solution was put into a 100 mL volumetric flask, and the volume was sufficient to the limit mark with methanol p. a., and then stored in a dark room.

Determination of DPPH Maximum Wavelength

Determination of the maximum wavelength aims to achieve optimal absorption. The maximum wavelength measurement process is carried out by placing 4 mL of 0.05 mM DPPH solution in a cuvette, then measured using a spectrophotometer UV-Vis (*Shimadu UV-1601*) in the 510-520 nm wavelength range (Dewi et al., 2018). The maximum absorbance value was recorded as the blank absorbance value.

Preparation of Extract Test Solution

The mother liquor was prepared by dissolving 10 mg of the extract in 10 mL of methanol p. a. until homogeneous. A certain amount of this solution (0.124, 0.25, 0.5, 1, and 2 mL) was added using a pipette, and then placed into a 10 mL volumetric flask to obtain a test solution with consecutive concentrations of 12.5, 25, 50, 100, and 200 ppm. Furthermore, the volume was increased to 10 mL with methanol.

Determination of Operating Time

The first step was to take 50 mL of extract test solution, then add 4.0 mL of 0.05 mM DPPH solution, then place the mixture in a vortex and measure the absorbance at minutes 0, 5, 10, 15, 20, 25, and 30 using the maximum wavelength that was previously obtained. The operating time was the time at which the longest stable DPPH free radical-scavenging absorbance was achieved.

Preparation of Vitamin C Solution

A total of 10 mg of vitamin C powder was dissolved in 100 mL of methanol p.a in a 100 mL volumetric flask, producing a parent solution with a concentration of 100 ppm, this solution was then used to make a concentration series of 1, 2, 3, 4, and 5 ppm in a volumetric flask, with the volume adjusted to reach 5 mL using methanol p.a. each concentration of vitamin C comparison solution as much as 2 mL was put into a test tube. mM DPPH solution (2 mL) was added, homogenized by vortexing, and placed in a dark room during the operating time (Fathurachman, 2014). The absorbance was measured at the maximum wavelength that had been previously obtained.

Antioxidant Activity Testing of Extracts with DPPH

The sample solution (2 mL) was placed into a test tube, followed by the addition of 0.05 mM DPPH solution (2 mL), which was then homogenized using a vortex. The mixture was then placed in the dark during the operation (Fathurachman, 2014). The absorbance was measured at the maximum wavelength that had been previously obtained.

Antioxidant Activity Testing of *Face Spray* Preparations

A 1000 ppm stock solution was prepared with 10 mg of krokot plant extract in face spray, which was then dissolved in methanol p. a. until homogeneous, and diluted to a volume of 100 mL. Dilutions were then made with concentration variations of 10, 20, 30, 40, and 50 ppm. The test was carried out by taking each comparison sample solution of various concentrations as much as 2 mL using a micropipette, which was then placed into a vial. Each concentration solution (2 mL) was added to each solution, homogenized using a vortex, and left in the dark place at 37° C for 30 minutes. The absorbance was then measured using a UV-Vis spectrophotometer at the maximum wavelength.

Data Analysis

A descriptive method, which describes the situation objectively, was used to analyze the data. These data can be presented in the form of tables, graphs, or percentages to evaluate the preparation physically. Linear regression and percentage inhibition calculations were also performed. Antioxidant activity (percentage difference of each treatment) was analyzed using SPSS software.

RESULTS AND DISCUSSION

The results showed that 5 kg of wet krokot (*Portulaca oleracea* L.) plants was required to produce 405 grams of krokot plant simplisia powder. Then, as much as ± 355 grams of Simplisia powder was used for the maceration process, and the remaceration process was performed 3 times. The maceration method was chosen as the extraction technique because of its ease of implementation, easy availability of equipment, and simple nature. This method is generally used to extract active compounds that are not heat-resistant. It is usually used to extract simplisia, containing active ingredients that are easily soluble in solvents and do not contain substances that easily expand in the liquid. Extraction of krokot plants (*Portulaca oleracea* L.) produces a thick extract with a brownish green color and a distinctive aroma. The yield of krokot plant extract (*Portulaca oleracea* L.) is 4.718%.

The extract obtained was tested for extract quality parameters, including specific parameter tests, namely organoleptic, which aimed to describe the shape, color, and odor of the extract. In addition, the non-specific parameter test of the extract aims to determine the amount of secondary metabolites dissolved in the solvent, but it is not able to identify the type (Sari et al., 2021). Moisture content aims to provide a minimum limit or range for the amount of water content in the material (Indonesian Ministry of Health, 2000). The ash content aims to provide an overview of the internal and external mineral content derived from the initial process until the formation of the extract (MOH RI, 2000) (Indonesian Ministry of Health, 2000). The test results obtained the following data:

Table II. Quality Parameter Test Results

Testing Type	Results	Requirements
Organoleptics	Form: Thick extract Color: Brownish green Odor: Typical of extracts	-
Yield	4,718%	-
Water Content	4,540%	<10%
Ash Content	7,560%	<10%

The purpose of phytochemical screening was to identify secondary metabolite compounds present in the 96% ethanol extract of krokot plants. It also aims to qualitatively detect the active compounds that may be present in plants, especially those that have potential as antioxidants. Based on the results of phytochemical screening that has been carried out, secondary metabolite compounds detected in 96% ethanol extract of krokot plants are as follows:

Table III. Phytochemical Screening Results

No.	Phytochemical Screening	Results
1	Flavonoid Test	+
2	Tannin Test	+
3	Saponin Test	+
4	Alkaloid Test	+

Description:
(+) : Secondary metabolites present
(-) : No secondary metabolites present

Based on Table III**.** The 96% ethanol extract of krokot (*Portulaca oleracea* L.) was positive for flavonoids, tannins, saponins, and alkaloids. Antioxidant tests on krokot plant extracts were performed using the DPPH (2,2-diphenyl-1-picrylhydrazyl) method, a stable free radical. DPPH free radicals interact with the antioxidant compounds in the sample through a hydrogen atom donation mechanism, which causes a change in the color of the DPPH solution from purple to yellow. Color change was measured at a wavelength of 516 nm. The antioxidant potential of the extract was evaluated by determining its IC_{50} value (Amin et al. 2021). Based on the calculations that have been carried out, the IC_{50} value of the 96% ethanol extract of krokot plants was 124.006 ppm. This shows that 96% ethanol extract of krokot plants as an active ingredient has moderate antioxidant power because the IC_{50} value is in the range of 100-150 ppm (Molyneux 2004). Krokot plants contain antioxidants, in the form of vitamin C and flavonoids (Yuniastri et al., 2020). Flavonoids are one of the compounds that act as antioxidants because they have an -OH group attached to an aromatic carbon ring. Thwasstructure effectively stabilizes the free radical products, making them less reactive than most other free radicals. Thus, flavonoids can function as effective antioxidants (Matheos, 2014).

Indonesia, which has a tropical climate, often has dry skin. Therefore, face sprays that can refresh and moisturize the skin are in high demand. The ease of use and cleansing of face spray wasalso a factor contributing to its increasing popularity among the public. The formulation of a face spray containing natural antioxidants wasexpected to help Indonesian people prevent dry skin and protect the skin from free radical damage. The process of making face spray with 96% ethanol extract of the krokot plant starts by weighing the ingredients according to the predetermined formulation. The results of the face spray formulation are presented in Figure 1. The face spray preparation was then subjected to physical evaluation tests, including organolpetis, pH, homogeneity, dry time test, viscosity, and spray spreadability.

Figure 1. Krokot Plant (Rahmatika, 2015) and Face spray of 96% Ethanol Extract Of Krokot Plant

The organoleptic test aims to visually evaluate the preparation of face spray containing 96% ethanol extract of krokot (*Portulaca oleracea* L.), including the observation of the color, aroma, and texture of the prepared product. The results of the organoleptic test on *face spray* containing 96% ethanol extract of the krokot plant are shown in Table IV**.**

Table IV. Results of Orgnanoleptic Test Of Face Spray

Formula	Parameters		
	Shape	**Color**	**Smell**
F1	Liquid	Clear	Typical
F2	Liquid	Brownish Green	Typical
F3	Liquid	Brownish Green	Typical
F4	Liquid	Clear Brown	Typical

Based on Table IV. The results of organoleptic observations showed differences only in color. The color of the preparation became darker as the percentage of extract used in the formulation increased (Puspita et al., 2020). The face spray wasformulated in a liquid dosage form to facilitate its use on the face.

pH testing was performed to determine whether the pH of the preparation was in accordance with the pH of the skin. Face spray preparations are expected to meet skin pH standards which are in the range of 4.5 to 6.5 (Anggriani, 2022).

Table V. Results of pH Test Of Face Spray Preparation (In Triplo)

Formula	Average pH	±SD
F1	6.19	0.015
F2	6.09	0.025
F3	5.98	0.026
F4	5.79	0.01

Based on Table V. The difference in pH values among the four formulations wasrelated to variations in the concentration of active substances used, where the concentration of acidic substances from krokot plants increases. It wasknown that krokot plants have acidic properties because of the malic acid and oxalic acid content present in krokot plants. In thwasstudy, the pH values obtained ranged between 5.79 to 6.19. Thus, *face spray* preparations containing 96% ethanol extract from krokot plants are considered safe for use on the skin (Apritasari et al., 2018). The pH of face spray preparation must meet the recommended pH range for facial care products. Low pH can cause facial skin irritation, whereas high pH can cause facial skin to become dry and flaky (Cendana et al., 2021).

The homogeneity test wasan important step in the formulation of pharmaceutical preparations and aims to assess the extent of the homogeneity of a preparation. Homogeneity of the preparation wasindicated by the presence or absence of particles or foreign bodies.

Table VI. Homogeneity Test Results Of Face Spray Preparation

Formula	Homogeneity
F1	Homogeneous
F2	Homogeneous
F3	Homogeneous
F4	Homogeneous

Based

Table VI**.** The observation results show that the homogeneity wasgood because no coarse particles are visible when face spray *was*applied to the preparation glass. The prepared formula met the homogeneity test criteria because it had an even composition, indicating that all the ingredients contained in the preparation were well mixed**.**

Dry time testing was conducted to determine the time required for the face spray to dry after application to the skin. The procedure involved spraying the preparation on the inner side of the volunteer's forearm and calculating the time required for the sprayed liquid to dry completely. The desired standard drying time was < 5 minutes (Anggriani, 2022). The test results showed some variation in the drying time of each face spray formula, and all four formulas met the desired drying time standard of less than 5 minutes (Anggriani, 2022).

Viscosity was determined using a *Lamy Rheology* viscometer with spindle number R-5 at 50 rpm for 25 seconds. The range of viscosity values considered good according to the Indonesian National Standard (SNI) is–38-396 cp (Shavira et al., 2021).

Table VII. Viscosity Test Results Of Face Spray Preparation (In Triplo)

Formula	Average viscosity (cP)	±SD
F1	60.57	0.85
F2	59.84	1.72
F3	54.78	2.10
F4	52.67	1.66

Based on Table VII. The results obtained from the four formulations met the viscosity standards for *face wash* preparations. Several factors affect the viscosity of the four formulations, including solution concentration, temperature, and pressure. According to Shavira et al. (2021), the optimal viscosity range for face spray preparations is–38-396 cp.

The spray spreadability test was performed by spraying the preparation on plastic mica from a distance of 5 cm, followed by measuring the diameter of the spray using a ruler (Anggriani, 2022).

Table VIII. Results Of Spray Spreadability Test Of Face Spray Preparation (In Triplo)

Formula	Average spray power (cm)	±SD
F1	5.1 cm	0.58
F2	5.3 cm	0.58
F3	5.3 cm	0.58
F4	5.1 cm	0.29

Based Table VIII. The data obtained from the spray spreadability test of face spray preparations of the four formulations showed that the spray diameter was in a good range,

which was between 5 and 7 cm. The larger the diameter of the resulting spray, the better wasthe ability of the active substance to distribute and come into contact with the skin (Helmi et al., 2018). Thwaswasbecause the further the spray distance, the greater wasthe spread of the spray applied, which wasdirectly proportional.

Antioxidant activity test results of DPPH method

The samples that were tested for antioxidant activity included vitamin C, 96% ethanol extract of the krokot plant, and face spray preparations with F1, F2, F3, and F4 formulations, along with brand X preparations (K+). Antioxidant measurements will be carried out using the DPPH method because thwasmethod has advantages such as simplicity, fast measurement, and requires few reagents (Sayuti & Yenrina, 2015). The first step wasto determine the maximum wavelength. The maximum wavelength was determined to obtain the optimal absorption. The measurement results show that the maximum wavelength of DPPH solution was516 nm. The results are included in the visible light wavelength range, which was400-800 nm, and in accordance with the DPPH special wavelength range, which usually ranges from to 510-520 nm (Molyneux, 2004).

The determination of the operating time aims to identify the optimal time at which the test solution stably absorbs DPPH free radicals. The time at which the DPPH free radical absorbance remained stable was considered the operating time (Mulangsri et al., 2017). The test material reacted with the DPPH solution, and the absorbance was observed at a wavelength of 516 nm at certain time intervals, namely at minutes 0, 5, 10, 15, 20, 25, and 30. The results showed that at minute 20, 96% ethanol extract of krokot plants showed stable DPPH absorbance.

Vitamin C was used as a comparative control in the face spray preparation of 96% ethanol extract of krokot plant because vitamin C has been proven to have very strong antioxidant activity. If the IC_{50} value of the sample wasclose to or equal to the IC_{50} value of the positive control of vitamin C, it can be concluded that the sample has potential as a strong antioxidant alternative (Mulangsari et al., 2017). The absorbance measurement results and % inhibition of the vitamins are shown in Table IX**.**

Table IX. Absorbance and % Inhibition of Vitamin C

Concentration (ppm)	Absorbance	% Inhibition	Linear Regression	IC_{50}
1	0.403	2.892		
2	0.389	6.265	y = 2.9398x + 0.0964	16.976
3	0.378	8.916		
4	0.367	11.566		
5	0.353	14.94		

Based on Table IX, the results of absorbance measurements on various vitamin C concentration series were carried out using a UV-VwasSpectrophotometer with three repetitions. The results were calculated using the % inhibition formula. The higher the concentration, the lower the absorbance, and the higher the percentage of inhibition, the stronger wasthe antioxidant activity. The IC_{50} value of vitamin C as a positive control was 16.976 ppm (µg/ml), indicating that vitamin C has strong antioxidant activity.

Data from the comparison of absorbance and % inhibition of Vitamin C in Table 10 are presented in the form of a curve, which can be seen in Figure 1.

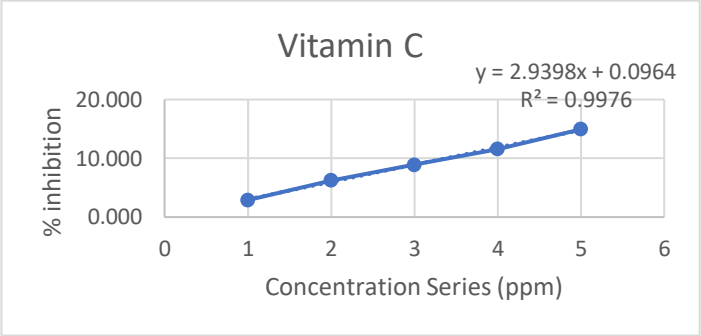

Figure 2. Vitamin C Antioxidant Activity Inhibition % Curve

Antioxidant activity testing of face spray preparations on (K+), F1, F2, F3, and F4 was carried out to determine the antioxidant activity in the form of the IC_{50} of each formula. F1 as a face spray base that does not use krokot plant extract as a negative control, F2 (0.7%), F3 (1.4%), and F4 (2.8%) as test controls with krokot plant extract content respectively 0.7 grams, 1.4 grams and 2.8 grams.

Table X. Measurement Results of IC50 in Antioxidant Activity Test Of Face Spray

Formula	IC_{50}				Antioxidant Category
	U1	U2	U3	Average	
K(+)	37.778	37.778	37.778	37.778	Very strong
F1	110.004	111.123	111.615	110.914	Medium
F2	106.581	106.581	106.581	106.581	Medium
F3	85.261	85.261	85.261	85.261	Strong
F4	64.93	64.93	64.949	64.936	Strong

Face spray brand X (K+) as a comparison, which wasa market preparation containing HI-Grade Vitamin C. Each face-spray formula was prepared at a concentration of 100 ppm, and then a concentration series of 10, 20, 30, 40, and 50 ppm was prepared. Antioxidant testing was performed using the DPPH method with a UV-Vwasspectrophotometer. The results of the IC_{50} measurement in the antioxidant activity test of the face spray for each formula are shown in Table IX**.** Based on Table X. it can be seen that the IC_{50} results in the antioxidant activity test on the face spray using the DPPH method were carried out in three repetitions. The positive control, which wasthe face spray available in the market, had an IC_{50} value of 47.225 ppm, showing very strong antioxidant activity. Because it contains HIGrade Vitamin C, which has a very strong antioxidant activity. F1, as a negative control, obtained an IC_{50} value of 110.914 ppm, which wasclassified as a moderate antioxidant activity because of the manufacture of face spray base using fragrance from lemon. Lemon fruit wasone type of fruit that produces antioxidant compounds that are effective in neutralizing free radicals (Krisnawan et al., 2018). F2, as the test control, obtained an IC_{50} value of 106.581 ppm, which wasclassified as moderate antioxidant activity because F2 contains 96% ethanol extract of krokot plants, as much as 0.7 grams. F3, as the test control, had an IC_{50} value of 85.261 ppm, which wasclassified as a strong antioxidant activity because F3 contains 96% ethanol extract of krokot plants as much as 1.4 grams. The F4 test control showed an IC_{50} value of 64.936 ppm, which wasclassified as strong antioxidant activity because F4 contains 96% ethanol extract of krokot plants as much as 2.8 grams. The higher the concentration value of 96% ethanol extract of krokot plant, the stronger the antioxidant activity.

Based on the antioxidant activity test data obtained, a normality test was carried out using the Shapiro-Wilk method. The normality test was carried out to find out whether the

antioxidant activity data from each group or the observed treatment was normally distributed or not, the results of the normality test (Shapiro-Wilk) showed that the data was not normally distributed with a significance value of <0.05 or p<0.05. Because the data wasnot normally distributed, the Kruskal-Wallwastest wascarried out to find out whether there wasa significant difference between variables in the data that does not meet the requirements of the ANOVA test. The results showed that there was a significant difference in the average between each formula (Asympt. Sig. 0.005 or p<0.05). Thwasshows that there wasa significant difference between the six treatment groups.

CONCLUSIONS

From the results of the study, it can be concluded that the extract and formulation of face spray preparations of ethanol extract of 96% krokot plant (*Portulaca oleracea* L.) show antioxidant activity. In addition, face spray ethanol extract of 96% krokot plant (*Portulaca oleracea* L.) fourth formulation with an ethanol extract concentration of 2.8% showed optimal physical characteristics.

ACKNOWLEDGMENTS

We would like to thank Muhammadiyah A.R Fachruddin University for their support in conducting thwasresearch.

LITERATURE

Amin, A., Paluseri, A., & Linggotu, R. P. 2021. Antioxidant Activity Test of Ethanol Extract of Leaf Stem and Jumpai Flower (Glinus oppositifolius (L.) Aug. DC.). Fullerene Journal of Chemistry, 6(1), 14-19. https://doi.org/10.37033/fjc.v6i1.237.

Anggriani, R. (2022). Formulation and Physical Stability Test of Face Mist Containing Cucumber Extract. (*CucumwasSativus* L.) as antioxidant (*Doctoral dissertation*, Universitas 17 agustus 1945).

Apristasari, O., Yuliyani, S. H., Rahmanto, D., & Srifiana, Y. (2018). FAMIKU (Face Mist-Ku) Utilizing Purple Cabbage and Bengkuang Extracts as Antioxidants and Facial Moisturizers. *Faculty of Pharmacy and Science, University of Muhammadiyah Prof DR Hamka*, *5*(2), 35-40.

Cendana, Y., Adrianta, K. A., & Suena, N. M. D. S. (2021). Spray Gel Formulation of Sandalwood Essential Oil (Santalum album L.): as an Anti-Inflammatory Preparation Candidate. *Medicamento Scientific Journal*, *7*(2), 84-89.

Ministry of Health of the Republic of Indonesia, 2000. *General standard parameters of medicinal plant extracts*. Ministry of Health, Directorate General of Food and Drug Control, Directorate of Traditional Medicine Control. First Printing: Jakarta. pp. 10-12.

Dewi, N. N. A., & Mustika, I. W. (2018). *Nutrition content and antioxidant activity of black garlic*. *International Journal of Health Sciences*, *2*(1), 11-20.

Fathurrachman, D. A. (2014). Effect of Solvent Concentration on Antioxidant Activity of Soursop Leaf Ethanol Extract (*Annona muricata* Linn) with DPPH Free Radical Inhibition Method. *Thesis.* Jakarta: Syarief Hidayatullah State Islamic University .

Helmi, F., & Khaldun, I. (2018). Characteristics of Leaf Powder and Spray Preparation of Starfruit Wuluh (*Averrhoa Bilimbi* L.) Leaf Extract as a Facial Cleanser. *Scientific Journal of Chemistry Education Students*, *3*(2).

Krisnawan, A. H., Budiono, R., Sari, D. R., & Salim, W. (2018). Antioxidant potential of local and imported peel and pulp juice of local and imported lemon (*Citrus lemon*). *Proceedings of SEMNASTAN*, 30-34.

Matheos, H. (2014). Antioxidant activity of moon tree leaf extract (*Pisonia alba*). *Pharmacon*, 3 (3).

Molyneux, P. *The use of stable free radical diphenylpicril-hydrazyl* (DPPH) *for estimating antioxidant activity*. (2004). Songklanakarin J. *Sci. Technol*. 26(2):211-219.

Mulangsri, D. A. K., Budiarti, A., & Saputri, E. N. (2017). Antioxidant activity of fraction diethylether fraction of arumanwasmango fruit (*Mangifera indica* L.) with DPPH method . *Journal of Pharmascience*, *4*(1).

Puspita, W., Puspasari, H., & Restanti, N. A. 2020. Formulation And Physical Properties Test Of Spray Gel From Ethanol Extract Of Buas Buas Leaf (*Premna serratifolia* L.) Weni. *Scientific Journal of Pharmaco Bahari*, 11(2), 145-152.

Rahmatica, Pritarani. 2015. Extraction and Antioxidant Stability Test of Krokot (*Portulaca oleracea* L.) as a Free Radical Scavenger. 2015. PhD Thesis. Muhammadiyah University of Malang.

Sari, A. N. 2015. Alternative antioxidants to counteract the dangers of free radicals on the skin. *Elkawnie: Journal of Islamic Science and Technology*, *1*(1), 63-68.

Sari, Y., Syahrul & Iriani, D. 2021. Phytochemical Screening and Antioxidant Activity in Mussels (*Pilsbryoconcha* Sp.) With Different Solvents. *Indonesian Journal of Agricultural Technology and Industry*. 13(1). 16-20.

Sayuti, K, & Yenrina, R, 2015, *Natural* and *Synthetic Antioxidants*, Andalas University Press, Padang.

Shavira, S., Margaretta, A. D., Sandra, A. D., Sitorus, R. U., & Fatmaria, F. (2021). Formulation and Stability Test of *Zingiber zerumbet* Rhizome Extract Suspension Preparation. Herb-Medicine *Journal: A Scientific Periodical of Herbs, Medicine and Health*, 4(4), 7-13.

Sicari, V., Loizzo, M. R., Tundis, R., Mincione, A., & Pellicano, T. M. (2018). *Portulaca oleracea L. (Purslane) extracts display antioxidant and hypoglycaemic effects*. J. Appl. Bot. Food Qual, 91(1), 39-46.

Widaryanto, E., & Azizah, N. (2018). *Perspectives on nutritious medicinal plants: Opportunities, cultivation, processing, and utilization*. Brawijaya University Press.

Yuniarsih, N., Haryani, A., Farmasi, F., Karawang, U., & Karawang, I. (2022). Formulation And Physical Stability Test of krokot Extract Facial Serum (*Portulaca* oleracea Linn). In *Journal of Buana Farma* (Vol. 2).

Yuniastri, R., Hanafi, I., & Sumitro, E. A. (2020). Antioxidant Potential of krokot (*Portulaca oleracea*) as Functional Food. Journal of *Agricultural Engineering Tropical and Biosystems*, 8(3), 284-290.

Yuslianti, E. R. (2018). *Introduction to free radicals and antioxidants*. Deepublish.