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Research Article

Determination Antioxidant Activity of *Coffea Arabica*, *Coffea Canephora*, *Coffea Liberica* and Sunscreens Cream Formulation for Sun Protection Factor (SPF)

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ABSTRACT

Coffee is a highly consumed and popular beverage consumed in many countries. Its ingredients have a powerful antioxidant capacity and have the potential as sunscreen to protect the skin. This study aimed to examine the antioxidant activity of Arabica, robusta and liberica coffee extracts and the SPF activity of the coffee extract cream formulation. Coffee were macerated with three types of solvents with polarity gradients. The fraction of each coffee was analyzed for antioxidant activity using DPPH and formulated into the cream. Furthermore, the cream was analyzed for its SPF activity. The results of this study indicated that the methanol fraction of Robusta and Arabica coffee has the best antioxidant activity with IC50 values of 8.98 (ppm) and 13.13, respectively. Meanwhile, Liberica coffee has the best antioxidant in the ethyl acetate fraction, IC50 = 10.90 (ppm). In addition, the best SPF values for Robusta, Liberica and Arabica coffees were found in F3 cream with the category of Very Good Protection; 36.087 ± 0.0005; 35.007 ± 0.0005; 36,867 ± 0.0005 respectively.

Key words: Coffee, Antioxidant, Cream, SPF.

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INTRODUCTION

The aging process is a physiological process that cannot be avoided and affects all organs of the body including the skin. Sun exposure generally consists of ultraviolet (UV) rays which contribute to making the skin wrinkled, rough and also experiencing thickening.[1] DNA is the main molecule that regulates the function of every cell in the body. According to dermatologists, the main target that is damaged due to UV light exposure is cell DNA.[2] The impact of DNA damage by UV rays can be seen from the damage to the growth of cells.[3] When skin is exposed repeatedly to UV rays, the skin will produce MMP expression in RNA and protein, which causes premature aging. Mice that give UV exposure regularly cause thickening of the skin epidermis, then resulting in skin wrinkling and gelatinase activity from MMP-2 and MMP.[4]

Exposure to UV rays, namely UV B or UV A wavelengths on the skin, triggers the synthesis and transfer of melanin into keratonocytes. This results in thickening of the epidermis of the skin. Melanocytes are dendrite cells that are near the basal layer of the stratum germinativum.[5] Melanocytes will synthesize melanin, a large polymer that is bound to proteins. This melanin absorbs light with very large wavelengths between 200-2,400 nm, so it is a good screen to protect the destructive effects of UV rays. The keratinization process also acts as a physiological mechanical protection and lasts a lifetime.[6]

Antioxidants have been shown to help reduce the skin's intrinsic (natural) signs of aging while restoring and protecting the skin from extrinsic (environmental) aging.[7,8] Natural antioxidants (applied topically or ingested) can strengthen the activity of the body's endogenous antioxidant defense systems, thus providing additional protection from oxidative stress. The chlorogenic acid compounds, condensed proanthocyanidin, quinic acid, and ferulic acid in arabica coffee resulted in an increase in the accumulation of structural proteins such as collagen I and collagen IV, as well as a reduction in collagenase (MM P-1) and IL-1b1 (inflammatory mediators).[9]

The use of natural ingredients from plants for the treatment of diseases is still widely practiced this is based on the consideration that the side effects of natural medicines are smaller than pure chemical drugs. In addition, coffee is one of the agro industry commodities in Jambi Province. However, the fundamental problem in the coffee bean roasting process is the accuracy in selecting the roasting time (t) and temperature (T) in the thermal process that matches the characteristics of the coffee beans.[10] Roasting temperature and time greatly affect chemical and physical changes which generally result in changes in chemical composition, namely: (i) changes or degradation of polysaccharide compounds, (ii) decreases or increases in caffeine content, (iii) decreases in protein content, (iv) decreases in compounds -compounds that are not palafified and (v) increase in free fatty acids. The main constituent elements of coffee beans are Carbon, Hydrogen, Nitrogen, Oxygen and various other minerals and metals.[11]

Coffee beans naturally contain various types of volatile compounds such as aldehydes, furfural, ketones, alcohols, esters, formic acid, and acetic acid which have volatile properties. Compounds that cause a sharp or sour taste such as tannins and acetic acid will be lost and some will react with amino acids to form melancidin compounds which give a brown

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color. The results of preliminary analysis studies using LC-MS on Liberica, Arabica, and Robusta coffees show that the roasting process causes changes in the composition of the compounds contained in coffee. In Liberica coffee after roasting, there are no derivative compounds of chlorogenic acid, quinic acid, and ferulic acid.[12] Green arabica coffee contains chlorogenic acid and green robusta coffee contains ferulic acid derivatives. Meanwhile, roasted robusta coffee contains derivative compounds of ferulic acid and quinic acid.[13]

The content of chlorogenic acid, quinic acid, and ferulic acid derivatives contained in coffee has the potential to be used for anti-aging because of its antioxidant properties that can bind to free radicals.[13] These antioxidant properties can also strengthen the activity of the body's endogenous antioxidant defense system, thus providing additional protection from oxidative stress, thereby preventing premature aging.

The phenolic compounds extracted from Arabica coffee can reduce levels of MMP-1 and IL-1b. In addition, upregulated gene expression for four collagen structural proteins and derived gene expression for three MMPs were also observed to infer the reparative effect of Coffeaarabica extract on photoaged skin. Preliminary studies that have been conducted have shown that arabica, robusta and liberal coffee contain derivatives of chlorogenic acid, ferulic acid and caffeine which are antioxidants and can be used as anti-aging.[12]

Unfortunately, research about the compound content of robusta, arabica and liberica of both total extract and fraction has not been studied yet in more depth. It is necessary to isolate and characterize antiaging bioactive compounds. The results are expected to answer the potential of bioactive compounds that are antiaging potential by Antioxidant and SPF activity.

MATERIALS AND METHODS

Materials

The coffee beans used in this research were robusta, liberica and Arabica. Robusta and Arabica were collected from Kerinci Regency, while Liberica from Tanjung Jabung Barat Regency, Jambi Province. The solvents that were used for extraction and chromatography were technical solvents that have been distilled were methanol, n-hexane, benzene, diethyl ether, methylene chloride, acetone, ethyl acetate, and chloroform (Sigma-Aldrich). Vacuum liquid chromatography (VLC) with Merck 60 GF254 silica gel, gravity chromatography with Merck 60 silica gel (230 - 400 mesh), and purity analysis of compounds by thin-layer chromatography on plates coated with Merck 60 GF254 silica gel 0.25mm carried out according to the previous studies.[14]

Determination of the melting point was carried out with the Fisher John melting point apparatus, for the prospective chemical structure determination of compounds as antibiotics, and UV-Vis was required. Incubator, autoclave, laminar air flow cabinet, loop, ELISA plate and other equipment commonly used in microbiology labs were used.

METHODS

Extraction and isolation

50g of roasted Arabica, Robusta, and Liberica coffee were extracted with methanol as a solvent, followed by fractionation using ethanol, n-hexane, dichloromethane and ethyl acetate as solvents. The initial methanol extract was also tested for antibiotics against ethanol fraksinate, n-hexane, dichloromethane and ethyl acetate. Bioactive compounds were isolated by column chromatography. The extracted compounds were first simplified using vacuum liquid column chromatography. Fraction 1 was subjected to gravity column chromatography to obtain four subfractions. Fraction 1.3 was selected because it had the best TLC stain pattern in the form of a single-tailed stain. Figure 1 shows the TLC results of fraction 1.3 in three eluents: n-hexane: ethyl = 9: 1; n-hexane: chloroform = 6: 4; and n-hexane: acetone = 8:2.[14]

Antioxidant activity test

DPPH radical scavenging

The radical scavenging abilities of extract and fraction of three different of coffee were based on reaction with 1,1-diphenyl-2-picrylhydrazyl radical (DPPH) and compared to standards, ascorbic acid (AA). Determination of antioxidant activity by DPPH method was adapted for use with microplates.[7] Briefly, 10 mg of the sample was dissolved in 10 ml of methanol, so that a concentration of 1 mg/ml was obtained for the determination of antioxidant activity. 0.2 ml of the sample solution was piped with a micro pipette into the vial, then added 3.8 ml of 50 µM DPPH solution. The solution mixture was homogenized and left for 30 minutes in the dark at room termperature (25°C). Absorption was measured by a UV-Vis spectrophotometer at a wavelength of 517 nm.[7] Scavenging ability was expressed as the inhibition percentage and was calculated by the following equation:

Scavenging ability (%)

$$\frac{(Abs_{control} - Abs_{sample})}{Abs_{control}} x\ 100$$

Where $Abs_{control}$ = absorbance of DPPH radical in 80% methanol and Abs_{sample} = absorbance of samples and standards in 80% methanol + DPPH. The antioxidant activity of all samples was expressed as IC_{50}, which was defined as the concentration (ppm) of samples required to inhibit the formation of DPPH radicals by 50%.[8] IC_{50} were calculated by Prohibit analysis, and were performed in triplicate.

Preparation of anti-aging of the ethanol extract of arabica coffee, liberica and robusta

Preparation of cream preparations was carried out by melting the oil phase, then adding to the water phase with an emulsifier, and stirring until a mass of cream mixture forms.[15] The oil phase consists of stearic acid, cetyl alcohol, and propyl paraben. The water phase consists of triethanolamine, glycerin, methyl paraben and aquadest. Each phase was melted at 70 ° C on a hot plate. After melting, the oil phase is slowly added to the water phase at 70 ° C while continuing to stir. The two masses are crushed in a heated mortar to form a creamy mass. After it is cool enough, then add the extract of arabica coffee, liberica coffee and robusta coffee slowly into the cream base while continuing to stir until it is homogeneous. The formula for the antiaging cream is presented in Table 1.

Sun Protection Factor (SPF) activity test

The SPF was determined *in vitro* by spectrophotometer, as described by previous research.[16] Cream formulation was dissolved at a concentration

Table 1: The formula for the antiaging cream.

Material	Weight (%)		
	F1	F2	F3
Arabica Coffee, Liberica Coffee, Robusta Coffee	1	3	5
Stearic acid	10	9	8
Cetyl alcohol	3	4	5
TEA	1,5	1,5	1,5
Glycerine	1,8	1,8	1,8
Methyl Paraben	0,3	0,3	0,3
Propyl Paraben	0,3	0,3	0,3
Aquadest	Ad 100	Ad 100	Ad 100

of 2 mg/mL in ethanol (0.2%) followed by ultrasonication for 5 min and then filtered. 1 mL aliquot was transferred to a 10 mL volumetric flask and diluted to ethanol (0.02%). The absorption spectra of the samples were recorded in the range of 290 to 320 nm for every 5 nm using 1cm quartz cell and ethanol as a blank. Three determinations were made at each point, followed by the application of the Mansur equation.[17]

$$SPF = CF \times \sum EE(\lambda \times I(\lambda) \times ABS(\lambda),$$

where: EE (λ) is erythemal e ect spectrum; I (λ) is solar intensity spectrum; ABS (λ) is absorbance of Test material; CF is correction factor (–10).

RESULT AND DISCUSSION

The sample extraction process (Arabica, Robusta and Liberica) was carried out by maceration using methanol, then partitioned with n-hexane and ethyl acetate. Separation of chemical compounds for all fractions was carried out using isolation techniques. The initial stage is to separate using LVC (Liquid Vacuum Column) and gravity chromatography (KKG) column. Silica gel for column packing was activated at 1100C for 15 minutes. Every 15 grams of the sample is impregnated with 15 grams of silica gel. Column packing is done by wet. The sample extraction process (Arabica, Robusta and Liberica) was carried out by maceration using methanol as a solvent, then partitioned with n-hexane and ethyl acetate (Table 2).

Antioxidant activity

The antioxidant activity was carried out by the DPPH method. The essay was carried out for all coffee fractions of the three types of coffee, namely the hexane fraction, ethyl acetate fraction, and methanol fraction from Arabica, liberica and Robusta coffee.

Test results of coffee extract cream

The test consisted of organoleptic test, pH, spread ability, homogeneity, and cream type. The test results are presented as follows. The parameters observed in this organoleptic were aroma, color, texture and consistency of the preparation. The results of observations on physical appearance (organoleptic) showed that the dosage forms of all formulas had cream forms (semi-solid) according to the specifications made. Organoleptic observations were carried out visually using the five senses to describe the sunscreen cream preparations that had been made. The organoleptic test result is presented in Table 3.

The Robusta coffee in all formulas has a distinctive aroma of Arabica coffee, semi-solid texture and soft consistency (Table 4). While the F1 color has a pale brown color, while the F2 has a light brown color and F3 has a dark brown color. The evaluation of the pH value is carried out to determine the pH value of the cream preparation which will affect the safety and comfort of using the preparation on the skin. Following are the results of the evaluation of the pH value of the cream preparations are presented in the Table 4.

Table 2: The percent of yield.

No.	Extract	Extract mass (g)	% Yield
	Arabica		
1.	n-Hexane	92.00	38.17
2.	Ethyl Acetate	85.00	35.27
3.	Methanol	62.00	25.73
	Robusta		
4	n-Hexane	21.81	10.32
5	Ethyl Acetate	44.59	21.09
6	Methanol	53.77	25.29
	Liberica		
7	n-Hexane	21.23	9.40
8	Ethyl Acetate	31.21	13.63
9	Methanol	46.53	20.32

Table 3: Antioxidant activity of coffee fraction.

Sample	Concentration (ppm)	Robusta		Liberica		Arabica	
		% Inhibition	IC_{50} (ppm)	% Inhibition	IC_{50} (ppm)	% Inhibition	IC_{50} (ppm)
Ascorbic Acids	0	0.00	**IC_{50} = 32.29**	0.00	**IC_{50} = 32.29**	0.00	**IC_{50} = 32.29**
	10	21.77		21.77		21.77	
	30	14.11		14.11		14.11	
	50	97.37		97.37		97.37	
n-hexane	0	0.00	**IC_{50} = 15.35**	0.00	**IC_{50}= 73.65**	0	**IC_{50}= 165.66**
	100	72.25		16.84		2.87	
	300	77.03		17.94		11.48	
	500	96.41		36.84		14.83	
Ethyl acetate	0	0.00	**IC_{50}= 11.12**	0.00	**IC_{50}= 10.90**	0	**IC50 = 21.66**
	100	83.54		83.54		30.86	
	300	91.62		93.54		82.78	
	500	93.30		93.78		94.26	
Methanol	0	0.00	**$IC_{50=}$ 8.98**	0.00	**IC_{50} = 11.02**	0.00	**IC50 = 13.13**
	100	89.95		52.15		22.01	
	300	96.17		92.34		49.52	
	500	96.88		93.78		77.27	

Table 4: Organoleptic evaluation.

	R1	R2
F1	**Aroma**: Coffee **Color**: Pale brown **Texture**: Semi solid **Consistence**: Soft	**Aroma**: Coffee **Color**: Pale brown **Texture**: Semi solid **Consistence**: Soft
F2	**Aroma**: Coffee **Color**: Light brown **Texture**: Semi solid **Consistence**: Soft	**Aroma**: Coffee **Color**: Light brown **Texture**: Semi solid **Consistence**: Soft
F3	**Aroma**: Coffee **Color**: Dark brown **Texture**: Semi solid **Consistence**: Soft	**Aroma**: Coffee **Color**: Dark brown **Texture**: Semi solid **Consistence**: Soft

Table 5: Coffee pH test results.

Arabica Coffee			
	F1	F2	F3
R1	7.00	6,6	5,9
R2	6.90	6,5	6,0
Liberica Coffee			
	F1	F2	F3
R1	6,6	6,5	6,3
R2	6,8	6,4	6,1
Robusta Coffee			
	F1	F2	F3
R1	6,7	6,3	6,3
R2	6,8	6,2	6,3

Based on the table 4, the pH results obtained by each coffee preparation from 3 different variations have a pH range of 5.9-7.0. This shows that the three coffee preparations have a pH that is safe and according to the standards based on the requirements of standarts, which ranges from 3.5-8. So it is hoped that the formulated cream will not irritate the skin (Table 5).

The spread ability evaluation was conducted to determine the spreading ability which affects the comfort of the cream when applied to the skin. In this study, the observed parameter to measure the spread ability of the cream preparations was to measure the diameter of the spread of the cream resulting from the application of a certain load (Table 6).

Based on the table above, the dispersion power value obtained is in the range of 5-7 cm. This value indicates that the spread ability of all formulas has a good value because according to previous studies, the value of the spread ability ranges from 5-7 cm.[18] Therefore, a good cream preparation must meet the predetermined dispersion evaluation requirements. The spread ability test is used to describe the ease with which the cream is applied to the skin.

Homogeneity evaluation aims to determine the distribution of active substances in cream preparations where the cream must have a homogeneous structure and do not show any spots and coarse particles. The results of the homogeneity evaluation of the cream are presented in the Table 7.

Based on the Table 7, for each Arabica coffee, Liberica coffee and Robusta coffee in formula 3 the cream preparation is not homogeneous. Meanwhile, formula 1 and formula have homogeneous preparations. The cream must show a homogeneous structure and do not show any spots. Moreover, the evaluation of the type of cream aims to determine the type of emulsion from the cream preparation made. Tests carried out by the method of staining with methylene blue. The evaluation results of the type of cream are presented in the Table 8.

In testing the methylene blue dripped on the cream preparation visually showed the results of the homogeneous distribution of the blue color in the cream preparation. So that the cream can be categorized into the type of oil in water (M/A). Methylene blue or brilliant blue FCF has good solubility in water. So that it can be used as an indicator of the type of cream where if the cream preparation has a water dispersing phase (M / A) the positive results form a blue or greenish color which is easily spread evenly in the cream preparation (Table 8).

Sun protection factor (SPF) assay

During the product development process, the *in vitro* SPF determination method acts as a screening test and reduces the number of *in vivo* experiments.[16,19] Measurement Results SPF values at wavelengths 290 – 320 were analyzed using the Mansur Question and categorized their activities (Table 9). The highest average SPF value for liberica, robusta and Arabica coffee is formula F3 with a value 35.007 ± 0.005; 36,087 ± 0.001; 36,887 ± 0.0005 respectively.

One of the most critical factors affecting skin physiology is exposure to solar radiation, specifically UV radiation.[16] Sunlight is a mixture of different UV radiations, with each component exerting a distinct effect

Table 6: Result of spread ability test.

Arabica Coffee			
	F1	F2	F3
R1	5,9 cm	5,7 cm	5 cm
R2	6 cm	5,5 cm	5 cm
Liberica Coffee			
	F1	F2	F3
R1	6,8 cm	6,5 cm	5,3 cm
R2	6,7 cm	6,3 cm	5,5 cm
Robusta Coffee			
	F1	F2	F3
R1	7 cm	6,5 cm	6 cm
R2	7 cm	6,4 cm	6,1 cm

Table 7: The result of homogeneity test.

Arabica Coffee			
	F1	F2	F3
R1	homogeneous	homogeneous	inhomogeneous
R2	homogeneous	homogeneous	inhomogeneous
Liberica Coffee			
	F1	F2	F3
R1	homogeneous	homogeneous	inhomogeneous
R2	homogeneous	homogeneous	inhomogeneous
Robusta Coffee			
	F1	F2	F3
R1	homogeneous	homogeneous	inhomogeneous
R2	homogeneous	homogeneous	inhomogeneous

Table 8: Evaluation results of coffee cream types.

Arabica Coffee			
	F1	F2	F3
R1	M/A	M/A	M/A
R2	M/A	M/A	M/A
Liberica Coffee			
	F1	F2	F3
R1	M/A	M/A	M/A
R2	M/A	M/A	M/A
Robusta Coffee			
	F1	F2	F3
R1	M/A	M/A	M/A
R2	M/A	M/A	M/A

Table 9: The average SPF value of coffee cream.

Coffe	Concentration (ppm)	SPF			SPF (Average)	Grade
		I	II	III		
Liberica	F1R1	6.419 ± 0.005	6,412± 0.005	6,446± 0.005	6,425± 0.005	LP
	F1R2	6. 270 ± 0.005	6,270± 0.005	6,267± 0.005	6,269± 0.005	LP
	F2R1	21.485 ± 0.001	21,504± 0.001	21,439± 0.001	21,476± 0.001	GP
	F2R2	31.895 ± 0.001	31,858± 0.001	30,910± 0.001	31,554± 0.001	GP
	F3R1	35. 007 ± 0.0005	35,007± 0.0005	35,007± 0.0005	35,007± 0.0005	VGP
	F3R2	35.007 ±0.0005	35,007±0.0005	35,007±0.0005	35,007± 0.0005	VGP
Robusta	F1R1	6.720 ± 0.005	7.00 ± 0.005	7.00 ± 0.005	6.907± 0.005	LP
	F1R2	6.024 ± 0.005	6.00 ± 0.005	6.00 ± 0.005	6.008± 0.005	LP
	F2R1	28.237 ± 0.0001	28.327 ± 0.0001	28.007 ± 0.0001	28.190± 0.0001	GP
	F2R2	32.307 ± 0.0001	32.007 ± 0.0001	32.078 ± 0.0001	32.130± 0.0001	GP
	F3R1	35,007 ± 0.0005	35,007 ± 0.0005	36,007 ± 0.0005	36.087 ± 0.0005	VGP
	F3R2	35,007 ± 0.0005	35,007 ± 0.0005	36,007 ± 0.0005	36.087 ± 0.0005	VGP
Arabika	F1R1	7.16 ± 0.002	7.30 ± 0.002	7.28 ± 0.002	7.2509 ± 0.001	LP
	F1R2	9.78756 ± 0.002	9.80 ± 0.002	9.83 ± 0.002	9.8082± 0.001	LP
	F2R1	25.212 ± 0.0003	21.321 ± 0.0003	23.332 ± 0.0003	23.288± 0.0003	GP
	F2R2	31.556 ± 0.0003	31.653 ± 0.0003	31.453 ± 0.0003	31.554 ± 0.0003	GP
	F3R1	35.007 ± 0.0003	35,007 ± 0.0003	35,007 ± 0.0003	36.877 ± 0.0005	VGP
	F3R2	35.007 ± 0.0005	35,007 ± 0.0005	35,007 ± 0.0005	36.867 ± 0.0005	VGP

(LP: Low-protection; GP: Good Protection; VGP: Very Good Protection)[20]

on the skin. UVA is an inducer of oxidative free radical damage to DNA and the other macromolecules in the cells. At the same time, UVB is a potent activator of inflammation and is highly mutagenic, and causes DNA dimers.[21] Exposure to UV radiation causes skin inflammation, ROS-mediated DNA damage, oxidative stress, dysregulation of cellular signaling pathways, and immunosuppression.[22] The primary health risk associated with these changes includes aging, cataracts, non-melanoma, immune system damage, and melanoma cancer. The sun protection factor (SPF) is the measure to determine the efficacy of a UV protectant. It indicates the ratio of time needed to produce sunburn on sunscreen-protected skin to the time required to cause sunburn on unprotected skin (31.49). Coffee is a plant with wide bioactivity potential including anti-inflammatory, anti-carcinogenic, antioxidant, and antibacterial agents.[23,24]

Coffee is a highly consumed, especially in Indonesia. Coffee is a popular beverage consumed in many countries and its ingredients have a powerful antioxidant capacity.[25] An animal study showed that coffee improved liver oxidative balance in rats fed a high-fat diet.[26,27] Also, an epidemiological study has found an inverse association between coffee consumption and severe periodontitis.[28] On the other hand, some studies showed that coffee and some of its components might have detrimental effects on periodontal tissues.[29-31] Coffee contains several antioxidant compounds. Among them, chlorogenic acid is a major coffee polyphenol with a powerful antioxidant capacity.[27] Coffee's active compounds can act as antioxidants and have the potential as sunscreen to protect skin.

The SPF value is a quantitative measurement of the effectiveness of a sunscreen formulation. The efficacy in preventing sunburn and other skin damage a sunscreen product should have a wide range of absorbance between 290 and 400 nm. The evaluation of the efficiency of a sunscreen formulation has been assessed through *in vitro* tests.

In this research, three different coffee, Arabica, Robusta, and Liberica, and three different fractions were formulated onto the cream. The SPF values of samples obtained using the UV spectrophotometric method were shown in **Table 10**. It can be observed that the SPF value found for F1 for all coffee cream have low protection grade, following F2 good protection, and F3 Very Good Protection. F3 contains 5% of coffee extract. It seems that the concentration of 1% (F1) and 2% (F2) has low and good protective activity. Among F3's three types of coffee, the difference in SPF value is not so significant.[20,32]

CONCLUSION

Our finding in this research is that the ethyl acetate fraction of arabica coffee, hexane fraction, ethyl acetate, and methanol of robusta coffee and liberica coffee has antioxidant activity. The test results of the anti-aging cream product of coffee extract from the three types of coffee showed that the organoleptic test of pH, dispersibility, and homogeneity at a concentration of 3% for all types of coffee gave the best results. The SPF values of the three formulations of coffee show that F1 (low protection), F2 (good protection), and F3 (very good protection).

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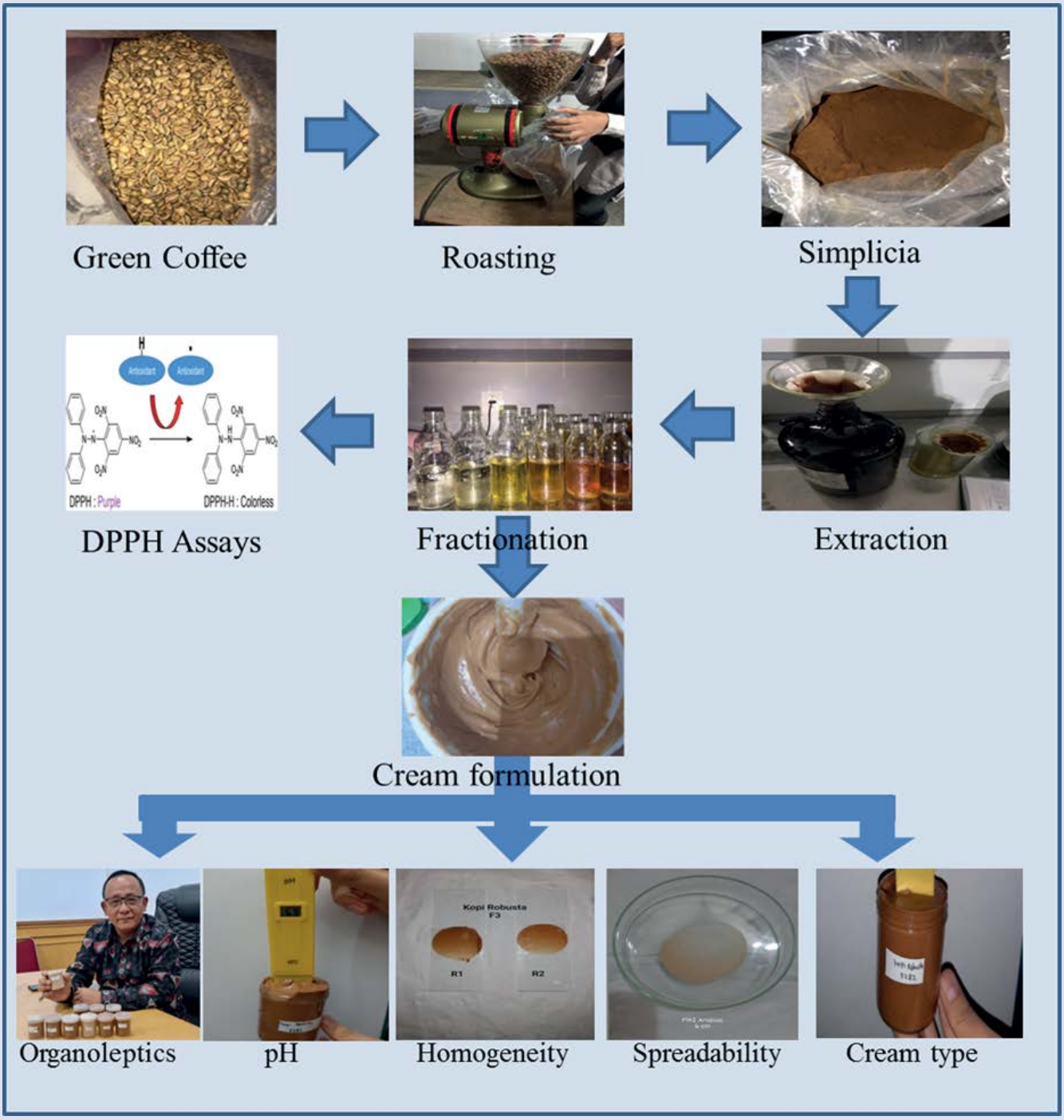
GRAPHICAL ABSTRACT



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