

RESEARCH ARTICLE

Antioxidant Activities and Antioxidant Cream Formulation of Corn Silk (*Zea Mays L*) Extract

Fifia Wulan Safitri*, Ardhan Syahreza, Siti Farah H, M Cahyo Satrio B, Indriyati Hadi S

Pharmacy Department, Medical Faculty, Sultan Agung Islamic University Semarang
Jl. Kaligawe KM 4 Semarang 50012 Telp (+6224) 6583584 Fax (+6224) 6594366 Semarang

*corresponding author, email: fifiwulansafitri@gmail.com

ABSTRAK

Pendahuluan : Salah satu radikal bebas yang dapat merusak kulit wajah berasal dari radiasi sinar UV matahari. Radikal bebas merupakan faktor utama yang mempercepat proses penuaan dini. Pemberian antioksidan dipercaya dapat meningkatkan ketahanan tubuh khususnya mencegah penuaan kulit. **Tujuan:** Penetapan aktivitas antioksidan ekstrak rambut jagung dan formulasi sediaan krim.

Metode: Rambut jagung diekstraksi secara maserasi menggunakan etanol 80% selama 5 hari. Penetapan aktivitas antioksidan dilakukan dengan metode 2,2-diphenil 1-picrylhydrazyl (DPPH). Optimasi formula sediaan menggunakan *design expert* dengan memvariasikan konsentrasi tween 80 dan span 80. Sediaan krim dievaluasi meliputi evaluasi homogenitas, organoleptik, pH, daya lekat, daya sebar dan daya iritasi pada kulit.

Hasil: IC50 ekstrak rambut jagung diperoleh 30,03 ppm dengan nilai *AAI* (*antioxidant activity index*) 3,33 ppm. Dari hasil evaluasi dari 8 formula menunjukkan sediaan krim yang homogen, evaluasi organoleptis memiliki warna putih kecoklatan dan tidak berbau tengik, pH 6,5, daya lekat rentang 0,12-0,5 detik, daya sebar 5,1-7,3 cm, dan daya iritasi kulit yang menunjukkan hasil sedikit mengiritasi. Formula optimum sediaan krim mengandung ekstrak rambut jagung 0,0315%; propilen glikol 2,1%; tween 80 4,5%; span 80 5,5%; sorbitol 20%; asam stearat 5%; VCO 20%; asam sitrat 0,7%; asam askorbat 0,06%; Trietanolamin 2,45%; metil paraben 0,25%; propil paraben 0,15% dan aquadest add 100ml.

Kesimpulan: Ekstrak rambut jagung (*Zea mays L*) memiliki aktivitas antioksidan yang sangat kuat dan untuk formula krim yang optimum memiliki pebandingan antara emulgator tween 80 dan span 80 adalah, 4,5% banding 5,5%.

Kata Kunci : Rambut Jagung (*Zea mays L*), DPPH, *Design Expert*, IC50, *Antioxidant Activity Index*

ABSTRACT

Introduction : One of the free radicals that can damage facial skin is coming from UV radiation of the sun. Free radicals are the main factors that can accelerate the early aging process. The administration of antioxidants are believed to increase immune system of the body especially to prevent skin aging.

Objectives: to determine the antioxidant activity of the corn silk extract and cream formulation.

Methods: Corn silk were extracted by maceration using 80% ethanol for 5 days. Determination of antioxidant activity was conducted using 2,2-diphenil 1-picrylhydrazyl (DPPH). Optimization of the preparation formula using expert design by varying the concentration of tween 80 and span 80. The evaluation of cream preparation included homogeneity, organoleptic, pH, adhesion, dispersive power and power irritation to the skin.

Results: IC50 corn silk extract were obtained 30,03 ppm with *AAI*(*antioxidant activity index*) values of 3,33 ppm. From the results of evaluation of 8 formula indicates a homogenous cream preparation, evaluation of the organoleptic have a brownish-white color and does not become rancid, pH 6.5, adhesion range of 0.12 to 0.5 seconds, dispersive power from 5.1 to 7.3 cm and power irritation of the skin that show the results slightly irritating. Formula optimum dosage creams containing extracts of corn silk 0.0315%; propylene glycol 2.1%; 4.5% tween 80; Span 80 5.5%; sorbitol 20%; 5% stearic acid; VCO 20%; citric acid 0.7%; ascorbic acid 0.06%; Triethanolamine 2.45%; methyl paraben 0.25%; propyl paraben 0.15% and add distilled water 100ml.

Conclusion: Corn silk extract (*Zea mays L*) has a very strong antioxidant activity and for the optimum cream formulation has comparison between emulsifier tween 80 and span 80 is 4.5% to 5.5%.

Keywords : Corn silk (*Zea mays L*), DPPH, *Design Expert*, IC50, *Antioxidant Activity Index*

INTRODUCTION

One of the free radicals that can damage facial skin is the solar ultraviolet (UV) radiation. Solar UV radiation on living cells can cause varieties of risks such as chemical photograph, photo-isomerization, and photo-oxidation. Photo-oxidation reaction occurs due to the release (ROS) in the form, superoxide anion (O_2^-),

hydrogen peroxide (H_2O_2) and hydroxyl radicals (OH) by a chromophore that absorbs ultraviolet light (Gadri et.al, 2012). The reaction of the skin to UV radiation, among which are, the formation of free radicals (O_2^- and OH-), and cell death directly. Ultraviolet (UV-A and UV-B) can generate free radicals and cause damage to DNA, these free radicals are the main factors that

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$$\% \text{ inhibition} = \left(\frac{\text{control absorbance} - \text{sample absorbance}}{\text{control absorbance}} \right) 100\%$$

accelerate the aging process (Tjandrawinata, 2011).

Administration of antioxidants is believed to increase body endurance especially to prevent facial skin aging. Facial aging requires special treatment which can be started by prevention process. Awareness of the behavior of self protection against the negative impact of UV should be applied and emphasize. Primary prevention of negative impact of the UV includes the selection of clothes, hat and protective glasses and also application of sunscreen (both organic, anorganic; narrow or broad spectrum). Secondary prevention can use the application of topical retinoic acid (which also function in the treatment of photoaging condition when it occurs) and the use of both topical and systemic antioxidants (Tjandrawinata, 2011).

In a previous study, corn silk potentially used as an antioxidant because it is rich in bioactive compounds such as phenolic compounds, especially flavonoid. The compounds has conjugated tie to resonate when exposed to ultraviolet (UV) that is photoprotective (Prasiddha et.al., 2016). Application of corn silk directly to facial skin is unfavorable, therefore cream formulation of its extract, which is easy and convenient to apply, is needed. This study aims to create an effective antioxidant cream of corn silk extract.

METHODS

Kits: this research used rotary evaporation, spektrosimatzu UV-vis, vortex, and glass equipments
Materials: this research was using fresh corn silk from sweet corn plant around Gunung Pati, Semarang, Central Java area aged 80-90 days. Propylene materials pharmaceutical grade glycol tween 80 pharmaceutical grade, a pharmaceutical grade span 80, pharmaceutical grade sorbitol from, stearic acid pharmaceutical grade, pharmaceutical grade VCO, a pharmaceutical grade citric acid, ascorbic acid pharmaceutical grade, triethanolamine pharmaceutical grade, methyl paraben pharmaceutical grade, propyl paraben pharmaceutical grade were manufactured from PT. Barco, technical aquades of MKR, 1,1- Diphenyl-2-2- Picril Hydrazil (DPPH) p.a. from Surakarta Muhammadiyah University and technical ethanol pharmaceutical grade from PT. Barco.

1. Qualitative Test for Flavonoid Content

A total of 200 mg of corn silk extract reconstituted with 5 ml of ethanol and heated for five minutes in a test tube. Then added concentrated HCl, and 0.2 g of

powdered magnesium. A positive result is indicated by the onset of a yellow color.

2. Antioxidant Activity Test by DPPH Method

Antioxidant activity test by DPPH testing was conducted by weighing 10 mg of DPPH solution and then added 10 ml of 80% ethanol in a flask after it was taken 2.5 ml of ethanol was added 25 ml of 80% means a concentration of 80 ppm, the solution for comparison. Furthermore, the manufacture of the sample solution by means of weighing 0.01 g was added 10 ml of 80% ethanol in a flask, and then created a sample concentration of 5 ppm in a way taken 2 µl was added 5 ml 80% ethanol in a flask, 10 ppm taken 50 µl added 5 ml of 80% ethanol solvent in a flask, 15 ppm taken 75 µl added 5 ml 80% ethanol in a flask, 20 ppm taken 100 µl added 5 ml of ethanol 80% in the flask. Determination of the maximum wavelength that is by taking 2 ml of DPPH solution concentration of 80 ppm plus 2 ml of 80% ethanol and then incubated for 30 minutes. Further reading of the sample used in the lambda maximum of 80% ethanol is 515.5 nm, and then control the resulting absorbance wavelength is 0,629 nm. At a concentration of 5 ppm 0.616 nm, 10 nm 0,579 ppm, 15 ppm 0.539 nm, 20 nm 0.417 ppm. Next calculate inhibition percentage using the following formula on top.

3. The Making of Preparation Cream

Formulation and optimization cream using expert design software version 10 and obtained 8 formulas with different emulsifier concentrations. Preparation step of a cream: 1. water phase consisting of: propylene glycol, tween 80, sorbitol, citric acid, triethanolamin, ascorbic acid, methyl paraben, propyl paraben and distilled water; 2. Oil phase consisting of: namely stearic acid, VCO and span 80 heated at 70 ° C for five minutes or until evenly mixed. Each phase is mixed in different bowls, then the oil phase is placed in mortar and mixed with the water phase and stir continuously. Cream formulation evaluation included organoleptic test, adhesion, dispersive power, pH and power irritation of the skin (Widyaningrum *et al.*, 2015).

4. Evaluation Test of Preparations Cream

a. Organoleptic Appearance

Observing the appearance on preparation color change, phase separation or rupture of the emulsion, very strong smell of rancid, and how it feels on the

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skin.

b. Observation of Homogeneity

Observing particles sizes on the slide to find the coarse particles. As much as 0,25 gram cream were put on the slide for observation of coarse particles.

c. pH Observation

pH values were measured using pH paper. A total of 0.5 grams of the cream was dissolved in 5ml distilled water. Then observed using a pH indicator attached. The measurements were conducted three times for each formula.

d. Dispersion Test

Cream weighing 0.5 grams and is placed in the middle of a round glass scale. Another round glass which has been weighed placed there on and left for 5 minutes. After being awarded a load of 50 grams, let stand for 1 minute and record the diameter of the spreaded cream. Followed by delivering a load of 100 grams let stand for 1 minute and record the diameter of the deployment of the cream. This test is done 3 times for each formula.

e. Adhesion Test

A total of 0.5 grams of preparation were spread on the disc glass determined the area of(2 x 2 cm), on top of it other glass objects placed and pinned under a load of 1 kg for 1 minute. Then disc glass mounted on test equipment, weighing 1kg load is released and the time is recorded up to the second object of the glasses fallen off.

f. Skin Irritation Test

This study uses six albino male rabbits aged two months and a weight of 1.5 kg. Research irritation test using rabbit Draize test .The fur shaved on the back until clean. Shaving were done carefully so it cause no damage to the rabbit. Rabbit’s backs were divided into the nine sections of a square, in this trial the box smeared corn silk extract, one box smeared a cream base and a box smeared with cream that has been obtained from the optimum simplex lattice design software that creams containing extracts of corn silk 0.0315%; propylene glycol 2.1%; 4.5% tween 80; Span 80 5.5%; sorbitol 20%; 5% stearic acid; VCO 20%;citric acid 0.7%; ascorbic acid 0.06%; Triethanolamine 2.45%; methyl paraben 0.25%; propyl paraben 0.15% and add

distilled water 100ml. Evaluation was conducted for 24 hours to see the changes that occur as erythema where the skin becomes red and raised patches of skin spots. Next, for each state were rated as follows:

Table 1. Value Power of Draize Skin Irritation Test Method

Skin Conditions	Value
No erythema	0
very mild erythema	1
Mild Erythema	2
Moderate erythema	3
Severe erythema	4
No edema	0
Very mild edema	1
Mild Edema	2
Moderate edema	3
Severe Edema	4

Irritation index is calculated by adding up the value of the rabbits after 24 hours, the administration of irritant samples and then divided by 4. The assesments of irritiations are as follows:

- 0,00 = Non irritation
- 0,04 - 0,99 = Slightly irritation
- 1,00 - 2,99 = Mild irritation
- 3,00 - 5,99 = Moderate irritation
- 6,00-8,00 = Severe irritation

RESULTS

Optimal design obtained through the output program 10 design experts (trial version). In this study, a qualitative test results on corn silk extract provides yellow color, meaning the extract contained flavonoid. While antioxidant activity test’s result on hair extracts using the method (DPPH) 1,1- Diphenyl-2 -2- Picril Hydrazil (DPPH) for 5ppm, 10 ppm, 15 ppm, and 20 ppm inhibition concentration were 2.07%, 7.95%, 14.31%, and 33.70%. The results showed the value of the linear regression, which is to the value of A is -10.805; B is 2.025 and the R value is 0.951. From the results of linear regression can be concluded that the results is linier. The IC50 value of thick corn silk extract:

$$IC50 = bx + a$$

$$50 = 2, 025 x - 10, 805$$

$$x = 30,03 \text{ ppm (0,03 mg/ml)}$$

The value of AAI (antioxidant activity index) can be determined from IC50, with the formula:

$$AAI = \frac{\text{konsentrasi ppm DPPH}}{\text{nilai IC50}} = \frac{100}{30.03} = 3.33 \text{ ppm (sangat kuat)}$$

Emulsifier optimization, tween 80 and span 80, on cream formula using expert design method with simplex lattice design (SLD) and the obtained results were eight formula cream preparations with different emulsifier composition as listed on table 2 as follows:

Table 2. Composition of Emulsifier used for the preparation of Corn Silk Antioxidant Cream.

Formula	Tween 80	Span 80
1	1	0
2	0,25	0,75
3	0	1
4	1	0
5	0	1
6	0,75	0,25
7	0,5	0,5
8	0,5	0,5

Furthermore, eight formula of cream preparations were obtained as seen on table 3.

Evaluation Test of Cream Formula

a. Organoleptic Appearance

Observing appearance on preparation color change, phase separation or rupture of the emulsion, very strong smell of rancid, and how it feels on the skin. The result given that all formula has a creamy-

white color, phase separation did not occur, no rancid smell and felt damp when applied to the skin. Thus, all formula are in accordance with the appearance of cream on the market.

b. Homogeneity Observation

The observation of homogeneity indicated that all formulas did not contain particles or already homogen. This was as expected because it proved that the cream formula showed no separation phase between the oil phase and the water phase.

c. pH Observation

pH observation of all formula showed a constant pH value of 6.5. This proved that the difference concentration of emulsifier did not produce differences in the average value pH. The pH result of all formula illustrated in Table 4.

d. Dispersion Test

The dispersion test by adding and without adding weight have an average of more than 4cm to all creamy formula with each of three replicate for each formula. The average yield of the dispersion shown in Table 4.

f. Examination of Adhesion

Examination of adhesion all creamy formula

Table 3. Eight Formulas of Corn silk extract antioxidant cream

Components (gram)	F1	F2	F3	F4	F5	F6	F7	F8
Extract	0,0315	0,0315	0,0315	0,0315	0,0315	0,0315	0,0315	0,0315
Propilenglikol	2,1	2,1	2,1	2,1	2,1	2,1	2,1	2,1
Tween 80	9	2,5	1	9	1	7,5	5	5
Span 80	1	7,5	9	1	9	2,5	5	5
Sorbitol	20	20	20	20	20	20	20	20
Asam Stearat	5	5	5	5	5	5	5	5
VCO	20	20	20	20	20	20	20	20
Stearic Acid	0,7	0,7	0,7	0,7	0,7	0,7	0,7	0,7
Ascorbic Acid	0,06	0,06	0,06	0,06	0,06	0,06	0,06	0,06
Trietanolamine	2,45	2,45	2,45	2,45	2,45	2,45	2,45	2,45
Methyl paraben	0,25	0,25	0,25	0,25	0,25	0,25	0,25	0,25
Propil paraben	0,15	0,15	0,15	0,15	0,15	0,15	0,15	0,15
Aquadest	Add	Add	Add	Add	Add	Add	Add	Add
	100	100	100	100	100	100	100	100

Table 4. Test result of dispersive power, adhesion and the pH of the corn silk antioxidant cream extract formula

Formula	1	2	3	4	5	6	7	8	p
Mean of the dispersion (cm)	7.00	5,1	6,37	6,83	7,3	6,83	7,03	6,43	0.394253
Mean of Adhesion (cm)	0,44	0,12	0,38	0,51	0,42	0,38	0,41	0,40	5.58194
pH	6,5	6,5	6,5	6,5	6,5	6,5	6,5	6,5	6,5

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with each of three replicate showed an average yield of less than 1 second. Result of average adhesion shown in Table 4.

Optimum Formula Result

Evaluation of creams formula such as: adhesion test, dispersive power and pH respectively inputted results in Design Latice Design to get the optimum formula. The results of the optimum formula presented in Table 5:

Table 5. Optimum Formula of Corn Hair Antioxidant Cream Extract

Ingredients (gram)	Formula
Corn silk extract	0,0315
Propilenglikol	2,1
Tween 80	4,5
Span 80	5,5
Sorbitol	20
Stearic acid	5
VCO	20
Citric Acid	0,7
Ascorbic acid	0,06
Triethanolamine	2,45
Methyl paraben	0,25
Propyl paraben	0,15
Aquadest	Add 100

Values of dispersive power test, adhesion test and pH were verified by comparing with lattice design predictive values. Optimum data verification were illustrated in table 6.

The analysis result using One Sample t-test shows that between three responses which are adhesive power, dispersive power and pH all have trials values and predictive values did not significantly differ ($p > 0,05$) so the equation of optimization result is valid.

Skin Irritation Power test

The result of the skin irritation showed that the extract has a value of 0.5; a cream base have a value of 0.25 and cream optimum dosage has the result of

0.75. From the extract, the base and cream preparations according to the method of Draize test showed results included into a slightly irritating. This is proved by the onset of erythema or redness on testing animals. These results need to be re-evaluated.

DISCUSSION

Corn silk (*Zea mays L*) is one of the natural materials initially regarded as waste and the wider community use it as animal feed. Corn silk contain phenolic compounds, that are especially useful as an antioxidant flavonoids (Prasiddha, I., et al., 2016). For easier use, the corn silk is formulated as a cream preparation, because it has many advantages that active substances absorbed rapidly into the skin, easy to clean and easy to spread evenly over the surface of the skin (Harun, 2014) .Formulation of cream preparations using the oil type water (O / W).

The results of the optimum formula has the effect of slightly irritating to the skin it is because the solvent used in the extract using ethanol 80% and can be mitigated by a decrease in the concentration of the solvent and can be done fractionation so that the extract obtained absolutely pure extracts in the powder form (Widyaningrum, 2015).

This study has its limitations because it has not tested the inhibitory effect on UV. Require future research to prove the anti UV activity of corn silk extract. Furthermore, it is necessary for the test to determine the effectiveness of corn silk extract cream preparations as anti-aging and anti-UV.

CONCLUSION

Corn silk (*Zea mays L*) contains a powerful antioxidant proven by 3.33 ppm antioxidant activity index with IC50 value of 30.03 ppm.

Emulsifier optimization is done to get a formula that is compatible and can improve adhesion and the dispersion power. From expert design optimization using version 10 obtained optimum ratio tween span 80 to 80 compared to 4.5% compared to 5.5%.

Table 6. Data verification of Dispersive Power Test, Adhesive Power and pH of corn silk extract cream

No	Respon	Trial values	Predictive values
1	Dispersive Power no load	5.72375	5.58194
2	Dispersive power load 50 gr	6.22125	6.29351
3	Dispersive power load 100 gr	6.61125	6.6577
4	Adhesive power	0.3825	0.394253
5	pH	6.5	6.5

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