



Research article

Formulation of nano spray preparation from Purslane Leaf Extract (*Portulaca oleracea* L.) as sunscreen

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Received - 11-11-2023, Revised - 17-01-2024, Accepted - 28-01-2024 (DD-MM-YYYY)

Refer This Article

Diva Adela Sabrien, Ni'matun Nada Azzahra, Puspa Cinta Ratri Paramitha, Flora Katerrina Desta, Elmiawati Latifah, 2024. Formulation of nano spray preparation from purslane leaf extract (*portulaca oleracea* L.) as sunscreen. Journal of medical pharmaceutical and allied sciences, V 13 - I 1, Pages- 6303 – 6308. Doi: <https://doi.org/10.55522/jmpas.V13I1.5890>.

ABSTRACT

Purslane plant (*Portulaca oleracea* L.) is a type of wild plant that grows abundantly, but its benefits are rarely known. Purslane contains saponins, which have high antioxidant and biological activity. Skin that is sensitive to sun exposure is the issue this nanospray is targeting. The aim of this research is to determine the optimal formulation of herbal nanospray products with Purslane sunscreen to treat skin problems caused by sun exposure. The research method used was an experimental method with research stages starting from plant determination, extract making, nanospray formulation determination, preparation evaluation, in vivo UV protection activity test, and in vitro SPF test.

INNOVATION OF NANO SPRAY PREPARATION FROM PURSLANE LEAF EXTRACT (*PORTULACA OLERACEA* L.) AS SUNSCREEN



Organoleptic test

Formulas	Color	Form	Smell	Texture
F1	Bright dark green	Liquid	Like leaves (no sting)	Fine
F2	Dark green	Liquid	Like leaves (more pungent than F1)	Fine

Adhesion test

Replication	Observation Result		Standard
	F1	F2	
I	1.30	1.34	>1 second
II	1.52	1.40	
III	1.35	0.66	
Average	1.39	1.13	

Homogeneity test

Replication	Observation Result		Standard
	F1	F2	
I	Few particles	Homogeneous	Homogeneous (There isn't any particle)
II	Homogeneous	Homogeneous	
III	Homogeneous	Homogeneous	

PH test

Replication	Observation Result		Standard
	F1	F2	
I	5.44	5.46	4.5 - 7
II	5.44	5.50	
III	5.51	5.43	
Average	5.46	5.46	

Nanoparticle evaluation

	Size (nm)	PDI
F1	76.4	0.397
F2	73.5	0.379

SPF Values

Optimum Formulation

Sample			SPF Value	UV Protection Power Category
Portulaca	Nano	SpraySunscreen	9.9	Maximum
Purslane dose 0.8 grams				
Portulaca	Nano	SpraySunscreen	8.43	Extra
Purslane dose 0.4 grams				

Qualitative Test

- Tanin
- Alkaloid
- Steroid
- Flavonoid
- Saponin

Quantitative descriptive analysis was carried out using Microsoft Excel and SPSS Version 24. There were significant differences in erythema (lesions on the skin) between the 4 treatment groups, with significant differences in the negative control and positive control groups, as well as the group given dose treatment (the amount of extract in the nanoparticles) of 0.8 g and 0.4 g of purslane extract with the negative control. So, it can be concluded that doses of 0.8 g (SPF value: 9.9) and 0.4 g (SPF value: 8.43) of purslane extract have the potential to be sunscreens with maximum category and extra protective power. Purslane extract nanospray sunscreen has the advantage of being made from herbal ingredients, which have less potential for irritation than chemical sunscreen products.

Keywords: Nanospray, sunscreen, Purslane leaf extract, particle size, SPF Value.

INTRODUCTION

The Purslane plant (*Portulaca oleracea* L.) is a type of wild plant that grows abundantly and its benefits are rarely known. Many people still ignore the benefits of the Purslane plant. Based on research, Purslane contains KCl, K₂SO₄, KNO₃, nicotinic acid, saponin, vitamins A, B, and C, noradrenaline, l-noradrenaline, dopamine, and dopa which have high antioxidant biological activity.

As a tropical country, Indonesian people are often exposed to sunlight. Too much sun exposure can cause health problems, especially on the skin. With its antioxidant content, the Purslane plant is able to ward off free radicals caused by exposure to sunlight and has good potential for protecting the skin. Skin is one of the largest and outermost parts of the body that interacts directly with the environment. In everyday life, the skin continuously interacts with various products, substances, foreign materials, and surrounding objects. In current conditions, skin needs products that can ward off free radicals caused by pollution and sun exposure. Based on the data obtained, it was found that the ethanol extract and antioxidants of the Purslane herb had an SPF value of 19.5 [1]. Humans need sunlight to help form vitamin D, which is needed by bones, but excessive exposure to sunlight can have detrimental effects on human skin due to ultraviolet rays.

Nanotechnology is the most popular and interesting research field [2]. Nanoparticles are nanometer technology that converts water into atomic particles in a few seconds so that nutrients and oxygen levels in the water can enter the skin pores. Nanoparticles are defined as nano-scale particles ranging in size from 1 to 100 nm. Nanotechnology can be the recommendation chosen because it is a non-invasive model, so it can be attractive to the public because it can accelerate the onset of action of the target compound and is easily absorbed by the skin. The limitations of sunscreens currently available are mostly in the form of creams and gels, which are less practical to use, sticky, and take a long time to be absorbed. Nanoparticle technology has many advantages, including the smaller particle size causing the surface area to be larger in the same amount so that the affinity of the system increases. Based on the background that has been described, the aim of this research is to determine the optimum formulation of an herbal product nano spray with Purslane leaf extract to treat skin problems due to sun exposure.

MATERIAL AND METHODS

Plant Determination

The determination test was carried out to determine the correct identity of the plants used in the research. Plant determination was carried out at the Biology Laboratory, Faculty of Applied Science and Technology, Ahmad Dahlan University, with registration number 316/Lab.Bio/B/VI/2023

Purslane leaf extraction

The extraction process of Purslane leaves is carried out using the maceration method. Purslane plants were obtained in the Mungkid of Magelang. Extraction of Purslane leaves (*Portulaca oleracea* L.) begins with drying the Purslane leaves in an oven at a temperature of 57°C for 1x24 hours. In a maceration vessel, Purslane leaf simplicia is soaked in 96% ethanol at a ratio of 1:10 for three days, while being stirred every so often. The liquid is then filtered out, and the leaves are dried in a water bath until a thick extract of Purslane leaves (*Portulaca oleracea* L.) is made. The yield obtained is weighed and recorded [3]:

$$\text{Yield (\%)} = \frac{\text{Extract weight}}{\text{Dried simplicia weight}} \times 100\%$$

Qualitative Phytochemical Screening Test

The sample used in phytochemical screening was an ethanol extract of Purslane leaves. Phytochemical screening is carried out for color tests using various reagents to identify alkaloids, tannins, flavonoids, saponins, steroids, and terpenoids [4].

a) Alkaloid Test

A solution of 2 mL of Purslane leaf ethanol extract was put into a beaker, and Mayer's reagent was added. If a white precipitate forms, then the sample contains alkaloids.

b) Tannin Test

The ethanol extract solution of Purslane leaves was heated in 10 mL of distilled water for 3 minutes, then cooled and filtered. The filtrate is dripped with 1% gelatin solution; if a white precipitate forms, it shows that the ethanol extract of Purslane leaves contains tannin.

c) Flavonoid Test

A total of 2 mL of Purslane leaf ethanol extract solution was added to hot water, then boiled for 5 minutes. After that, it was filtered, and the filtrate was added with a little magnesium powder and 1 mL of concentrated HCl, then shaken. The test result is positive if the color changes to red, yellow, or orange.

d) Saponin Test

A solution of ethanol extract of Purslane leaves added to hot water was placed in a test tube and waited until it cooled. Then shake vigorously for 10 minutes. Positive test results are indicated by the presence of stable foam or foam as high as 1 cm for \pm 10 minutes.

e) Terpenoid Steroid Test

A total of 2 mL of ethanol extract solution, was added the reagent, namely 2 mL of chloroform, 10 drops of acetic anhydride, and 3 drops of concentrated H₂SO₄. A positive reaction to steroids is indicated by the formation of a green solution and a red color is formed if it is positive for terpenoids.

Making nano spray gel preparations from Purslane leaf extract

Making nano spray gel preparations from purslane leaf extract uses the formulation shown in Table 1.

Table 1: Formulation ^[5]

F1		F2	
Karbopol 940	0.13 g	0.13 g	Gel base
Trietanolamin	0.067 g	0.067 g	Gel base neutralizer
Ad aquadest	25 ml	25 ml	Solvent
Nanoemulsion Formula			
Leaf extract Purslane	0.8 g	0.4 g	Active substance
Propil paraben	0.003 g	0.003 g	Preservative
Metil paraben	0.0167 g	0.0167 g	Preservative
Sorbitol	0.67 g	0.67 g	Surfactant
Tween 80	1 g	1 g	Surfactant
Ad aquadest	25 ml	25 ml	Solvent

a) Making spray gel base preparations from Purslane leaf extract

Carbopol 940 (powder) is added with hot distilled water, then crushed until homogeneous and forms a gel base. Then add little by little TEA (liquid) as a gel base neutralizer.

b) Making nano emulsions

Purslane leaf extract is added with sorbitol (liquid) until mixed or dissolved. Next, heat methylparaben (powder) and propylparaben (powder) plus distilled water in a water bath until dissolved. Then cool it, place it in a mortar, add Tween 80 little by little, and add distilled water until it doesn't form a white color. Then add the extract mixture with sorbitol to the mixture above little by little until homogeneous. Then the preparation was mixed at 6000 rpm using turrax for 5 hours until homogeneous and clear.

c) Making nano-spray gel preparations

To make spray gel nanoparticle preparations, the gel base is added to the nanoemulsion preparation, then stirred again until homogeneous.

Evaluate preparations**a) Organoleptic and homogeneity tests**

The organoleptic test was carried out by looking at the physical appearance of the nanospray preparation in the Purslane nanospray product for sunscreen. Homogeneity is checked using a visual method, namely spraying the preparation on a glass preparation,

then leveling it by attaching another glass preparation and observing. Observations were made by looking at the presence or absence of particles that had not been mixed homogeneously ^[6].

b) Test pH

The pH value of the preparation is measured using a pH meter dipped in the nano spray preparation, left until the pH value that appears on the screen stabilizes. It is noted that each formula must meet the skin pH range requirements, namely 4.57 ^[7].

c) Test adhesion

The adhesion test was carried out by placing the nano spray preparation in the PNS (Portulaca Nano Spray) product on top of a glass object, then covering it with another glass object and applying a load of 0.5 kg for 5 minutes. The load is released, thereby pulling the bottom glass object. The time required for the two glass objects to separate is recorded as the sticking force.

d) Particle size test

The procedure for determining the particle size in nano spray is to first dilute the nano spray preparation with 1 ml of distilled water. Then take 1 ml and put it into the cuvette, then put the cuvette in the sample holder on the particle size analyzer. The tool will measure the sample within 15 minutes.

In Vivo Protective UV Activity Test

In vivo data was collected, and the anti-inflammatory properties of the compound were observed as measured by a score of 0-4 for skin areas that responded to erythema. The diameter of the erythema was calculated using a caliper. The erythema score used is: Score 0 = no erythema;

Score 1 = very slight erythema (diameter \leq 25.00 mm);

Score 2 = obvious erythema (diameter 25.10 - 30.00 mm);

Score 3 = moderate to severe erythema (diameter 30.10 - 35.00 mm);

Score 4 = crust forming (diameter \geq 35 mm).

The results obtained were then analyzed using a quantitative descriptive analysis method to determine whether the nano spray preparation of Purslane leaf extract met the standard requirements for a good preparation by processing the data using Microsoft Excel and SPSS Version 24.

In-Vitro Test to Determine Sun Protection Factor (SPF)

Determining the effectiveness of sunscreen is done by determining the SPF value in vitro with a UV-Vis spectrophotometer. The Purslane leaf extract to be used is diluted and divided into several concentrations, namely 100 ppm, 150 ppm, and 200 ppm. Read using a UV-Vis spectrophotometer with a wavelength between 290-320 nm every 5 nm interval, the blank used is distilled water. Then the absorbance results are calculated to calculate the SPF value ^[8]:

$$320$$

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

$$290$$

CF: Correlation Factor

EE: Erythermic Efficiencyation

I: Solar ray simulation spectrum

Abs: Read absorption value

RESULTS AND DISCUSSION

Plant determination

The Purslane leaves used were subjected to a determination test first at the Biology Laboratory, Faculty of Applied Science and Technology, Ahmad Dahlan University (SK. Number: 316/Lab. Bio/B/VI/2023, with results showing that the simplicia used was indeed a plant *Portulaca oleracea* L.

1b – 2b – 3b – 4b – 12b – 13b – 14b – 16a (g01 10 : Single leaf opposite)
239b – 243b – 244b – 248b – 249b – 250b – 266b – 267a – 268b –
271a – 272b *Portulacaceae*

1a *Portulaca*

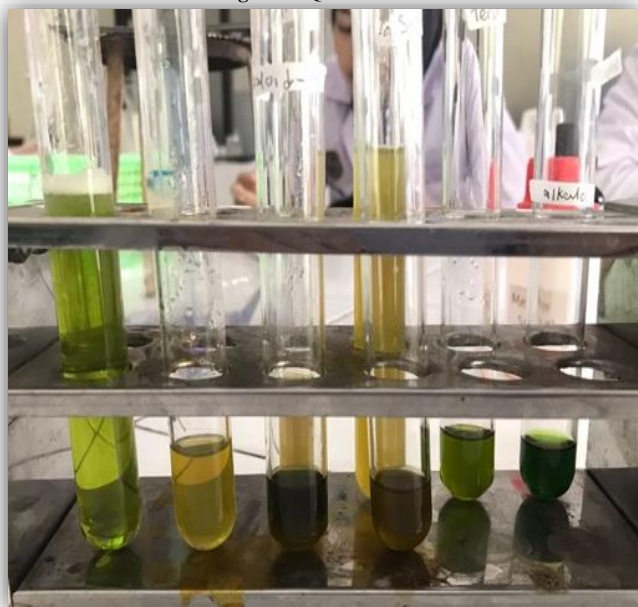
1a *Portulaca oleracea* L.

Purslane leaf extraction

The maceration extraction begins with a 1x24 hour drying process to optimize the drying of the simplicia. Drying aims to reduce the water content of a simplicia. Drying can also reduce microbial activity and minimize physical and chemical changes while dry materials are stored [9]. Extraction was carried out by maceration for 3 days, where 60 grams of Purslane leaf simplicia were soaked with 600 ml of 97% ethanol in a maceration vessel, then soaked for 3 days with occasional stirring. After that the liquid was removed by filtration, then evaporated over a water bath until the extract was obtained. Thick Purslane leaves (*Portulaca oleracea* L.) after weighing the thick extract was 1.64 grams, with a yield of 2.74%.

Qualitative Test (Phytochemical Screening)

Figure 1: Qualitative Test



The results of the phytochemical screening of the ethanol extract of Purslane leaves are shown in Table 2. The results were positive for tannins, alkaloids, steroids, flavonoids, saponins, and negative for terpenoids. Flavonoid

Table 2: Results of phytochemical screening tests

Test	Result	Observation
Tanin	+	White precipitate
Alkaloid	+	White precipitate
Steroid	+	Green color
Flavonoid	+	Yellowish green color
Saponin	+	Froth/foam
Terpenoid	-	There is no color change to red

Inventory Evaluation

a) Organoleptic test

Organoleptic tests are tests carried out by naked eye or observation directly to describe the preparation. Organoleptic tests include form or consistency, color and odor of the resulting preparation (Table 3).

Table 3: Organoleptic test

Formulas	Color	Form	Smell	Texture
F1	Bright dark green	Liquid	Like leaves (no sting)	Fine
F2	Dark green	Liquid	Like leaves (more pungent than F1)	Fine

b) Homogeneity test

This homogeneity test is carried out with the aim of determining homogeneity by looking at the uniformity of the particles in the preparation (Table 4). Based on the homogeneity test, the second dosage formulation has homogeneity that is good and meets the requirements of the Indonesian Pharmacopoeia Edition III, namely that if the preparation is applied topically to a piece of glass or other suitable transparent material, must show the correct arrangement of homogeneity which can be seen by the absence of particles that are clustered and spread out randomly equally.

Table 4: Homogeneity test

Replication	Observation Result		Standard
	F1	F2	
I	Few particles	Homogeneous	Homogeneous (There isn't any particle)
II	Homogeneous	Homogeneous	
III	Homogeneous	Homogeneous	

c) pH Test

The pH of the gel preparation was measured using the Universal pH Indicator. The pH of a preparation that meets the skin pH criteria is in the range 4.5 – 6.5 [12]. Based on the PH data produced, the pH of the extract is in accordance with existing standards.

Table 5: pH Test

Replication	Observation Result		Standard
	F1	F2	
I	5.44	5.46	4.5 -7
II	5.44	5.50	
III	5.51	5.43	
Average	5.46	5.46	

The results of this phytochemical screening were compared with previous research on Purslane plants using ethanol solvent, showing that the ethanol extract of Purslane leaves contains flavonoids. Flavonoids are phenolic compounds and phenolic compounds are a

secondary metabolite that acts as an antioxidant ^[10]. Thus, the flavonoids in the ethanol extract of Purslane leaves have the potential to act as antioxidants.

d) Adhesion Test

The adhesion test is carried out with the aim of finding out how long it lasts attachment of the preparation to the surface of the skin so that the active substance is absorbed preparations. The longer it stays attached to the skin, the more effects it will have, and the more it spreads over the surface of the skin. There are special requirements regarding adhesion, but the adhesion should be semi-solid for more than 1 second ^[13]. The average adhesion strength of the two formulations complies with existing standards (Table 6).

Table 6: Test adhesion

Observation Result			
Replication	F1	F2	Standard
I	13.0	1.34	More than 1 second
II	1.52	1.40	
III	1.35	0.66	
Average	1.39	1.13	

e) Particle size test

Sample testing includes particle size, poly-dispersity index (PDI) value. The results of particle size and PDI values are presented in Table 7.

Table 7: Nanoparticle evaluation results

	Size (nm)	PDI
Formulation 1	76.4	0.397
Formulation 2	73.5	0.379

Referring to Table 7, the sample meets the nanoparticle size requirements where the size is between 10-1,000 nm. A PDI value of 0.3 indicates a narrower and more uniform particle size distribution. Size of the particles and the PDI of nano-delivery systems are the main physical factors that affect how well active ingredients pass through cell membranes ^[14]

The *In-Vivo* UV Protective Activity Test

The research protocol has met ethical requirements and received approval from the ethical commission before conducting experiments on animals. The ethical clearance registration number for this research is, No. 036/EC-HC-KEPK FKIK UMY/VII/2023. This test was carried out to see the sunscreen capabilities of the Portulaca Nano Spray preparation using Wistar rats as experimental animals. In this testing process, 3 stages were carried out, namely, dividing the mice into 4 groups that were adapted for 7 days, the first day of treatment (shaving the mice's back hair, spraying the preparation, and exposing them to UV B lamps for 3x24 hours), and the second day (observing the results by measuring the diameter of the erythema). The results obtained were the appearance of erythema in the negative control group (Figure 2) with diameters of 3 mm, 5 mm and 15 mm (score 1). Meanwhile, erythema did not form in the other three treatment groups. These results prove that Portulaca Nano Spray has activity as a sunscreen.

Figure 2: The control erythema results were negative



Kruskal Wallis SPSS Analysis Data

Kruskal-Wallis is a non-parametric statistical test that can be used to test whether there are significant differences between groups of independent variables and the dependent variable ^[15]. Based on Table 8, there are significant differences in erythema between the positive control group and the negative control group, the 0.8 g dose control group and the 0.4 g dose with the negative control group, so that the Purslane leaf extract nanoparticle formulation has Sunscreen potential like the positive control group. This is relevant to previous research where the highest SPF value of 7.862 was found in Purslane leaf ethanol extract cream with 1% extract ^[16].

In Vitro Test to Determine Sun Protection Factor (SPF)

Determining the SPF value in this research is a parameter in determining the potential of sunscreen which is carried out in vitro using a UV-VIS spectrophotometer. The SPF calculation results from the Portulaca Nano SpraySunscreen preparation containing 0.8 grams and 0.4 grams of Purslane leaf extract can be seen in Table 9.

The purpose of using sunscreen is based on its ability to absorb, reflect or scatter sunlight. SPF measurement is the main way to determine the effectiveness of sunscreen formulations. The higher the SPF value, the better the sunscreen's protection against UV rays ^[17].

The SPF value obtained by this preparation in nano spray

form was higher compared to previous research on Purslane extract in

cream form ^[17].

Table 9: SPF values

Sample	SPF Value	UV Protection Power Category
Portulaca Nano SpraySunscreen Purslane dose 0.8 grams	9.9	Maximum
Portulaca Nano SpraySunscreen Purslane dose 0.4 grams	8.43	Extra

CONCLUSION

There were significant differences in erythema between the 4 treatment groups, with significant differences in the negative control and positive control groups, and the group given a dose of 0.8 g and 0.4 g of purslane extract with a negative control so that it can be concluded that a dose of 0.8 g and 0.4 g of purslane extract has the potential as a sunscreen with maximum and extra protective power categories.

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