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1149

EVALUATION AND ANTIOXIDANT TEST OF SAFFRON FACE MIST AS A SKIN MOISTURIZER

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ABSTRACT

Saffron (Crocus sativus L.) is a medicinal plant used as a spice in food, but recent times have shown that has a role in cosmetics. The saffron stigma has antioxidant activity and is useful in cosmetics. Antioxidants can protect the skin from various types of cell damage due to free radicals from ultraviolet light, motor vehicle fumes, chemicals in food, chemicals, and drugs. The antioxidant content of saffron stigma makes it an ingredient in cosmetics such as face mist. Face mist is a cosmetic skin freshener that refreshes facial skin, removes residual oil from the skin, kills some skin microorganisms, and helps close the pores. The aim of this study was to evaluate the antioxidant properties of the saffron face mist. The evaluation of saffron face mist includes an organoleptic test based on odor, color, and shape/texture, a pH test using a universal pH indicator paper, a specific gravity test using a pycnometer, and a moisture test using a skin moisture meter. The saffron stigma antioxidant test was performed using the DPPH assay. The results showed that the face mist had a rose aroma, golden yellow color, and moist form, the specific gravity ranged from 1.0637 to 1.0689 g/mL, the pH was 6, the moisture value of the facial skin after the application of the face mist to the face ranged from $47.77 \pm 3.81 - 56.52 \pm 0.72$ %. The IC₅₀ value of saffron stigma was 248.82 ppm. The physical quality of saffron face mist follows the pH of the skin and can moisturize the skin. The antioxidant activity of the saffron stigma was weak.

Keywords: antioxidant, saffron, face mist

INTRODUCTION

Saffron (Crocus sativus L.) is a medicinal plant predominantly cultivated in Iran, with other significant producers including countries like India, Greece, and Italy. Its stigma contains several important compounds such as crocin, crocetin, picrocrocin, and safranal. Crocetin, a carotenoid compound, contributes to the vibrant yellow hue of saffron, while picrocrocin imparts its distinctive taste, and safranal gives saffron its characteristic aroma (Ferrara, Naviglio, and Gallo, 2014; Dwitiyanti, Astuti and Hayati, 2022). Traditionally, saffron has been mainly utilized as a culinary spice, but recent studies have highlighted its potential role in the pharmaceutical industry, particularly in the field of cosmetics. Research has shown that the saffron stigma possesses antioxidant properties that are highly valuable in cosmetic formulations (Mzabri, Addi, and Berrichi, 2019). Antioxidants are compounds that help to prevent oxidation reactions caused by free radicals, which are highly reactive atoms or molecules capable of damaging healthy cells in the body (Ramadhan, 2020). These antioxidants act as protective agents for the skin, shielding it from various types of cellular damage triggered by free radicals that come from ultraviolet radiation, vehicle exhaust, chemicals in food, and other substances, including pharmaceuticals. The presence of free radicals in the skin can lead to health and cosmetic concerns. Skin problems such as melasma, dryness, and even an increased long-term risk of skin cancer have been linked to these reactive molecules (Sari, 2015).

In Indonesia, the incidence of skin cancer - one of the many diseases attributed to free radicals - remains notably high, with approximately 10 million cases recorded annually, ranking it as the third most common cancer (Dampati and Veronica, 2020). Using antioxidant-rich cosmetics is one of the most effective strategies to combat skin damage caused by free radicals (Jani, Hakim, and Juliantoni, 2020). Antioxidant cosmetics can be classified into three types: endogenous, exogenous, and plant-based. Plant-based antioxidants are particularly popular in skincare because they are natural, have fewer side effects, and are considered safer (Haerani, Chaerunisa, and Subarnas, 2018). The antioxidant compounds found in saffron stigmas make saffron a valuable ingredient in the formulation of cosmetic products such as sunscreens, face mists, whitening creams, facial masks, and others (Arsitowati, 2017).

Face mist is a cosmetic product designed to refresh the skin and is primarily used to hydrate the face, remove excess oil, eliminate certain skin microorganisms, and help close the pores. Classified as a lotion, face mist can come in various forms such as solutions, suspensions, or emulsions intended for topical application (Apristasari et al., 2018). Incorporating natural ingredients, such as saffron, into face mist formulations has become increasingly popular. Over the past year, the saffron face mist has gained significant traction as a lucrative business venture. Market analysis of a popular marketplace revealed that nearly hundred different saffron face mist products were available last year alone; however, only about 12 of these products were officially registered with BPOM. The popularity of saffron face mist is largely attributed to its antioxidant content and moisturizing properties. In a tropical country like Indonesia, which experiences continuous sun exposure year-round, skin issues like dryness are quite prevalent (Wadoe et al., 2020). Dry skin ranks among the top ten most common dermatological conditions in Indonesia, affecting approximately 50-80% of the population (Sinulingga, 2017). Research focusing on adolescents aged 15-17 years has shown that 20.4% of them suffer from dry skin, and 52.63% of those with dry skin also experience acne vulgari (Tamba, 2019). These findings underscore the importance of face mist products that serve as effective moisturizers, offering a viable solution to combat dry skin issues (Kamilah, 2021). This study aimed to assess and analyze the antioxidant properties of the saffron face mist.

RESEARCH METHODS

Equipment and Materials

The equipment used in this study includes a B-One RE-2010 rotary evaporator, Faithful DK-2000-IIIL water bath with 2 holes, Universal indicator from Suncare brand, vortex mixer Oregon, Ohaus CP 214 analytical balance, Cheyi_N Skin Moisture Meter Analyzer, and B-One UV-Vis 100DA spectrophotometer. All glassware used was the branded Pyrex. The materials used in this study included high-quality super negin saffron stigma from Iran, obtained from an online marketplace, with Société Générale de Surveillance (SGS) certificate number CG19-021661.001, ethanol, methylparaben, propylene glycol, rose-scented perfume, PEG-40, 2,2-Diphenyl-1-Picrylhydrazyl (DPPH) from Sigma Aldrich, and methanol. All chemicals used were of proanalytical grade.

Research Procedure

- 1. Saffron Extraction
 - The extraction of saffron stigmas is performed using the maceration method, which involves immersing saffron in a solvent to allow its components to be drawn out.
- 2. Preparation of Saffron Face Mist
 - The resulting saffron infusion was then combined with distilled water, followed by gradual incorporation of methylparaben, propylene glycol, PEG-40, and a rose-scented fragrance until the mixture reached a final volume of 100 ml. The blend was then transferred to a spray bottle. In this study, three distinct formulas were developed, each with varying amounts of saffron. Formula 1 (F1) contains 20 mg of saffron extract,

Formula 2 (F2) includes 30 mg, and Formula 3 (F3) contains 40 mg (Chowdhury, Ray and Sengupta, 2020).

3. Evaluation of Saffron Face Mist

The assessment of the physical and chemical properties of saffron face mist includes organoleptic evaluation, pH measurement, specific gravity determination, and moisture analysis. The detailed procedure for each test is as follows:

a. Organoleptic Evaluation

The organoleptic test serves as a sensory analysis to observe the fragrance, color, and texture of the product. During this evaluation, the facial mist formulation was assessed by examining its scent, appearance, and consistency. This test is conducted by untrained panelists, allowing for an unbiased review of the product's sensory characteristics (Puspita, Puspasari, and Restanti, 2020).

b. pH Measurement

The pH of the face mist was determined using universal pH indicator paper. This process involved immersing the indicator paper in the solution to be tested. After a brief period, the paper is retrieved and compared against a color reference chart to ascertain the pH value (Handayani, Warnida, and Nur, 2016).

c. Specific Gravity Determination

Specific gravity tests were performed using a pycnometer. This involves measuring the weight of the empty pycnometer, then the pycnometer filled with distilled water, and finally the pycnometer filled with the sample solution. The specific gravity was calculated using Equation (1).

$$Specific\ gravity = \frac{weight\ of\ pycnometer\ +\ weight\ of\ pycnometer\ with\ distilled\ water}{weight\ of\ pycnometer\ +\ weight\ of\ pycnometer\ filled\ with\ sample}\dots\dots\dots(1)$$

d. Moisture Test

Facial moisture levels were assessed using a Skin Moisture analyzer (Cheyi_N Skin). Measurements were taken before applying the face mist, and then again at intervals of 1 minute, 1 hour, 2 hours, and 8 hours after the application of the face mist. These values are recorded and compared to evaluate the difference in skin moisture levels before and after using the face mist (Apristasari *et al.*, 2018).

4. Antioxidant Activity Testing of Saffron

The antioxidant activity of saffron was tested by weighing 50 mg of saffron stigmas and dissolving them in 10 ml of methanol p.a., followed by stirring, until a homogeneous solution was obtained. The mixture was left to stand for 30 minutes in a dark room. Test solutions with concentrations of 20, 40, 60, and 80 ppm were prepared by pipetting 2, 4, 6, and 8 ml of each filtrate and 100 ppm methanol saffron macerate precipitate solution. Each amount was then transferred into a 10 ml volumetric flask, and methanol p.a. was added up to the mark. The solutions were thoroughly mixed until they were homogeneous. A standard stock solution of DPPH at a concentration of 100 ppm was prepared by weighing 10 mg of DPPH powder, dissolving it in methanol p.a., and filling a 100 ml volumetric flask up to the mark. The solution was shaken until homogeneous. To determine the maximum wavelength of the DPPH standard solution, 4 ml of a 40 ppm DPPH solution was pipetted into a cuvette, and its absorption spectrum was observed at wavelengths ranging from 400 to 800 nm using a Vis spectrophotometer. A blank solution containing 4 ml of methanol p.a. is used as the reference. The maximum wavelength was obtained from the absorption curve.

The absorbance of DPPH was measured by pipetting 2 ml of a 40 ppm DPPH solution into a test tube, adding 2 ml of methanol p.a., vortexing, and allowing it to stand for 30 minutes before transferring it into a cuvette. The absorbance was measured at the maximum wavelength using a UV-Vis spectrophotometer. To evaluate the free radical scavenging activity, 2 ml of 40 ppm DPPH solution was mixed with 2 ml of each test

solution at various concentrations (20, 40, 60, 80, and 100 ppm), vortexed, and left to stand for 30 minutes. The absorbance was measured sequentially for each concentration at the maximum wavelength using a UV-Vis spectrophotometer, and the absorbance values were recorded. The percentage of radical-scavenging activity was calculated using Equation 2.

$$\% attenuation = \frac{DPPH \ absorbance - sample \ absorbance}{DPPH \ absorbance \ test} x \ 100 \ \dots \dots \dots (2)$$

Based on the scavenging percentages at each concentration, a regression curve was constructed, resulting in the equation: y = bx + a. The IC₅₀ value, which represents the concentration required to inhibit 50% of the free radicals, was determined using linear regression analysis, where the sample concentration (ppm) was plotted on the x-axis and the scavenging percentage on the y-axis. The IC₅₀ value is derived from the calculation of a 50% scavenging activity (Suena and Antari, 2020).

RESULTS AND DISCUSSION Evaluation of Saffron Face Mist

In this study, three formulations of saffron face mist were developed, each containing varying amounts of saffron extract. Formula 1 (F1) contains 2% saffron extract of the total formulation, Formula 2 (F2) includes 3% saffron extract, and Formula 3 (F3) has 4% saffron extract. These percentages were chosen based on a study by Apristasari et al. (2018), which utilized a 3% extract concentration across all face mist formulations. The organoleptic evaluation considered aspects such as color, scent, and texture, performed on finished face mist products packaged in spray bottles, with observations made by untrained panelists. The results of these evaluations are summarized in Table I. As shown in Table I, the fragrance of all three face mist formulations is characterized by a rose scent, which is attributed to the rose-scented perfume used in their preparation. The intensity of the rose aroma remained consistent across all formulations, as the same amount of rose extract was used for each formulation. Saffron itself has a distinctive aroma derived from its safranal content, which is a cyclic terpenic aldehyde produced from picrocrocin. The amount of safranal in saffron can be influenced by processing conditions, with temperatures above 80°C and durations of less than 30 minutes leading to higher safranal production (Rezaee and Hosseinzadeh, 2013). In this face mist production, the blending process was conducted at room temperature, which likely limited the formation of safranal, resulting in a less pronounced saffron scent in the final product. Additionally, the relatively low saffron concentration of 2-4% in the formulations may have further contributed to the subtle aroma.



Figure 1. Saffron Face Mist in Spray Bottles. Formulations: a. F1, b. F2, and c. F3.

All three formulas (F1, F2, and F3) displayed a golden-yellow color, with the highest intensity observed in formula 3 (F3), which had the highest saffron concentration. This color

is derived from saffron's active compound, crocin, which is a water-soluble carotenoid responsible for the rich yellow-red hue of saffron. Crocin (monoglycosyl or di-glycosyl polyera) is a key component in saffron, known for its high glycosyl content and significant coloring properties (Afifah and Hasanah, 2020). The golden hue of face mist formulations is enhanced by the clarity of the base ingredients, allowing crocin to effectively impart its characteristic color. The texture of the face mist in all three formulas is fluid, which is expected given that most ingredients in the formulations are liquids. The appearance of the saffron face mist is shown in **Figure 1**.

The specific gravity test aims to determine the viscosity and purity of the substance, and was carried out using a pycnometer. Factors influencing specific gravity include temperature, particle size, viscosity, and molecular weight of the substance (Ilyas, 2018) According to **Table I**, the specific gravity of the saffron face mist across all formulations ranges from 1.0637 to 1.0689 g/mL. These results align with those reported by Herliningsih and Anggraini (2021), indicating that the specific gravity of face mist exceeds that of water, and as the concentration of the extract increases, so does the specific gravity.

Table I. Results of Saffron Face Mist Evaluation

Parameter Test	Formula				
Farameter Test	F1 F2		F3		
Organoleptic					
Color	Golden yellow (+)	Golden yellow (++)	Golden yellow (++)		
Odor	Rose	Rose	Rose		
Phase	Thin liquid	Thin liquid	Thin liquid		
Spesific Grafity	1.0637	1.0668	1.0689		
pН					
Week 0	6 ± 0.00	6 ± 0.00	6 ± 0.00		
Week 1	6 ± 0.00	6 ± 0.00	6 ± 0.00		
Week 2	6 ± 0.00	6 ± 0.00	6 ± 0.00		
Week 3	6 ± 0.00	6 ± 0.00	6 ± 0.00		
Moisture					
Before applied	47.64 ± 1.51	47.42 ± 5.99	47.46 ± 4.65		
1 minute	48.7 ± 0.63	48.5 ± 5.37	51.86 ± 2.54		
1 hour	56.52 ± 0.72	50.4 ± 3.34	52.12 ± 2.02		
2 hour	55.88 ± 0.54	49.68 ± 3.67	50.9 ± 4.05		
8 hour	52 ± 2.57	47.77 ± 3.81	48.76 ± 1.57		

The pH test results revealed that all three formulas—Formula 1 (F1), Formula 2 (F2), and Formula 3 (F3)—had a pH of 6. This value indicates that the face mist formulations have a suitable pH for facial skin, as the normal skin pH range is between 4.5 and 6.5 (Herliningsih and Anggraini, 2021). The purpose of testing the pH of the formulation is to determine whether it might cause skin irritation; a pH below 4.5 or above 6.5, can lead to skin issues such as redness and inflammation. The pH of a formulation can be influenced by its ingredients, including both active substances and excipients. The pH of 6 likely results from the presence of ethanol and propylene glycol, as their combination tends to produce a higher pH than either substance used alone (Qisti, Nurahmanto, and Rosyidi, 2018). The pH is an indicator of the stability of the formulation. In this observation, pH was monitored from week 0 to week 4 (Sayuti, 2015), and the results showed no significant changes, indicating that the saffron face mist was stable over the four-week storage period. Stability can also be influenced by the presence of humectants in the formulation (Tsabitah *et al.*, 2020). In comparison, this formulation used propylene glycol as a humectant, which likely contributed to the stability of face mist.

The saffron face mist offers benefits, such as maintaining and enhancing skin moisture. The results showed that the skin moisture levels after applying the face mist ranged from $47.77 \pm$

3.81 to 56.52 ± 0.72 %. These values indicated that the skin moisture level remained within the normal range, which considered normal skin moisture levels to be between 35-60%. The moisturizing effect of the formulation was primarily due to its humectant ingredients. Humectants, which are common in cosmetic products, attract and retain the moisture in the skin. Common humectants used in moisturizing formulations include glycerin, propylene glycol, and hyaluronic acid, all of which help draw water into the skin (Butarbutar and Chaerunisaa, 2020). The concentration of propylene glycol in the formulation was based on development studies by (Chowdhury, Ray, and Sengupta, 2020). During the moisture test, face mist was sprayed on the faces of five consenting participants who volunteered for the study. None of the participants reported any signs of skin inflammation, such as erythema or edema, following application. Ingredients that commonly pose a risk of skin irritation include preservatives, antioxidants, ultraviolet (UV) protectants, fragrances, and colorants. However, the ingredients used in face mist were within safe limits, minimizing the risk of irritation. Moreover, with a pH of 6, which falls within the range of 4 to 8, the face mist formulation also helps reduce the potential for skin irritation (Savitri, 2018).

Table II. Hasil Uji Aktivitas Antioksidan Putik Bunga Saffron dan Quercetin

Sample	Concentration (ppm)	Inhibition (%)	Regression Equation	IC ₅₀	Antioxidant Category*
Saffron	20	6,90		248,82±4,4	Weak
	40	7,52	v- 0.2022v		
Stigma	60	8,78	y=0,2022x +0,094		
Extract	80	14,11	+0,094		
	100	23,82			
Quercetin (Standart)	20	50,47		19,25±9,5	Strong
	40	53,92	v=0.6201		
	60	87,15	y=0,6301 +37,868		
	80	92,79	+37,000		
	100	94,04			

^{*(}Fauziah, Sudirga and Parwanayoni, 2021)

Antioxidant Result

The antioxidant activity test was conducted using the DPPH method, a quantitative technique that measures antioxidant activity by evaluating the ability of an antioxidant compound to scavenge DPPH free radicals, utilizing a visible spectrophotometer at a wavelength of 517 nm (Novatama, Kusumo, and Supartono, 2016). The antioxidant activity was expressed as the IC₅₀ value. The results of the antioxidant tests for the saffron stigma are presented in **Table II**. Based on the data from this table, it was found that the antioxidant activity of saffron fell into the weak category. Saffron contains antioxidant compounds derived from phenolic compounds, carotenoids, crocetin, and crocin (Zakiyah *et al.*, 2021). The low antioxidant activity of an extract may be influenced by the presence of secondary metabolites with antioxidant properties and the fact that the extract has not been purified (Ismail and Ningtyas, 2020). In this study, a phytochemical screening process was not performed; therefore, the specific metabolites present in the saffron used were not identified. Moreover, since the extract was not purified, its antioxidant activity was categorized as weak.

CONCLUSION

The physical quality of the saffron face mist formulation is compatible with the pH of the skin and it can moisturize the skin. However, the antioxidant activity of saffron in face mist formulation was weak.

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