

DEVELOPMENT AND PHYSICOCHEMICAL EVALUATION OF ANTIOXIDANT PEEL-OFF MASKS CONTAINING ETHANOL EXTRACT OF CURRY LEAVES**Tri Susanti Sirait^{1*}, Yunda Fachrunniza², Desni Rinanda Silitonga³, Sry Ulina Karo-Karo⁴, Yulia Safitri Limbong⁴**¹Public Health Study Program, Universitas Teuku Umar, Meulaboh, Indonesia²Pharmacy Study Program, Universitas Syiah Kuala, Banda Aceh, Indonesia³Pharmacy Study Program, Universitas Sari Mutiara Indonesia, Medan, Indonesia⁴Pharmacy Study Program, Institut Kesehatan Helvetia, Medan, Indonesia*Email: trisantisirait@utu.ac.id**Article History:**

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DOI: <https://doi.org/10.22373/8fb0n491>**ABSTRACT**

Curry leaves (*Bergera koenigii*) are widely recognized for their rich phytochemical content and antioxidant properties; however, their application as active ingredients in cosmetic delivery systems remains relatively underexplored. This study aimed to evaluate the phytochemical profile and antioxidant activity of the ethanol extract of *B. koenigii* leaves and to investigate its potential incorporation into a peel-off mask formulation through physicochemical characterization. Antioxidant activity was assessed using the DPPH (2,2-diphenyl-1-picrylhydrazyl) assay, while formulation quality was evaluated through organoleptic properties, homogeneity, pH, viscosity, spreadability, drying time, stability testing for 28 days, cycling test, and skin irritation assessment. Phytochemical screening confirmed the presence of tannins, flavonoids, steroids, and saponins. The ethanol extract exhibited very strong antioxidant activity with an IC_{50} value of 8.23 $\mu\text{g/mL}$, indicating a high free radical scavenging capacity. The formulated peel-off masks demonstrated stable physicochemical properties during storage, with viscosity values of 6,740–13,729 cPs, spreadability of 5.3–6.6 cm, drying time of 14.03–16.36 minutes, and pH values within the acceptable skin-compatible range (6.40–6.63). No irritation responses were observed in the preliminary skin test. These findings highlight the potential of *B. koenigii* ethanol extract as a promising natural antioxidant ingredient for peel-off mask cosmetic formulations.

Keywords: antioxidant, *Bergera koenigii*, peel-off mask**INTRODUCTION**

The skin is the outermost tissue that covers and protects the entire surface of the human body and is continuously exposed to various environmental factors, including air pollution and solar radiation. Such exposures can trigger the formation of reactive compounds that disrupt

the biological balance of the skin and accelerate cellular damage (Dunaway et al., 2018; Ryšavá et al., 2021). In the long term, this condition not only leads to a decline in skin function but also triggers various aesthetic problems, including dull skin, premature aging, and reduced skin elasticity (Ramakrishnan et al., 2020; Singh et al., 2020). Therefore, topical antioxidant-based skin protection has become an increasingly important strategy, not only for cosmetic purposes but also as an effort to maintain long-term skin health (Burke, 2024).

In recent years, attention to natural antioxidants has continued to increase, along with growing concerns about the potential side effects of synthetic compounds (Oderinde et al., 2023). Tropical plants are considered a promising source owing to their high diversity of secondary metabolites (Samal et al., 2024). Curry leaves (*Berbera koenigii*) are rich in natural polyphenols, phenolic compounds, and flavonoids, which contribute to their strong antioxidant properties. A previous study reported that the total phenolic content reached 77.00 μg GAE/mg, while the flavonoid content was recorded at 21.02 μg RU/mg (Bhatt et al., 2022). Another study reported a total phenolic content of 2.591 mg/g, along with a total flavonoid content of 5.248 mg/g (Phatak et al., 2018). These compounds play an essential role in neutralizing free radicals and inhibiting lipid peroxidation, thereby protecting cells from damage induced by oxidative stress (Parashar et al., 2020).

In the field of cosmetic science, various plant-derived antioxidants have been incorporated into topical formulations such as creams, gels, and peel-off masks to enhance skin protection against oxidative damage. Recent studies have demonstrated that plant extracts rich in phenolic compounds can improve the antioxidant capacity and functional properties of cosmetic formulations when applied topically (Vo et al., 2019). In particular, peel-off mask formulations containing plant-based antioxidants have attracted increasing attention because they provide a convenient delivery system that allows active compounds to remain in contact with the skin for a longer period while simultaneously removing impurities from the skin surface. Several studies have reported that peel-off masks formulated with botanical extracts exhibit promising antioxidant activity and favorable physicochemical characteristics for topical use.

In the extraction of plant-derived bioactive compounds, solvent selection is a critical factor influencing extraction efficiency, compound selectivity, and biological activity. Various organic solvents such as methanol, ethanol, acetone, and aqueous mixtures have been widely used to extract phenolic and flavonoid compounds from medicinal plants. Polar solvents generally demonstrate higher efficiency in extracting antioxidant-related phytochemicals

because many phenolic compounds exhibit polar or semi-polar characteristics. Previous studies have shown that ethanol is particularly advantageous because it effectively extracts polyphenols and flavonoids while presenting lower toxicity and better safety for pharmaceutical, food, and cosmetic applications compared with other organic solvents (Bhuyan & Handique, 2022; Shen et al., 2022). Consequently, ethanol is widely considered an appropriate solvent for obtaining antioxidant-rich extracts intended for topical cosmetic formulations.

The antioxidant activity of curry leaves may help reduce oxidative stress, which is one of the primary factors contributing to skin aging. By neutralizing free radicals, curry leaves have the potential to prevent premature aging, wrinkles, and other signs of skin damage (Agrawal et al., 2024). However, its utilization remains largely confined to the food industry and traditional medicine. The limited number of studies linking the biological activity of curry leaves to cosmetic applications indicates a research gap that warrants further investigation, particularly in the development of natural active ingredients for skin care. Recent studies have increasingly explored plant-derived antioxidants in topical cosmetic formulations, including peel-off masks and gel-based delivery systems, demonstrating promising antioxidant activity, good physicochemical stability, and favorable skin compatibility (Nazar et al., 2025; Ukhty et al., 2021; Winingrum & Zai, 2024). Despite these advances, the use of *Bergera koenigii* extract as an active ingredient in peel-off mask formulations has not been widely investigated. This limited evidence indicates that the potential of curry leaf extract in cosmetic delivery systems remains insufficiently explored, particularly in relation to formulation stability and physicochemical performance.

Although several studies have reported the antioxidant potential of *Bergera koenigii* extracts, most investigations focus on pharmacological or nutritional applications rather than their incorporation into cosmetic delivery systems. In addition, studies examining the formulation of curry leaf extract in topical cosmetic products, particularly peel-off mask systems, remain very limited. This lack of formulation-based research highlights a clear gap between the known antioxidant potential of curry leaves and their practical application in cosmetic products designed for skin protection against oxidative stress.

Peel-off masks possess physical properties and functional characteristics that are well suited for skin application. The gel system allows active compounds to be distributed homogeneously, while the peel-off mechanism provides a mechanical cleansing effect that may enhance user comfort and satisfaction. In addition, the formation of a film layer on the skin surface has the potential to increase the contact time of active ingredients, thereby supporting

the effectiveness of antioxidant activity. The incorporation of ethanol extract of curry leaves into a peel-off mask is expected not only to function as a cosmetic product but also to contribute more optimally to skin protection against oxidative stress (Ukhty et al., 2021; Winingrum & Zai, 2024).

Based on the above considerations, this study aims to evaluate the phytochemical profile and antioxidant activity of ethanol extract obtained from curry leaves and to develop a peel-off mask formulation containing the extract. Furthermore, this research seeks to investigate the physicochemical characteristics, stability, and preliminary skin compatibility of the formulated peel-off mask. By integrating phytochemical evaluation with cosmetic formulation development, this study addresses the limited evidence regarding the application of *Bergera koenigii* extract in peel-off mask systems and provides scientific insight into its potential as a natural antioxidant ingredient for topical cosmetic products.

METHODS

Materials

All materials used in this study were of analytical grade. Ethanol 96% (Merck), was used as the extraction solvent. 2,2-diphenyl-1-picrylhydrazyl (DPPH) (Sigma-Aldrich, USA) was used for antioxidant activity analysis. Distilled water, FeCl₃ (Merck, Germany), ethanol (Merck, Germany), concentrated HCl (Merck, Germany), Mg powder (Merck, Germany), H₂SO₄ (Merck, Germany), Mayer's reagent (Merck, Germany), Wagner's reagent (Merck, Germany), Dragendorff's reagent (Merck, Germany), were used for phytochemical screening. Polyvinyl alcohol (PVA, Merck, Germany), hydroxypropyl methylcellulose (HPMC, Sigma-Aldrich, USA), propylene glycol (Merck, Germany), methyl paraben (Merck, Germany), propyl paraben (Merck, Germany), and distilled water (Brataco Chemical, Indonesia).

Equipments

The equipment used in this study included standard laboratory glassware (Pyrex) mortar and pestle, spatula, pestle rod, spatula scoop, digital analytical balance (Ohaus) aluminum foil, formulation tubes, plastic containers, evaporating dish, gloves and masks for laboratory safety, filter paper (Whatman), vacuum rotary evaporator (Buchi Rotavapor R-3), micropipettes (Dragonlab), hot plate (Nesco), oven (B-One), incubator (Memmert), UV-Vis spectrophotometer (Shimadzu).

Preparation and Extraction of Curry Leaves

Curry leaves were collected using a purposive sampling technique based on predefined criteria, namely fresh, intact leaves with a dark green color. The samples were obtained from Meulaboh, West Aceh, and were subjected to botanical identification at the National Research and Innovation Agency (BRIN), as confirmed by an official identification certificate (No. B-4534/II.6.2/DI.05.07/12/2022). The sampling location was specifically carried out in Gunong Kleng, Meureubo, West Aceh, Indonesia (Latitude : 4.1313022, Longitude : 96.1808959). The coordinates were determined using Google Maps to ensure accurate documentation of the plant sampling site. The curry leaves were washed and dried using a drying cabinet at a temperature range of 40–60 °C until completely dried. After drying, the samples were sorted then ground into a fine powder.

The extraction process was carried out using the maceration technique with 96% ethanol as the solvent. The powdered plant material was placed into a suitable container, and 75 parts of ethanol were added. The container was tightly sealed and stored for five days under light-protected conditions with periodic stirring. Subsequently, the residual marc was remacerated using 2.5 parts of ethanol for two days. Ethanol was selected as the extraction solvent because of its intermediate polarity, which allows it to effectively dissolve a wide range of bioactive compounds, including phenolic and flavonoid constituents known to possess antioxidant activity. In addition, ethanol is widely used in phytochemical extraction due to its relatively low toxicity, good extraction efficiency for antioxidant compounds, and compatibility with cosmetic and pharmaceutical applications. The combined filtrates were then concentrated using a rotary evaporator at 40–50°C, followed by further evaporation in a water bath until a thick extract was obtained. The steps of curry leaf extraction can be seen in Figure 1.

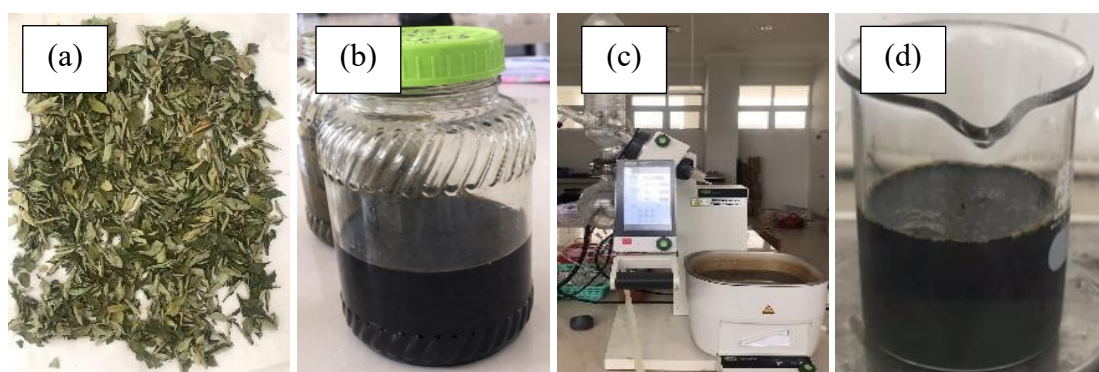


Figure 1. Steps of Curry Leaf Extraction Process: (a) Drying of simplicia, (b) Extraction, (c) Extract concentration, (d) Concentrated extract

Phytochemical Screening

Alkaloid screening was performed by heating 50 mg of the ethanol extract of curry leaves in a water bath for two minutes, followed by cooling and filtration. The resulting filtrate was then transferred into three separate test tubes, with three drops added to each tube. Subsequently, two drops of Mayer, Bouchardat, and Dragendorff reagents were added to the respective test tubes. The presence of alkaloids was identified by the formation of a precipitate. A white or yellow precipitate was observed with Mayer reagent, while dark brown and yellowish-orange precipitates were formed with Bouchardat and Dragendorff reagents, respectively (Aristyawan et al., 2024)

Flavonoid screening was carried out by placing 50 mg of the ethanol extract of curry leaves into a beaker, followed by heating and filtration. Subsequently, 100 mg of magnesium powder was added to the filtrate, and 1 mL of 2 N hydrochloric acid (HCl) was added dropwise. A positive result for flavonoids was indicated by a color change from yellow to red (Aristyawan et al., 2024).

Saponin screening was performed by mixing 50 mg of the ethanol extract of curry leaves with 10 mL of hot water. The mixture was allowed to cool host, then vigorously shaken for 10 seconds. The formation of stable foam indicated the presence of saponins when the foam remained stable for at least 10 minutes and reached a height of 1–10 cm. The foam was considered stable if it persisted after the addition of 2 N hydrochloric acid (HCl) (Sudarwati & Ariastuti, 2024).

Tannin screening was conducted by dissolving 50 mg of the ethanol extract of curry leaves in 20 mL of distilled water, followed by heating. After filtration, several drops of a 10% ferric chloride (FeCl₃) solution were added, and the resulting color change was observed. The formation of a blue or greenish-black coloration signified the presence of tannins (Sudarwati & Ariastuti, 2024).

Steroid and triterpenoid screening was performed by adding 2 mL of chloroform to 50 mg of the ethanol extract of curry leaves, followed by the addition of 0.5 mL of acetic anhydride. Subsequently, 2 mL of concentrated sulfuric acid (H₂SO₄) was slowly added dropwise along the inner wall of the test tube, and the resulting color changes were monitored. The presence of steroids was indicated by the formation of a bluish-green color, reflecting the presence of a steroid ring, whereas the appearance of a brown or purple a ring formed at the boundary between the two solvents indicated a positive result for triterpenoids (Sudarwati & Ariastuti, 2024).

Antioxidant Activity Assay

Preparation of 0.4 mM DPPH Stock Solution

The DPPH stock solution was prepared by accurately weighing 15.8 mg of DPPH powder in ethanol in a 100 mL volumetric flask. After complete homogenization, the solution was stored in a dark, airtight container to prevent light exposure (Sirait et al., 2023).

Preparation of 0.1 mM DPPH Working Solution

A 0.1 mM DPPH solution was obtained by diluting 25 mL of the 0.4 mM stock solution to a final volume of 100 mL in a volumetric flask. The solution was subsequently brought to the calibration mark using ethanol and mixed thoroughly to ensure homogeneity.

Preparation of Sample Solutions

An amount of 2.5 mg of the sample was dissolved in ethanol and brought to the final volume of 50 mL to obtain a 50 ppm stock solution, which was subsequently diluted to concentrations of 10, 15, 20, 25, and 30 ppm.

Determination of the Maximum Wavelength of the DPPH Solution

The absorbance of the 0.1 mM DPPH working solution was measured using a UV–Vis spectrophotometer at wavelengths ranging from 400 to 600 nm, with ethanol used as the blank solution. The wavelength exhibiting the highest absorbance was determined as the maximum wavelength (λ_{max}).

Preparation of Vitamin C Standard Solution

An amount of 4 mg of vitamin C was dissolved in ethanol until completely homogenized, transferred into a 100 mL volumetric flask and diluted to the calibration mark using ethanol to obtain a 40 ppm vitamin C stock solution. The stock solution was then diluted to produce a series of concentrations of 2, 4, 6, 8, and 10 ppm.

Antioxidant Activity Testing of the Sample and Vitamin C

A total of 2 mL of the 0.1 mM DPPH the solution was measured using a pipette into a test tube, followed by the addition of 2 mL of the sample at various concentrations. The mixture was then incubated for 30 minutes under light-protected conditions. Absorbance was measured at the predetermined maximum wavelength using a UV–Vis spectrophotometer, with ethanol serving as the blank. Antioxidant activity was reported as the percentage of inhibition (% inhibition), and the extract was subsequently compared with vitamin C as a reference standard.

All measurements were performed in triplicate to ensure the reliability of the results. The obtained data were expressed as mean \pm standard deviation (SD). The percentage of DPPH radical scavenging activity was calculated using the following equation, where A_0 represents the absorbance of the control (DPPH solution without sample) and A_s represents the absorbance of the sample solution.

$$\% \text{ Inhibition} = \frac{(A_0 - A_s)}{A_0} \times 100$$

Determination of IC₅₀ Value

The IC₅₀ (Inhibitory Concentration 50%) value was calculated using a linear regression model ($y = ax + b$) derived from the calibration curve representing the relationship between sample concentration (x) and the percentage of antioxidant activity (y). The IC₅₀ value was determined by substituting $y = 50$ into the regression equation.

The regression analysis was performed using the relationship between the natural logarithm (\ln) of sample concentration and the percentage of inhibition to obtain a linear regression equation. The goodness-of-fit of the regression model was evaluated using the coefficient of determination (R^2). A higher R^2 value indicates a better fit between the experimental data and the regression model. The IC₅₀ value was subsequently calculated from the regression equation corresponding to 50% inhibition. Statistical analysis and regression calculations were performed using Microsoft Excel software.

Formulation of Peel-Off Masks

This formulation was developed with reference to previous studies (Pradiningsih & Mahida, 2019). However, adjustments were made pertinent to the active compound and the concentration of polyvinyl alcohol (PVA). The materials used in this formulation are listed in Table 1. The formulation process of the peel-off mask is detailed in Table 1, beginning with weighing each ingredient. Polyvinyl alcohol (PVA) and hydroxypropyl methylcellulose (HPMC) were separately dispersed and allowed to swell in hot distilled water (80°C) using a mortar and pestle to ensure proper hydration of the polymer components. After both PVA and HPMC had fully swelled, they were combined and stirred until a homogeneous mixture was obtained (Mass I). The curry leaf ethanol extract was dissolved in propylene glycol in a beaker to form Mass II, ensuring complete dissolution of the extract. Methyl paraben and propyl paraben were first triturated in a mortar to obtain a fine and uniform powder before being incorporated into Mass II, and the mixture was stirred until a homogeneous preservative

solution was formed (Mass III). Subsequently, Mass I (polymer base) was gradually incorporated into Mass II under continuous stirring to produce a uniform gel base. Finally, Mass III was added to the mixture, followed by the gradual addition of the remaining distilled water while continuously mixing until a homogeneous peel-off mask formulation was obtained. The final formulation was stirred until a uniform gel consistency was achieved to ensure even distribution of the active extract and excipients within the formulation matrix. The formulation steps of the ethanol extract peel-off mask are illustrated in Figure 2.

Table 1. Peel-Off Mask Formulation of Curry Leaf Ethanol Extract

Chemicals	Formula (%)			Function
	F1	F2	F3	
Ethanol Extract of Curry Leaves	0	1	2	Active ingredient
PVA	12	12	12	Film-forming agent
HPMC	2	2	2	Viscosity enhancer
Propylene glycol	15	15	15	Humectant
Methyl paraben	0.05	0.05	0.05	Preservative
Propyl paraben	0.05	0.05	0.05	Preservative
Aquadest	Ad 100	Ad 100	Ad 100	Solvent

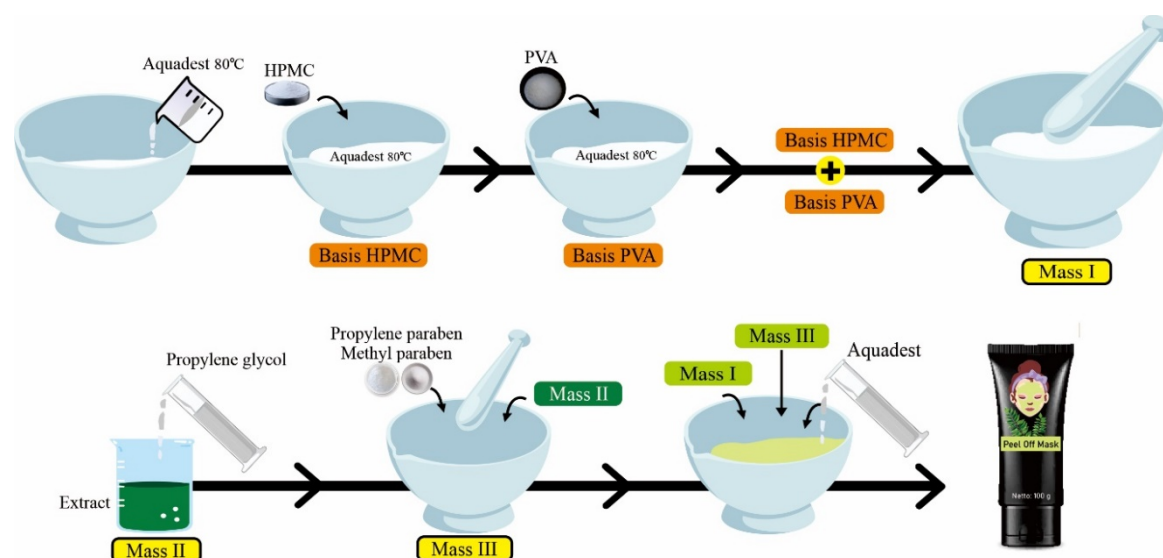


Figure 2. Formulation Steps of Peel-Off Mask Containing Ethanol Extract of Curry Leaves

Physicochemical Evaluation

Organoleptic evaluation of the peel-off mask formulation was carried out by assessing its physical characteristics, including color, viscosity, and odor, both before storage and after a specified storage period. Homogeneity testing was performed by applying 0.1 g of the

formulation onto a transparent glass surface, followed by visual observation to determine the presence or absence of unevenly mixed particles (Nurhikma et al., 2023).

The acidity level of the formulation was determined using a pH meter. Before measurement, the instrument was calibrated using standard buffer solutions, namely a neutral buffer (pH 7.01) and an acidic buffer (pH 4.01), until stable and accurate readings were achieved. The electrode was then rinsed with distilled water and carefully dried using tissue paper. Subsequently, the electrode was placed into the sample until a stable pH reading was obtained. pH measurements were conducted at several time intervals, namely on days 0, 1, 7, 14, 21, and 28 (Nurhikma et al., 2023).

Viscosity testing was performed by placing the formulation into a 100 mL beaker. The appropriate spindle was selected based on the characteristics of the formulation, and viscosity measurements were carried out using a Brookfield viscometer (DV-E type). The viscosity of the formulation was determined at room temperature on days 0, 1, 7, 14, 21, and 28 (Nurhikma et al., 2023).

Spreadability testing was performed by weighing 0.5 g of the peel-off mask formulation and placing it at the center of a graduated circular glass plate. Another circular glass plate or a similar transparent material, along with an additional weight to achieve a total load of 150 g, was placed on top of the gel. The formulation was allowed to stand for 1 minute before recording the resulting spread diameter. Spreadability measurements were conducted on days 0, 1, 7, 14, 21, and 28 (Nurhikma et al., 2023).

Drying time testing was carried out by applying 1 g of the peel-off mask formulation onto the skin surface of the arm over an application area of 7 cm × 7 cm. The drying time required for the formation of a film layer was measured using a stopwatch, with an acceptable drying time ranging from 15 to 30 minutes. Drying time measurements were conducted on days 0, 1, 7, 14, 21, and 28 (Nurhikma et al., 2023).

Cycling test evaluation was conducted by storing the samples at a temperature of 4 ± 2 °C for 24 hours, followed by placement in an oven at 40 ± 2 °C for an additional 24 hours. This sequence of treatments was defined as one cycle. The test was carried out for six cycles, during which the samples were observed for any signs of phase separation. The physical condition of the formulation after the cycling test was then compared with its initial condition prior to treatment (Nurhikma et al., 2023).

Irritation testing was conducted by applying a certain amount of the peel-off mask formulation to the dorsal area of the hands of 30 different respondents. The formulation was

left in place for at least 15 minutes, after which the skin was observed for any visible signs of irritation, as indicated by the presence of edema and erythema (Ade et al., 2021).

RESULTS AND DISCUSSION

Extraction

The extraction process was conducted using the maceration method, followed by a concentration step employing a rotary evaporator to obtain an optimal extract. The rotary evaporator functions to remove the solvent from the solution, thereby yielding the desired concentrated extract. This process is achieved by lowering the boiling point of the solvent, allowing evaporation to occur at a relatively low temperature (Shen et al., 2022). A total of 500.44 g of curry leaf simplicia was used as the initial material for extraction, which produced 90.78 g of extract with a percentage yield of 18.15%. In comparison, a previous study that performed curry leaf extraction using ethanol reported a yield of 2.83% (Diana & Ukhty, 2018). These results indicate that the extraction conditions applied in the present study were able to produce a higher extraction yield, suggesting a more effective extraction process in obtaining bioactive compounds from curry leaves.

Phytochemical Screening

Phytochemical screening was performed to identify the presence of various secondary metabolites in the plant extract, including alkaloids, flavonoids, saponins, tannins, phenolic compounds, and glycosides. These compounds are widely recognized for their pharmacological activities, particularly their anti-inflammatory effects, antioxidant, antimicrobial, and anticancer activities (Syafi et al., 2023). The phytochemical screening results of the curry leaf extract are presented in Table 2. Based on these findings, flavonoid compounds were identified in the extract. Flavonoids are known to play a role in neutralizing free radicals in the body through an electron transfer mechanism to free radical compounds (Sirait et al., 2023). This mechanism stabilizes the electrons of free radicals, thereby reducing their likelihood of reacting in ways that damage healthy cells. In addition, the extract was found to contain tannins, saponins, and triterpenoids. The phytochemical screening results of the curry leaf extract are presented in Table 2.

Table 2. Phytochemical Screening Results of Curry Leaf Extract

Secondary Metabolites	Reagents	Observations	Result
Alkaloid	Bouchardat	No precipitate formed	-
	Dragendorff	No precipitate formed	-
	Mayer	No precipitate formed	-
Tannin	FeCl ₃ 10% + HCl 2 N	Green-colored solution	+
Flavonoid	Mg Powder + HCl 2 N	Orange-colored solution	+
Saponin	H ₂ O + HCl 2 N	Foam formation of 2 cm	+
Steroid	CH ₃ COOH	Green-colored solution	+
Triterpenoid	H ₂ SO ₄ 98%	Green-colored solution	-

The presence of these phytochemical constituents is closely associated with the antioxidant activity of the extract. Polyphenolic compounds such as flavonoids and tannins are widely reported as major contributors to antioxidant activity because of their ability to donate hydrogen atoms or electrons to neutralize reactive oxygen species (ROS). These compounds can terminate radical chain reactions and protect biomolecules from oxidative damage. Therefore, the detection of flavonoids and tannins in the ethanol extract of curry leaves provides a plausible explanation for the strong antioxidant activity observed in the DPPH assay.

Alkaloid testing was considered positive when a precipitate formed after the addition of Mayer, Dragendorff, and Bouchardat reagents. Prior to testing, the extract was treated with hydrochloric acid (HCl) to facilitate alkaloid extraction due to the basic nature of alkaloids. The formation of a white precipitate upon reaction with Mayer reagent indicated the presence of alkaloid compounds, resulting from the formation of an insoluble potassium–alkaloid complex (Cheng et al., 2024). A similar mechanism occurs in tests using Dragendorff and Bouchardat reagents. The use of Dragendorff reagent typically produces an orange–reddish precipitate, while Bouchardat reagent results in a brown precipitate. However, in this study, the extract did not show a positive result for alkaloid content.

Saponin testing was indicated by the presence of stable foam with a height of 1–10 cm after the addition of 2 N HCl. The saponin test of the ethanol extract of curry leaves showed a positive response, as evidenced by the formation of foam with a height of approximately 2 cm. Saponins are amphiphilic compounds composed of both hydrophilic and hydrophobic moieties. When the solution is shaken, the Hydrophilic groups exhibit affinity toward water, while

hydrophobic groups preferentially interact with air, resulting in foam formation. In micelle structures, the polar groups are oriented outward, while the nonpolar groups are oriented inward, thereby stabilizing the foam (Barbosa, 2014; Böttcher & Drusch, 2017). The addition of HCl to the saponin solution can lead to a reduction in surface tension and an increase in surface hydrophobicity, which are important factors for improved foam stability (Chen et al., 2023). In addition to their chemical properties, saponins may also contribute to the functional characteristics of cosmetic formulations. Their amphiphilic structure allows them to act as natural surfactants that improve the dispersion of active compounds and enhance the stability and texture of gel-based formulations such as peel-off masks (Chen et al., 2019; Morais et al., 2025). In addition to their chemical properties, saponins may also contribute to the functional characteristics of cosmetic formulations. Their amphiphilic structure allows them to act as natural surfactants that improve the dispersion of active compounds and enhance the stability and texture of gel-based formulations such as peel-off masks (X.-W. Chen et al., 2019; Morais et al., 2025)

Tannin testing was carried out by adding FeCl_3 reagent. The addition of FeCl_3 caused the solution to change to a dark green color. This colour change occurred as a result of complex formation between tannins and Fe^{3+} ions. The test results indicated that the ethanol extract of curry leaves showed a positive result for the presence of tannin compounds (Khattab et al., 2018). Tannins are known to exhibit strong antioxidant properties due to their multiple hydroxyl groups, which enable them to donate hydrogen atoms and scavenge free radicals effectively. In cosmetic formulations, tannins also possess astringent properties that may contribute to skin tightening effects and help reduce excess sebum on the skin surface (De Souza Schaumlöffel et al., 2021; Ditthawutthikul et al., 2021)

Flavonoid testing was conducted by the addition of magnesium powder and hydrochloric acid, which induced the reduction of the benzopyran nucleus in the flavonoid structure. This reaction resulted in a color change from yellow to reddish orange, indicating the presence of flavonoids (Sun et al., 2020). The test results showed that the ethanol extract of curry leaves tested positive for flavonoids, as evidenced by a color change to orange. Flavonoids are among the most important natural antioxidants because they can donate hydrogen atoms and stabilize free radicals through resonance structures. In topical cosmetic applications, flavonoids are also known to protect skin cells from oxidative stress caused by ultraviolet radiation and environmental pollutants, making them valuable active ingredients in antioxidant-based skincare products (Czaplińska et al., 2012; Ma & Khachemoune, 2023)

Steroid/triterpenoid testing was performed using the Liebermann–Burchard reagent, which is a mixture of acetic anhydride and concentrated sulfuric acid. The test results showed that the ethanol extract of curry leaves contained steroid compounds, as indicated by the appearance of a green color. The reaction involved was an esterification reaction, in which concentrated sulfuric acid acted as a catalyst (Campos, 2011) and acetate anhydride functions as a carboxylic acid derivative that reacts with the hydroxyl groups of steroid/triterpenoid compounds.

Although steroids and triterpenoids are not primarily responsible for antioxidant activity, these compounds may contribute to additional biological effects such as anti-inflammatory and skin-protective properties, which are beneficial in cosmetic formulations designed to improve skin health (Yoo et al., 2025).

Overall, the phytochemical composition identified in this study supports the antioxidant activity observed in the DPPH assay, where the ethanol extract of curry leaves exhibited a very strong antioxidant capacity with an IC_{50} value of 8.23 $\mu\text{g/mL}$. The presence of polyphenolic compounds, particularly flavonoids and tannins, likely plays a dominant role in this antioxidant performance and highlights the potential of curry leaf extract as a natural active ingredient for antioxidant-based cosmetic formulations.

Antioxidant Activity

The antioxidant activity assay using the DPPH method was performed to evaluate the antioxidant capacity of the ethanol extract of curry leaves. DPPH is a free radical molecule that significantly contributes to oxidative damage in living organisms (Chandimali et al., 2025). Free radicals are unstable molecules due to the presence of unpaired electrons, making them highly reactive and capable of inducing oxidative damage to lipids, proteins, and DNA. This instability plays an important role in cellular aging processes and the development of various degenerative diseases (Ahmad et al., 2024). Antioxidants act by donating an electron to free radicals, allowing the previously unpaired electron to become paired and restoring molecular stability. An illustration of this mechanism is shown in Figure 3

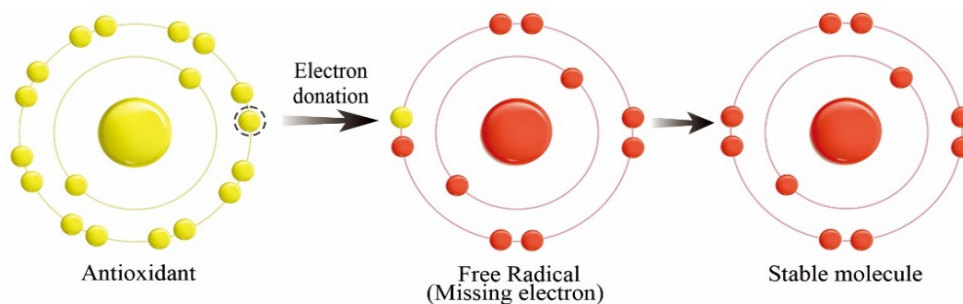


Figure 3. Mechanism of Antioxidant Action in Neutralizing Free Radicals

In line with the electron donation mechanism, the findings of this study indicate that the interaction between antioxidants and free radicals is reflected in the changes in radical characteristics observed in the DPPH assay. The solution, which was initially purple, turned into a stable yellow color after reacting with antioxidants. The results of the antioxidant activity evaluation of the ethanol extract of curry leaves are presented in Table 3.

Table 3. Antioxidant Activity Test Data Of The Ethanol Extract Of Curry Leaves and Vitamin C

Sample	Concentration	Absorbance	% Inhibition	IC_{50} value
Ethanol Extract of Curry Leaves	10	0.447	52.95	8.232
	15	0.391	58.77	
	20	0.357	62.35	
	25	0.327	65.56	
	30	0.292	69.21	
Vitamin C	2	0.529	45.92	3.247
	4	0.476	51.40	
	6	0.434	55.66	
	8	0.413	57.79	
	10	0.392	59.96	

The linear regression equation for antioxidant activity was determined based on the relationship between the natural logarithm (\ln) of concentration and the percentage of inhibition or antioxidant activity. Subsequently, the IC_{50} value was calculated using the obtained linear equation for the ethanol extract of curry leaves as well as the positive control, vitamin C. The test results for the ethanol extract of curry leaves indicated that the extract was able to neutralize DPPH free radicals, as evidenced by a decrease in absorbance compared to the blank, namely the solution without sample addition. The absorbance measurement results were analyzed through the relationship between sample concentration and percentage of inhibition, showing that antioxidant activity increased with increasing concentration. A linear regression equation

was then established to obtain the effective concentration (IC_{50}) value. The IC_{50} value represents the concentration of the ethanol extract of curry leaves required to achieve 50% inhibition of oxidation. The linear regression curve is presented in Figure 4.

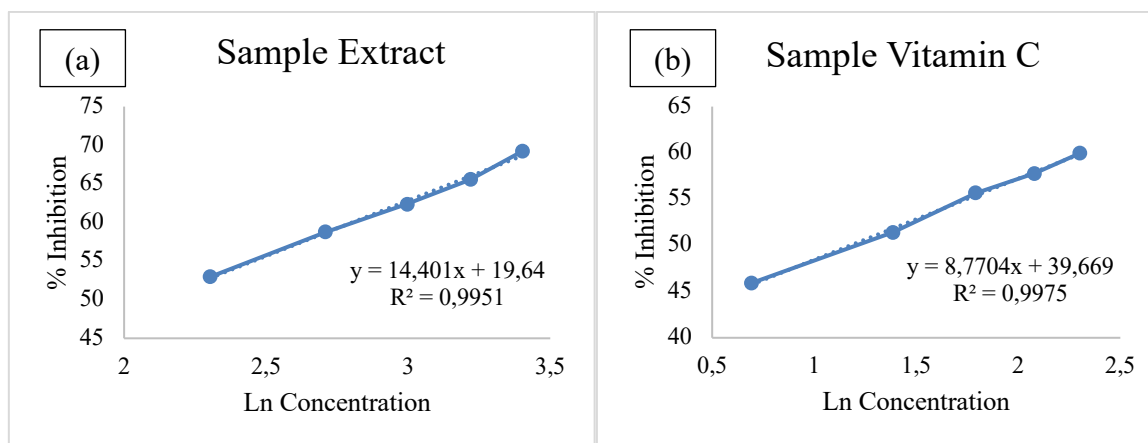


Figure 4. The Percentage of Antioxidant Activity Potential of: (a) the Ethanol Extract of Curry Leaves; and (b) Sample of Vitamin C

Based on the data presented in Table 3 above, the ethanol extract of curry leaves exhibited an IC_{50} value of 8.232, while vitamin C (positive control) showed an IC_{50} value of 3.247. The effectiveness of antioxidant compounds can be evaluated based on their IC_{50} values. An IC_{50} value below 50 ppm indicates very strong antioxidant activity in neutralizing free radicals, whereas compounds with IC_{50} values in the range of 50–100 ppm are classified as strong antioxidants. Furthermore, IC_{50} values of 100–150 ppm and 150–200 ppm are categorized as moderate and weak antioxidant activities, respectively (Zamzani et al., 2021). Based on the IC_{50} value categories described above, the IC_{50} value of the ethanol extract of curry leaves is classified as effective in scavenging free radicals.

These findings are consistent with previous studies reporting strong antioxidant activity of *Bergera koenigii* extracts. For instance, (Parashar et al., 2020) reported that methanolic extracts of curry leaves exhibited significant free radical scavenging activity due to the presence of polyphenolic compounds. Similarly, (Bhatt et al., 2022) reported high phenolic and flavonoid contents in *Bergera koenigii*, which are strongly associated with antioxidant potential.

The IC_{50} value obtained in the present study (8.23 $\mu\text{g/mL}$) indicates very strong antioxidant activity according to commonly used antioxidant classification standards. Previous research (Franyoto et al., 2025) reported a higher IC_{50} value for curry leaves extract, namely 28.82 $\mu\text{g/mL}$, which also falls within the strong antioxidant category. However, the lower IC_{50}

value observed in the present study suggests that the extract used in this research exhibits stronger antioxidant capacity compared to that reported in earlier studies.

This difference may be attributed to variations in extraction methods, solvent polarity, plant origin, or differences in phytochemical composition, particularly the levels of phenolic and flavonoid compounds that contribute significantly to free radical scavenging activity. The comparison with vitamin C as a positive control further highlights the antioxidant capacity of the extract. Vitamin C exhibited a lower IC_{50} value (3.247 $\mu\text{g/mL}$), indicating stronger antioxidant activity compared to the curry leaf extract. This difference is expected because vitamin C is a pure compound with a well-established radical scavenging ability, whereas plant extracts contain complex mixtures of bioactive compounds with varying antioxidant capacities. However, based on antioxidant activity classification standards, both vitamin C and the curry leaf extract still fall into the category of very strong antioxidants because their IC_{50} values are below 50 $\mu\text{g/mL}$ (Sirait et al., 2023). This indicates that although vitamin C demonstrates higher potency, the curry leaf extract still possesses a highly significant antioxidant capacity.

Nevertheless, the strong antioxidant activity of the curry leaf extract suggests that it may serve as a natural alternative antioxidant source in cosmetic formulations, where plant-derived ingredients are often preferred due to their additional biological activities, natural origin, and higher consumer acceptance. This antioxidant activity is consistent with the phytochemical screening results, which confirmed that the ethanol extract of curry leaves was found to contain flavonoids and tannins. These compounds belong to the polyphenol group, which is known to play a significant role in antioxidant activity (Bhuyan & Handique, 2022).

The antioxidant activity of flavonoids is primarily associated with two main mechanisms: hydrogen atom transfer (HAT) and single electron transfer (SET). In the HAT mechanism, flavonoids donate a hydrogen atom (consisting of both a proton and an electron) to neutralize free radicals (Hou et al., 2025). In the SET mechanism, antioxidants transfer an electron to the radical species, reducing its reactivity and forming a more stable molecular structure. These mechanisms enable flavonoids and other polyphenolic compounds to effectively scavenge DPPH radicals, resulting in the observed decrease in absorbance during the assay (Bhuyan & Handique, 2022; Vo et al., 2019).

Therefore, the strong antioxidant activity observed in this study can be attributed to the presence of polyphenolic compounds, particularly flavonoids and tannins, which are capable of stabilizing free radicals through electron donation and hydrogen atom transfer mechanisms. In the context of cosmetic applications, such antioxidant properties are important because they

may help protect skin cells from oxidative stress induced by ultraviolet radiation and environmental pollutants (Tyrrell, 2012) thereby supporting the development of natural antioxidant-based skincare products.

Physicochemical Evaluation

Organoleptic evaluation was performed on the formulation, including observations of color, odor, form, and phase separation. This test was carried out to evaluate the stability of the formulation. The results of the organoleptic and homogeneity evaluation are presented in Table 4 below.

Table 4. Organoleptic and Homogeneity Evaluation of Curry Leaf Peel-Off Mask

Formula	Time (Days)	Organoleptic			Homogeneity
		Color	Odor	Form	
F1	0	Clear	Extract odor	Gel	Homogen
	1	Clear	Extract odor	Gel	Homogen
	7	Clear	Extract odor	Gel	Homogen
	14	Clear	Extract odor	Gel	Homogen
	21	Clear	Extract odor	Gel	Homogen
	28	Clear	Extract odor	Gel	Homogen
F2	0	Clear green	Extract odor	Gel	Homogen
	1	Clear green	Extract odor	Gel	Homogen
	7	Clear green	Extract odor	Gel	Homogen
	14	Clear green	Extract odor	Gel	Homogen
	21	Clear green	Extract odor	Gel	Homogen
	28	Clear green	Extract odor	Gel	Homogen
F3	0	Clear green	Extract odor	Gel	Homogen
	1	Clear green	Extract odor	Gel	Homogen
	7	Clear green	Extract odor	Gel	Homogen
	14	Clear green	Extract odor	Gel	Homogen
	21	Clear green	Extract odor	Gel	Homogen
	28	Clear green	Extract odor	Gel	Homogen

The homogeneity test of the peel-off mask was conducted to evaluate the uniformity of ingredient mixing within the mask formulation and to confirm that the formulation exhibited consistent composition and homogeneous texture. Based on the outcomes of the homogeneity test presented in Table 4 above, all three peel-off mask formulations exhibited homogeneous characteristics, as indicated by the lack of visible particles or granules when observed on a glass slide. The homogeneous appearance of the formulations indicates that the dispersion of the curry leaf extract within the polymer matrix was well achieved. Uniform distribution of active

ingredients is essential in topical formulations because it ensures consistent delivery of bioactive compounds to the skin surface during application.

pH measurement was performed to assess the safety of the formulation when applied to the skin, ensuring that it does not cause adverse reactions such as irritation or other skin responses. Formulations with pH values below the physiological pH may potentially impair the skin's protective barrier function, thereby leading to skin irritation (Farage et al., 2018). Conversely, pH values higher than the physiological pH may lead to an increase in transepidermal water loss (TEWL) accompanied by a reduction in skin hydration (Goh et al., 2020). Table 5 and Figure 5 show that during storage, all formulations experienced a decrease in pH values; however, the values remained within the established pH range of 4.5 to 6.5 (Luki, 2021). The slight reduction in pH observed during storage may be associated with gradual chemical interactions between the plant extract components and the gel matrix, or minor oxidation processes occurring within the formulation. However, since the pH remained within the acceptable range for topical cosmetic products, these changes are unlikely to affect product safety or skin compatibility.

Viscosity is a key parameter that influences the ease of application of a formulation. Viscosity testing was performed to determine the thickness or flow behavior of the mask. The results obtained met the viscosity criteria for semi-solid preparations. During the 28-day storage period, a gradual decrease in viscosity was observed in all formulations. This phenomenon may be attributed to relaxation of the polymer network formed by PVA and HPMC. Over time, polymer chains may undergo structural rearrangement or partial weakening of intermolecular hydrogen bonding interactions within the gel matrix, resulting in reduced resistance to flow (Bercea et al., 2025).

The optimal viscosity range for a high-quality peel-off mask formulation is between 5,000 and 50,000 cPs (Nazar et al., 2025). Despite the slight decrease in viscosity, the observed values remained within the acceptable range for peel-off mask formulations, indicating that the structural integrity of the gel matrix was maintained during storage (Leblanc, 2017). An increase in viscosity is directly correlated with a corresponding increase in resistance. Higher gel viscosity results in a greater force being required to maintain flow at a given rate. In addition, increased viscosity reduces the rate of dispersed phase separation, thereby enhancing the stability of gel formulations (Ao et al., 2025).

The spreadability test results before and after storage showed an increase for each formulation. This condition occurred due to a decrease in formulation viscosity, which resulted

in an increased ability of the formulation to spread. Spreadability testing was conducted to determine the extent to which the formulation can spread and evenly coat the skin surface. Spreadability is an important parameter because it influences the uniform distribution of the mask on the skin surface. Improved spreadability facilitates easier application and enhances user comfort during cosmetic use.

The increase in spreadability may also be influenced by the concentration of curry leaf extract. As the extract concentration increased from 0% (F1) to 2% (F3), slight variations in rheological properties were observed. The presence of extract components may interfere with the polymer network structure, leading to subtle modifications in gel elasticity and film-forming properties (Kola & Carvalho, 2023). Based on the evaluation results, the peel-off gel mask was considered to meet the required criteria. The acceptable spreadability range for topically applied semi-solid preparations is 5–7 cm (Santoso et al., 2020). According to Mandal & Ap (2024) spreadability is inversely related to viscosity, where a decrease in viscosity increases the spreading ability of a formulation. The findings of this study are in accordance with this theory.

The drying time test was conducted to determine the duration required for the mask to reach a completely dry state and form a thin film on the skin surface following application. Drying time is a critical parameter in peel-off mask formulations because it determines the practicality and user acceptance of the product. Masks that dry too quickly may form uneven films, while excessively long drying times may reduce consumer comfort during use. The results demonstrated that all three mask formulations exhibited drying times within the acceptable range of 15–30 minutes. The drying behavior is strongly influenced by the viscosity and polymer concentration of the formulation. Higher viscosity formulations generally require longer drying times because the thicker gel matrix slows down water evaporation. (Nazar et al., 2025).

Table 5. Peel-off Mask Evaluation (Curry Leaf Ethanol Extract)

Formula	Time (Days)	Parameter			
		pH	ViscoSity (cPs)	Spreadability (cm)	Drying time (minutes)
F1	0	6.53	7140	5.9	15.59
	1	6.53	7131	5.9	15.55
	7	6.51	7056	6.1	15.23
	14	6.49	6950	6.3	15.15
	21	6.45	6878	6.4	14.83
	28	6.40	6740	6.6	14.03

	0	6.56	11416	5.3	16.09
	1	6.56	11400	5.5	16.05
	7	6.53	11320	5.5	15.98
F2	14	6.50	11030	5.6	15.80
	21	6.48	10978	5.8	15.69
	28	6.44	10120	5.9	15.55
	0	6.63	13729	5.5	16.36
	1	6.63	13701	5.6	16.29
F3	7	6.61	13005	5.7	16.03
	14	6.57	12780	5.8	15.89
	21	6.55	12007	5.9	15.83
	28	6.51	11864	5.9	15.71

The cycling test was conducted to evaluate the stability of the formulation under extreme temperature conditions with a high level of stress. Accelerated stability testing, such as the cycling test, is widely used in cosmetic formulation studies to predict the long-term stability of products under normal storage conditions. The absence of phase separation, color change, or odor alteration after repeated temperature cycles suggests that the polymer matrix of the formulation remained structurally stable under thermal stress (Akdogan, 2023). These findings indicate that the peel-off mask formulation possesses good physical stability and is likely to maintain its quality during storage and distribution.

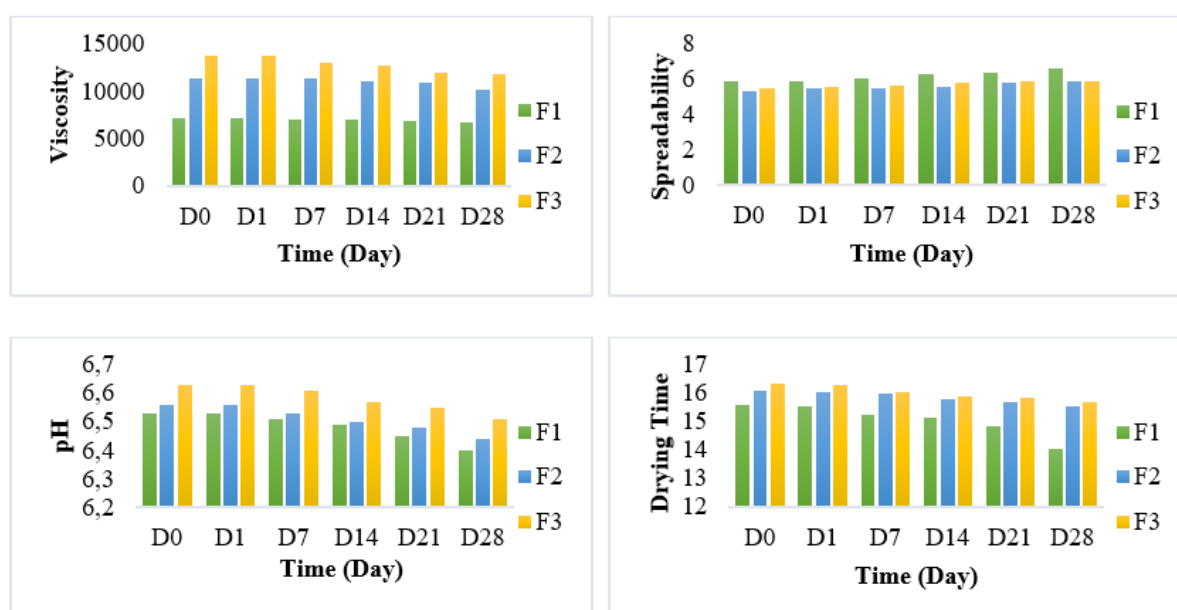


Figure 5. Evaluation Results Of Viscosity, Spreadability, pH, And Drying Time of The Peel-Off Mask (Curry Leaf Ethanol Extract)

The irritation test results from 30 volunteer respondents (Table 6) using a simple patch test method. Patch testing is commonly used in preliminary cosmetic safety evaluations to detect potential skin irritation responses such as erythema or edema. The irritation test results from 30 respondents showed no signs of irritation, including erythema or edema, among the volunteers. Although these findings indicate that the formulation is well tolerated, additional dermatological evaluations and standardized clinical testing protocols would be required to further confirm the safety profile of the formulation. Based on these findings, it can be concluded that the peel-off gel mask formulation is safe for application.

Table 6. Peel-off Mask Irritation Test (Curry Leaf Ethanol Extract)

Participants	Evaluation results					
	F1		F2		F3	
	Erythema	Edema	Erythema	Edema	Erythema	Edema
1	-	-	-	-	-	-
2	-	-	-	-	-	-
3	-	-	-	-	-	-
4	-	-	-	-	-	-
5	-	-	-	-	-	-
6	-	-	-	-	-	-
7	-	-	-	-	-	-
8	-	-	-	-	-	-
9	-	-	-	-	-	-
10	-	-	-	-	-	-
11	-	-	-	-	-	-
12	-	-	-	-	-	-
13	-	-	-	-	-	-
14	-	-	-	-	-	-
15	-	-	-	-	-	-
16	-	-	-	-	-	-
17	-	-	-	-	-	-
18	-	-	-	-	-	-
19	-	-	-	-	-	-
20	-	-	-	-	-	-
21	-	-	-	-	-	-
22	-	-	-	-	-	-
23	-	-	-	-	-	-
24	-	-	-	-	-	-
25	-	-	-	-	-	-
26	-	-	-	-	-	-
27	-	-	-	-	-	-

28	-	-	-	-	-	-
29	-	-	-	-	-	-
30	-	-	-	-	-	-

CONCLUSION

The antioxidant assay of the ethanol extract of curry leaves demonstrated strong antioxidant potential, as indicated by an IC_{50} value of 8.23. Physicochemical evaluation of the peel-off mask formulation showed that all formulations met acceptable quality parameters, including viscosity, spreadability, drying time, and pH stability during storage. These findings indicate that *Bergera koenigii* ethanol extract has promising potential as a natural antioxidant ingredient for cosmetic formulations. The peel-off mask system provides a practical topical delivery platform that supports the distribution of antioxidant compounds on the skin surface while maintaining suitable physicochemical stability. The irritation test suggested that the formulation was well tolerated under the tested conditions; however, this preliminary assessment was limited in scope and should be interpreted cautiously. Further dermatological evaluation and long-term stability studies are recommended to confirm the safety and effectiveness of the formulation and to optimize the concentration of curry leaf extract for cosmetic applications.

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