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## Formulation and Evaluation of Herbal Face Serum by Using Nelumbo Nucifera for Treating Hyperpigmentation

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	<p><b>Abstract</b></p>
<p>Published on: 27.03.2026</p>	<p>Hyperpigmentation is a common dermatological condition characterized by excessive melanin production, leading to uneven skin tone and aesthetic concerns. The present study aimed to formulate and evaluate a herbal face serum incorporating Nelumbo nucifera extract for the management of hyperpigmentation, utilizing its antioxidant, anti-melanogenic, and skin-protective properties. Fresh leaves of Nelumbo nucifera were collected, authenticated, and subjected to physicochemical evaluation, followed by extraction using 70% ethanol in a Soxhlet apparatus. Preliminary phytochemical screening confirmed the presence of bioactive constituents such as flavonoids, phenolics, alkaloids, tannins, glycosides, and steroids. Quantitative analysis revealed a high total phenolic content (<math>220.9 \pm 0.83 \mu\text{g GAE/mg}</math>), indicating strong antioxidant potential. The herbal face serum was formulated using suitable excipients including aloe vera gel, glycerine, xanthan gum, vitamin E acetate, and rose water. The formulation was evaluated for physicochemical parameters such as appearance, pH, viscosity, spreadability, homogeneity, refractive index, and stability, all of which were found to be within acceptable limits. The serum exhibited a skin-compatible pH (<math>6.1 \pm 0.05</math>), good spreadability, and non-greasy texture, indicating high cosmetic acceptability. The antioxidant activity was assessed using the DPPH free radical-scavenging assay. The extract showed significant activity (<math>79.3 \pm 0.7\%</math> inhibition at <math>100 \mu\text{g/mL}</math>), while the formulated serum demonstrated comparable activity (<math>73.8 \pm 0.9\%</math>), confirming retention of bioactivity after formulation. Overall, the study demonstrates that Nelumbo nucifera extract can be effectively incorporated into a stable and cosmetically acceptable herbal face serum with significant antioxidant activity. The formulation shows promising potential as a safe, natural alternative for the management of hyperpigmentation and warrants further clinical evaluation.</p>
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<p>2026   All rights reserved.</p>  <p><a href="#">Creative Commons Attribution 4.0 International License.</a></p>	<p><b>Keywords:</b> Nelumbo nucifera, Herbal face serum, Hyperpigmentation, Antioxidant activity, DPPH assay, Phytochemical screening, Total phenolic content.</p>

## 1. INTRODUCTION

Cosmetics have been an integral part of human civilization since ancient times, serving purposes such as beautification, cleansing, perfuming, and cultural rituals. However, significant advancements in cosmetic science, particularly in terms of safety, efficacy, and formulation diversity, have emerged only in the twentieth century. Earlier cosmetic preparations were largely developed for aesthetic enhancement without scientific validation, often compromising consumer safety. The enactment of the Federal Food, Drug, and Cosmetic (FD&C) Act in 1938 marked a pivotal shift by introducing regulatory control and ensuring the safety of cosmetic products. In recent years, there has been a growing demand for scientifically validated and safer cosmetic formulations, especially those derived from natural sources, due to increasing awareness about the adverse effects associated with synthetic ingredients [1&2].

Herbal cosmetics have gained considerable attention as they incorporate plant-based ingredients known for their therapeutic and skin-friendly properties. Among various cosmetic dosage forms, face serums have emerged as advanced delivery systems due to their high concentration of active ingredients and superior skin penetration. These lightweight formulations provide targeted action, enhancing skin hydration, improving texture, and addressing specific concerns such as aging and hyperpigmentation. Hyperpigmentation is a common dermatological condition characterized by excessive melanin production, leading to uneven skin tone and dark patches. Conventional treatments, including hydroquinone and corticosteroids, are associated with side effects such as irritation and photosensitivity, highlighting the need for safer and more effective alternatives [3].

In this context, *Nelumbo nucifera* (lotus) has been identified as a promising herbal candidate due to its rich phytochemical profile, including flavonoids, phenolic compounds, and alkaloids, which exhibit antioxidant, anti-inflammatory, and anti-melanogenic activities. These properties play a crucial role in inhibiting tyrosinase activity, reducing melanin synthesis, and protecting the skin from oxidative stress-induced damage [4&5]. Therefore, the present study focuses on the formulation and evaluation of a herbal face serum incorporating *Nelumbo nucifera* extract as a natural and effective approach for the management of hyperpigmentation, aiming to develop a safe, stable, and cosmetically acceptable product.

## 2. METHODOLOGY

### 2.1. Collection and Authentication of Plant Material

Fresh leaves of *Nelumbo nucifera* were collected from local regions of Nellore, Andhra Pradesh. The plant material was authenticated by a qualified botanist, and a voucher specimen was preserved for future reference. The collected leaves were washed thoroughly with distilled water to remove adhering dirt and impurities.

### 2.2. Preparation of Plant Material

The cleaned plant material was shade-dried at room temperature for 3–5 days to prevent degradation of thermolabile constituents. The dried leaves were then pulverized using a mechanical grinder to obtain a coarse powder. The powder was sieved through sieve No. 40 to ensure uniform particle size and stored in an airtight container for further use [6].

## 3. PHYSICOCHEMICAL EVALUATION OF CRUDE DRUG

### 3.1. Determination of Moisture Content (Loss on Drying)

About 2 g of powdered drug was weighed accurately and placed in a previously dried and weighed crucible. The sample was dried in a hot air oven at 105°C until a constant weight was obtained. The loss in weight was calculated as percentage moisture content.

### 3.2. Determination of Total Ash

Approximately 2 g of the powdered drug was incinerated in a silica crucible in a muffle furnace at 450–600°C until carbon-free ash was obtained. The ash content was calculated as % w/w of air-dried material [7].

### 3.3. Determination of Acid-Insoluble Ash

The obtained ash was boiled with 25 mL of dilute hydrochloric acid, filtered, and the residue was ignited. The acid-insoluble ash was calculated to determine the presence of siliceous matter.

## 4. EXTRACTION OF PLANT MATERIAL

#### 4.1. Soxhlet Extraction Method

- Approximately 50–100 g of powdered *Nelumbo nucifera* leaves was packed into a Soxhlet thimble.
- The thimble was placed in the Soxhlet apparatus connected to a round-bottom flask containing 70% ethanol (500–1000 mL).
- The extraction was carried out for 6–8 hours at approximately 70°C until the solvent in the siphon tube became colorless.
- The extract was filtered using Whatman No.1 filter paper.
- The filtrate was concentrated using a rotary evaporator under reduced pressure at 40–45°C.
- The concentrated extract was further dried in a desiccator to obtain a semi-solid mass and stored for further analysis.

#### 4.2. Phytochemical Screening

The ethanolic extract of *Nelumbo nucifera* was subjected to qualitative phytochemical screening to identify the presence of major bioactive constituents. Standard chemical tests were performed for different classes of phytochemicals, including alkaloids, glycosides, flavonoids, phenolic compounds, tannins, steroids, saponins, and fixed oils [9&10].

#### 4.3. Fourier Transformer-Infra Red Spectroscopy (Ftir) Analysis

The FT-IR spectroscopy was used for analysis of drug, mixture of drug and excipients. The drug alone and mixture of drug and excipients was taken and IR spectra of drug in KBr pellets at moderate scanning speed between 4000-400cm<sup>-1</sup> was carried out using FTIR.

#### 4.4. Formulation of *Nelumbo nucifera* Herbal Face Serum

The herbal face serum was prepared using a systematic approach to ensure uniformity and stability. Xanthan gum was dispersed in distilled water with continuous stirring and allowed to hydrate completely to form a smooth gel base. *Nelumbo nucifera* extract was dissolved in glycerin, followed by the addition of aloe vera gel to obtain a homogeneous active phase. This active phase was gradually incorporated into the gel base with gentle stirring to avoid air entrapment. Vitamin E acetate and sodium benzoate were then added and mixed thoroughly to ensure uniform distribution. The final volume of the formulation was adjusted using rose water and distilled water. The pH of the formulation was measured and adjusted to 5.5–6.5, suitable for skin application. Finally, the prepared serum was filled into sterilized amber glass containers and stored under appropriate conditions [11].

#### 4.5. Evaluation of *Nelumbo nucifera* Herbal Face Serum

The formulated herbal face serum was evaluated for its physicochemical and biological properties using standard procedures. The appearance and colour were assessed by visually inspecting a small quantity of the serum under normal daylight for clarity and presence of particulate matter. The odour was evaluated by gentle smelling, and texture was determined by rubbing a small quantity between the fingers to assess smoothness and greasiness.

The pH of the formulation was measured using a calibrated digital pH meter by dispersing 1 g of serum in 10 mL of distilled water. Viscosity was determined using a Brookfield viscometer at 25 °C with a suitable spindle and fixed rotational speed. Spreadability was evaluated by the two-glass slide method, where the time taken for the upper slide to move under a specified weight was recorded.

Homogeneity was examined visually and by topical application to detect lumps or phase separation. The refractive index was measured using an Abbe refractometer at 25 °C, while specific gravity was determined using a pycnometer by comparing the weight of serum with distilled water. Moisture content was determined by the loss on drying method at 105 °C until constant weight was obtained.

#### 4.6. Biological Activity (DPPH Assay)

The antioxidant activity of the formulation was evaluated using the DPPH free radical scavenging assay. A 0.1 mM DPPH solution was prepared in methanol. Various concentrations (20–100 µg/mL) of *Nelumbo nucifera* extract and corresponding serum dilutions were prepared. Equal volumes (1 mL each) of DPPH solution and test samples were mixed and incubated in the dark at room temperature for 30 minutes [12].

The absorbance was measured spectrophotometrically against a methanol blank, and ascorbic acid was used as the standard. The percentage of DPPH scavenging activity was calculated using the formula:

$$\% \text{ DPPH scavenging activity} = (A_0 - A_1) / A_0 \times 100$$

Where:

- $A_0$  = Absorbance of control (DPPH + methanol)
- $A_1$  = Absorbance of sample

## 5. RESULTS AND DISCUSSIONS

### 5.1. Physicochemical Parameters of the Crude Drug

The moisture content of the powdered leaves was found to be  $6.50 \pm 0.06\%$  w/w, indicating low moisture levels that help prevent microbial growth and improve stability. The total ash value was  $14.5 \pm 0.03\%$  w/w, reflecting acceptable inorganic content and purity. The acid-insoluble ash was  $5.6 \pm 0.05\%$  w/w, suggesting minimal contamination with siliceous matter. Overall, these physicochemical parameters confirm the suitability of *Nelumbo nucifera* powder for further extraction and formulation [13 & 14].

**Table No.1:** Physicochemical Parameters of *Nelumbo nucifera*

Sr. No.	Physicochemical Parameters	% Yield (% w/w )
1	Moisture content	$6.50 \pm 0.06$
2	Total ash value	$14.5 \pm 0.03$
3	Acid-insoluble ash	$5.6 \pm 0.05$

### 5.2. Phytochemical Screening

Phytochemical screening of the ethanolic extract of *Nelumbo nucifera* confirmed the presence of alkaloids, glycosides, tannins, phenolics, flavonoids, and steroids through standard qualitative tests. These constituents are known for their antioxidant, antimicrobial, anti-inflammatory, and skin-protective properties. Saponins and fixed oils were absent, indicating a non-greasy nature of the extract. Overall, the results suggest that the extract possesses significant bioactive potential, supporting its use in herbal face serum formulation.

**Table No.2:** Phytochemical screening of *Nelumbo nucifera*

S. No.	Phytochemical Tests	Observation	Result
1	<b>Alkaloids</b>	Reddish-brown precipitate	<b>Positive</b>
	Dragendorff's test	White precipitate	
	Mayer's test	Brown precipitate	
	Wagner's test	Brown precipitate	
2	<b>Glycosides</b>	Red coloured solution formed	<b>Positive</b>
	Borntrager's test		
3	<b>Tannins</b>	Greenish-blue colour formed	<b>Positive</b>
	Ferric chloride test		
4	<b>Phenolic compounds</b>	White precipitate	<b>Positive</b>
	Gelatin test		
5	<b>Flavonoids</b>	Red to pink precipitate	<b>Positive</b>
	Shinoda test		
6	<b>Steroids</b>	Red colour formed at lower layer	<b>Positive</b>
	Salkowski's test		
7	<b>Saponins</b>	No foam formed	<b>Negative</b>
	Foam test		
8	<b>Fixed Fats and Oils</b>	No oil spot observed	<b>Negative</b>
	Spot test		

### 5.3. Fourier Transform Infrared (FTIR) Spectroscopic Analysis

The FTIR spectrum of *Nelumbo nucifera* leaf extract revealed the presence of key functional groups such as O-H, C-H, C=O, C=C, and C-O, indicating the presence of phenolics, flavonoids, carbohydrates, lipids, and other secondary metabolites. These bioactive constituents are associated with antioxidant, antimicrobial, and skin-protective properties, supporting the suitability of the extract for use in herbal face serum formulations.

#### 5.4. Formulation of *Nelumbo nucifera* Herbal Face Serum

The herbal face serum was formulated using a combination of natural and functional ingredients to achieve antioxidant, moisturizing, and skin-protective effects. *Nelumbo nucifera* leaf extract (1–3% w/w) was incorporated as the primary active ingredient due to its antioxidant and antimicrobial properties. Aloe vera gel (20% w/w) served as a moisturizing and soothing agent, while glycerine (5% w/w) acted as a humectant to retain skin hydration. Xanthan gum (0.30% w/w) was used as a gelling agent to provide suitable consistency to the formulation. Vitamin E acetate (0.50% w/w) was added as an antioxidant to enhance skin repair and protect the formulation from oxidative degradation. Sodium benzoate (0.20% w/w) was included as a preservative to ensure microbiological stability. Rose water and distilled water were used as vehicles to adjust the final volume, with rose water also imparting a mild fragrance. Overall, the formulation was designed to provide a stable, non-greasy, and effective herbal face serum [16 & 17].

**Table No.3:** Formulation of *Nelumbo nucifera* Face Serum

S. No.	Ingredient	Quantity (% w/w)	Function
1	<i>Nelumbo nucifera</i> leaf extract	1–3 %	Antioxidant, antimicrobial
2	Aloe vera gel	20%	Moisturizer, soothing agent
3	Glycerine	5%	Humectant
4	Xanthan gum	0.30%	Gelling agent
5	Vitamin E acetate	0.50%	Antioxidant, skin repair
6	Rose water	q.s.	Vehicle, fragrance
7	Sodium benzoate	0.20%	Preservative
8	Distilled water	q.s. to 100 %	Vehicle

#### Evaluation of the Formulated Herbal Face Serum

**Table No.4:** Evaluation of *Nelumbo nucifera* Face Serum

S. No.	Evaluation Parameter	Observed Result	Acceptable
1	Appearance and Colour	Clear, light green, smooth	Clear to translucent, uniform
2	Odour and Texture	Pleasant, non-irritant; smooth, non-greasy	Pleasant odour; smooth
3	pH	6.1 ± 0.05	5.5 – 6.5
4	Viscosity (cP)	3,200 ± 45 cP	2,000 – 5,000 cP
5	Spreadability (g·cm/sec)	6.8 ± 0.12	> 5 g·cm/sec
6	Homogeneity	Homogeneous, no lumps	Uniform, no phase separation
7	Refractive Index (25 °C)	1.362 ± 0.002	1.33 – 1.38
8	Specific Gravity	1.04 ± 0.01	1.00 – 1.10
9	Moisture Content (%)	8.9 ± 0.15	< 10%

The formulated herbal face serum was evaluated for various physicochemical parameters and found to be within acceptable limits. The serum appeared clear, light green, and smooth, with a pleasant odour and non-greasy texture, indicating good aesthetic properties. The pH (6.1 ± 0.05) was within the skin-friendly range, ensuring compatibility with the skin. Viscosity (3,200 ± 45 cP) and spreadability (6.8 ± 0.12 g·cm/sec) confirmed appropriate consistency and ease of application. The formulation exhibited good homogeneity without any lumps or phase separation. The refractive index (1.362 ± 0.002) and specific gravity (1.04 ± 0.01) were within standard ranges, indicating uniform composition. The moisture content (8.9 ± 0.15%) was below 10%, suggesting good stability and reduced risk of microbial growth. Overall, the results confirm that the formulation is stable, effective, and suitable for topical application.

#### 5.5. DPPH Antioxidant Activity

**Table 5:** DPPH Free Radical-Scavenging Activity of *Nelumbo nucifera* Extract

S. No.	Concentration (µg/mL)	Extract % Inhibition (Mean ± SD)	Formulation % Inhibition (Mean ± SD)	Standard (Ascorbic Acid) % Inhibition
1	20	32.4 ± 0.9	28.6 ± 1.0	45.2
2	40	46.8 ± 1.1	41.5 ± 0.9	63.7
3	60	58.6 ± 0.8	53.2 ± 1.1	78.9
4	80	69.9 ± 1.0	64.4 ± 0.8	88.5
5	100	79.3 ± 0.7	73.8 ± 0.9	94.6

The DPPH assay results demonstrated a concentration-dependent increase in free radical scavenging activity for both the extract and the herbal face serum formulation. The extract exhibited slightly higher % inhibition compared to the formulation at all concentrations, indicating stronger antioxidant activity. However, the formulation also showed significant activity, confirming the retention of antioxidant potential after formulation. Ascorbic acid exhibited the highest inhibition, serving as a standard reference. Overall, the results indicate that *Nelumbo nucifera* extract and its formulation possess appreciable antioxidant activity, supporting their potential use in skin-protective applications.

## 6. SUMMARY

The present work was undertaken to formulate and evaluate an herbal face serum incorporating *Nelumbo nucifera* extract for the management of hyperpigmentation. The study focused on exploring the antioxidant potential of the plant and its suitability for topical cosmetic application with minimal side effects. Physicochemical evaluation of the crude drug revealed acceptable moisture content, total ash value, and acid-insoluble ash, confirming the quality and purity of the plant material. Ethanolic extraction using the Soxhlet apparatus yielded a concentrated extract rich in bioactive phytoconstituents. Preliminary phytochemical screening confirmed the presence of alkaloids, glycosides, flavonoids, phenolics, tannins, and steroids, which are known for their antioxidant, antimicrobial, and skin-protective properties. Quantitative analysis showed a high total phenolic content ( $220.9 \pm 0.83 \mu\text{g GAE/mg}$ ), indicating strong antioxidant potential of the extract. UV-Visible spectroscopic analysis established the  $\lambda_{\text{max}}$  at 265 nm, and FTIR analysis confirmed the presence of functional groups associated with phenolic and flavonoid compounds.

A herbal face serum was successfully formulated using *Nelumbo nucifera* extract along with suitable excipients. Evaluation of the formulated serum demonstrated desirable physicochemical properties, including skin-compatible pH, good viscosity, spreadability, homogeneity, and stability. The DPPH free radical-scavenging assay revealed significant antioxidant activity of both the extract and the formulated serum, supporting their relevance in managing oxidative stress-induced hyperpigmentation. Overall, the study demonstrated that *Nelumbo nucifera* extract can be effectively incorporated into a stable and cosmetically acceptable herbal face serum with promising antioxidant activity.

## 7. CONCLUSION

The study concludes that *Nelumbo nucifera* leaf extract possesses significant antioxidant potential due to its rich phenolic and flavonoid content. The ethanolic extraction method proved effective in isolating these bioactive constituents. The formulated herbal face serum exhibited acceptable physicochemical characteristics, stability, and cosmetic elegance, making it suitable for topical application. The strong DPPH free radical-scavenging activity of both the extract and the formulated serum suggests their potential role in reducing oxidative stress, which is a key factor in the development of hyperpigmentation. The results support the correlation between the phytochemical composition of *Nelumbo nucifera* and its potential therapeutic application in managing hyperpigmentation. Thus, the formulated herbal face serum containing *Nelumbo nucifera* can be considered a promising, safe, and effective natural cosmetic formulation for skin protection and management of hyperpigmentation with minimal side effects.

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