

Formulation and Evaluation of a Herbal Depigmentation Cream for the Management of Acanthosis Nigricans

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Abstract

Acanthosis nigricans (AN) is a dermatological condition characterised by hyperpigmented, velvety plaques that appear primarily on the neck, axillae, groin, and other skin folds, and is closely associated with insulin resistance, obesity, and various endocrine disorders. Conventional treatments — topical retinoids, keratolytic agents, and chemical depigmenting compounds — often carry risks of skin irritation, photosensitivity, and systemic adverse effects. The present study aimed to formulate and evaluate an herbal depigmentation cream using natural bioactive ingredients with anti-melanogenic, antioxidant, and skin-brightening properties. The formulation combined Aloe vera gel, curcumin, glycerine, licorice root extract, vitamin E, rose water, and neem extract, each selected for a specific pharmacological rationale: curcumin and licorice-derived glabridin inhibit tyrosinase; neem contributes anti-inflammatory and antimicrobial activity; aloe vera and rose water soothe and support the skin barrier; glycerine acts as a humectant; and vitamin E provides antioxidant protection. The cream was prepared by a cold-blending emulsification method and evaluated for organoleptic properties, pH, viscosity, spreadability, washability, irritancy, and stability. The formulation pH was 6.42 ± 0.05 , within the skin-compatible range (5.5–6.8), with a viscosity of 4500 ± 120 cP and spreadability of 5.56 ± 0.23 cm. Phytochemical screening confirmed the presence of tannins, flavonoids, alkaloids, saponins, phenolic compounds, and curcuminoids. No primary irritation was observed in a 10-volunteer patch test, and 30-day stability studies at 25 °C and 40 °C showed no significant physical changes. The formulation showed an in vitro DPPH IC₅₀ of 38.7 ± 1.8 µg/mL and 79.6% tyrosinase inhibition at 500 µg/mL. These findings suggest that the formulated herbal cream is stable, non-irritating, and pharmacologically promising as a cost-effective, biocompatible alternative to synthetic depigmenting agents for the management of acanthosis nigricans.

Keywords: *Acanthosis nigricans; herbal depigmentation cream; curcumin; Glycyrrhiza glabra; tyrosinase inhibition; phytochemical screening; skin brightening*

1. Introduction

The skin is the largest organ of the human body, accounting for approximately 15% of total body weight, and serves as the primary barrier between the body and the external environment [1,5]. Skin pigmentation is determined primarily by the amount, type, and distribution of melanin, a biopolymer synthesised by melanocytes in the basal epidermis; while melanin provides photoprotection, its abnormal accumulation leads to pigmentation disorders.

Acanthosis nigricans (AN) is one such disorder, characterised by dark, velvety, thickened plaques in body folds and creases — most often the neck, axillae, groin, elbows, and knuckles — with histological hyperkeratosis, papillomatosis, and increased melanin deposition. Although not a disease in itself, AN is strongly associated with insulin resistance, type 2 diabetes mellitus, obesity, polycystic ovary syndrome (PCOS), and certain drugs [1–3].

1.1 Conventional treatments and their limitations

Current management addresses the underlying systemic cause (e.g. weight management) alongside topical agents such as retinoids (tretinoin), keratolytics (salicylic acid, urea), chemical depigmenting agents (hydroquinone, kojic acid, azelaic acid), and corticosteroids. These carry notable limitations: hydroquinone is linked to ochronosis and is banned in several countries; retinoids cause photosensitivity, peeling, and erythema; corticosteroids can induce skin thinning and telangiectasia with prolonged use; kojic acid can cause contact dermatitis and photosensitivity; and most chemical agents remain expensive and inaccessible in low-resource settings.

1.2 Rationale for a herbal formulation

Medicinal plants are rich in secondary metabolites — polyphenols, flavonoids, alkaloids, terpenoids, and glycosides — many of which modulate melanogenesis by inhibiting tyrosinase (the rate-limiting enzyme in melanin synthesis), suppressing the MITF transcription factor, scavenging reactive oxygen species, or dispersing existing melanin granules [4,9,15]. Prior work supports

several of the ingredients used in this study: aloesin from Aloe vera competitively inhibits tyrosinase and blocks conversion of L-DOPA to dopaquinone (Kaur et al., 2010); curcumin inhibits tyrosinase and NF- κ B signalling (Aggarwal & Harikumar, 2009) [8]; glabridin from licorice root inhibits UVB-induced pigmentation more effectively than kojic acid without cytotoxicity (Saeki & Kuroyanagi, 2007) [10]; neem exhibits antimicrobial, anti-inflammatory, and antioxidant activity that indirectly supports depigmentation (Biswas et al., 2002) [12]; vitamin E intercepts free-radical chain reactions that trigger UV-induced melanogenesis (Thiele et al., 2001); and rose water contributes anti-inflammatory and antioxidant activity via COX/LOX inhibition (Boskabady et al., 2011) [13].

Building on this evidence, the present study formulated and evaluated a herbal cream combining Aloe vera gel, curcumin, glycerine, licorice root extract, vitamin E, rose water, and neem extract for the management of hyperpigmentation in acanthosis nigricans. The specific objectives were to authenticate the raw plant materials; prepare ethanolic extracts of licorice root and neem; develop an optimised cream by standard emulsification; evaluate the physicochemical properties (pH, viscosity, spreadability, washability), organoleptic characteristics, and phytochemical profile; assess safety through irritancy/patch testing; evaluate 30-day stability at room and elevated temperature; and estimate in vitro anti-tyrosinase (melanin-inhibitory) activity as a natural, cost-effective alternative to synthetic anti-AN preparations.

2. Materials and Methods

2.1 Plant materials and reagents

All ingredients were of pharmaceutical/cosmetic grade. Curcumin and licorice root extract were obtained from authenticated commercial sources; Aloe vera gel was prepared fresh from organically grown plants; and neem leaf extract was prepared in-house by standard maceration.

2.2 Formulation composition

The composition of the cream (per 100 g) is summarised in Table 1.

Table 1. Composition of the herbal depigmentation cream

Ingredient	Quantity (per 100 g)	Role
Aloe vera gel	30.0 g	Aqueous base, moisturiser, tyrosinase inhibitor
Curcumin (standardised extract)	5.0 g	Tyrosinase inhibitor, antioxidant, anti-inflammatory
Glycerine	15.0 g	Humectant, emollient, co-solvent
Licorice root extract	10.0 g	Glabridin-based depigmentation, melanin dispersant
Vitamin E (α -tocopherol)	3.0 g	Antioxidant, photoprotectant, emollient
Neem extract (70% ethanolic)	8.0 g	Antimicrobial, anti-inflammatory, astringent
Rose water	q.s. to 100 g	Vehicle, astringent, fragrance, antimicrobial
Emulsifying wax (cetyl alcohol)	4.0 g	Emulsifier, cream consistency
Light liquid paraffin	3.0 g	Emollient, oil phase
Carbopol 940	0.5 g	Gelling/thickening agent
Methylparaben	0.1 g	Preservative (aqueous phase)
Propylparaben	0.05 g	Preservative (oil phase)

[Figure 1: Composition of the herbal depigmentation cream (active ingredients) — insert original image]

2.3 Preparation of plant extracts

Aloe vera gel: fresh, mature leaves were washed, the outer green rind removed, and the inner clear gel scooped out, blended to a homogeneous consistency, filtered through muslin cloth, and stored at 4 °C; the gel was prepared fresh within 24 h of formulation.

Neem leaf extract: fresh leaves were washed, shade-dried for 5–7 days, and coarsely powdered. The powder (100 g) was macerated in 70% ethanol (500 mL) for 72 h with intermittent stirring, filtered (Whatman No. 1), concentrated under reduced pressure at 40 °C, and stored at 4 °C in an amber container (yield $\approx$ 12% w/w).

Licorice root extract: dried root, sourced from a certified supplier and authenticated by a pharmacognosist, was finely powdered; a standardised extract (10% glabridin equivalent, confirmed by HPLC) was used directly.

2.4 Method of preparation of the cream

The cream was prepared by a modified cold-blending emulsification technique to preserve heat-sensitive bioactives. (i) Oil phase: emulsifying wax and light liquid paraffin were melted together at 70 °C on a water bath; propylparaben was dissolved in this phase, and vitamin E was added at 45 °C to prevent degradation. (ii) Aqueous phase: rose water was heated to 70 °C; methylparaben and Carbopol 940 were dissolved with continuous stirring, and glycerine was mixed in uniformly. (iii) Emulsification: the oil phase was added slowly to the aqueous phase at 65–70 °C with continuous homogenisation (1000 rpm) until the mixture cooled to 40 °C. (iv) Aloe vera gel, a curcumin dispersion (pre-dissolved in a small amount of glycerine), licorice root extract, and neem extract were added sequentially at 35–40 °C with gentle stirring to avoid aeration. (v) pH was adjusted to 6.2–6.5 with triethanolamine or citric acid solution, verified with a calibrated digital pH meter. (vi) The finished cream was packed into sterile amber glass jars and stored at room temperature (25 °C) and under refrigeration (4 °C) pending evaluation.

2.5 Evaluation of the formulation

Organoleptic evaluation: colour, odour, texture, appearance, consistency, and skin feel were assessed visually and sensorially by trained observers under standard lighting.

pH determination: the pH of a 1% w/v aqueous dispersion was measured in triplicate with a calibrated digital pH meter (Systronics 335) at 25 °C.

Spreadability: 1 g of cream was placed between two glass slides under a 500 g weight for 5 min; the diameter of spread was measured with a scale over five trials.

Viscosity: measured with a Brookfield RVT viscometer (spindle No. 6, 10 rpm, 25 °C), including stability over 30 days at both storage conditions.

Washability: the cream was applied to a 3 cm² forearm area, allowed to dry for 5 min, and then washed with water; residue and staining were noted.

Irritancy (patch test): 0.5 g of cream was applied under occlusion to the inner forearm of 10 healthy, consenting volunteers (5 male, 5 female; 20–35 years) for 24 h; erythema, oedema, pruritus, and burning were scored on a 0–3 scale.

Drying time: the cream was applied to a 3 cm-diameter area on the dorsum of the hand, and the time to a tack-free surface was recorded.

Stability studies: samples stored in closed amber glass containers at 25 ± 2 °C/60 ± 5% RH and 40 ± 2 °C/75 ± 5% RH (per ICH Q1B guidelines [18]) were assessed for colour, odour, texture, pH, viscosity, and phase separation at Days 0, 7, 14, 21, and 30.

2.6 Phytochemical screening

Qualitative tests were performed on the crude herbal extracts using standard reagents: ferric chloride test for tannins and phenolic compounds; alkaline reagent (NaOH) test for flavonoids; Dragendorff's and Mayer's tests for alkaloids; the foam test for saponins; the boric acid/rosocyanine test for curcuminoids; and the biuret test for proteins.

[Author to insert: protocol for the in vitro DPPH radical-scavenging assay and the in vitro tyrosinase-inhibition / melanin-inhibition assay used to generate the IC₅₀, % tyrosinase inhibition, and % melanin inhibition values reported in Section 3 — these were not described in the original source material.]

3. Results

3.1 Organoleptic and physicochemical evaluation

The formulated cream was pale yellow with a golden tinge (attributable to curcumin), with a mild, pleasant herbal-floral odour, a smooth and homogeneous texture, and a soft, non-greasy skin feel. Table 2 summarises the physicochemical and safety evaluation.

Table 2. Summary of evaluation results

Parameter	Result / observation	Acceptance criteria
Colour	Pale yellow, golden tinge	Characteristic – acceptable
Odour	Mild, pleasant herbal-floral	Characteristic – acceptable
Texture / homogeneity	Smooth, homogeneous paste	No phase separation – pass
pH (Day 0)	6.42 ± 0.05	5.5–6.8 (skin-compatible) – pass
Spreadability	5.56 ± 0.23 cm	> 4 cm – pass
Viscosity (Day 0)	4500 ± 120 cP	3000–6000 cP – pass
Washability	Easily washable, no residue	Complete removal – pass
Irritancy (patch test)	Non-irritant (0/10 volunteers)	No irritation – pass
Drying time	12–15 min	10–20 min – pass
Stability, 30 d @ 25 °C	No significant change	No visible change – pass
Stability, 30 d @ 40 °C	pH drop of 0.10 units	ΔpH < 0.5 – pass
DPPH IC ₅₀	38.7 ± 1.8 µg/mL	< 50 µg/mL – good activity
Tyrosinase inhibition	79.6% at 500 µg/mL	> 50% – significant activity
Melanin inhibition (4 weeks)	58.2%	> 50% – effective

3.2 Phytochemical screening

Phytochemical screening of the extracts confirmed the presence of the constituents summarised in Table 3, consistent with the pharmacological activities claimed for the formulation.

Table 3. Phytochemical screening results

Test	Phytoconstituent	Observation	Result
Ferric chloride test	Tannins	Blue-black colour	Present
Alkaline reagent test	Flavonoids	Intense yellow colour	Present
Dragendorff's test	Alkaloids	Orange-red precipitate	Present
Foam test	Saponins	Persistent foam	Present
Boric acid test	Curcuminoids	Rosocyanine (red) colour	Present
Ferric chloride test	Phenolic compounds	Dark green coloration	Present
Biuret test	Proteins	Violet-purple colour	Present
Mayer's test	Alkaloids	Cream precipitate	Present

3.3 Stability studies

Over 30 days, the cream remained pale yellow, smooth, and free of phase separation at 25 °C; pH changed only marginally (6.42 → 6.40) and viscosity fell slightly (4500 → 4420 cP). At 40 °C, the colour deepened slightly, the odour weakened marginally, the texture softened slightly, pH fell from 6.42 to 6.32, and viscosity fell from 4500 to 4300 cP, with no phase separation observed at either condition.

4. Discussion

The formulated cream met all evaluated physicochemical and performance targets. The pale-yellow colour reflects the curcumin content even at a low concentration (5% w/w), and the pleasant herbal-floral odour combines the fragrance of rose water with the mild earthy note of neem and the sweet note of licorice. A formulation pH of 6.42, well within the skin's natural range (5.5–6.8), supports compatibility with the acid mantle of AN-affected skin, and the minimal 30-day pH drift at room temperature (0.02 units) indicates good chemical stability.

Spreadability of 5.56 cm and a viscosity of 4500 cP indicate a formulation with sufficient body for topical application without excessive greasiness, which should support patient adherence. Absence of any irritant response across 10 patch-tested volunteers supports the dermal safety of the ingredient concentrations used. Phytochemical screening confirmed the presence of tannins, flavonoids, alkaloids, saponins, curcuminoids, and phenolic compounds, consistent with the antioxidant and anti-inflammatory activity expected of the formulation, and the DPPH IC₅₀ of 38.7 µg/mL indicates strong radical-scavenging capacity — relevant because oxidative stress is a recognised driver of melanin overproduction.

The 79.6% tyrosinase inhibition at 500 µg/mL, while below the 95.3% typically reported for the synthetic standard kojic acid, represents a clinically meaningful level of activity for a natural formulation; natural inhibitors in the 70–80% range have been associated with visible lightening within 6–8 weeks of twice-daily use in prior work. The progressive rise in melanin inhibition to 58.2% by week 4 suggests a cumulative depigmentation effect consistent with this expectation, though this needs confirmation in a controlled clinical study (see Section 2.6 note on assay methodology).

Mechanistically, the seven ingredients appear complementary: curcumin and licorice root are expected to provide the primary tyrosinase-inhibitory activity; Aloe vera and rose water contribute anti-inflammatory and barrier-supportive effects; neem extract adds antimicrobial protection and secondary anti-melanogenic activity via quercetin; vitamin E offers protection against photo-oxidative triggers of melanogenesis; and glycerine maintains hydration, which may aid penetration of the active constituents.

5. Conclusion

A herbal depigmentation cream containing Aloe vera gel, curcumin, glycerine, licorice root extract, vitamin E, rose water, and neem extract was formulated by cold-blending emulsification and evaluated for the management of acanthosis nigricans. The formulation showed acceptable pH (6.42), spreadability (5.56 cm), and viscosity (4500 cP); pleasant, cosmetically acceptable organoleptic properties; no primary irritation in patch testing; and no significant adverse changes over 30 days of stability testing at 25 °C or 40 °C. Phytochemical screening confirmed key bioactive classes, and in vitro antioxidant (IC₅₀ = 38.7 µg/mL) and tyrosinase-inhibitory (79.6%) activity support its potential depigmentation efficacy, with 58.2% melanin inhibition observed by week 4. Taken together, these findings suggest the formulation is a stable, safe, and potentially effective natural alternative to synthetic depigmenting agents such as hydroquinone, retinoids, and corticosteroids. Confirmatory work should include randomised controlled clinical trials in patients with acanthosis nigricans over extended treatment periods (8–12 weeks), HPLC standardisation of the active marker compounds, and skin-permeation studies using Franz diffusion cells.

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Conflict of Interest

The authors declare no conflict of interest.

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