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Formulation, Physicochemical Evaluation and Antimicrobial Activity of a Polyherbal Topical Cream Containing *Tridax procumbens*, *Aloe vera*, *Curcuma longa* and *Mentha* spp.

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Abstract

This study aimed to formulate and evaluate a polyherbal topical cream containing *Tridax procumbens*, *Aloe vera*, *Curcuma longa*, and *Mentha* spp. for antimicrobial skincare applications. An oil-in-water emulsion was prepared using beeswax and liquid paraffin as the base. Physicochemical parameters including pH, viscosity, spreadability, phase stability, and skin irritation were evaluated. Antimicrobial activity was assessed against *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Salmonella typhi*, *Klebsiella pneumoniae*, and *Candida albicans* using the agar well diffusion method.

The cream exhibited good homogeneity, stability without phase separation, suitable pH (7.68–7.97), and no skin irritation. Significant antimicrobial activity was observed, with maximum inhibition zones of 25 mm against *Klebsiella pneumoniae* and *Candida albicans*. Preliminary phytochemical analysis revealed the presence of flavonoids and phenolic compounds, which may contribute to the antimicrobial effect.

The developed polyherbal cream demonstrates promising antimicrobial potential, acceptable physicochemical stability, and suitability for topical use. Further studies on long-term stability and clinical efficacy are recommended.

Keywords: Polyherbal cream, Topical formulation, Antimicrobial activity, Herbal cosmetics, *Tridax procumbens*.

1. Introduction

Creams are semisolid emulsions intended for topical application and are commonly formulated as oil-in-water (O/W) or water-in-oil (W/O) systems depending on the nature of the continuous phase [1]. Such formulations are widely used in pharmaceutical and cosmetic preparations to deliver active ingredients, maintain skin hydration, and protect the skin from environmental stressors [2].

In recent years, there has been a growing consumer preference for herbal cosmetic and pharmaceutical products due to their perceived safety, biocompatibility, and reduced risk of adverse effects when compared to synthetic formulations [3]. Medicinal plants are rich sources of bioactive phytochemicals such as flavonoids, phenolics, terpenoids, and alkaloids, which are known to exhibit antimicrobial, anti-inflammatory, antioxidant, and wound-healing properties, making them suitable candidates for topical applications [4].

The formulation of stable, antimicrobial herbal topical preparations is of increasing pharmaceutical interest due to rising antimicrobial resistance and growing consumer preference for plant-based products. Several international studies indexed in PubMed and Scopus have demonstrated that polyherbal topical formulations exhibit significant antimicrobial activity along with acceptable physicochemical stability and skin compatibility. For example, Patil *et al*. reported enhanced antibacterial effects in a polyherbal cream containing *Allium sativum*, *Moringa oleifera*, and *Thymus vulgaris*. Similarly, Nipanikar *et al*. demonstrated that a herbal cream formulation showed both antimicrobial and anti-inflammatory activities without causing skin irritation. These findings support the scientific rationale for combining multiple herbal extracts in topical dosage forms to achieve synergistic therapeutic effects.

Aloe vera (*Aloe barbadensis*) is widely recognized for its moisturizing, soothing, anti-acne, and wound-healing effects, primarily attributed to polysaccharides, glycoproteins, and vitamins present in the gel [5]. Turmeric (*Curcuma longa*) contains curcumin as its principal bioactive compound, which possesses well-documented antioxidant, anti-inflammatory, and antimicrobial properties beneficial for skin health [6]. Mint (*Mentha* spp.) is known for its cooling sensation and antimicrobial potential due to the presence of phenolic compounds and flavonoids, making it a valuable ingredient in dermatological and cosmetic formulations [7].

Tridax procumbens is a traditionally used medicinal herb for the treatment of wounds and skin infections and has been

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reported to exhibit significant antibacterial and anti-inflammatory activities against various skin pathogens, which are attributed to its rich phytochemical profile [8]. Despite extensive documentation of these individual herbs, scientific reports on their combined formulation into a single polyherbal topical cream with synergistic antimicrobial and skin-protective effects remain limited [9].

The novelty of the present study lies in the formulation of a polyherbal topical cream incorporating a specific combination of *Tridax procumbens*, *Aloe vera*, *Curcuma longa*, and *Mentha* spp., aimed at achieving enhanced antimicrobial efficacy along with desirable cosmetic and pharmaceutical properties. Therefore, this study was undertaken to formulate and evaluate a polyherbal cream and to assess its physicochemical characteristics, stability, skin compatibility, and antimicrobial activity against selected bacterial and fungal pathogens.

2. Materials and Methods

2.1. Materials

Fresh leaves of *Tridax procumbens* and *Aloe vera* (Aloe barbadensis) were collected locally and authenticated. Turmeric (Curcuma longa) powder and dried mint (Mentha spp.) leaves were procured from a certified herbal supplier. Beeswax, liquid paraffin, glycerin, rose water, and fragrance oil were of pharmaceutical grade. All chemicals and solvents were of analytical grade.

2.2. Preparation of Plant Extracts

i). ***Tridax procumbens* Extract:** Leaves were washed, chopped, and dried in a hot air oven at 80 °C for 3–4 days. The dried leaves were powdered. Ten grams of powder were macerated with 100 mL of 70% ethanol for 48 hours with intermittent shaking, filtered through Whatman No. 1 filter paper, and stored at 4 °C.

ii). **Turmeric Extract:** One gram of turmeric powder was mixed with 10 mL distilled water, heated at 80–100 °C for 5–10 minutes, cooled, and filtered.

iii). **Mint Extract:** One gram of dried mint leaves was mixed with 10 mL distilled water, heated at 80–100 °C for 5–10 minutes, cooled, and filtered.

iv). **Aloe vera Gel:** Fresh Aloe vera leaves were washed, outer rind removed, and gel collected. Fibrous material removed by filtration; gel stored at 4 °C.

2.3. Preliminary Phytochemical Screening

Preliminary phytochemical screening was carried out using standard qualitative methods as described by Harborne (1998). This provides rationale for antimicrobial activity in the cream.

2.4. Formulation of Polyherbal Cream

An oil-in-water emulsion was prepared. Beeswax and liquid paraffin formed the oil phase; plant extracts, glycerin, rose water, and fragrance oil formed the aqueous phase. Both phases were heated to ~70 °C, aqueous phase gradually added to oil phase with continuous stirring, and cooled to room temperature to obtain a homogeneous cream. Three formulations (C1, C2, C3) were prepared by varying extract concentrations.

2.5. Evaluation of Polyherbal Cream

i). **Physical and Physicochemical Properties**

- **Color, Odor, Consistency:** Visual and sensory inspection
- **pH:** Measured in 5 g cream + 50 mL distilled water using a calibrated digital pH meter
- **Washability & greasiness:** Assessed on skin
- **Phase Stability:** Monitored for 30 days
- **Viscosity:** Measured using a Brookfield viscometer
- **Spreadability:** Slip and drag method; $S = (M \times L)/T$, where S = spreadability, M = applied weight, L = distance moved, T = time (s)
- **Extrudability:** Measured as the amount of cream extruded from a tube under fixed pressure

2.5.2 Microbial Limit Test

Standard plate count method for bacterial and fungal load; results expressed as CFU/mL.

2.5.3 Determination of Phytochemical Content

- Total phenolic content: Folin-Ciocalteu method; expressed as mg gallic acid equivalents/g extract.
- Total flavonoid content: Aluminum chloride colorimetric method; expressed as mg quercetin equivalents/g extract.

2.5.4 *In-vitro* Antioxidant Activity

DPPH free radical scavenging assay performed on extracts. Percentage inhibition calculated and expressed as mean ± SD.

2.6. Antimicrobial Activity

Agar well diffusion method against *S. aureus*, *P. aeruginosa*, *S. typhi*, *K. pneumoniae*, and *C. albicans*.

- Wells loaded with 2%, 5%, 10% extract concentrations.
- Positive controls: Standard antibiotic discs (e.g., ampicillin for bacteria) and antifungal discs (e.g., fluconazole for *C. albicans*).
- Incubation: 37 °C for 24 h (bacteria) or 25 °C for 48 h (*C. albicans*).
- Zones of inhibition measured in mm.
- All tests performed in triplicate; results expressed as mean ± SD.
- Statistical analysis: ANOVA, $p < 0.05$ considered significant.

2.7. Accelerated Stability Study

Formulations stored at 4 °C, room temperature, and 40 °C with 75% relative humidity for 30–90 days. Observed periodically for color, odor, pH, viscosity, phase separation, and microbial growth.

2.8. Microbiological Challenge Test

Cream inoculated with standard test strains (*S. aureus*, *E. coli*, *P. aeruginosa*, *C. albicans*) and monitored over 7–14 days to assess microbial stability and preservative efficacy.

2.9. Patch Test for Skin Irritation

Applied to 20–30 healthy volunteers under dermatological supervision; observed for 24–72 hours for redness, itching, or inflammation. The patch test was conducted with informed consent from all volunteers, in accordance with institutional ethical guidelines.

3. Results and Discussion

The formulated polyherbal cream containing extracts of *Tridax procumbens*, *Aloe vera*, mint (*Mentha* spp.), and turmeric (*Curcuma longa*) exhibited satisfactory physicochemical properties, stability, skin compatibility, and biological activity. The results confirm the successful

development of a stable, cosmetically acceptable, and biologically active herbal topical formulation.

3.1. Physical Evaluation

All formulations (C1, C2, and C3) showed a faint yellow color, smooth semisolid consistency, and a pleasant lavender odor. The creams were homogeneous, free from grittiness or phase separation, and showed uniform texture, indicating proper emulsification and formulation stability.

Table 1: Physical parameters of cream formulations

Sr. No.	Parameter	C1	C2	C3
1	Color	Faint yellow	Faint yellow	Faint yellow
2	Odor	Pleasant	Pleasant	Pleasant
3	State	Semisolid	Semisolid	Semisolid
4	Consistency	Smooth	Smooth	Smooth

Fig 1: Physical appearance of the formulated polyherbal cream.

3.2. Irritancy, Washability and Greasiness

All formulations were found to be non-irritant when applied to the skin. The creams were easily washable with water and exhibited non-greasy behavior, indicating suitability for topical application and good patient acceptability.

Table 2: Irritancy test results

Sr. No.	Formulation	Result
1	C1	Non-irritant
2	C2	Non-irritant
3	C3	Non-irritant

3.3. pH and Phase Stability

The pH of the formulations ranged between 7.68 and 7.97. Although slightly alkaline, no signs of irritation or discomfort were observed during skin application studies. No phase separation was observed during the study period, indicating good formulation stability.

Table 3: pH and phase separation

Formulation	pH	Phase Separation
C1	7.68	None observed
C2	7.97	None observed
C3	7.80	None observed

Although the pH was slightly alkaline, no skin irritation was observed, indicating acceptable compatibility for short-term topical application.

3.4. Viscosity and Spreadability

Viscosity measurements using a Brookfield viscometer ranged from 4500–4800 cP, indicating appropriate consistency for topical application. Spreadability studies demonstrated smooth and uniform spreading of the cream with minimal resistance, ensuring ease of application and adequate skin coverage.

3.5. After-Feel Properties

All formulations exhibited good emollience, slipperiness, and minimal residue after application. These sensory properties confirm acceptable cosmetic characteristics and user compliance.

Table 4: After-feel evaluation

Formulation	After Feel
C1	Good
C2	Good
C3	Good

3.6. Antimicrobial Activity

The formulated polyherbal cream demonstrated significant antimicrobial activity against both Gram-positive, Gram-negative bacteria, and fungal strains. Maximum zones of inhibition were observed against *Klebsiella pneumoniae* (25 mm) and *Candida albicans* (25 mm). The antimicrobial efficacy can be attributed to the synergistic effect of phytoconstituents present in the herbal extracts.

Table 5: Antimicrobial activity of cream (zone of inhibition, mm)

Organism	2% Extract
Staphylococcus aureus	9
Pseudomonas aeruginosa	21
Salmonella typhi	15
Klebsiella pneumoniae	25
Candida albicans	25

3.7. Preliminary Phytochemical Screening

Qualitative phytochemical analysis of the individual plant extracts revealed the presence of flavonoids, phenolics, tannins, saponins, and terpenoids. These bioactive compounds are known for their antimicrobial and antioxidant properties and provide scientific justification for the observed biological activity of the formulation.

Table 6: Phytochemical screening of extracts

Phytochemical	Tridax	Aloe	Turmeric	Mint
Alkaloids	+	–	+	–
Flavonoids	+	+	+	+
Phenolics	+	+	+	+
Tannins	+	–	–	+
Saponins	–	+	–	+
Terpenoids	+	+	+	+

(+ present, – absent)

3.8. Total Phenolic and Flavonoid Content

The combined herbal extract exhibited appreciable phenolic and flavonoid contents, expressed as gallic acid equivalents (GAE) and quercetin equivalents (QE), respectively. These phytochemicals are associated with enhanced antimicrobial and antioxidant activities.

Table 7: Phenolic and flavonoid content

Parameter	Content (Mean ± SD)
Total phenolic content	68.4 ± 2.1 mg GAE/g
Total flavonoid content	42.7 ± 1.8 mg QE/g

3.9. Antioxidant Activity (DPPH Assay)

The herbal extracts showed concentration-dependent free radical scavenging activity in the DPPH assay, indicating moderate to strong antioxidant potential. This activity may contribute to skin protection and formulation stability.

Table 8: DPPH radical scavenging activity

Concentration (µg/mL)	% Inhibition
25	31.2 ± 1.4
50	48.6 ± 1.9
100	67.8 ± 2.3

3.10. Extrudability

Extrudability studies confirmed that the cream could be easily extruded from collapsible tubes with uniform pressure, indicating appropriate viscosity and ease of application for topical use.

3.11. Microbial Load and Safety

The total viable microbial count of the formulated cream was 3.15×10^3 cfu/mL, which is within acceptable limits for topical formulations, confirming microbiological safety.

3.12. Accelerated Stability Studies

Formulations stored at 40 °C ± 2 °C and 75% ± 5% RH for up to 90 days showed no significant changes in color, odor, pH, viscosity, consistency, or phase separation. Microbial counts also remained within acceptable limits, indicating good physical and microbiological stability.

4. Conclusion

A stable polyherbal topical cream containing extracts of *Tridax procumbens*, *Aloe vera*, mint (*Mentha* spp.), and turmeric (*Curcuma longa*) was successfully developed and evaluated. The formulation exhibited acceptable physicochemical properties, good spreadability, suitable viscosity, and favorable sensory characteristics.

The cream was non-irritant, non-greasy, and easily washable, indicating suitability for topical application. Significant antibacterial and antifungal activities were observed against selected microorganisms, which may be attributed to the presence of bioactive phytochemicals such as flavonoids, phenolics, tannins, and terpenoids. The formulation complied with microbial safety limits and remained stable under accelerated storage conditions.

Overall, the developed polyherbal cream demonstrates promising antimicrobial potential and acceptable stability, supporting its potential use as a topical herbal formulation. Further studies involving extended stability testing and in vivo evaluation are recommended to confirm its clinical applicability.

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