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DESIGN AND DEVELOPMENT OF HERBAL FORMULATIONS FOR THE TREATMENT OF TOPICAL SKIN DISORDERS

Swaroopa Rani Gurram[1], Mandula Sravanthi[2], Kothoju Maheshwari[3], Kuchanapally Sangeetha[3], Kota Sravani[3], Kuntigorla Saikiran[3]

[1]Associate Professor, Vikas College of Pharmaceutical Sciences, Rayanigudem, Suryapet.
[2]Assistant Professor, Vikas College of Pharmaceutical Sciences, Rayanigudem, Suryapet.
[3]Student, Vikas College of Pharmaceutical Sciences, Rayanigudem, Suryapet.

Abstract:
The present study was carried out to formulate, characterize, and evaluate a topical antibacterial herbal cream using Piper beetle and Ocimum tenuiflorum extracts as major herbal sources. Herbal medicines are increasingly preferred due to their safety, effectiveness, and fewer side effects compared to synthetic drugs. Piper beetle possesses antimicrobial, antioxidant, and anti-inflammatory properties, while Ocimum tenuiflorum is well known for antibacterial, wound-healing, and antioxidant activities. The herbal extracts were incorporated into a cream base prepared using suitable emulsifying agents and excipients. The cream formulation was evaluated for physicochemical parameters such as appearance, pH, viscosity, spread ability, homogeneity, washability, stability, and antibacterial activity. The formulation exhibited satisfactory antibacterial activity against common skin pathogens and showed acceptable physicochemical properties suitable for topical application. The study suggests that the prepared herbal cream can be used as an effective natural antibacterial topical preparation.The formulation and evaluation of a herbal cream containing extracts from Betel leaf (Piper beetle) and Tulsi (Ocimum sanctum) were carried out to explore their potential therapeutic properties in skin care. .The current study's goal was to create a herbal cream that will moisturize, nourish, lighten, protect, and treat a range of skin conditions.Betel leaf and Tulsi are known for their antimicrobial, anti-inflammatory, and antioxidant properties, whichare beneficialfor treating skin disorders. Products used to enhance one's appearance are known as herbal cosmetics. Creating a herbal cream that would moisturize, nourish, whiten, and treat a variety of skin conditions was the aim of the study. Among the fundamental ingredients used to produce the cream are Betel Leaf(Piper betel) and Ocimum sanctum (tulsi leaves).

Keywords: *Betel leaf, Piper betle, Tulsi, Ocimum sanctum, Herbal cream, Topical formulation, Antibacterial activity, Antioxidant activity, Anti-inflammatory activity, Herbal cosmetics, Skin care, Wound healing.*

Corresponding author:
Swaroopa Rani Gurram,
Associate Professor, Vikas College of Pharmaceutical Sciences, Rayanigudem, Suryapet, Telangana, India
Mobile:

QR CODE

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INTRODUCTION:

Herbal medicines have been used since ancient times for the treatment of various diseases. The increasing awareness about the side effects of synthetic drugs has lead to a growing interest in herbal formulations. Topical preparations are commonly used for treating skin infections because they provide direct action at the site of infection. Herbal creams are semisolid preparations that offer easy application, better absorption, and improved patient compliance.

Piper beetle belongs to the family Piperaceae and is commonly known as Betel leaf. The leaves contain active constituents such as chavicol, eugenol, hydroxychavicol, tannins, and flavonoids which contribute toantimicrobial activity. Ocimum tenuiflorum belongs to the family Lamiaceae and is commonly known as Tulsi or Holy Basil. It contains eugenol, rosmarinic acid, ursolic acid, and essential oils responsible for antibacterial and anti-inflammatory activities.The combination of Piper beetle and Ocimum tenuiflorum in a herbal cream may provide synergistic antibacterial effects and promote wound healing.

A herbal cream is a semisolid topical preparation containing natural plant extracts, oils, or herbal ingredients used for application on the skin. Herbal creams are widely used in cosmetic and medicinal formulations because they provide therapeutic effects with fewer side effects compared to synthetic products. Herbal creams help in moisturizing the skin, treating minor skin infections, reducing inflammation, promoting wound healing, and improving overall skin health. Common herbal ingredients used in creams include aloe vera, neem, turmeric, betel leaf, tulsi, and calendula, which possess antimicrobial, antioxidant, and anti-inflammatory properties.

Due to their safety, effectiveness, and natural origin, herbal creams have gained significant popularity in skincare and pharmaceutical industries. They are easy to apply, non-greasy, and enhance the delivery of active herbal constituents to the skin. "Herbal cream is a semisolid preparation containing medicinal plant extracts intended for external application on the skin. It is used for moisturizing, protecting, and treating various skin conditions. Herbal creams are preferred because of their natural ingredients, therapeutic benefits, and minimal side effects."

Types of Herbal Creams

1.**Oil-in-Water (O/W) Cream**

- Water is the continuous phase and oil is the dispersed phase.
- Non-greasy and easily washable with water.
- Suitable for oily skin and daytime use. Example: Moisturizing and vanishing creams.

2.**Water-in-Oil (W/O) Cream**

- Oil is the continuous phase and water is the dispersed phas e.
- Greasy and provides better moisturizing effect.
- Suitable for dry skin and cold weather.
- Example: Cold creams and nourishing creams.

3.Medicated Herbal Cream

- Contains herbal drugs for treating skin diseases.
- Used for antifungal, antibacterial, anti-inflammatory, or wound-healing purposes.
- Example: Neem cream, turmeric cream.

4.Cosmetic Herbal Cream

- Used mainly for beauty and skincare purposes.
- Helps in fairness, anti-aging, sun protection, and skin nourishment.
- Example: Aloe vera cream, sandalwood cream.

5.Moisturizing Herbal Cream

- Prevents skin dryness and keeps skin soft.
- Contains natural moisturizers like aloe vera and coconut oil.

6.Antiseptic Herbal Cream

Prevents microbial infections and promotes healing of cuts and wounds.Common ingredients include neem and tulsi extracts.

Betel leaf (Piper betle) is an excellent addition to herbal cream formulations due to its potent antimicrobial, anti-inflammatory, and antioxidant properties. It contains bioactive compounds like chavicol, eugenol, and hydroxychavicol, making it highly effective in treating acne, fungal infections, and minor wounds.

Why Betel Leaf in Skincare

➢ Antimicrobial: Effectively combats bacteria (e.g., Staphylococcus aureus) and fungi, preventing infection in acne or eczema-prone skin.

➢ Anti-inflammatory: Helps soothe swelling, redness, and itching associated with skin irritations.

➢ Antioxidant: Fights free radicals, protecting the skin from premature aging and damage.

Tulsi (Ocimum tenuiflorum), commonly known as Holy Basil, is an aromatic shrub native to India. Highly revered in Ayurveda as the "Queen of Herbs," it holds deep spiritual significance. The leaves are a powerhouse of antioxidants and essential oils, widely used to boost immunity, relieve respiratory issues, and reduce stress.

MATERIALS:

Tulasi leaf extract was procured from Synpharma Research Labs, Hyd, Aloe vera, Borax, Methyl paraben, Bee's wax, Liquid paraffin and Rose Oil were obtained from Synpharma Research Labs,

Hyderabad. Other chemicals and the reagents used were of analytical grade.

METHODOLOGY:

Preparation of Betel Leaf Extract by using (Maceration method)

Take the betel leaves and wash the leaves thoroughly with distilled water to remove dirt and impurities. Air-dry the leaves in the shade or in an oven at a low temperature (40°C - 50°C) until completely brittle. Grind the dried leaves into a fine powder and sift it (using a mesh sieve) to ensure a uniform particle size. Weigh out exactly 20g of betel leaf powder. Place the 20g powder into a clean, dark glass container (e.g., an amber jar) or maceration chamber. Add a solvent (typically70% to 96% ethanol or methanol). A standard leaf-to-solvent ratio is 1g of powder to 10-15mL of solvent. For 20g of powder, you will need 200mL to 300mL of solvent. Stir or agitate the mixture thoroughly, ensuring the powder is completely submerged. Seal the container and leave it at room temperature for 24 to 72 hours, shaking it periodically (a few times a day) to maximize extraction efficiency. After the soaking period, filter the mixture using a funnel lined with Whatman No. 1 filter paper into a clean flask. Press the remaining leaf residue slightly to extract trapped solvent. Transfer the filtered liquid (filtrate) into a Rotary Evaporator to remove the solvent at 40°C to 50°C under reduced pressure. Once the solvent has entirely evaporated, you will be left with a dark green/blackish-green thick crude extract with the characteristic, pungent odor of betel leaf.

Preparation of Tulasi Leaf Extract by using (Maceration method)

Take the thulasi leaves and wash the leaves thoroughly with distilled water to remove dirt and impurities. Air-dry the leaves in the shade or in an oven at a low temperature (40°C - 50°C) until completely brittle. Grind the dried leaves into a fine powder and sift it (using a mesh sieve) to ensure a uniform particle size. Weigh out exactly 20g of thulasi leaf powder. Place the 20g powder into a clean, dark glass container (e.g., an amber jar) or maceration chamber. Add a solvent (typically 70% to 96% ethanol or methanol). A standard leaf-to-solvent ratio is 1g of powder to 10-15mL of solvent. For 20g of powder, you will need 200mL to 300mL of solvent. Stir or agitate the mixture thoroughly, ensuring the powder is completely submerged. Seal the container and leave it at room temperature for 24 to 72 hours, shaking it periodically (a few times a day) to maximize extraction efficiency. After the soaking period, filter the mixture using a funnel lined with Whatman No. 1 filter paper into a clean flask. Press the remaining leaf residue slightly to extract trapped solvent. Transfer the filtered liquid (filtrate) into a Rotary Evaporator to remove the solvent at 40°C to 50°C under reduced pressure. Once the solvent has entirely evaporated, you will be left with a dark green/blackish-green thick crude extract with the characteristic, pungent odor of tulasi leaf.

Preparation of herbal cream

Table:1 Formulation

S.No	Ingredients	Quantities	Uses
1	Tulasi leaf extract	3ml	Antiseptic and Anti-inflammatory agent
2	Aloe vera	6ml	Moisturizer &Hydrating agent
3	Borax	2g	Emulsifier
4	Methyl paraben	1g	Preservative
5	Bee's wax	4g	Thickening agents
6	Liquid paraffin	5ml	Emollient
7	Rose Oil	1ml	Fragment

1. **Preparation Oil Phase:** Heat liquid paraffin 5ml and beeswax 4mg in a borosilicate beaker on a water bath or heating mantle until it reaches 75° C
2. **Preparation of Aqueous Phase:** In a separate beaker, dissolve borax and methyl paraben in distilled water and heat to 75° C until a clear solution is formed.
3. **Emulsification:** Slowly add the aqueous phase to the heated oily phase while stirring vigorously.
4. **Add Actives:** Allow the mixture to cool slightly (around 40° - 50° C), then stir in your and tulsi extracts.
5. **lab Technique:** Pour the mixture onto a clean porcelain ointment slab and mix thoroughly with a stainless-steel spatula in a geometric manner until it achieves a smooth, homogenous cream texture.

Table: 2 Formulation of Betel Leaf Extract

S.No	Ingredients	Quantities	Uses
1	Betel leaf extract	3ml	Antibacterial activity
2	Aloe vera	6ml	Moisturizer & hydrating agent
3	Borax	2g	Emulsifier
4	Methyl paraben	1g	Preservative
5	Bee's wax	4g	Thickening agents
6	Liquid paraffin	5ml	Emollient
7	Rose Oil	1ml	Fragment

1. **Preparation Oil Phase:** Heat liquid paraffin 5ml and beeswax 4mg in a borosilicate beaker on a water bath or heating mantle until it reaches 75° C
2. **Preparation of Aqueous Phase:** In a separate beaker, dissolve borax and methylparaben in distilled water and heat to 75° C until a clear solution is formed.
3. **Emulsification:** Slowly add the aqueous phase to the heated oily phase. While stirring vigorously
4. **Add Actives:** Allow the mixture to cool slightly (around 40° - 50° C), then stir in your and tulsi extracts.
5. **Slab Technique:** Pour the mixture onto a clean porcelain ointment slab and mix thoroughly with a stainless-steel spatula in a geometric manner until it achieves a smooth, homogenous cream texture.

Preparation Of Herbal Cream By Using F1 And F2 Process

Table: 3 Formulation of Herbal cream

S.No	Ingredients	Quantities	Uses
1	Betel leaf extract	3ml	Antibacterial activity
2	Tulasi leaf extract	3ml	Antiseptic and Anti inflammatory
3	Aloe vera	3ml	Moisturizer & hydrating agent
4	Borax	1g	Emulsifier
5	Methyl paraben	0.5g	Preservative
6	Bee's wax	2g	Thickening agents
7	Liquid paraffin	2.5ml	Emollient
8	Rose Oil	0.5ml	Fragment

1. **Preparation Oil Phase:** Heat liquid paraffin 5ml and beeswax 4mg in a borosilicate beaker on a water bath or heating mantle until it reaches 75° C
2. **Preparation of Aqueous Phase:** In a separate beaker, dissolve borax and methylparaben in distilled water and heat to 75° C until a clear solution is formed.
3. **Emulsification:** Slowly add the aqueous phase to the heated oily phase while stirring vigorously.
4. **Add Actives:** Allow the mixture to cool slightly (around 40° - 50° C), then stir in your betel and tulsi extracts.
5. **Slab Technique:** Pour the mixture onto a clean porcelain ointment slab and mix thoroughly with a stainless-steel spatula in a geometric manner until it achieves a smooth, homogenous cream texture.

Evaluation Tests

1.Physical Appearance

- The cream was evaluated for color, odor, texture, and consistency.
- pH Determination
- Measured using a digital pH meter. The pH should fall between 4.5 and
- 6.5 to match the skin's natural acidic mantle and prevent irritation.

2.Viscosity Measured using a viscometer to ensure the cream is thick enough to stay on the skin but soft enough to pump or squeeze from the tube.

3.Spreadability
Spreadability was determined by the apparatus which consists of a wooden block, which was provided by a pulley at one end. By this method spreadability was measured on the basis of slip and drag characteristics of creams. An excess of creams (about 2 g) under study was placed on this ground slide. The cream was then sandwiched between this slide and another glass slide having the dimension of fixed ground slide and provided with the hook. Weight of 1 kg was placed on the top of the slide for 5 minutes to expel air and to provide a uniform film of the cream between the slides.
Excess of the cream was scrapped off from the
Homogeneity

A visual or microscopic inspection to ensure no clumps or separated oils are present.
Washability
Washability was checked by washing the applied cream with water.

Stability Studies
The cream was stored at different temperatures and observed for changes.
Antibacterial Activity
Anti-bacterial Activity In Vitro Techniques
1. Preparation of Agar Medium

1. Prepare nutrient agar (2.8 gm nutrient agar dissolve in 100 ml of distilled water). Media should be preparedusing distilled water or deionized water Heat with frequent agitation and boil to dissolve the medium completely. Sterilize by autoclaving at 121 °C for15 min. Check the pH of each preparation after it is sterilized, which should be between 7.2 and at room temperature. This is done by macerating a small amount of medium in a little distilled water or by allowing a little amount of medium to gel around a pH meter electrode. Cool the agar medium to 40-50 °C. Pour the agar into sterile glass or plastic petri dish on a flat surface to a uniform depth of 4 mm.Allow to solidify.Prior to use, dry plates at 30-37 °C in an incubator, with lids partly ajar, for not more than 30 minutes or until excess surface moisture has evaporated. Media must be moist but free of water droplets on the surface. Presence of water droplets may result to swarming bacterial growth, which could give inaccurate results. They are also easily contaminated.

In vitro studies
In vitro antibacterial screening of Formulation

(i). Determination of minimum inhibitory concentration (MIC)
The antibacterial activity study of the formulations was performed bacterial strains, *Escherichia coli* NCIM 2065 following the guidelines of Clinical Laboratories Standard Institute (CLSI 2007). The edges. The top plate was then subjected to pull of 50 g. With the help of string attached to the hook and the time (in seconds) required by the top slideto cover a distance of 6.5 cm be noted. A shorter interval indicates better spreadability.Spreadability was calculated using the following formula:
$S = M \times L / T$

S = Spreadability,M = Weight in the pan (tied to the upper slide),

L = Length moved by the glass slide and T= Time (in sec.) taken to separate the slide completely each othe
tests were performed in Mueller Hinton medium (Hi-media) by broth micro dilution method, in 96-well micro titer plates. Formulations was dissolved in sterile dimethyl sulfoxide (DMSO) to screen their antibacterial activity. Ciprofloxacin were dissolved in sterile DMSO and used as positive control, while sterile DMSO served as a negative control. The final volume for MIC protocols was 100 µL, whereas DMSO concentration in assays well was less than 1%. formulations were tested in the concentration range of 19.74 to 221.98 µM. The bacterial suspensions at 10^5 Colony Forming Unit/mL (CFU/mL) concentrations were inoculated to the corresponding wells. Following inoculation 96-well microtitre plates was incubation at 37 °C for 24 h. Plates were then agitated and read for the absorbance at a wavelength of 600 nm. All tests were performed in triplicate, and the results were taken as a mean. After MIC determination, 50 µL aliquots from each well was sub-cultured on Mueller-Hinton agar plates and were further incubated at 37 °C for 24 h.All experiments were performed in triplicate. The wells are visually inspected for turbidity under black background to determine the growth of the organism. The wells containing the antimicrobial agent in concentration sufficient to inhibit the growth remain clear. In experimental terms the MIC is the concentration of the drug present in the last clear well, i.e. the well having the lowest antibiotic concentration in which growth is not observed, MIC values (µM)

(ii) Determination of Minimum bactericidal concentration (MBC)
The Minimum Bactericidal Concentration (MBC) is the lowest concentration of an antibacterial agent required to kill a bacterium over a fixed, somewhat extended period, such as 18 hours or 24 hours, under a specific set of conditions. It can be determined from the broth dilution of MIC tests by sub-culturing to agar plates that do not contain the test agent. The MBC is identified by determining the lowest concentration of antibacterial agent that reduces the viability of the initial bacterial inoculum by a pre-determined reduction such as ≥99.9%. The MBCs were determined by sub-culturing aliquots

(20 µl) from the bacterial suspension wells with no visible bacterial growth at concentrations greater than the defined MlC and from control wells onto nutrient agar plates. The plates were incubated aerobically at 37 °C for 24 hours and colonies were comparatively counted with the starting inoculum. MBC was determined as the concentration at which a ≥99.9% decrease in bacterial counts (i.e., 3 log l0 reduction in cfu/ml) was achieved or the lowest concentrations that did not produce any bacterial growth in the agar plates were regarded as MBC values. All experiments were repeated in triplicate for each strain. The ratio MBC/MIC ≤2 indicates bactericidal activity, while MBC/MIC ratio ≥4 shows bacteriostatic activity of test formulations MIC and MBC values (µM) of the tested compounds are presented in Table 28-30 respectively.

Irritancy: First, we took a measurement of one centimeter squared for a small area on the back of the left hand. After the application of cream, we took note of the time. After up to a day, we looked to see whether there was any swelling or redness. We told someone about it if it was true.

RESULTS & DISCUSSION:

Table: 4 Evaluation parameters of formulation

S.No	Evaluation test	F1	F2	F3
1	Color	Deep greenish	Green	Pale greenish
2	Odor	Pleasant	Pleasant	Pleasant
3	Texture	Smooth	Smooth	Smooth
4	State	Semi solid	Semi solid	Semi solid
5	Irritant effect	Nil	Nil	Nil
6	Edema	Nil	Nil	Nil
7	Washability	Washable	Washable	Washable
8	Spreadability	4 (gmc /sec)	5 (gmc/sec)	5 (gmc /sec)
9	Homogeneity	Good	Good	Good
10	pH	7.1	7.9	7.5
11	Viscosity	Moderate	Good	Good
12	Stability	25°C	25°C	25°C

In vitro studies

(I). In vitro antibacterial screening of Formulation

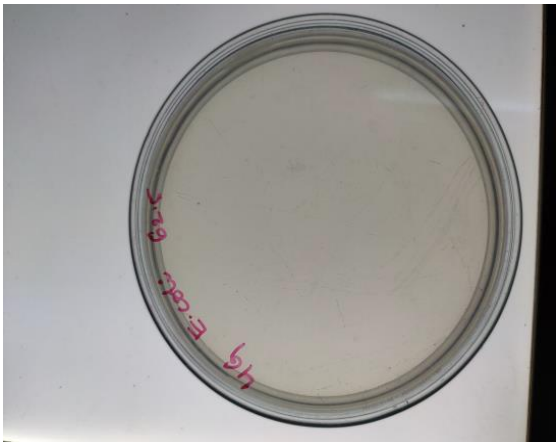
(i). Determination of minimum inhibitory concentration (MIC)

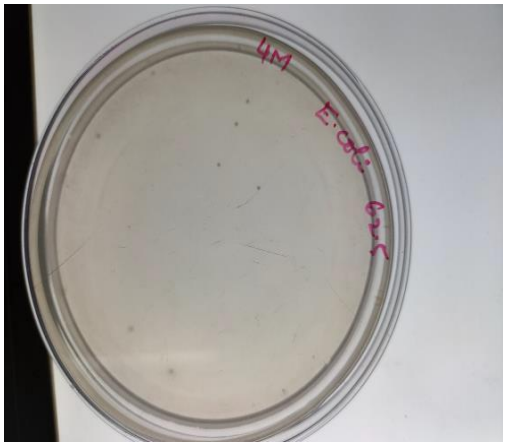
Antibacterial activity of formulations F1, F2 and F3 was evaluated by broth microdilution method (CLSI 2007) using Mueller Hinton medium (Hi-media). The MIC value of tested formulation is presented, with the standard drugs ciprofloxacin. formulations F1, F2 and F3 exhibited variable activity against the tested bacterial strains. Among all tested formulations F3 exhibited maximum activity against E.coli µM. Formulations were tested in the concentration range of 19.74 to 221.98 µM..The formulations F1, F2 and F3 we got 32 µM, 64 µM and 19.74 µM

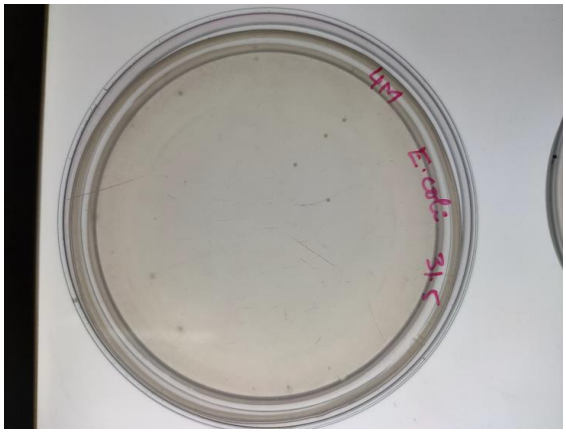
(ii) Determination of Minimum bactericidal concentration (MBC)

The Minimum Bactericidal Concentration (MBC) is the lowest concentration of an antibacterial agent required to kill a bacterium over a fixed, somewhat extended period, such as 18 hours or 24 hours, under a specific set of conditions. It can be determined from the broth dilution of MIC tests by sub-culturing to agar plates that do not contain the test agent. The MBC is identified by determining the lowest concentration of antibacterial agent that reduces the viability of the initial bacterial inoculum by a pre-determined reduction such as ≥99.9%. The MBCs were determined by sub-culturing aliquots (20 µl) from the bacterial suspension wells with no visible bacterial growth at concentrations greater than the defined MlC and from control wells onto nutrient agar plates. The plates were incubated aerobically at 37 °C for 24 hours and colonies were comparatively counted with the starting inoculum. MBC was determined

as the concentration at which a ≥99.9% decrease in bacterial counts (i.e., 3 log l0 reduction in cfu/ml) was achieved or the lowest concentrations that did not produce any bacterial growth in the agar plates were regarded as MBC values. All experiments were repeated in triplicate for each strain. The ratio MBC/MIC ≤2 indicates bactericidal activity, while MBC/MIC ratio ≥4 shows bacteriostatic activity oftest formulations MIC and MBC values (μM) of the tested formulation is F1, F2 and F3- 4, 4 and 2 respectively.







- Irritancy: First, we took a measurement of one centimeter squared for a small area on the back of the left hand. After the application of cream, we took note of the time. After up to a day, we looked to see whether there was any swelling or redness. Prepared formulation was no swelling or redness
- Packaging and Storage: Transfer the cream into sterilized jars or containers. Let it cool completely before sealing. Storage. Conditions. Active compounds in tulsi and betel leaves are highly sensitive to environmental factors.
- Temperature: Store in a cool, dry place. Temperatures should ideally range between 15° C and 25° C. For enhanced shelf life and to prevent natural oils from turning rancid, refrigeration at 4° C to 8° C is best.
- Light: Keep the product away from direct sunlight or intense indoor lighting, a s UV rays break down antioxidants and phytochemicals.

SUMMARY & CONCLUSION:

The herbal cream formulated with betel leaf and tulsi leaf proves to be an effective and stable product that combines the best of nature's ingredients for healthy, rejuvenated skin. Its moisturizing, soothing, and healing properties, along with the natural antimicrobial action of its active ingredients, make it an excellent choice for consumers seeking a natural alternative to conventional skincare products. The cream was made using a special method, and itsquality was checked by testing it. Considerations were made for the cream's texture, acidity, spreadability, skin-friendliness, ease of washing off, thickness, and cohesion.The betel and tulsi leaf herbal cream was successfully formulated using natural ingredients known for their medicinal and skin-protective properties. The cream showed good texture, spreadability, stability, and skin compatibility during evaluation. Both betel leaf and tulsi leaf extracts possess antimicrobial, which make the herbal cream beneficial for protecting and

nourishing the skin.

The preparation demonstrated that herbal ingredients can be effectively used as a safer and eco-friendly alternative to synthetic skin creams with fewer side effects. The study also highlighted the potential of traditional medicinal plants in modern cosmetic and pharmaceutical applications. Overall, the formulated herbal cream can be considered effective for maintaining healthy skin and may be useful for treating minor skin infections, irritation, dryness, and inflammation. Further clinical and stability studies can be carried out to improve and standardize the product for commercial use

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