

Formulation and Evaluation of Herbal Antimicrobial Basil Seed Gel Enriched with Tea Tree Oil

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Abstract

Skin infections and microbial contamination are common health concerns that require safe and effective topical treatments. The increasing use of synthetic antimicrobial agents has raised concerns regarding adverse effects and antimicrobial resistance, creating a need for natural alternatives. The present study was undertaken to formulate and evaluate a herbal antimicrobial gel enriched with basil seed gel (*Ocimum basilicum*) and tea tree oil (*Melaleuca alternifolia*) for topical application. Basil seed mucilage was extracted and utilized as a natural gelling agent, while tea tree oil was incorporated for its well-known antimicrobial properties. The gel was formulated using basil seed gel, tea tree oil, aloe vera gel, glycerin, carbopol 940, vitamin E, rose water, and suitable preservatives. The prepared formulation was evaluated for various physicochemical parameters, including appearance, pH, spreadability, viscosity, washability, phase separation, irritancy, and antimicrobial activity. The developed gel exhibited a transparent appearance, pleasant odor, smooth texture, and good consistency. The pH of the formulation was found to be 6.12, indicating compatibility with normal skin. The gel showed satisfactory spreadability, excellent washability, high viscosity, and no evidence of phase separation. Irritancy studies confirmed that the formulation was non-irritating and safe for topical use. Phytochemical screening revealed the presence of phenols, flavonoids, terpenes, and fatty acids, which contribute to its therapeutic activity. Antimicrobial evaluation demonstrated significant inhibitory activity against selected microorganisms. The results indicate that the formulated basil seed gel enriched with tea tree oil possesses desirable physicochemical characteristics, good stability, safety, and promising antimicrobial potential, suggesting its suitability as a natural topical antimicrobial formulation for skincare and wound-care applications.

Keywords: Basil Seed Gel, Tea Tree Oil, Herbal Gel, Antimicrobial Activity, Topical Formulation, *Ocimum basilicum*, *Melaleuca alternifolia*.

1. Introduction

1.1 Skin and Skin Disorders

The skin is the largest organ of the human body and serves as the primary barrier against physical,

chemical, and microbial insults. It performs several vital functions, including protection against pathogens, regulation of body temperature, prevention of excessive water loss, and sensory perception [1]. The skin consists of three major layers: epidermis, dermis, and hypodermis, each contributing to its structural integrity and physiological functions [2].

Skin infections caused by bacteria, fungi, and other microorganisms are among the most common dermatological disorders worldwide. Pathogens such as *Staphylococcus aureus*, *Escherichia coli*, and *Pseudomonas aeruginosa* are frequently associated with wound infections, dermatitis, and other skin conditions [3]. The increasing prevalence of microbial resistance and adverse effects associated with synthetic antimicrobial agents has highlighted the need for safer and more effective topical formulations [4]. Consequently, the development of herbal antimicrobial products has gained considerable attention due to their efficacy, biocompatibility, and reduced side effects.

1.2 Herbal Gels

Herbal gels are semisolid topical preparations containing plant-derived bioactive compounds incorporated into a suitable gel base. These formulations are widely used in cosmetic and pharmaceutical applications because of their ease of application, non-greasy nature, excellent spreadability, and enhanced patient compliance [5]. Herbal gels provide antimicrobial, anti-inflammatory, antioxidant, moisturizing, and wound-healing benefits while minimizing the risks associated with synthetic chemicals [6].

Compared with conventional synthetic formulations, herbal gels are generally safer, eco-friendly, cost-effective, and better tolerated by sensitive skin. The incorporation of herbal ingredients into gel systems can improve therapeutic efficacy by ensuring prolonged contact with the skin and controlled release of active constituents [7].

1.3 Basil Seed Gel

Basil seed gel is obtained from the seeds of *Ocimum basilicum* L., a medicinal plant belonging to the family Lamiaceae. When hydrated, basil seeds produce a mucilaginous polysaccharide-rich gel that exhibits excellent thickening, emulsifying, and water-retention properties [8]. Basil seed mucilage has attracted significant interest in pharmaceutical and cosmetic formulations due to its natural origin, biodegradability, and biocompatibility [9].



Fig No. 1 : Basil Seed Gel

Studies have reported that basil seed gel possesses antioxidant, soothing, moisturizing, and wound-healing properties. The mucilage forms a protective film over the skin surface, helping maintain hydration and supporting tissue repair processes [10]. Additionally, basil contains various bioactive constituents that contribute to its therapeutic potential, making it a promising natural gelling agent for topical drug delivery systems [11].

1.4 Tea Tree Oil

Tea tree oil is an essential oil obtained from the leaves of *Melaleuca alternifolia*, a plant native to Australia and belonging to the family Myrtaceae. The oil contains several bioactive terpenes,

Fig No. 2 : Tea Tree Oil



including terpinen-4-ol, α -terpineol, and γ -terpinene, which are primarily responsible for its antimicrobial and anti-inflammatory activities [12].

Tea tree oil has demonstrated broad-spectrum antimicrobial activity against a variety of Gram-positive and Gram-negative bacteria, fungi, and yeasts. It has been extensively used in the management of acne, wound infections, fungal diseases, and other dermatological disorders [13]. In addition to its antimicrobial effects, tea tree oil exhibits anti-inflammatory properties that help reduce erythema, irritation, and skin inflammation [14]. These characteristics make tea tree oil a valuable ingredient in topical pharmaceutical and cosmetic preparations.

1.5 Rationale of the Study

The emergence of antimicrobial resistance has become a major global health concern, reducing the effectiveness of conventional antibiotics and increasing the need for alternative therapeutic approaches [4]. Natural products have gained considerable attention as potential sources of novel antimicrobial agents because of their safety, accessibility, and diverse biological activities.

Basil seed gel and tea tree oil represent two promising natural ingredients with complementary properties. Basil seed mucilage acts as a natural gelling and moisturizing agent, while tea tree oil provides potent antimicrobial activity. The combination of these components is expected to produce a stable herbal formulation capable of delivering sustained antimicrobial effects while maintaining skin hydration and comfort [15]. Therefore, the present study was undertaken to formulate and evaluate an herbal antimicrobial basil seed gel enriched with tea tree oil for potential topical applications.

2. Aim and Objectives

2.1 Aim

The aim of the present study was to formulate and evaluate a herbal antimicrobial gel enriched with basil seed gel (*Ocimum basilicum*) and tea tree oil (*Melaleuca alternifolia*) for topical application. The formulation was designed to combine the natural gelling and moisturizing properties of basil seed mucilage with the potent antimicrobial activity of tea tree oil. The developed gel was intended to provide a safe, effective, and natural alternative to conventional topical antimicrobial preparations while improving skin hydration, protection, and overall skin health [16,17].

2.2 Objectives

- 1. To Extract Basil Seed Mucilage:** Basil seeds contain a mucilage-rich outer layer that swells upon hydration and forms a viscous gel. The objective was to extract this mucilage efficiently and utilize it as a natural gelling agent in the formulation. Basil seed mucilage possesses excellent water-holding capacity, biocompatibility, and film-forming properties, making it suitable for topical applications [18,19].
- 2. To Formulate Herbal Gel Using Basil Seed Gel and Tea Tree Oil:** The formulation was developed by incorporating basil seed gel, tea tree oil, aloe vera gel, glycerin, carbopol, vitamin E, and suitable excipients to obtain a stable and aesthetically acceptable herbal gel. The combination was selected to achieve moisturizing, soothing, antioxidant, and antimicrobial effects in a single topical formulation [20,21].
- 3. To Evaluate Physicochemical Properties:** The prepared gel was evaluated for various physicochemical characteristics such as appearance, color, odor, texture, pH, spreadability, washability, viscosity, and phase separation. These parameters are important for determining formulation quality, stability, and patient acceptability [22].
- 4. To Assess Antimicrobial Activity:** The antimicrobial efficacy of the herbal gel was evaluated against selected pathogenic microorganisms using agar diffusion techniques. The study aimed to determine the ability of tea tree oil incorporated within the basil seed gel matrix to inhibit microbial growth and provide effective protection against skin infections [23,24].
- 5. To Determine Safety and Skin Compatibility:** The safety profile of the formulation was assessed through irritancy testing and phytochemical evaluation. The objective was to ensure that the prepared gel is non-irritating, biocompatible, and suitable for regular topical application on human skin [25].

3. Materials and Methods

3.1 Materials

The materials used for the preparation of the herbal antimicrobial gel included basil seeds (*Ocimum basilicum*), tea tree oil (*Melaleuca alternifolia*), aloe vera gel, carbopol 940, glycerin, vitamin E oil, rose water, methyl paraben, propyl paraben, triethanolamine, and distilled water. Basil seeds served as a natural source of mucilage and acted as the primary gelling agent. Tea tree oil was incorporated as the active antimicrobial component due to its broad-spectrum antibacterial and antifungal properties. Aloe vera gel was added for its moisturizing and soothing effects on the skin. Carbopol 940 was used as a viscosity-enhancing and gel-forming polymer, while glycerin functioned as a humectant to maintain skin hydration. Vitamin E acted as an antioxidant, rose water imparted fragrance and skin-conditioning properties, and methyl paraben and propyl paraben were included as preservatives to enhance the stability and shelf life of the formulation. Triethanolamine was used for pH adjustment, and distilled water served as the vehicle for the formulation [26–30].

3.2 Equipment

The formulation and evaluation studies were carried out using standard laboratory equipment. A digital pH meter was used for pH determination, while viscosity measurements were performed using a Brookfield viscometer. An incubator was employed for microbial culture growth during antimicrobial studies. A laminar airflow chamber provided aseptic conditions for microbiological procedures, and a magnetic stirrer was used to ensure uniform mixing of ingredients during gel preparation [31].

3.3 Extraction of Basil Seed Gel

Basil seed gel was extracted from hydrated basil seeds through a simple extraction process. Initially, basil seeds were soaked in distilled water for 2–3 hours to allow swelling and mucilage formation. The soaked seeds were then boiled for approximately 6–8 minutes to facilitate complete release of the mucilage. The resulting mass was ground to obtain a smooth mucilaginous paste. Finally, the mixture was filtered through a suitable filter medium to separate the mucilage from seed residues and obtain a clear basil seed gel extract [32,33].

3.4 Formulation of Herbal Gel

Preparation of Gel Base

Carbopol 940 was dispersed in distilled water and stirred continuously until a homogeneous gel base was formed. Aloe vera gel and glycerin were subsequently added and mixed thoroughly to obtain a smooth gel matrix.

Incorporation of Active Ingredients

The extracted basil seed gel was gradually incorporated into the gel base under continuous stirring. Tea tree oil, vitamin E oil, and rose water were then added and mixed uniformly. Preservatives were dissolved separately and incorporated into the formulation.

pH Adjustment

The pH of the formulation was adjusted to the range of 5.5–6.5 using triethanolamine to ensure compatibility with normal skin and maintain formulation stability.

Final Gel Preparation

Continuous stirring was carried out until a smooth, homogeneous, and lump-free herbal gel was obtained. The prepared gel was then stored in suitable containers for further evaluation studies [34,35].

3.5 Formulation Composition

Table 1: Composition of Herbal Antimicrobial Basil Seed Gel

Sr. No.	Ingredient	Quantity	Function
1	Basil seeds (Sabja)	2 g	Natural gelling agent
2	Distilled water	80 mL	Vehicle
3	Aloe vera gel	10 g	Moisturizer and soothing agent
4	Tea tree oil	0.5 mL	Antimicrobial agent
5	Glycerin	5 mL	Humectant
6	Carbopol 940	0.5 g	Gel thickener
7	Triethanolamine	q.s.	pH adjustment
8	Vitamin E oil	0.5 mL	Antioxidant
9	Methyl paraben	0.2 g	Preservative
10	Propyl paraben	0.02 g	Preservative
11	Rose water	1 mL	Fragrance and skin conditioner

4. Evaluation of Herbal Gel

4.1 Organoleptic Evaluation

Organoleptic evaluation was carried out to assess the physical and sensory characteristics of the formulated herbal gel. Parameters such as colour, odour, texture, consistency, and overall appearance were visually examined. The prepared gel was observed for transparency, homogeneity, smoothness, and the

presence of any particulate matter. Organoleptic evaluation is an important quality control parameter that influences consumer acceptability and product aesthetics [36,37].

Parameters Evaluated

- Colour
- Odour
- Texture
- Consistency
- Physical appearance

4.2 pH Determination

The pH of the herbal gel was determined using a calibrated digital pH meter. Approximately 1 g of gel was dispersed in 10 mL of distilled water and mixed thoroughly to obtain a uniform solution. The electrode was immersed in the sample, and the pH was recorded. The pH of topical formulations should remain within the physiological skin pH range (5.5–6.5) to minimize irritation and maintain skin compatibility [38].

4.3 Washability Study

Washability was evaluated by applying a small quantity of gel onto the skin surface and observing the ease of removal with water. A formulation with good washability ensures convenience of use and improved patient compliance. The prepared herbal gel was assessed manually and categorized based on the ease of washing [39].

4.4 Spreadability Study

Spreadability determines the extent to which a gel can be easily spread on the skin surface. A known quantity of gel was placed between two glass slides, and a specified weight was applied. The time required for the upper slide to move a certain distance under the influence of the applied weight was recorded. Spreadability was calculated using the following equation:

$$S = (M \times L) / T$$

Where:

S = Spreadability (g·cm/sec)

M = Weight tied to upper slide (g)

L = Length moved by the glass slide (cm)

T = Time taken (sec)

A formulation with higher spreadability is preferred because it facilitates easy application and uniform distribution over the skin [40].

4.5 Phase Separation Study

Phase separation studies were conducted to evaluate the physical stability of the herbal gel. A sample of the formulation was placed on a transparent glass slide and observed visually for signs of separation, precipitation, aggregation, or instability. The absence of phase separation indicates good formulation stability and compatibility among ingredients [41].

4.6 Viscosity Measurement

Viscosity is a critical parameter affecting the application and stability of topical gels. The viscosity of the prepared gel was measured using a Brookfield viscometer equipped with a suitable spindle. Measurements were carried out at controlled temperature and rotational speed. Appropriate viscosity ensures good spreadability, retention at the site of application, and patient acceptability [42].

4.7 Irritancy Test

The irritancy test was performed to assess the safety of the herbal gel for topical use. A small quantity of the formulation was applied to a marked area on the dorsal surface of the hand of healthy volunteers. The application site was observed periodically for up to 24 hours for signs of erythema (redness), edema (swelling), itching, or any other adverse skin reactions. The absence of irritation indicates the suitability of the formulation for dermal application [43].

4.8 Preliminary Phytochemical Screening

Preliminary phytochemical screening was performed to identify the presence of bioactive constituents responsible for the therapeutic properties of the formulation.

Determination of Phenols

Phenolic compounds were identified using the Ferric Chloride Test. The development of a characteristic dark blue or green coloration indicated the presence of phenolic compounds [44].

Determination of Flavonoids

Flavonoids were detected using Thin Layer Chromatography (TLC) with a suitable solvent system. After development, the chromatographic plate was sprayed with aluminium chloride reagent and examined under ultraviolet light for fluorescent spots [45].

Determination of Terpenes

Terpenes were identified using TLC analysis with a hexane–ethyl acetate solvent system. Developed plates were treated with sulfuric acid reagent and visualized under UV light [46].

Determination of Fatty Acids

Fatty acids were analyzed using TLC with a hexane–ethyl acetate solvent system. Visualization was achieved by exposing the developed plate to iodine vapours, followed by UV examination [47].

4.9 Antimicrobial Activity

Agar Well Diffusion Method

The antimicrobial activity of the herbal gel was evaluated using the agar well diffusion method. Sterile nutrient agar medium was prepared, sterilized, and poured into Petri plates under aseptic conditions. After solidification, the microbial cultures were uniformly inoculated onto the agar surface. Wells of approximately 4 mm diameter were created in the agar medium, and equal quantities of the gel formulation were introduced into each well. The plates were incubated at 37°C for 24–48 hours. Following incubation, the diameter of the zone of inhibition surrounding each well was measured in millimetres. Larger zones of inhibition indicated stronger antimicrobial activity [48,49].

Test Microorganisms

The antimicrobial efficacy of the formulation was assessed against the following microorganisms:

- *Staphylococcus aureus* (Gram-positive bacterium)
- *Escherichia coli* (Gram-negative bacterium)
- *Bacillus subtilis* (Gram-positive bacterium)
- *Pseudomonas aeruginosa* (Gram-negative bacterium)

These microorganisms were selected because they are commonly associated with skin infections, wound contamination, and opportunistic microbial infections [50].

5. Results

The formulated herbal antimicrobial basil seed gel enriched with tea tree oil was evaluated for organoleptic characteristics, physicochemical properties, irritancy potential, phytochemical constituents, and ant-

imicrobial activity. The results obtained from the evaluation studies are presented below.

5.1 Organoleptic Properties

The prepared herbal gel exhibited desirable organoleptic characteristics with a transparent appearance, pleasant odour, smooth texture, and good consistency, indicating acceptable aesthetic quality and patient compliance.

Table 2: Organoleptic Evaluation of Herbal Gel

Parameter	Result
Colour	Transparent
Odour	Pleasant
Texture	Smooth
Consistency	Good
State	Semi-solid

The formulation remained homogeneous throughout the study period and did not exhibit any visible signs of instability or aggregation.

5.2 Physicochemical Evaluation

The physicochemical properties of the formulated gel were evaluated to determine its suitability for topical application.

Table 3: Physicochemical Evaluation of Herbal Gel

Parameter	Result
pH	6.05–6.12
Washability	Easily washable
Spreadability	4–5 cm/g
Phase Separation	Absent
Viscosity	9348–9365 cps

The pH values were found to be within the normal physiological range of skin, suggesting good compatibility. The formulation exhibited satisfactory spreadability, excellent washability, and high viscosity without any evidence of phase separation, indicating good physical stability.

5.3 Irritancy Study

The irritancy study was performed to assess the dermal safety of the prepared gel formulation.

Table 4: Irritancy Study of Herbal Gel

Sr. No.	Time (Hours)	Irritant Effect	Erythema (Redness)	Edema (Swelling)
1	4	No	No	No
2	8	No	No	No
3	12	No	No	No
4	16	No	No	No
5	20	No	No	No
6	24	No	No	No

No erythema, edema, itching, or other adverse skin reactions were observed during the study period, indicating that the formulation is safe and non-irritating for topical use.

5.4 Phytochemical Screening

Preliminary phytochemical screening confirmed the presence of important bioactive constituents responsible for the therapeutic activity of the formulation.

Table 5: Preliminary Phytochemical Screening

Phytochemical Constituent	Result
Phenols	Positive
Flavonoids	Positive
Terpenes	Positive
Fatty Acids	Positive

The presence of phenols and flavonoids contributes to antioxidant activity, while terpenes and fatty acids are associated with antimicrobial, anti-inflammatory, and skin-protective properties.

5.5 Antimicrobial Activity

The antimicrobial activity of the formulated gel was evaluated using the agar well diffusion method against selected pathogenic microorganisms.

Table 6: Test Microorganisms Used for Antimicrobial Evaluation

Sr. No.	Microorganism	Type
1	<i>Staphylococcus aureus</i>	Gram-positive
2	<i>Escherichia coli</i>	Gram-negative
3	<i>Bacillus subtilis</i>	Gram-positive
4	<i>Pseudomonas aeruginosa</i>	Gram-negative

The formulation produced clear zones of inhibition against the tested microorganisms, indicating effective antimicrobial activity. The antimicrobial effect is primarily attributed to the presence of tea tree oil and bioactive phytoconstituents present in basil seed gel. The results demonstrate the potential of the formulation as a natural topical antimicrobial preparation for skincare and wound management applications.

6. Future Scope

The developed herbal antimicrobial basil seed gel enriched with tea tree oil demonstrated promising physicochemical properties, safety, and antimicrobial activity. However, further research and development are necessary to establish its long-term effectiveness, stability, and commercial viability. The following areas represent potential future directions for the advancement of this formulation.

6.1 Stability Studies According to ICH Guidelines

Comprehensive stability studies should be conducted in accordance with the International Council for Harmonisation (ICH) guidelines to evaluate the physical, chemical, microbiological, and therapeutic stability of the formulation under different storage conditions. Parameters such as pH, viscosity, colour, odour, phase separation, microbial load, and active constituent retention should be monitored over time.

Stability studies will help determine the shelf life and appropriate storage conditions of the herbal gel [61,62].

6.2 Clinical Evaluation in Human Volunteers

Although preliminary irritancy and antimicrobial studies demonstrated favorable results, controlled clinical studies involving human volunteers are required to confirm the safety, efficacy, and dermatological acceptability of the formulation. Clinical evaluation can provide scientific evidence regarding wound healing potential, antimicrobial effectiveness, skin hydration, and long-term user compliance [63].

6.3 Scale-Up and Commercialization

The formulation can be further optimized for industrial-scale manufacturing. Process validation, quality assurance, packaging compatibility studies, and regulatory compliance should be investigated to facilitate commercial production. Large-scale manufacturing may increase product availability while maintaining consistency, efficacy, and safety [64].

6.4 Incorporation of Additional Herbal Actives

The therapeutic effectiveness of the formulation may be enhanced through the incorporation of additional herbal ingredients possessing antimicrobial, antioxidant, anti-inflammatory, and wound-healing properties. Natural extracts such as *Azadirachta indica* (Neem), *Curcuma longa* (Turmeric), *Centella asiatica*, and *Calendula officinalis* may provide synergistic benefits and broaden the spectrum of biological activity [65,66].

6.5 Development of Nano-Herbal Gel Systems

Recent advances in nanotechnology have enabled the development of nano-herbal gel systems that improve the delivery and bioavailability of herbal active constituents. Nanoemulsions, liposomes, phytosomes, and nanostructured lipid carriers can enhance skin penetration, controlled release, stability, and therapeutic efficacy of tea tree oil and other phytoconstituents. Future research may focus on developing nano-herbal gel formulations to achieve superior antimicrobial activity and improved patient outcomes [67,68].

Overall, the formulated basil seed gel enriched with tea tree oil possesses significant potential for further pharmaceutical and cosmeceutical development. Continued research in these areas may contribute to the development of a safe, effective, and commercially viable natural topical antimicrobial product.

Conclusion

The present study successfully formulated and evaluated a herbal antimicrobial gel enriched with basil seed gel (*Ocimum basilicum*) and tea tree oil (*Melaleuca alternifolia*). The developed formulation exhibited desirable physicochemical characteristics, including suitable pH, good consistency, smooth texture, satisfactory spreadability, high viscosity, and excellent washability, making it appropriate for topical application. The absence of phase separation indicated good formulation stability, while irritancy studies confirmed its safety and skin compatibility.

Preliminary phytochemical screening revealed the presence of important bioactive constituents such as phenols, flavonoids, terpenes, and fatty acids, which contribute to the therapeutic potential of the formulation. The antimicrobial evaluation demonstrated effective inhibitory activity against common skin pathogens, indicating the ability of the gel to prevent microbial growth and support skin health. The combined action of basil seed mucilage and tea tree oil provided both moisturizing and antimicrobial benefits, enhancing the overall effectiveness of the formulation.

The use of natural ingredients offers several advantages over conventional synthetic antimicrobial products, including reduced risk of adverse effects, improved biocompatibility, and better patient acceptability. Therefore, the formulated herbal gel may serve as a promising natural alternative for topical antimicrobial therapy and skincare applications.

In conclusion, the developed basil seed gel enriched with tea tree oil showed satisfactory quality, safety, and antimicrobial efficacy. However, further studies involving long-term stability testing, advanced characterization, and clinical evaluation in human subjects are recommended to establish its therapeutic effectiveness and commercial potential on a larger scale.

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