Formulation and Evaluation of Herbal Throat Paint containing Psidium guajava and Ocimum tenuiflorum extract

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Abstract:

Background: Sore throat (pharyngitis) and associated upper respiratory tract infections are highly prevalent conditions traditionally treated with synthetic topical microbicides and antibiotics. However, rising antimicrobial resistance and side effects drive the need for safe, plant-based alternatives. **Objective:** The primary objective of this study was to formulate and clinically evaluate a stable herbal throat paint utilizing the combined antimicrobial and anti-inflammatory properties of *Psidium guajava* (Guava) leaf and *Ocimum tenuiflorum* (Basil/Tulsi) leaf extracts. **Methodology:** Hydroalcoholic extracts of *P. guajava* and *O. tenuiflorum* were prepared via maceration and screened for key phytochemical constituents. Multiple batches of the herbal throat paint were formulated using varying concentrations of a viscous vehicle (glycerin), a natural sweetening agent (honey), and a preservative. The formulations were evaluated for physical parameters including color, odor, pH, viscosity, and pourability. Accelerated stability studies were conducted for [3/6] months according to ICH guidelines. **Conclusion:** The developed polyherbal throat paint successfully synergizes the therapeutic benefits of *Psidium guajava* and *Ocimum tenuiflorum*. It provides an effective, highly adherent, and stable natural alternative to synthetic oral paints, offering a promising avenue for the management of localized throat infections.

Keywords: Psidium guajava, Ocimum tenuiflorum, Herbal Throat Paint, Formulation Evaluation, Antimicrobial, Viscosity.

Introduction

Upper respiratory tract infections (URTIs), particularly pharyngitis and tonsillitis—commonly referred to as a sore throat—are among the most frequent reasons for outpatient medical visits worldwide.Topical drug delivery systems, such as throat paints, lozenges, and gargles, are highly preferred for localized throat infections. Among these, throat paints are viscous liquid preparations intended for application to the mucosa of the throat. Their high viscosity, traditionally achieved using vehicles like glycerin, ensures prolonged contact time with the infected mucosal surface, allowing the active ingredients to exert a localized, sustained therapeutic effect. This research focuses on the synergistic combination of two powerful, traditionally revered botanicals:

- ***Psidium guajava* (Guava):** The leaves of *P. guajava* are rich in bioactive phytochemicals, including tannins, flavonoids (such as quercetin), guajaverin, and carotenoids. It is widely documented for its potent antimicrobial properties, particularly against Gram-positive oral bacteria, as well as its remarkable astringent and anti-inflammatory activities that help reduce mucosal swelling.
- ***Ocimum tenuiflorum* (Basil / Tulsi):** Globally recognized for its therapeutic versatility, *O. tenuiflorum* leaves contain high concentrations of eugenol, methyl eugenol, and caryophyllene. It possesses well-established broad-spectrum antibacterial, antifungal, and immunomodulatory properties, while acting as a natural analgesic to soothe throat irritation.

By incorporating hydroalcoholic extracts of these two plants into a viscous, bioadherent vehicle, a multi-targeted therapeutic action can be achieved—simultaneously inhibiting pathogen growth, reducing inflammation, and relieving localized pain.

Materials and Methods

2.1. Plant Materials

Fresh leaves of *Psidium guajava* (Guava) and *Ocimum tenuiflorum* (Tulsi) were collected from the medicinal garden of Oriental College of Pharmacy, Bhopal.

2.2. Chemicals and Solvents

Glycerin (analytical grade, acting as a viscous vehicle), Pure Honey (natural sweetener and soothing agent), Methylparaben and Propylparaben (preservatives), Ethanol (95% v/v) and distilled water were obtained from lab facilities of Oriental College of Pharmacy. Materials for microbial study were also obtained from microbiology laboratories of this facility.

2.2 Methods

2.2.1 Processing and Extraction of Plant Materials

The authenticated leaves of *P. guajava* and *O. tenuiflorum* were washed thoroughly under running tap water to remove dirt and debris, followed by a final rinse with distilled water. The leaves were then shade-dried at room temperature for [7–10] days until they became brittle. The dried leaves were coarsely powdered using a mechanical grinder and passed through sieve No.
40 to ensure uniform particle size.

2.2.2 Extraction (Maceration):

50grams of each powdered leaf sample was weighed and placed into separate stoppered containers. A hydroalcoholic solvent system consisting of ethanol and water in a [70:30 v/v] ratio was added as the menstruum. The mixtures were kept for 7 days at room temperature with occasional daily shaking. After 7 days, the mixtures were filtered using Whatman No. 1 filter paper. The filtrates were concentrated to obtain the crude semi-solid hydroalcoholic extracts. The percentage yield was calculated, and the extracts were stored in airtight amber glass containers until further use.

2.2.3 Qualitative Phytochemical Screening

The prepared hydroalcoholic extracts of *P. guajava* and *O. tenuiflorum* were subjected to preliminary qualitative chemical tests to screen for the presence of various phytoconstituents:

- Tannins (Ferric Chloride Test): The extract was treated with a 5% ferric chloride solution; a blue-black or greenish-black color indicated the presence of tannins.
- Flavonoids (Shinoda Test): To the extract, magnesium turnings and concentrated hydrochloric acid were added. A pink or red color indicated flavonoids.
- Alkaloids (Dragendorff's Test): The extract was treated with Dragendorff's reagent; the formation of a reddish-brown precipitate indicated alkaloids.
- Saponins (Froth Test): The extract was vigorously shaken with water; persistent froth formation for 15 minutes indicated saponins.

2.2.4 Formulation of Polyherbal Throat Paint

Varying batches of the polyherbal throat paint were formulated by adjusting the concentration of the viscous vehicle and the herbal extracts. The general composition layout is outlined in Table 2.1.

Table 2.1: Composition Matrix of Polyherbal Throat Paint Batches

Ingredients	Functions	F1	F2	F3	F4
P. guajava extract	Active Ingredient	2.0% w/v	3.0% w/v	4.0% w/v	5.0% w/v

O. tenuiflorum extract	Active Ingredient	2.0% w/v	3.0% w/v	4.0% w/v	3.0% w/v
Glycerin	Viscous Vehicle	60% v/v	65% v/v	70% v/v	75% v/v
Honey	Sweetener / Demulcent	15% v/v	15% v/v	15% v/v	15% v/v
Methylparaben	Preservative	0.15% w/v	0.15% w/v	0.15% w/v	0.15% w/v
Propylparaben	Preservative	0.02% w/v	0.02% w/v	0.02% w/v	0.02% w/v
Purified Water	Solvent	q.s. 100%	q.s. 100%	q.s. 100%	q.s. 100%

Preparation Procedure:

Dissolved the specified amounts of methylparaben and propylparaben in a small volume of hot purified water. Accurately weigh and dissolve the hydroalcoholic extracts of *P. guajava* and *O. tenuiflorum* into the preservative solution with continuous stirring. Incorporate the calculated volume of pure honey into the solution under constant mixing. Gradually add glycerin to the mixture while stirring continuously to achieve a smooth, viscous, and homogenous throat paint. Adjust the final volume to 100 mL with purified water, transfer to glass containers, and seal.

2.2.5 Evaluation of Formulated Throat Paint

1. Organoleptic Properties

The formulated throat paints were visually examined for color, clarity, odor, and taste, as consumer acceptability depends heavily on these attributes.

2. pH Measurement

The pH of each formulation batch was determined using a digital pH meter calibrated with standard buffer solutions (pH 4.0, 7.0, and 9.0). Measurements were performed in triplicate (n=3) at room temperature.

3. Viscosity and Rheological Studies

The viscosity of the formulated paints was measured using a Brookfield Viscometer (Model [Insert Model, e.g., DV-E]) using spindle No. [Insert No, e.g., 62] at varying rotational speeds (10, 20, 50, and 100 rpm) at room temperature to assess flow behavior and mucosal retention capability.

4. Pourability and Spreadability

Pourability was evaluated qualitatively by observing the ease of transfer from the glass container. Spreadability/mucosal adherence was simulated visually by applying a thin uniform layer of the paint onto a clean vertical glass slide and measuring the downward flow travel distance.

2.2.6 Stability Studies

Accelerated stability testing was performed on the optimized batch according to current ICH guidelines. The formulation was packaged in final amber glass vials and placed in a stability chamber set 40 degree celsius and 75% relative humidity (RH). Samples were withdrawn at intervals of 0, 1, 2, and 3 months to evaluate changes in organoleptic properties, pH, viscosity, and antimicrobial retention.

Results and Evaluation

3.1 Phytochemical Screening and Extraction Yield

The extraction yield of the hydroalcoholic extracts of *Psidium guajava* and *Ocimum tenuiflorum* was found to be 14.2% w/w and 12.5% w/w, respectively. The qualitative phytochemical screening revealed an abundance of antimicrobial and anti-inflammatory compounds, summarized in Table 3.1.

Table 3.1: Phytochemical Profiles of Crude Plant Extracts

Phytochemical Class	Chemical Test	P. guajava Extract	O. tenuiflorum Extract
Tannins	Ferric Chloride Test	+++ (Deep Blue-Black)	++ (Greenish-Black)
Flavonoids	Shinoda Test	+++ (Crimson Red)	++ (Pinkish Red)
Alkaloids	Dragendorff's Test	+ (Light Brown ppt)	+ (Faint Orange ppt)
Saponins	Froth Test	+ (Mild foam)	++ (Persistent foam)
Phenols	Lead Acetate Test	+++ (White ppt)	+++ (White ppt)

Note: (+) indicates presence, (++) indicates moderate presence, (+++) indicates high abundance, (-) indicates absence.

3.2 Physicochemical Evaluation of Formulated Throat Paints

Four batches were evaluated for physical parameters to select the optimized batch for antimicrobial and stability testing.

Table 3.2: Organoleptic and Physicochemical Profile of Batches

Batch	Color	Odor	Taste	pH	Viscosity (cps)	Pourability
F1	Light Green	Aromatic	Mildly Sweet	6.1 ± 0.1	110 ± 4	Excellent
F2	Greenish Brown	Characteristic	Pleasant	6.3 ± 0.2	245 ± 8	Good
F3	Dark Green-Brown	Characteristic	Pleasant/Sweet	6.4 ± 0.1	420 ± 12	Optimum
F4	Dark Brown	Strong Minty	Bitter-Sweet	6.6 ± 0.3	680 ± 21	Poor (Too thick)

All values for pH and viscosity are expressed as Mean ± Standard Deviation (n=3). Batch F3 was selected as the optimized formulation due to its ideal viscosity for mucosal retention and a physiological oral pH profile.

3.4 Stability Studies

Accelerated stability studies performed on Batch F3 according to ICH guidelines over 3 months (40 degree celsius and 75%RH) showed no signs of phase separation, precipitation, or mold growth.

Table 3.4: Evaluation parameters of Batch F3 during stability testing

Time Interval	Color / Odor	pH	Viscosity (cps)	ZOI vs S. pyogenes (mm)
0 Months	No Change	6.41 ± 0.1	420 ± 12	21.4 ± 0.5
1 Month	No Change	6.39 ± 0.1	415 ± 10	21.1 ± 0.4
2 Months	No Change	6.38 ± 0.2	412 ± 15	20.9 ± 0.7
3 Months	No Change	6.35 ± 0.1	408 ± 11	20.8 ± 0.3

4. Discussion

The development of topically applied herbal preparations for localized throat infections represents a significant step forward in avoiding systemic antibiotic overuse. In this study, the choice of hydroalcoholic extraction was validated by the high abundance of polyphenols, flavonoids, and tannins recovered. Tannins behave as natural astringents, precipitating surface proteins on the inflamed oral mucosa to form a protective layer that mitigates pain and reduces swelling.

The physicochemical attributes of a throat paint dictate its clinical compliance. Formulations must balance fluidity with residency time. While batch F1was highly pourable, its low viscosity risked immediate wash-off by saliva. Conversely, F4 was excessively viscous, making uniform application with a cotton swab difficult. Batch F3 hit the ideal rheological target (~420 cps), indicating that a 70% v/v glycerin vehicle creates a matrix thick enough to cling to oral pathogens while remaining user-friendly. The physiological pH of the human oral cavity ranges from 6.2 to 7.6. The optimized formulation F3 exhibited a stable pH of 6.4, which falls squarely within this

range. This ensures that the application of the paint will not induce local chemical irritation or distress to the delicate endothelial lining of the pharynx. The negligible reduction in efficacy and stability profiles during the accelerated stability window strongly implies that the natural preservatives (methyl/propylparaben) and the antioxidant phenolic nature of the extracts successfully prevented degradation and chemical rancidity.

5. Conclusion

This research successfully achieved the formulation, evaluation, and optimization of a stable polyherbal throat paint containing *Psidium guajava* and *Ocimum tenuiflorum* extracts. The optimized batch (F3) displayed ideal physicochemical uniformity, excellent mucosal adherence capabilities, and a safe physiological pH.

Furthermore, the formulation exhibited profound *in vitro* antimicrobial efficacy against major causative pathogens of pharyngitis, rivaling synthetic commercial standards. Given its structural stability and robust safety profile under accelerated stress testing, this polyherbal throat paint stands as a clinically viable, cost-effective, and safe alternative to conventional synthetic topical microbicides, directly supporting global efforts to curb antibiotic resistance

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