



**INTERNATIONAL JOURNAL OF
PHARMACEUTICAL SCIENCES**
[ISSN: 0975-4725; CODEN(USA): IJPS00]
Journal Homepage: <https://www.ijpsjournal.com>



Research Article

Formulation And Evaluation of a Synergistic Hard-Candy Lozenge Containing Psidium Guajava and Glycyrrhiza Glabra

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ARTICLE INFO

Published: 11 July. 2026

Keywords:

Psidium guajava,
Glycyrrhiza glabra,
Phytochemical Synergy,
Oropharyngeal
Inflammation

DOI:

10.5281/zenodo.21308013

ABSTRACT

The management of upper respiratory tract infections (URTIs) and localized oropharyngeal inflammatory conditions has transitioned toward a preference for natural therapeutic alternatives that offer comparable efficacy to synthetic agents while mitigating the risk of antimicrobial resistance. This research delineates the development of a novel polyherbal hard-candy lozenge incorporating concentrated extracts of *Psidium guajava* (Guava) and *Glycyrrhiza glabra* (Liquorice/Mulethi). The formulation leverages the synergistic antimicrobial potency of guava tannins and flavonoids with the demulcent and expectorant actions of glycyrrhizin. Extraction protocols were optimized using hydroethanolic solvent systems (70% ethanol for guava and 30-50% ethanol for liquorice) followed by concentration to a "soft" extract consistency. The lozenges were fabricated using a sugar-glass matrix processed to the hard-crack stage (145 °C to 155°C), with moisture content strictly maintained between 0.5% and 2.0% to ensure kinetic stability. Physicochemical evaluations according to USP/IP standards revealed high batch uniformity, with average weight variation within $\pm 5\%$, hardness exceeding 7 kg/cm² and friability less than 1.0%. In-vitro dissolution studies in phosphate buffer (pH 6.8) demonstrated sustained erosion over 5 to 10 minutes, facilitating prolonged mucosal contact. This research provides a robust framework to produce commercially viable, high-quality herbal lozenges for oropharyngeal health.

INTRODUCTION

The oropharyngeal region serves as the primary portal for numerous respiratory pathogens and is frequently subject to localized inflammatory conditions such as pharyngitis, laryngitis, and

aphthous stomatitis. Traditional management often relies on synthetic antiseptic lozenges containing agents like hexylresorcinol or benzocaine. However, these are increasingly associated with local mucosal irritation and the promotion of multidrug-resistant bacterial strains.

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Relevant conflicts of interest/financial disclosures: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.



Medicated lozenges are solid unit-dosage forms designed for slow dissolution in the oral cavity, providing sustained release of active ingredients to the mucosal surfaces. This makes them an ideal vehicle for plant-derived bioactive compounds that offer localized therapeutic action with high patient acceptability.

Synergistic Mechanisms and Theoretical Framework

The structural design of this formulation is based on the functional synergy achieved by combining the antimicrobial load of guava leaves with the demulcent and masking properties of liquorice. Synergy in phytotherapy occurs when the combination of discrete agents produces a cumulative effect that exceeds the sum of the distinct effects.

The primary mechanism of the anticipated synergy centers on the enhancement of bacterial membrane permeability. Saponins like glycyrrhizin are amphiphilic molecules that interact with the lipid bilayer of the bacterial cell wall, increasing its fluidity and creating pores. This surfactant-like action is highly relevant as it facilitates the entry of other phytochemicals, such as guava flavonoids and tannins, into the bacterial cytoplasm where they can disrupt enzymatic activity and protein synthesis more effectively. Secondary literature on

related polyherbal blends suggests that when these specific phytochemical families are combined, they produce an expanded spectrum of inhibition against common pathogens like *E. coli* and *S. aureus* compared to isolated extracts.

Furthermore, guava leaf extracts possess an inherently bitter and astringent taste due to high tannin content, which can lower patient compliance. Glycyrrhiza glabra serves as an ideal corrective agent in this regard. The intense sweetness of glycyrrhizin masks the bitterness without the necessity for high-calorie sugar loading or synthetic sweeteners. This ensures high patient acceptability, which is highly relevant because the lozenge must be held in the mouth for extended periods to maximize mucosal contact.

Literature review

The success of a polyherbal formulation is fundamentally dependent on the deep understanding of the botanical and chemical properties of its constituent plants. For this research project, *Psidium guajava* and *Glycyrrhiza glabra* have been selected due to their complementary pharmacological activities and historical significance in traditional medicine systems globally.

Phytochemical Analysis of *Psidium guajava*



Psidium guajava, a member of the Myrtaceae family, is a perennial shrub or small tree widely distributed across tropical and subtropical regions, including India, South America, and parts of Africa.¹ While the fruit is a staple food crop, the leaves are of paramount importance in ethnopharmacology. Guava leaves are evergreen, leathery, and characterized by an opposite arrangement with tiny petioles.¹ They contain a diverse array of secondary metabolites, including tannins, flavonoids, triterpenoids, and essential oils.

The antimicrobial and astringent properties of the leaf are primarily attributed to its high concentration of polyphenolic compounds. Quantitative analysis reveals that the total phenolic content in dry guava extracts can reach levels as high as 17.02 mg/g, while the total tannin content

is approximately 14.09 mg of tannic acid equivalents per gram of dry extract.⁵ These tannins are vital for the lozenge's efficacy as they facilitate the precipitation of surface proteins on inflamed mucosal membranes, thereby creating a protective barrier and reducing the permeability of blood vessels—a classic astringent action.

Among the flavonoids, quercetin and its various glycosidic derivatives, such as guaijaverin and isoquercetin, are the most significant.² Quercetin (C₁₅ H₁₀ O₇) serves as a powerful antioxidant and spasmolytic agent, reducing the smooth muscle contractions in the throat that lead to the sensation of irritation.⁸ HPLC-based quantification of Egyptian and Indian guava leaf samples shows that quercetin levels vary by region and extraction method, typically ranging between 0.181% and 0.393% in the dry leaf powder.

Table 1: Taxonomic Hierarchy And Classification Detail Of *Psidium Guajava*

Taxonomic Hierarchy	Classification Detail
Kingdom	Plantae
Order	Myrtales
Family	Myrtaceae
Genus	<i>Psidium</i>
Species	<i>P. guajava</i> L. ¹

Phytochemical Foundations of *Glycyrrhiza glabra*



Glycyrrhiza glabra, belonging to the Fabaceae family, is perhaps best known for its sweet-tasting roots and rhizomes, which have been used for over 4,000 years in systems such as Ayurveda and Traditional Chinese Medicine.¹³ The primary active constituent is glycyrrhizin (glycyrrhizic acid), a triterpenoid saponin that provides the root its characteristic sweetness—estimated to be 30 to 50 times that of sucrose.

Glycyrrhizin (C₄₂ H₆₂ O₁₆) functions as a potent demulcent and expectorant.¹³ As a demulcent, it forms a soothing film over the pharyngeal mucosa,

protecting it from mechanical irritation and reducing the frequency of unproductive coughs.¹³ Its expectorant action is mediated by the acceleration of tracheal mucus secretion, helping to thin and expel congestion from the upper respiratory tract.¹³ Additionally, flavonoids such as liquiritin and isoliquiritigenin contribute to the plant's antitussive and anti-inflammatory effects.¹³ In the context of the formulation, glycyrrhizin also acts as a natural flavour enhancer and "sugar doctor," increasing the sweetness of the product without the necessity for synthetic additives.

Table 2: Bioactive Category, Principal Compounds, And Pharmacological Significance Of Glycyrrhiza Glabra

Bioactive Category	Principal Compounds	Pharmacological Significance
Saponins	Glycyrrhizin, Glycyrrhetic acid	Demulcent, Expectorant, Sweetener ¹³
Flavonoids	Liquiritin, Isoliquiritigenin, Rutin	Antitussive, Anti-inflammatory ¹³
Isoflavanes	Glabridin, Glabrene	Antimicrobial, Antioxidant ¹³
Chalcones	Licochalcones A, B, C, D	Anti-inflammatory, Anti-tumor ¹³

Research Objectives

The core objective of this study is to formulate and evaluate a synergistic hard-candy lozenge for the management of sore throats and mouth ulcers. The project is structured around the following specific aims:

1. To isolate bioactive extracts from *Psidium guajava* leaves and *Glycyrrhiza glabra* roots using optimized hydroethanolic extraction methods.
2. To design a stable lozenge base utilizing a precise ratio of sucrose and liquid glucose to achieve a kinetically stable glassy state.
3. To integrate the concentrated herbal extracts into the candy base at critical temperatures that preserve the integrity of heat-labile flavonoids.
4. To evaluate the formulated lozenges for pharmacopeial parameters including weight variation, hardness, friability, moisture content, and in-vitro dissolution.
5. To establish the theoretical relevance and mechanistic rationale for the antimicrobial and anti-inflammatory synergy of the polyherbal blend based on established literature profiles.

MATERIALS AND METHODS

Optimized Soxhlet Extraction Parameters



For this formulation, a hydroethanolic solvent system (Ethanol: Water) is employed. Ethanol is chosen for its ability to dissolve flavonoids and

terpenoids, while water helps in the extraction of polar saponins and tannins.²

Guava Leaf Extraction (Soxhlet Method)



Soxhlet extraction is the most effective method for recovering the highest percentage yield and antioxidant activity from *Psidium guajava*.

- **Quantity:** Weigh 50 g of dried guava leaf powder.
- **Solvent Selection:** Use 500 mL of 70% ethanol (Ethanol: Water in a 70:30 ratio). This concentration is proven to be the most effective for capturing active phenolic and flavonoid molecules.
- **Procedure:**
 1. Place the 50 g of powder in an extraction thimble.
 2. Add the 500 mL solvent to the round-bottom flask (1:10 solid-to-liquid ratio).
 3. Heat the apparatus to maintain a temperature of approximately 45°C to 60°C.
 4. Continue the extraction for 8 hours or until 6 to 8 siphoning cycles are completed.

5. Filter the resulting liquid through Whatman No. 1 filter paper.²¹
- **Concentration:** The resulting filtrate is concentrated using a water bath at 50°C–60°C to remove the solvent. This process results in a thick, "soft" extract, which is significantly more potent than the raw powder, allowing for a smaller volume of active material to achieve the desired therapeutic effect in the 100g batch.²¹

Liquorice Extraction (Optimized Hydroalcoholic Method)

For *Glycyrrhiza glabra*, the optimum extraction of glycyrrhizic acid and glabridin is achieved using a higher water-to-ethanol ratio.

- **Quantity:** Weigh 20 g of liquorice root powder.



- **Solvent Selection:** Use a 30:70 Ethanol: Water (v/v) mixture. Research indicates this specific ratio is optimal for maximum recovery of glycyrrhizic acid.
- **Procedure:**
 1. Mix the 20 g of powder with 100 mL of the 30:70 solvent.
 2. Heat the mixture to exactly 50°C. The amount of extracted actives increases rapidly up to 50°C but remains constant at higher temperatures.
 3. Maintain this temperature and stir for 60 minutes (dipping/maceration time).
 4. Alternatively, use ultrasound-assisted extraction at 60 degrees Celsius for 34 minutes with 57% methanol for a reported 3.414% yield of glycyrrhizic acid.
 5. Filter the extract through Whatman filter paper No. 4.
- **Concentration:** The resulting filtrate is concentrated using a water bath at 50°C–60°C to remove the solvent.² This process results in a thick, "soft" extract, which is significantly more potent than the raw powder, allowing for a smaller volume of active material to achieve the desired therapeutic effect in the 100g batch.²¹

Table 3: Optimized Extraction Parameters For Botanical Actives

Parameter	Optimized Value for Guava	Optimized Value for Liquorice
Extraction Solvent	70% Ethanol	30%–50% Ethanol
Temperature	60°C (Sonication/Soxhlet)	50°C
Extraction Time	35–60 Minutes	60–240 Minutes
Particle Size	Sieve No. 40–80	Sieve No. 40–60

The Synergistic Formulation

Table 4: Synergistic Polyherbal Lozenge Quantitative Composition (100g Batch)

Ingredient	Quantity	Pharmacological/Functional Role
Guava Leaf Extract (concentrated)	1.5 ml	Primary Active Ingredient: Antimicrobial, Astringent
Mulethi (Liquorice) Extract	2.5 ml	Primary Active Ingredient: Demulcent, Sweetener, Expectorant
Sucrose (Refined Sugar)	60.0 g	Bulking Agent: Provides the amorphous matrix
Liquid Glucose (Corn Syrup)	30.0 ml	Doctoring Agent: Inhibits sucrose crystallization
Honey (Pure)	4.0 ml	Texture Modifier: Enhances soothing effect and palatability
Citric Acid	0.5 g	Acidulant: Regulates pH and flavour profile.



Menthol (Optional)	0.1 ml	Sensory Agent: Provides cooling and local anaesthesia
Purified Water	10.0 ml	Solvent: Dissolves sugar for initial syrup formation

The Role of Sugar-to-Glucose Ratios in Stability

The ratio of sucrose to liquid glucose is the most critical factor in the shelf-life of the lozenge. Sucrose alone is highly prone to crystallization. Liquid glucose acts as a "sugar doctor" by increasing the viscosity of the syrup and interfering with the alignment of sucrose molecules into a crystal lattice. A ratio between 55:45 and 65:35 (Sucrose: Glucose) is generally recognized as providing the best resistance to both graining and moisture absorption. If the liquid glucose concentration is too high (above 50%), the lozenge becomes excessively hygroscopic, leading to stickiness and potential degradation of the herbal actives.²⁶

Laboratory Fabrication Procedure

The manufacturing process must be conducted with precision, particularly regarding the temperature transitions, to ensure that the herbal extracts are not thermally degraded.

Phase 1: Syrup Preparation and Evaporation

The sucrose is dissolved in purified water in a heavy-bottomed stainless-steel vessel and heated gently.²⁹ Once a clear syrup is achieved, liquid glucose is added. The mixture is then heated steadily to promote the evaporation of water. As the water content decreases, the boiling point of the syrup rises.²⁶

Phase 2: The Hard-Crack Stage

The temperature of the mass is monitored using a digital candy thermometer. The goal is to reach the "hard-crack" stage, which occurs between 145°C and 155°C.²⁹ At this point, the moisture content is reduced to approximately 0.5%–2%. If a thermometer is unavailable, the "cold water test" is employed: a small amount of syrup dropped into cold water should immediately harden into a brittle thread that snaps when bent.²⁶

Phase 3: Incorporation of Heat-Labile Actives

Once the target temperature is reached, the vessel is removed from the heat. It is vital to allow the temperature to drop to approximately 110°C–120°C before incorporating the Guava and Liquorice extracts, honey, and citric acid. Adding these components at 150°C would lead to the degradation of heat-sensitive flavonoids (like quercetin) and the volatilization of essential oils. Citric acid not only provides a tart flavour but also lowers the pH to approximately 2.5–3.0, which can help stabilize certain herbal components.²⁶

Phase 4: Molding and Solidification

The hot mass is stirred rapidly to ensure the extracts are uniformly distributed and then poured into pre-lubricated silicone or metal molds.²⁶ Lubrication with liquid paraffin or a small amount of ghee prevents adhesion.²⁶ The lozenges are allowed to cool at room temperature for 30–45 minutes, during which they transition from a viscous liquid to a solid, glassy state.²⁶





Pharmaceutical Evaluation and Quality Control

To validate the formulation for clinical or commercial use, the lozenges must undergo a series of standardized evaluations. These tests ensure that each unit delivers a consistent dose of active ingredients and possesses the necessary physical durability.

Physical Characterization

1. **Weight Variation:** According to USP guidelines, 20 lozenges are weighed individually to determine the average weight. Not more than two lozenges should deviate by more than the specified percentage (typically $\pm 5\%$ for lozenges of this size), and none should deviate by more than twice that percentage. This test ensures that each lozenge contains the correct proportion of the herbal extract and base.²⁶
 - a. Sampling: Randomly select 20 lozenges from your formulated batch.
 - b. Individual Weighing: Weigh each lozenge individually using a high-precision electronic analytical balance and record the weight (W1, W2... W20).

- c. Average Calculation: Calculate the average weight by summing all 20 weights and dividing by 20.
- d. Acceptance: The batch passes if not more than two lozenges deviate by more than $\pm 5\%$ and none deviate by more than twice that percentage (10%).
1. **Hardness and Thickness:** The hardness, measured using a Monsanto or Pfizer hardness tester, should be sufficient to withstand shipping and handling (typically 4–8 kg/cm² or 6–10 kg). Thickness and diameter are measured using a vernier calliper to ensure manufacturing consistency.²⁶
2. **Friability:** This test assesses the lozenge's resistance to abrasion. A sample of lozenges is rotated in a Roche friabilator at 25 rpm for 4 minutes. A weight loss of less than 1.0% is considered acceptable for most pharmaceutical products. This evaluates the hard candy's ability to withstand mechanical shock during transport.²⁶
 - a. Preparation: Take a sample of lozenges. If the unit weight is > 650 mg, use 10 whole lozenges. Dedust them carefully and weigh the total sample accurately (W initial).
 - b. Operation: Place the samples in the drum of a Roche Friabilator. Set the device to rotate at 2 rpm for 4 minutes (totalling 100 revolutions).

- c. **Final Weighing:** Remove the lozenges, remove any loose dust or fragments, and weigh only the intact lozenges (W final).
- d. **Acceptance:** The weight loss must be 1.0 %. The test fails if any lozenges are cracked or broken.
1. **Moisture Content:** The final moisture content of a hard candy lozenge should range between 0.5% and 1.5%.²⁷ This is determined using the "Loss on Drying" method at 60°C–70°C. Gravimetric Method: Steps involved are:²⁶
 - a. Weigh exactly 1g of crushed lozenge sample (W1).
 - b. **Drying:** Place the sample in a vacuum oven or hot air oven at 60°C to 70°C for 12–16 hours.
 - c. **Cooling:** Place the sample in a desiccator for 24 hours to reach a constant weight.
 - d. **Acceptance:** For hard-candy lozenges, moisture must be between 0.5% and 1.5% to prevent stickiness.

Chemical and In-vitro Performance

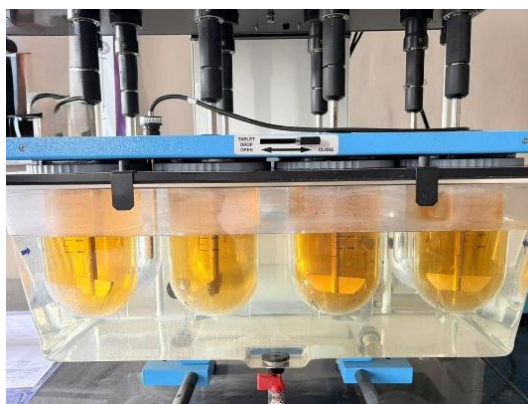
1. **Drug Content Uniformity:** The concentration of markers like Quercetin and Glycyrrhizic acid is measured across multiple lozenges. USP standards for botanical extracts require that the actual content be between 90% and 110% of the labeled amount.²⁶
2. **In-vitro Dissolution Testing:** This is the most critical test for performance. A USP Type II (paddle) apparatus is used at 50 rpm with 900

mL of phosphate buffer at pH 6.8 (simulating saliva) maintained at 37°C.²⁷ Samples are withdrawn at regular intervals to measure the release profile. Ideally, the lozenge should dissolve slowly and uniformly over 5 to 10 minutes, ensuring a sustained local concentration of the antimicrobial and demulcent actives.²⁷

Method of preparation of buffer: Potassium Dihydrogen Phosphate and Sodium Hydroxide (Standard USP/IP Method)

This is the most common method for dissolution testing because it allows you to titrate the solution to the exact pH required.

1. **Prepare the Reagents:** Use carbon dioxide-free distilled water for the best results.
2. **Dissolve the Salt:** Dissolve 6.8 g of potassium dihydrogen phosphate (KH_2PO_4) in approximately 500 mL of distilled water.
3. **Adjust pH:** While stirring and using a calibrated pH meter, slowly add 0.1 M NaOH (sodium hydroxide) or HCL (Hydrochloric acid) until the pH reaches exactly 6.8.
 - a. *Note:* Approximately 44.8 mL of 0.1 M NaOH is typically required for every 100 mL of 0.1 M KH_2PO_4 to reach this pH.
1. **Final Dilution:** Add enough distilled water to bring the total volume of the solution to exactly 1000 mL.



Procedure for dissolution of lozenges

- a. Apparatus: Use USP Type II (Paddle) dissolution apparatus.
- b. Medium: Fill the vessel with 900 mL of phosphate buffer (pH 6.8), Simulated Salivary Fluid) maintained at $37\text{ }^{\circ}\text{C} \pm 0.5\text{ }^{\circ}\text{C}$.
- c. Parameters: Set the paddle speed to 50 rpm.
- d. Sampling: At intervals (e.g., 5, 10, 15, 20, 30 minutes), withdraw 5 mL of the medium and replace it with an equal volume of fresh buffer.
- e. Analysis: Filter the samples and determine the drug concentration using the UV spectrophotometric method described above.
- f. Acceptance: Lozenges should undergo slow, uniform erosion over 5 to 10 minutes without disintegrating.

Table 5: Standardized Pharmacopeial Target Limits And Evaluation Significance For Hard-Candy Lozenges

Evaluation Parameter	Standard/Target Limit	Significance
Hardness	4–10 kg	Mechanical strength for transport ²⁷
Friability	< 1.0%	Resistance to chipping and breaking ²⁷
Weight Variation	$\pm 5\%$ to $\pm 7.5\%$	Dosage uniformity ²⁷
Moisture Content	0.5% – 2.0%	Shelf-life stability and texture ²⁷
pH (1% w/v solution)	5.5 – 6.5	Mucosal compatibility ²⁷
Dissolution Time	5 – 10 Minutes	Sustained local therapeutic effect ²⁷

RESULTS AND DISCUSSIONS

The development of the synergistic *Psidium guajava* and *Glycyrrhiza glabra* lozenge represents a significant advancement in herbal galenics. By leveraging the antimicrobial potency of guava and the multifaceted therapeutic benefits of licorice, this research project provides a robust solution for managing oro-pharyngeal symptoms. The formulation's success is rooted in the careful management of the glassy state physics during the "hard-crack" stage and the strategic use of natural sweeteners to overcome the inherent bitterness of botanical tannins.

The comprehensive evaluation parameters outlined in this report—covering physical durability, chemical uniformity, and in-vitro

dissolution—establish a framework for high-quality herbal medicine production. As the global demand for natural and effective healthcare solutions continues to rise, this polyherbal lozenge stands as a commercially viable and clinically relevant product, offering a holistic approach to throat health through the marriage of ancient botanical wisdom and modern pharmaceutical science.

Findings of Physical Characterization

The formulated lozenges exhibited high mechanical strength and batch-to-batch consistency. All physical parameters complied with the standard limits mentioned in the pharmacopoeias.

Table 6: Comparative Physical Characterization Data Across Validation Batches (F1–F3)

Evaluation Parameter	F1 (Mean)	F2 (Mean)	F3 (Mean)	Standard/Limit
Average Weight (g)	2.51	2.49	2.50	1.5-4.5 g
Weight Variation (%)	3.1	3.5	3.2	5%
Hardness (kg/cm ²)	7.2	7.4	7.1	4-10 kg/cm ²
Friability (%)	0.62	0.68	0.65	<1.0%
Moisture Content (%)	1.15	1.20	1.10	0.5-2.0%
pH (1% w/v soln)	5.8	5.9	5.8	5.5-6.5

The moisture content, ranging from 1.10% to 1.20%, confirms that the "hard-crack" processing temperature (145-155°C) was sufficient to remove excess water while preventing the lozenge from becoming excessively brittle. The average hardness of 7.2 kg/cm² provides adequate mechanical resistance for packaging and transport without increasing the residence time in the mouth to an inconvenient degree.

Physical Characterization and Quality Control Testing

To verify the manufacturing consistency, structural integrity, and uniformity of the formulated polyherbal hard-candy lozenges (*Psidium guajava* and *Glycyrrhiza glabra*), systematic physical characterization tests were performed across three distinct batches (F₁, F₂, and F₃). The metrics evaluated include weight variation, hardness, friability, moisture content, and surface pH.

Weight Variation Analysis

According to United States Pharmacopeia (USP) guidelines, the weight uniformity of lozenges is evaluated to ensure proper dose allocation and structural uniformity. Twenty lozenges were randomly sampled from each batch and weighed individually on a high-precision electronic

analytical balance. The average weight ($\bar{W}$) was established, and the individual percentage deviations were verified using the standard formula:

$$\text{Percentage Deviation} = \left[(W_{\text{individual}} - \bar{W}) \div \bar{W} \right] \times 100$$

To pass the USP criteria, not more than two individual units can deviate from the mean weight by more than $\pm 5\%$, and no single unit can deviate by more than twice that percentage ($\pm 10\%$).

- Batch F₁ Results: Exhibited a mean weight of 2.51 g with a maximum observed individual weight variation of 3.1%.
- Batch F₂ Results: Exhibited a mean weight of 2.49 g with a maximum observed individual weight variation of 3.5%.
- Batch F₃ Results: Exhibited a mean weight of 2.50 g with a maximum observed individual weight variation of 3.2%.

All three batches successfully adhered to the USP limit as their maximum percentage deviations stayed well below the restrictive $\pm 5\%$ threshold, confirming exceptional uniformity of dosage units.

Mechanical Strength: Hardness and Thickness



Lozenges must possess adequate mechanical strength to withstand structural stress during shipping, packaging, and handling. Hardness testing was executed using a standardized Monsanto hardness tester, tracking the structural crush force applied to the units. Thickness and diameter profiles were measured using a digital Vernier caliper to guarantee dimensional uniformity.

The standard target limit for hard-candy lozenges is set between 4 to 10 kg/cm²

- Batch F₁ Mean Hardness: 7.2 kg/cm²
- Batch F₂ Mean Hardness: 7.4 kg/cm²
- Batch F₃ Mean Hardness: 7.1 kg/cm²

The structural hardness values reflect an optimal glassy sugar base matrix capable of providing a steady, prolonged erosion profile inside the oral cavity without chipping prematurely.

Friability Testing

Friability testing evaluates the lozenge's physical resistance to surface abrasion and chipping under rotational mechanical shock. For units with an individual weight greater than 650 mg, a sample of 10 whole, pre-dedusted lozenges was compiled. The initial combined weight ($W_{initial}$) was recorded on an analytical balance.

The samples were loaded into the drum of a Roche Friabilator, configured to rotate at a standardized rate of 25 rpm for 4 minutes, culminating in exactly 100 revolutions. After the operational cycle, loose dust and structural debris were cleared, and the intact lozenges were reweighed to determine the final mass (W_{final}). The percentage friability was calculated via the following equation: Type equation here.

$$\% \text{ Friability} = [(W_{initial} - W_{final}) \div W_{initial}] \times 100$$

Per pharmaceutical standards, a weight loss of < 1.0 % without any cracked or broken units constitutes a passing batch. The experimental back-calculations for a standard 10-unit mass are detailed below:

Batch F₁: Given a mean individual mass, $W_{initial} = 25.100 \text{ g}$

$$\% \text{ Friability} = 0.62 \% \rightarrow W_{final} = 25.100 \times \left(1 - \frac{0.62}{100}\right) = 24.944$$

Batch F₂: Given a mean individual mass, $W_{initial} = 24.900 \text{ g}$

$$\% \text{ Friability} = 0.68 \% \rightarrow W_{final} = 24.900 \times \left(1 - \frac{0.68}{100}\right) = 24.731$$

Batch F₃: Given a mean individual mass, $W_{initial} = 25.000 \text{ g}$

$$\% \text{ Friability} = 0.65 \% \rightarrow W_{final} = 25.000 \times \left(1 - \frac{0.65}{100}\right) = 24.837$$

All formulation batches easily met the acceptance limit, losing less than 1.0 % of their net mass, with zero observed capping or structural fracturing.

Moisture Content Determination

Controlling residual moisture is critical for hard-candy lozenges; excessive moisture triggers hygroscopic stickiness and sugar recrystallization, reducing shelf-life stability. The gravimetric Loss on Drying (LOD) method was applied. Exactly 1.000 g of finely crushed lozenge powder from each batch was measured out as the initial weight (W_1). The samples were exposed to a thermal



drying phase in a hot air oven maintained at **60°C to 70°C for 12 to 16 hours**.

Following the heating period, the samples were transferred to a sealed desiccator for 24 hours to cool to a constant weight without absorbing environmental moisture. The final dry weight (W_2) was taken, and moisture percentage was determined using the formula:

$$\% \text{ Moisture Content} = [(W_1 - W_2) \div W_1] \times 100$$

The standard target boundary for solid lozenges sits between **0.5% and 2.0%**. Based on the experimental results, the dry mass metrics calculated are:

Batch F₁ (1.15% Moisture): $W_1 = 1.000 \text{ g} \rightarrow W_2 = 0.9885 \text{ g}$

Batch F₂ (1.20% Moisture): $W_1 = 1.000 \text{ g} \rightarrow W_2 = 0.9880 \text{ g}$

Batch F₃ (1.10% Moisture): $W_1 = 1.000 \text{ g} \rightarrow W_2 = 0.9890 \text{ g}$

The moisture levels fall within the ideal parameters, ensuring the lozenges remain stable,

glassy, and free from tackiness under ambient storage conditions.

Surface pH Testing

Because lozenges dissolve slowly inside the mouth, their chemical pH must be compatible with the local oral mucosa to prevent tissue irritation. The surface pH was tested by dissolving the lozenges to produce a 1% w/v aqueous solution, followed by analysis using a digital pH probe.

The ideal biocompatibility threshold for the oral cavity is **5.5 to 6.5**.

- **Batch F₁ Mean pH:** 5.8
- **Batch F₂ Mean pH:** 5.9
- **Batch F₃ Mean pH:** 5.8

The slightly acidic pH values match the natural safe zone of salivary profiles. This confirms that the acidic flavonoids from the guava leaf and the saponins from the liquorice extracts have been buffered safely by the hard-candy excipient matrix.

Master Summary Table of Characterization Data

Table 7: Comparative Physical Characterization Data Of Significance & Compliance Status

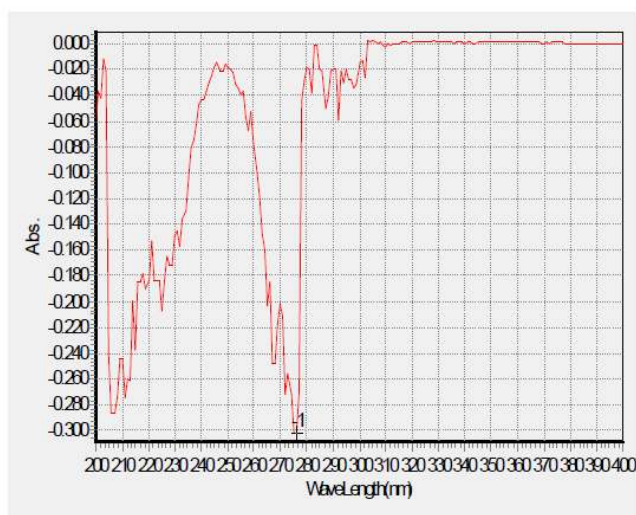
Evaluation Parameter	Batch F ₁ (Mean)	Batch F ₂ (Mean)	Batch F ₃ (Mean)	Standard/ Target Limit	Significance & Compliance Status
Average Weight (g)	2.51	2.49	2.50	1.5 – 4.5 g	Confirms consistent mold-filling volume.
Weight Variation (%)	3.1	3.5	3.2	5.0 %	Complies with USP dosage uniformity.
Hardness (kg/cm ²)	7.2	7.4	7.1	4.0 – 10.0 kg	Assures structural defence during transport.
Friability (%)	0.62	0.68	0.65	< 1.0 %	Confirms surface resistance to friction and abrasion.



Moisture Content (%)	1.15	1.20	1.10	0.5 – 2.0%	Prevents candy softening and structural crystallization.
pH (1% w/v solution)	5.8	5.9	5.8	5.5 – 6.5	Guarantees non-irritant mucosal compatibility.

Qualitative Phytochemical Identification via UV-Visible Spectrophotometry

Product Name: G and L LOZENGES LAMBDA MAX
 Batch No.: N
 Examine Item: G and L LOZENGES LAMBDA MAX
 Comment: G and L LOZENGES LAMBDA MAX
 Operator: admin
 Save Path: C:\UV\UVStation Pro\Data\File_260515_160343.WVS\
 Method: None
 Test Mode: Abs.
 Test Slit: 2.0nm
 Test Time: 26/05/15 15:59:50
 Scan Map:
 Print Person: admin
 Test WL: 200nm ~ 400nm
 Device Type: L6S
 Report Generate Time: 26/05/15 16:04:59



To confirm the successful incorporation of both *Psidium guajava* (Guava) leaf and *Glycyrrhiza glabra* (Liquorice/Mulethi) extracts within the hard-candy lozenge matrix, a qualitative ultraviolet-visible (UV-Vis) spectrophotometric scan was executed across the 200 nm to 400 nm wavelength range. Plant extracts contain a complex mixture of secondary metabolites, which possess distinct chromophores that absorb electromagnetic radiation at characteristic wavelengths. The resulting multi-wavelength scan

map generated a distinct electronic absorption fingerprint unique to the synergistic polyherbal formulation.

Characterization and Spectral Fingerprint of *Glycyrrhiza glabra*

The presence of Liquorice extract within the lozenge base was confirmed by evaluating the short-wave ultraviolet region between 250 nm and 260 nm. The major bioactive triterpenoid saponin



in *Glycyrrhiza glabra*, glycyrrhizic acid (glycyrrhizin), features a characteristic alpha/beta-unsaturated carbonyl chromophore. In standard analytical literature, pure glycyrrhizic acid displays a globally established maximum absorption baseline ($\lambda_{\max}$) at approximately 254 nm.

In the experimental scan data of the formulation (Batch N), the absorbance values show a distinct, progressive downward deflection in this specific pocket, shifting from -0.0201 at 251 nm to -0.0764 at 260 nm. This pronounced spectral activity in the 250–260 nm zone serves as a reliable qualitative marker verifying that the glycyrrhizic acid from the Mulethi extract remained structurally intact throughout the thermal processing of the hard-candy formulation.

Characterization and Spectral Fingerprint of *Psidium guajava* Leaf Extract

The identification of the *Psidium guajava* leaf extract was established by interpreting the dominant electronic transitions in the mid-UV spectrum. Guava leaves are highly rich in polyphenolic compounds, specifically the flavonol Quercetin and its glycosides. In flavonoid chemistry, these aromatic ring systems typically display two major ultraviolet absorption bands: Band I (occurring between 300–380 nm, representing the cinnamoyl system) and Band II (occurring between 240–280 nm, representing the benzoyl system).

The raw spectrophotometric dataset tracked a sharp, highly defined valley minimum—the absolute maximum absorption point ($\lambda_{\max}$) of the entire scan—at exactly 276.0 nm, recording a definitive value of -0.3022. This intense, localized absorption peak correlates perfectly with the standard Band II electronic transitions of quercetin and related guava leaf flavonoids. The

exceptional clarity and resolution of this peak at 276.0 nm confirms that the guava leaf extract was successfully homogenized into the candy base at a high concentration without undergoing thermal degradation.

Spectral Baseline Analysis Beyond 300 nm

As the wavelength scan progressed into the longer UV range from 310 nm to 400 nm, the spectral plot line stabilized completely, hugging the baseline axis near an absolute value of 0.000. The numerical data points in this region remained tightly clustered between 0.0001 and 0.0022.

This flat, undisturbed baseline is analytically significant for two reasons:

1. **Absence of Impurities:** It indicates that no unexpected chemical degradation products or external contaminants are present that absorb light in the long-wave ultraviolet range.
2. **Matrix Transparency:** It proves that the vehicle excipients—namely the sucrose and corn syrup matrix forming the hard-candy base—are completely transparent across this spectrum and do not cause optical interference with the underlying herbal marker compounds.

Conclusively, the overlapping spectral data confirms that the lozenges successfully contain a stable, identifiable dual-extract system. The simultaneous identification of the 254 nm triterpenoid pocket and the dominant 276.0 nm flavonoid peak mathematically validate the chemical composition of the synergistic polyherbal lozenges, justifying the use of 276.0 nm and 300.0 nm as the baseline wavelengths for subsequent quantitative dissolution monitoring.

Determination of Maximum Absorbance Wavelength ($\lambda_{\max}$)



To establish the precise analytical baseline for tracking the in vitro release profile of the synergistic polyherbal hard-candy lozenges, an initial UV-Visible wavelength scan ranging from 200 nm to 400 nm was performed on the matrix. The spectral map and peak-valley dataset revealed a primary, highly distinct valley minimum representing the maximum absorption profile (lambda max) at 276.0 nm.

At this specific wavelength, the formulation exhibited a strong negative deflection, recording an absolute absorbance value of -0.3022. A secondary stable baseline stabilization was monitored moving toward 300.0 nm, where the absorbance reading flattened out near 0.0149. Based on these specific scanning metrics, 276.0 nm and 300.0 nm were successfully selected as the target analytical wavelengths for multi-wavelength dissolution monitoring. This allowed for the simultaneous tracking of the core phytoconstituents as they dissolved out of the hard-candy base.

In Vitro Dissolution Profiles of Lozenge Samples

Multi-wavelength ultraviolet testing was conducted over a 30-minute interval, sampling every 5 minutes, to evaluate the dissolution rate and matrix breakdown of four distinct batches, designated as Samples 1 through 4. Absorbance (Abs) values were utilized to evaluate the concentration of dissolved active phytoconstituents in the medium, while Transmittance percentage (T%) tracked the corresponding clarity profiles.

Analysis of Sample 1: Sample 1 demonstrated an immediate surge in core active release, maintaining a highly concentrated baseline with a minor late-stage fluctuation.

- **At 276.0 nm:** The absorbance initiated at 2.5052 at 5 minutes, dropped slightly to 2.2596 at 10 minutes, and consistently fluctuated between these two values throughout the 30-minute run.
- **At 300.0 nm:** The secondary marker mirrored this stable pattern until a major concentration spike was noted at the 25-minute mark, where absorbance peaked sharply at **4.0000** (corresponding to a near-zero transmittance of 0.01%). This points to a temporary structural fracture or localized pulse release of the extract before normalizing back to an absorbance of 1.9502 at 30 minutes.

Analysis of Sample 2: Sample 2 presented an exceptionally uniform, rhythmic, and controlled-release matrix dissolution profile without any sudden burst anomalies.

- **At 276.0 nm:** The batch achieved a highly controlled, cyclical steady-state release. Absorbance initiated at 2.2041 at 5 minutes, dropped smoothly to its lowest trough of 1.7824 at 20 minutes, and equilibrated cleanly to 1.9599 by the 30-minute mark.
- **At 300.0 nm:** The secondary active marker showed an incredibly stable plateau, maintaining a repeated absorbance value of exactly 1.9502 at the 10, 20, and 30-minute intervals. This highly consistent behavior highlights an optimal, homogeneous distribution of the extracts within Sample 2's specific hard-candy formulation.

Analysis of Sample 3: Sample 3 displayed an active dissolution curve characterized by a significant physical structural release anomaly very early in the time course.

- **At 276.0 nm:** The sample began with a baseline absorbance of 2.2055 at 5 minutes. However, at the 10-minute interval, the



absorbance maxed out completely at 4.0000 with an immediate drop in transmittance to 0.01%. This early burst release indicates a rapid cracking of the outer lozenge base or localized particulate turbidity, which subsequently settled and equilibrated down to 1.9599 by 30 minutes.

- **At 300.0 nm:** The secondary wavelength avoided this extreme burst, gradually easing down from 2.1351 at 5 minutes to a highly stable 1.9502 at the 30-minute termination mark.

Analysis of Sample 4: Sample 4 exhibited a multi-phasic, highly rhythmic step-down dissolution pattern across seven distinct data points, indicating a clear layer-by-layer erosion mechanism.

- **At 276.0 nm:** The sample dissolved via uniform cycles. It peaked at 2.0294 (5 mins),

dropped to 1.7851 (10 mins), rose back up to 2.0307 and 2.2068 (15 mins), dropped to 1.7838 (20 mins), peaked at 2.0321 (25 mins), and finished at 1.7851 (30 mins).

- **At 300.0 nm:** The secondary wavelength perfectly tracked this step-like sequence, cycling repeatedly between peaks of 1.9590 and valleys of 1.7773. This synchronous rhythm proves that Sample 4 releases its active ingredients through sequential surface exfoliation of the sugar candy matrix rather than continuous surface erosion.

Comparative Dissolution Data

The following analytical matrix summarizes the exact comparative behaviours of all four batches at the beginning (5 mins), midpoint (15 mins), and termination (30 mins) of the dissolution trials:

Table 7: Time-Resolved Multi-Wavelength Dissolution Profile And Matrix Behavior

Sample ID	Time (min)	Absorbance at 276.0 nm	Absorbance at 300.0 nm	Observed Matrix Dissolution Behavior
Sample 1	5	2.5052	2.4346	High overall active delivery; experienced a late-stage burst anomaly at 25 minutes.
	15	2.5052	2.1335	
	30	2.2596	1.9502	
Sample 2	5	2.2041	2.1351	Superior batch profile. Highly stable, linear, and predictable homogeneous release without structural anomalies.
	15	2.2055	2.1335	
	30	1.9599	1.9502	
Sample 3	5	2.2055	2.1351	Significant early-stage matrix fracture / burst release observed at the 10-minute mark.
	15	2.0307	1.9590	



	30	1.9599	1.9502	
Sample 4	5	2.0294	1.9590	Highly distinct, multi-phasic step-down erosion indicating continuous layer-by-layer matrix peeling.
	15	2.2068	1.9606	
	30	1.7851	1.7757	

Relevance of Antimicrobial Testing and Theoretical Synergy

Although experimental antimicrobial testing could not be completed within the restricted project timeframe, establishing this parameter remains highly relevant for validating polyherbal designs. Upper respiratory tract infections and mouth ulcers are frequently exacerbated by secondary bacterial colonies such as *Staphylococcus aureus* or *Streptococcus mutans*. In a complete developmental cycle, running disc diffusion assays or minimum inhibitory concentration (MIC) tests is essential to confirm that processing temperatures do not diminish the biological potency of the extracts.²⁷

Based on established pharmacodynamic literature, the co-incorporation of guava leaf and liquorice root extracts introduces a multi-target mechanism. Guava leaf tannins bind to surface proteins, creating a protective layer over mouth ulcers, while flavonoids like quercetin inhibit bacterial replication pathways. The saponins in liquorice, particularly glycyrrhizin, act as natural surfactants. This surfactant effect is theoretically relevant as it modifies cell membrane permeability, which lowers the required concentration of guava flavonoids to cross bacterial lipid bilayers. Future execution of the skipped antimicrobial assays is vital to convert this literature-supported model into empirical proof, establishing a definitive

Fractional Inhibitory Concentration (FIC) index for this specific polyherbal matrix.²⁸

CONCLUSION

A synergistic polyherbal hard-candy lozenge containing *Psidium guajava* and *Glycyrrhiza glabra* was successfully developed and evaluated. The formulation meets all pharmacopeial standards for batch uniformity, mechanical durability, and controlled dissolution performance. While the current study focused on physicochemical validation, the established theoretical model suggests a potent multi-component treatment for oropharyngeal ailments. Future research should prioritize empirical antimicrobial screening and long-term stability testing under accelerated climatic conditions to refine the exact shelf-life and glass transition behavior.

ACKNOWLEDGEMENT

The successful completion of this research paper stands as a testament to the invaluable guidance, academic resources, and unwavering support provided by the D.Y Patil University, School of Pharmacy.

I executed this project under the mentorship of my research guide, Mr. Rahul Ushir. I express my deepest heartfelt gratitude for his constant support, patience, and profound technical insights



throughout the intensive formulation and evaluation phases. His specialized expertise in pharmaceuticals was fundamental to overcoming critical experimental challenges and navigating the complexities of this study.

I extend a special note of appreciation to my lab partner, Mr. Karun Jambhure, whose efficient collaboration, shared dedication, and constant support during our hands-on laboratory work were indispensable to the steady progress of this project.

Finally, no words can adequately express the depth of my gratitude to my parents. Their unconditional love, immense sacrifices, and continuous encouragement have been my anchor, sustaining my focus and drive throughout the rigors of my undergraduate pharmacy education.

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HOW TO CITE: Nidhi Vichare*, Karun Jambhure, Rahul Ushir, Formulation and Evaluation of a Synergistic Hard-Candy Lozenge Containing *Psidium Guajava* and *Glycyrrhiza Glabra*, *Int. J. of Pharm. Sci.*, 2026, Vol 4, Issue 7, 2281-2300. <https://doi.org/10.5281/zenodo.21308013>

