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Formulation and Evaluation of Polyherbal Mouth Ulcer Gel

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Abstract: *Mouth ulcers are painful lesions commonly caused by stress, poor oral hygiene, nutritional deficiencies, infections, and irritation of the oral mucosa. Herbal medicines are increasingly preferred for oral care due to their safety, effectiveness, and minimal side effects compared to synthetic formulations. The present study focuses on the formulation and evaluation of a herbal mouth ulcer gel using Giloy (Tinospora cordifolia) as the principal active ingredient. Giloy is well known for its anti-inflammatory, antimicrobial, antioxidant, and wound-healing properties, making it a suitable natural remedy for the treatment of mouth ulcers. The herbal gel was prepared by incorporating Giloy extract along with other natural ingredients such as turmeric, Tulsi, and aloe vera into a suitable gel base. Turmeric provides anti-inflammatory and antiseptic activity, Tulsi exhibits antimicrobial and soothing effects, while aloe vera promotes cooling and healing of the ulcerated tissue.*

The formulation was prepared using suitable excipients and evaluated for various physicochemical parameters including pH, homogeneity, spreadability, viscosity, stability, appearance, and irritation test. The prepared gel showed good consistency, smooth texture, satisfactory spreadability, and was found to be stable and non-irritant. The herbal mouth ulcer gel demonstrated promising effectiveness in reducing pain, inflammation, and promoting faster healing of oral ulcers. The study concludes that the Giloy based herbal gel can serve as a safe, effective, and economical natural alternative for the management of mouth ulcers.

Keywords: - Herbal Gel, Mouth Ulcer, Giloy (Tinospora cordifolia), Turmeric, Tulsi, Aloe Vera, pH, Spreadability, Wound Healing

I. INTRODUCTION

Concept of natural healing and herbal medicine has existed since ancient times, forming an integral part of traditional systems of medicine such as Ayurveda. Medicinal plants have been widely used for the treatment of various oral and systemic disorders due to their safety, effectiveness, and holistic healing properties. In recent years, there has been a growing interest in herbal formulations for oral care, especially for the management of mouth ulcers, as they are considered safer alternatives to synthetic drugs. Mouth ulcers are small, painful lesions that develop in the oral cavity and are commonly associated with factors such as stress, nutritional deficiencies, hormonal changes, minor injuries, or microbial infections. These ulcers often cause discomfort while eating, drinking, and speaking, thereby affecting the quality of life. Although several synthetic formulations are available for treatment, they may provide only temporary relief and can sometimes lead to adverse effects on prolonged use. Hence, herbal alternatives are gaining importance due to their safety, effectiveness, and minimal side effects.[1]

The present herbal mouth ulcer gel is formulated using natural medicinal herbs such as Giloy, Tulasi, Aloe vera, and Turmeric. These herbal ingredients possess significant anti-inflammatory, antimicrobial, antioxidant, antibacterial, immunomodulatory, and wound healing properties that help in the treatment and prevention of mouth ulcers. Giloy is known for its immunomodulatory and anti-inflammatory activities, helping reduce pain and supporting the body's natural healing process. Tulasi exhibits strong antimicrobial, antiseptic, and antibacterial properties that control oral infections and inhibit microbial[2] growth. Aloe vera provides soothing, cooling, and moisturizing effects, accelerates

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wound healing, and promotes regeneration of damaged oral tissues. Turmeric contains curcumin, a bioactive compound with strong anti-inflammatory, antioxidant, and antibacterial activities that promote rapid healing and prevent secondary infections.[3]

The formulated gel is designed to provide localized action at the affected site, ensuring prolonged contact with the oral mucosa and improved therapeutic effectiveness. It aims to reduce pain, inflammation, microbial load, and irritation while promoting faster healing in a natural and safe manner. Due to herbal ingredients, the gel offers better patient compliance with minimal side effects compared to synthetic preparations[4].

Herbal plant used in mouth ulcer gel:

1 Giloy (Tinospora cordifolia):

Fig no [1] Giloy (Tinospora cordifolia)

Synonyms	Guduchi, Amrita, Amritavalli Chakralaksha, Chinnodbhava, Madhuparni, Heart-shaped moonseed
Biological source	Giloy consists of the dried mature stem of Tinospora cordifolia (Willd.) Miers, helonging to the family Menispermaceae
Key constituents	Alkaloids, glycosides, flavonoids, terpenoids, polysaccharides.

Giloy (Tinospora cordifolia) is a traditional Ayurvedic herb known as "Amrita" (root of immortality) and has long been used to trvat ulcers and digestive issues. Taditionally used for the treatment of me and gastric ulcers. Its pharmacological actions include enhancing white blood cell activity, scavenging free radicals, reducing inflammatory mediators, inhibiting certain bacteria and vinases and protecting the gastrointestinal lining all of which validate its traditional uses

Medicinal uses:

It boosts immunity munomodulator

Anti-inflammatory, antioxidant, and antipyrotic.

Helps in mouth ulcers, wound healing and infections.

It Supports digestive health and ruducus acidity.

Useful in fever, diabetes and liver disorders

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2.Turmeric (Curcuma longa):

Fig no [2] Turmeric (Curcuma longa)

Synonyms	Haridra (Ayurveda), Haldi (Hindi), Kanchani/Kanchani Haridra, Indian saffron.
Biological source	Turmeric is obtained from the dried, mature rhizomes of the plant Curcuma longa Linn... a perennial herb belonging to the family Zingiberaceae
Key constituents	Curcuminoids volatile oil, polysaccharides, phenolic compound.

Turmeric is a well-known medicinal herb widely used in traditional Indian medicine for oral and skin disorders. In mouth ulcers turmeric acts natural anti-inflammatory, antimicrobial, antioxidant and wound-healing agent. Curcumin inhibits inflammatory mediators such as prostaglandins and cytokines, thereby reducing pain, redness and swelling. Turmeric also promotes faster epithelial regeneration, aiding in ulcer healing, Traditionally, turmeric is used as a paste with water, honey or glycerin, or Turmeric mouth rinse, providing symptomatic relief and accelerating healing. Because of its safety, affordability and minimal side effects, alternative or adjunct therapy for the management of mouth ulcers. considered an effective herbal.

Medicinal uses:

Antiinflammatory: Turmeric contains curcumin, which reduces inflammation in oral tissues, helping ulvers heal faster.
Antimicrobial: Curcumin has antibacterial and antifungal properties, preventing infections in the ulcer.
Antioxidant: Helps in scavenging free raclicals, reducing oxidative stress Pain Relief Turmeric can reduce discomfort and buming sensation associated with ulcers.
Healing Promotion: Stimulates tissue regeneration, aiding in faster recovery of oral mucosa.

3. Aloe vera:

Fig no [3] Aloe vera

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Synonyms	Aloe barbadensis, Aloe indicates, Kumari (Ayurveda), Ghritkumari, Ghritkumarinas, Musabber/Musabber (Unani), Indian aloe, True aloe, Barbudos aloe.
Biological source	biological source of Aloe vera is the fresh or dried leaves and the clear mucilaginous gel obtained from the plant Aloe vera (L.) Burm. I belonging to the familly Asphodelaceae.
Key constituents	Polysaccharides, Anthraquinon (aloin, alocmodin), vitamins A,C,E b complex, enzymes, amino acid minerals.

Aloe vera (Aloe barbadensis Mill) is a widely recognized medicinal plant commonly employed in traditional medicine systems including Ayurveda, Siddha, Unani, and folk practices. The therapeutically active part of the plant is its fresh leaves, which contain a clear, jelly-like gel and a yellow-colored latex.

Medicinal uses:

- Promotes healing: Enhances epithelial cell migration and accellerates wound healing Reduces inflammation.
- Its anti-inflammatory compounds help decrease pain and swelling.
- Antimicrobial action: Helps prevent secondary infections by inhibiting bacterial and fungal growth.
- Antioxidant support: Protects tissues from oxidative stress, aiding faster recovery.

4. Tulasi (Ocimum sanctum Linn):

Fig no [4] Tulasi (Ocimum sanctum Linn)

Synonyms	Holy fruit, Ocimum tenuiflorum, Tulasi/Tulsi (Ayurveda), Krishna Tulasi, Rama Tulasi, Holybasil, Sacred results.
Biological source	The biological source of Tulasi consists of the fresh or dried leaves obtained from the plant Ocimum sanctum (Family: Lamiaceae). The larves contain volatile oils and bioactive phytoconstituents responsible for its therapeutic activity .
Key constituents	Essential oils (cugenol, methyl cugenol, linakool), Flavonoids (orientin, vicenin), Phenolic compounds, Triterpenoids, Tannins, Vitamins (A, C) and minerals

Tulasi (Ocimum sanctum Linn) Despite remarkable advances in science and technology. Modern life is increasingly dominated by stress and lifestyle-related disorders. Rapid urbanization, unhealthy dietary habits, environmental pollution and reduced contact with nature have significantly affected human health in this context Tulasi (Ocimum sanctum Linn.). It serves as a natural protective herb aiding the body in coping with stress and enhancing imme strength. Regular use of Tulasi supports overall well-being and helps in the prevention of lifestyle-related chronic diseases due to its antioxidant, anti inflammatory and detoxifying properties.

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Medicinal uses:

- Tulasi is traditionally used in the treatment of mouth ulcers due to its antimicrobial anti-inflammatory, analgesic, and wound-healing properties.
- Chewing fresh Tulasi leaves or using Tulasi lenf extract as a mouth rinse helps reduce pain, inflammation, and ulcer durution.

Herbal Medicinal Gel:

Herbal medicinal gels are semisolid preparations used for the treatment and relief of various oral and skin disorders. These formulations are prepared using natural herbs and plant extracts possessing therapeutic properties. The term "gel" is derived from the Latin word gelu, meaning "frozen" or "jelly-like substance." Herbal medicinal gels are widely used due to their soothing action, easy application, better patient compliance, and fewer side effects compared to synthetic preparations. In recent years, herbal medicines have gained global acceptance because of their safety, effectiveness, and natural origin. There is an increasing demand for herbal therapeutic formulations in the pharmaceutical and healthcare industries. Traditional systems of medicine such as Ayurveda, Siddha, and Unani have long used medicinal plants for the treatment of wounds, ulcers, infections, and inflammation.

Herbal medicinal gels are important dosage forms as they provide prolonged contact with the affected site and promote faster healing. Mouth ulcer gel is considered a herbal therapeutic oral preparation used for treating ulcers, pain, inflammation, and infections in the oral cavity. Herbal ingredients such as Giloy, Turmeric, Tulsi, and Aloe vera are commonly used due to their antimicrobial, anti-inflammatory, antioxidant, and wound-healing properties. These formulations are gaining popularity due to their biocompatibility, affordability, and minimal adverse effects. They help in reducing irritation, promoting tissue repair, and maintaining oral hygiene naturally.

Gel:

Gel preparations are semisolid dosage forms in which a liquid phase is entrapped within a three-dimensional polymer network, forming a jelly-like structure. The term "gel" originates from the Latin word gelu, meaning frozen or solidified mass. Gels are widely used in pharmaceutical and cosmetic formulations due to their smooth consistency, non-greasy nature, and ease of application. Gels can be prepared using either natural or synthetic polymers and are widely utilized for the topical and mucosal administration of drugs. They are transparent or translucent in nature and provide better drug release compared to many conventional semisolid dosage forms. Gels are used for cleansing, soothing, moisturizing, and therapeutic purposes.In pharmaceutical applications, gels are designed to deliver active ingredients directly to the site of action, ensuring localized treatment with minimal systemic absorption. They are commonly used in dermatological and oral formulations for conditions such as inflammation, infections, burns, and ulcers.Due to their high water content and biocompatibility, gels are non-irritating and provide a cooling and soothing effect. They are widely preferred for patient-friendly topical drug delivery systems.

Ideal Properties Of Herbal Mouth Ulcer Gel:

1) It should be non-toxic and safe for application on oral mucosa.
2) It should have a smooth and fine particle size for easy application.
3) It should provide a soothing and protective coating over ulcers.
4) It should have proper gel consistency to stay at the site of action without flowing easily.
5) It should spread uniformly over the affected area inside the mouth.
6) It should contain suitable preservatives to maintain stability and shelf life.
7) It should be compatible with oral pH and not cause irritation or burning sensation.
8) It should be non-irritant and safe
9) herbal formulations are safer than synthetic formulations
10) It should have wound healing property.

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Benefits Of Herbal Mouth Ulcer Gel:

1. Provides relief from pain and burning sensation caused by mouth ulcers.
2. Promotes faster healing of oral ulcers and damaged mucosa.
3. Reduces inflammation and redness in the affected area.
4. Forms a protective layer over ulcers, preventing further irritation.
5. Helps maintain oral hygiene and reduces microbial growth.
6. Suitable for regular use with minimal or no side effects.
7. Made from natural herbal ingredients, making it safer than synthetic products.
8. Easily available and cost-effective.

Improves overall comfort while eating and speaking.

9. Can include various medicinal plant extracts with healing and anti-inflammatory properties.

Anatomy of Oral Mucosa:

Oral Mucosa:

Epithelium

- Outermost protective layer
- Stratified squamous cells
- Protects from injury, microbes, chemicals
- Site of ulcer formation

Lamina Propria

- Connective tissue layer
- Blood vessels + nerves
- Supports and nourishes epithelium
- Helps in healing of ulcers

Submucosa

- Present in some areas
- Contains blood vessels, glands, fat
- Provides support and flexibility

Anatomy of Oral Mucosa (T-Tree Short Explanation)

Oral Mucosa:

Epithelium

It is the outermost protective layer of the oral mucosa made of stratified squamous cells. It protects the underlying tissues from mechanical injury, microorganisms, and chemicals. In many areas, it is non-keratinized and is the primary site of mouth ulcer formation.

Lamina Propria

It is a connective tissue layer present below the epithelium. It contains blood vessels, nerves, and collagen fibers that provide support and nutrition to the epithelium. It plays an important role in healing and inflammation of ulcers due to rich blood supply.

Submucosa

It is a deeper supportive layer present in some regions of the oral cavity. It contains blood vessels, minor salivary glands, and fat tissue. It connects the mucosa to underlying muscles and provides flexibility and cushioning

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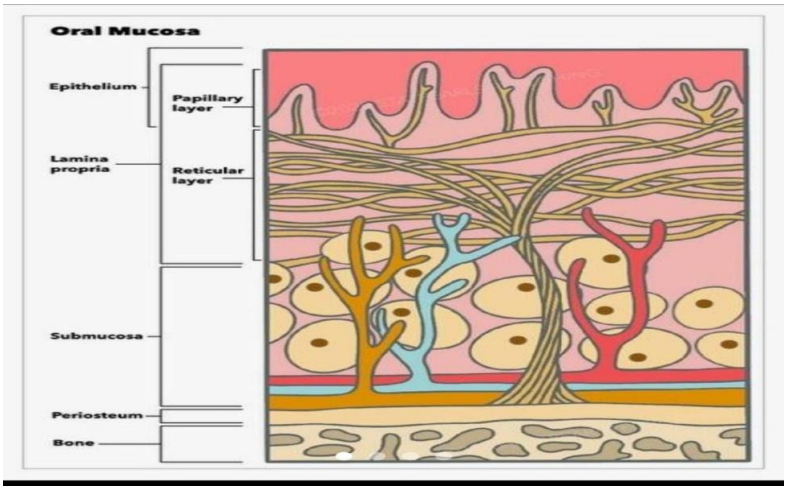

Fig no [5] Anatomy of oral MucosaPreparation of Extracts

Preparation Method of Herbal Extract:

Preparation of Giloy extracts :

Preparation of Giloy extract .Fresh giloy leaves were collected, washed thoroughly, and shade dried to remove moisture. The dried material was then powdered, and 20 g of powder was placed in a clean, closed container. A hydroalcoholic solvent consisting of 70% ethanol and 30% distilled water (200 mL) was added to the powder. The mixture was kept for 24–48 hours at room temperature with occasional shaking to enhance extraction. After maceration, the mixture was filtered using muslin cloth followed by Whatman filter paper to remove plant residues. The filtrate was concentrated on a water bath below 60°C until a semi-solid extract was obtained. The final extract was stored in an airtight container under refrigerated conditions at 2–8°C for further use.

Fig no [6] Extraction of Giloy

Preparation of Turmeric Extract:

Rhizomes of Curcuma longa were washed, shade dried, and ground into coarse powder. About 10 g of powder was macerated in 200 ml of hydroalcoholic solvent (70% ethanol:30% distilled water) for 24–48 hours with intermittent shaking. The mixture was then filtered through muslin cloth, and the filtrate was concentrated on a water bath below 60°C to obtain a thick extract rich in curcuminoids. The extract was stored in an airtight container at 4°C (refrigerator temperature) until further formulation use.

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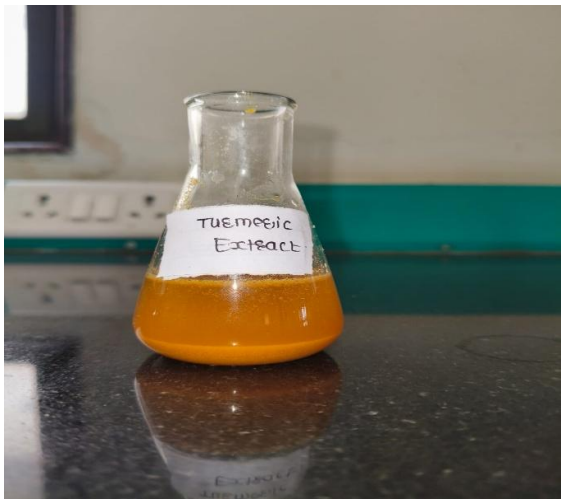

Fig no [7] Extraction of Turmeric

Preparation of Tulsi Extract:

Leaves of Ocimum tenuiflorum were washed thoroughly, shade dried, and powdered. About 10 g of the powder was macerated with 200 ml of hydroalcoholic solvent (70% ethanol:30% distilled water) for 24 hours with occasional shaking to enhance extraction. After maceration, the mixture was filtered using muslin cloth. The filtrate was concentrated on a water bath below 60°C until a semisolid extract was obtained. The prepared extract was stored in an airtight container at 4°C (refrigerator temperature) until further use in gel formulation.

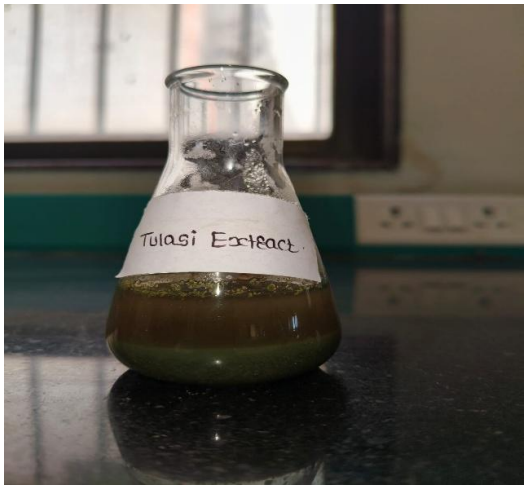

Fig no [8] Extraction of Tulsi

Preparation of Aloe vera extracts:

Fresh leaves of Aloe vera were collected and washed thoroughly with distilled water. The outer green rind of the leaves was carefully removed, and the inner mucilaginous gel was separated aseptically. About 15 g of fresh gel was collected and homogenized using a blender to obtain a uniform extract. The homogenized gel was filtered to remove fibrous material and other impurities. The obtained extract was transferred into a sterile airtight container and stored under refrigerated conditions at 4°C until further use in gel preparation.

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Fig no [9] Extraction of Aloe vera

Preparation of Mouth Ulcer Gel (Herbal Gel Formulation – 100 g):

The mouth ulcer gel was prepared using a two-step dispersion method followed by gel formation. Initially, the gel base was prepared by dispersing **Carbopol 934** in a portion of purified water with continuous stirring until it was completely swollen and a uniform dispersion was obtained. In a separate beaker, the aqueous phase was prepared by dissolving **Giloy extract, Aloe vera gel, Tulsi extract, and Turmeric extract** in purified water along with **glycerin and propylene glycol** under gentle stirring to obtain a homogeneous solution.

The prepared aqueous phase was then slowly incorporated into the Carbopol dispersion with continuous stirring to ensure uniform distribution of herbal extracts. After complete mixing, **methyl paraben** (previously dissolved in a small amount of warm water or propylene glycol) was added as a preservative. The formulation was then neutralized using **triethanolamine (q.s.)**, which converted the dispersion into a clear, smooth gel by adjusting the pH and enabling gel network formation. Stirring was continued until a uniform, lump-free gel consistency was achieved. Finally, the gel was made up to **100 g with purified water**, mixed thoroughly, and stored in a clean, airtight container.

Sr.no	Ingredient	Quantity	Role
1	Giloy extract	20 g	Main active ingredient (anti-inflammatory, antimicrobial)
2	Aloe vera gel	15 g	Soothing agent, wound healing, moisturizing
3	Turmeric extract	10 g	Anti-inflammatory and antioxidant agent
4	Tulsi extract	10 g	Antimicrobial and healing enhance
5	Carbopol 934	1 g	Gelling agent (provides gel structure)
6	Glycerin	10 g	Humectant (retains moisture, softens tissue)
7	Propylene glycol	5 g	Penetration enhancer (improves absorption)
8	Methyl paraben	0.2 g	Preservative (prevents microbial growth)
9	Triethanolamine	q.s	pH adjuster and gel neutralizer
10	Purified water	q.s to 100 g	Vehicle / solvent base

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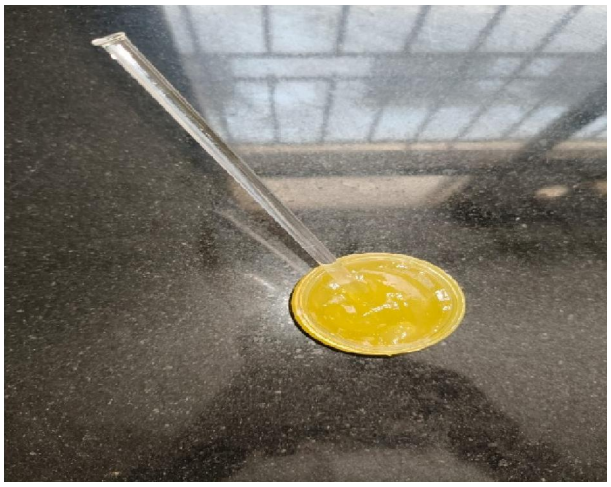

Fig no [9] Formulated Gel

Fig no [10] Mouth ulcer gel with label

Phytochemical Screening of Herbal Extract:

Phytochemical Screening of Giloy (Tinospora cordifolia) Extract:

The crude extract of Giloy was subjected to preliminary phytochemical screening to identify the presence of various bioactive constituents using standard qualitative methods.

1**. Alkaloids Test (Dragendorff's / Mayer's Test):** A small amount of the extract was treated with Dragendorff's reagent. Formation of a precipitate indicated the presence of alkaloids. The extract was mixed with sodium hydroxide solution, which produced an intense yellow color that became colorlesson addition of dilute acid, indicating flavonoids.

2. **Tannins Test (Ferric Chloride Test):** A small quantity of the extract was treated with 1% ferric chloride solution. Formation of a blue-black or greenish coloration confirmed the presence of tannins.

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3. **Saponins Test (Froth Test):** The extract was shaken vigorously with distilled water. Persistent froth formation indicated the presence of saponins.

4. **Glycosides Test:** The extract was mixed with glacial acetic acid and ferric chloride, followed by the addition of concentrated sulfuric acid:

The extract was treated with chloroform and concentrated sulfuric acid. A reddish-brown interface indicated the presence of steroids/terpenoids.

Conclusion:
Preliminary phytochemical screening of Giloy extract confirmed the presence of important bioactive compounds such as alkaloids, flavonoids, tannins, saponins, glycosides, and terpenoids, which may contribute to its anti inflammatory and wound-healing properties

Phytochemical Screening of Turmeric (Curcuma longa) Extract:
The crude extract of Turmeric was subjected to preliminary phytochemical screening using standard qualitative methods to identify bioactive constituents. The following result were observed:

1. **Alkaloids Test (Dragendorff's / Mayer's Test):** The extract was treated with Dragendorff's reagent. Formation of an orange or reddish-brown precipitate confirmed the presence of alkaloids.

2. **Flavonoids Test (Alkaline Reagent Test):** The extract was mixed with sodium hydroxide solution, producing an intense yellow color that became colorless on addition of dilute acid, indicating the presence of flavonoids.

3. **Tannins Test (Ferric Chloride Test):** The extract was mixed with 1% ferric chloride solution, and the appearance of a greenish-black color confirmed the presence of tannins.

4. **Saponins Test (Froth Test):** The extract was vigorously shaken with distilled water. Formation of stable froth confirmed the presence of saponins

5. **Steroids / Terpenoids Test (Salkowski Test):** The extract was combined with chloroform and concentrated sulfuric acid. A reddish-brown interface indicated the presence of steroids/terpenoids.

Conclusion:
The phytochemical screening of Turmeric (Curcuma longa) extract confirmed the presence of alkaloids, flavonoids, tannins, saponins, and terpenoids/steroids.
For mouth ulcer gel formulation, the most important constituents are:
Curcumin (Flavonoid-like compound) → strong anti-inflammatory and antioxidant activity

Tannins → wound healing and protective effect

Terpenoids → antimicrobial and anti-inflammatory activity

Saponins → cleansing and antimicrobial properties

These bioactive compounds make Turmeric highly effective for reducing inflammation, pain, and microbial infection in oral ulcers, supporting faster healing in gel formulations.

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Phytochemical Screening of Tulsi (Ocimum tenuiflorum) Extract:

The crude extract of Tulsi was subjected to preliminary phytochemical screening using standard qualitative methods to identify the presence of bioactive constituents. The results showed the following outcomes.

1. Alkaloids Test (Dragendorff's / Mayer's Test): The extract was treated with Dragendorff's reagent. Formation of an orange or reddish-brown precipitate confirmed the presence of alkaloids.
2. Flavonoids Test (Alkaline Reagent Test): The extract was mixed with sodium hydroxide solution, producing an intense yellow color that became colorless on addition of dilute acid, indicating the presence of flavonoids.
3. Tannins Test (Ferric Chloride Test): The extract was treated with 1% ferric chloride solution. A blue-black or greenish coloration confirmed the presence of tannins.
4. Saponins Test (Froth Test): The extract was shaken vigorously with distilled water. Persistent froth formation indicated the presence of saponins.
5. Glycosides Test: The extract was treated with glacial acetic acid, ferric chloride, and concentrated sulfuric acid. A brown ring at the interface indicated the presence of glycosides.
6. Steroids / Terpenoids Test (Salkowski Test): The extract was mixed with chloroform and concentrated sulfuric acid. A reddish-brown interface indicated the presence of steroids/terpenoids.

Conclusion:

The preliminary phytochemical screening of Tulsi (Ocimum tenuiflorum) extract confirmed the presence of alkaloids, flavonoids, tannins, saponins, glycosides, and terpenoids/steroids. For mouth ulcer gel formulation, the most important bioactive constituents are: Flavonoids → strong anti inflammatory and antioxidant activity, promotes healing Tannins → astringent effect, helps in wound contraction and protective layer formation Saponins → antimicrobial and cleansing action Terpenoids → antibacterial and anti-inflammatory properties Glycosides → support tissue repair and healing process These phytochemicals collectively contribute to antimicrobial, anti-inflammatory, analgesic, and wound healingproperties, making Tulsi extract highly suitable for oral ulcer gel formulation.

Phytochemical Screening of Aloe vera (Aloe barbadensis Miller) Extract:

The crude gel extract of Aloe vera was subjected to preliminary phytochemical screening using standard qualitative methods. The results showed the following outcomes:

1. Flavonoids Test (Alkaline Reagent Test): The extract was treated with sodium hydroxide solution. Yellow coloration that faded on acid addition confirmed the presence of flavonoids.
2. Saponins Test (Froth Test): The extract was shaken with distilled water. Stable froth formation indicated the presence of saponins.
3. Glycosides Test: The extract was treated with glacial acetic acid, ferric chloride, and concentrated sulfuric acid. A brown ring at the interface indicated the presence of glycosides.
4. Steroids / Terpenoids Test (Salkowski Test): The extract was mixed with chloroform and concentrated sulfuric acid. A reddish-brown interface indicated the presence of steroids/terpenoids.

Conclusion:

The phytochemical screening of Aloe vera extract confirmed the presence of flavonoids, saponins, glycosides, and terpenoids/steroids.

For mouth ulcer gel formulation, the most important constituents are:

Aloe polysaccharides (e.g., acemannan) → promotes tissue repair and healing

Flavonoids → anti-inflammatory and antioxidant activity

Saponins → antimicrobial and cleansing action

Glycosides & terpenoids → support healing and reduce inflammation

These components make Aloe vera highly effective in soothing irritation, promoting epithelial regeneration, and accelerating wound healing in oral ulcers.

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DPPH Radical Scavenging Assay (For Polyherbal Mouth Ulcer Gel):

Principle:

The DPPH (2,2-diphenyl-1-picrylhydrazyl) assay is based on the ability of antioxidants present in the polyherbal mouth ulcer gel to donate hydrogen atoms or electrons to the stable DPPH free radical. DPPH is deep violet in color, and upon reduction by antioxidants, it changes to a yellow-colored compound (DPPH-H). The decrease in absorbance at 517 nm is measured, which indicates the radical scavenging activity of the gel.

Materials Required:

DPPH solution (0.1 mM in methanol)
Polyherbal gel extract (aqueous or hydroalcoholic dilution)
Methanol (analytical grade)
UV-Visible spectrophotometer
Test tubes
Micropipettes

Chemicals Required – DPPH Assay:

DPPH (2,2-diphenyl-1-picrylhydrazyl) – Free radical reagent
Methanol (analytical grade) – Solvent for preparing DPPH solution and sample dilution
Distilled water – For preparation of aqueous gel extract (if used)
Ascorbic acid (Vitamin C) – Standard antioxidant for comparison
Polyherbal mouth ulcer gel extract (test sample containing Giloy, Tulsi, Turmeric, Aloe vera)
Ethanol (optional, 70–95%) – For hydroalcoholic extraction if required

Procedure:

Fresh polyherbal mouth ulcer gel extract was prepared by dissolving a known quantity of gel in suitable solvent (methanol or distilled water).
Serial dilutions of the sample were prepared in the range of 10, 20, 30, 40, and 50 µg/ml.
1 ml of freshly prepared 0.1 mM DPPH solution was added to each test tube.
1 ml of different concentrations of gel sample was added to each tube.
A control was prepared by adding 1 ml of DPPH solution with 1 ml of methanol (without sample).
The reaction mixtures were shaken well and incubated in the dark for 30 minutes at room temperature to prevent light degradation.
After incubation, absorbance was measured at 517 nm using a UV-Visible spectrophotometer.
Ascorbic acid or standard antioxidant was used as reference standard.
All readings were taken in triplicate for accuracy.

Calculation:

% Radical Scavenging Activity
Where:
A control = absorbance of control
A sample = absorbance of test sample
IC_{50} Determination
IC_{50} is defined as the concentration of sample required to inhibit 50% of DPPH radicals.
It was calculated from the graph plotted between % inhibition vs concentration.

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Observation Table:

Concentration	Absorbance
10	0.78
20	0.65
30	0.50
40	0.38
50	0.25

Step 1: calculation of % activity

Formula: % Activity =A sample / A max ×100

Calculated values

Concentration	%Activity
10	25.2%
20	38.5%
30	55.1%
40	68.4%
50	82.0%

Step 2: IC50 Calculation

Formula: $IC50 = C_1 + \frac{(50-A_1)(C_2-C_1)}{A_2-A_1}$

IC50= 25.2µg/ml

Result:

The polyherbal mouth ulcer gel showed a concentration-dependent increase in DPPH radical scavenging activity. The IC_{50} value of the formulation was found to be approximately 25–30 µg/ml, indicating strong antioxidant potential.Conclusion. The DPPH assay confirmed that the polyherbal mouth ulcer gel possesses significant free radical scavenging activity. This antioxidant property may contribute to reducing oxidative stress, inflammation, and accelerating the healing process of mouth ulcers.

Evaluation Parameters of Mouth Ulcer Herbal Gel:

1. Organoleptic Evaluation

The formulated herbal mouth ulcer gel was evaluated visually and manually for different physical characteristics:

- Colour: Determined by visual inspection of the formulation.
- Odor: Assessed by smelling the formulation; should be pleasant and characteristic of herbal ingredients.
- Consistency: Evaluated by tactile application; the gel should be smooth and non-irritant.
- State: The formulation should be a homogeneous semisolid gel.
- Grittiness: Checked by rubbing between fingers to ensure absence of particulate matter.

Observation Table (Organoleptic Evaluation):

Sr.no	Property	F1	F2	F3
1	Colour	Light greenish gel	Light greenish gel	Light yellowhish gel
2	Odor	Herbal (pleasant)	Herbal (pleasant)	Herbal (pleasant)
3	State	Semisolid gel	Semisolid gel	Semisold gel

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4	Texture	Smooth	Smooth	Smooth
5	Grittiness	No grittiness	NO grittiness	NO grittiness

2. PH Determination

- 1 g of gel was dispersed in 10 mL of distilled water.
- The pH was measured using a calibrated digital pH meter.
- Measurements were performed in triplicate and the mean value was recorded.
- The ideal pH range for oral gel formulations is 6.0–7.0, which ensures compatibility with the oral mucosa and prevents irritation.

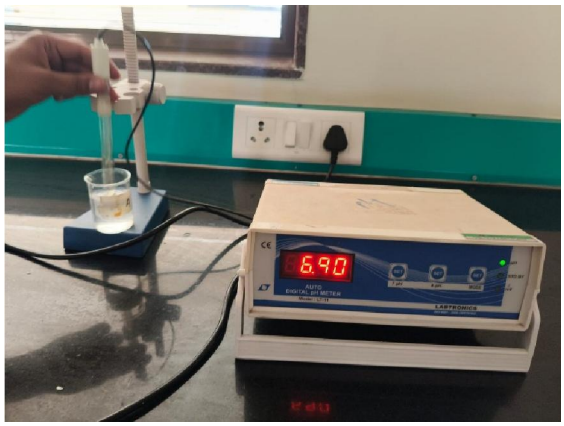

Fig no [10] Ph of Gel

3. Spreadability Test

Spreadability was determined to evaluate the ease of application of the gel.

Procedure:

- 1 g of gel was placed between two clean glass slides.
- A 500 g weight was placed on the upper slide for 1 minute.
- The diameter of spread was measured and recorded.
- Higher spreadability indicates better application and uniform distribution over oral mucosa.

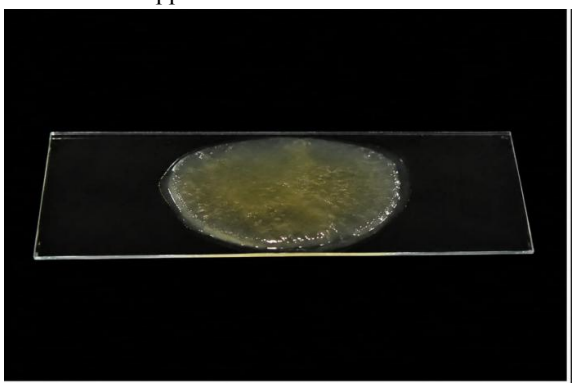

Fig no [11] Spreadability Test of Mouth Ulcer

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4. Consistency and Homogeneity

- The gel was manually inspected for uniformity.
- It should show smooth texture without phase separation.
- No oily or solid residue should be observed after application.

5. Washability Test

- The gel was applied to a clean surface and allowed to remain for a few minutes.
- It was then washed with tap water.
- **Observation:** The gel was **easily washable with water**, indicating good removal properties without residue.

Fig no [12] Washability Test

6. Irritancy Test (Patch Test)

Objective: To evaluate any irritation or allergic reaction on skin/mucosa.

Procedure:

A small amount of gel was applied to the skin (forearm or behind the ear) and observed for 24 hours.

Observation:

No redness, itching, swelling, or inflammation was observed. The formulation was found to be **non-irritant and safe**.

7. Stability Evaluation

- The gel was observed for physical stability at room temperature over a period of time.
- **Observation:** The formulation remained **stable with no phase separation, discoloration, or odor change**.

8. Antioxidant Activity

The gel exhibited antioxidant potential due to the presence of herbal extracts such as turmeric, tulsi, and giloy.

Observation: The formulation showed **good antioxidant activity**, indicating potential in wound healing and anti-inflammatory action.

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9. Overall Evaluation Summary

Sr.no	Evaluation Parameter	Observation
1	Colour	Light Yellow
2	Odour	PleasantHerbal odour
3	Texture	Smooth and Uniform
4	PH	6.0-7.0
5	Spreadability	Good
6	Washability	Easily washable with water
7	Stability	Stable with no Phase separation
8	Irritation	No irritation observed
9	Antioxidant activity	Good antioxidant property

II. CONCLUSION

In conclusion, the present study successfully formulated and evaluated a herbal mouth ulcer gel containing extracts of Tulsi, Giloy, Aloe vera, and Turmeric. The antioxidant activity assessed by the DPPH/FRAP assay confirmed that the herbal extracts possess significant free radical scavenging and reducing potential, indicating the presence of bioactive compounds responsible for healing and protective effects. The prepared gel showed satisfactory physicochemical properties, including suitable pH, good consistency, spreadability, homogeneity, and stability, making it appropriate for oral topical application. The formulation also demonstrated promising soothing and healing potential, which may help in reducing pain, inflammation, and irritation associated with mouth ulcers. Therefore, the developed herbal gel can be considered a promising natural formulation for the management of mouth ulcers. However, further studies such as long-term stability testing, antimicrobial studies, and clinical evaluation are recommended to confirm its safety, efficacy, and therapeutic effectiveness. concept of natural healing and herbal medicine has existed since ancient times, forming an integral part of traditional systems of medicine such as Ayurveda. Medicinal plants have been widely used for the treatment of various oral and systemic disorders due to their safety, effectiveness, and holistic healing properties. In recent years, there has been a growing interest in herbal formulations for oral care, especially for the management of mouth ulcers, as they are considered safer alternatives to synthetic drugs. Mouth ulcers are small, painful lesions that develop in the oral cavity and are commonly associated with factors such as stress, nutritional deficiencies, hormonal changes, minor injuries, or microbial infections. These ulcers often cause discomfort while eating, drinking, and speaking, thereby affecting the quality of life. Although several synthetic formulations are available for treatment, they may provide only temporary relief and can sometimes lead to adverse effects on prolonged use. Hence, herbal alternatives are gaining importance due to their safety, effectiveness, and minimal side effects.

The present herbal mouth ulcer gel is formulated using natural medicinal herbs such as Giloy, Tulasi, Aloe vera, and Turmeric. These herbal ingredients possess significant anti-inflammatory, antimicrobial, antioxidant, antibacterial, immunomodulatory, and wound healing properties that help in the treatment and prevention of mouth ulcers. Giloy is known for its immunomodulatory and anti-inflammatory activities, helping reduce pain and supporting the body's natural healing process. Tulasi exhibits strong antimicrobial, antiseptic, and antibacterial properties that control oral infections and inhibit microbial growth. Aloe vera provides soothing, cooling, and moisturizing effects, accelerates wound healing, and promotes regeneration of damaged oral tissues. Turmeric contains curcumin, a bioactive compound with strong anti-inflammatory, antioxidant, and antibacterial activities that promote rapid healing and prevent secondary infections. The formulated gel is designed to provide localized action at the affected site, ensuring prolonged contact with the oral mucosa and improved therapeutic effectiveness. It aims to reduce pain, inflammation, microbial load, and irritation while promoting faster healing in a natural and safe manner. Due to herbal ingredients, the gel offers better patient compliance with minimal side effects compared to synthetic preparations.

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