Formulation And Evaluation of Chebulinic Acid for Mouth Gargle

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ABSTRACT

Oral infections and dental diseases are common health concerns, often linked to microbial biofilms. Chebulinic acid, a bioactive tannin derived from *Terminalia chebula*, possesses potent antimicrobial, anti-inflammatory, and antioxidant properties.

This study focuses on the formulation and evaluation of a chebulinic acid-based mouth gargle for oral health applications. The formulation was designed to ensure optimal solubility, stability, and patient compliance. Various excipients were incorporated to enhance the effectiveness and palatability of the gargle while maintaining a pH range suitable for oral use. The gargle aims to prevent and manage oral infections, gingivitis, and dental plaque while promoting overall oral hygiene.

The formulation was optimized for pH, viscosity, and stability to ensure efficacy and patient compliance. Antimicrobial studies demonstrated significant inhibitory activity against oral pathogens such as *Streptococcus mutans*.

The results highlight the therapeutic potential of chebulinic acid as a natural alternative to conventional mouthwashes. Additionally, accelerated stability studies confirmed the formulation's integrity over time, making it a promising candidate for commercial production. Further clinical investigations are recommended to establish its long-term benefits and safety in diverse populations.

Keywords: Terminalia chebula, Gingivitis, Dental plaque, Oral hygiene, Mouth gargle.

1. INTRODUCTION

1.1 HIRDA [TERMINALIA CHEBULA]

Hirda (*Terminalia chebula*), commonly known as Haritaki in Sanskrit and Harad in Hindi, is a highly revered medicinal plant in Ayurveda and traditional medicine. Belonging to the Combretaceae family, this deciduous tree is found in tropical and subtropical regions of India, Nepal, Sri Lanka, and China. The dried fruits of Hirda are widely used for their therapeutic benefits, owing to their rich phytochemical composition, which includes tannins (chebulinic acid, chebulagic acid), flavonoids, glycosides, alkaloids, and terpenoids. It is known for its powerful antioxidant, antimicrobial, anti-inflammatory, and digestive properties. ^[1]

Hirda is considered a "Rasayana" or rejuvenator, balancing all three doshas—Vata, Pitta, and Kapha making it a versatile remedy. It is extensively used for promoting digestion, relieving constipation, boosting immunity, supporting liver function, managing diabetes, and improving brain health. As a key



ingredient in Triphala, it plays a crucial role in detoxification and overall well-being. Additionally, Hirda is applied externally for wound healing and skin care, making it a valuable component in both pharmaceutical and cosmetic formulations. Modern research continues to explore its potential in neuroprotection and anti-aging therapies. Available in various forms such as powder, capsules, decoctions, and oils, Hirda remains an essential part of herbal medicine, proving its significance in both traditional and contemporary healthcare.

Hirda is highly valued in traditional medicine and is known for the following therapeutic effects:^[5]



Fig. No.1: - Hirda Herbal Powder

- 1. Digestive Health: Acts as a mild laxative, relieves constipation, and promotes digestion.-
- 2. Antioxidant Effects: Protects cells from oxidative damage, slowing aging and reducing the risk of chronic diseases.
- 3. Liver Protection: Supports liver detoxification and improves hepatic function.
- 4. Immunity Booster: Strengthens the immune system and enhances the body's resistance to infections.
- 5. Respiratory Benefits: Helps in treating cough, asthma, and bronchitis.
- 6. Anti-Inflammatory and Analgesic: Reduces inflammation and pain in conditions like arthritis.
- 7. Diabetes Management: Helps regulate blood sugar levels.
- 8. Wound Healing: Applied externally to heal wounds, cuts, and ulcers.
- 9. Brain Health: Used in Ayurvedic formulations for cognitive enhancement and neuroprotection.
- 10. Eye Care: Beneficial in treating eye infections and improving vision.

1.2 Chebulinic Acid: A Multi-Targeted & Potent Natural Tannin

Chebulinic acid is a hydrolyzable tannin and one of the major bioactive compounds found in *Terminalia chebula* (Hirda or Haritaki). It is known for its diverse pharmacological properties, including antioxidant, antimicrobial, anti-inflammatory, anti-diabetic, hepatoprotective, and neuroprotective effects. Chebulinic acid is classified as an ellagitannin, a type of polyphenol known for its strong free radical scavenging and therapeutic potential.^[1]

1.2.1 Chemical Structure and Properties



- IUPAC Name: [(2R,3R)-2,3,4,5-tetrahydroxy-6-oxooxan-2-yl] 3,4,5-trihydroxybenzoate ^[1]
- Molecular Formula: C41H32O27
- Molecular Weight: 956.68 g/mol



- Chemical Classification: Hydrolysable tannin (Ellagitannin)
- Solubility: Water-soluble, soluble in polar solvents like ethanol and methanol
- Appearance: White to light brown amorphous powder
- **1.2.2 Sources and Extraction**



Fig.No.2 Fruits of T. Chebula

Chebulinic acid's primary source is: ^[2]

- Fruits of Terminalia chebula (Highest concentration)
- Other Terminalia species (T. bellirica, T. arjuna)
- Phyllanthus emblica (Amla) in smaller amounts

Extraction Methods:^[4]

- Solvent extraction (Methanol, Ethanol, Water)
- Chromatographic techniques (HPLC, TLC, LC-MS) for purification
- 1.2.3 Pharmacokinetics and Bioavailability
- Absorption: Limited due to its large molecular structure; gut microbiota convert it into smaller metabolites.
- Metabolism: Hydrolysed in the intestine to ellagic acid and gallic acid, which retain bioactivity.
- **Excretion:** Primarily through urine and bile.
- **1.2.4 Industrial and Commercial Applications**
- **Pharmaceutical Industry:** Used in Ayurvedic medicines (Triphala), anti-diabetic formulations, and neuroprotective supplements.
- Cosmetic Industry: Included in anti-aging creams due to its collagen-protecting properties.
- Food Industry: Used as a natural preservative for its antimicrobial effects.

1.2.5 Safety, Toxicity, and Side Effects

- Generally safe in moderate doses.
- High doses may cause:
- a) Mild gastrointestinal issues (diarrhea, bloating).

b) Potential hypoglycemia in diabetic patients.

• Pregnancy and lactation safety not well studied—use with caution.

1.3 Oral Cavity-Related Problems:

The oral cavity is the gateway to overall health, and maintaining proper oral hygiene is crucial for preventing various dental and periodontal diseases. Common oral cavity issues include **gingivitis**, **bad breath (halitosis)**, **plaque buildup**, **cavities**, **and periodontitis**, all of which can significantly impact oral and systemic health if left untreated.^[3]



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Fig.No.3 Common Oral Cavity Related Problems

1. Gingivitis: The Early Stage of Gum Disease

Gingivitis is the mildest form of gum disease, characterized by inflammation of the gums due to plaque accumulation along the gumline.

2. Bad Breath (Halitosis)

Halitosis refers to persistent bad breath, usually caused by bacterial activity in the oral cavity, poor oral hygiene, or systemic health issues.

3. Plaque and Tartar Buildup

Plaque is a soft, sticky film of bacteria that accumulates on teeth and gums. If not removed, it hardens into tartar (calculus), which can lead to cavities and gum disease.

4. Cavities (Dental Caries)

Cavities are permanently damaged areas on the surface of teeth caused by acid-producing bacteria that erode tooth enamel.

5. Periodontitis (Advanced Gum Disease)

Periodontitis is a severe gum infection that destroys the bone and soft tissues supporting teeth, leading to tooth loss if untreated.

1.3.1 Causes

- Poor oral hygiene (improper brushing and flossing)
- Bacterial infections
- Hormonal changes (pregnancy, puberty, menopause)
- Smoking and tobacco use
- Vitamin deficiencies (especially Vitamin C deficiency)
- Consumption of pungent foods (onions, garlic, dairy, coffee)
- High sugar and carbohydrate intake

1.3.2 Sign and Symptoms

- Persistent unpleasant odour
- Dry mouth
- Thick coating on the tongue
- Bleeding gums, especially during brushing or flossing
- Tooth sensitivity to hot, cold, or sweet foods

1.3.3 Treatment with aid of mouth gargles

Mouth gargles are an effective way to manage various oral health issues such as gingivitis, bad breath (halitosis), plaque buildup, and oral infections. They help in reducing bacteria, freshening breath, soothing inflammation, and preventing dental diseases.

Comprehensive guide to treat these conditions effectively are: -



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- 1. Proper oral hygiene practices (Brushing, Flossing, Mouth Rinsing, Tongue cleaning)
- 1. 2. Medicinal Treatments (Antiseptic, Antibiotics, Antifungal Mouth gargles)
- 2. Professional Dental Treatments (Scaling & Root Planing, Dental Fillings)
- 3. Home Remedies & Natural Treatments (Clove oil application)

1.4. MOUTH GARGLES

Mouth gargles are liquid solutions used to rinse the mouth and throat to maintain oral hygiene, reduce infections, soothe throat irritation, and freshen breath. Mouth gargles serve as antiseptics by killing bacteria, viruses, and fungi, helping to prevent infections. Regular use of gargles aids in oral hygiene by reducing plaque, bad breath, and gum diseases. They also provide a soothing effect, relieving sore throat, inflammation, and irritation. Some gargles support healing, especially after dental procedures or oral surgeries.^[6]

1.4.1 Ideal Properties of Gargles

- 1. It should be non-toxic and non-irritant.
- 2. It should have a refreshing flavour to encourage use.
- 3. It should be economical.
- 4. It should not cause discoloration of teeth or oral tissues.
- 5. It should be environmentally friendly.
- 6. It should be quality supreme and effective.

1.4.2 Classification of Gargles

- Based on its function:
- Antiseptic Gargles
- Analgesic Gargle
- Analgesic Gargle
- Fluoride Gargles
- Based on Composition:
- o Herbal Gargles
- Chemical-Based Gargles
- Based on the Mechanism of Action:
- Anti-Inflammatory Gargles
- Bactericidal Gargles
- Bacteriostatic Gargles
- Based on Usage:
- o Cosmetic Gargle
- Therapeutic Gargles
- Prescription Gargle
- o Over-the-Counter (OTC) Gargle



2. PLANT PROFILE



Fig No. 4 SEEDS OF HIRDA Fig. No. 5 LEAVES OF HIRDA Fig. No. 6 FLOWERS OF HIRDA

Hirda also known as Haritaki, is a powerful medicinal plant widely used in Ayurveda. It is one of the key ingredients of **Triphala** and is known for its **rejuvenating**, **digestive**, **and detoxifying** properties. ^[2]

2.1 SYNONYM (COMMON NAMES): - Harad (Hindi), Haritaki (Sanskrit), Hirda (Marathi)

2.2 SCIENTIFIC NAME: - Terminalia chebula

2.3 BIOLOGICAL SOURCE: -It is obtained from the dried fruits of *Terminalia chebula*.

2.4 GEOLOGICAL SOURCE: -South Asia, including India, Nepal, Sri Lanka, China.^[9]

2.5 TAXONOMICAL CLASSIFICATION

- Kingdom: Plantae
- Phylum: Angiosperms
- Class: Eudicots
- Order: Myrtales
- Family: Combretaceae
- Genus: Terminalia
- Species: Terminalia chebula

2.6 BOTANICAL DESCRIPTION

Morphology- It is a medium to large deciduous tree, growing up to 20–30 meters in height with a broad, spreading canopy. The bark is dark brown and often cracked.

Leaves- The leaves are simple, ovate to elliptic, with a smooth margin and prominent veins, arranged in opposite pairs.

Flowers- The flowers are small, pale yellow to greenish, and occur in terminal or axillary spikes.

Fruits- The fruit is a drupe, oval to ellipsoidal in shape, about 2–4 cm long, and turns yellowish-brown upon ripening.

Seeds- The seed is single, enclosed within the fleshy pericarp of the fruit.

2.7 CHEMICAL CONSTITUENTS ^[5]

- 1. Tannins: (Chebulagic acid, Chebulinic acid, Gallic acid, Ellagic acid)
- 2. Flavonoids
- 3. Terpenoids
- 4. Glycosides
- 5. Saponins

2.8 USES:

1. Antioxidant



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- 2. Anti-inflammatory
- 3. Antimicrobial
- 4. Hepatoprotective
- 5. Laxative
- 6. Digestive Stimulant
- 7. Immunomodulatory

3. MATERIAL & METHODS 3.1 LIST OF MATERIALS

Sr. No.	Name of Chemical	Category	Company
1.	Hirda Powder	API	Saurveda
2.	Ethanol	Solvent	Advita Lifesciences
3.	Glycerine	Humectant	Neurochem
4.	Sodium Benzoate	Preservative	Research Labfine Chem
5.	Peppermint Oil	Flavoring agent	Suvidhinath Laboratories
6.	Distilled Water	Solvent	BRM Chemicals

Table 1: List of Chemicals and Grade

3.2 LIST OF INSTRUMENTS

Sr. No.	Name of Instrument	Brand Name
1.	pH meter	Systronics, Globe Instruments
2.	Brookfield viscometer	Brookfield Engineering Laboratories
3.	Digital Balance	Kerro Series P6D
4.	Mixer Grinder	Jyoti
5.	Digital Autoclave	ASI-254
6.	B.O.D Incubator	HMG India
7.	Heating Mantle	Biotechnics India
8.	Water Bath	Labline
9.	Microscope	Metzer M, Imperial India
10.	Muffle Furnance`	Bio Technics India
11.	Hot Air Oven	Insif India

Table 2: Instrument Used

3.3 DRUG, EXCIPIENTS AND INSTRUMENTS

Fresh powder of Hirda (Terminalia chebula) was selected for the experimental study. The powder was



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acquired from offline market source.

All the other ingredients like: Ethanol, Distilled Water, Methanol (HPLC grade), Hydrochloric acid, Ethyl acetate, Sodium benzoate, Glycerine, Menthol, Sorbitol, Sodium chloride, Tween 80, Saccharin sodium etc. were supplied from the Laboratories of Pharmaceutics & Pharmacognosy, MIBP, Gondia.

All the chemicals used in the formulation are of analytical grade.

All the Instrumentation like: - Rotary evaporator, UV-Spectrophotometer, HPLC, FTIR, pH meter, Viscometer, etc. are calibrated.^[7]

Preparation of Chebulinic Acid Extract

The extraction of Chebulinic Acid is done using Soxhlet apparatus.

Solvent used for extraction is Ethanol.

The process is continued for several hours to ensure maximum yield.

The extract is filtered to remove solid residues and then concentrated using a rotary evaporator or under reduced pressure to obtain a semi-solid or pure extract. ^[7]

Role of Drugs & Excipients

- Chebulinic Acid Extract: It is used for its antimicrobial & antioxidant property.
- Ethanol: It is used as solubilizer as well as solvent.
- Glycerine: It is used as Humectant & Texture Enhancer.
- Menthol: It is used for cooling effect.
- Sodium Benzoate: It is used as preservative.
- **Distilled water**: It is used as solvent.
- **Peppermint oil**: It is used as Flavoring agent.

3.4 CHARECTERIZATION OF HIRDA POWDER

Objective: To evaluate the physiochemical properties of Hirda (Terminalia Chebula) powder.

3.4.1 Moisture Content Determination [Loss on Drying/ LOD]^[12]

Procedure:

Weigh 2.5g of Hirda powder. Place the sample in pre-weighed moisture dish. Dry in Hot air oven at 105°C for 5hrs. Cool the dish in desiccator to room temperature.. Weigh the dried sample.Calculate the moisture content by using following formula:

$$Moisture\ Content[LOD](\%) = \frac{Initial\ wt. -Dried\ wt.}{Initial\ wt.} \times 100$$

3.4.2 Ash Value [Total ash value]

Procedure:

Weigh 2g of Hirda powder. Place the sample in pre-weighed silica crucible. Incinerate to 450°C in muffle furnance until it turns white (indicating absence of carbon). Allow it to cool at room temperature. Weigh the residue ash. Calculate the ash value by following formula:

$$Total Ash Value(\%) = \frac{Weigh of Ash}{Weigh of sample} \times 100$$



3.4.3 Determination of foreign matter

Procedure: 1g of sample is weighed & spread over the butter paper. Observe the foreign particles by naked eye & separate it. Then calculate the % of foreign particle.

3.4.4 Determination of Extractive value

Procedure:

Weigh 10g of Hirda powder.

Extraction: Extract it with 95% Ethanol in stoppered flask. Filtration: Filter the solution using filter paper & discard first 10ml of filtrate. Evaporation: Evaporate the filtrate to dryness in drying oven at $105^{\circ}C \pm 2^{\circ}C$. Weighing: Weigh the residue accurately.

$$Extractive \ value(\%) = \frac{Weigh \ of \ Residue}{Weigh \ of \ sample} \times 100$$

3.5 EXTRACTION OF HIRDA POWDER^[4]

Extraction of Hirda powder is done using Soxhlet apparatus using Ethanol (90%) as solvent.



Fig. No. 7 Soxhlet Apparatus

Procedure: Soak 50g of Hirda powder in 250ml Ethanol for 24 hrs. After soaking or extraction, filter the solution using filter paper to remove solid material. The filtrate is crude extract. The filtered extract is then concentrated by distillation (evaporation) of solvent to obtain concentrated resin or oily extract. Triturate it to obtain solid powder. Calculate the % yield using following formula:

% Yield = $\frac{Final wt. of Hirda(after extraction)W2}{Initial wt. of Hirda(before extraction)W1} \times 100$

3.6 PRELIMINARY PHYTOCHEMCAL SCREENING OF CHEBILINIC ACID EXTRACT

Objective: To identify and analyse the bioactive compounds present in plant extracts. This process helps in determining the potential medicinal and therapeutic properties of a plant.^[15]

3.6.1 TEST FOR ALKALOIDS

Dragendorff's Test: To 2-3ml filtrate, add few drops Dragendorff's reagent.



Wagner's Test: 2-3ml filtrate with few drops Wagner's reagent gives reddish brown colour.

3.6.2 TEST FOR PHENOLS

FeCl₃ solution: Deep blue-black colour.

3.6.3 TEST FOR FLAVONOIDS

Sulphuric Acid Test: On addition of sulphuric acid, flavones and flavono dissolve into it and give a deep yellow solution. Chalcones and aurones give red or red-bluish solutions. Flavanes give orange to red colours.

3.6.4 TEST FOR PROTEINS

Millon's Test: Mix 3ml T.S. with 5ml Millon's reagent. White ppt. turns brick red or the ppt. dissolves giving red coloured solution.

3.6.5 TEST FOR CARBOHYDRATES

Fehling's Test (For Reducing sugar): Mix 1ml Fehling's A & 1ml Fehling's B solutions, boil for one minute. Add equal volume of test solution. Heat in boiling water bath for 5-10 min. First yellow, then brick red ppt. is observed.

Barfoed's Test (For Monosaccharides): Mix equal volume of Barfoed's test and test solution. Heat for 1-2 min.in boiling water bath and cool. Red ppt. should observe.

3.6.5 TEST FOR AMINO ACIDS

Ninhydrin Test (General test): Heat 3ml T.S. and 3 drops 5% Ninhydrin solution in boiling water bath for 10min.Purple or bluish colour appears.

3.6.6 TEST FOR GLYCOSIDES

Foam Test: Shake the drug extract or dry powder vigorously with water.

3.7 PREPARATION OF MOUTH GARGLE

- 1. The Chebulinic Acid Mouth Gargle can be prepared by following procedure: -
- 2. Dissolve chebulinic acid extract in a small amount of ethanol (10-15%) with stirring.
- 3. Heat gently (below 50°C) if needed to ensure complete dissolution.
- 4. Dissolve sodium benzoate in warm distilled water (40-50°C) with continuous stirring.
- 5. Add glycerin to the sodium benzoate solution and mix well.
- 6. Add peppermint oil dropwise into the ethanol-chebulinic acid mixture while stirring.
- 7. Slowly add the chebulinic acid-ethanol-peppermint oil solution into the aqueous base (sodium benzoate + glycerin + water) while stirring.
- 8. Make up the final volume with distilled water and mix until uniform.
- 9. Filter the solution to remove any undissolved particles.
- 10. Filter the solution to remove any undissolved particles.
- 11. Transfer to amber-colored sterilized bottles to protect from light.^[9]



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Fig. No.8 PREPARATION OF MOUTH GARGLE

3.7 Table NO.3 FORMULA FOR COMPOSITION OF MOUTH GARGLE

Sr. No.	Ingredients	F1	F2	F3
1)	Chebulinic Acid Extract	0.1g	0.5g	1g
2)	Ethanol	10ml	15ml	13ml
3)	Glycerine	12ml	9ml	15ml
4)	Peppermint Oil	0.1ml	0.1ml	0.1ml
5)	Sodium Benzoate	0.2ml	0.2ml	0.2ml
6)	Distilled Water	q.s.	q.s.	q.s.
Total Fo	rmulation	100ml	100ml	100ml

3.8 EVALUATION PARAMETERS OF GARGLE> PHYSIOCHEMICAL EVALUATION OF PARAMETERS:

Name of Study	Observed Result	Reported Standard
Appearance	Clear and Uniform	Clear and uniform
Colour & Odor	Pale Yellow and pleasant	Characteristic color and odor



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Name of Study	Observed Result	Reported Standard	
Taste & Mouthfeel	No foul taste and unpleasant aftertaste	No burning sensation, irritation, or unpleasant aftertaste	
Homogeneity	Uniform	Uniform without phase separation	
рН	5.5	5.0 - 7.5	
Viscosity	60	1-200 cP (Depending on formulation)	
Acid Value Within limit		Should be within acceptable limits	
Saponification Value		Should be within acceptable limits	
Irritancy Test	No irritancy	No redness, swelling, ulcers, lesions, or irritation	
Antimicrobial Activity	Present	Zone of inhibition should be present against S. aureu	
boamability Test Mild Foaming Foam height should be stable and within range		Foam height should be stable and within acceptable range	

4. RESULT

4.1 Moisture Content Determination [LOD]

Calculation: Initial wt.=36.24g & Dried wt.=36.06g

Moisture Content[*LOD*](%) =
$$\frac{36.24g - 36.06g}{36.24g} \times 100 = 0.49\%$$

4.2 Ash value [Total Ash Value]

Calculation: Wt. of Ash=35.95 & Wt. of sample=37.73g

$$Total Ash Value(\%) = \frac{35.95g}{37.73g} \times 100 = 95.28\%$$

4.3 Determination of foreign particle

Result: No foreign particle was observed in Hirda powder.

4.4 Determination of Extractive Value

Calculation: Wt. of Residue=0.23g &

Wt. of hirda powder used for extraction=30g

Extractive value(%) =
$$\frac{0.23g}{30g} \times 100 = 0.7\%$$

Table No. 4	Pre-formulation	Studies
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Sr. No.	Property	Result
1.	Moisture Content Determination [LOD%]	0.49%
2.	Ash value (Total)	95.28%
3.	Determination of foreign particle	No Foreign particle
4.	Extractive Value	0.7%



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Sr.	Phytochemical	Test Performed	Colour	Inference
No.	Test		Observed	
1.	Alkaloids	A] Dragendorff's	Reddish Brown	Presence of alkaloids
		Test	ppt	
		B] Wagner's Test	Reddish Brown	Presence of alkaloids
			ppt	
2.	Phenols	C] FeCl ₃ solution	Deep Blue-	Presence of phenol
			Black	
3.	Flavanoids	D] Sulphuric Acid	Deep yellow	Presence of
		Test		flavonoids
4.	Proteins	E] Millon's Test	Brick Red	Presence of proteins
5.	Carbohydrates	F] Fehling's Test	Brick Red	Presence of reducing
		(Reducing Sugar)		sugars
		G] Barfoed's Test	Red ppt.	Presence of
		(Monosaccharides)		Monosaccharides
6.	Amino acids	H] Ninhydrin Test	Purple/Bluish	Presence of Amino
				acid
7.	Glycosides	I] Foam Test	Foaming	Presence of
			observes	glycosides

Table No. 5 Pre-liminary Phytochemical Screening

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Fig. No. 9 Pre-liminary Phytochemical Screening



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Table No. 6 Determination of Organoleptic Characteristics of Prepared Gargle

Sr no.	Properties	Observations
1.	Colour	Pale Yellow
2.	Appearance	Clear or slightly turbid liquid
3.	Odour	Pleasant, characteristic herbal odour
4.	State	Liquid
5.	Texture	Smooth, slightly viscous
6.	Clarity	Transparent to slightly translucent
7.	Foaming Ability	Mild foaming
8.	Homogeneity	Homogenous



Fig. No. 10 Formulated Gargle [F1, F2 & F3]

	Table No.	7 pH Determination	
Property	F1	F2	F3
pН	4.52	6.01p	5.56



Fig. No. 11 pH meter



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Table No. 8 Viscosity Determination

Property	F1	F2	F3
Viscosity	34 cP	36 cP	42 cP

Fig. No. 12 Brookfield Viscometer



Table No. 9 Irritancy Testing

Property	F1	F2	F3
Irritancy	No Irritancy	No Irritancy	No Irritancy

Table No. 10 Specific Gravity Test

Property	F1	F2	F3
Specific Gravity	1.01 g/mL	0.98 g/mL	1.05 g/mL

Table No. 12 Anti-microbial Evaluation ^[10]

Sr.no	Zone of Inhibition		
	Marketed formulation	Herbal Preparation	
	(Streptomycin)	(Chebulinic Acid Gargle)	
F1	18mm	17mm	
F2	21mm	23mm	
F3	19mm	16mm	

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