

Formulation and Evaluation of Herbal Mouthwash of Acmella Olaraceae

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Abstract

Since herbal mouthwash acts on oral bacteria and pathogens, instantly relieves pain, and has no unfavorable side effects, it is more widely used than chemical mouthwash. At various stages of their life, the most common infectious diseases brought on by a variety of viruses and microbes include tooth caries and periodontal disorders. The present work aims to develop it and evaluate its effectiveness against germs in the oral cavity. The mouthwash was made with four different herbs Acemella Oleracea (jambu), clove (laung), gauva leaves (peru) and tulsi (tulas). Physical properties like color, stability, and pH were then evaluated for the created product. The mouthwash that is now available has strong antimicrobial qualities. Under a range of temperature conditions, this formulation stays stable. The liquid mouthwash that is currently on the market usually contains analgesic and local anesthetic components. These treatments can reduce microbial growth and infections of the oral cavity.

Keywords: Acmella Oleracea, Clove, Analgesic, Local Anesthatic

***** INTRODUCTION-

In India and other impoverished countries, a significant portion of dental problems are caused by microbial disease.oral hygiene is an essential part of overall health since the condition of one's mouth has a significant influence on one's overall wellbeing^{(1).} Nowadays, when dietary choices and lifestyle choices often result in a range of oral health problems, there is a greater need than ever for effective dental care solutions Practicing good oral hygiene is essential because issues like cavities, gum disease, and foul breath are widespread and impact millions of individuals worldwide⁽²⁾.Dental plaque is a complex biofilm that forms on the surface of teeth and is made up of more than 500 different types of bacteria. These bacteria first colonize the salivary film of the enamel before colonizing again through antibacterial adherence. Disorders of prenominals affect the tissues that support teeth, and because gingivitis is characterized by bleeding and inflammation of the gums, it is important to ensure that the least amount of discomfort during treatment is experienced. A significant medicinal plant, Spilanthes acmella (S. acmella) is also known as the toothache plant. Its antibacterial, anti-inflammatory, analgesic, and anesthetic qualities are all present. In dentistry, we talk about how well S. acmella extract works as a local anesthetic⁽³⁾.

International Journal on Science and Technology (IJSAT)

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> ROLE OF HERBAL MOUTHWASH IN DIFFERENT ORAL DISORDERS:

- Gingivitis: Herbal mouthwashes can help reduce inflammation and plaque accumulation, which are key factors in gingivitis. Ingredients like neem and clove oil are known for their antibacterial properties, aiding in the control of oral bacteria and promoting gum health.
- Periodontitis: The anti-inflammatory and antimicrobial effects of herbal mouthwashes can assist in managing periodontitis by reducing gum inflammation and preventing further tissue damage. Regular use can help maintain periodontal health and support healing.
- Halitosis (Bad Breath): Herbal mouthwashes containing ingredients peppermint and fennel can effectively combat bad breath by neutralizing odor-causing bacteria providing a fresh scent. These natural agents to maintaining oral freshness throughout the day⁽⁴⁾.
- Oral Mucositis: For patients undergoing chemotherapy or radiate on therapy, herbal mouthwashes can provide soothing relief from oral mucositis. Ingredients like aloe vera and chamomile can help reduce irritation and promote healing of the oral mucosa
- Oral Candidiasis: Herbal mouthwashes with antifungal properties, such as those containing tea tree oil, can be effective in managing oral thrush by inhibiting the growth of Candida species in the oral cavity⁽⁵⁾.

*** OBJECTIVES:**

- 1. The primary goal of making a herbal mouthwash is to keep your mouth clean.
- 2. Prevention, control and reduction of oral infection
- 3.To reduce side effects by promoting herbal $use^{(6)}$.

PLAN OF WORK

Proposed Methodology:

- 1. Selection of the topic by reading research articles & reviews.
- 2. Finalise the topic & collect information about its methodology
- 3. List of materials & equipment.
- 4. Collection & Extraction of herbs
- 5. Identification test for extract
- 6. Formulation of herbal mouthwash
- 7. Evaluation of herbal mouthwash $(^{7, 8})$

***** Active ingredient used in herbal mouthwash:

1: AcmellaOleracea:





Fig. No.1: Acmella Oleracea

- Class: Magnoliopsida
- Family: Asteraceae
- Genus: Acmella
- Uses: Analgesic, local anesthetic
- Species: Acmella oleracea L3
- Common Name: Sanskrit: Akal kara

Marathi: Pipulka

English: Toothache plant, Paracress, Jambu.

- **Morphology:** The leaves of the plant are egg- shaped to triangular with a toothed leaf margin (5-11 cm long 4-8 cm wide) flowers emerging maroon to red, turning yellow-golden as they mature.
- **Chemical Constituent**: phenolic acids, glycosylated flavonoids, alkamides and fatty acids, steroidals, terpene alcohols, polyenoic fatty acids, palmitic acid ethyl esters, and hydrocarbons^{(9).}

2: Guava Leaves:



Fig. No.2: Guava Leaves

- Class: Magnoliopsida
- Family: Myrtceae
- Genus: Psidium Linnaeus
- Uses: Antibacterial properties
- **Species:** guajava Linnaeus
- Common Name: Sanskrit: Amratafalam or Perala
- English:Psidium guajava

Marathi: peru

• **Morphology:** The leaves of the guava Size are typically 7–15 cm long and 3–5 cm. Shape Guava leaves are elliptic to oblong in shape dark green in color. The upper surface of the leaf is smooth, while the lower surface is slightly downy

Chemical Constituent: Guava leaves contain a variety of chemical constituents, Essential oils, flavonoid, sesquiterpene, triterpenoid, coumarin, alkaloid, and tannin molecules^(10, 11).

3. Tulsi:





Fig. No.3 : Tulsi

- Class: Magnoliopsida
- Family: Lamiaceae
- Genus: Ocimum
- Uses: Antimicrobial Activity
- Species: Ocimum sanctum
- Common Name: Tulas and Tulsi (in Marathi)
- **Morphology:** Tulsi is an herbaceous plant. The leaves are simple, opposite, and have an ovate or elliptical shape. The margins of the leaves can be smooth or slightly toothed. The flowers of Tulsi are small, arranged in terminal spikes or clusters. The fruit is a nutlet and is produced after flowering. The root system is generally fibrous. The leaves release a pleasant and distinct aroma when crushed.
- Chemical Constituent: Tulsi contains essential oils, phenolic compound, flavonoids, triterpenoids, alkaloids, vitamins, adaptogens, Antimicrobial compounds^(12, 13).

4 Clove:



Fig. No.4: Clove

- Class: Magnoliopsida
- Family: Myrtaceae
- Genus: Syzygium
- Common Name: Clove (English)

Lavang (Marathi)

Laung (Hindi)

- **Species:** Syzygium aromaticum
- **Morphology:** The leaves are opposite, simple, and aromatic. The leaves are dark green and glossy on the upper surface and lighter green on the lower surface. Clove flowers are small and are produced in terminal clusters
- **Chemical Constituent:** Clove contains eugenol, acetaeugenol, caryophyllene, isoeugenol, methyleugenol, tannins, flavonoids, gallic acid, quercetin⁽¹⁴⁾.



***** EXCIPIENT PROFILE:

1 Propylene glycol:⁽¹⁵⁾



Fig No.5 propylene glycol

- **IUPAC Name:** Propylene glycol
- Chemical Formula: C3H8O2
- Appearance: Colourless liquid
- Melting point: -59 °c
- Boiling Point: 188.2 °c
- **Density:** 1.036 g/cm3
- Viscosity: 0.042 Pa·s

2.Glycerin:⁽¹⁵⁾



Fig. no. 6: Glycerin

- IUPAC Name: Propane-1,2,3-triol
- Chemical Formula: C3H8O3
- Appearance: Colourless hygroscopic liquid
- Melting point: 17.8 °C
- Boiling Point: 290 °C
- Density: .261 g/cm3
- Viscosity: 1.412 Pa·s

3. Sodium Lauryl Sulphate (SLS):⁽¹⁶⁾





Fig no.7: Sodium Lauryl Sulphate (SLS)

- IUPAC Name: α-Sulfo-ω-(dodecyloxy)-poly(oxyethane-1,2-diyl), sodium salt
- Chemical Formula: CH3(CH2)11(OCH2CH2)nOSO3Na
- Appearance: Crystalline powder
- Melting point: 206°C
- **Boiling Point:** 288.4°C
- **Density:** 1.01 g/cm3
- Viscosity: 1.39mPa·s

4 Menthol:⁽¹⁷⁾



Fig no.8: menthol

- **IUPAC Name:** 5-Methyl-2-(propan-2-yl) cyclohexan-1-ol
- ChemicalFormula: C10H20O
- **Odor:** mint-licorice
- Melting point: 36-38°C
- **Boiling Point:** 214.6°C
- **Density:** 0.890 g·cm-3
- Viscosity: 17 mPa·s

Extraction method:

The aerial part was dried under shade. Coarse powder Acmella Oleraceais prepared with the help of mixer grinder. Dried powder of tulsi, guava leaves menthol and clove are also prepared by grinder. These powders are separately packed into an airtight container and stored in a cool and dry place. This material was used for further study⁽¹⁸⁾.

✤ PREPARATION OF PLANT EXTRACTS

- 1. The powder drug (sample)was placed in the extraction thimble. The Soxhlet apparatus was assembled by placing the thimble in the extractor and attaching it to⁽¹⁹⁾.
- 2. The Ethanolin the round-bottom flask was heated. The solvent vaporized and traveled up into the condenser. the condenser cooled the vapor, converting it back into liquid, which dripped onto the solid sample in the thimble. The solvent extracted the desired compounds as



it filtered through the sample. Once the solvent level reached the siphon arm, it automatically siphoned back into the flask, carrying the extracted compounds with it.

- 3. The process was repeated multiple times, continuously cycling the solvent through the sample until extraction was complete.
- 4. After the desired number of cycles, the extraction process was stopped. The solvent

containing the extracted compounds was collected from the round-bottom flask⁽²⁰⁾.



Fig No .9 Soxhlet apparatus

Formulation Table of Herbal Mouthwash

Sr.No	Ingredients	Uses	F1	F2
1	Acmella Oleracea	Analgesic,Local anesthatic	5ml	5ml
2	Guava leaves	Antioxidant	2.5ml	2.5ml
3	Tulsi	Antimicrobial	2.5ml	2.5ml
4	Clove	Analgesic	2ml	2ml
5	Sodium Lauryl Sulphate	Foaming agent	0.25gm	0.5gm
6	Propylene Glycol	Surfactant	5ml	7.5ml
7	Glycerin	Emulsifying agent	5ml	6.25ml
8	Menthol	Flavouring agent	Q. S	Q.S
9	Water	Vehicle	Q. S	Q. S

*** PREPARATION OF MOUTHWASH:**

Acmella Oleracea extract, Clove extract, guava leaves extract, and, Tulsiextractwas taken in beaker. Then sodium lauryl sulphate (foaming agent), menthol and propylene glycol (surfactant) wasadded with continuous stirring, followed by the addition of glycerin to mask the bitter taste.Finally, water was added to make up the volume, as well as a preservative, and the product was packaged in an attractive, wellclosed container.



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Fig No.10 Herbal mouthwash

& EVALUATION OF HERBAL MOUTHWASH:

1. Physical Evaluation:

- **Colour:** The mouthwash was observed for consistent color and the absence of discoloration.
- **Odor:** The mouthwash was evaluated for a pleasant and characteristic odor, typically minty or herbal, depending on the formulation.
- **Clarity:** The solution was observed for clarity, ensuring no suspended particles or cloudiness were present.
- **Texture:** The formulation was observed for texture.

1. pH:

The pH of prepared herbal mouthwash was measured by using digital pH meter. The pH meter was calibrated using standard buffer solution. About 1ml of mouthwash was weighed and dissolved in 50 ml of distilled water and its pH was measured^{(21).}

2. Foam Ability:

1ml of mouthwash was taken and dissolved in distilled water (about 50ml) in 100 ml graduated measuring cylinder. The measuring cylinder was shaken for about 10 minutes. Foam height was measured after 10 minutes and observation were recorded⁽²²⁾

3. Test for microbial growth:

The mouthwash formulation was streak plate inoculated on agar media plates, and a control was made. In the incubator, the plates were put. Then it was incubated for 24 hours at 37°C.Plates were removed after the incubation time and tested for microbiological growth⁽²³⁾.

4. Stability studies:

A stability study was performed on herbal mouthwash, following ICH guidelines. The mouthwash was stored in a closed container at room temperature for 30 days. The evaluation was based on change in colour, odour, texture and pH analysis to assess the stability of the herbal mouthwash⁽²⁴⁾.

5. Determination of Viscosity:

The internal barrier to fluid flow is known as viscosity. the ostwald viscometer was utilized to ascertain the viscosity of prepared formulation. the ostwald viscometer, dried, cleaned, and clamped



vertically. using an ostwald viscometer, lmounted vertical position on a suitable stand. Mouthwash was filled in to the viscometer up to mark a. the time was counted for mouthwash to flow from A to mark B. the time was measured with a stopwatch⁽²⁵⁾.

*** RESULT:**

Sr.No	Evaluation parameter	Formulation 1	Formulation 2
1	colour	Brownish	Brownish
2	odour	Sweet and spicy	Sweet and spicy
4	clarity	clear	clear
5	Viscosity	1.28	2.27

6. pH of formulation:

The formulation's pH was found to be 6.70, which is in line with the scheme's acidic pH of roughly 5.5. This pH range of formulation is suitable for oral problems. Eating the right foods is the best method to keep your mouth's pH level in a healthy range. Our gums and teeth are directly impacted by the pH level in our mouths.

You can lower your risk of tooth decay, gum disease, and cavities by managing the pH in your mouth.





7. Growing microbial growth in plate

After preparing the soil solution sample in the test tube, transfer the solution onto the plate using a nichrome wire. After carefully rubbing the lid over the agar in a few zigzag motions, replace the dish's lid. Allow the dish to remain in the incubator for three to seven days before the bacteria start to grow. Make a drawing and write a note every day description to record the expansion. A single bacterium cannot be seen without a strong microscope, however bacterial colonies can be seen. You can identify different bacterial species by looking at the colonies' color and structure. Next, make a hole in the battery plate so that the solution from the formulation may enter.



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Sr.no	Parameter		Observation	
		Time	F1	F2
		(days)		
	Colour	0	Brownish	Brownish
1		15	Brownish	Brownish
		30	Brownish	Brownish
	Odour	0	Pleasant	Pleasant
2		15	Pleasant	Pleasant
		30	Pleasant	Pleasant
	Texture	0	Liquid	Liquid
3		15	Liquid	Liquid
		30	Liquid	Liquid

Figno.12Microbial growth

8. Stability study:

The stability studies of the formulated mouthwash batches (F1 to F2) over 30 days at room temperature revealed consistent observations. All batches maintained a Brownishcolour and pleasant odour throughout the study period.

Conclusion:

Herbal mouthwashes demonstrate significant effectiveness in addressing a wide range of dental conditions, including decay, gingivitis, sensitive teeth, root infections, bad breath, and enamel erosion, among others. They are formulated without hazardous substances, making them a safer alternative to synthetic mouthwashes, which can lead to dry mouth, tooth discoloration, and altered taste perception over time. The ease of use and preventive capabilities of herbal mouthwashes make them a convenient solution for maintaining oral hygiene. However, while this study underscores their potential for widespread use, further research with larger sample sizes is necessary to fully establish their efficacy, particularly in low-socioeconomic settings.

REFERENCES

1. Peres MA, Macpherson LM, Weyant RJ, Daly B, Venturelli R, Mathur MR, et al. Oral diseases: a global public health challenge. The Lancet. 2019;394(10194):249-60.



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- 2. Chan WSH. The role of oral health in the prevention of systemic diseases. Universal Library of Medical and Health Sciences. 2024;1(1).
- 3. Dubey S, Maity S, Singh M, Saraf SA, Saha S. Phytochemistry, pharmacology and toxicology of Spilanthes acmella: a review. Advances in Pharmacological and Pharmaceutical Sciences. 2013;2013(1):423750.
- 4. De Geest S, Laleman I, Teughels W, Dekeyser C, Quirynen M. Periodontal diseases as a source of halitosis: a review of the evidence and treatment approaches for dentists and dental hygienists. Periodontology 2000. 2016;71(1):213-27.
- 5. Worthington H, Clarkson J. Prevention of oral mucositis and oral candidiasis for patients with cancer treated with chemotherapy: cochrane systematic review. Journal of Dental Education. 2002;66(8):903-11.
- 6. Kukreja BJ, Dodwad V. Herbal mouthwashes-a gift of nature. Int J Pharma Bio Sci. 2012;3(2):46-52.
- 7. Chan K, Shaw D, Simmonds MS, Leon CJ, Xu Q, Lu A, et al. Good practice in reviewing and publishing studies on herbal medicine, with special emphasis on traditional Chinese medicine and Chinese materia medica. Journal of Ethnopharmacology. 2012;140(3):469-75.
- Nazliniwaty N, Laila L. Formulation and antibacterial activity of Plectranthus amboinicus (Lour.) Spreng leaves ethanolic extract as herbal mouthwash against halitosis caused bacteria. Open access Macedonian journal of medical sciences. 2019;7(22):3900.
- 9. Aktar MA, Bhuia MS, Molla S, Chowdhury R, Sarkar C, Al Shahariar M, et al. Pharmacological and phytochemical review of Acmella oleracea: a comprehensive analysis of its therapeutic potential. Discover Applied Sciences. 2024;6(8):412.
- 10. Kumar M, Tomar M, Amarowicz R, Saurabh V, Nair MS, Maheshwari C, et al. Guava (Psidium guajava L.) leaves: Nutritional composition, phytochemical profile, and health-promoting bioactivities. Foods. 2021;10(4):752.
- 11. Mitra S, Irenaeus T, Gurung M, Pathak P, editors. Taxonomy and importance of Myrtaceae. III International Symposium on Guava and other Myrtaceae 959; 2012.
- 12. Nahak G, Mishra R, Sahu R. Taxonomic distribution, medicinal properties and drug development potentiality of Ocimum (Tulsi). Drug Invention Today. 2011;3(6).
- 13. Mandal AK, Poudel M, Neupane NP, Verma A. Phytochemistry, pharmacology, and applications of Ocimum sanctum (Tulsi). Edible Plants in Health and Diseases: Volume II: Phytochemical and Pharmacological Properties: Springer; 2022. p. 135-74.
- 14. Cheikhyoussef A, Cheikhyoussef N, Rahman A, Hussein AA. Clove (Syzygium aromaticum) phenolics: Extraction, compositions, and biological activities. Clove (Syzygium aromaticum): Elsevier; 2022. p. 215-33.
- 15. Shya LC. Study of Glycerol Electrochemical Conversion into Added-Value Compounds: University of Malaya (Malaysia); 2016.
- 16. Paul T, Taylor T, Babu RS A. Sodium lauryl sulphate. British dental journal. 2019;227(12):1012-.
- 17. LAKE NL, AMARGADH-BHICHARI R. HERBAL NASAL ROLL-ON.
- 18. Pore AV, Bais SK, Shinde DN. Review on Herbal Monograph Preparation.



- 19. Shabna V. A Comparative Study of Microwave Assisted Extraction Versus Soxhlet Extraction in the Analysis of in Vitro Antioxidant Study of Michelia Champaca Linn Flowers: Rajiv Gandhi University of Health Sciences (India); 2010.
- 20. Joy P, Surya S, Aswathy C. Laboratory Manual of Biochemistry. Kerala Agricultural University, Pineapple research station, Vazhakkulam. 2015:124-5.
- 21. Kalyani P, Leelavathi L. Comparison between the effect of plain water, herbal mouthwash, and chlorhexidine mouthwash on salivary pH. Drug Invention Today. 2019;11(5).
- 22. Řeháková P. Spectrophotometric determination of chlorhexidine in mouthwash employing Lab-In-Syringe automated ion-pair extraction and back-extraction. 2020.
- 23. Da Silva NB, Alexandria AK, De Lima AL, Claudino LV, Carneiro TFDO, Valença AM, et al. In vitro antimicrobial activity of mouth washes and herbal products against dental biofilm-forming bacteria. Contemporary clinical dentistry. 2012;3(3):302-5.
- 24. Nafea J, Edbeib M, Notarte KIR, Huyop F, Yaakub H. Stability and antibacterial property of polyherbal mouthwash formulated using local ingredients. Biosaintifika: Journal of Biology & Biology Education. 2020;12(3):288-96.
- 25. Anderson AM, Bruno BA, Smith LS. Viscosity measurement. AA. 2014;2(t3):809.