The Bioscan
AN INTERNATIONAL QUARTERLY JOURNAL OF LIFE SCIENCES
20(2): S2: 55-61, 2025
www.thebioscan.com

FORMULATION AND EVALUATION OF HERBAL MOUTHWASH FROM KADAMBA LEAVES EXTRACT

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DOI: 10.63001/tbs.2025.v20.i02.S2.pp55-61

KEYWORDS
Neolamarckia cadamba, Oral Hygine, Mouthwash, Staphylococcus aureus, E. coli bacteria.

Received on:
12-02-2025

Accepted on:
09-03-2025

Published on:
16-04-2025

ABSTRACT

Natural substitutes are sought out based on the accumulation of oral infectious microorganisms and the increase in resistance to synthetic antimicrobial agents. In this study, we aimed to formulate and evaluate an herbal mouthwash from Kadamba (Neolamarckia cadamba) plant leaves against common oral pathogens. Extracts were prepared from the leaves using ethanol and water, and were shown to contain phytochemical analytes including bioactive compounds such as flavonoids, tannins, and alkaloids that increase antimicrobial effectiveness. Using agar method, the extract was tested for involvement against Staphylococcus aureus, E. coli bacteria. The results indicated excellent antimicrobial activity and effectiveness, which was higher in the ethanolic extract compared to the aqueous extract. Moreover, pH value of the formulation was assessed and the formulation was stable and safe with respect to oral mucosal tissues. Conclusion combined extracts of Kadamba leaves may serve as a good mouthwash owing to the anti-bacterial activity on oral microorganisms.

Traditional mouthwash, especially containing alcohol, chlorhexidine, or synthetic antimicrobial agents, do have some side effects. These include irritation of the oral mucosa, dryness and burning sensation owing to high alcohol content. Extended use of chlorhexidine-based mouthwashes may cause staining of the teeth, altered taste sensation (dysgeusia) and disturbance of the normal oral microbiome, which might increase the risk of secondary infections such as oral thrush. Synthetic additives like parabens and artificial flavors can also cause allergic responses in certain people. Alcohol-based formulations also pose a toxicity risk if accidentally ingested, especially in children.

INTRODUCTION

Herbal and allopathic mouthwashes differ primarily in their composition and approach to oral care. Allopathic mouthwashes typically contain synthetic chemicals like chlorhexidine, alcohol, and fluoride, which are designed to kill bacteria, reduce plaque, and prevent cavities but chlorhexidine cause side effects like mouth irritation, dry mouth. On the other hand, herbal mouthwashes are formulated with natural ingredients like neem, clove, peppermint which have antimicrobial properties. These natural compounds not only combat oral pathogens but also promote gum health and freshen breath without the harsh side effects often associated with synthetic chemicals.

Materials and methods:

Plant Profile of Kadamba Leaves (Neolamarckia cadamba):

Kadamba, scientifically known as *Neolamarckia cadamba*, is a large evergreen tree belonging to the Rubiaceae family. The tree is known for its glossy, dark green, ovate leaves, fragrant orange-yellow spherical flowers, and grayish-brown bark. Culturally Medicinally, the leaves are used in Ayurveda for their antipyretic, anti-inflammatory, antimicrobial, and wound-healing properties. They contain bioactive compounds like alkaloids, flavonoids, and tannins, which contribute to their antioxidant and analgesic effects.

Materials:

The main material used in this study was cadamba leaf powder All solvents used were of analytical grade or liquorice powder, cinnamon oil, mentha peprita oil, glycerol, SLS, alcohol, water. The Ragatech Microwave was used for extraction. The Jasco V-630 spectrophotometer was used for UV-Vis spectrophotometric analysis. The Shimadzu FTIR 8400S was used For IR analysis.

Method:

Extraction preparation

Kadamb leaf powder (10 g) of was mixed in RBF with 100 mL of methanol. It subject to microwave for 10 minutes at 280 W, and

filtered through a 0.45µ filter. The extract was concentrated by rotavapor and the residue was stored for further study in a tightly closed container.

Extract evaluation test:

1) UV-Vis spectrophotometric analysis:

Extracts of *Kadamb* showed three absorbance peaks at at 208 nm, 282 nm and 316 nm in the UV-vis spectrum (Fig. 1).

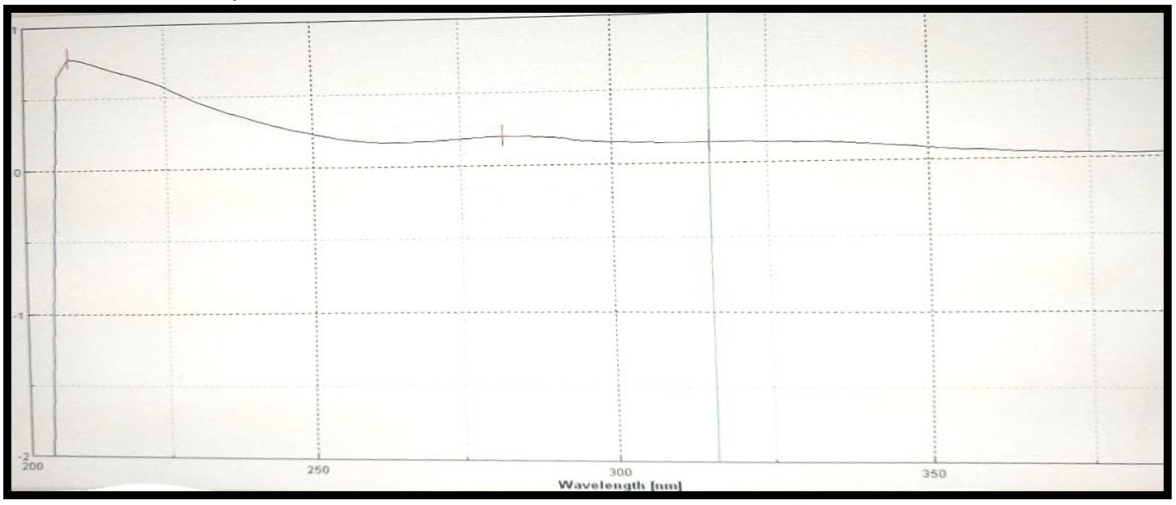
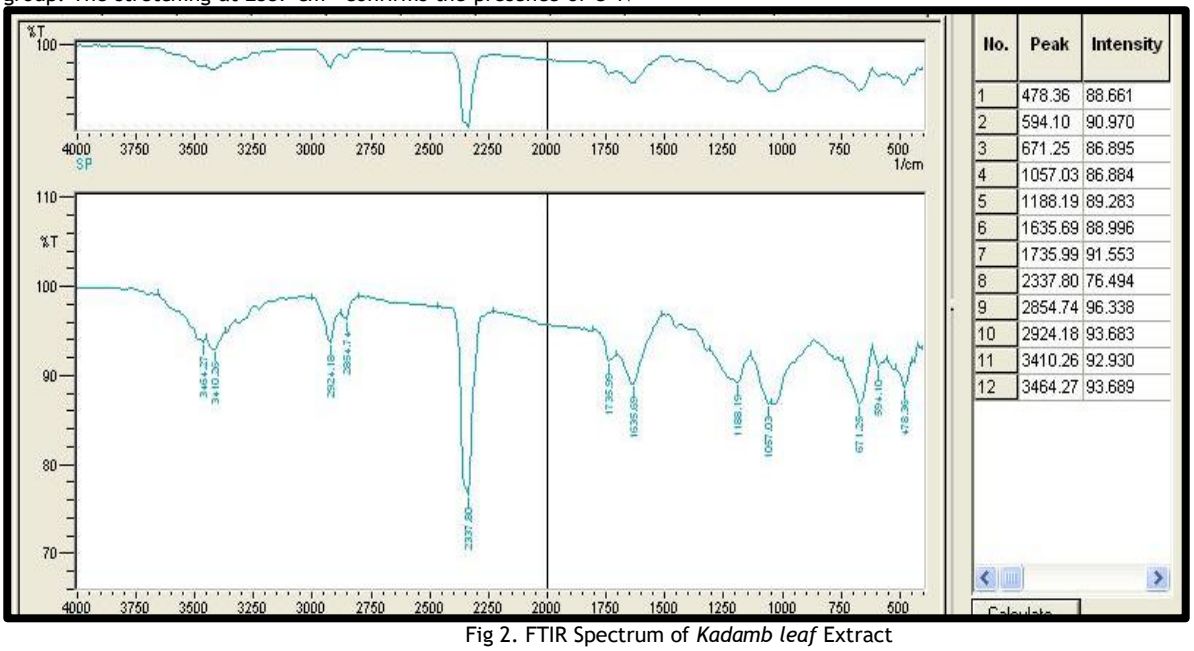


Fig 1. UV-Vis Spectrum of *Kadamb* leaf Extract

2) FT-IR spectrophotometric analysis:

The FTIR spectrum of *Kadamb* leaf extract was showed a characteristics peaks of N-H group at 3410 & 3464 cm^{-1}. The stretching at 2924 & 2854 cm^{-1} confirms the presence of C-H group. The stretching at 2337 cm^{-1} confirms the presence of C=N group. The stretching at 1735 & 1635 cm^{-1} confirms the presence of C=O group. The stretching at 1600-1610 cm^{-1} confirms the presence of C=C group. The stretching at 1188 & 1057 cm^{-1} confirms the presence of the C-H deformation.



No.	Peak	Intensity
1	478.36	88.661
2	594.10	90.970
3	671.25	86.895
4	1057.03	86.884
5	1188.19	89.283
6	1635.69	88.996
7	1735.99	91.553
8	2337.80	76.494
9	2854.74	96.338
10	2924.18	93.683
11	3410.26	92.930
12	3464.27	93.689

Fig 2. FTIR Spectrum of *Kadamb leaf* Extract

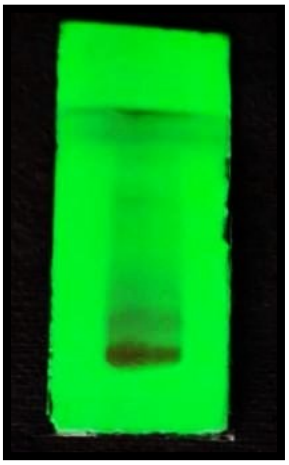


Fig. TLC of extract

4)Anti-microbial activity:

The well diffusion method was used to investigate antimicrobial activity. Nutrient agar medium was used for S. aureus and E. coli. The culture plates with extract and ofloxacin (100 µg/ mL) were then incubated at 37° C for 48 hrs. The antimicrobial activity of extract at different concentrations was determined by measuring the diameter of zone of inhibition.

Name of Organism	**Zone of Inhibhition (mm)**		
Concentration	**50 mg/ mL**	**100 mg/ mL**	**Ofloxacin (100 µg/ mL)**
Control	0 mm	0 mm	--
S. aureus	16 mm	14 mm	22 mm
E. Coli	18 mm	16 mm	23 mm

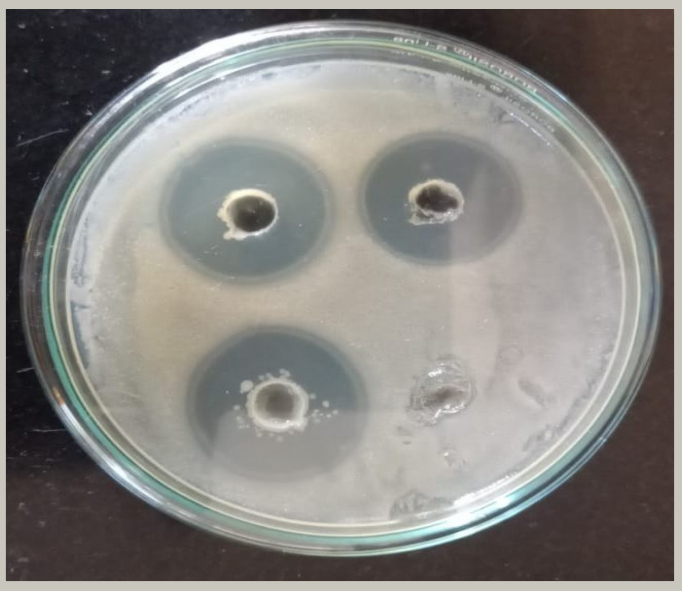


Fig: - Antibacterial activity of Kadamba leaf against gram +ve bacteria-S. aureus

Fig: Antibacterial activity of Kadamba leaf against gram -ve bacteria- E. Coli

Formulation table:

Ingredients	Functions	F1	F2	F3
Kadamba extract	Antimicrobial activity	5ml	7ml	3ml
Liquorice powder	Sweetner	045.gm	0.30gm	0.5gm
Peppermint oil	Flavouring agent	0.1ml	0.1ml	0.1ml
alcohol	Preservative	0.5ml	0.2ml	0.3ml
water	Solvent	Q.S (25ml)	Q.S (25ml)	Q.S (25ml)

[TABLE NO. 1]

Preparation Method

1. **Extract of Kadamba Leaves.**
2. **Preparation of the Base Solution:**
    - In a clean beaker, dissolve 1 gm extract of Kadamba in 10ml ethanol.

Take Kadamba leaves extract base solution. According to formula

3. **Addition of Excipients:**
    - Add peppermint oil for flavor and liquorice powder for sweetness (According to formula). Stir well to ensure homogeneity.
4. **Filtration and Packaging:**
    - Filter the final formulation through a 0.22 µm membrane filter to remove any particulate matter.
    - Transfer the solution into a 20 ml amber glass bottle to protect it from light and ensure stability.
5. **Labeling and Storage:**
    - Label the bottle with ingredients, usage instructions, and expiry date.
    - Store the gargle in a cool, dry place away from direct sunlight. [5]

Fig. Formulation of Herbal Mouthwash

Result and discussion:

1. Physical Evaluation:

All three formulations of mouthwash were evaluated for physical evaluation and the results were shown in the table 5.

Physical characteristics:

Sr.no	Formulation	Parameters	Observation

1	F1	Color Odour Appearance Texture	Golden yellow Pleasant (cool mint) Visuial appearance Liquid
2	F2	Color Odour Appearance Texture	Vibrant Orange Pleasant odour Visual appearance Liquid
3	F3	Color Odour Appearance Texture	Light Orange Pleasant odour Visual appearance Liquid

[TABLE NO. 2]

2. pH of Herbal mouthwash preparation:

pH of all three formulation was recorded by digital pen style pH meter. The results of Ph were shown in Table 3.

Sr.no	Formulation	pH
1	F1	6.37
2	F2	5.35
3	F3	5.29

[TABLE NO. 3]

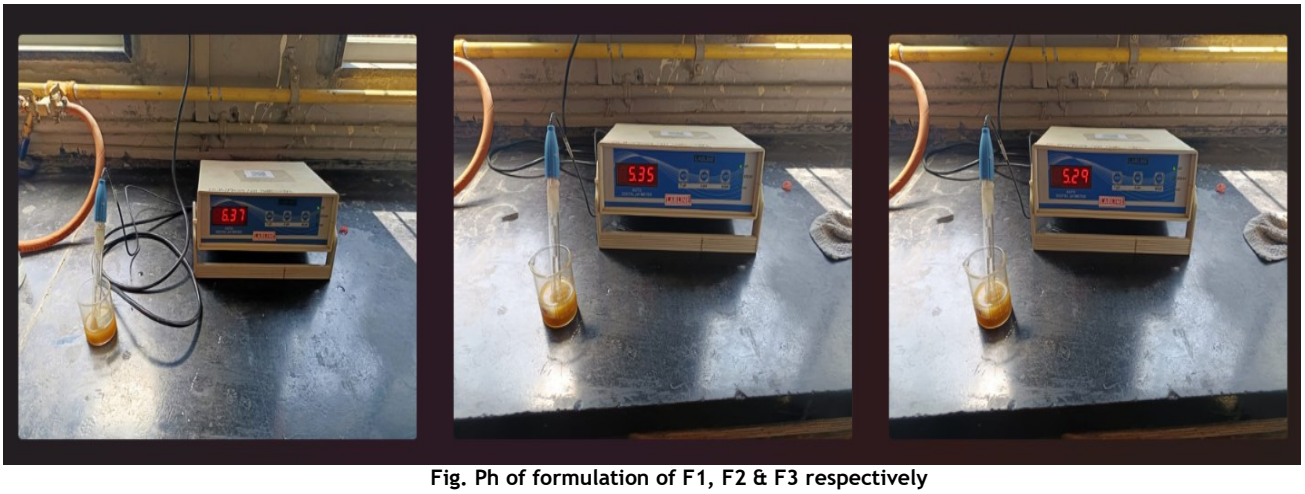


Fig. Ph of formulation of F1, F2 & F3 respectively

3. Viscosity:

We used an Ostwald viscometer to measure the viscosity of each of three mouthwash formulations**.**

[TABLE NO. 4]

Sr.no	Formulation	Viscosity (centipoises)
1	F1	1.53 cP
2	F2	2.10 cP
3	F3	1.2 cP

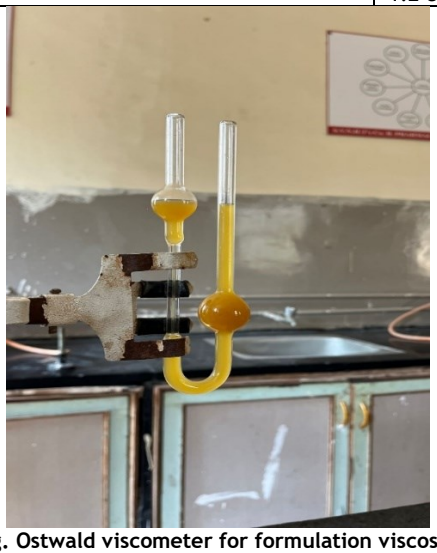


Fig. Ostwald viscometer for formulation viscosity

4. Stability studies:

The result of physical stability studies was shown in the table. Change in colour, odour; physical separation and homogeneity of all three formulated mouthwash was observed at 12^0 C and 25^0 C.

Formulations	Parameters	Temperature (12^0 C)	Temperature (25^0 C)
F1	Change in colour Odour Physical separation	No Change	No Change
F2	Change in colour Odour Physical separation	No Change	No Change
F3	Change in colour Odour Physical separation	No Change	No Change

[TABLE NO. 5]

5. Confirmative test for Terpenoids (Salkowski's test) - Procedure:

1) Mix 2ml of extract with 2ml of chloroform in a test tube.
2) Carefully add 3ml of conc. Sulfuric acid (H_2SO_4) along the side of test tube to form a layer.
3) Observe the interface between the two layers.
4) A reddish-brown coloration at the interface indicates in the presence of terpenoids.

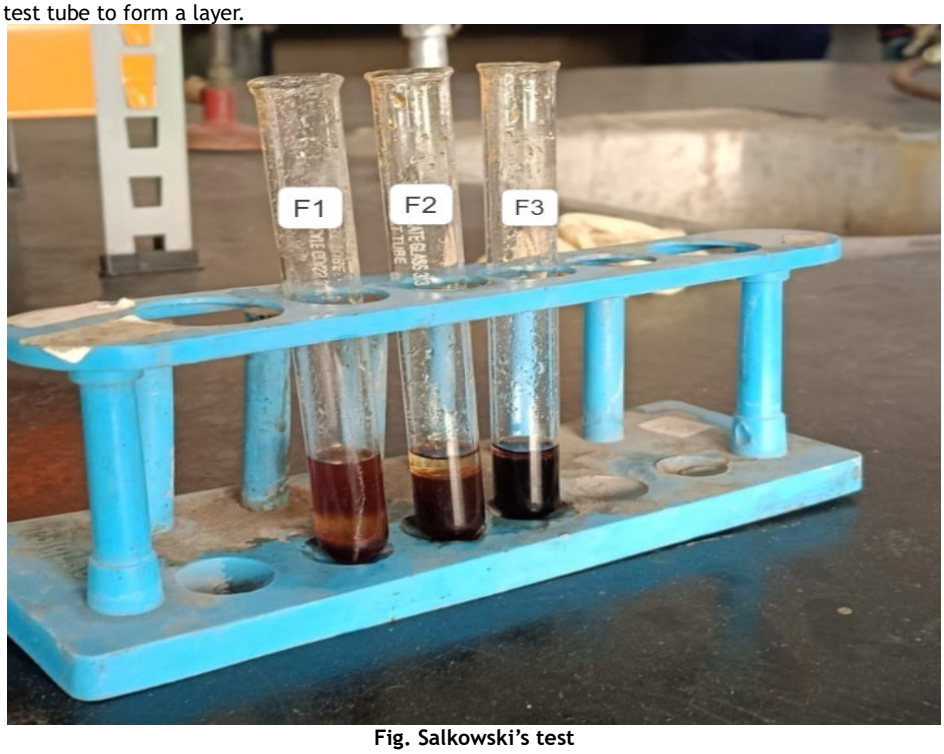


Fig. Salkowski's test

6. Foam Height:

The height of the foam of three formulation F1, F2 & F3 was found to be 5ml, 4ml & 5ml respectively.

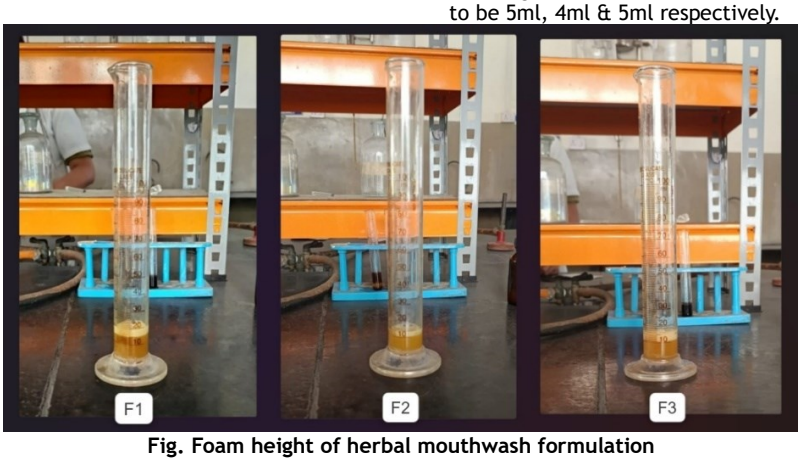


Fig. Foam height of herbal mouthwash formulation

CONCLUSION

The present study successfully formulated and evaluated an herbal mouthwash using *Kadamba (Neolamarckia cadamba)* leaf extract as a natural alternative to conventional synthetic mouthwashes. Phytochemical analysis confirmed the presence of bioactive compounds such as flavonoids, tannins, and alkaloids,

which contributed to the antimicrobial efficacy of the extract. Ethanolic extracts demonstrated superior antibacterial activity against *Staphylococcus aureus* and *Escherichia coli* compared to aqueous extracts, validating their potential in combating oral pathogens.

The formulated mouthwashes (F1, F2, F3) exhibited favorable physical characteristics, including optimal pH (5.29–6.37), acceptable viscosity (1.2–2.1 cP), and stability under varying temperatures. The absence of adverse changes in color, odor, or homogeneity during stability studies further confirmed their suitability for oral use. Additionally, the presence of terpenoids, as confirmed by Salkowski's test, may contribute to the antimicrobial and anti-inflammatory properties of the formulation.

In contrast to traditional alcohol- and chlorhexidine-based mouthwashes—which are associated with side effects such as mucosal irritation, tooth staining, and microbiome disruption—the *Kadamba*-based herbal mouthwash offers a safer, biocompatible alternative. Its natural composition, combined with effective antimicrobial action, positions it as a promising candidate for oral hygiene applications. Future studies should focus on clinical trials to assess long-term efficacy, safety, and consumer acceptability in human subjects.

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