

FORMULATION AND EVALUATION OF A NOVEL POLYHERBAL ANTIDANDRUFF
SHAMPOO

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DOI: <https://doi.org/10.5281/zenodo.17471393>

How to cite this Article: Sowjanya A.R.*, T. Tamizh Mani, L. Shiju, Vageesh Revadigar, Pavithra T. (2025). Formulation and Evaluation of A Novel Polyherbal Antidandruff Shampoo. European Journal of Pharmaceutical and Medical Research, 12(11), 109–117.

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Article Received on 22/09/2025

Article Revised on 13/10/2025

Article Published on 01/11/2025

ABSTRACT

This study was aimed to formulate and evaluate a polyherbal antidandruff shampoo using plant extracts such as *Calotropis gigantea*, *Pongamia pinnata*, *Limonia acidissima*, *Solanum nigrum*, and *Putranjiva roxburghii*. The formulations (F1–F5) were assessed for pH, viscosity, foam stability, solid content, dirt dispersion, antimicrobial activity, and stability. Results showed suitable pH, good viscosity, effective foam stability, and solid content within 20–30%. Antimicrobial tests demonstrated significant activity against *Staphylococcus aureus* and, especially in F3 and F4. Stability studies confirmed that F4 and F5 remained stable *Malassezia furfur* over three months. These findings suggest F4 and F5 as promising candidates for commercial herbal antidandruff shampoo development.

KEYWORDS: Formulations, Antimicrobial activity, *Staphylococcus aureus*, *Malassezia furfur*.

INTRODUCTION

“A cosmetic is an item intended to be rubbed, poured, sprinkled or sprayed on, introduced in to or otherwise applied to the human body or any part thereof for cleansing, beautifying, promoting attractiveness or altering the appearance.”

HERBAL COSMETICS

Herbal cosmetics are personal care products formulated using plant-derived ingredients such as leaves, roots, fruits, flowers, and seeds. These products are designed to improve, enhance, and maintain the appearance of the skin, hair, and nails by cleansing, moisturizing, promoting attractiveness, or treating common disorders. Unlike synthetic cosmetics, which rely heavily on chemical compounds, herbal cosmetics utilize bioactive phytochemicals that offer both cosmetic and therapeutic benefits.^[1]

The variety of items referred to as “hair cosmetics” are used to maintain the cleanliness of hair. Depending on a person’s culture and physical attributes of their hair, several hair care regimens are used, like shampoo, hair dyes, and hair sprays.^[2]

DANDRUFF

A significant cosmetic issue that is extremely concerning for public health in both industrialized and developing nations is dandruff. Dandruff is a chronic scalp condition leading to scaling, itching, redness of scalp by shedding epidermal cells. Scalp sheds dead cells in nearly invisible way but sometimes sheds as visible flakes called dandruff.^[3]

CAUSES OF DANDRUFF

- Dry skin.
- Irritated, oily skin.
- Not shampooing often enough
- Other skin conditions: A. Eczema B. Psoriasis C. Seborrheic dermatitis
- Malassezia-yeast like fungus
- Sensitivity to hair products (contact dermatitis)

TREATMENT

- FOLLOW A HEALTHY DIET.
- AVOID STRESS.
- SHAMPOO USE A COMBINATION OF SPECIAL INGREDIENTS TO CONTROL DANDRUFF.⁴

HERBAL ANTI-DANDRUFF SHAMPOOS

Herbal anti-dandruff shampoos are the cosmetic formulations which contain herbal ingredients such as plant extracts and essential oil. These herbal shampoos are generally used to remove the dandruff, to add natural colour to the hair, to remove the extra oil content of the hair, for the healthy growth of the hair, to remove the dust, dirt, and scales of the scalp, to prevent hair falling, to impart softness and smoothness to the hair shaft, etc. It is assumed that they can penetrate to the root shafts, stimulate the sebaceous glands, enhance the blood circulation, and impart greater strength to the hair root and the shaft. They are also used against alopecia, thinning, clubbing, and greying of hair and hair shaft roughness and breaking.

Advantages of Herbal Shampoo

Nowadays, traditional herbal shampoos are most widely used hair products. Because it is believed that these products are safe and free from side effects. Chemical shampoos might appear to be improving hair texture along the length but eventually end up with damaging the roots and cause:

- Scalp dryness and itchiness
- Premature aging, greying of hair
- Split ends and excessive hair loss.

To overcome all such problems, it is best to switch to an herbal shampoo which will make up for the loss of nutrients and nullify the damage way.^[5]

Preparation of Polyherbal antidandruff Shampoo^[6]

Table No. 1: Formulation of herbal anti dandruff shampoo.

SL NO	Ingredients	Purpose	Qty (100%)
1	Triethanolamine lauryl sulphate	Primary surfactant	45%
2	Coconut monoethanolamide	Foam Stabilizer	2%
3	Herbal extract	Antidandruff agent	10-15%
4	Xanthan gum	Thickening agent	Q. S
5	Perfume	Flavoring agent	Q. S
6	Colour	Appearance	Q. S
7	Water	vehicle	Q. S

Procedure

Take Triethanolamine lauryl sulphate under constant stirring to avoid excessive foam formation. Then dissolve the Cocamide MEA and herbal extracts (different concentration) in hydro-alcoholic

MATERIALS AND METHODOLOGY

Collection and authentication of plant materials

The plant materials of *Calotropis Gigantea* leaves, *Pongamia Pinnata* seeds, *Limonia Acidissima* Fruit, and *Solanum nigrum* Fruit, was collected from K Bellur village, Maddur taluk, Mandya District, Karnataka, and *Putranjiva Roxburghii* leaves was collected from the Medical Plant Garden of Shree Sindhagi Shantaveereshwar Ayurvedic College and Hospital Haveri, Karnataka, India in the month of April 2025. The plant was identified and authenticated by Dr. Thejesh Kumar M.P Assistant professor and Head (Botany), Post graduate research Centre Bharathinagara, Maddur taluk, Mandya district, Karnataka. An herbarium voucher specimen was preserved in the Department of Pharmacognosy, Bharathi College of Pharmacy, Bharathinagara for further reference, after collection and authentication of plant materials. All the fresh ingredients were dried in shade, converted into coarse powder.

METHODOLOGY

The method was carried out by using the closed vessel container. Accurately weigh 250g of powdered dried plants materials separately and then poured into the vessel to that add 1000ml of Hydroalcoholic solvent (50% alcohol and 50% distilled water) soaked for 72hours with occasional shaking. After this period filter the mixture with the help of filter paper. Evaporate the solvent by heating on a water bath until to get a semisolid extract (concentrated extract).

solution(5ml) separately. Then mix it together and adjust the thickness using Xanthan gum slowly by stirring. Then add fragrance and colour with gentle mixing. Adjust pH using Sodium Chloride to around 4.5-5.5.

Table No. 2: Different concentration of extract used for the preparation of herbal Shampoo(mg/50ml).

Name of herbal ingredients	F1	F2	F3	F4	F5
<i>Calotropis gigantea</i> (Leaves)	500mg	750mg	1000mg	750mg	500mg
<i>Limonia Acidissima</i> (Fruit pulp)	500mg	750mg	1000mg	500mg	250mg
<i>Pongamia Pinnata</i> (Seeds)	500mg	750mg	1000mg	500mg	250mg
<i>Putranjiva Roxburghii</i> (Leaves)	500mg	750mg	1000mg	1250mg	1500mg
<i>Solanum nigrum</i> (Fruit)	500mg	750mg	1000mg	1250mg	1500mg

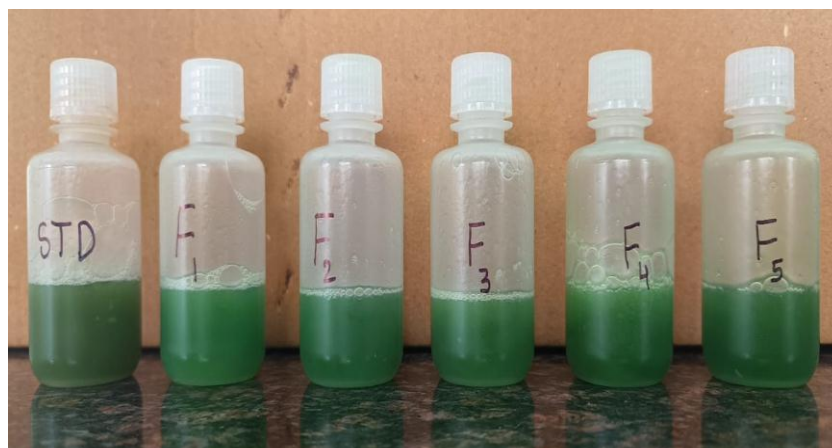


Fig. 1: Different concentration of formulated herbal antidandruff Shampoo.

Evaluation of prepared polyherbal antidandruff shampoo

To evaluate the prepared formulations, quality control tests including visual assessment and physicochemical controls such as pH, density, viscosity, surface tension, foam volume, foam stability, and wetting time were performed using standard protocols.

Physical appearance/visual inspection^[7]

The prepared formulations were evaluated for the clarity, colour, odour and foam producing ability and fluidity. To ensure aesthetic and sensory acceptability.

pH^[8]

The pH of shampoos has been shown to be important for improving and enhancing the qualities of hair, minimizing irritation to the eyes, and stabilizing the ecological balance of the scalp.

The pH will be calculated using pH meter. The pH will be measured by using 1ml of shampoo into 5 ml distilled water then calculate the pH.

Viscosity^[9]

Viscosity has an important role in explaining and controlling many attributes like shelf-life stability and product aesthetics such as clarity, ease of flow, on removal from packing and spreading when applied to hair.

Procedure: The viscosity of the prepared formulations was measured at room temperature using a Brookfield viscometer (R/S plus rheometer model, LV, USA). 100 ml of the tested shampoo was poured in a beaker and an appropriate spindle was immersed into it. Readings were recorded after 5 min. of rotation at a speed of 10 rpm.

Dirt dispersion^[10]

Shampoo should remove dirt effectively without foam darkening.

Procedure: In a large test tube, two drops of shampoo will be added to another test tube containing 10 ml of distilled water. To this, 1 drop of Indian ink will be added;

the test tube will be stoppered and shaken for 10 times. The amount of ink in the foam will be estimated as None, Light, Moderate, or heavy.

Determination of solid content percentage^[11]

Determines total dissolved/undissolved content; should be 20–30% ideally.

Procedure: A clean dry evaporating dish will be weighed and 4 grams of shampoo will add to the evaporating dish. The evaporating dish with shampoo will be placed on the hot plate until the liquid portion will evaporate. The weight of the solid contents present in the shampoo will be calculated after drying.

Foam stability test^[12]

Evaluates cleansing efficiency and consumer appeal.

Procedure: The stability of foam will determine by using cylinder shakes method. About 25 ml of shampoo will be taken in 250 ml measuring cylinder and shaken for 10 minutes.

The total foam volume will measure after 1 minute and foam stability was determined by recording foam volume from 1 to 4 minute.

Antimicrobial Activity

Determination of Anti-microbial activity of formulated shampoo by agar well diffusion c method.

The increasing incidence of drug-resistant pathogens raises an urgent need to identify and isolate new bioactive compounds from medicinal plants using standardized modern analytical procedures. Medicinal plant-derived compounds could provide novel straightforward approaches against pathogenic bacteria. Plants are rich in a wide variety of secondary metabolites, such as tannins, terpenoids, alkaloids, and flavonoids, which are responsible for antimicrobial properties. Hence, the formulated shampoos were subjected to antimicrobial screening by agar well diffusion method.

Antibacterial Activity

Microorganisms used: *Staphylococcus aureus* were procured from JSS college of Pharmacy, Ooty, India and are used for determining antimicrobial activity.

Preparation of standard: Ciprofloxacin and ketoconazole are dissolved in DMSO solution to get the concentration of (5µg/well) and (30µg/well) respectively. Determination of zone of inhibition: Evaluation of antimicrobial activity was carried out by agar well diffusion or cup-plate technique using nutrient agar medium and the antimicrobial activity was measured in terms of zone of inhibition in millimeter (mm).

Preparation of Inoculums: Suspension of organism was prepared as per McFarland standard. A 24-hrs old culture was used for the preparation of bacterial suspension. Suspension of organism was made in a sterile isotonic solution of sodium chloride (0.9% w/v) and the turbidity was adjusted such that it contained approximately 1.5×10^8 cells/ml. It was obtained by adjusting the optical density of the bacterial suspension equivalent to mixture of 0.05ml of 1.175% of barium chloride and 9.95ml of 1% Sulphuric acid.^[13,14]

Antifungal Activity^[15,16,17]

Microorganism Used: *Malassezia furfur* were cultured by (invitro) scraping method from the humans.

Procedure for scraping

Infected lesion was identified, Fungal presence is confirmed with the help of microscopic examination, Infected region was washed with 70% ethanol, Collection of samples by disposable scalpel and cotton swab Collected sample placed in sterile Petri dish, Specimen was transferred into dark sampling paper to prevent exposure to light the sample collected were cultured on SDA (Sabouraud dextrose agar) Chloramphenicol was incorporated to media to get rid of contaminants, small amount of samples collected was introduced into Petri plates containing media. The Petri dishes were then labeled Incubated at RT (25 ° C) for the three days and upto the week.

Culture Media Preparation

- Prepare Sabouraud Dextrose Agar (SDA) with 1% olive oil or Tween 60, which is essential for *Malassezia* growth.
- Sterilize the media and pour into sterile Petri dishes. Allow to solidify.

Shampoo Sample Preparation

- Prepare different concentrations of the shampoo using sterile distilled water
- Use the undiluted shampoo as another test concentration.

Inoculation Procedure

- Take *Malassezia furfur* culture and grow it on SDA with olive oil for 48–72 hrs at 32–35°C.
- Prepare a fungal suspension in sterile saline or broth, adjusting to 0.5 McFarland standard ($\sim 1 \times 10^6$ CFU/mL).
- Swab the entire surface of solidified SDA plate with the fungal suspension.

Agar Well Diffusion Method

- Use a sterile cork borer to punch wells (~6 mm diameter) in the agar plate.
- Fill the wells with:
 - Test shampoo (various concentrations)
 - Positive control (e.g., ketoconazole shampoo)
 - Incubate the plates at 32–35°C for 5–7 days in a humidified chamber.

OBSERVATION AND MEASUREMENT

- After incubation, measure the zone of inhibition (clear zone around the well) in millimetres.

Stability studies^[18]

The stability studies for the polyherbal anti-dandruff formulations will be performed according to ICH guidelines. The formulations will be tested for their physical appearance, % solid content, transparency, and pH.

RESULTS AND DISCUSSION

Evaluation of polyherbal antidandruff Shampoo

The prepared Herbal Shampoo formulations (Standard, F1, F2, F3, F4, F5) were evaluated for their physicochemical properties and performance parameters. Results are presented below.

Physical Appearance

All formulations were homogeneous, free from phase separation, and had a pleasant herbal fragrance.

Table No. 3: Evaluation of Formulation for physical appearance.

SL.NO	Formulation	Odour	Colour
1	F1	Pleasant	Peacock green
2.	F2	Pleasant	Peacock green
3.	F3	Pleasant	Peacock green
4.	F4	Pleasant	Peacock green
5.	F5	Pleasant	Peacock green
6.	Standard	Pleasant	Peacock green

All the formulations exhibited desirable physical characteristics including homogeneity, pleasant odor, and stable peacock green color.

pH Measurement**Table No. 4: Determination of pH.**

SL.NO.	Formulation	pH (25 °C) Trials(n)			Average pH (25 °C)
		1	2	3	
1.	F1	5.3	5.2	5.3	5.3
2.	F2	5.3	5.1	5.2	5.2
3.	F3	5.3	5.3	5.3	5.3
4.	F4	5.2	5.3	5.3	5.3
5.	F5	5.2	5.3	5.3	5.3
6.	Standard	5.3	5.2	5.2	5.2

The pH of formulations (5.2–5.3) was within the recommended range for hair care products (4.5–5.5),

indicating suitability for scalp application without causing irritation or dryness.

Viscosity**Table No. 5: Determination of Viscosity.**

SL. NO.	Formulation	Viscosity (cP) Trials(n)			Average Viscosity(cp)
		1	2	3	
1.	F1	2800	2750	2850	2800
2.	F2	2900	2950	2800	2883
3.	F3	3200	3150	3300	3216
4.	F4	4700	4650	4800	4716
5.	F5	5100	5000	5200	5100
6.	Standard	4200	4150	4300	4216

Viscosity measurements revealed that F3–F5 formulations were within the ideal viscosity range for shampoos, providing good consistency and spread ability

compared to the standard. Higher viscosities in F4 and F5 may be attributed to increased extract concentration and stabilizer interaction.

Foam hight and Foam stability**Table No. 6: Foam Height and Stability.**

SI. No.	Formulation	Initial Foam Height (cm)	Foam Height after 5min (cm)
1	F1	8	7.2
2	F2	7.6	6.9
3	F3	7.6	6.7
4	F4	9.3	8.4
5	F5	9.1	8.2
6	Standard	8.5	7.6

Foamability and foam stability are essential for consumer acceptability. Among the formulations, F4 and F5 showed maximum foam height (9.3 cm and 9.1 cm initially) and retained significant stability after 5 minutes (8.4 cm and 8.2 cm, respectively), which were superior to the standard formulation. This suggests an adequate surfactant–herbal extract balance that enhances foaming properties.

Solid content determination

$$\%Total\ Solids = \frac{B - A}{4} \times 100$$

Where,

A= Wight of empty China dish

B= Wight of China dish after evaporation

B-A= Wight of Sample

Table No. 7: Solid content determination.

Formulation	Total Solid Content Trials(n)			% Solid Content Trials(n)		
	1	2	3	1	2	3
F1	0.83g	0.82g	0.83g	20.75	20.5	20.75
F2	0.82g	0.83g	0.83g	20.5	20.75	20.75
F3	0.86g	0.85g	0.85g	21.5	21.25	21.25
F4	0.95g	0.94g	0.95g	23.75	23.5	23.75
F5	0.95g	0.96g	0.96g	23.75	24	24
Standard	0.82g	0.82g	0.83g	20.5	20.5	20.75

Solid content determination revealed values ranging from 20.5% to 24%, with F4 and F5 exhibiting the highest solid contents. These values are in line with

reported optimal ranges for shampoos (20–30%), ensuring effective cleansing without leaving residues.

Dirt dispersion

Table No. 8: Dirt Dispersion Test.

Formulation	Observation	Inference (Ink in Foam)
F1	Dirt present in water layer	Light
F2	Dirt present in water layer	Light
F3	Dirt present in water layer	Light
F4	Dirt present in water layer	Light
F5	Dirt present in water layer	Light
Standard	Dirt present in water layer	Light

Dirt dispersion tests further confirmed the cleansing ability, with F1 and F5 performing comparably to the standard shampoo by producing clear foam and entrapping dirt in the water layer.

activity against bacterial strain such as *Staphylococcus aureus*, and, fungal strains such as *Malassezia furfur*. In vitro antimicrobial activity performed by agar well diffusion method.

Antimicrobial Study

Formulated shampoo was studied for its antimicrobial

Table No. 9: Antimicrobial activity (against bacterial strain) of formulated shampoo by Agar well diffusion method.

Formulation	Organisms used	Standard	Zone of Inhibition (ZOI) in mm		
		Ciproflaxacin (200 µl/well(std)) ZOI in mm	100 µl/well	200 µl/well	300 µg/well
F1	<i>Staphylococcus aureus</i>	22.3	9.5	11.2	14.9
F2		22.4	9.3	11.4	14.5
F3		22.4	9.5	11.1	15.0
F4		22.4	9.5	11.1	15.2
F5		22.4	9.1	11.0	14.1
Standard		22.4	9.5	11.1	15.2

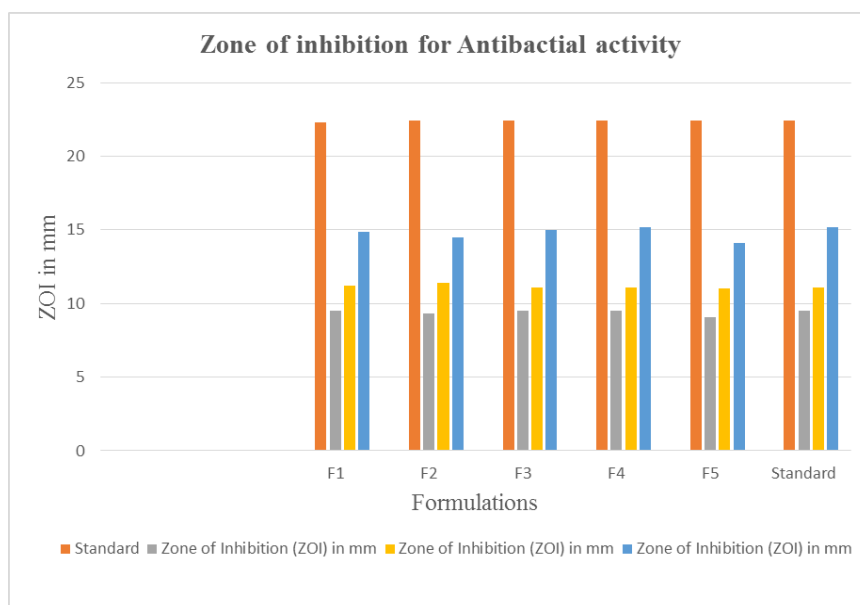


Fig. 2: Zone of inhibition for Antibacterial activity.

Table No. 10: Antimicrobial activity (fungal strains) of formulated shampoo by Agar well diffusion method.

Formulation	Organisms used	Standard	Zone of Inhibition (ZOI) in mm		
		Ketoconazol (200 µl/well (std)) ZOI in mm	100 µl/well	200 µl/well	300 µg/well
F1	<i>Malassezia furfur</i>	21.3	9.3	11.4	15.1
F2		21.3	9.4	11.5	15.3
F3		21.2	10.3	12.2	16.2
F4		21.3	10.4	12.4	16.4
F5		21.3	9.5	11.6	15.4
Standard		21.3	10.4	12.3	16.1

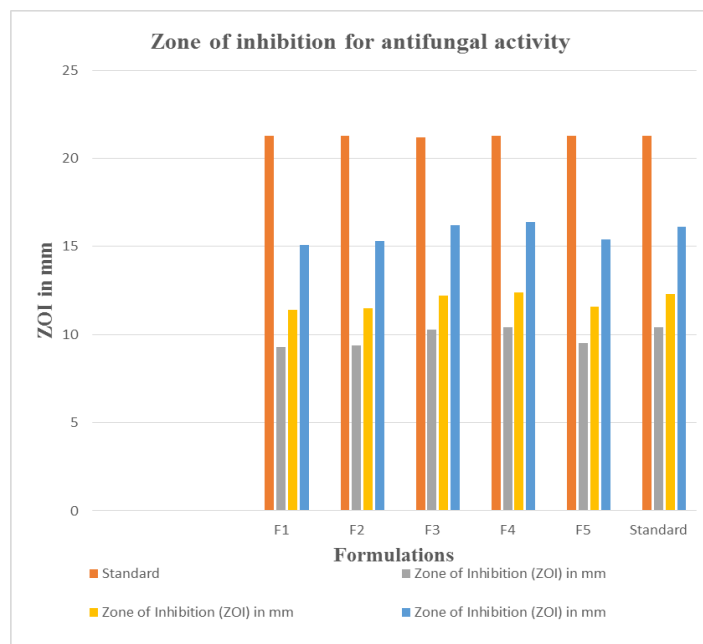


Fig. 3: Zone of inhibition for Antifungal activity.

The zone of inhibition increased in a concentration-dependent manner, with F3 and F4 showing maximum activity, nearly comparable to standard ciprofloxacin (for

bacteria) and ketoconazole (for fungi). This indicates that the selected herbal extracts contributed synergistically to antimicrobial efficacy.

Stability test

Table No. 11: Evaluation of Formulation for physical appearance.

Formulation	1 month	2 months	3 months
F1	No change was observed	No change was observed	No change was observed
F2	No change was observed	No change was observed	No change was observed
F3	No change was observed	No change was observed	Separation of two layers
F4	No change was observed	No change was observed	No change was observed
F5	No change was observed	No change was observed	No change was observed
Standard	No change was observed	No change was observed	No change was observed

Table No. 12: Determination of pH.

Formulation	1 month	2 months	3 months
F1	5.2	5.3	5.4
F2	5.1	5.2	5.4
F3	5.3	5.5	6.2
F4	5.3	5.4	5.5
F5	5.3	5.4	5.5
Standard	5.2	5.3	5.4

Table No. 13: Foam hight and Foam stability.

Formulation	1 month		2 months		3 months	
	Initial Foam Height (cm)	Foam Height after 5min (cm)	Initial Foam Height (cm)	Foam Height after 5min (cm)	Initial Foam Height (cm)	Foam Height after 5min (cm)
F1	8.0	7.2	7.8	6.0	7.4	5.2
F2	7.6	6.9	7.4	5.5	6.8	4.9
F3	7.6	6.7	7.0	5.4	6.5	4.3
F4	9.3	8.4	8.9	7.9	8.4	7.2
F5	9.1	8.2	8.8	7.8	8.5	7.2
Standard	8.5	7.6	8.0	7.2	7.0	5.5

Stability studies conducted over three months indicated that most formulations retained their physical characteristics, pH, foam stability, and antimicrobial properties. However, F3 showed phase separation after three months, suggesting that formulations with higher extract concentration may require stabilizers to maintain long-term integrity. F4 and F5 remained stable throughout the study, highlighting their suitability for further development as commercial formulations.

CONCLUSION

The polyherbal antidandruff shampoo formulations using *Calotropis gigantea*, *Pongamia pinnata*, *Limonia acidissima*, *Solanum nigrum*, and *Putranjiva roxburghii* showed good physical properties, optimal pH (4.5–5.5), suitable viscosity, and effective foam stability. Solid content and dirt dispersion tests confirmed effective cleansing. Antimicrobial studies demonstrated strong activity against *Staphylococcus aureus* and *Malassezia furfur*, especially in F3 and F4. Stability tests showed F4 and F5 remained stable over three months. F4 and F5 were identified as the most promising formulations for further commercial development.

Based on the comparative antimicrobial data, F4 is considered the best formulation because it demonstrates the most potent and balanced antibacterial and antifungal activity, with results that match or exceed the standard antimicrobial agents. Therefore, F4 stands out as the most effective formulation for controlling scalp pathogens, making it the ideal choice for a polyherbal antidandruff shampoo formulation.

Further in vivo studies and clinical evaluations are recommended to validate its therapeutic effectiveness and safety, paving the way for potential commercial production and widespread use as an effective, safe, and natural antidandruff solution.

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