



Original Research Article

Formulation, Optimization, and In-vitro Evaluation of a Polyherbal Facewash Containing Rosmarinus officinalis, Rubia cordifolia, and Gomphrena globosa for Anti-acne Activity

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Abstract: **Aim:** Formulation, Optimization, and In-vitro Evaluation of a Polyherbal Facewash to access anti-acne Activity **Background:** Acne vulgaris is a chronic inflammatory skin disorder caused by the proliferation of *Propionibacterium acnes*. Synthetic facewash formulations often cause dryness or irritation, thus increasing the demand for herbal alternatives. This study focuses on the formulation and evaluation of a polyherbal facewash containing *Rosmarinus officinalis*, *Rubia cordifolia*, and *Gomphrena globosa* for anti-acne potential. **Material and Methods:** Ethanolic extracts of selected herbs were incorporated into gel-based facewash formulations (F1–F3) and optimized for physicochemical properties such as pH, viscosity, foaming capacity, and spreadability. The final formulation was evaluated for antimicrobial activity against *P. acnes* using the agar well diffusion method. **Results:** The optimized formulation showed pH 4.07–4.9, viscosity 344.2 cps, foam stability up to 23 min, and spreadability 6.56 g•cm/sec. It exhibited a significant zone of inhibition (21 mm) and lower IC₅₀ value (0.61) than the marketed product (0.63). Statistical analysis confirmed the significant antimicrobial effect ($p < 0.05$). **Conclusion:** The formulated polyherbal facewash demonstrated excellent physicochemical properties and potent anti-acne activity, making it a promising natural alternative for acne management.

Keywords: *Propionibacterium acnes*, *Rosmarinus officinalis*, *Rubia cordifolia*, *Gomphrena globosa*, herbal facewash.

INTRODUCTION

Acne is a prevalent dermatological condition that arises when hair follicles become obstructed by

sebum and dead skin cells. The pathophysiology of acne involves four primary factors: inflammation, proliferation of *Propionibacterium acnes* (*P. acnes*), hyper-keratinization of pilosebaceous follicles, and

excessive sebum production. Acne vulgaris is clinically characterized by both inflammatory lesions (papules, pustules, nodules, and cysts) and non-inflammatory lesions (blackheads and whiteheads). Its onset is primarily driven by bacterial activity and inflammation within the pilosebaceous glands, often exacerbated by hormonal fluctuations that alter glandular function.

Globally, acne affects approximately 20.5% of individuals across different age groups, with over 85% of adolescent boys and 75% of adolescent girls experiencing the condition. Nearly 30% of affected adolescents suffer from moderate-to-severe acne, while about 40% continue to experience active acne into adulthood. A systematic analysis from the Global Burden of Disease Study ranked acne as the eighth most prevalent disease worldwide.

Several herbal interventions, including tablets, capsules, creams, soaps, gels, lotions, facewashes, and emulgels, have been utilized for acne management.^{1,2} Among these, facewashes are particularly popular in daily skincare routines due to their ease of use and ability to address oily skin, dryness, itching, and irritation. Acne treatments generally follow two primary approaches: herbal-based anti-acne facewashes, which leverage natural extracts with soothing and antibacterial properties, and chemical-based formulations containing active agents such as salicylic acid, benzoyl peroxide, or triclosan for more rapid effects.^{3,4}

Herbal facewashes are designed to cleanse the skin, unclog pores, and prevent acne through bioactive plant-derived constituents. Their function extends beyond cleansing to include the removal of excess sebum, environmental pollutants, and impurities while providing nourishment to the skin. Being plant-based, they offer a safer and more cost-effective alternative to synthetic formulations.^{5,6} The growing preference for herbal skincare products is driven by their reduced likelihood of adverse effects. Among the most promising herbal ingredients, *Rosmarinus officinalis* (rosemary), *Rubia cordifolia* (manjistha), and *Gomphrena globosa* have been recognized for their anti-acne efficacy.⁷

Rosmarinus officinalis, a Mediterranean herb now cultivated globally, is valued both as a culinary spice and a medicinal plant due to its diverse bioactive composition. It is rich in polyphenols and bioactive compounds that confer multiple pharmacological properties, including antioxidant, anti-inflammatory, antimicrobial, antifungal, antiviral, anti-obesity, anticancer, antidiabetic, neuroprotective, cardioprotective, gastrointestinal, dermatological, and other health benefits. Key bioactive constituents include lipophilic diterpenes such as carnosic acid and carnosol, potent antioxidants; rosmarinic acid,

which enhances overall antioxidant capacity; and oleanolic acid, exhibiting antiviral and antiproliferative effects. Ursolic acid contributes to chemopreventive activity, while flavonoids (cirsimaritin, diosmin, genkwanin) and volatile oils provide additional therapeutic and organoleptic advantages.^{8,9}

Gomphrena globosa (L.), commonly known as Globe Amaranth or Bachelor's Button, has been traditionally used in North America, Bangladesh, and India for a broad spectrum of ailments, including hypertension, diabetes, hepatic dysfunctions, bronchitis, fever, diarrhea, and urinary complications. The plant's medicinal activity is attributed to its phytoconstituents—flavonoids, betacyanins, phytosterols, phenolics, and terpenoids—which exhibit antioxidant, anti-inflammatory, antimicrobial, anticancer, analgesic, cytotoxic, and hemostatic effects. Ethnomedical applications extend to malaria, bacterial infections, dysuria, and hypercholesterolemia, underscoring its pharmacological relevance alongside its ornamental and natural colorant uses.^{10,11}

Rubia cordifolia (Manjistha) holds considerable significance in Ayurveda, classified by Charaka as varnya (complexion enhancer), jvarahara (antipyretic), visaghna (detoxifier), and rasayana (rejuvenator), and by Sushruta as pittasamsamana (pitta pacifier). The red roots, rich in anthraquinones, have traditionally been used to improve blood circulation, manage bleeding disorders, and treat gynecological conditions, allergic purpura, and renal hemorrhage. Phytochemical analyses have identified over 100 compounds, including quinones, flavonoids, peptides, terpenes, and polysaccharides. Modern pharmacological investigations report anti-tumor, antimicrobial, antioxidant, anti-inflammatory, and anti-platelet aggregation properties, although most findings remain preclinical. Limited pharmacokinetic and toxicity data highlight the need for further clinical validation.^{12,13}

Based on the collective literature evidence, *Rosmarinus officinalis*, *Gomphrena globosa*, and *Rubia cordifolia* were selected in combination for the development of a herbal facewash targeting acne management. The formulated product was subsequently subjected to comprehensive physicochemical and biological evaluation to assess its efficacy and substantiate its therapeutic potential.

MATERIAL AND METHOD

Materials

Rosmarinus officinalis and *Rubia cordifolia* were supplied by Manikarnika Aushadhalaya, Chinchwad, Pune. *Gomphrena globosa* flowers were

collected from Dehuroad, Pune. Rose oil and glycerin were procured from Research Fine Lab Chem Industry, Mumbai. Carbopol and methyl paraben were sourced from Chemydes Corporation, Rajkot, and vitamin E was obtained from Pharvax Biosciences, Chandigarh.

Preparation of Extract

For extract preparation, 20 g each of dried *Rosmarinus officinalis*, *Rubia cordifolia*, and *Gomphrena globosa* powders were placed in 150 mL of ethanol in a 250 mL conical flask. The mixture was agitated on an orbital shaker for 2–4 hours and allowed to soak for 24–48 hours. The solution was then filtered through filter paper, and the filtrate was collected in a pre-weighed petri dish. The solvent was evaporated at room temperature to obtain a dried extract, which was subsequently weighed.^{14,15}

Optimization of Anti-Acne Facewash Formulation

The herbal facewash was prepared by combining two phases (A and B) using controlled sonication to ensure homogeneity and stability.

- **Phase A:** Carbopol was dispersed in 20 mL of distilled water and sonicated at 25.5 °C. Subsequently, 15 mL and 5 mL of water were added sequentially with intermittent sonication and gentle stirring to minimize air entrapment and improve formulation stability.
- **Phase B:** Sodium lauryl sulfate was sonicated in 5 mL of water, followed by stepwise incorporation of an aqueous solution of methyl paraben, herbal extracts, vitamin E, rose oil, and glycerin. Each component was individually sonicated to ensure uniform dispersion.

Phase B was gradually added to Phase A, and the combined mixture was further sonicated at 25.5 °C to obtain a stable, homogenous formulation. The optimized composition of the facewash is presented in Table 1, and the final product appearance is illustrated in Figure 1. This method ensured uniform distribution of all excipients and active constituents, enhancing stability, reproducibility, and therapeutic efficacy.^{2,16}

Evaluation Parameters

The prepared formulations were evaluated for physical characteristics, surface tension, pH, viscosity, foam properties, spreadability, and antimicrobial efficacy.

Physical Characteristics

The color, odor, and clarity of the formulations were visually assessed to determine overall appearance and sensory acceptability.¹⁷

Surface Tension

Surface tension was measured to evaluate cleansing efficiency and spreading behavior. A pycnometer was used to determine the density of 10% v/v facewash solutions at room temperature. Samples were then analyzed using a stalagmometer to measure surface tension.^{18,19}

pH Determination

The pH of a 10% v/v solution of each facewash was measured using a digital pH meter, calibrated with phosphate buffers of pH 4 and 7. This ensured skin compatibility and topical safety.^{20,21}

Viscosity

The flow properties and consistency of each formulation were assessed using a digital viscometer. To prevent spillage, bottles were placed in a protective container, and viscosity measurements were recorded at multiple rotational speeds.^{14,22}

Foam Test

Foaming capacity, reflecting cleansing ability, was determined by adding 10% v/v facewash solutions to test tubes, with and without oil. Tubes were shaken 20 times, and foam height was measured at 0, 5, 10, 15, 20, 25, and 30 minutes using a reference scale.^{23,24}

Foam Stability

Foam stability was assessed to determine the persistence of foam during use. A 10% v/v solution was added to a burette and allowed to flow into a measuring cylinder to produce foam. Foam height was recorded after complete transfer. The experiment was repeated using oil to ensure reproducibility.^{25,26}

Spreadability

Spreadability was measured to evaluate the ease of application and coverage of the formulation. After a 24-hour preparation period, 1 g of facewash was placed between two 7 × 3 cm glass plates, with a 20 g weight attached to the upper plate. The spreading diameter was measured, and spreadability (S) was calculated using:

$$S = \frac{m \times l}{t}$$

where S = spreadability (g.cm/sec), m = weight of the upper plate (g), l = length of glass plate (cm), and t = time taken for the upper plate to slide the entire length (sec).²⁷

Antimicrobial Activity

The antimicrobial efficacy against *P. acnes* was evaluated using agar well diffusion. Muller Hinton medium (7.5 g) and nutritional agar (16 g) were dissolved in 500 mL distilled water, autoclaved, and poured into sterilized Petri plates. Wells were created in the solidified medium, and facewash solutions at concentrations of 40%, 20%, 10%, and 5% were carefully added. Plates were incubated at 28 °C for

20–24 hours. Zones of inhibition around the wells were measured to determine antimicrobial activity, with larger zones indicating greater efficacy.^{15, 28}

RESULTS

Polyherbal facewash formulations incorporating *Rosmarinus officinalis* (Rosemary), *Rubia cordifolia* (Manjistha), and *Gomphrena globosa* were successfully developed and evaluated for physicochemical, antimicrobial, and organoleptic properties. All formulations exhibited favorable aesthetic characteristics, including a transparent gel structure, smooth and uniform consistency, and a mild rose-like aroma, which collectively contribute to cosmetic appeal and user compliance. Surface tension values were recorded between 36.5 and 38.1 dynes/cm. These values, comparable to the marketed product, indicate effective cleansing ability and optimal wetting action, which are essential for removing dirt and excess sebum from the skin. The pH of the formulations ranged from 4.07 to 4.9, aligning with the natural acidic environment of the skin. This pH range is beneficial for maintaining the integrity of epidermal barrier and reducing the potential for microbial colonization or irritation. Rheological analysis confirmed viscosity values between 344.2 and 377.9 cps, suggesting the development of a stable gel with adequate thickness for easy handling and uniform application. Spreadability studies further revealed enhanced performance, with values up to 6.56 g•cm/sec compared to the marketed formulation (4.6 g•cm/sec) given in Figure 2A to 2D. This reflects the formulation’s ability to distribute evenly across the skin surface, ensuring effective coverage with minimal effort. Foamability and stability tests demonstrated that the polyherbal blends produced foam volumes comparable or superior to the marketed product, with the optimized formulation retaining foam stability for 23 minutes given in Figure 3A to 3B. This sustained foam stability is an important indicator of prolonged cleansing action during use. The consolidated results in Table 2 highlight the optimized blend as having a balanced physicochemical profile along with significant antimicrobial efficacy. Zone of inhibition measurements confirmed in Table 3 having the strong antibacterial potential of the formulation details given in Figure 4A and 4B. Antimicrobial evaluation against *Propionibacterium acnes* showed that the optimized 2% herbal combination exhibited the lowest IC₅₀ value (0.61), surpassing the marketed product (0.63) details given in Table 4.

Table 1: Optimization of antiacne facewash

Sr No	Ingredients	F1	F2	F3	Final
1	Carbapol 934(g)	0.37	0.4	0.44	0.44
2	Sodium Lauryl Sulphate (g)	1	0.5	0.5	0.5
3	Methyl paraben (g)	0.1	0.1	0.1	0.1
4	Active agent (g)	4	4	4	4
5	Rose oil(drops)	1-2	2-3	1-2	1-2
6	Glycerin (ml)	2	2	1.5	1.5
7	Vit E(drops)	1-2	1-2	1-2	1-2
8	Water(ml)	qs	qs	qs	qs

Table 2: Evaluation study of Antiacne facewash

Parameter	R+M	M+G	R+G	Mixture	Marketed
Color	Yellow	Slight orange	White	Yellow	Orange
Odour	Rose like	Rose like	Rose like	Rose like	Rose like
Clarity	Clear gel	Clear gel	Clear gel	Clear gel	Clear gel
Foaming ability	3.8	3.7	3.6	3.6	3
Foam Stability	23	21	21	22	19
pH	4.77	4.56	4.07	4.9	4.96
Viscosity (cps)	367.3	377.9	347.4	344.2	295.7
Surface Tension	36.5	38.1	37.7	36.9	36.77
Spreadability (g•cm/sec)	6.4	6.5	6.4	6.56	4.6
IC ₅₀	0.65	0.9	1.01	0.61	0.63

Note: R: *Rosmarinus officinalis*; M: *Rubia cordifolia*; G: *Gomphrena globosa*; D: [G+M] = *Gomphrenaglobosa* + *Rubia cordifolia*, E: [R+M] = *Rosmarinus officinalis* + *Rubia cordifolia*, F: [G+R] = *Gomphrena globosa* + *Rosmarinus officinalis*, S: Marketed formulation, Q,T [M+G +R] = 1% and 2 % mixture of *Rosmarinus officinalis* + *Rubia cordifolia* + *Gomphrena globosa*

Table 3: Zone of inhibition of antiacne facewash

Abbreviation	5%	10%	20%	40%
D	14mm	18mm	18 mm	19 mm
E	15 mm	20 mm	19mm	20 mm
F	16 mm	30 mm	22 mm	25 mm
S	7 mm	9 mm	10 mm	16 mm
Q	6 mm	8 mm	8 mm	17 mm
T	8 mm	10 mm	15 mm	21 mm

Note: R: *Rosmarinus officinalis*; M: *Rubia cordifolia*; G: *Gomphrena globosa*; D: [G+M] = *Gomphrenaglobosa* + *Rubia cordifolia*, E: [R+M] = *Rosmarinus officinalis* + *Rubia cordifolia*, F: [G+R] = *Gomphrena globosa* + *Rosmarinus officinalis*, S: Marketed formulation, Q,T [M+G +R] = 1% and 2 % mixture of *Rosmarinus officinalis* + *Rubia cordifolia* + *Gomphrena globosa*

Table 4: IC₅₀ values of antiacne facewash

Sr. no.	Formulation	IC ₅₀
1	Marketed	0.63
2	D	0.9
3	E	0.65
4	F	1.01
6	Q (1%)	0.9
7	T (2%)	0.61

Note: R: *Rosmarinus officinalis*; M: *Rubia cordifolia*; G: *Gomphrena globosa*; D: [G+M] = *Gomphrenaglobosa* + *Rubia cordifolia*, E: [R+M] = *Rosmarinus officinalis* + *Rubia cordifolia*, F: [G+R] = *Gomphrena globosa* + *Rosmarinus officinalis*,

S: Marketed formulation, Q,T [M+G +R] = 1% and 2 % mixture of *Rosmarinus officinalis* + *Rubia cordifolia* + *Gomphrena globosa*

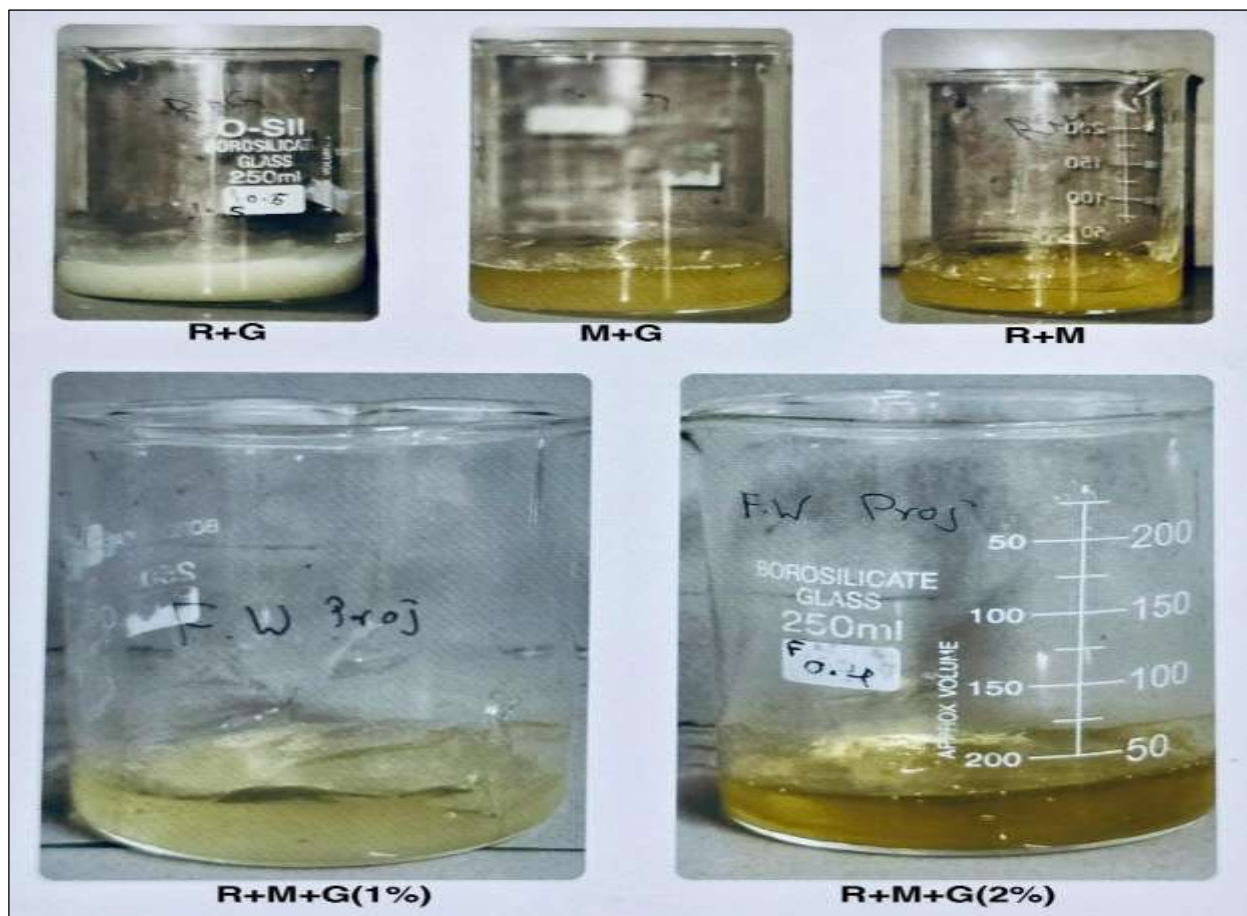


Figure 1: Herbal antiacne facewash formulation of various combinations of *Rosmarinus officinalis*, *Gomphrena globosa* and *Rubia cordifolia*

Note: R: *Rosmarinus officinalis*; M: *Rubia cordifolia*; G: *Gomphrena globosa*; D: [G+M] = *Gomphrenaglobosa* + *Rubia cordifolia*, E: [R+M] = *Rosmarinus officinalis* + *Rubia cordifolia*, F: [G+R] = *Gomphrena globosa* + *Rosmarinus officinalis*, S: Marketed formulation, Q,T [M+G +R] = 1% and 2 % mixture of *Rosmarinus officinalis* + *Rubia cordifolia* + *Gomphrena globosa*

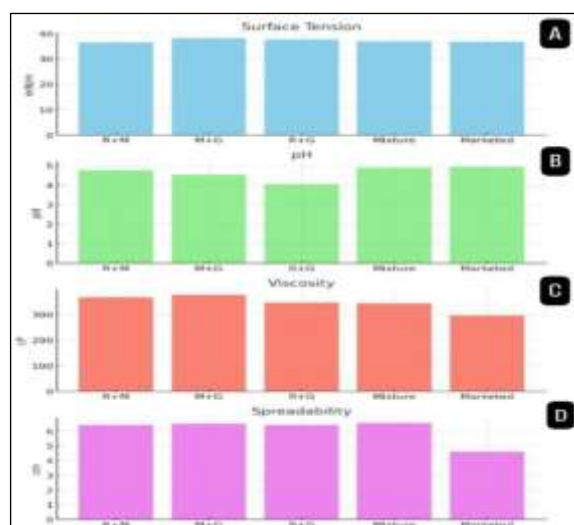


Figure 2A: Surface tension of antiacne facewash; **Figure 2B:** Determination of pH of antiacne facewash; **Figure 2C:** Viscosity determination of antiacne facewash; **Figure 2D:** Spreadability of antiacne facewash

Note: R: *Rosmarinus officinalis*; M: *Rubia cordifolia*; G: *Gomphrena globosa*; D: [G+M] = *Gomphrenaglobosa* + *Rubia cordifolia*, E: [R+M] = *Rosmarinus officinalis* + *Rubia cordifolia*, F: [G+R] = *Gomphrena globosa* + *Rosmarinus officinalis*, S: Marketed formulation, Q,T [M+G +R] = 1% and 2 % mixture of *Rosmarinus officinalis* + *Rubia cordifolia* + *Gomphrena globosa*

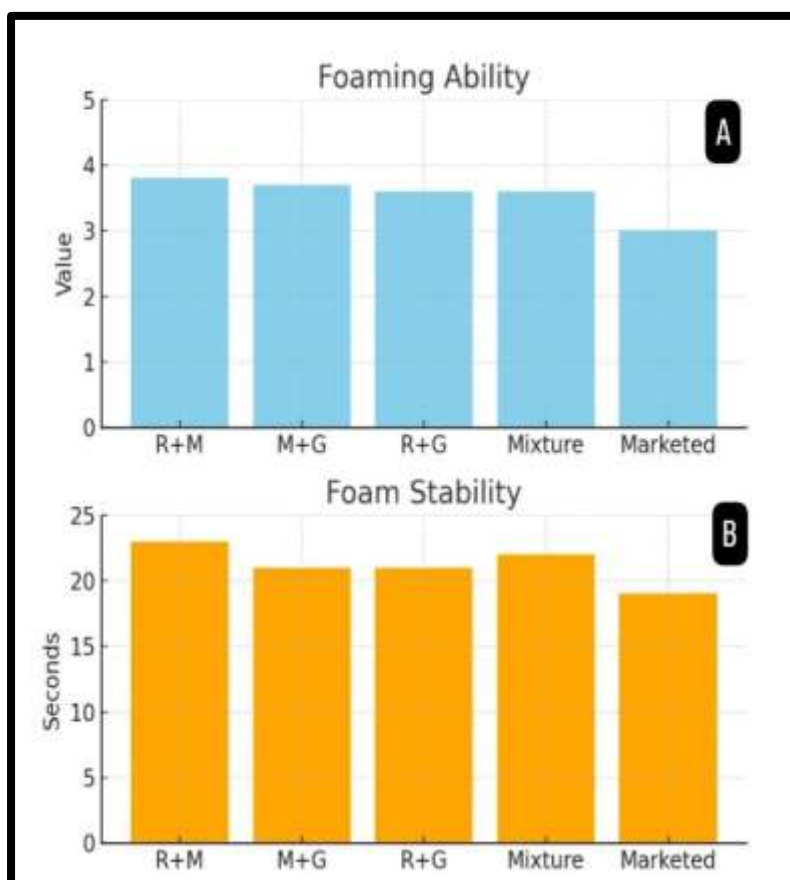


Figure 3A: Foaming ability of antiacne facewash; **Figure 3B:** Foam Stability of antiacne facewash

Note: R: *Rosmarinus officinalis*; M: *Rubia cordifolia*; G: *Gomphrena globosa*; D: [G+M] = *Gomphrenaglobosa* + *Rubia cordifolia*, E: [R+M] = *Rosmarinus officinalis* + *Rubia cordifolia*, F: [G+R] = *Gomphrena globosa* + *Rosmarinus officinalis*, S: Marketed formulation, Q,T [M+G +R] = 1% and 2 % mixture of *Rosmarinus officinalis* + *Rubia cordifolia* + *Gomphrena globosa*

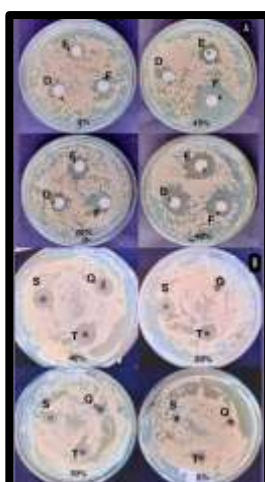


Figure 4A: Antimicrobial activity of antiacne facewash 2% combination; **Figure 4B:** Antimicrobial activity of combination and Standard antiacne facewash

Note: R: *Rosmarinus officinalis*; M: *Rubia cordifolia*; G: *Gomphrena globosa*; D: [G+M] = *Gomphrenaglobosa* + *Rubia cordifolia*, E: [R+M] = *Rosmarinus officinalis* + *Rubia cordifolia*, F: [G+R] = *Gomphrena globosa* + *Rosmarinus officinalis*,

S: Marketed formulation, Q,T [M+G +R] = 1% and 2 % mixture of *Rosmarinus officinalis* + *Rubia cordifolia* + *Gomphrena globosa*

DISCUSSION

The results demonstrate that the polyherbal facewash offers a unique combination of physicochemical stability, antimicrobial activity, and user-friendly properties, positioning it as a strong contender to synthetic anti-acne formulations. Maintaining an acidic pH range of 4.07–4.9 is a critical advantage, as it not only supports the skin's natural defense mechanisms but also helps in preventing overgrowth of acne-causing bacteria. This compatibility ensures that the product is safe for repeated daily use without compromising skin health. The viscosity values recorded suggest an ideal gel-like consistency that enhances both application and adherence, thereby prolonging contact time with the skin and increasing the effectiveness of active herbal components. Coupled with superior spreadability, the formulation ensures uniform coverage, which is particularly beneficial in addressing acne-prone areas that require thorough cleansing. Foam stability studies highlight the formulation's ability to provide effective surfactant activity for cleansing, while avoiding the excessive foaming associated with synthetic surfactants that can strip natural oils and cause dryness. The optimized foam stability of 23 minutes indicates prolonged cleansing action, which enhances product efficiency during use. Antimicrobial assays revealed that herbal formulation exerts stronger inhibitory effects against *P. acnes* than the marketed control. The significantly lower IC_{50} values suggest synergistic activity of the bioactive constituents from Rosemary (rich in phenolic diterpenes), Manjistha (containing anthraquinones and flavonoids), and *Gomphrena globosa* (noted for saponins and antimicrobial peptides). Together, these phytoconstituents may contribute to antibacterial, anti-inflammatory, and antioxidant effects, all of which are essential in acne management. In addition to performance-related parameters, the organoleptic qualities (clarity, uniform gel texture, and pleasant fragrance) enhance the consumer acceptability of the product. The avoidance of harsh chemical surfactants further supports its skin-friendly nature, reducing the risks of irritation, barrier damage, or excessive dryness, which are common drawbacks of conventional facewashes. Taken collectively, these findings emphasize that the optimized polyherbal facewash not only matches but, in many aspects, surpasses marketed formulations in terms of efficacy, safety, and user acceptability. Its integration of favorable physicochemical properties with potent antimicrobial activity makes it a scientifically supported and consumer-appealing natural alternative for long-term acne care.

SUMMARY:

In summary, the optimized polyherbal facewash demonstrates a well-balanced combination of physicochemical stability, antimicrobial potency, and favorable sensory attributes, making it a promising natural alternative to conventional synthetic anti-acne products. Its skin-compatible acidic pH, ideal gel-like viscosity, enhanced spreadability, and prolonged foam stability ensure effective cleansing while preserving the skin's natural barrier. The synergistic activity of bioactive constituents from *Rosmarinus officinalis*, *Rubia cordifolia*, and *Gomphrena globosa* contributes to significant antibacterial, anti-inflammatory, and antioxidant effects, offering superior efficacy against *Propionibacterium acnes* compared to marketed formulations. Collectively, these findings support the polyherbal facewash as a safe, effective, and consumer-friendly option for long-term acne management, highlighting its potential for integration into daily skincare routines.

CONCLUSION

The optimized polyherbal facewash exhibited stable physicochemical properties and strong antimicrobial activity, outperforming a marketed formulation against *Propionibacterium acnes*. Its skin-compatible profile and synergistic herbal constituents support its potential as a safe, effective, and natural alternative for long-term acne management.

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ABBREVIATIONS

R: *Rosmarinus officinalis*; M: *Rubia cordifolia*; G: *Gomphrena globosa*; D: [G+M] = *Gomphrena globosa* + *Rubia cordifolia*, E: [R+M] = *Rosmarinus officinalis* + *Rubia cordifolia*, F: [G+R] = *Gomphrena globosa* + *Rosmarinus officinalis*, S: Marketed formulation, Q,T [M+G +R] = 1% and 2 % mixture of *Rosmarinus officinalis* + *Rubia cordifolia* + *Gomphrena globosa*

CONFLICT OF INTEREST

The authors declare that there are no conflicts of interest.

FUNDING SOURCE

There is no funding source.

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