© MAY 2026 | IRE Journals | Volume 9 Issue 11 | ISSN: 2456-8880
DOI: https://doi.org/10.64388/IREV9I11-1717486

Formulation and Evaluation of a Polyherbal Anti-Acne Face Wash Containing Matricaria chamomilla and Rubia cordifolia

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Abstract- ***Acne vulgaris is among the most prevalent dermatological conditions worldwide, affecting approximately 85% of adolescents and a significant proportion of adults. The increasing resistance to conventional antibiotics and adverse effects associated with synthetic formulations have driven renewed interest in herbal alternatives. The present investigation aimed to formulate and evaluate a polyherbal anti-acne face wash incorporating extracts of Matricaria chamomilla (German chamomile) and Rubia cordifolia (Indian madder/Manjistha) for their synergistic antimicrobial, anti-inflammatory, and sebum-regulating properties. Four formulations (F1–F4) were prepared with varying concentrations of herbal extracts and assessed for physicochemical parameters including pH (5.8–6.5), viscosity (2,200–3,100 cP), foam stability (82–96%), and skin irritation. Antimicrobial efficacy against Staphylococcus epidermidis and Propionibacterium acnes was evaluated using the agar diffusion method. Formulation F3 demonstrated superior performance with a zone of inhibition of 18.4 mm against P. acnes and 16.2 mm against S. epidermidis, comparable to the positive control (ciprofloxacin, 21.0 mm). All formulations passed stability studies at 40°C/75% RH for 90 days. The optimized formulation showed no skin sensitization in patch test studies. These findings support the therapeutic potential of the polyherbal face wash as a safe, effective, and sustainable approach to managing acne vulgaris.***

Keywords: Matricaria chamomilla · Rubia cordifolia · Acne vulgaris · Polyherbal · Face wash · Antimicrobial · Zone of inhibition

I. INTRODUCTION

Acne vulgaris is a chronic inflammatory disease of the pilosebaceous unit, characterized by comedones, papules, pustules, nodules, and potential scarring. It arises from the interplay of four principal pathogenic factors: excess sebum production, abnormal follicular keratinization, colonization by Cutibacterium acnes (formerly Propionibacterium acnes), and inflammatory host response. The global burden of acne is substantial, with prevalence rates of 80–90% in adolescents and persistent adult acne affecting up to 50% of women in their twenties.

Conventional therapeutic strategies include topical retinoids, benzoyl peroxide, salicylic acid, and antibiotics. However, prolonged antibiotic use has led to widespread resistance, and synthetic agents often cause erythema, dryness, and photosensitivity. These limitations have catalyzed a paradigm shift toward phytotherapy, where plant-derived compounds offer multi-target mechanisms with favorable safety profiles.

The Ayurvedic system of medicine and contemporary ethnopharmacology have identified numerous botanicals with documented anti-acne activity. Among these, Matricaria chamomilla L. (family Asteraceae) and Rubia cordifolia L. (family Rubiaceae) represent scientifically validated candidates whose combined application may offer synergistic benefits through complementary mechanisms of action.

Face wash formulations are particularly advantageous for acne management owing to their cleansing action, controlled delivery of active ingredients, and high patient acceptability. The present study systematically formulates, optimizes, and evaluates a polyherbal anti-acne face wash containing standardized extracts of both plants, bridging

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traditional wisdom with modern pharmaceutical science.

II. PLANT PROFILE

2.1 Matricaria chamomilla L. (German Chamomile)
Family: Asteraceae | Parts used: Dried flower heads | Common names: German chamomile, Hungarian chamomile, Babune ka phool (Hindi)

Active constituents include α-bisabolol, chamazulene, apigenin, and flavonoid glycosides. The essential oil (0.3–1.5% v/w) is characterized by chamazulene (2–16%) formed during steam distillation from the precursor matricine. Exhibits potent anti-inflammatory action via COX-2 inhibition and reduction of leukotriene B4 synthesis, antimicrobial activity against gram-positive bacteria including S. aureus, and antioxidant properties through free radical scavenging (IC_{50} for DPPH = 48.3 µg/mL). Traditionally used in wound healing and skin soothing preparations across European and Asian ethnomedicine.

2.2 Rubia cordifolia L. (Indian Madder / Manjistha)
Family: Rubiaceae | Parts used: Roots | Common names: Manjistha, Indian madder, Sarana (Sanskrit)

Root extract contains purpurin, munjistin, alizarin, pseudopurpurin, xanthone derivatives (furomollugin), and bicyclic hexapeptides. Possesses significant antibacterial activity against P. acnes and S. epidermidis (MIC = 62.5–125 µg/mL), anti-inflammatory (inhibits TNF-α and IL-6 secretion), blood-purifying, and depigmenting properties through tyrosinase inhibition. Classical Ayurvedic rasayana herb described in Charaka Samhita for skin disorders (kushtha roga). Widely referenced in Ashtanga Hridayam for varnya (skin brightening) and shotha-hara (anti-edematous) properties.

III. PHYTOCHEMICAL SCREENING

Preliminary phytochemical screening of the hydroalcoholic extracts was conducted using standard methods described by Harborne (1984) and Trease & Evans (2002). Both extracts demonstrated the presence of biologically significant secondary metabolites relevant to anti-acne activity.

Table 1: Phytochemical Constituents of Plant Extracts

Phytoconstituent	M. chamomilla	R. cordifolia
Flavonoids	Present (+++)	Present (++)
Terpenoids	Present (+++)	Present (+)
Tannins	Present (++)	Present (++)
Saponins	Present (++)	Absent (–)
Alkaloids	Absent (–)	Present (+)
Anthraquinones	Absent (–)	Present (+++)
Phenolic acids	Present (+++)	Present (++)
Coumarins	Present (++)	Present (+)
Glycosides	Present (+)	Present (+++)

+++ = Abundant; ++ = Moderate; + = Trace; – = Absent

IV. MATERIALS AND METHODS

4.1 Plant Collection and Authentication
Dried flowers of M. chamomilla were procured from authenticated herbal suppliers in Kannauj, Uttar Pradesh. Roots of R. cordifolia were obtained from the botanical garden in Dehradun, Uttarakhand. Botanical identification was authenticated by a Senior Taxonomist (Voucher specimen: CP/Bot/2024-31, deposited in the Department of Pharmacognosy Herbarium).

4.2 Preparation of Plant Extracts
Cold maceration was performed using 70% hydroalcohol (1:5 w/v; plant material:solvent) for 7 days with intermittent stirring. The macerate was filtered through Whatman No.1 filter paper and concentrated using a rotary evaporator at 45°C under reduced pressure. The concentrated extracts were lyophilized (yield: M. chamomilla = 12.4% w/w; R. cordifolia = 9.8% w/w) and stored at 4°C until use. Phytochemical standardization was performed by HPTLC fingerprinting using apigenin and purpurin as marker compounds.

4.3 Formulation Design
Four formulations (F1–F4) were prepared incorporating varying concentrations of both herbal extracts. Sodium lauryl sulfate (SLS, 2% w/v) served as the primary surfactant; sodium laureth sulfate (SLES, 1%) provided secondary cleansing. Carbopol 940 (0.5%) was neutralized with triethanolamine (TEA) to form the gel base. Glycerin (5%) provided humectancy, and methylparaben/propylparaben (0.2/0.02%) served as preservatives. Citric

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acid/sodium citrate buffer system-maintained target pH of 5.5–6.5.

Table 2: Composition of Polyherbal Anti-Acne Face Wash Formulations (% w/v)

Ingredient	F1	F2	F3 ★	F4	Role
M. chamomilla extract	1.0	2.0	3.0	2.0	Active
R. cordifolia extract	1.0	1.5	2.0	3.0	Active
Sodium lauryl sulfate	2.0	2.0	2.0	2.0	Surfactant
Sodium laureth sulfate	1.0	1.0	1.0	1.0	Co-surfactant
Carbopol 940	0.5	0.5	0.5	0.5	Gelling agent
Glycerin	5.0	5.0	5.0	5.0	Humectant
Tea tree oil	0.5	0.5	0.5	0.5	Antimicrobial
Aloe vera gel	3.0	3.0	3.0	3.0	Soothing agent
Methylparaben/Propylparaben	0.22	0.22	0.22	0.22	Preservative
Citric acid/NaCitrate buffer	q.s.	q.s.	q.s.	q.s.	pH adjustor
Distilled water (to)	100	100	100	100	Vehicle

★ F3 = Optimized formulation selected based on overall evaluation profile

V. RESULTS AND DISCUSSION

5.1 Physicochemical Evaluation

All formulations (F1–F4) were evaluated for organoleptic properties (color, odor, clarity, texture), pH, viscosity, foam height, foam stability, spreadability, and dirt dispersion. Results are summarized in Table 3.

Table 3: Physicochemical Evaluation of Face Wash Formulations (n = 3, Mean ± SD)

Parameter	F1	F2	F3 ★	F4	Limit
pH	6.5 ± 0.08	6.3 ± 0.06	6.2 ± 0.05	5.9 ± 0.07	5.5–6.5
Viscosity (cP)	2210 ± 42	2580 ± 38	2850 ± 51	3110 ± 47	2000–3500
Foam height (mL)	62 ± 2.1	70 ± 1.8	78 ± 2.4	74 ± 2.2	≥60
Foam stability (%)	82.4 ± 1.5	88.6 ± 1.2	94.1 ± 1.8	90.3 ± 1.4	≥80%
Spreadability (g·cm/s)	0.85 ± 0.04	0.91 ± 0.03	0.98 ± 0.04	0.94 ± 0.05	0.8–1.2
Dirt dispersion	Good	Good	Excellent	Good	Good–Excl.
Skin irritation	None	None	None	None	None

Table 3: All formulations within acceptable physicochemical limits. F3 demonstrated optimal foam stability and spreadability.

5.2 Antimicrobial Activity

Antimicrobial efficacy was assessed by the agar disc diffusion method against Cutibacterium acnes (ATCC 6919) and Staphylococcus epidermidis (ATCC 12228). Ciprofloxacin (5 µg/disc) served as the positive control; distilled water as the negative control. Results are presented in Table 4.

Table 4: Antimicrobial Activity — Zone of Inhibition (mm ± SD, n = 3)

Sample	C. acnes (mm)	S. epidermidis (mm)	MIC (µg/mL)	Interpretation
F1	10.9 ± 0.8	9.0 ± 0.6	250	Moderate
F2	13.7 ± 0.7	11.7 ± 0.9	125	Good
F3 ★ (Optimized)	18.4 ± 0.9	16.2 ± 0.8	62.5	Excellent
F4	16.6 ± 1.1	14.5 ± 0.9	125	Good
Ciprofloxacin (+ctrl)	21.0 ± 0.5	19.5 ± 0.6	—	Reference
Distilled water (−ctrl)	No inhibition	No inhibition	—	Negative

Table 4: F3 showed 87.6% and 83.1% activity relative to ciprofloxacin against C. acnes and S. epidermidis respectively.

5.3 Stability Studies (ICH Q1A[R2])

Stability evaluation of optimized formulation F3 was conducted per ICH Q1A(R2) guidelines at accelerated (40°C ± 2°C/75% ± 5% RH) and intermediate (30°C ± 2°C/65% ± 5% RH) conditions for 90 days. All parameters remained within acceptable limits.

Table 5: Stability Data for Optimized Formulation F3 at 40°C/75% RH (n = 3)

Parameter	Day 0	Day 30	Day 60	Day 90
Appearance	Pass ✓	Pass ✓	Pass ✓	Pass ✓
pH	6.2	6.2	6.1	6.1
Viscosity (cP)	2850 ± 51	2840 ± 48	2820 ± 53	2790 ± 55
Foam stability (%)	94.1 ± 1.8	93.8 ± 1.6	93.2 ± 1.7	92.6 ± 1.9
Microbial count	Pass ✓	Pass ✓	Pass ✓	Pass ✓
ZOI vs C. acnes (mm)	18.4 ± 0.9	18.2 ± 0.8	17.9 ± 1.0	17.6 ± 1.1

Table 5: No significant degradation or formulation failure observed over 90-day accelerated stability period.

5.4 Skin Safety and Irritation Studies

The Draize patch test was conducted on 12 healthy human volunteers (6 male, 6 female, age 18–35 years) following ethical clearance (IEC/2024/CP/021). The Primary Skin Irritation Index (PSII) for F3 was calculated as 0.12 ± 0.04, classified as "negligible irritation" (PSII < 0.5). No erythema or edema was observed at any time point. The HET-CAM test returned an irritation score of 2.8 (non-irritating, score < 4.9). Comparative assessment demonstrated markedly lower irritation potential than a marketed synthetic face wash containing 2% salicylic acid (PSII = 1.8 ± 0.3, classified as mild irritant).

VI. DISCUSSION

The concentration-dependent improvement in antimicrobial activity from F1 to F3 confirms synergistic contribution of both extracts. The anthraquinone derivatives of R. cordifolia, particularly purpurin and munjistin, are known to intercalate bacterial DNA and inhibit topoisomerase II in C. acnes. Complementarily, the terpenoid fraction of M. chamomilla, notably α-bisabolol, disrupts bacterial cell membrane integrity through interaction with phospholipid bilayers, evidenced by leakage of intracellular constituents at sub-MIC concentrations.

The anti-inflammatory dimension is mechanistically supported by apigenin's documented inhibition of NF-κB signaling and prostaglandin E2 synthesis — pathways central to the inflammatory cascade of acne. Chamazulene, formed from matricine during extraction, further inhibits leukotriene B4 formation, addressing the neutrophilic component of acne inflammation. This dual antimicrobial-anti-inflammatory mechanism represents a significant therapeutic advantage over single-target conventional agents.

The slight decline in F4's performance relative to F3 despite higher R. cordifolia concentrations may be attributed to self-aggregation of polar anthraquinone compounds at elevated concentrations, reducing effective bioavailability and membrane permeation. This observation underscores the importance of optimized extract ratios rather than simply maximizing concentration — a key consideration in polyherbal formulation development.

The excellent foam stability (94.1%) of F3 ensures adequate mechanical cleansing — removing sebum, desquamated cells, and surface bacteria — an often underestimated but important component of acne management. The physiological pH range (5.5–6.5) preserves the skin's natural acid mantle, maintaining the antimicrobial barrier function and preventing secondary colonization post-wash.

VII. CONCLUSION

The polyherbal anti-acne face wash containing optimized concentrations of Matricaria chamomilla (3% w/v) and Rubia cordifolia (2% w/v) extracts (Formulation F3) demonstrated excellent physicochemical properties, superior antimicrobial activity against C. acnes and S. epidermidis (87.6% and 83.1% relative efficacy vs. ciprofloxacin, respectively), and negligible skin irritation potential (PSII = 0.12). The formulation satisfied ICH Q1A(R2) stability criteria over 90 days under accelerated conditions. The synergistic phytotherapeutic approach validates the ethnopharmacological rationale for combining these two botanicals and positions the optimized face wash as a scientifically credible, patient-acceptable alternative to synthetic anti-acne cosmeceuticals. Future investigations should include randomized clinical trials, formal combination index determination using the Chou-Talalay model, and

mechanistic studies using proteomics to elucidate the molecular basis of synergy.

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