# Formulation And Evaluation of Polyherbal Anti-Acne Cream

Vaishnavi. R. Bhimewad[1], Dr. A. D. Kshirsagar[2], S.M. Ambore[3]
[1,2,3]*D K Patil Institute of Pharmacy, Loha 431708 Maharashtra*

***Abstract*—Acne vulgaris is a chronic inflammatory skin disorder involving the pilosebaceous units and is highly prevalent among adolescents and young adults. The condition develops due to multiple factors, including excessive sebum secretion, follicular blockage, bacterial colonization, and inflammatory reactions. Although conventional anti-acne medications are effective, prolonged use may lead to adverse effects such as skin irritation, dryness, erythema, and the emergence of antimicrobial resistance. These limitations have encouraged the exploration of herbal formulations as safer and more biocompatible alternatives for topical acne therapy. The present investigation focused on the development and evaluation of a polyherbal anti-acne cream prepared from extracts of Psidium guajava (Guava), Azadirachta indica (Neem), Ocimum sanctum (Tulsi), and Aloe vera. These medicinal plants were selected because of their well-documented antimicrobial, anti-inflammatory, antioxidant, wound-healing, and skin-protective properties. The herbal extracts were incorporated into an oil-in-water cream base using a conventional emulsification method to obtain a stable topical formulation. The prepared formulations were evaluated for physicochemical characteristics, including appearance, Odor, pH, homogeneity, viscosity, spreadability, washability, consistency, irritancy, and stability. Among the developed formulations, the optimized batch demonstrated satisfactory physicochemical properties, good spreadability, skin-compatible pH, excellent homogeneity, and stability during storage. The combined action of the herbal ingredients is expected to inhibit acne-causing microorganisms, reduce inflammation, and promote healthy skin. Overall, the findings indicate that the formulated polyherbal cream possesses desirable pharmaceutical and cosmetic properties and may serve as a safe and effective topical preparation for acne management. The study supports the potential use of medicinal plant extracts in the development of herbal dermatological and cosmeceutical products.**

***Index Terms*—Acne, Cream, Polyherbal Formulation, Extraction, Guava, Neem, Tulsi, etc**

## I. INTRODUCTION:

### 1.1. Herbal Cosmetics:

Herbal cosmetics are cosmetic products prepared using natural ingredients obtained from medicinal plants, herbs, flowers, fruits, seeds, roots, and other botanical sources. Unlike conventional cosmetics, which often rely on synthetic chemicals for their activity, herbal cosmetics combine aesthetic benefits with therapeutic effects. These products are designed not only to improve appearance but also to maintain the health of the skin, hair, and other body tissues through the biological activities of plant-derived compounds such as flavonoids, phenolic acids, tannins, alkaloids, terpenoids, glycosides, and essential oils [1,22,26,29]

The growing awareness of the adverse effects of prolonged use of synthetic cosmetic ingredients has significantly increased consumer interest in herbal and naturally derived skincare products. Modern consumers increasingly prefer products that are environmentally friendly, biodegradable, and less likely to cause skin irritation or allergic reactions. As a result, herbal cosmetics have become an important segment of the cosmetic and pharmaceutical industries, with applications in skin care, hair care, oral hygiene, anti-aging, wound healing, and dermatological therapy [22,32,33,40]

Medicinal plants possess a wide range of pharmacological properties that make them suitable for cosmetic formulations. Many plant extracts exhibit antioxidant, antimicrobial, anti-inflammatory, moisturizing, photoprotective, and wound-healing activities. These properties help prevent skin damage, delay the aging process, promote tissue regeneration, and improve the overall appearance and health of the skin [7,20,27,37]

Based on their dosage forms, herbal cosmetics are available as creams, lotions, gels, ointments, face

packs, shampoos, soaps, powders, oils, and emulsions. Among these, creams are one of the most preferred topical dosage forms because they spread easily on the skin, provide prolonged contact with the affected area, and are cosmetically elegant. Depending on the nature of the continuous phase, creams are classified as oil-in-water (O/W) or water-in-oil (W/O) emulsions. Oil-in-water creams are non-greasy, easily washable, and generally preferred for acne-prone and oily skin because they provide good patient acceptability without blocking skin pores. [26,29,30,31]

1.2. Skin and Its Functions

The skin is the largest organ of the human body and serves as the first protective barrier against external environmental factors such as microorganisms, ultraviolet radiation, chemical irritants, and mechanical injury. It covers approximately 1.5–2.0 square meters of body surface and performs numerous physiological functions essential for maintaining homeostasis. [23,24,39]

Structurally, the skin consists of three major layers: the epidermis, dermis, and hypodermis (subcutaneous tissue).

The epidermis is the outermost layer composed primarily of keratinocytes arranged in several strata. It provides protection against pathogens and prevents excessive water loss from the body. Melanocytes present in the basal layer synthesize melanin, which protects the skin from ultraviolet radiation. [23,24]

The dermis lies beneath the epidermis and consists mainly of connective tissue rich in collagen and elastin fibers. It contains blood vessels, lymphatics, sebaceous glands, sweat glands, nerve endings, and hair follicles. The dermis provides mechanical strength, elasticity, nutrition, and sensory perception.

The hypodermis is composed predominantly of adipose tissue that cushions internal organs, stores energy, and acts as thermal insulation.

Besides acting as a protective barrier, the skin regulates body temperature, participates in immune responses, synthesizes vitamin D upon sunlight exposure, excretes metabolic waste through sweat glands, and functions as an important sensory organ.

1.3. Acne Vulgaris

Acne vulgaris is a chronic inflammatory disorder of the pilosebaceous unit and represents one of the most prevalent dermatological conditions worldwide. It affects individuals of all age groups but occurs most frequently during adolescence due to hormonal changes associated with puberty. Although acne is generally not life-threatening, it can significantly impair quality of life by causing permanent scarring, psychological stress, anxiety, depression, and reduced self-esteem. [24,25]

The disease develops through a complex interaction of multiple pathogenic mechanisms, including increased sebum production, follicular hyperkeratinisation, colonization of hair follicles by *Cutibacterium acnes* (formerly *Propionibacterium acnes*), and activation of inflammatory pathways. These events ultimately result in the formation of comedones, papules, pustules, nodules, and cysts of varying severity. [24,25]

Several endogenous and exogenous factors contribute to acne development. Hormonal fluctuations during puberty, menstrual cycles, pregnancy, and endocrine disorders stimulate sebaceous gland activity and increase sebum secretion. Genetic predisposition also plays a significant role in determining an individual's susceptibility to acne. Additional contributing factors include psychological stress, high-glycemic diets, excessive cosmetic use, environmental pollution, certain medications, and improper skincare practices. [24,25]

Although acne most commonly affects the face, lesions may also occur on the neck, chest, shoulders, and upper back, where sebaceous glands are abundant.

1.4. Pathophysiology of Acne

The pathogenesis of acne involves four interconnected biological processes.

The first stage involves excessive sebum production, primarily stimulated by androgenic hormones. Increased sebum creates a favorable environment for bacterial growth within hair follicles. [24,25]

The second stage is follicular hyperkeratinisation, during which abnormal shedding of keratinocytes blocks the follicular opening and leads to come done formation.

The third stage involves colonization by Cutibacterium acnes, an anaerobic Gram-positive bacterium that proliferates within blocked follicles. The microorganism produces lipases that hydrolyse triglycerides present in sebum into free fatty acids, triggering local inflammation. [24,25]

The fourth stage is the activation of inflammatory mediators such as cytokines, chemokines,

prostaglandins, and reactive oxygen species, resulting in erythema, swelling, pain, and formation of inflammatory lesions.

1.5. Types of Acne [24,25]

Acne lesions are broadly categorized into non-inflammatory and inflammatory lesions.

1.5.1.Non-inflammatory Acne

I. Whiteheads (Closed Comedones)

Whiteheads develop when hair follicles become blocked by keratin and sebum while the follicular opening remains closed. These lesions appear as small, flesh-colored papules without surrounding inflammation.

II. Blackheads (Open Comedones)

Blackheads occur when the follicular opening remains open, allowing the accumulated keratin and sebum to undergo oxidation upon exposure to atmospheric oxygen. The characteristic dark color results from oxidation rather than the presence of dirt.

1.5.2.Inflammatory Acne

I. Papules

Papules are small, erythematous, raised lesions resulting from inflammatory responses surrounding blocked follicles. They are usually tender upon palpation.

II. Pustules

Pustules resemble papules but contain visible purulent material composed of inflammatory cells and bacterial debris.

III. Nodules

Nodules are larger, deeper inflammatory lesions extending into the dermis. They are painful and often persist for prolonged periods.

IV. Cysts

Cysts are severe inflammatory lesions characterized by pus-filled cavities located deep within the skin. They frequently heal with permanent scarring if not adequately treated.

1.6. Conventional Treatment of Acne [24,25]

The therapeutic management of acne depends on disease severity and patient characteristics. Mild acne is commonly treated using topical agents such as benzoyl peroxide, salicylic acid, azelaic acid, topical antibiotics, and retinoids. Moderate to severe acne often requires systemic therapy including oral antibiotics, hormonal therapy, or isotretinoin.

Although these treatments are clinically effective, prolonged use may lead to adverse effects including skin dryness, irritation, erythema, photosensitivity, peeling, and bacterial resistance. These limitations have encouraged researchers to explore safer therapeutic alternatives derived from medicinal plants.

1.7. Role of Herbal Medicines in Acne Management

Medicinal plants represent an important source of bioactive compounds with antimicrobial, antioxidant, anti-inflammatory, wound-healing, and sebum-regulating activities. Unlike many synthetic drugs, herbal preparations often exhibit multiple pharmacological actions simultaneously while producing fewer adverse effects.

Psidium guajava leaves contain flavonoids, tannins, quercetin, and phenolic compounds that possess antibacterial and antioxidant activities. [4,18,20]

Azadirachta indica (Neem) contains azadirachtin, nimbidin, nimbolide, and quercetin, which exhibit antimicrobial, anti-inflammatory, and wound-healing properties. [7,9,27,28]

Ocimum sanctum (Tulsi) is rich in eugenol, ursolic acid, rosmarinic acid, and flavonoids that contribute to antibacterial and antioxidant activities. [7,9,27]

Aloe vera contains polysaccharides, vitamins, amino acids, and glycoproteins that promote wound healing, moisturization, collagen synthesis, and reduction of inflammation. [7,27,28]

The combination of these herbal ingredients in a single formulation may produce synergistic effects, enhancing therapeutic efficacy against acne while improving skin hydration and repair.

1.8. Need for the Present Study [2,5,22]

The growing demand for safe, effective, and naturally derived skincare products has increased interest in the development of herbal anti-acne formulations. Conventional therapies are often associated with adverse effects and the emergence of antimicrobial resistance, highlighting the need for alternative topical preparations.

The present study was therefore undertaken to formulate and evaluate a polyherbal oil-in-water anti-acne cream containing extracts of Psidium guajava, Azadirachta indica, Ocimum sanctum, and Aloe vera. The selected medicinal plants were chosen because of their complementary antimicrobial, anti-inflammatory, antioxidant, and wound-healing properties. The developed formulation was evaluated for physicochemical characteristics, stability, and suitability as a topical anti-acne preparation.

It is anticipated that the synergistic action of these botanical ingredients will provide a safe, effective, and cosmetically acceptable herbal alternative for acne management.

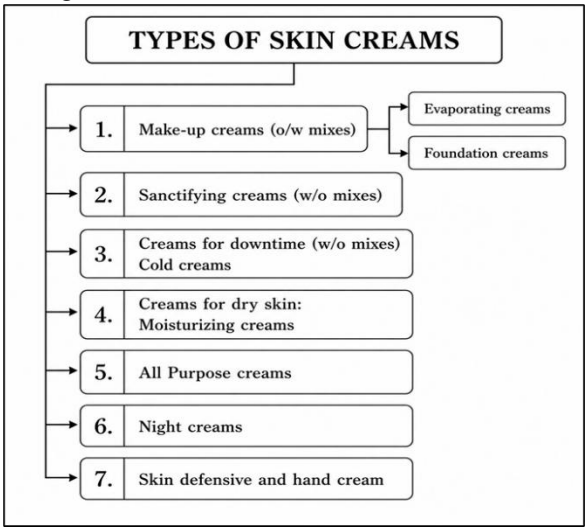


Fig No. 1. Types of Skin

1.9. Advantages of Herbal Anti-Acne Creams

1. Natural Ingredients [22,32,33]
- Prepared from plant-derived extracts and bioactive compounds.
- Free from harsh synthetic chemicals.

2. Fewer Side Effects [22,40]
- Gentle on the skin with a lower risk of irritation, redness, dryness, and allergic reactions.
- Suitable for long-term topical application.

3. Multiple Therapeutic Actions [7,20,37]
- Possess antimicrobial, anti-inflammatory, antioxidant, wound-healing, and moisturizing properties.
- Help control acne-causing bacteria while promoting skin repair.

4. Cost-Effective [32,33]
- Herbal ingredients are generally economical and readily available.
- Suitable for prolonged treatment at a lower cost.

5. Eco-Friendly and Sustainable [22,40]
- Derived from renewable natural resources.
- Biodegradable and environmentally safe compared to many synthetic cosmetic ingredients.

6. Improves Overall Skin Health [7,20,23]
- Nourishes and hydrates the skin.
- Enhances skin texture, reduces inflammation, and protects against oxidative stress.

7. Customizable Formulations [26,29,30,31]
- Herbal extracts can be combined in different proportions to suit various skin types and dermatological conditions.
- Allows the development of targeted skincare products.

8. Long-Term Benefits [2,5,15]
- Supports healthy skin by addressing the underlying causes of acne.
- Regular use may reduce acne recurrence and improve skin appearance over time.

9. Suitable for Sensitive Skin [23,40]
- Most herbal formulations are well tolerated and less likely to cause skin sensitivity compared with synthetic products.

10. Rich Source of Bioactive Phytochemicals [20,27,28]
- Contains flavonoids, tannins, phenolic compounds, alkaloids, saponins, and terpenoids that contribute to therapeutic efficacy.

11. Non-Greasy and Easy to Apply [26,29,34]
- Oil-in-water (O/W) herbal creams spread easily, are readily washable, and provide better patient compliance.

12. Reduced Risk of Antimicrobial Resistance [2,5,13,16]
- The presence of multiple phytochemicals with different mechanisms of action may reduce the likelihood of resistance development compared with prolonged use of single synthetic antibiotics.

## II. MATERIAL AND METHODS

Table No. 1 Chemicals Used

| SR. NO | CHEMICALS |
|---|---|
| 1 | Guava Extract |
| 2 | Neem Extract |
| 3 | Tulsi Extract |
| 4 | Beeswax |
| 5 | Liquid Paraffin |
| 6 | Borax |
| 7 | Methyl Paraben |
| 8 | Rose water |
| 9 | Aloe vera gel |
| 10 | Almond oil |
| 11 | Glycerin |
| 12 | Water |

Table 2. List of Equipment

| SR NO. | LIST OF EQUIPMENT |
|---|---|
| 1 | Electronic weighing balance |
| 2 | Mortar and pestle |
| 3 | Soxhlet Apparatus |
| 4 | Beakers |
| 5 | Stirrer |
| 6 | Water Bath |
| 7 | Pipettes |
| 8 | Filter Paper |
| 9 | Thermometer |
| 10 | Hot Air Oven |
| 11 | Refrigerator |
| 12 | Digital pH Meter |
| 13 | Brookfield Viscometer |
| 14 | Microbial Agar Plates |
| 15 | Storage Container |

### 2.1. Collection, identification and authentication of plant material:

Fresh leaves of *Psidium guajava* (Guava), *Azadirachta indica* (Neem), and *Ocimum sanctum* (*Ocimum tenuiflorum*, Tulsi) were collected from healthy and disease-free plants from the local area during the early morning hours. The collected plant materials were authenticated by Dr. Vishal R. Marathe, Professor of Botany and Botanist at the Plant Taxonomy Research Laboratory, Department of Botany, N.E.S. Science College. The authenticated plant specimens were used for further research and formulation studies.

The collected leaves were thoroughly washed with distilled water to remove dust, soil particles, and other extraneous matter. The cleaned leaves were shade-dried at room temperature for 7–10 days to preserve their phytoconstituents and prevent degradation of thermolabile compounds. After complete drying, the leaves were separately pulverized into coarse powders using a mechanical grinder. The powdered materials were passed through a suitable sieve to obtain uniform particle size and stored in airtight containers in a cool and dry place until further extraction, formulation, and evaluation of the polyherbal anti-acne cream.

### 2.2. Plant Profile:

1. NEEM

FigNo. 2- Neem Leaves

Table No. 3 Description of Neem

| | Description (Short) |
|---|---|
| Active Compounds | Contains tannins contributing to therapeutic effects |
| Antibacterial | Effective against *Staphylococcus aureus* and *Propionibacterium acnes* (acne-causing bacteria) |
| Anti-acne Action | Reduces pimples, controls sebum, prevents infection |
| Antifungal | Treats fungal infections like ringworm and athlete's foot |
| Anti-inflammatory | Reduces redness, swelling, irritation (acne, eczema, dermatitis) |
| Wound Healing | Promotes tissue regeneration and faster healing |

| | |
|---|---|
| Antioxidant | Protects from free radicals and premature aging |
| Skin Health | Maintains elasticity and improves overall skin condition |
| Cleansing Action | Removes impurities, unclogs pores, detoxifies skin |
| Uses in Formulation | Used in creams, gels, ointments for acne and skin disorders |

2. GUAVA

Fig. No. 3 Guava Leaf

Table No. 4 Description of Guava

| Property | Description (Short) |
|---|---|
| Plant Name | *Psidium guajava* (Guava) |
| Active Compounds | Rich in flavonoids, tannins, and vitamin C |
| Antibacterial | Effective against *Staphylococcus aureus* and *Propionibacterium acnes* |
| Anti-acne Action | Reduces pimples, prevents infection, maintains skin hygiene |
| Antifungal | Helps treat fungal skin infections |
| Anti-inflammatory | Reduces redness, swelling, irritation (acne, eczema, dermatitis) |
| Astringent | Tightens skin, reduces pores, controls excess oil |
| Antioxidant | Protects against oxidative stress and premature aging |
| Anti-aging Effect | Improves skin texture, reduces wrinkles, promotes youthful skin |
| Wound Healing | Enhances collagen synthesis and tissue regeneration |
| Cleansing Action | Removes impurities and detoxifies skin |
| Uses in Formulation | Used in creams, gels, face packs for acne and skin disorders |

3. TULSI

Fig No. 4 Tulsi Leaf

Table No. 5 Description of Tulsi Leaf

| Property | Description (Short) |
|---|---|
| Plant Name | *Ocimum tenuiflorum* (Tulsi / Holy Basil) |
| Active Compounds | Contains eugenol, flavonoids, tannins, phenolics |
| Antibacterial | Effective against acne-causing bacteria |
| Anti-acne Action | Reduces pimples and prevents skin infections |
| Antifungal | Helps treat fungal skin infections |
| Anti-inflammatory | Reduces redness, swelling, irritation |
| Antioxidant | Protects skin from oxidative stress and aging |
| Skin Purifying | Cleanses skin and removes toxins |
| Oil Control | Helps regulate excess sebum production |
| Wound Healing | Promotes faster healing of cuts and minor wounds |
| Skin Health | Improves overall skin texture and glow |
| Uses in Formulation | Used in creams, gels, face packs for acne and skin care |

2.3. Excipient Profile:

1.Beeswax

Fig No. 5 Beeswax

Table No. 6 Description of Beeswax

| Property | Description (Short) |
|---|---|
| Source | Natural wax from honeybees (*Apis mellifera*) |
| Chemical Composition | Esters (myricyl palmitate), fatty acids, hydrocarbons, alcohols |
| Appearance | Yellow to off-white solid |
| Odor | Mild honey-like odor |
| Melting Point | 62–65°C |
| Solubility | Insoluble in water; soluble in oils & organic solvents |
| Texture | Firm but pliable |

2. Liquid Paraffin:


Fig No. 6 Liquid Paraffin

Liquid paraffin, also known as mineral oil, is a highly refined fraction obtained from petroleum. It consists primarily of saturated hydrocarbons and is widely used in pharmaceutical and cosmetic formulations due to its chemical stability, safety, and excellent emollient properties.

3. Borax:

Fig No. 7 Borax

Borax, chemically known as sodium borate, is a naturally occurring mineral compound widely used in pharmaceutical and cosmetic formulations. It functions primarily as a buffering agent, emulsifying aid, and preservative enhancer, making it valuable in topical cream formulations.

4.Methyl Paraben:


Fig No. 8 Methyl Paraben

Methyl paraben is a widely used antimicrobial preservative in pharmaceutical and cosmetic preparations. It belongs to the class of parabens (esters of p-hydroxybenzoic acid) and is commonly incorporated into topical formulations to prevent microbial contamination and enhance product shelf life.

5.Rose water:

Fig No. 9 Rose Water

Rose water is a natural aqueous distillate obtained from the petals of *Rosa* species, commonly *Rosa damascena*. It is widely used in cosmetic and pharmaceutical preparations due to its soothing, anti-

inflammatory, and mild antiseptic properties. In polyherbal anti- acne creams, rose water serves both as a functional excipient and a therapeutic agent.

6.Aloe vera gel [3,22]

Fig No. 10 Aloe Vera gel

Aloe vera gel is a natural mucilaginous substance obtained from the leaves of *Aloe barbadensis Miller*. It is widely used in dermatological and cosmetic formulations due to its anti- inflammatory, antimicrobial, moisturizing, and wound-healing properties. In polyherbal anti- acne creams, Aloe vera gel serves both as an active therapeutic agent and a base component.

7.Almond oil:

Fig No. 11 Almond Oil

Almond oil is a natural fixed oil obtained from the kernels of *Prunus amygdalus* (commonly *Prunus dulcis*). It is widely used in pharmaceutical and cosmetic formulations due to its emollient, nourishing, and skin-soothing properties. In polyherbal anti-acne creams, almond oil functions as both a carrier oil and a skin-conditioning agent.

8.Glycerine:


Fig No. 12 Glycerine

Glycerin, also known as glycerol, is a widely used humectant in pharmaceutical and cosmetic formulations. It is a trihydroxy alcohol known for its excellent moisturizing, hygroscopic, and skin-protective properties. In polyherbal anti-acne creams, glycerin plays a crucial role in maintaining skin hydration and improving formulation stability.

2.4. Preformulation Study:

The pre-formulation study is the first step in the development of dosage forms of a drug substance. Before using any drug for the preparation of a dosage form, pre-formulation studies are done, which focus on physicochemical properties of the substance like organoleptic properties, Solubility, Calibration curve, compatibility studies, etc.

It provides important information for designing the formulation. Hence, pre-formulation studies were performed for the obtained sample of the drug for identification and compatibility studies as well as for the selection of a vehicle.

The following pre-formulation studies were done for the polyherbal drug.

1. Organoleptic Properties:

Table No. 7 Organoleptic Properties

| Plant Name | Extract Type | Appearance | Odor | Texture |
|---|---|---|---|---|
| *Psidium guajava* (Guava) | Ethanolic Extract | Dark green, semi-solid | Characteristic herbal odor | Slightly coarse |
| *Azadirachta indica* (Neem) | Ethanolic Extract | Dark brownish-green | Strong, pungent odor | Sticky consistency |
| *Ocimum sanctum* (Tulsi) | Ethanolic Extract | Greenish-brown | Pleasant aromatic odor | Smooth |

2. Flow Property Evaluation:

1. Angle of Repose (θ):

The angle of repose is a crucial pre-formulation factor utilized to assess the flow characteristics of powders and granules. It is characterized as the largest angle created between the surface of a powder mound and the horizontal plane.

The angle of repose refers to the angle created between the flat surface and the slope of the pile formed when powder flows easily through a funnel.

2.Bulk Density:

Bulk density is a crucial pre-formulation factor that indicates how well a powder can be packed. It is described as the mass of the powder divided by the entire bulk volume filled by the powder, including the gaps between the particles

Bulk density is the ratio of the weight of powder to its bulk volume before tapping.

3.Tapped Density:

Tapped density serves as a crucial pre-formulation metric that reflects the packing capacity and compressibility of a powder following mechanical tapping. It is defined as the mass of the powder divided by the volume occupied following tapping

Tapped density is the ratio of the weight of powder to the tapped volume obtained after mechanically tapping the measuring cylinder containing the powder.

4. Carr's Compressibility Index:

Carr's index, or the Compressibility Index, is a key pre-formulation factor utilized to assess the flow characteristics and compressibility of powders and granules. It is determined by utilizing values for bulk density and tapped density.

5.Hausner Ratio:

Hausner's ratio is a crucial preformulation factor utilized to assess the flow characteristics of powders and granules. It is established from the connection between bulk density and tapped density

2.5. Formulation Study:

Extraction Process For:

1. Guava
2. Neem
3. Tulsi

- Fresh green leaves were collected and washed thoroughly with distilled water. The leaves were separated from the branches and air-dried at room temperature. The dried leaves were pulverized using a grinder and stored in an airtight glass container until further use.
- Approximately 50 g of powdered Tulsi leaves was macerated with 200 mL of absolute ethanol in a covered beaker. The beaker was sealed with aluminum foil and kept in a dark place for 7 days with intermittent stirring to ensure complete extraction. After maceration, the mixture was filtered through clean muslin cloth, and the filtrate was collected. The filtrate was concentrated using a rotary evaporator and further air-dried to obtain the dried extract.

Give in point wise

- Extraction of Tulsi Leaves by Cold Maceration Method
- Fresh green Tulsi leaves were collected from the plant.
- The leaves were washed thoroughly with distilled water.
- Leaves were separated manually from the branches.
- The leaves were air-dried at room temperature until complete drying.
- The dried leaves were pulverized into a fine powder using a grinder.
- The powdered material was stored in an airtight glass container until further use.
- About 50 g of Tulsi leaf powder was transferred into a beaker.

- The powder was macerated with 200 mL of absolute ethanol.
- The beaker was sealed with aluminum foil and kept in a dark place for 7 days.
- The mixture was stirred intermittently during the maceration period to ensure complete extraction.
- After 7 days, the mixture was filtered through clean muslin cloth.
- The filtrate was collected in a clean beaker.
- The collected filtrate was concentrated using a rotary evaporator.
- The concentrated extract was further air-dried to obtain the dried Tulsi extract.
- The dried extract was stored in a suitable airtight container for further studies.


Fig No. 13 Extraction of Guava, Neem, Tulsi

Formulation:

The present study was carried out to develop a polyherbal anti-acne cream using ethanolic extracts of *Psidium guajava* (Guava), *Azadirachta indica* (Neem), and *Ocimum sanctum* (Tulsi). These plants were selected based on their well-established antimicrobial, anti-inflammatory, antioxidant, and wound-healing properties, which are beneficial in the treatment of acne.

Table No. 8 Ingredients & role in Formulation

| Ingredient | Role in Formulation |
|---|---|
| Guava extract | Antioxidant, anti-acne |
| Neem extract | Antibacterial, anti-inflammatory |
| Tulsi extract | Antimicrobial, soothing agent |
| Beeswax | Thickening agent, stabilizer |
| Liquid paraffin | Emollient, enhances spreadability |
| Emulsifier | Stabilizes oil and water phases |
| Preservatives | Prevent microbial growth |
| Water | Vehicle |

2.6. Method of Preparation:

The polyherbal anti-acne cream was formulated by the emulsion method, which is widely used for preparing stable topical creams. The formulation involved the preparation of two separate phases, namely the oil phase and the aqueous phase, followed by emulsification.

Step 1: Preparation of Oil Phase

The oil-soluble ingredients, namely beeswax, liquid paraffin, and almond oil, were accurately weighed and transferred into a clean, dry beaker. The mixture was heated on a water bath at 70–75°C until the beeswax melted completely and a clear, homogeneous oily phase was obtained.

Step 2: Preparation of Aqueous Phase

In a separate beaker, the required quantity of rose water was heated to the same temperature (70–75°C). Borax was dissolved completely in the heated rose water with continuous stirring. The ethanolic extracts of Neem (Azadirachta indica), Tulsi (Ocimum sanctum), and Guava (Psidium guajava) leaves were then added gradually with constant stirring to obtain a uniform dispersion.

Step 3: Emulsification

The hot aqueous phase was added slowly to the oil phase with continuous stirring. The addition was carried out gradually while maintaining the temperature to ensure proper emulsification and to obtain a stable oil-in-water (O/W) cream.

Step 4: Homogenization

The resulting emulsion was stirred continuously using a mechanical stirrer or glass rod until a smooth,

uniform, and creamy consistency was achieved. This step improved the texture and stability of the formulation.

Step 5: Cooling
The prepared cream was allowed to cool gradually to room temperature with continuous stirring to prevent phase separation and ensure uniform distribution of all ingredients.

Step 6: Addition of Heat-Sensitive Ingredients
After the cream temperature decreased to below 40°C, Aloe vera gel and glycerine were incorporated with gentle stirring. These ingredients were added during the cooling stage to preserve their moisturizing and soothing properties.

Step 7: Packaging
The finished polyherbal cream was transferred into clean, dry, airtight containers, properly labelled, and stored at room temperature until further physicochemical, stability, and antimicrobial evaluation.

Fig No. 14 Formulated Cream

Table No. 9 Formula for Cream (20gm)

| Ingredients | F1 | F2 | F3 | F4 (Optimized) |
|---|---|---|---|---|
| Guava extract | 1.5 ml | 2 ml | 1 ml | 2.5 ml |
| Neem Extract | 1.5 ml | 1.5 ml | 1 ml | 1 ml |
| Tulsi Extract | 1.5 ml | 1 ml | 2.5 ml | 1 ml |
| Bees wax | 3.0 gm | 3.0 gm | 3.0 gm | 3.0 gm |
| Liquid paraffin | 3.0 ml | 3.0 ml | 3.0 ml | 3.0 ml |
| Borax | 0.3 gm | 0.3 gm | 0.3 gm | 0.3 gm |
| Methyl paraben | 0.2 gm | 0.2 gm | 0.2 gm | 0.2 gm |
| Rose water | 1 ml | 1 ml | 1 ml | 1 ml |
| Aloe vera gel | 2 ml | 2 ml | 2 ml | 2 ml |
| Almond oil | 0.5 ml | 0.5 ml | 0.5 ml | 0.5 ml |
| Glycerine | 0.5 ml | 0.5 ml | 0.5 ml | 0.5 ml |
| Water | q.s | q.s | q.s | q.s |

## III. EVALUATION OF CREAM

1. Homogeneity:
The homogeneity of the formulation was examined by visual appearance and by touch.

2. Dye test:
The dye test is used for the evaluation to determine the type of formulation i.e., w/o type or w/o type. The prepared formulation was mixed with the scarlet red dye and placed on the microscopic slides.

The slide is covered with a coverslip and examined under the microscope. If the dispersed globules appear red and the ground appears colorless, then the cream is o/w type and vice versa, i.e., the disperse globules appear colourless in the red ground.

3. pH of the cream:
The pH of the cream was measured by using pH meter. The pH meter was calibrated by using a standard buffer solution .0.5 g of weighed cream is taken and dissolved in 50ml of distilled water and the pH is measured.

4. Appearance:
The cream's appearance can be determined by its colour, pearlescence, and roughness.

5. Type of smear:
The type of smear or film formed on the skin is checked after the application.

6. Removal of applied cream:
The easy removal of applied cream was checked by washing the part where the cream was applied with tap water or distilled water.

7. Viscosity:
The formulation viscosity was measured by using a Brookfield viscometer with spindle no 7 at 100 rpm.

8. Spreadability:
The spreadability was expressed in terms of time in seconds taken by two slides to slip off from the cream, placed in between the slides, under a certain load. The lesser the time taken for the separation of the two slides, the better the spreadability. Then a weight or certain load was placed on the upper slide so that the cream between the two slides was pressed uniformly to form a thin layer.

The spreadability was calculated using the following formula
Spreadability (S)=tm×l
Where,
*m*= Standard weight, which is tied to or placed over the upper slide
*l*= length of a glass slide
*t*= time taken in seconds

9. Irritancy test:
Mark an area (1sq.cm) on the left-hand dorsal surface. The cream was applied to the specified area and time was noted. Irritancy, erythema, oedema, was checked if any for regular intervals up to 24 hours and repeated.

## IV. RESULTS AND DISCUSSION:

Preformulation Result:
*1. Psidium guajava*

Table No. 10 Guava

| PARAMETER | A | B | C | D |
|---|---|---|---|---|
| Sieve No. (#) | 44 | 60 | 80 | 100 |
| Bulk density (g/ml) | 0.33 | 0.39 | 0.37 | 0.30 |
| Tapped density (g/ml) | 0.4 | 0.46 | 0.48 | 0.40 |
| Carr's index | 17.5 | 15.2 | 22.9 | 25.00 |
| Hausner's ratio | 1.21 | 1.17 | 1.29 | 1.33 |
| Angle of repose (°) | 33 | 35 | 39 | 41 |
| Ash value (%) | 7.5 | 8.0 | 9.0 | 9.5 |

The micromeritic properties of formulations A–D showed variation with particle size. Formulation B exhibited the best flow properties, with the lowest Carr's index (15.2%) and Hausner's ratio (1.17), indicating good compressibility and flowability. In contrast, formulation D showed poor flow behavior, as indicated by higher Carr's index (25%) and Hausner's ratio (1.33), likely due to smaller particle size.
The angle of repose increased from A (33°) to D (41°), confirming a decrease in flowability. Bulk and tapped density values also supported that formulation B had better packing ability.
Ash values ranged from 7.5% to 9.5%, with formulation A showing the highest purity and formulation D containing more inorganic residue.
Overall, formulation B demonstrated optimal micromeritic properties, while formulation D showed poor flow characteristics.

*2. Azadirachta indica*

Table No. 11 Neem

| PARAMETER | A | B | C | D |
|---|---|---|---|---|
| Sieve No. (#) | 44 | 60 | 80 | 100 |
| Bulk density (g/ml) | 0.38 | 0.37 | 0.37 | 0.32 |
| Tapped density (g/ml) | 0.4 | 0.40 | 0.46 | 0.46 |
| Carr's index | 5.05 | 7.5 | 19.5 | 30.4 |
| Hausner's ratio | 1.05 | 1.08 | 1.24 | 1.43 |
| Angle of repose (°) | 34 | 36 | 41 | 43 |
| Ash value (%) | 8.0 | 8.5 | 9.0 | 9.0 |

The micromeritic evaluation showed that formulation A had the best flow properties, with the lowest Carr's index (5.05%) and Hausner's ratio (1.05). Formulation B also showed good flow, while formulations C and D exhibited poorer flow due to higher compressibility values.
The angle of repose increased from A (34°) to D (43°), indicating a decrease in flowability with decreasing particle size. Bulk and tapped density values were comparable, though formulation D showed relatively lower bulk density.

Ash values ranged from 8.0% to 9.0%, with formulation A being more pure and formulations C and D showing slightly higher inorganic content.

Overall, formulation A demonstrated the best micromeritic properties, while formulation D showed the poorest flow characteristics.

*3. Ocimum sanctum*

Table No. 12 Tulsi

| PARAMETER | A | B | C | D |
|---|---|---|---|---|
| Sieve No. (#) | 44 | 60 | 80 | 100 |
| Bulk density (g/ml) | 0.33 | 0.33 | 0.34 | 0.38 |
| Tapped density (g/ml) | 0.44 | 0.46 | 0.42 | 0.50 |
| Carr's index | 25 | 28 | 19 | 24 |
| Hausner's ratio | 1.33 | 1.39 | 1.23 | 1.31 |
| Angle of repose (°) | 33 | 37 | 40 | 41 |
| Ash value (%) | 7.0 | 8.5 | 9.0 | 9.5 |

The micromeritic evaluation indicates variation in flow properties among formulations A–D. Formulation C showed relatively better flowability, with the lowest Carr's index (19%) and Hausner's ratio (1.23), whereas formulation B exhibited the poorest flow due to the highest Carr's index (28%) and Hausner's ratio (1.39). The angle of repose increased from A (33°) to D (41°), indicating reduced flowability with decreasing particle size. Bulk and tapped density values suggest that formulation D has better packing ability due to higher density values.

Ash values ranged from 7.0% to 9.5%, with formulation A showing higher purity and formulation D containing more inorganic residue.
Overall, formulation C demonstrated comparatively better micromeritic properties, while formulation B showed poor flow characteristics.

Organoleptic Properties of Polyherbal Cream

Table No. 13 Organoleptic Properties of Polyherbal Cream

| Parameter | Observation | Inference |
|---|---|---|
| Appearance | Smooth and homogeneous, no lumps or phase separation | Indicates proper formulation and uniform mixing |
| Color | Light green to pale green | Due to presence of herbal extracts (Neem, Tulsi, Guava) |
| Odor | Pleasant, mild herbal odor | Acceptable and non-irritating to users |
| Texture | Smooth and non-gritty | Shows uniform dispersion of ingredients |
| Consistency | Good, neither too thick nor too runny | Ensures easy application on skin |

The organoleptic evaluation showed that the formulation possessed desirable physical characteristics. The smooth and homogeneous appearance without lumps indicates proper mixing and formulation stability. The light green color confirms the presence of herbal ingredients such as neem, tulsi, and guava. The pleasant herbal odor suggests good acceptability and non-irritating nature for users. The smooth, non-gritty texture reflects uniform dispersion of ingredients, while the good consistency ensures ease of application on the skin.
Overall, the formulation exhibited acceptable organoleptic properties suitable for topical use.

Evaluation Parameter Result:

Table No. 14 Evaluation Parameter

| Parameter | F1 | F2 | F3 | F4 |
|---|---|---|---|---|
| Homogeneity | Slightly uniform, few lumps | Uniform, no lumps | Smooth and uniform | Smooth, uniform, no lumps |
| Appearance | Light green, slightly dull | Light green, smooth | Light green, glossy | Light green, smooth, glossy |
| Odor | Mild herbal odor | Pleasant herbal odor | Pleasant herbal odor | Mild pleasant herbal odor |
| pH | 6.5 ± 0.1 | 6.3 ± 0.1 | 6.2 ± 0.1 | 6.2 ± 0.1 |
| Type of Emulsion | O/W | O/W | O/W | O/W |
| Viscosity (cps) | 11,600 ± 180 | 12,000 ± 150 | 12,300 ± 180 | 12,500 ± 200 |
| Spreadability (g·cm/sec) | 5.8 ± 0.2 | 6.1 ± 0.2 | 6.3 ± 0.2 | 6.5 ± 0.3 |

| | | | | |
|---|---|---|---|---|
| Smear Type | Slightly greasy | Thin film | Non-greasy | Non-greasy, thin film |
| Washability | Washable with water | Easily washable | Easily washable | Easily washable with water |
| Greasy Property | Slightly greasy | Slightly non-greasy | Non-greasy | Non-greasy |
| After Feel | Slightly sticky | Smooth | Soft and smooth | Soft, smooth, non-sticky |
| Irritancy Test | No irritation | No irritation | No irritation | No irritation, erythema, or edema |
| Phase Separation | Absent | Absent | Absent | Absent |
| Thermal Stability | Stable | Stable | Stable | Stable |

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