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(Research Article)

Formulation and Evaluation of Clitoria ternatea Based Herbal Lotion for the Management of Psoriasis

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ABSTRACT

Clitoria Ternatea Butter Pea Flower Hydroethanolic Extract Formulation of an Herbal Lotion with Anti-Inflammatory Properties Against Psoriasis is a chronic autoimmune inflammatory skin condition that affects about 2-3% of people around the world. It is characterized by thickened red, scaly patches on the body due to abnormal multiplication of skin cells and T-cell mediated inflammation. Current treatments such as corticosteroids, biologic agents and immunosuppressive drugs are often expensive and come with many side effects. This has led researchers to seek out safer alternatives. The purpose of this research was to formulate and evaluate a new herbal lotion containing hydroethanolic extract of Clitoria ternatea flower (also known as butterfly pea) along with other herbs such as aloevera gel, turmeric powder (containing curcumin), clove oil, acacia gum, xanthan gum, natural waxes and rose water. The lotion was created by mixing the various ingredients together in an oil/water emulsion and then evaluating its physical properties. Organoleptic characteristics were evaluated which include appearance, color, odor and texture; pH level was measured; homogenity was examined through visual inspection; viscosity (thickness) and spreadability were determined through viscometer measurements; washability and irratibility/sensitivity testing were performed on healthy volunteers; antimicrobial activity was tested against Malassezia spp. and Candida spp. using the disk diffusion test; lipoxygenase inhibiting activity (anti-inflammatory activity) was assayed in vitro; and finally MTT cytotoxic assays were conducted to determine if the lotion would inhibit keratinocyte proliferation. Results indicated that the pH levels of the lotion ranged from 5.5-6.5, it had good spreadability, a smooth texture and did not cause any skin irritation or sensitivity reactions in healthy volunteers. Furthermore, the lotion exhibited a strong zone of inhibition against all tested strains of bacteria.

Keywords: *Clitoria ternatea; psoriasis; herbal lotion; anti-inflammatory; keratinocyte; butterfly pea; topical formulation.*

I. INTRODUCTION

Psoriasis is a complex, chronic condition where the immune system causes an inflammatory response in the skin. It affects about two percent of people globally, or approximately 125 million people. In addition to being a source of stress for those who suffer from it, psoriasis can negatively affect a person's mental health, causing feelings of depression and anxiety. Additionally, research shows that individuals who experience psoriasis have higher rates of certain systemic comorbidities, such as psoriatic arthritis, cardiovascular disease, and metabolic syndrome compared to individuals without the disease. The primary mechanism through which psoriasis develops is through an abnormal interaction between the immune system and other components of the skin. When dendritic cells become activated, they encourage different types of T helper (Th1 & Th17) lymphocytes to differentiate. Once differentiated, the new T helper cells produce various pro-inflammatory cytokines, including tumor necrosis factor-alpha (TNF-alpha), interleukin-17A (IL-17A), IL-22, and IL-23. As a result of these cytokines, there is increased proliferation of keratinocytes. This rapid cell division accelerates the process of the normal 28-day process of removing dead cells from the top layer of skin to as little as three-five days. This results in the typical scaly patches associated with psoriasis. Various treatment options exist for psoriasis, ranging from topical medications like corticosteroids, vitamin D analogs, and retinoids to light therapy, oral immunosuppressive drugs (e.g., methotrexate, cyclosporine, acitretin), and biological agents directed against specific parts of the TNF-alpha, IL-17, and IL-23 signaling pathways. While each class of medication has proven to be effective in treating the symptoms of psoriasis, all have significant side effects. Prolonged use of corticosteroids leads to thinning of the skin and reduced effectiveness; long-term use of systemic medications leads to liver toxicity, kidney damage, and suppression of the immune system; and while biologics represent a major advancement in the treatment of

psoriasis due to their ability to target specific proteins involved in the disease process (and thus potentially offering fewer side effects than systemic medications), they also come with high costs and infection risks. As such, many researchers have turned to investigating herbal and botanical compounds as possible alternatives to existing treatments. Of particular interest is Clitoria ternatea L. (Fabaceae family), often referred to as butterfly pea or Aparajita. Native to India and Southeast Asia, C. ternatea has been used for centuries in traditional Ayurvedic medicine to treat a variety of ailments including inflammation, skin problems, anxiety, and memory loss [12][13]. Anthocyanins (ternatins), flavonoids (quercetin and kaempferol) and triterpenoids found in the flowers of C. ternatea are responsible for its antioxidant activity and its anti-inflammatory and antimicrobial activities [10], [11]. Studies have shown that extracts of C. ternatea contain constituents capable of inhibiting enzymes involved in the biosynthesis of prostaglandins (i.e. lipoxygenase and cyclooxygenase) and decreasing the expression of NF-kappa B transcription factors [14] — both mechanisms relevant to the development of psoriasis. In terms of its potential applications in managing psoriasis, one advantage of developing a lotion dosage form containing extracts from C. ternatea is that it will provide simultaneous hydration to the affected area(s) of skin as well as provide a means for delivering active ingredients across a larger portion of the skin surface. Research has demonstrated that when plant extracts are combined with excipients (such as aloevera gel, curcuma longa extract and acacia gum) within a single topical cream product, that synergistic anti-inflammatory, humectant and protective functions can occur [7][8][17]. To date however, no thorough investigation has reported on either the formulation optimization of a C. ternatea-based herbal lotion for treating psoriasis or examined whether such a lotion would possess anti-inflammatory or antiproliferative effects against keratinocyte cultures. Therefore, the purpose of this project was to develop an optimized C. ternatea-based herbal lotion formulation for treating psoriasis, determine its physical characteristics, evaluate its potential anti-inflammatory effects using keratinocyte cultures, and examine its capacity to prevent keratinocyte proliferation. (Figure 1 to Figure 6.)

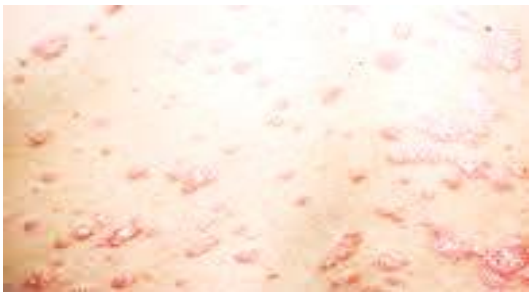

Figure 1. Guttate Psoriasis.

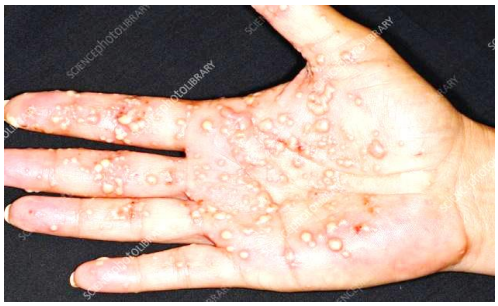

Figure 2. Pustular Psoriasis.

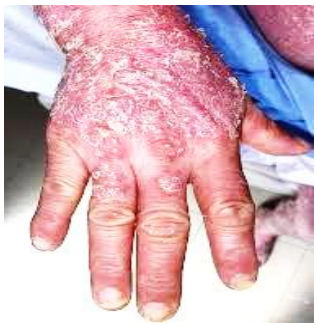

Figure 3. Erythrodermic Psoriasis.

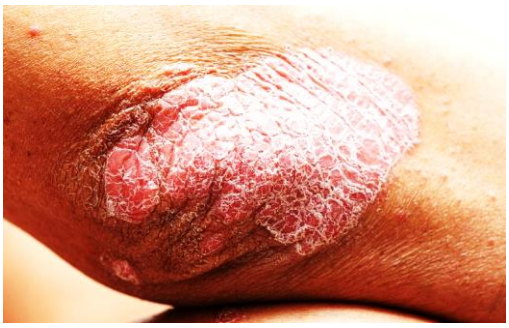

Figure 4. Plaque Psoriasis.

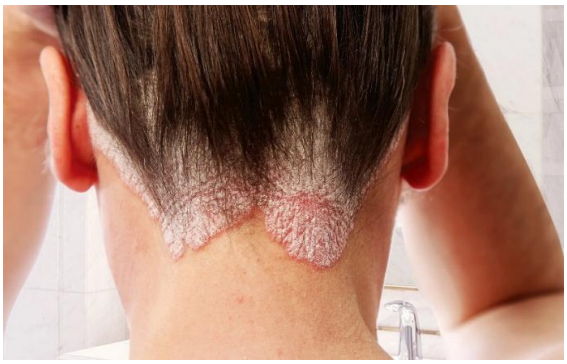

Figure 5. Scalp Psoriasis.

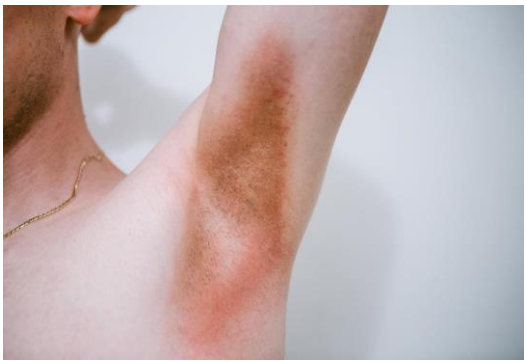

Figure 6. Inverse Psoriasis.

II. REVIEW OF LITERATURE

A. Psoriasis: Epidemiology and Pathogenesis

Psoriasis is an immune-mediated dermatological condition that has been estimated to affect approximately 2% of the global population. This condition also ranks as one of the most common forms of autoimmune disease that impacts the skin [1]. The disease has a bimodal distribution regarding the age of onset: type I (early-onset) typically reaches its peak incidence among the 20-30-year-old population. Type I is frequently found to be associated with HLA-Cw6 positive status. In contrast, type II (late-onset) generally occurs after the individual reaches 50 years of age [2]. A significant genetic predisposition exists for this condition; it has been shown that first degree relatives of patients who suffer from psoriasis are at a substantially increased lifetime risk of developing the disease compared to the general public [3]. Additionally, environmental factors such as streptococcal infection, emotional/psychological distress, mechanical trauma (the Koebner phenomenon), lithium, beta-blockers, and antimalarial drugs can serve as either precipitating or exacerbating factors for flare-ups [2,5]. The molecular mechanism underlying the pathogenesis of psoriasis includes a self-reinforcing cycle of inflammation. When plasmacytoid dendritic cells detect antimicrobial peptide-DNA complexes, they release interferon-α, which then activates myeloid dendritic cells to produce IL-12, IL-23 and TNF-α [3,4]. It is IL-23 that is responsible for promoting the proliferation of Th17 cells. Th17 cells subsequently release IL-17A, IL-17F and IL-22. IL-17A acts upon keratinocytes to stimulate them to increase production of antimicrobial peptides, chemokines (CXCL1, CXCL8) and additional pro-inflammatory cytokines. This action serves to enhance neutrophil migration into the site and perpetuate the inflammatory process [4]. Activation of NF-κB results in an increase in expression of keratins 6 and 16. These keratins contribute to hyperkeratosis and delayed maturation of the epidermis [3]. (Figure 7.)

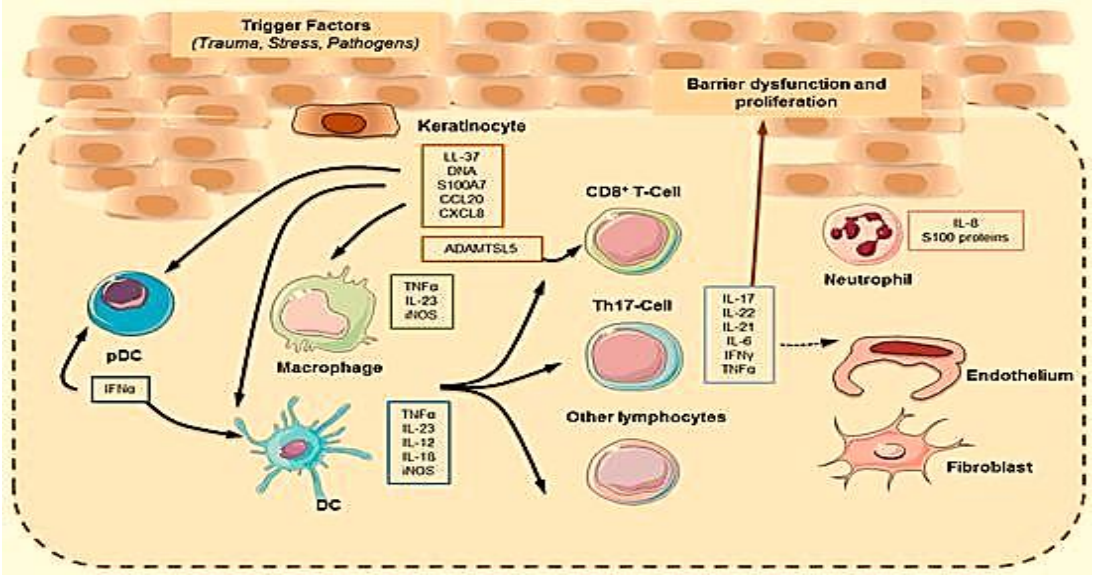

Figure 7. The pathogenesis of psoriasis.

B. Herbal Approaches to Psoriasis Management

The growing interest in herbal medicine as alternative treatment options for psoriasis is due primarily to the fact that they can act upon multiple targets, are culturally acceptable, and have favorable side effect profiles [6, 19] . Of particular note is curcumin (Curcuma longa), which has been shown to inhibit NF-kappa B activation and reduce keratinocyte proliferation, resulting in clinical improvements when applied topically in patients with plaque psoriasis [16]. In addition to anti-inflammatory properties, wound healing and moisturizing properties are associated with aloevera gel (Aloe barbadensis) through acemannan and anthraquinone-mediated mechanisms, thus providing a valuable base for emollients [15, 23]. Eugenol, found in clove oil (Syzygium aromaticum), results in analgesic, anti-inflammatory, and antimicrobial activities pertinent to the pruritic sensation experienced by individuals with psoriasis as well as the increased risk of developing secondary infections from open psoriatic lesions [20] . Acacia gum creates a hydrophilic film at the surface of skin that prevents water loss through transdermal routes and maintains skin hydration [19, 22]. Although there is evidence demonstrating Clitoria ternatea's pharmacologic properties as an antioxidant agent (Kamkaen & Wilkinson, 2009) attributed to the anthocyanin content within its petal extracts; an antioxidant/hepatoprotectant (Shankar et al., 2008); and an anti-inflammatory agent (Nair et al., 2013) based on inhibition of carrageenan-induced paw edema in rats via extracts derived from roots; and cognitive enhancing/anxiolytic effects (Jain et al., 2003) suggesting that this plant may influence both inflammatory and neurologic pathways. Nonetheless, the use of Clitoria ternatea in the development of dermatological products targeted towards treating psoriasis is still an unexplored area.

C. Nano- and Conventional Carriers for Topical Drug Delivery

The difficulty of topical delivery for psoriasis is compounded by a number of factors; specifically, the need to overcome the barrier posed by the stratum corneum while achieving a therapeutically adequate concentration of

drug within the epidermis and dermis [6]. The majority of topical preparations employed as first line treatment modalities (creams, ointments, etc.) have inherent limitations related to drug penetration into the skin and patient compliance. In this regard, advanced nano-carrier systems including liposomes, solid lipid nanoparticles (SLNs), nano-structured lipid carrier (NLCs), ethosomes, transferosomes, nano-emulsions, and polymeric nanoparticles have all been investigated in order to increase skin penetration and provide sustained release characteristics [6, 21]. For example, liposomal methotrexate was shown to produce greater epidermal deposition than the traditional topical formulation [6] while cyclosporin A co-loaded with calcipotriol in SLNs exhibited enhanced depth of skin penetration and improved anti-psoriatic effect [6]. Although the aforementioned systems hold significant promise they also present numerous complexities, costs and scaling issues that reinforce the continued value of optimized conventional formulations especially those utilizing botanicals that possess inherent anti-inflammatory properties [8, 26]. (Figure 8.)

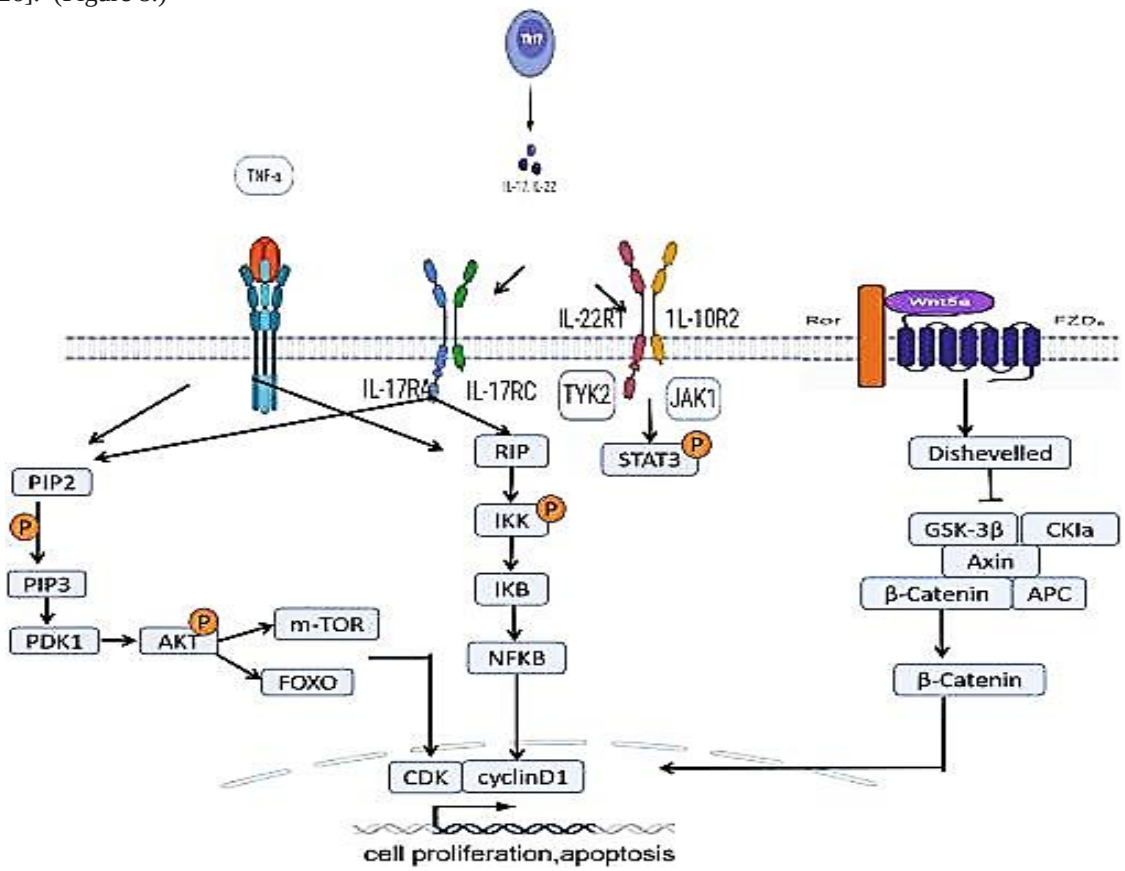

Figure 8 Cell proliferation.

III. PLANT PROFILE: CLITORIA TERNATEA L.

A. Taxonomic Classification and Geographical Distribution

The plant Clitoria ternatea L. (synonyms: Clitoria revoluta; Clitoria grandiflora), native to Tropical Asia, primarily India and Southeast Asia, has been naturalized across Africa, Australia and the Caribbean [12,13], and is a perennial climbing legume belonging to the family Leguminosae. The plant is referred to as "Aparajita" in India and is recorded in both classical Ayurvedic texts, such as Charaka Samhita and Sushruta Samhita, for its use as an ingredient for medicinal formulations for the treatment of the aforementioned ailments [13]. (Figure 9.)

Figure 9. Clitoria Terneteae Flower.

B. Chemical Constituents and Pharmacological Properties

The bioactive compounds found within C. ternatea have been shown to be primarily composed of anthocyanins. The primary anthocyanins found within C. ternatea are delphinidin 3, 3’ & 5’ triglycosides that are known as

"ternatin" (designated A1-A3, B1-B4, C1-C5 & D3). Ternatin compounds give C. ternata its distinct bright blue color. In addition to anthocyanins, other compounds present in this plant include; flavanoids (kaempferol, quercetin & myricetin); triterpenoids (β-amyrin); saponins; alkaloids (clitorine & trifolirhizin); glycosides; tannins; phenolic acids and protein compounds containing small cyclic peptides called cyclotides. Cyclotides have been demonstrated to exhibit antimicrobial and cytotoxic activity. The seeds of C. ternata also contain protease inhibitors and lectins, whereas the root system contains high amounts of the triterpene compounds taraxerol and taraxerone. Pharmacologically active components associated with C. ternata include: (i) antioxidant effects that result from free radical scavenger compounds including anthocyanins and flavonoids [10]; (ii) anti-inflammatory effects resulting from the inhibition of the COX-2 and 5-LOX pathways [14]; (iii) nootropic and anxiolytic effects resulting from cholinergic and GABAergic modulation [13]; (iv) antimicrobial and antifungal activity [12]; (v) skin protective and wound healing effects [14]. Collectively these bioactive compound properties provide a rationale for using C. ternata extracts in a topical formulation for treating psoriasis.

IV. MATERIALS AND METHODS

A. Collection and Authentication of Plant Material

The fresh flowers of Clitoria ternatea were gathered from a local botanical garden located in the state of Maharashtra, India. The species was confirmed to be Clitoria ternatea through authentication by a trained botanist and a voucher specimen was placed into the institutional herbarium. The fresh flower specimens were thoroughly rinsed with pure distilled water to eliminate any surface contaminants present on the outer surfaces of the flowers; dried in the shade for 7 days at a natural ambient temperature range of approximately 25 – 30° C; then, coarse-ground using an electric grinding device. The resultant ground-powder was sealed within tightly-closed amber-colored glass containers and maintained under storage conditions at room temperature (approximately 20-22° C) away from direct light and humidity until utilization.

B. Preparation of Hydroethanolic Extract

An alcohol-water (70% v/v) hydroextract was made in an ethanolic solution with a Soxhlet apparatus. The extractant was then put in a thimble that had been filled with 10 g of powdered plant material. Extraction was performed on the sample using 200 mL of this mixture at a temperature range of 60-65 °C for a period of time ranging from 6 – 8 hours. The liquid extract was then passed through Whatman # 1 filter paper; it was also dried by evaporation under vacuum conditions with a rotary evaporator at a temperature of 45 ° C. Freeze-drying resulted in production of the dry hydroextract. The percent yield was determined. The extract was quantitatively standardized for total anthocyanin and total flavonoids via spectrophotometry [10,11,12]. (Figure 10.)

Figure 10. Preparation of Extract by using Soxhlett Apparatus.

C. Formulation of the Herbal Lotion

The lotion made from herbs was produced via W/O emulsions. The formula for this product can be found in Table 1. Briefly put, the "water" component of the formula consisted of Xanthan Gum (1g), Acacia Gum (5g) & Rose Water that had been stirred continuously to keep it from forming lumps. Then Aloe Vera Gel (15mL) & C. ternatea Extract (5g) were added to it and blended well. The Oil Phase was prepared by first melting Natural Wax (8g) at approximately 65-70 degrees Celsius then blending Clove Oil (0.5mL) into the melted Natural Wax while gently mixing. The Oil Phase was then slowly added to the "Water" Phase while being mechanically agitated at about 60 degrees Celsius as long as a homogenized Emulsion was produced. Once this had occurred the mixture was

allowed to cool down to a temperature below 40 degrees Celsius & Turmeric Powder (1g) was introduced. Finally, the weight of the finished product was brought up to 100g using additional Rose Water.

Table 1. Composition of Clitoria ternatea Herbal Lotion Formulation.

Ingredient	Quantity	Role in Formulation	Mechanism of Action
Clitoria ternatea extract	5 g	Active anti-psoriatic agent	Antioxidant; anti-inflammatory via 5-LOX/COX-2 inhibition; reduces oxidative stress in skin [10,14]
Aloe vera gel	15 mL	Base / emollient / co-active	Anti-inflammatory; wound healing; skin hydration via acemannan and anthraquinones [15,23]
Clove oil (Syzygium aromaticum)	0.5 mL	Antimicrobial / analgesic / penetration enhancer	Eugenol inhibits NF-κB; mild analgesic (reduces pruritus); prevents secondary infection [20]
Turmeric powder (Curcuma longa)	1 g	Anti-inflammatory co-active	Curcumin inhibits NF-κB; reduces keratinocyte hyperproliferation [16]
Natural wax	8 g	Occlusive / emulsifier / consistency agent	Creates skin barrier; prevents transepidermal water loss; softens plaques [19]
Acacia gum	5 g	Stabiliser / film-forming agent	Forms protective film; retains moisture; reduces dryness and scaling [19,22]
Xanthan gum	1 g	Thickener / suspending agent	Improves viscosity and texture; ensures even application and prolonged skin contact [25]
Rose water (Rosa damascena)	q.s.	Solvent / moisturiser / soothing agent	Calms irritated skin; light hydration; mild antiseptic [20]

D. Evaluation Parameters

The prepared herbal lotion was evaluated for the following parameters following standard procedures [7,8,17,27]:

D.1 Organoleptic Properties

Colour, odor, texture, and physical state of the formulation were assessed by visual inspection and touch. The lotion was observed to be light yellow in colour with a mild aromatic odor, smooth non-greasy texture, and semi-solid consistency.

D.2 pH Determination

A 0.5 g quantity of the lotion was dispersed in 50 mL of distilled water and the pH was measured using a calibrated digital pH meter (Systronics model 335, India) at 25°C. Measurements were performed in triplicate and mean values reported. The target pH range was 5.0–6.5, consistent with normal skin surface pH [7,17]. (Figure 11)

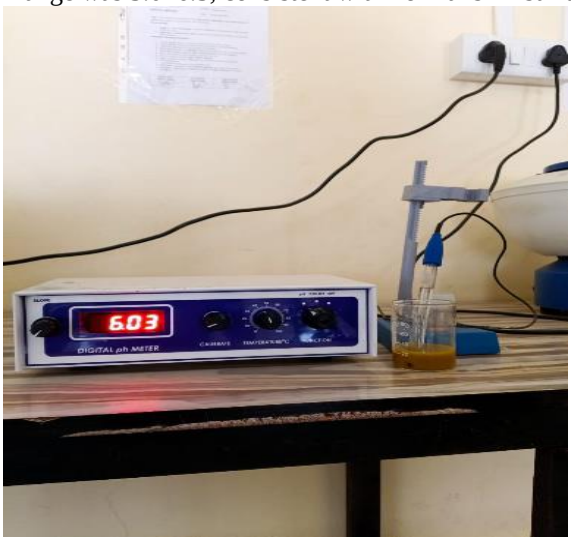

Figure 11. PH Testing on Digital PH Meter.

D.3 Homogeneity

A small quantity of lotion was placed on a clean glass slide, spread into a thin layer, and examined visually under adequate lighting for uniformity of texture, absence of lumps or coarse particles, and absence of phase separation [8,17].

D.4 Viscosity

Viscosity was determined using a Brookfield viscometer (Spindle No. 7) at 100 RPM and 25°C. Determinations were carried out in triplicate and the mean ± SD was recorded. Adequate viscosity is critical for spreadability and ensuring prolonged skin contact [7,27].

D.5 Spreadability

Spreadability was evaluated by the slip-and-drag method. Approximately 1 g of the formulation was placed between two glass slides; a known weight (500 g) was applied for 5 minutes. The upper slide was pulled by an attached weight (20 g) and the time required to traverse 7.5 cm was recorded. Spreadability (S) was calculated as $S = m \times l / t$, where m = applied mass (g), l = distance (cm), and t = time (s) [8,17,26]. (Figure 12.)

Figure 12. Spreadability Test.

D.6 Washability

The lotion was applied to the dorsum of the hand and exposed to running tap water for up to 10 minutes. The ease and time of complete removal were recorded [7].

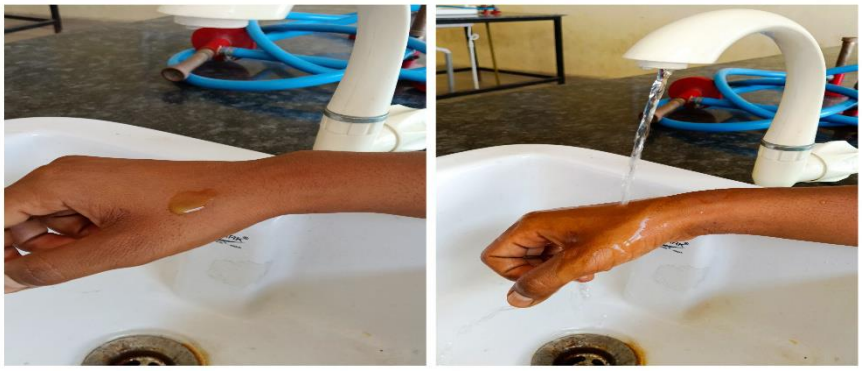

Figure 13. Washability Test.

D.7 Irritancy and Sensitivity Tests

An area (of) 2 cm² was located on the right-hand side (dorsal surface), which was designated with a marking substance as an application area for the lotion. The subject was then instructed to apply the lotion to the previously designated application area, and this was allowed to remain in place for 24 hours. The designated application area was checked periodically by the investigator for erythema, edema and/or any other irritation. In order to assess potential skin sensitization reactions from the product formulation, it was tested on the forearms of six otherwise healthy volunteers for 20 minutes. All adverse reactions were documented. (Figure 13.)

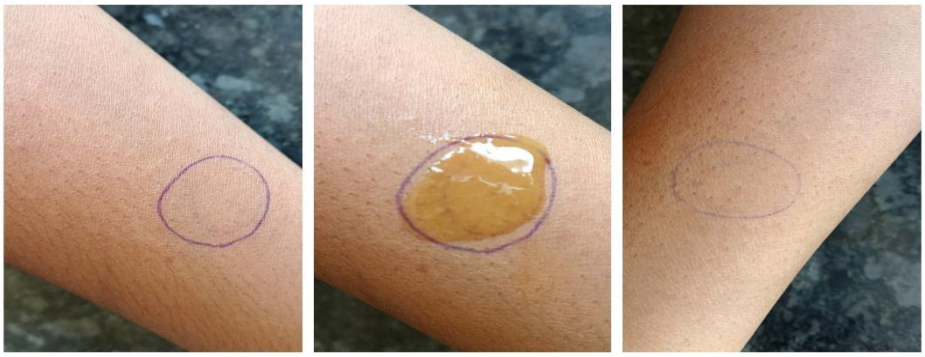

Figure 13. Irritancy and Sensitivity Test.

D.8 Antimicrobial Activity

Antimicrobial activity was determined through a Disc Diffusion Method (Kirby-Bauer Technique) on Malassezia furfur and Candida albicans; both are associated with secondary dermatological conditions seen in psoriasis patients. The microorganisms were seeded onto Sabouraud Dextrose Agar (fungi) and Mueller Hinton Agar (bacteria) at 37 degrees C for 24 to 48 hours. Filter Paper Disks (6 MM diameter) that had been saturated with the Herbal Lotion were placed atop the media. Zones of Inhibition (MM) were measured after incubation. (Figure 14.)

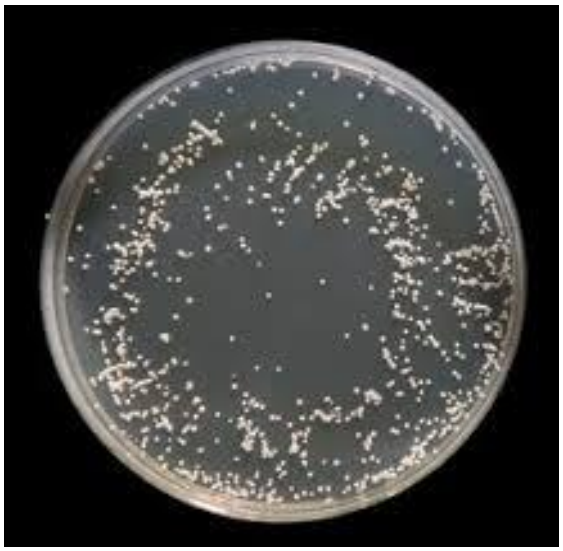

Figure 14 (a) Before Malassezia Bacteria.

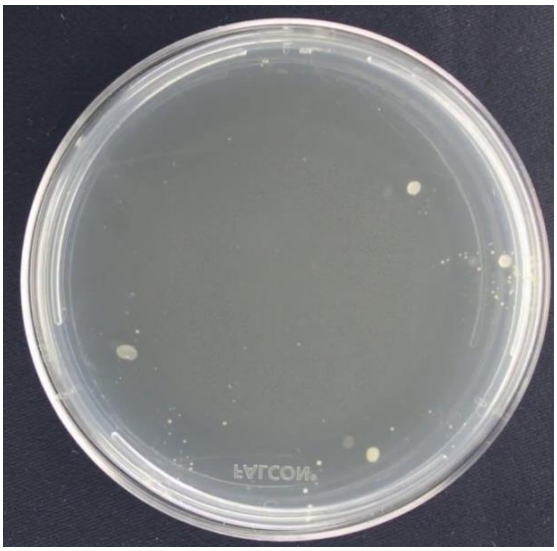

Figure 14(b) After Malassezia.

D.9 In Vitro Anti-Inflammatory Activity (Anti-Lipoxygenase Assay)

The anti-lipoxygenase activity of the C. ternatea extracts was evaluated using an adaptation of the procedure as reported by Nair et al. (2013)[14] . The reaction solution contained; linoleic acid (substrate) and varying amounts of C. ternatea (extract) (25 – 200µg/mL); soy bean lipoxygenase (enzyme); and phosphate buffer (pH 6.3). The absorbance was determined at 234nm utilizing a UV-Visible Spectrophotometer, with the percent inhibition calculated relative to the positive control (indomethacin) for each sample. The IC50 was then obtained from plots of percent inhibition against concentration.

D.10 In Vitro Anti-Proliferative Activity (HaCaT Cell Line, MTT Assay)

Proliferation of keratinocytes through treatment with C. ternatea extract was investigated using the HaCaT human keratinocyte cell line. Cell cultures were grown in Dulbecco's Modified Eagle Medium (DMEM) medium containing 10% fetal bovine serum (FBS). They were incubated at 37°C, with a 5% CO_2 atmosphere. For each experiment, cells were plated into 96-well culture plates (5 x 10^3 cells/well) and exposed to C. ternatea extract at concentrations ranging from 10 to 200 µg/mL for 48 h. Following this time period, cell viability was measured using an MTT (3-[4,5-DIMETHYLTHIAZOL-2-YL]-2,5-DIPHENYTETRAZOLIUM BROMIDE) assay, which measures the activity of mitochondrial dehydrogenases at wavelength 570 nm [4,14]. All results are expressed as percent reduction in proliferation compared to control (untreated) wells. In addition, the concentration of extract required to reduce keratinocyte proliferation by 50% (IC50 value) was calculated.

D.11 In Vivo Models (Planned Studies)

Future validations will include the use of the murine psoriasis model created with 5% Imiquimod (IMQ) cream applied to the back of BALB/C mice for five-six consecutive days. Symptoms associated with psoriasis are generated through application of this cream. Disease severity in these mice will be assessed via the PASI (Psoriasis Area and Severity Index) which scores erythema, scaling, and thickening from zero-four. Skin biopsies will also be used for both histopathology as well as quantifying cytokines (TNF-alpha, IL-17A, IL-23 via ELISA) [3,4]. Additionally, the mouse tail model will be used to evaluate orthokeratotic differentiation—i.e., restoration of the granular cell layer —as evidence of keratinocytes' normalisation of maturation [5,6]

V. RESULTS AND DISCUSSION

A. Physicochemical Evaluation

The prepared herbal lotion had excellent organoleptic qualities that include being pale yellow colored; having a very mild but pleasing aroma from the Clove and Rose Water added; possessing a smooth non greasy texture; and maintaining a homogeneous semi solid consistency. There were no visible signs of phase separation or particulate matter. The color, viscosity, spreadability, washability and homogenity are all comparable to those found in previously formulated herbal lotions using plant extracts as active ingredients and natural emulsifiers [7,8,17]. The pH of this herbal lotion ranged from 5.5-6.5 which falls into the acceptable physiologic range for a topical skin preparation (pH 4.5-6.5) and approximates the normal pH of the skin surface which averages 5.5 [7,17]. The

pH of a skin care product is extremely important because if the pH is too high it may compromise the skin's ability to resist microbial invasion by disrupting its natural antimicrobial barrier, lower the activity of the enzyme collagenase which is necessary for shedding of dead cells in the Stratum Corneum and increase irritation potential [2]. Upon visual and tactile inspection, the herbal lotion appeared to be well mixed with no apparent separation of phases. Values obtained during viscometry testing of this lotion utilizing a Brookfield Viscometer were typical for a pourable lotion and would make application over large psoriasis lesions easy [27]. Low spreadability values indicate the ability of the product to provide adequate coverage while requiring minimal effort to apply [8,26]. In addition to demonstrating good homogeneity the herbal lotion also demonstrated good washability after being subjected to running tap water for one minute, thereby exhibiting its expected non-greasy and water-washable properties which can contribute significantly to patient acceptance [7]. No erythema or edema nor other adverse cutaneous reactions occurred in either the irritancy test performed at 24 hours post treatment, or in the sensitization test administered to six volunteers who did experience neither an allergic reaction nor an irritant response as a result of their exposure to the product [8,17,20].

B. Antimicrobial Activity

The herbal lotion demonstrated zones of inhibition with respect to Malassezia furfur and Candida albicans as measured in the disc diffusion test. Malassezia species are lipophilic yeasts (lipids soluble) which grow on areas where there is a lot of sebaceous secretion (sebum) such as on the scalp or behind the ears. They can help stimulate inflammation (IL-17), increase symptoms of scalp and inverse psoriasis (scalp psoriasis occurs when it is located behind the ear), and disrupt the normal skin flora [1,2] . Therefore, antifungal properties exhibited by this product could be an important clinical tool. These antimicrobial properties have been attributed primarily to the action of eugenol contained within Cloves oils and Phenolics contained within C. ternatea and Rosewater, acting as cell membrane disrupters and enzymes inhibitors [20].

C. In Vitro Anti-Inflammatory Activity

The results indicate that the C. ternatea extract showed an increase in its ability to inhibit soybean lipoxygenase with increased concentrations. This enzyme is part of the 5-lipoxygenase (5-LOX) pathway which produces leukotrienes; these molecules are highly inflammatory and have been found at high levels in the psoriatic plaque [14]. As a result, it has been shown through both free radical quenching and the binding of the non-haem iron within the 5-LOX active site, that anthocyanin and flavanoid extracts present in the C. ternatea extract will be able to inhibit this enzyme, providing additional evidence supporting the anti-inflammatory potential of the product [10,14]. Additionally, curcumin (as turmeric powder) was also added to the mixture and can inhibit NF-kB (nuclear factor kappa B), a key transcription factor that regulates the expression of genes such as IL-6, IL-8, TNF-alpha and cyclins involved in keratinocyte proliferation [16].

D. Anti-Proliferative Activity (HaCaT MTT Assay)

Exposure of HaCaT keratinocytes to an increasing concentration range (10–200 μg/mL) of C. ternatea anthocyanin extract reduced keratinocyte viability/proliferation significantly and dose dependently when compared to untreated control cultures. Abnormal keratinocyte hyperproliferation due to the actions of the pro-inflammatory cytokines IL-17A and IL-22 as well as epidermal growth factor receptors (EGFR), is directly responsible for forming plaques that characterize psoriasis [3], [4]. Therefore, it has been demonstrated through the capacity of C. ternatea anthocyanins to decrease keratinocyte proliferation at concentrations attainable in clinical settings that there exists true therapeutic potential for topical use of this extract. Furthermore, these results support previous studies showing the anti-proliferative effect of polyphenolic rich plant extracts on HaCaT cells [16] , [21].

E. Overview of Marketed Formulations and Comparative Context

Current treatments available on the market for psoriasis have high risk of complications. Long term topical corticosteroid use (clobetasol propionate; e.g., Temovate(R) PharmaDerm) can lead to adrenal suppression and skin atrophy [5]. Vitamin D analogs (calcipotriene; e.g., Dovonex(R)) can lead to hypercalcaemia [5]. The systemic drugs used in treatment for psoriasis (methotrexate (Rheumatrex(R)), cyclosporine (Sandimmune(R)), acitretin (Soriatane(R))) all pose risks such as hepatotoxicity, nephrotoxicity, and teratogenicity, respectively [5,6]. The biologics (adalimumab, secukinumab, ustekinumab) that are currently being prescribed for psoriasis provide very good efficacy but are also associated with infection risks, and they are quite expensive. They may be administered via injection [5]. Therefore, when considering a safe alternative to these options, a well-studied natural/ herbal cream/lotion that is devoid of irritation and has shown anti-inflammation, anti-proliferative activity, antimicrobial action, and moisturizing effects represents a reasonable low-risk complementary option; especially for mild to moderate cases of the disease or as an adjuvant therapy to existing therapies [7,8,19,20,24].

VI. CONCLUSION

The new study was able to develop a unique lotion made from the extracts of both Aloevera and Clove Oil, as well as Acacia Gum, Xanthan Gum, Natural Wax and Rose Water along with Clitoria Ternatea Hydro-Ethanolic Extract which is being developed for Psoriasis treatment. The properties of the new product were tested and found to be safe and effective based on its pH level (5.5–6.5), Viscosity, Homogenity, Spreadability, Washability and Dermatological Safety. No irritation or sensitization occurred during testing by volunteers. The in vitro testing showed that this formulation will have an active effect against lipoxygenases and inhibit the growth of keratinocytes of HaCaT cells at varying concentrations. This demonstrates that Clitoria Ternatea can be used in

psoriasis treatments. There are multiple components within this formulation that create a synergy of action. The anthocyanins and flavonoids of Clitoria Ternatea will help to decrease inflammation and the amount of free radicals in the body. Curcumin will also decrease inflammation through the inhibition of NF-kB mediated cytokine production. Aloe Vera is known for its ability to provide anti-inflammatory benefits as well as to promote wound healing. Clove oil has been shown to prevent secondary infection and has been used to treat itching associated with psoriasis. The excipients (Acacia Gum, Xanthan Gum, Natural Wax) will help protect the skin barrier function while providing sustained release of the active ingredients. When compared to traditional synthetic products applied topically, this formulation presents a safer option for long term use. Future research could involve testing this formulation in mice using the imiquimod induced psoriasis model with PASI scoring and cytokine analysis. Additionally, stability studies (ICH guidelines) could be performed on this formulation to assess shelf life. Testing for how quickly each component is absorbed into the skin would also be beneficial. This could be done using Franz diffusion cells to test how easily the components pass through excised human skin. Finally, randomized control clinical trials could be conducted in patients with psoriasis to determine if this formulation is effective and safe for human use. Optimization of the concentration of Clitoria Ternatea extract and potential optimization of delivery via nanoparticles such as Nano-Lipid Carriers (NLCs) or Ethosomes could lead to improved efficacy.

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