

# Evaluation of Anti-Psoriatic Activity of Panchgavya Ghrita and Panchgavya Ghrita Cream in an Imiquimod-Induced Psoriasis-Like Mouse Model

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## ABSTRACT

**Background:** Psoriasis is a chronic immune-mediated inflammatory skin disorder characterised by excessive keratinocyte proliferation, oxidative stress, and dysregulated cytokine production. Although Panchgavya Ghrita, a traditional Ayurvedic formulation, has been reported to possess antioxidant and anti-inflammatory properties, its anti-psoriatic potential has not been scientifically evaluated.

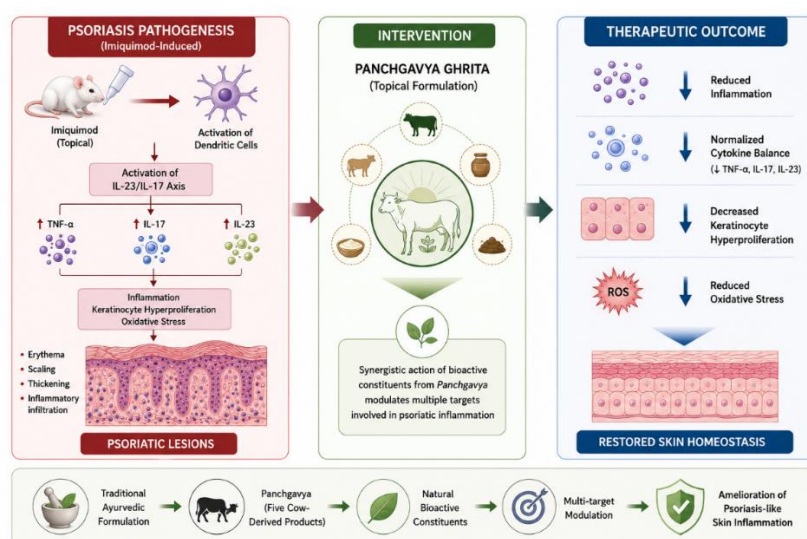
**Objective:** The present study aimed to develop a topical Panchgavya Ghrita cream and investigate the anti-psoriatic activity of Panchgavya Ghrita and its cream formulation in an imiquimod-induced psoriasis-like mouse model.

**Methods:** Panchgavya Ghrita cream was prepared using the fusion method and evaluated for physicochemical characteristics, including organoleptic properties, homogeneity, spreadability, pH, and potential for skin irritation. Antioxidant activity was assessed using the DPPH radical scavenging assay. Anti-psoriatic activity was evaluated in Swiss albino mice with imiquimod-induced psoriasis-like skin inflammation using Psoriasis Area and Severity Index (PASI) scoring, estimation of inflammatory cytokines (TNF- $\alpha$ , IL-17, and IL-23), and histopathological examination of skin tissue.

**Results:** The prepared cream exhibited satisfactory physicochemical characteristics, acceptable spreadability, skin-compatible pH, and no evidence of dermal irritation. Panchgavya Ghrita cream exhibited higher DPPH radical-scavenging activity than conventional Panchgavya Ghrita. Topical treatment significantly reduced erythema, scaling, and skin thickening in imiquimod-treated mice. The total PASI score decreased from  $2.43 \pm 0.11$  in the disease control group to  $0.50 \pm 0.07$  in the Panchgavya Ghrita cream-treated group. Treatment also significantly reduced tissue levels of TNF- $\alpha$ , IL-17, and IL-23 and improved histopathological features, including epidermal hyperplasia and inflammatory cell infiltration. The cream formulation produced greater therapeutic effects than conventional Panchgavya Ghrita.

**Conclusion:** Panchgavya Ghrita and Panchgavya Ghrita cream exhibited significant anti-psoriatic activity in an imiquimod-induced psoriasis-like mouse model. The observed effects may be attributed to their antioxidant and anti-inflammatory properties, as evidenced by enhanced radical-scavenging activity, suppression of pro-inflammatory cytokines, and improvement in histopathological alterations. Panchgavya Ghrita cream demonstrated superior efficacy and may serve as a promising topical therapeutic approach for psoriasis management.

## GRAPHICAL ABSTRACT



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**Keywords:** Panchgavya Ghrita; Panchgavya Ghrita cream; psoriasis; imiquimod; antioxidant activity; TNF- $\alpha$ ; IL-17; IL-23; PASI score.

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**Conflict of interest:** Nil.

## HIGHLIGHTS

- Panchgavya Ghrita cream exhibited significant anti-psoriatic activity in an imiquimod-induced mouse model.
- Treatment effectively reduced PASI scores, erythema, scaling, and skin thickening.
- Pro-inflammatory cytokines TNF- $\alpha$ , IL-17, and IL-23 were significantly downregulated.
- Histopathological abnormalities, including epidermal hyperplasia and inflammatory infiltration, were markedly improved.
- Panchgavya Ghrita cream demonstrated superior therapeutic efficacy compared with conventional Panchgavya Ghrita

## INTRODUCTION

Psoriasis is a common chronic inflammatory skin disease whose core pathological manifestations include excessive keratinocyte proliferation and abnormal epidermal proliferation and differentiation. The global prevalence of this disease currently ranges from 2% to 3%. In addition to typical cutaneous clinical manifestations such as scaly erythema and refractory pruritus, psoriasis is often accompanied by multiple comorbidities, including metabolic syndrome and cardiovascular disease, imposing a heavy physical, psychological, and economic burden on patients. Its pathogenesis stems from the interplay among genetic susceptibility, immune dysregulation, and environmental triggers (1,2).

Currently, the first-line preferred clinical treatment for mild-to-moderate psoriasis is topical therapy. Among the 7 commonly used conventional treatment modalities, widely adopted drugs include topical glucocorticoids and vitamin D analogues. However, existing therapies generally have clear limitations. Long-term topical use can easily trigger local adverse reactions such as skin atrophy; systemic medication may carry risks of systemic toxicity; and the disease is highly likely to relapse after treatment discontinuation, which fails to meet the clinical demand for long-term, safe disease control. Against this backdrop, natural traditional medicines that balance safety and pharmacological activity have become the core focus of research in alternative therapies. Among these, lipid-based topical preparations have also been shown to exert triple skin-repair effects: moisturising and retaining skin moisture, repairing the skin barrier, and exerting anti-inflammatory and soothing effects (3,4).

Panchgavya Ghrita, a classic formulation from Indian Ayurveda, uses ghee (ghrita) as its processing base, with core ingredients derived from cattle. Its traditional applications and prior studies have reported antioxidant,

anti-inflammatory, immunomodulatory, and wound-healing properties (7,8,9). However, to date, the anti-psoriasis activity of this formulation has not been scientifically verified in experimentally induced psoriasis models, leaving a distinct research gap in the field (5,6). Converting the traditional medicinal ghrita formulation into a topical cream can improve spreadability, ease of use, and patient acceptance while retaining the therapeutic activity of the original preparation. However, systematic evaluations of such modified formulations in psoriasis-like skin inflammation models remain insufficient (7,8).

Recent advances in psoriasis research have identified the interleukin (IL)-23/IL-17 axis as a key pathogenic pathway involved in disease progression. IL-23 promotes the differentiation and maintenance of T helper 17 (Th17) cells, which subsequently secrete IL-17 and other pro-inflammatory mediators that stimulate keratinocyte proliferation and amplify cutaneous inflammation. Tumour necrosis factor- $\alpha$  (TNF- $\alpha$ ) further synergises with IL-17 to sustain chronic inflammatory responses and epidermal hyperplasia. Consequently, therapeutic strategies that modulate these cytokines have emerged as effective approaches for psoriasis management. Therefore, evaluation of traditional Ayurvedic formulations against these inflammatory mediators may provide valuable insights into their anti-psoriatic potential (28,29).

In this study, an imiquimod-induced Swiss albino mouse model was used, and PASI scoring and skin histopathological assessment were applied to verify the anti-psoriasis activity of the original Panchgavya Ghrita preparation and its cream formulation.

## 2. MATERIALS AND METHODS

### 2.1 Materials

The core active ingredient of the topical preparation used in this study's experiments, Panchgavya Ghrita, was purchased from Go-Vigyan Anusandhan Kendra, Deolapar. The semi-solid cream base was formulated with beeswax

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and lanolin, and included the preservative benzoic acid, the stabiliser sodium phosphate, and peppermint water. For psoriasis model establishment, commercially available 5% imiquimod cream was used. All chemical reagents were of analytical grade and received no further purification before use (9,10).

## 2.2 Ethical Approval

All experimental procedures involving animals were approved by the Institutional Animal Ethics Committee (IAEC) of Smt. Kishoritai Bhoyar College of Pharmacy, Kamptee, Nagpur, under Approval No. 853/IAEC/2025-26/22. The study was conducted in accordance with the guidelines of the Committee for Control and Supervision of Experiments on Animals (CPCSEA), Government of India (11).

## 2.3 Characterisation and Standardisation of Panchgavya Ghrita

Before formally initiating formulation development, this study measured the acid value, iodine value, pH, and refractive index of the medicinal raw material, Panchgavya Ghrita, in accordance with standard analytical procedures to assess its quality and suitability for developing topical pharmaceutical preparations.

### 2.3.1 Physicochemical Evaluation of Panchgavya Ghrita

Panchgavya Ghrita underwent physicochemical evaluation to assess its quality and suitability for developing topical formulations. The formulation was analysed for acid value, saponification value, iodine value, peroxide value, specific gravity, pH, and refractive index using standard analytical procedures (31,32).

The acid value was determined by titration with a standard potassium hydroxide solution and expressed as the mg of KOH required to neutralise the free fatty acids present in 1 g of the sample. The saponification value was determined by refluxing the sample with alcoholic potassium hydroxide, followed by back-titration with standard hydrochloric acid. It was expressed as mg KOH required to saponify 1 g of the sample. The iodine value was estimated using Wij's method and expressed as grams of iodine absorbed per 100 g of sample. Peroxide value was determined by iodometric titration and expressed as milliequivalents of active oxygen per kilogram of sample. Specific gravity was determined at 25°C using a specific gravity bottle. The apparent pH of Panchgavya Ghrita was measured using a calibrated digital pH meter. Refractive index was determined at 25°C using an Abbe refractometer. All determinations were carried out in triplicate, and the mean values were recorded.

### 2.3.2 HPTLC Fingerprint Analysis

HPTLC fingerprinting of Panchgavya Ghrita was performed using a CAMAG HPTLC system. Samples were applied to pre-coated silica gel 60 F254 plates and developed in a twin-trough chamber using Toluene: Ethyl acetate: Formic acid (5:4:0.2, v/v/v) as the mobile phase. The developed plates were scanned at 254 nm and 366 nm using a CAMAG TLC Scanner, and the chromatographic profile was evaluated based on R<sub>f</sub> values and peak area percentages.

### 2.3.3 Gas Chromatographic Analysis of Fatty Acid Profile

The fatty acid composition of Panchgavya Ghrita was determined by gas chromatography following conversion of fatty acids to their corresponding methyl esters. Chromatographic separation was performed using a flame ionisation detector (FID), and individual fatty acids were identified by retention time and comparison with standard fatty acid methyl ester profiles. The relative abundance of each fatty acid was expressed as a percentage of area.

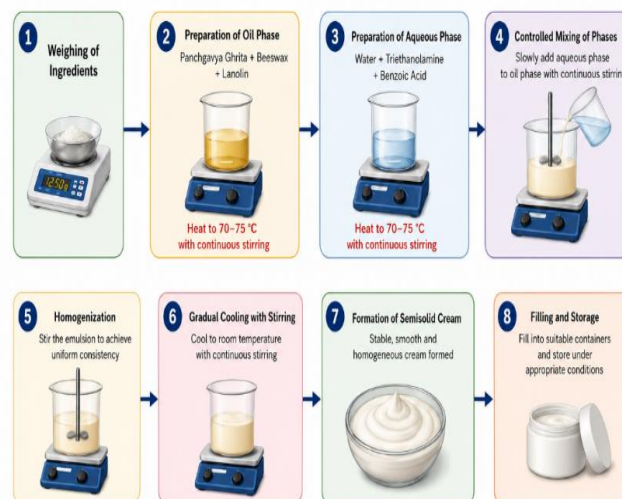
## 2.4 Preparation of Topical Panchgavya Ghrita Cream

The topical Panchgavya Ghrita cream prepared in this study was prepared using the fusion method. The oil phase, Panchgavya Ghrita (16.67% w/w), beeswax (10% w/w), and lanolin (6.67% w/w) and the aqueous phase, sodium phosphate (1% w/w) and benzoic acid (0.5% w/w), were dissolved in peppermint water and heated to the same temperature. Both phases were processed separately, stirred and homogenised at 70–75°C. The proportions of each ingredient are precisely documented, in accordance with pharmaceutical specifications, ensuring the experiment can be traced and reproduced.

**Table 1. Composition of Panchgavya Ghrita Cream**

Ingredient	Quantity (g)	% w/w
Panchgavya Ghrita	5.0	16.67
Beeswax	3.0	10.00
Lanolin	2.0	6.67
Benzoic acid	0.15	0.50
Sodium phosphate	0.30	1.00
Peppermint water	q.s. to 30 g	65.16

The aqueous phase was gradually added to the oily phase while stirring continuously to obtain a uniform emulsion. The resulting preparation was homogenised mechanically to improve consistency and uniformity. The formulation was allowed to cool gradually with continuous stirring until a smooth semisolid cream was formed. The prepared cream was transferred into a suitable airtight container and stored under standard conditions until further evaluation (12,13).



**Figure 1. Formulation of Panchgavya Ghrita Cream**

## 2.5 Physicochemical Evaluation

### 2.5.1 Organoleptic Evaluation

The prepared Panchgavya Ghrita cream was visually evaluated for colour, odour, texture, consistency, and the presence of phase separation (14).

### 2.5.2 Homogeneity

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Homogeneity of the formulation was assessed by visual inspection and tactile examination for lumps, grittiness, or phase separation (15).

### 2.5.3 Spreadability

Spreadability of the prepared cream was determined using the glass slide method. Approximately 1 g of cream was placed between two glass slides, and a known weight was applied to obtain a uniform film. The time required for the upper slide to move a specified distance was recorded (15). Spreadability was calculated using the following equation:

$$S = \frac{W \times L}{t}$$

Where:

S = Spreadability

W = Applied weight (g)

L = Distance moved by the slide (cm)

T = Time taken (s)

### 2.5.4 pH Determination

The apparent pH of the cream formulation was determined by dispersing 1 g of cream in distilled water and allowing the system to equilibrate before measurement. The pH electrode was immersed in the aqueous dispersion, and readings were recorded using a calibrated digital pH meter. All measurements were performed in triplicate and reported as mean  $\pm$  standard deviation (16,17,18).

## 2.6 In Vitro Antioxidant Activity

### 2.6.1 DPPH Radical Scavenging Assay

The 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay was performed to evaluate the antioxidant activity of Panchgavya Ghrita and Panchgavya Ghrita cream. Test preparations were prepared at various concentrations, mixed with DPPH solution, and incubated in the dark at room temperature for 30 minutes. The absorbance was measured at 517 nm using an ultraviolet-visible spectrophotometer, with ascorbic acid as the positive control, and the radical-scavenging rate was subsequently calculated using the specified formula (26,27).

$$\% \text{ Inhibition} = \frac{A_0 - A_1}{A_0} \times 100$$

Where:

$A_0$  = Absorbance of control

$A_1$  = Absorbance of sample

The antioxidant activity was expressed as a percentage inhibition of DPPH radicals.

### 2.7 Skin Irritation Study

The dermal irritation potential of the prepared Panchgavya Ghrita cream was evaluated using Swiss albino mice weighing approximately 25–30 g. Animals were maintained under standard laboratory conditions with free access to food and water.

The dorsal surface of each animal was shaved 24 h before topical application. Approximately 0.5 g of formulation was applied uniformly over the shaved skin area. The treated sites were observed at 24, 48, and 72 h for erythema, oedema, redness, or any visible signs of dermal irritation (17).

## 2.8 In Vivo Anti-Psoriatic Activity

### 2.8.1 Experimental Animals

Swiss albino mice weighing 25–30 g were used for the study. Animals were housed under standard laboratory conditions at  $25 \pm 2$  °C with a relative humidity of 60–70%

and a 12 h light/dark cycle. Animals were provided a standard pellet diet and water ad libitum and allowed to acclimatise before initiation of the study (11).

### 2.8.2 Induction of Psoriasis

Psoriasis-like skin inflammation was induced by the topical application of commercially available 5% imiquimod cream over the shaved dorsal region of mice once daily for 16 consecutive days. Imiquimod-induced skin inflammation is a well-established experimental model that closely resembles human plaque psoriasis in terms of erythema, scaling, epidermal hyperplasia, and inflammatory cell infiltration (18,19).

### 2.8.3 Experimental Design

Animals were randomly divided into four groups containing six animals in each group (n = 6):

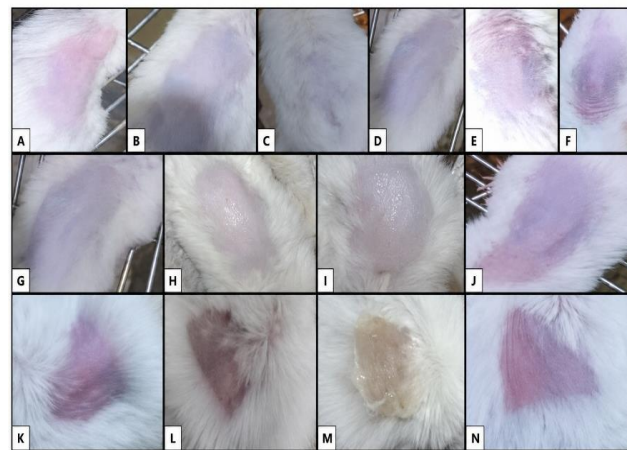
Group I: Normal control

Group II: Disease control (IMQ-induced)

Group III: Panchgavya Ghrita treatment group

Group IV: Panchgavya Ghrita cream treatment group

Following imiquimod induction, treatment groups received topical application of Panchgavya Ghrita or Panchgavya Ghrita cream once daily to the affected skin region for the duration of the study.



**Figure 2. Representative photographs of psoriasis-like skin lesions in experimental groups during the treatment period.**

(A–D) Normal control group showing intact skin architecture without erythema or scaling; (E–G) Disease control (IMQ-induced) group showing severe erythema, scaling, thickening, and psoriasis-like lesions; (H–J) Panchgavya Ghrita-treated group showing moderate reduction in erythema, scaling, and lesion severity; (K–N) Panchgavya Ghrita cream-treated group showing marked improvement in skin appearance with reduced scaling, inflammation, and restoration of near-normal skin texture following topical treatment.

### 2.8.4 PASI Scoring

Severity of psoriasis-like lesions was assessed using a modified Psoriasis Area Severity Index (PASI) scoring system based on erythema, scaling, and skin thickening. Each parameter was scored independently on a scale from 0 to 4, where 0 indicated the absence of symptoms and 4 indicated severe manifestation. The cumulative score was used to evaluate disease severity and therapeutic response (20).

### 2.8.5 Cytokine Estimation

At the end of the experimental period, skin tissue samples were collected to estimate inflammatory cytokines. Levels of tumour necrosis factor-alpha (TNF- $\alpha$ ), interleukin-17 (IL-17), and interleukin-23 (IL-23) were determined using commercially available ELISA kits according to the manufacturer's instructions. Cytokine concentrations were measured spectrophotometrically and expressed as pg/mL (28,29).

### 2.9 Histopathological Evaluation

At the end of the experimental period, animals were sacrificed, and skin tissue samples were excised from the dorsal region. The tissues were fixed in 10% neutral buffered formalin, dehydrated in ascending grades of alcohol, cleared in xylene, and embedded in paraffin wax. Sections of approximately 5  $\mu$ m thickness were prepared using a microtome and stained with haematoxylin and eosin (H&E). The stained sections were examined under a light microscope for epidermal hyperplasia, hyperkeratosis, inflammatory cell infiltration, vascular congestion, and restoration of normal skin architecture (21,22).

### 2.10 Statistical Analysis

All experimental data were expressed as mean  $\pm$  standard deviation (SD). Statistical analysis was performed using one-way analysis of variance (ANOVA) followed by Tukey's post hoc test using GraphPad Prism software. Differences were considered statistically significant at  $p < 0.05$ .

## 3. RESULTS

### 3.1 Characterisation and Standardisation of Panchgavya Ghrita

#### 3.1.1 Physicochemical Characterisation of Panchgavya Ghrita

Before formulation development, Panchgavya Ghrita was characterised for selected physicochemical parameters to assess its quality and suitability for topical formulation. The results are presented in Table 2.

**Table 2. Physicochemical Characteristics of Panchgavya Ghrita**

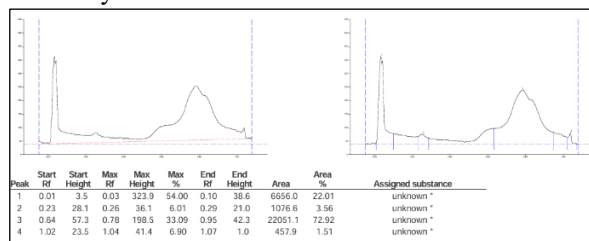
Parameter	Value
Acid Value (mg KOH/g)	3.51
Saponification Value (mg KOH/g)	210.13
Iodine Value (g I <sub>2</sub> /100 g)	32.20
Peroxide Value (meq O <sub>2</sub> /kg)	1.042
Specific Gravity (25°C)	0.9628
pH	5.2
Refractive Index (nD <sup>25</sup> )	1.44

The physicochemical characteristics of Panchgavya Ghrita indicated acceptable quality attributes for formulation development. The acid value suggested minimal hydrolytic degradation, while the iodine value reflected the presence of unsaturated fatty constituents. The refractive index was consistent with the lipidic nature of the formulation. The apparent pH was within a range suitable for topical application.

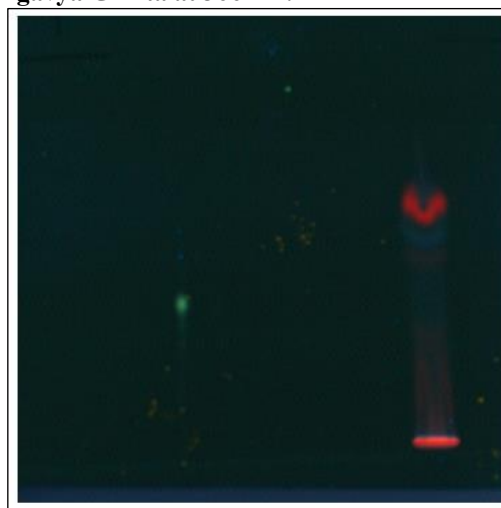
#### 3.1.2 HPTLC Fingerprint Profile

HPTLC fingerprint analysis of Panchgavya Ghrita revealed a characteristic chromatographic profile with multiple resolved peaks, indicating the presence of diverse phytochemical and lipid-derived constituents. At 366 nm,

four major peaks were detected with Rf values of 0.03, 0.26, 0.78, and 1.04. The predominant peak was observed at Rf 0.78, accounting for 72.92% of the total peak area, suggesting the presence of a major constituent in the formulation. Additional peaks with area percentages of 22.01%, 3.56%, and 1.51% were also observed, indicating the complex chemical composition of Panchgavya Ghrita. The obtained chromatographic fingerprint may serve as a reference profile for quality control and batch-to-batch consistency of the formulation.



**Figure 3A. HPTLC fingerprint chromatogram of Panchgavya Ghrita at 366 nm.**



**Figure 3B. Developed HPTLC plate of Panchgavya Ghrita visualised under UV light (366 nm)**

**Table 3. Major HPTLC Peaks of Panchgavya Ghrita**

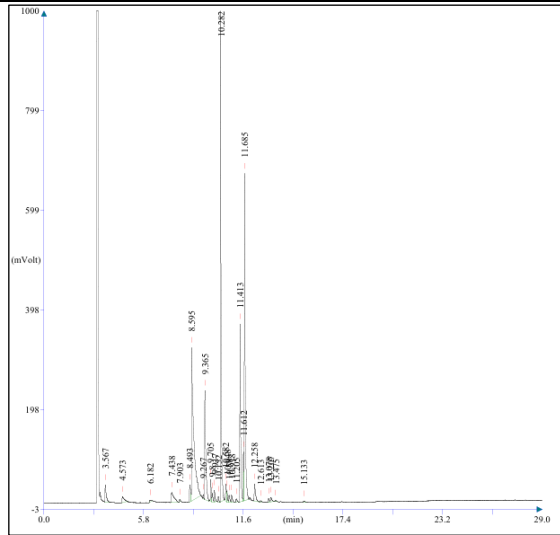
Peak	Rf Value	Area %
1	0.03	22.01
2	0.26	3.56
3	0.78	72.92
4	1.04	1.51

#### 3.1.3 Fatty Acid Composition of Panchgavya Ghrita

Gas chromatographic analysis demonstrated that Panchgavya Ghrita contained a diverse range of saturated and unsaturated fatty acids. The predominant fatty acids identified were palmitic acid (23.8%), myristic acid (23.4%), oleic acid (19.2%), and stearic acid (9.2%). Moderate amounts of myristoleic acid (8.0%) and elaidic acid (2.4%) were also detected. The formulation contained 63.27 g/100 g saturated fatty acids, 29.69 g/100 g monounsaturated fatty acids, and 1.8 g/100 g polyunsaturated fatty acids. The presence of oleic acid and other unsaturated fatty acids may contribute to skin barrier maintenance and anti-inflammatory effects. In contrast, the lipid-rich composition may enhance emollient and moisturising properties relevant to psoriasis management.

**Table 4. Major Fatty Acids Identified in Panchgavya Ghrita**

Fatty Acid	Composition (%)
Palmitic Acid (16:0)	23.8
Myristic Acid (14:0)	23.4
Oleic Acid (18:1)	19.2
Stearic Acid (18:0)	9.2
Myristoleic Acid (14:1)	8.0
Elaidic Acid (18:1 trans)	2.4
Linoleic Acid (18:2)	1.5



**Figure 4. Gas chromatogram of Panchgavya Ghrita showing fatty acid composition.**

**3.2 Physicochemical Evaluation of Panchgavya Ghrita Cream**

The prepared Panchgavya Ghrita cream exhibited satisfactory organoleptic characteristics, including a smooth, semisolid consistency, characteristic odour, and uniform appearance. The formulation showed good homogeneity, with no visible signs of grittiness or phase separation, indicating acceptable physical stability of the cream base. The pH of the formulation was found to be within the physiological skin range, suggesting suitability for topical application and reduced possibility of skin irritation. The prepared cream also demonstrated acceptable spreadability, facilitating uniform application over the affected skin surface.

**Table 5. Physicochemical Evaluation of Panchgavya Ghrita Cream**

Parameter	Observation
Color	Greenish-yellow
Odor	Characteristic
Texture	Smooth
Homogeneity	Good
Phase separation	Absent
pH	6.53 ± 0.01
Spreadability	6.91 ± 0.17 g·cm/s

The favourable physicochemical characteristics observed in the present study may be attributed to the lipid-rich composition of the formulation and proper incorporation of excipients, which contributed to improved consistency and topical applicability. Similar observations regarding semisolid topical preparations have been reported

previously in topical herbal formulations and emulsion-based cream systems (23,24).

**3.3 In Vitro Antioxidant Activity**

Panchgavya Ghrita and its cream formulation both displayed concentration-dependent DPPH radical scavenging activity, and the cream's antioxidant activity outperformed that of the traditional pure preparation. Existing research has confirmed that oxidative stress amplifies inflammatory responses by increasing reactive oxygen species production, thereby contributing to tissue damage in psoriasis (30). Based on this well-established pathological mechanism, this category of preparations may have therapeutic potential for psoriasis. The cream's superior activity stems from the improved dispersion and accessibility of active ingredients in its semi-solid formulation, and this in vitro observation aligns with the anti-psoriatic effect observed in the in vivo experiments of this study.

**Table 6. DPPH Radical Scavenging Activity of Panchgavya Formulations**

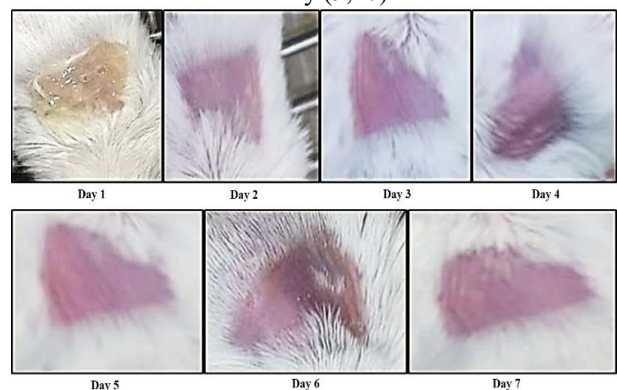
Concentration (µg/mL)	Panchgavya Ghrita (% Inhibition)	Panchgavya Ghrita Cream (% Inhibition)	Ascorbic Acid (% Inhibition)
20	28.4 ± 1.2	34.7 ± 1.5	48.2 ± 1.1
40	39.6 ± 1.4	46.8 ± 1.3	61.5 ± 1.4
60	51.2 ± 1.6	58.9 ± 1.7	73.8 ± 1.2
80	63.7 ± 1.5	71.4 ± 1.6	84.6 ± 1.5
100	74.5 ± 1.8	82.3 ± 1.4	92.7 ± 1.3

Values are expressed as mean ± SD (n = 3)

**3.4 Skin Irritation Study**

The dermal irritation potential of Panchgavya Ghrita cream was evaluated in Swiss albino mice following topical application over the shaved dorsal skin region. Throughout the observation period, no visible signs of erythema, oedema, redness, or inflammation were observed in the treated animals. The irritation score remained negligible throughout the study, indicating good dermal compatibility and non-irritant properties of the formulation.

The absence of dermal irritation may be attributed to the emollient and moisturising properties of the lipid-based formulation, which may help maintain skin hydration and barrier integrity. Similar findings have been reported for traditional ghee-based topical preparations exhibiting favourable dermal tolerability (9,17).



**Figure 5. Skin Irritation Study in Swiss Albino Mice**

**3.5 Anti-Psoriatic Activity**

**3.5.1 PASI Score Evaluation**

Topical application of imiquimod successfully induced psoriasis-like skin lesions characterised by erythema, scaling, and increased skin thickness in the disease control group. Progressive worsening of psoriatic symptoms was observed in untreated animals throughout the experimental period.

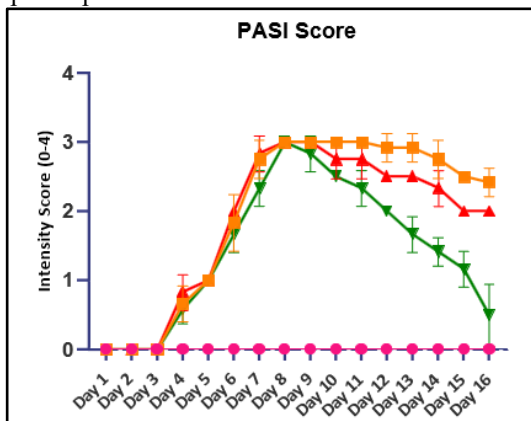
In contrast, animals treated with Panchgavya Ghrita and Panchgavya Ghrita cream demonstrated noticeable improvement in psoriasis-like symptoms compared with the disease control group. Reduction in erythema, scaling, and skin thickening was observed following topical treatment. The Panchgavya Ghrita cream-treated group exhibited comparatively greater reduction in PASI scores than the Panchgavya Ghrita treatment group, indicating improved therapeutic response.

**Table 7. PASI Score Evaluation**

Group	Erythem a	Scalin g	Thicknes s	Total PASI Scor e
Normal Control	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
Disease Control	0.85 ± 0.08	0.80 ± 0.07	0.78 ± 0.09	2.43 ± 0.11
Panchgavy a Ghrita Treatment	0.68 ± 0.06	0.65 ± 0.05	0.67 ± 0.08	2.00 ± 0.10
Panchgavy a Ghrita Cream Treatment	0.18 ± 0.04	0.15 ± 0.03	0.17 ± 0.05	0.50 ± 0.07

Values are expressed as mean ± SD (n = 6).

The enhanced therapeutic response observed with the cream formulation may be associated with improved spreadability and uniform topical application, which could facilitate better retention of the formulation over the affected skin surface. The findings suggest that conversion of traditional Panchgavya Ghrita into a semisolid cream formulation may improve topical applicability while preserving its therapeutic potential.



**Figure 6. Effect of Panchgavya Ghrita Formulations on PASI Score**

The figure shows the progression of psoriasis-like skin inflammation and the therapeutic response following topical treatment in Swiss albino mice over 16 days. Severity of psoriasis-like lesions was assessed using the modified Psoriasis Area Severity Index (PASI) scoring based on erythema, scaling, and skin thickening. Pink line (●) represents the Normal Control group, which showed no visible signs of psoriasis-like lesions throughout the study period and maintained a PASI score of 0. Orange line (■) represents the Disease Control (IMQ-induced) group, which exhibited a progressive increase in PASI score following imiquimod application, indicating severe psoriasis-like skin inflammation with persistent erythema, scaling, and thickening. The red line (▲) represents the Panchgavya Ghrita treatment group, which demonstrated a moderate reduction in PASI score compared with the disease control group, indicating attenuation of psoriasis-like symptoms following topical treatment. The green line (▼) represents the Panchgavya Ghrita cream treatment group, which showed a marked reduction in PASI score during the treatment period, suggesting comparatively improved anti-psoriatic activity and restoration of skin condition.

Values are expressed as mean ± SD (n = 6). A reduction in PASI score indicates improvement in psoriasis-like skin lesions following topical treatment. The observed reduction in PASI scores indicates significant attenuation of psoriasis-like inflammation following treatment with Panchgavya Ghrita formulations. Similar reductions in erythema, scaling, and epidermal thickening have been reported in previous investigations involving herbal and lipid-based anti-psoriatic topical preparations (20,25).

**3.5.2 Cytokine Estimation**

Disease-control animals showed significantly elevated levels of TNF-α, IL-17, and IL-23 compared with the normal control group. Treatment with Panchgavya Ghrita and Panchgavya Ghrita cream significantly reduced cytokine levels compared with the disease control group (p < 0.05). The Panchgavya Ghrita cream-treated group demonstrated comparatively greater reduction in inflammatory cytokines.

**Table 8. Effect of Panchgavya Formulations on Inflammatory Cytokine Levels**

Group	TNF-α (pg/mL)	IL-17 (pg/mL)	IL-23 (pg/mL)
Normal Control	18.6 ± 1.2	15.4 ± 1.1	12.8 ± 0.9
Disease Control	72.5 ± 2.8	68.7 ± 2.4	64.9 ± 2.1
Panchgavya Ghrita Treatment	46.8 ± 2.1	42.5 ± 1.9	39.7 ± 1.8
Panchgavya Ghrita Cream Treatment	28.9 ± 1.7	25.6 ± 1.5	22.8 ± 1.3

Values are expressed as mean ± SD (n = 6)

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The elevated levels of TNF- $\alpha$ , IL-17, and IL-23 observed in the disease control group confirm activation of inflammatory pathways associated with psoriasis-like skin inflammation. Treatment with Panchgavya Ghrita and Panchgavya Ghrita cream significantly reduced the levels of these cytokines, indicating suppression of key mediators involved in disease pathogenesis. The comparatively greater reduction observed in the Panchgavya Ghrita cream-treated group suggests enhanced anti-inflammatory efficacy and improved topical delivery of active constituents.

The reduction in pro-inflammatory cytokines observed following treatment suggests attenuation of inflammatory pathways associated with psoriasis-like skin inflammation. The findings correlated well with reductions in PASI scores and histopathological improvements observed in treated groups (28,29).

### 3.5.3 Histopathological Evaluation

Histopathological examination of skin sections from the normal control group revealed normal epidermal and dermal architecture, intact skin layers, and no inflammatory infiltrates. In contrast, the disease control group exhibited characteristic psoriatic alterations, including epidermal hyperplasia, hyperkeratosis, elongated rete ridges, inflammatory cell infiltration, and vascular congestion.

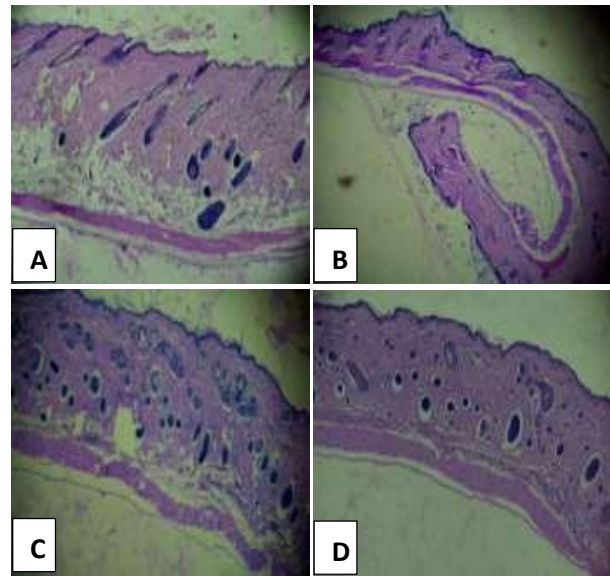
**Table 9. Histopathological Findings**

Group	Hyperplasia	Hyperkeratosis	Inflammation	Skin Architecture
Normal Control	Absent	Absent	Absent	Normal epidermal and dermal architecture
Disease Control	Severe	Severe	Severe inflammatory infiltration	Markedly disrupted skin architecture
PGG Treatment	Moderate	Moderate	Moderate inflammatory infiltration	Partial restoration of skin architecture
PGG Cream Treatment	Mild	Mild	Mild inflammatory infiltration	Near-normal restoration of skin architecture

Histopathological examination revealed marked pathological alterations in disease-control animals, including epidermal hyperplasia, hyperkeratosis, inflammatory cell infiltration, and disruption of normal skin architecture. These findings are characteristic features of psoriasis-like skin inflammation induced by imiquimod. Treatment with Panchgavya Ghrita and Panchgavya Ghrita cream substantially reduced these pathological abnormalities. The Panchgavya Ghrita cream-treated group exhibited near-normal epidermal architecture with minimal inflammatory infiltration, indicating significant restoration of skin morphology. These histopathological improvements closely correlated with reductions in PASI scores and

inflammatory cytokine levels, further supporting the anti-inflammatory activity of the formulation (18,20).

**Figure 7. Histopathological evaluation of skin tissue**



sections stained with haematoxylin and eosin (H&E). (A) Normal control showing intact epidermal and dermal architecture; (B) Disease control showing epidermal hyperplasia, hyperkeratosis, inflammatory infiltration, and disrupted skin architecture; (C) Panchgavya Ghrita-treated group showing partial restoration of skin morphology; (D) Panchgavya Ghrita cream-treated group showing near-normal epidermal structure and reduced inflammatory infiltration.

The histopathological findings correlated well with reductions in PASI scores and confirmed the anti-psoriatic activity of Panchgavya Ghrita formulations. Restoration of epidermal organisation and reduction of inflammatory alterations suggest attenuation of psoriasis-like skin inflammation following topical treatment. Comparable histopathological improvements have been reported previously in experimental studies involving anti-inflammatory and herbal topical formulations (20,21).

## 4. DISCUSSION

Psoriasis is a chronic immune-mediated inflammatory disorder characterised by excessive keratinocyte proliferation, epidermal hyperplasia, and dysregulated cytokine production. Among the various pathogenic pathways involved, the IL-23/Th17 axis has emerged as a central driver of disease progression. IL-23 promotes differentiation and survival of Th17 lymphocytes, which subsequently produce IL-17 and related cytokines that stimulate keratinocyte activation, inflammatory cell recruitment, and amplification of local immune responses. TNF- $\alpha$  acts synergistically with IL-17, contributing to chronic inflammation and tissue damage. Consequently, suppression of these cytokines has become a major therapeutic target in contemporary psoriasis management (28,29).

The significant reduction in TNF- $\alpha$ , IL-17, and IL-23 levels observed following treatment suggests that Panchgavya

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Ghrita formulations may modulate the IL-23/Th17 inflammatory pathway. Since IL-17 is a major effector cytokine responsible for keratinocyte hyperproliferation and epidermal thickening, suppression of this cytokine may explain the observed reductions in PASI scores and histopathological abnormalities. Similarly, the reduction of TNF- $\alpha$  may contribute to the attenuation of inflammatory cell infiltration and the restoration of normal skin architecture. These findings support the proposed anti-inflammatory mechanism underlying the anti-psoriatic activity of Panchgavya Ghrita formulations (18,28,29).

Oxidative stress is recognised as an important contributor to the pathogenesis of psoriasis. Increased production of reactive oxygen species can promote lipid peroxidation, cellular injury, inflammatory cytokine release, and abnormal keratinocyte proliferation. The concentration-dependent DPPH radical scavenging activity observed in the present study indicates the significant antioxidant potential of both Panchgavya Ghrita and Panchgavya Ghrita cream. The superior antioxidant activity demonstrated by the cream formulation may contribute to suppression of oxidative stress-mediated inflammatory responses and may partly explain its enhanced anti-psoriatic efficacy (26,27,30).

This study used Swiss albino mice as experimental subjects to conduct a series of *in vivo* and *in vitro* experiments. The test substances were the traditional preparation, Panchgavya Ghrita, and the cream formulation prepared from it. The detection metrics included body-surface PASI scores, histopathological staining of skin lesion tissues, DPPH free-radical scavenging antioxidant assays, and quantification of the core inflammatory cytokines TNF- $\alpha$ , IL-17, and IL-23. Results from all detection metrics showed that the cream formulation had superior activity to the original preparation, with all indicators mutually validating one another. In light of the confirmed pathogenic mechanisms of psoriasis, the findings suggest that the anti-psoriatic activity of Panchgavya Ghrita cream may involve modulating inflammatory pathways and attenuating oxidative stress, thereby contributing to the improvement of psoriasis-like skin lesions. It can regulate the abnormal activation of the core inflammatory axis, alleviate oxidative stress damage, inhibit excessive keratinocyte proliferation, and comprehensively intervene in the pathological progression of psoriasis.

The HPTLC fingerprint profile demonstrated the chemical complexity of Panchgavya Ghrita and provides a useful tool for future quality control and standardisation of the formulation. Furthermore, gas chromatographic analysis confirmed the presence of several biologically relevant fatty acids, including oleic acid, palmitic acid, stearic acid, and myristic acid. Oleic acid has been reported to enhance skin hydration and improve topical permeation, while the formulation's overall lipid-rich composition may help restore skin barrier function and reduce inflammation (6,23). These characteristics may partially explain the observed anti-psoriatic activity of Panchgavya Ghrita and its cream formulation.

The traditional medicinal preparation Panchgavya Ghrita can regulate oxidative and inflammatory mediators to

improve core pathological changes in psoriasis-like skin lesions, including excessive keratinocyte proliferation, epidermal thickening, and inflammatory infiltration. This study modified the traditional formulation into a cream dosage form and evaluated its anti-psoriasis activity, application potential, and limitations in an experimentally induced psoriasis-like skin inflammation model. The study found that the modified Panchgavya Ghrita cream achieved better treatment responses than the traditional dosage form. This advantage stems from the cream's superior spreadability and suitability for external use, which can increase drug retention and coating uniformity at lesion sites. This confirms that converting traditional oil-based ointments into semi-solid creams can improve patient acceptance and medication convenience without compromising the original therapeutic efficacy. Such lipid-rich formulations can also repair the impaired skin barrier in psoriasis, as supported by the findings of reference (6) and prior research on similar lipid-based topical preparations.

The histopathological assessment results of this study further reinforce the observed therapeutic efficacy: the disease control group exhibited typical psoriasis-related pathological changes, including acanthosis, hyperkeratosis, inflammatory infiltrates, and vascular congestion, whereas the treatment groups showed largely restored normal skin structure. This study also has clear limitations: only three inflammatory factors, TNF- $\alpha$ , IL-17, and IL-23, were measured, and no in-depth mechanistic studies were conducted on oxidative stress biomarkers, gene expression, immunohistochemistry, or molecular signalling pathways. Additional long-term safety and clinical studies are required to confirm the clinical value of this formulation.

### 5. CONCLUSION

The present study demonstrated significant anti-psoriatic activity of Panchgavya Ghrita and Panchgavya Ghrita cream in an imiquimod-induced psoriasis-like mouse model. Both formulations effectively reduced PASI scores, suppressed pro-inflammatory cytokines (TNF- $\alpha$ , IL-17, and IL-23), and improved histopathological alterations associated with psoriasis. The superior efficacy of Panchgavya Ghrita cream may be attributed to improved topical applicability and enhanced local delivery of active constituents. The findings suggest that modulation of inflammatory pathways and antioxidant activity contributes to the observed therapeutic effects. Panchgavya Ghrita cream may therefore represent a promising topical therapeutic approach for the management of psoriasis. Further mechanistic investigations and clinical studies are warranted to establish its long-term safety and clinical efficacy.

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### CONFLICT OF INTEREST

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“The authors declare that there is no conflict of interest regarding the publication of this manuscript.”

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