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IN VIVO EVALUATION OF *CHAKRAVHAYADI LEPA* FOR *KITIBHA KUSHTHA*: INSIGHTS FROM IMIQUIMOD-INDUCED PSORIASIS IN ALBINO MICE

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ABSTRACT

Introduction: Skin disorders are increasing due to lifestyle, dietary, and environmental factors, with psoriasis affecting 2–3% of the population. In *Ayurveda*, *Kitibha Kushta*, a *Vata-Kapha* dominant *Kshudra Kushta*, presents with hyperpigmentation, scaling, hardness, and severe itching, closely resembling psoriasis. *Chakravhayadi Lepa*, described in *Chakradatta*, was selected for local application in albino mice, offering a potential safe and effective approach for psoriasis management. **Objective:** To determine the efficacy of *Chakravhayadi lepa* in imiquimod induced Psoriasis in albino mice. **Material and Methods:** An in vivo study was conducted on albino mice following repeated administration. 14 days local application was done as per OECD guidelines. Assessment according to PASI score (erythema, scaling and thickness), inflammatory markers such as interleukin-6 (IL-6) and umornecrosis factor-alpha (TNF-α), histopathological investigation were carried out to evaluate efficacy of formulation. **Results**: IMQ induced stable body weight but caused erythema, scaling, thickening, increased spleen weight, and elevated **IL-6** and **TNF-α**. *Chakravhayadi Lepa* significantly ($P < 0.001$) reduced skin lesions, normalized spleen

weight, and lowered cytokine levels, with maximal effects from day 9. Histopathology showed minimal skin changes in the treated group, and spleen tissue remained normal, indicating no systemic toxicity. **Conclusion**: In view of Observations and on the basis of results obtained and statistical analysis we concluded that, 'Treatment with *Chakravhyadi Lepa* for seven days in *Kitibha Kushta* significantly reduced psoriasis-like symptoms in mice.'

KEYWORDS: *Kitibha Kushtha*, Psoriasis, *Chakravhyadi Lepa.*

INTRODUCTION

Skin, the largest organ of the body, serves as the first line of defense against physical, chemical, and biological agents, protecting against microbial invasion, mechanical injury, and environmental damage. It also reflects internal health, as many systemic disorders manifest through changes in skin texture, color, and integrity. Modern lifestyle factors—including sedentary habits, stress, unhealthy diet, disrupted sleep, environmental changes, and frequent exposure to chemicals—have led to a rising prevalence of skin disorders, causing physical, emotional, and economic burdens such as low self-esteem, anxiety, and social stigma.

Psoriasis is a chronic inflammatory skin disorder affecting 2–3% of the global population, characterized by erythema, scaling, thickening, and severe itching. In Ayurveda, *Kitibha Kushta*, classified under *Kshudra Kushta*[1], closely resembles psoriasis[2] and is predominantly *Vata-Kapha* in origin. Classical features include *Shyavam (*hyperpigmentation), *Kinkharsparsha (*roughness), *Paru*sha (hardening of lesions)[3], and, in chronic cases, darkened plaques and abnormal thickening. The condition arises due to factors such as incompatible diet, unhealthy daily and seasonal routines, and repeated exposure to chemicals or drugs, which vitiate *Rakta* and other *Dhatus*, leading to the manifestation of *Kitibha Kushta*.[4]

Ayurvedic texts describe numerous *Bahya Sanshaman Lepas*[5] (external applications) for managing skin disorders. Among these, *Chakravhayadi Lepa*[6], as cited in *Chakradatta*, was selected for evaluation in albino mice. Its formulation contains ingredients with anti-inflammatory, wound-healing, and immunomodulatory properties, making it a promising, safe, and accessible option for the management of psoriasis. Scientific validation of this *lepa* could represent a paradigm shift in treating psoriasis while aligning modern research with traditional Ayurvedic principles.

MATERIAL AND METHOD

The study was carried out in three steps i.e, collection of *Snuhi Ksheer* and preparation of *Chakramarda churna* and *Snuhi Ksheer Bhavit Chakramarda churna*. Secondly, to assess physiochemical analysis of *Snuhi Ksheer*, *Gomutra, Chakramarda churna* and *Snuhi Ksheer Bhavit Chakramarda churna*. Lastly, animal experimental study.

Analytical tests were done for *Snuhi Ksheer, Gomutra, Chakramarda churna* and *Snuhi Ksheer Bhavit Chakramarda churna* were as follows**:**

a. Determination of foreign matter.

b. Determination of pH values.

c. Moisture content

d. Determination of Total Ash.

e. Determination of Acid-Insoluble Ash.

f. Determination of Alcohol insoluble extract.

g. Determination of Water-soluble extract.

Preparation of *Snuhi Ksheera Bhavit Chakramard Churna*

Chakramarda seeds were washed, shade-dried, and ground into a fine powder, sieved repeatedly through sieve no. 85, and stored in an airtight container. *Snuhi Ksheera* was then used for *Bhavana* of the *churna* in a *Khalwa Yantra*, followed by drying in sunlight and a hot air oven. The final *Snuhi Ksheera Bhavit Chakramarda Churna* was stored in an airtight container and used with *Gomutra* for the animal study.

Ethics Approval

The study was initiated after the approval of protocol by Institutional Animal Ethics Committee (SCI/IAEC/2025-26/170).

MATERIALS

Chemicals used- isoflurane.

Interventions- *Gomutra, Snuhi Ksheera Bhavit Chakramarda Churna,* Imiquimod cream**.**

Apparatus used

Weighing machine, surgical gloves, depilatory cream, Autoclaved corn cob, non-adhesive tape, measuring cylinder, Spatula, Sterile bud, Dissecting microscope, Surgical instruments.

Experimental animals and sample size

Strain: Swiss Albino.

Animal species used	Mus musculus
Place of experiment	Research Center and animal shelter as per feasibility
Source of animal	Government Authorized Animal House
Sample Size	18
Group Size	Each Group 6 Albino mice.
Sex of animal to be used (Albino mice)	50% males and 50 % females in each group will be taken
No. of groups	3
Route of drug administration	Local Application
Duration of Study	14 days

Selection of animals

- A total of 18 animals, healthy Swiss Albino mice 8–10 weeks Old of either sex were used in this study.
- Swiss Albino mice was housed at standard environmental conditions.
- Body weight ranging from 20-30gm.

Acclimatization period

Acclimatization was done 7 days prior to the experimental study.

Diet: Standard laboratory Diet

Animals were marked with permanent marker for proper identification on their tail.

Housing and Feeding of Animals: Animals were housed in polypropylene cages. Bedding Materials-Autoclaved corn cob. Room temperature was maintained to 20 to 23ºC and relative humidity of 30-70%. Lighting was artificial with 12 hours light and dark cycle. The cages were cleaned on a daily basis. The illumination in animal house was controlled to give approximately a sequence of 12 hr. They were fed on standard chow and provided purified water and libitum.

The below table shows group of animals for the Study

Sr. No	Grouping	Treatment and Dose (Drug Administration)	No of Animals
1	Group I	Normal Control (No Application of any drug)	06
2	Group II	IMQ Disease group (Application of Imiquimod 62.5mg/day for 7 days to	06

		induce Psoriasis)	
3	Group III	IMQ+Test Drug (Application of Imiquimod to induce Psoriasis, followed by application of *Chakravhayadi lepa* once daily for next 7 days)	06
Total No		18	

Study Design

- Pre–clinical study / Animal Study: In-Vivo experiment.
- Healthy swiss albino mice in the study were divided into 3 groups, each containing of 6 animals.

- The groups were named as:

Group 1: Normal control group where there was no application of any drug.

Group 2: IMQ Disease group which received where there was application of Imiquimod 62.5mg/day for 7 days to induce Psoriasis.

Group 3: Application of Imiquimod to induce Psoriasis, followed by application of *Chakravhayadi lepa* once daily for next 7 days.

After 14 days of the study, body weight was observed and on the last day of the treatment, blood was withdrawn from the retroorbital plexus under mild isoflurane anaesthesia and used for estimation of inflammatory markers such as interleukin-6 (IL-6) and tumor necrosis factor-alpha (TNF-α) were quantified using ELISA, skin samples from psoriatic site were taken and subjected for histopathological investigation. Animals were sacrificed using CO2 euthanasia, spleen were isolated and weight of spleen was recorded.

Dose Fixation

1) Normal group: There will be no application of any drug.

2) Control group: Application of Imiquimod for 7 days to induce Psoriasis

3) Trial group: Application of Imiquimod to induce Psoriasis followed by application of *Chakravhayadi lepa* once in a day for next 7 days.

(The *lepa* will be applied uniformly over the affected skin surface.)

Thickness of *lepa*: According to *Sharangdhar samhita*, thickness of *vishaghna lepa* should be 1/3rd *angula* (1 *angula* is approximately 1.95 cm thick[7])

Duration of Study: 14 days.

Study protocol

PROCEDURE

1) In this study, healthy mice aged 8-10 weeks old were randomly assigned to three groups as per the above table: Normal Control, Disease Control (IMQ-treated), and Test Drug (*Chakravhayadi lepa* in *kitibha kushta*), with each group maintained under standard laboratory conditions and provided with ad libitum access to food and water.

2) On the day of Psoriasis induction, hair from mice were removed at the dorsal skin with 2x2 cm2 area using depilatory cream.

3) Psoriasis was induced to Group 2 and Group 3 by topically applying commercially available Imiquimod (IMQ) cream daily on the depilated area at a dose of 62.5mg/day for 7 days for a daily for 7 days.

Method of application of *Chakravhayadi lepa*

i) In trial group 6 albino mice were taken of both the genders and weighed before treatments.
ii) The test dose of local application was fixed according to the classics.
iii) *Chakravhayadi lepa* was applied on the affected area of animal models of Imiquimod induced psoriatic plaque for 7 days once a day.
iv) The effect of the *Chakravhayadi* lepa was observed on erythema, scaling and thickening of skin.
v) The *lepa* was removed by sterile bud before getting completely dried.
vi) Grading of scaling, erythema and thickening of skin was done.

Study Parameters

Clinical severity was evaluated by scoring three hallmark features of psoriasis according to PASI score[8]

- Erythema (redness),
- Induration (thickness), and
- Desquamation (scaling)

independently on a standardized scale from 0 to 4, where

- 0 indicated none,
- 1 indicated slight,

- 2 indicated mild or clearly visible,
- 3 indicated marked or moderate, and
- 4 indicated very marked or severe.

On the last day of the treatment, blood was withdrawn for estimation of inflammatory markers such as interleukin-6 (IL-6) and tumor necrosis factor-alpha (TNF-α) were quantified using ELISA.

After the blood withdrawal skin samples from psoriatic site were taken and subjected for histopathological investigation.

Animals were sacrificed, spleen were isolated and weight of spleen was recorded.

Statistical Analysis

Results were represented and expressed as mean ± SD (n=6). Data was analyzed by One-Way ANOVA followed by Bonferroni Post Hoc analysis. * P<0.5 was considered as statistically significant.

OBSERVATIONS AND RESULTS

Body weight

Table shows comparative data of Body weight between Control group, Disease group and Test Group by using ANOVA Test.

Mean body weight on various days

Group.no	Treatment and Dose	Body weights grams; Mean±SD		
		Day 0	Day 4	Day 7
1	Normal Control	20.50±1.64	23.17±2.14	24.83±2.79
2	Disease group (IMQ)	22.00±3.22	19.83±3.43	19.83±3.13
3	Test Chakravhayadi lepa in kitibha kushta	21.67±4.27	20.67±2.42	22.50±2.59

Erythema

Table showing comparative data of erythema between Control group, disease group and Test Group by using ANOVA Test.

Groups and Treatment	Erythema (Grade; Mean±SD)					
Normal Control	Day 0	Day5	Day 7	Day 9	Day 11	Day 14
	0.00±0.0	0.00±0.0	0.00±0.0	0.00±0.0	0.00±0.0	0.00±0.0
Disease group (IMQ)	0.00±0.0	0.50±1.2###	3.00±0.0###	3.50±0.5###	2.83±0.4###	2.33±0.8###
Test Chakravhaya di lepa in kitibha kushta	0.00±0.0	0.50±1.2	2.33±1.2	1.17±0.4***	1.33±0.8**	0.17±0.4***

Skin Scaling

Table showing comparative data of skin scaling between Control group, disease group and Test Group by using ANOVA Test.

Groups and Treatment	Scaling (Severity Score; Mean±SEM)					
Day 0		Day5	Day 7	Day 9	Day 11	Day 14
Normal Control	0.00±0.0	0.00±0.0	0.00±0.0	0.00±0.0	0.00±0.0	0.00±0.0
Disease group (IMQ	0.00±0.0	0.17±0.4###	0.17±0.4###	2.17±0.4###	3.17±0.8###	2.33±0.5###
Test *Chakravhaya di lepa in kitibha kushta*	0.00±0.0	0.17±0.4	0.00±0.0***	1.33±0.8**	1.00±0.9***	0.00±0.0***

Skin thickening

Table showing comparative data of skin scaling between Control group, disease group and Test Group by using ANOVA Test.

Groups and Treatment	Thickening (Severity Score; Mean±SEM)					
Day 0		Day5	Day 7	Day 9	Day 11	Day 14
Normal Control	0.00±0.0	0.00±0.0	0.00±0.0	0.00±0.0	0.00±0.0	0.00±0.0
Disease group (IMQ	0.00±0.0	0.50±1.2#	2.67±0.5###	2.67±0.5###	2.67±0.8###	2.00±1.1###
Test Chakravhaya di lepa in kitibha kushta	0.00±0.0	0.33±0.8	1.67±1.4*	1.00±0.9***	0.50±0.8***	0.00±0.0***

Result of spleen weight

Effects of Chakravhyadi Lepa on spleen weight in Imiquimod-induced Psoriasis in mice.

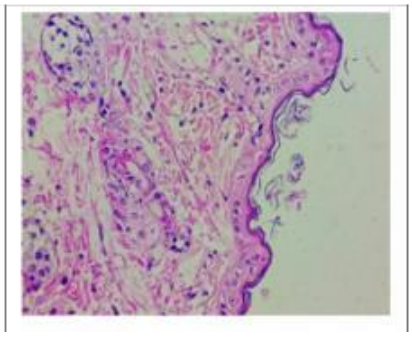
Groups and Treatment	Spleen weight (mg; Mean±SEM)
Normal Control	63.08±8.73
Disease group (IMQ	178.30±5.21###
Test Chakravhayadi lepa in kitibha kushta	100.00±15.29**

Result of Cytokines

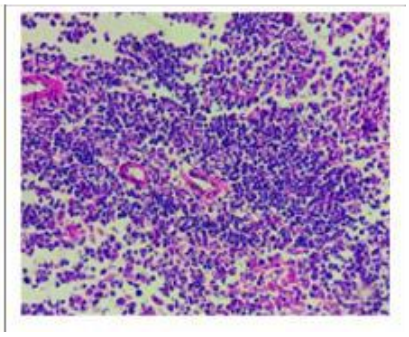
Effects of *Chakravhyadi Lepa* on IL-6 and TNF-α Levels in Imiquimod-Induced Psoriatic Mice.

Groups & Treatments	Serum cytokines (pg/mL) Mean ±SD	
	IL-6	TNF-α
Normal Control	6.52±2.67	16.57±6.48
Disease group (IMQ)	28.99±18.77#	66.75±8.30###
Test *Chakravhayadi lepa in kitibha kushta*	18.71±7.07	26.90±7.56***

Figure. Histopathology of Skin and Spleen

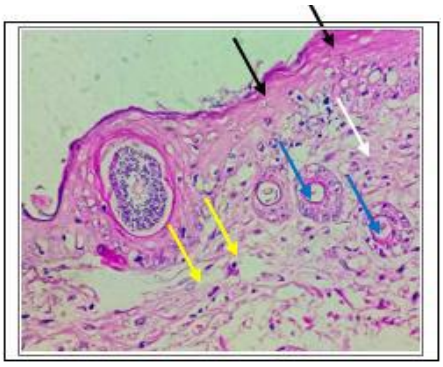


G1/1 (Skin)

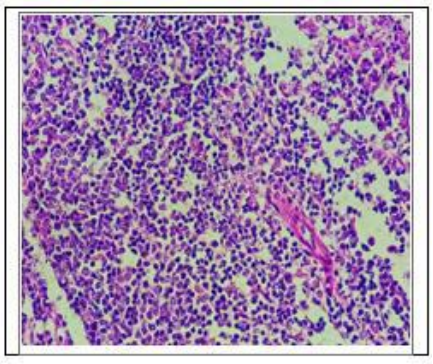


G1/1 (Spleen)

Group 1: Normal Control.

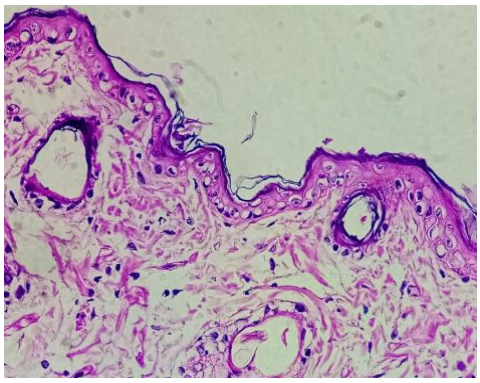


G2/1 (Skin)

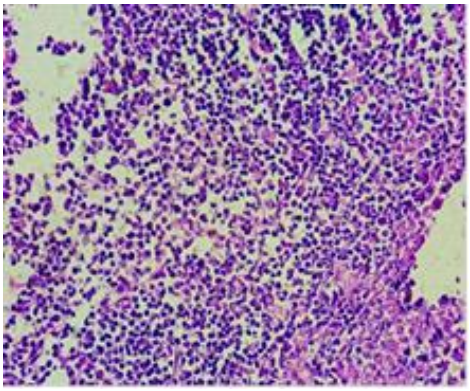


G2/1 (Spleen)

Group 2: Disease control (Imiquimod Appplied).



G3/1 (Skin)



G3/1 (Spleen)

Group 3: Test Group (*Chakravhayadi lepa*).

OBSERVATIONS

1. Skin

Group Name	Stratum basale/mitotic Activity	Acanthosis/ Average Epidermal thickness – prickle cell layer and granular cell layer	Rete ridges/ papillae/ Suprapapillary Thinning	Hyper/ Para/ Ortho - keratosis	Vascular changes	Leucocytic / Neutrophilic infiltration in para keratotic scale	Grade of lesion
Normal Control	Unremarkable	Not seen	NAD	NAD	NAD	NAD	Nil
Disease Control	Mild to moderate	Moderate to marked (black arrow)	Moderate to marked degree deepening of rete ridges (yellow arrow)	Moderate degree hyperkeratosis and mild degree parakeratosis (white arrow)	Minimal	Minimally Multi focal cluster of cocci	Moderate to marked degree (blue arrow)
Test Group I	Minimal	Mild (black arrow)	Minimal degree deepening of rete ridges	Minimal degree hyperkeratosis and minimal degree parakeratosis	Not seen	Not seen	Minimal

2. Spleen

Sample Code	Observations
Normal Control	No abnormalities were detected
Disease Control	No abnormalities were detected
Test Group I	No abnormalities were detected

DISCUSSION

According to *Acharya Charaka, Kushta* is a *Tridoshaja Vyadhi*, arising from the simultaneous vitiation of *Vata*, *Pitta***,** and *Kapha*, with varying manifestations based on their permutations (*Amsamsa Bheda*). *Kitibha Kushta* is primarily *Vata-Kapha* dominant, where aggravation of

Doshas due to specific *Nidana Sevana* leads to vitiation of *Dhatus—Tvak*, *Rakta*, *Mamsa*, and *Ambu*—together forming the *Saptaka Dravya Sangraha*, contributing to all eighteen types of *Kushta*. Dietary factors (*Aaharaja Hetu*) such as intake of Guru, Snigdha, *Ushna*, *Viruddha*, and *Dushivisha Aahara*, along with lifestyle factors (*Viharaja Hetu*) like *Divaswapa*, *Ratrojagarana*, and *Vegavrodha*, cause *Agnimandya* and *Ama* formation, further aggravating *Doshas* and *Rasadhatu*. Clinically, symptoms like erythema, scaling, and thickening closely correspond to classical descriptions of psoriasis.

The present study evaluated the efficacy of *Chakravhayadi Lepa* in albino mice with IMQ-induced psoriasis-like symptoms. Psoriasis is a chronic inflammatory skin disorder with recurrent relapses, for which conventional therapies provide only partial relief and are associated with serious adverse effects. *Chakravhayadi Lepa*, composed of *Chakramarda*, *Snuhi*, and *Gomutra*, is described in classical texts as a potent *Kushtaghna* formulation and offers a potentially safe and accessible alternative.

Experimental findings showed that IMQ application did not affect body weight. Treatment with *Chakravhayadi Lepa* significantly ($P<0.001$) reduced erythema, scaling, and skin thickening, with effects becoming more pronounced from day 9 onwards. Additionally, the *lepa* normalized spleen weight, attenuated serum inflammatory cytokines (IL-6 and TNF-α), and improved histopathological changes. These results indicate that *Chakravhayadi Lepa* exerts significant therapeutic effects in IMQ-induced psoriasis-like symptoms in mice.

Chakravhayadi Lepa, indicated by *Acharya Chakradatta* for *Kitibha Kushta*, contains *Chakramarda, Snuhi Ksheera*, and *Gomutra*. *Chakramarda* (*Katu Rasa, Ushna Virya*) pacifies *Kapha* and *Vata*, with *Kushtaghna, Tridosha Shamak, Kandughna, Vishahara*, and *Krimighna* properties, showing antifungal, antipsoriatic, anti-inflammatory, antioxidant, and antipruritic activities. *Snuhi Ksheera (Katu Rasa, Ushna Virya*) is *Vata-Kapha* pacifying, with *Kushtaghna*, *Vranaropaka, Rookshaghna*, and *Krimighna* actions, exhibiting anti-inflammatory, analgesic, and wound-healing effects in skin disorders like *Padvidarika, Dadru*, and *Vicharchika. Gomutra (Katu Rasa, Ushna Virya*) offers Kushtaghna, *Vranaropaka, Krimighna, Rasayana*, and *Shoolaghna* effects, with analgesic, antimicrobial, antifungal, immunomodulatory, and wound-healing activities. Collectively, these ingredients provide a multi-targeted therapeutic action in the management of psoriasis and other skin disorders.

Chakravhayadi Lepa, containing *Chakramarda, Snuhi Ksheera*, and *Gomutra*, exerts *Kapha-Vata* pacifying and multi-targeted effects in *Kitibha Kushta*. Its ingredients provide *Kushtaghna, Krimighna, Kandughna, Vranaropaka*, and Vata kapha shamak actions, reducing erythema, scaling, and thickening through anti-inflammatory, wound-healing, and bio-enhancing properties.

Body Weight: IMQ application did not affect body weight, and *Chakravhayadi Lepa* treatment also showed no significant changes, indicating safety. **Erythema, Scaling, and Thickening:** IMQ (Group 2) induced significant psoriasis-like symptoms from day 5–7. *Chakravhayadi Lepa* (Group 3) significantly reduced these symptoms ($P < 0.001$), with effects more pronounced from day 9 onwards. **Spleen Weight:** IMQ increased spleen weight, reflecting immune activation. *Chakravhayadi Lepa* normalized spleen size, suggesting immunomodulatory activity. **Cytokines:** IMQ elevated **IL-6** and **TNF-α**, promoting inflammation and keratinocyte proliferation. Treatment with *Chakravhayadi Lepa* significantly attenuated these cytokine levels. **Histopathology:** IMQ caused mild-to-moderate skin alterations, whereas *Chakravhayadi Lepa* treatment minimized these changes. Spleen histology remained normal in all groups, indicating no systemic toxicity.

CONCLUSION

The physico-chemical analysis of the individual ingredients and formulation revealed that all parameters were within permissible limits, confirming the quality and safety of the materials used. Specifically, *Chakramarda Churna, Snuhi Ksheera, Gomutra*, and the final preparation, *Snuhi Ksheera Bhavit Chakramarda Churna*, met the required standards, ensuring consistency and suitability for experimental use.

The animal study provided significant insights into the efficacy of *Chakravhayadi Lepa* in *Kitibha Kushtha* with reference to imiquimod (IMQ)-induced psoriasis in albino mice. IMQ application for seven consecutive days (Group 2) produced pronounced psoriatic symptoms, including erythema, scaling, and skin thickening, which became prominent from day 5 to day 7 compared to the normal control group. This confirmed successful induction of psoriasis-like lesions in the experimental model.

Treatment with *Chakravhayadi Lepa* (Group 3) demonstrated notable therapeutic effects. From day 9 onwards, mice treated with the formulation showed significant reductions in erythema, scaling, and skin thickening relative to the disease control group. Additionally,

IMQ-induced psoriasis was associated with an increase in spleen weight, reflecting systemic immune activation. *Chakravhayadi Lepa* treatment significantly normalized spleen weight, suggesting immunomodulatory effects and overall improvement in psoriatic pathology.

At the molecular level, IMQ application significantly elevated serum inflammatory cytokines, primarily IL-6 and TNF-α, indicative of heightened inflammatory responses. Administration of *Chakravhayadi Lepa* effectively attenuated these cytokine levels, highlighting its anti-inflammatory potential. Histopathological examination further confirmed these findings, showing marked reversal of epidermal hyperplasia, keratinocyte proliferation, and inflammatory cell infiltration in treated mice.

In summary, the study demonstrates that *Chakravhayadi Lepa*, when applied for seven days, effectively alleviates IMQ-induced psoriasis-like symptoms in albino mice. The treatment reduced erythema, scaling, and skin thickening, normalized spleen weight, suppressed pro-inflammatory cytokines, and improved histopathological alterations, indicating a multi-targeted therapeutic potential for *Kitibha Kushtha* management.

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