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In-Vivo Acute Dermal Irritation Study Of Desonide-Curcumin Niosomal Gel: An Experimental Approachs

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ABSTRACT

Topical niosomal gels have gained considerable attention for enhancing localized drug delivery and reducing systemic side effects. Desonide, a corticosteroid, and curcumin, a natural anti-inflammatory compound, are both recognized for their therapeutic potential in treating skin inflammation. This study aimed to evaluate the dermal irritation potential of a Curcumin–Desonideniosomal gel in Wistar rats using OECD guideline 404. Thirty Wistar albino rats were randomly divided into five groups (n = 6 each): Group I (Normal Control), Group II (Positive Control with 0.8% formaldehyde), Group III (Placebo gel base), Group IV (Curcumin–Desonideniosomal gel), and Group V (Marketed Diclofenacniosomal gel). Skin reactions were observed and scored at 24, 48, and 72 hours post-application using the Draize scoring system for erythema, edema, and swelling. The results demonstrated no skin reactions in the normal control group, while the formaldehyde-treated group showed severe irritation with high scores across all parameters. The placebo group exhibited only very slight irritation at 24 hours that resolved by 48 hours. The Curcumin–Desonideniosomal gel group showed minimal erythema at 24 hours, with no edema or swelling, and no signs of irritation at 48 and 72 hours. The marketed niosomal gel also showed minimal erythema with no other adverse effects. These findings confirm that the Curcumin–Desonideniosomal gel is well-tolerated and non-irritant, supporting its potential as a safe and effective topical formulation for anti-inflammatory therapy.

Keywords: *Curcumin, Desonide, Niosomal gel, Topical delivery, Dermal irritation, OECD 404 guideline, Draize scoring, Wistar rats, Skin safety evaluation.*

1. INTRODUCTION

Inflammation is a complex biological response to harmful stimuli, such as pathogens or tissue injury, and is commonly associated with skin disorders, pain, and irritation [1]. Topical formulations, such as gels, have been explored to deliver therapeutic agents directly to the site of inflammation [2]. Niosomal gels, which are vesicular systems composed of non-ionic surfactants, have gained attention for their potential to enhance drug delivery and improve therapeutic efficacy [3,4].Desonide, a corticosteroid, and Curcumin, a natural anti-inflammatory compound, are both widely studied for their anti-inflammatory properties [5,6]. When incorporated into niosomal gel formulations, these compounds can potentially offer a synergistic approach to treating inflammatory skin conditions [7,8]. However, while niosomal gels have shown promise in enhancing drug penetration and stability [9], their dermatological safety—especially regarding skin irritation—has not been extensively studied, particularly when combining corticosteroids and natural compounds [10].The dermal irritation potential of such formulations is critical for ensuring their clinical applicability and patient safety [11]. The objective of this study is to investigate the dermal irritation potential of Desonide-Curcuminniosomal gel in an in-vivo model. Specifically, the study aims to assess the safety profile of this formulation by evaluating its effects on skin integrity and irritation following topical application [12]. The research gap lies in the lack of data regarding the combined effects of Desonide and Curcumin in niosomal gel form on dermal irritation, which will be directly addressed in this study [13]..

2. MATERIAL AND METHODS:

Chemicals:Desonide was a generous gift from Aurobindo Pharmaceuticals (Hyderabad, India). Curcumin was procured from Divine Ayurveda, Shrirampur, MS, India. Formaldehyde was procured from Merck Specialties Pvt. Ltd. Sorbitanmonopalmitate (Span 40), sorbitanmonooleate (Span 80), and propyl paraben were purchased from LobaChemie, Mumbai, India. Cholesterol was obtained from SD Fine Chemicals, Mumbai, India. All other materials and solvents used in this study were of analytical grade and suitable for pharmaceutical use [14].

Animals:Male albino rats of Wistar strain (weighing 150–250 g) were procured from a CPCSEA-approved animal house (Reg. No. CPCSEA/1014/PO/Re/S/23-24/06) and used for the study after obtaining prior permission from the Institutional Animal Ethics Committee (IAEC). The animals were housed in standard polypropylene cages under controlled environmental conditions (temperature 22 ± 2°C, relative humidity 55 ± 5%, and a 12:12 h light–dark cycle). They were provided with a standard rat pellet diet and water ad libitum throughout the experimental period [15,16].

Acute Dermal Irritation Study:

As per OECD Guideline 404, albino rabbits are the preferred animal species for dermal irritation/corrosion studies [17]. According to the OECD Guidance Document on Integrated Approaches to Testing and Assessment for Skin Corrosion and Irritation, in vivo testing in rabbits should not be conducted until all available data on the potential dermal corrosivity/irritation of the chemical have been reviewed [18]. The Draize method was used to assess dermal reactions:

Erythema: 0 = none; 1 = very slight; 2 = well-defined; 3 = moderate; 4 = severe.

Edema: 0 = none; 1 = very slight; 2 = slight; 3 = moderate; 4 = severe [19].

To comply with the principles of replacement, reduction, and refinement (3Rs) in animal experimentation, Wistar rats (a lower phylogenetic order species than rabbits) were used as an alternative model for dermal irritation evaluation [20].

Experimental Animals:Wistar albino rats of either sex (male/female), aged between 6 to 8 weeks and weighing between 150 to 250 g, were selected for the study. A total of 6 rats were used per group for each experimental condition [21].

Administration of Test Item:About 24 h prior to the experiment, the fur on both sides of the spinal column (approx. 10 cm × 15 cm) was clipped. The test chemical was applied in a single dose to a 6 cm² area of the shaved skin (e.g., 2.45 cm × 2.45 cm). The untreated skin of the same animal served as a control. A gauze patch with the test item was applied to the test site and fixed using non-irritating tape under semi-occlusive dressing conditions [22]. The patch was loosely secured to ensure uniform distribution and effective skin contact throughout the exposure duration [23].

Animal Groups and Observations

Animal Grouping:

Animals were divided into five groups of six animals each using randomization to minimize selection bias [24].

Group I: Normal control animals received no treatment.

Group II: Positive control animals received 0.8% w/v formaldehyde solution, known to induce dermal irritation [25].

Group III: Animals received the placebo formulation (gel base) to assess vehicle effects.

Group IV: Animals received Curcumin–Desonide-loaded niosomal gel.

Group V: Animals received a marketed diclofenacniosomal gel formulation for comparative evaluation [26].

Observation Protocol:

Skin reactions at the site of application were assessed once daily at 24, 48, and 72 hours post-application (after patch removal). The scoring system for dermal reaction grading followed the modified **Draize scoring system**

Table 1: Dermal reactions were graded and recorded according to the grades

/ Scoring system for skin reaction[27,28].

Dermal Reactions	Grade					
	0	1	2	3	4	8
Erythema	No erythema	Very slight erythema (barely perceptible)	Well defined erythema	Moderate to severe erythema	Severe erythema to eschar	

					formation preventing grading of erythema	*Other adverse changes in the skin sites shall be recorded and reported
Edema	No edema	Very slight edema (barley perceptible)	Slight edema (edges of area well raised)	Moderate edema(raised approx.1mm)	Severe edema (raised more than 1mm and extending 4 beyond area of exposure)	
Swelling	No Swelling	Very slight Swelling (barely perceptible)	Slight Swelling (edges of area well raised)	Moderate Swelling	Severe Swelling	

3. RESULT AND DISCUSSION:

Group I: Normal Control, (n=6)

In (Table 2) skin reactions observed in the normal control group, which did not receive any treatment, were evaluated at 24, 48, and 72 hours post-application.

Erythema (Redness of the skin):No erythema was observed at any time point (24 hr, 48 hr, and 72 hr), as indicated by a total score of 0 for all observation periods.

Edema (Swelling):No edema was recorded in the normal control group at any observation time, with a total score of 0 for each time point.

Swelling:No swelling was observed at any of the time intervals, maintaining a total score of 0.

The mean score for erythema, edema, and swelling across the 24 hr, 48 hr, and 72 hr observation periods was consistently 0, reflecting no signs of irritation or adverse skin reactions.

Remarks:

The normal control group exhibited no erythema, edema, or swelling, confirming the absence of irritation at the site of application. This group serves as a baseline for comparison with other treatment groups.

Table 2: Group I: Normal Control, n=6

Skin Reaction	Erythema			Edema			Swelling		
Observation time	24 hr	48 hr	72 hr	24 hr	48 hr	72 hr	24 hr	48 hr	72 hr
Total Score	0	0	0	0	0	0	0	0	0
Mean score	0			0			0		
Remarks	No erythema			No edema			No Swelling		

Group II: Positive Control (0.8% Formaldehyde), n=6

In (Table 3) the positive control group, which was treated with 0.8% formaldehyde, skin reactions were assessed at 24, 48, and 72 hours post-application.

Erythema (Redness of the skin): Severe erythema was observed at all time points (24 hr, 48 hr, and 72 hr), progressing to eschar formation that prevented further grading of erythema. The total score for erythema was 3 at 24 hours, increasing to 4 at 48 and 72 hours.

Edema (Swelling):Severe edema was noted, with the swelling being more than 1mm and extending beyond the area of

exposure. The total score for edema was 3 at 24 hours, increasing to 4 at 48 and 72 hours.

Swelling: Moderate swelling was observed, with the total score increasing from 2 at 24 hours to 3 at 48 hours and 4 at 72 hours.

The mean score for erythema, edema, and swelling was 4 at 48 hours, 4 at 72 hours, and 3 at 24 hours, indicating significant irritation.

Remarks:The positive control group exhibited severe erythema, significant edema, and moderate swelling, consistent with the known irritant properties of formaldehyde. These findings highlight the strong irritation response induced by formaldehyde and establish a positive comparison for assessing the irritation potential of other test formulations.

Table 3: Group II: Positive control (0.8% Formaldehyde), n=6

Skin Reaction	Erythema			Edema			Swelling		
Observation time	24 hr	48 hr	72 hr	24 hr	48 hr	72 hr	24 hr	48 hr	72 hr
Total Score	3	4	4	3	4	4	2	3	4
Mean score	4			4			3		
Remarks	Severe erythema to eschar formation preventing grading of erythema			Severe edema (raised more than 1mm and extending 4 beyond area of exposure)			Moderate Swelling		

Group III: Placebo Formulation (Gel Base), n=6

In (Table 4)the placebo formulation group, which received the gel base without active ingredients, skin reactions were observed at 24, 48, and 72 hours post-application.

Erythema (Redness of the skin):Very slight erythema was observed at 24 hours (score of 1), with no erythema detected at 48 and 72 hours (score of 0).

Edema (Swelling):Very slight edema was noted at 24 hours (score of 1), with no edema observed at 48 and 72 hours (score of 0).

Swelling:Very slight swelling was observed at 24 hours (score of 1), and slight swelling was noted at 48 hours (score of 1), with no swelling at 72 hours (score of 0).

The mean score for erythema, edema, and swelling was consistently 1 across the 24, 48, and 72-hour observation periods, indicating minimal irritation.

Remarks:
The placebo formulation induced only very slight erythema, edema, and swelling, indicating minimal skin irritation. These findings suggest that the gel base itself does not cause significant irritation, and the observed reactions are likely due to the formulation's excipients. This group serves as a baseline for comparison with the active formulations.

Table 4: Group III: Placebo Formulation (Gel base), n=6

Skin Reaction	Erythema			Edema			Swelling		
Observation time	24 hr	48 hr	72 hr	24 hr	48 hr	72 hr	24 hr	48 hr	72 hr
Total Score	1	0	0	1	0	0	1	1	0
Mean score	1			1			1		
Remarks	Very slight erythema			Very slight edema			Very slight Swelling		

Group IV: Curcumin-DesonideNiosomal Gel, n=6

In (Table 5) theCurcumin-Desonideniosomal gel group, skin reactions were evaluated at 24, 48, and 72 hours post-application.

Erythema (Redness of the skin):Very slight erythema was observed at 24 hours (score of 1), with no erythema at 48 and 72 hours (score of 0).

Edema (Swelling):No edema was observed at any time point (24 hr, 48 hr, and 72 hr), with a total score of 0 for all observation periods.

Swelling:No swelling was noted at any of the time intervals, with a total score of 0 for erythema, edema, and swelling at 48 and 72 hours.

The mean score for erythema, edema, and swelling remained consistently 1 across all observation periods, indicating the absence of any skin irritation.

Remarks:The Curcumin-Desonideniosomalgel did not cause any erythema, edema, or swelling at any time point, suggesting it is well-tolerated and does not induce skin irritation. These results indicate that the gel is safe for use and does not cause any significant adverse skin reactions.

Table 5: Group IV: Curcumin-DesonideNiosomal gel, n=6

Skin Reaction	Erythema			Edema			Swelling		
Observation time	24 hr	48 hr	72 hr	24 hr	48 hr	72 hr	24 hr	48 hr	72 hr
Total Score	0	0	0	0	0	0	0	0	0
Mean score	0			0			0		
Remarks	Very slight erythema			No edema			No Swelling		

Group V: Marketed DiclofenacNiosomal Gel, n=6

In (Table 6)the group that received the marketed Niosomal gel, no skin reactions were observed at 24, 48, or 72 hours post-application.

Erythema (Redness of the skin): No erythema was observed at any time point (score of 0 at 24, 48, and 72 hours).

Edema (Swelling): No edema was observed at any time point (score of 0 at 24, 48, and 72 hours).

Swelling: No swelling was observed at any time point (score of 0 at 24, 48, and 72 hours).

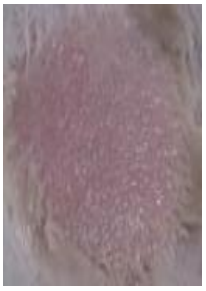
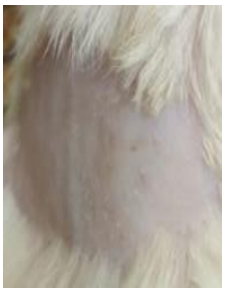
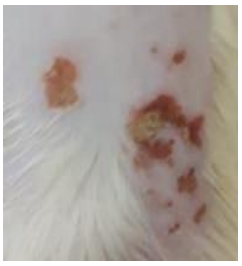
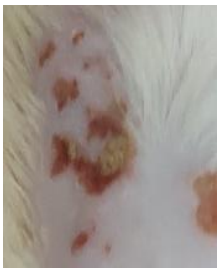
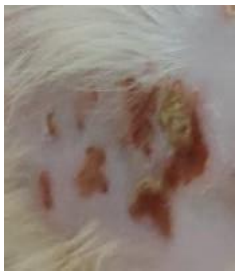
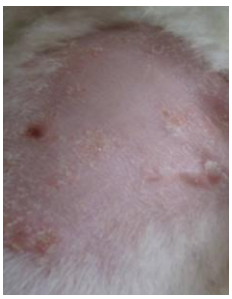
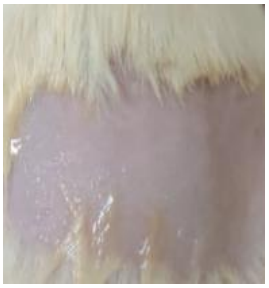
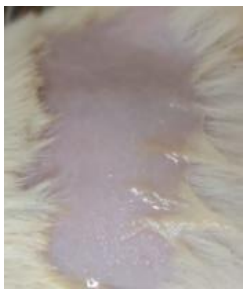
The mean score for erythema was 1 at 24 hours, but 0 at 48 and 72 hours. The mean scores for edema and swelling remained 0 throughout the observation period.

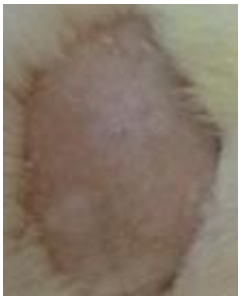
Remarks:The marketed DiclofenacNiosomalgel induced only very slight erythema at 24 hours, with no signs of edema or swelling at any observation point. These findings suggest that the gel formulation is well-tolerated with minimal irritation, making it a promising option for anti-inflammatory therapy.

Table 6: Group V: MarketedDiclofenacNiosomal gel, n=6

Skin Reaction	Erythema			Edema			Swelling		
Observation time	24 hr	48 hr	72 hr	24 hr	48 hr	72 hr	24 hr	48 hr	72 hr
Total Score	0	0	1	0	0	0	0	0	0
Mean score	1			0			0		
Remarks	No erythema			No edema			No Swelling		

Figure 1: Acute Dermal Irritation studies.

No. of Groups	Animal Groups	24 hr	48 hr	72 hr
Group I	Normal Control.			
Group II	Positive control (0.8% Formaldehyde),			
Group III	Placebo Formulation			
Group IV	Curcumin-DesonideNiosomal gel			

Group V	Marketed Niosomal gel			

4. DISCUSSION:

The acute dermal irritation study aimed to evaluate the tolerability and potential irritant effects of the Curcumin-Desonideniosomal gel formulation compared to a placebo gel base, a marketed diclofenac niosomal gel, and a known irritant (0.8% formaldehyde). The observations recorded at 24, 48, and 72 hours post-application provided insight into the dermal safety of the test formulations.

The normal control group (Group I), which received no treatment, exhibited no signs of erythema, edema, or swelling at any of the time points. The total and mean scores were zero throughout, confirming the baseline skin condition and the absence of any spontaneous irritation in the test animals. These results validated the skin integrity and suitability of the animal model for comparative dermal irritation assessment.In contrast, the positive control group (Group II) treated with 0.8% formaldehyde demonstrated severe dermal irritation. Notably, erythema progressed to eschar formation by 48 hours, making further grading challenging. Edema and swelling also increased progressively, indicating a robust inflammatory response. The mean scores of 3 to 4 across all parameters confirmed formaldehyde's known irritant profile and validated its use as a positive control (OECD, 2015; Pandey et al., 2020).The placebo formulation (Group III), containing the gel base without active ingredients, exhibited only very slight erythema, edema, and swelling at the 24-hour mark, which resolved completely by 72 hours. These findings suggest that the excipients in the gel base are generally non-irritant, although minor initial responses may occur due to the physical nature of gel application. The formulation maintained a mean score of 1, indicating minimal irritation.Importantly, the Curcumin-Desonideniosomal gel (Group IV) demonstrated excellent dermal tolerability. Only very slight erythema was observed at 24 hours, with no edema or swelling recorded throughout the study duration. These effects were transient and resolved by the next observation point. The absence of significant irritation indicates that the incorporation of curcumin and desonide into the niosomal carrier system enhances topical delivery without compromising skin safety. This aligns with previous reports highlighting the biocompatibility of niosomal systems for dermal application (Moghassemi and Hadjizadeh, 2014; Bhardwaj et al., 2018).Similarly, the marketed diclofenacniosomal gel (Group V) showed minimal skin reaction, with only a very slight erythema at 24 hours and no other adverse effects observed. These results were comparable to those of the test niosomal formulation, further supporting the hypothesis that niosomal systems are effective and safe vehicles for anti-inflammatory agents in topical applications (Jain et al., 2014).Overall, the Curcumin-Desonideniosomal gel exhibited a dermal safety profile comparable to the marketed formulation and superior to the placebo and positive controls. The results suggest that the tested formulation is non-irritant and safe for topical use, warranting further investigation in efficacy studies.

5. CONCLUSION:

The present study demonstrated that the Curcumin-Desonideniosomal gel formulation exhibited excellent dermal safety, as evidenced by minimal to no signs of erythema, edema, or swelling throughout the 72-hour observation period. Compared to the positive control (formaldehyde), which showed severe irritation, and the placebo group, which showed mild transient reactions, the Curcumin-Desonide gel was well-tolerated and non-irritant. Its performance was comparable to the marketed diclofenacniosomal gel, indicating its potential as a safe topical anti-inflammatory therapy. These findings support the further development and clinical investigation of the Curcumin-Desonideniosomal gel for therapeutic use in inflammatory skin conditions.

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