International Journal of Research in Dermatology
Sangle S et al. Int J Res Dermatol. 2026 Nov;12(6):633-639
https://www.ijord.com

DOI: https://dx.doi.org/10.18203/issn.2455-4529.IntJResDermatol20263742

Original Research Article

In vitro comparative evaluation of skin permeation and dermal retention of clobetasol 0.05% and salicylic acid 6% nano lotion and clobetasol 0.05% and salicylic acid 6% conventional lotion using excised swine skin and Franz diffusion cells

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Received: 18 August 2026
Revised: 29 September 2026
Accepted: 14 October 2026

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ABSTRACT

Background: Topical formulations aim to deliver drugs locally while minimizing systemic exposure. This study compared the in vitro skin permeation and dermal retention of Clobetasol Propionate 0.05% and Salicylic Acid 6% Nano Lotion with a Conventional Lotion.
Methods: Excised full-thickness dorsal swine skin was mounted on Franz diffusion cells with a diffusion area of 1.36 cm^2. A 100 mg dose of each formulation was applied and receptor-fluid samples were collected over 12 hours. Both active ingredients were quantified using reverse-phase high-performance liquid chromatography (RP-HPLC) with UV detection. Permeation, surface residue and skin retention were assessed using mass-balance calculations.
Results: Salicylic Acid showed time-dependent permeation from both formulations. At 12 hours, cumulative permeation was 0.969 mg (16.16%) for the nano lotion and 0.270 mg (4.50%) for the conventional lotion, indicating approximately 3.6-fold greater permeation with the nano lotion. Skin retention was 83.08% and 71.81%, respectively, while surface residues were 0.763% and 23.68%. Clobetasol propionate remained below the receptor-phase detection limit for both formulations. However, skin retention was higher with the nano lotion (86%) than with the conventional lotion (50%).
Conclusions: The nano lotion demonstrated greater salicylic acid permeation, higher dermal retention of both active ingredients and lower surface residue than the conventional lotion. These findings suggest improved dermal delivery and skin localization under the tested in vitro conditions. Further studies are needed to confirm these findings and establish their clinical relevance.

Keywords: Clobetasol propionate, Dermal retention, Franz diffusion cell, Nano lotion, OECD 428, Salicylic acid, Skin permeation, Swine skin

INTRODUCTION

Topical drug delivery is widely used in dermatological treatment because it can provide local exposure to active pharmaceutical ingredients while potentially reducing systemic exposure. The effectiveness of a topical product depends on the amount of drug released from its vehicle, its ability to enter the skin and its distribution within the relevant skin layers.[1,2] The stratum corneum, the outermost layer of the epidermis, represents the principal barrier to the passage of many externally applied substances. Consequently, the composition and physical properties of a formulation can influence drug

partitioning into the skin and subsequent movement through its layers.[1,2] Clobetasol propionate is a potent topical corticosteroid used to manage inflammatory dermatoses, including psoriasis and eczema. Salicylic Acid is a keratolytic agent that facilitates the removal of accumulated keratin and may improve the penetration of co-administered topical agents.[3,4] Combining these active ingredients in a topical formulation may therefore provide complementary pharmacological effects.

Nano-based formulations have been investigated as approaches to modify the release and distribution of drugs following topical application. Their performance depends on factors such as carrier composition, particle characteristics, drug solubility and interactions between the formulation and the skin barrier. Improved skin delivery should therefore be demonstrated experimentally rather than inferred from the designation of a formulation as man-sized.[5,6]

Franz diffusion cells provide an experimental system for investigating the movement of substances across excised skin by measuring drug concentrations in the receptor compartment over time. The distribution of the applied substance within the test system can also be assessed at the end of the experiment. OECD test guideline 428 describes principles for conducting in vitro skin absorption studies and evaluating absorption and distribution under defined experimental conditions.[7] Porcine skin is frequently used in percutaneous absorption research because it provides a useful model for investigating skin-barrier transport, although it does not completely reproduce human skin.[8]

The present study compared the in vitro skin permeation and dermal retention of clobetasol propionate 0.05% w/w and salicylic acid 6% w/w in nano lotion and conventional lotion using excised dorsal swine skin mounted on Franz diffusion cells.

The comparison focused on cumulative receptor-phase permeation, residual drug recovered from the skin surface and estimated drug retention within the skin over a 12 hours period.

METHODS

Study design

An in vitro dermal absorption study was conducted to compare clobetasol propionate 0.05% and salicylic acid 6% nano lotion with conventional lotion using excised full-thickness swine skin mounted on Franz diffusion cells over 12 hours.

The study was designed with reference to OECD test guideline 428 for in vitro skin absorption studies.[7]

Two independent Franz diffusion cells were used for each formulation. The reported values represent the mean of duplicate determinations.

Test formulations

Two formulations were evaluated.

Clop S nano lotion

Clobetasol propionate (0.05% w/w) and salicylic acid (6% w/w), designated as SRC/AB/25/12/01.

Conventional lotion

Clobetasol propionate (0.05% w/w) and salicylic acid (6% w/w), designated as SRC/AB/25/12/02.

Skin preparation

Full-thickness dorsal swine skin (Sus scrofa) was used as a surrogate model for human skin. Subcutaneous fat was removed and the skin was stored at −20°C until use. Before the experiment, the skin was thawed and mounted on Franz diffusion cells with a diffusion area of 1.36 cm^2.

Franz diffusion setup

The receptor compartment of each Franz diffusion cell was filled with 5.0 ml of phosphate buffer containing 20% ethanol and 0.5% Tween 20 to maintain sink conditions. The receptor medium was maintained at 35±1°C and continuously stirred at 150 rpm using a magnetic stirrer.

At predetermined time points (0.5, 1, 2, 4, 6, 8, 10 and 12 hours), 0.5 ml aliquots of receptor fluid were collected for HPLC analysis and immediately replaced with an equal volume of fresh receptor medium equilibrated to the study temperature. This procedure maintained a constant receptor volume during the experiment.

Dose application

A finite dose of 100 mg of each formulation was applied uniformly to the skin. The applied dose contained 6 mg of salicylic acid and 0.05 mg of clobetasol propionate.

HPLC analysis

Drug quantification was performed using reverse-phase high-performance liquid chromatography (RP-HPLC) with a C18 column (250×4.6 mm, 5 µm).

The mobile phase consisted of orthophosphoric acid solution (Phase A) and acetonitrile (Phase B) in a 50:50 (v/v) ratio. The flow rate was 1.0 ml/min and the injection volume was 20 µl.

UV detection was performed at approximately 230 nm for salicylic acid and 240 nm for clobetasol propionate.

Data analysis

Cumulative permeation (receptor phase)

The cumulative amount of drug recovered in the receptor fluid was considered the permeated fraction.

% Permeation= (Amount in receptor fluid×100)/Applied dose

Surface residue (unabsorbed fraction)

Residual formulation recovered from skin surface at 12 hours.

% Surface residue/Unabsorbed= (drug recovered from skin surface×100)/applied dose

Skin retention

Drug retained within the skin was calculated by mass balance using the difference between the applied dose and the sum of drug recovered in the receptor phase and on the skin surface after 12 hours.

% Skin retention= ((Applied dose−receptor amount−surface residue)×100)/applied dose.

RESULTS

Cumulative permeation of salicylic acid and clobetasol propionate

The cumulative permeation of salicylic acid and clobetasol propionate across excised swine skin over 12 hours is presented in Tables 1–3. A time-dependent increase in salicylic acid permeation was observed with both formulations; however, clobetasol 0.05% and Salicylic acid 6 % Nano lotion consistently demonstrated higher permeation at all evaluated time points. At 6 hours, permeation from clobetasol 0.05% and salicylic acid 6 % nano lotion was 4.59%, compared to 1.16% from clobetasol 0.05% and salicylic acid 6 % conventional lotion. The difference became more pronounced at 8 hours (7.05% vs. 2.17%) and 10 hours (10.22% vs. 3.42%).

At 12 hours, cumulative permeation of salicylic acid reached 0.969 mg (16.16% of the applied dose) from clop s nano lotion, whereas clobetasol 0.05% and salicylic acid 6 % conventional lotion showed cumulative permeation of 0.270 mg (4.50% of the applied dose). Thus, salicylic acid permeation from clobetasol 0.05% and salicylic acid 6 % nano lotion was approximately 3.6-fold higher than that from clobetasol 0.05% and salicylic acid 6 % conventional lotion. For clobetasol propionate, no measurable drug was detected in the receptor fluid for either formulation throughout the 12 hours study period, as concentrations remained below the detection limit of the analytical method. Consequently, no quantifiable receptor phase permeation of clobetasol propionate was observed under the experimental conditions employed.

Distribution of drug at 12 hours

The distribution of salicylic acid and clobetasol propionate following topical application is summarized in tables 4–6. For salicylic acid, clobetasol 0.05% and salicylic acid 6 % nano lotion demonstrated 83.08% retention within the skin and only 0.763% remaining on the skin surface after 12 hours. In contrast, clobetasol 0.05% and salicylic acid 6 % conventional lotion exhibited 71.81% skin retention and 23.68% remaining on the skin surface. These findings indicate greater uptake and localization of salicylic acid within the skin layers from the nano formulation.

Table 1: Cumulative permeation of salicylic acid and clobetasol propionate across excised swine skin from clobetasol 0.05% and salicylic acid 6 % nano lotion (SRC/AB/25/12/01) over 12 hours.

Clobetasol 0.05% and salicylic acid 6 % nano lotion (SRC/AB/25/12/01)				
	Salicylic acid		Clobetasol	
Time	Amount permeated (mg)	Amount permeated (%)	Amount permeated (mg)	Amount permeated (%)
30 min	0.014	0.24%	BDL	NA
1 hour	0.031	0.52%	BDL	NA
2 hours	0.059	0.99%	BDL	NA
4 hours	0.119	1.99%	BDL	NA
6 hours	0.275	4.59%	BDL	NA
8 hours	0.422	7.05%	BDL	NA
10 hours	0.613	10.22%	BDL	NA
12 hours	0.969	16.16%	BDL	NA

Table 2: Cumulative permeation of salicylic acid and clobetasol propionate across excised swine skin from clobetasol 0.05% and salicylic acid 6 % conventional lotion (SRC/AB/25/12/02) over 12 hours.

Clobetasol 0.05% & salicylic acid 6 % conventional lotion (SRC/AB/25/12/02)				
Time	Salicylic acid		Clobetasol	
	Amount permeated (mg)	Amount permeated (%)	Amount permeated (mg)	Amount permeated (%)
30 min	0.011	0.18%	BDL	NA
1 hour	0.020	0.35%	BDL	NA
2 hours	0.030	0.50%	BDL	NA
4 hours	0.042	0.71%	BDL	NA
6 hours	0.069	1.16%	BDL	NA
8 hours	0.130	2.17%	BDL	NA
10 hours	0.205	3.42%	BDL	NA
12 hours	0.270	4.51%	BDL	NA

Table 3: Comparative percentage permeation of salicylic acid across excised swine skin from clobetasol 0.05% and salicylic acid 6 % nano lotion and clobetasol 0.05% and salicylic acid 6 % conventional lotion at different time points.

Time	Salicylic acid (% amount permeated)	
	Clobetasol 0.05% and salicylic acid 6 % nano lotion (SRC/AB/25/12/01)	Clobetasol 0.05% and salicylic acid 6 % conventional lotion (SRC/AB/25/12/02)
30 min	0.24%	0.18%
1 hour	0.52%	0.35%
2 hours	0.99%	0.50%
4 hours	1.99%	0.71%
6 hours	4.59%	1.16%
8 hours	7.05%	2.17%
10 hours	10.22%	3.42%
12 hours	16.16%	4.51%

Table 4: Distribution of salicylic acid and clobetasol propionate following application of clobetasol 0.05% and salicylic acid 6 % nano lotion at 12 hours: permeated, surface residual and skin-retained fractions.

Clobetasol 0.05% and salicylic acid 6 % nano lotion (SRC/AB/25/12/01)				
Parameter (at 12 hours)	Salicylic acid		Clobetasol	
	mg	%	mg	%
Cumulative drug permeated (% permeated)	0.969	16.16	BDL	NA
Drug remaining on skin surface (% unabsorbed)	0.045	0.763	0.007	14
Drug retained in skin (% skin retention)	4.984	83.08	0.043	86
Amount present in 100 mg sample	6.000	NA	0.050	100

Table 5: Distribution of salicylic acid and clobetasol propionate following application of clobetasol 0.05% and salicylic acid 6 % conventional lotion at 12 hours: permeated, surface residual and skin-retained fractions.

Clobetasol 0.05% and salicylic acid 6 % conventional lotion (SRC/AB/25/12/02)				
Parameter (at 12 hours)	Salicylic Acid		Clobetasol	
	mg	%	mg	%
Cumulative drug permeated (% permeated)	0.270	4.50	BDL	NA
Drug remaining on skin surface (% unabsorbed)	1.420	23.68	0.025	50
Drug retained in skin (% skin retention)	4.308	71.81	0.025	50
Amount present in 100 mg sample	6.000	NA	0.050	100

Table 6: Comparative distribution of salicylic acid and clobetasol propionate at 12 hours following application of clobetasol 0.05% and salicylic acid 6 % nano lotion and clobetasol 0.05% and salicylic acid 6 % conventional lotion.

Parameter (at 12 h)	Salicylic Acid		Clobetasol	
	Clobetasol 0.05% and salicylic acid 6 % nano lotion	Clobetasol 0.05% and salicylic acid 6 % conventional lotion	Clobetasol 0.05% and salicylic acid 6 % nano lotion	Clobetasol 0.05% and salicylic acid 6 % conventional lotion
Cumulative drug permeated (% permeated)	16.16%	4.50%	BDL	BDL
Drug remaining on skin surface (% unabsorbed)	0.763%	23.68%	14%	50%
Drug retained in skin (% skin retention)	83.08%	71.81%	86%	50%

For clobetasol propionate, although receptor phase concentrations remained below the detection limit, differences in drug distribution were observed. Clobetasol 0.05% and salicylic acid 6% nano lotion showed 86% of the applied clobetasol retained within the skin and 14% remaining on the skin surface, whereas clobetasol 0.05% and salicylic acid 6% conventional lotion demonstrated 50% retention within the skin and 50% remaining on the skin surface.

Overall, the study demonstrated higher permeation and dermal retention of salicylic acid from clobetasol 0.05% and salicylic acid 6% nano lotion compared with clobetasol 0.05% and salicylic acid 6 % conventional lotion. For clobetasol propionate, receptor phase concentrations remained below the detection limit for both formulations; however, clobetasol 0.05% and salicylic acid 6 % nano lotion exhibited greater retention within the skin and lower residual drug remaining on the skin surface.

DISCUSSION

This study compared the in vitro permeation and distribution of salicylic acid and clobetasol propionate from nano lotion and conventional lotion using excised swine skin and Franz diffusion cells. The experimental approach was based on the principles described in OECD test guideline 428 for in vitro skin absorption studies, which supports the evaluation of receptor-phase absorption and the distribution of test substances within the experimental system.[7]

Salicylic acid permeation increased progressively with time for both formulations. At 12 hours, the cumulative amount recovered in the receptor compartment was 0.969 mg (16.16% of the applied dose) for nano lotion and 0.270 mg (4.50%) for conventional lotion. The approximately 3.6-fold difference indicates greater receptor-phase delivery of salicylic acid from the nano formulation under the conditions evaluated. This finding is consistent with literature indicating that topical drug delivery is influenced by formulation composition, drug partitioning and interactions between the vehicle and the skin barrier.[5,6,9,11]

However, the present experiment does not establish which formulation characteristic was responsible for the observed difference. Research on nanoparticle-based delivery systems has highlighted that skin penetration and drug distribution vary according to the properties of the carrier, the active ingredient and the formulation as a whole.[9] Therefore, the higher permeation observed with Nano Lotion should be interpreted as a formulation-specific finding rather than evidence that all nano-based formulations necessarily improve skin delivery.

The distribution estimates y, calculated by mass balance, was 83.08% for Nano Lotion and 71.81% for Conventional Lotion. The corresponding surface-residue values were 0.763% and 23.68%. These results indicate that a greater proportion of the applied dose was assigned to the skin-retained fraction and a smaller proportion was recovered from the surface after Nano Lotion application. Nevertheless, mass-balance-derived retention should not be interpreted as a direct measurement of drug concentration in specific skin layers or as proof of delivery to a particular therapeutic target. OECD Test Guideline 428 provides a framework for assessing absorption and the distribution of a test substance across the relevant compartments of an in vitro skin absorption system.[7] The use of complementary analytical methods to quantify drug recovered from the skin would provide additional evidence to confirm the retention estimates obtained in this study.[12]

Salicylic Acid is used in topical preparations for its keratolytic action. By promoting the removal of accumulated keratin, it can modify the condition of the stratum corneum and may influence the penetration of other topical agents.[3,4] The higher permeation observed in this study is compatible with differences in formulation-mediated delivery, but the individual contributions of Salicylic Acid, the vehicle and the nano formulation cannot be distinguished from the present experimental design. Wester et al, investigated Salicylic Acid absorption using an isolated perfused porcine skin flap

model and compared the findings with human in vivo absorption. The authors reported absorption values of 7.5±2.6% in the porcine skin model and 6.5±5.0% in humans.[10] These findings provide relevant background for understanding salicylic acid absorption and the use of porcine skin in percutaneous absorption research. However, direct numerical comparison with the present study is not appropriate because the formulation, experimental model, exposure conditions and study duration differ

Clobetasol propionate was below the analytical detection limit in receptor-fluid samples from both formulations throughout the 12 hours study. This finding means that quantifiable receptor-phase permeation was not demonstrated under the experimental conditions; it does not establish that no drug crossed the skin. Drug transport depends on the physicochemical properties of the active ingredient, the vehicle, the skin barrier and the experimental conditions.[1,2,7]

The calculated distribution of Clobetasol Propionate differed between formulations. Nano Lotion was associated with an estimated skin-retained fraction of 86% and a surface residue of 14%, whereas Conventional Lotion showed corresponding values of 50% and 50%. These estimates were derived from the applied dose after accounting for receptor-phase recovery and surface residue. Previous reviews have emphasized that vehicle composition, drug–vehicle interactions and formulation characteristics can influence the relative amounts of an active ingredient that penetrate the skin or remain within it.[9,11] Direct extraction and quantitative analysis of skin-associated Clobetasol Propionate would help determine whether the estimated difference represents a reproducible difference in skin deposition.

The use of excised porcine skin provides a practical model for comparative percutaneous absorption testing. Nevertheless, differences between porcine and human skin, as well as variation in skin preparation and experimental conditions, may affect the translation of in vitro findings to clinical use.[7,8] Reviews of skin penetration methods have highlighted the importance of selecting appropriate experimental models and considering their limitations when interpreting results and extrapolating findings to humans.[12] Wester et al, also demonstrated the relevance of comparing results from porcine skin models with human absorption data, while illustrating that the experimental model must be considered when interpreting absorption estimates.[10] In addition, the present study was conducted using duplicate determinations for each formulation. The results should therefore be considered preliminary descriptive findings; the available replication does not support a robust assessment of variability or statistical significance.

Overall, Nano Lotion showed greater Salicylic Acid permeation, higher mass-balance-derived retention estimates for both active ingredients and lower surface residue than Conventional Lotion in this experiment. These observations suggest that formulation characteristics may influence the distribution of active ingredients between the skin surface, the estimated skin-retained fraction and the receptor compartment. The findings are consistent with published literature describing the influence of formulation composition and delivery-system characteristics on dermal and transdermal drug delivery.[9,11] However, the specific mechanisms responsible for the observed differences remain undetermined. Further studies using additional skin donors and replicates, validated analytical sensitivity and direct measurement of drug recovered from the skin would help confirm these observations and determine their relevance to topical treatment.

CONCLUSION

Clobetasol 0.05% and salicylic acid 6 % nano lotion exhibited significantly higher salicylic acid permeation compared to clobetasol 0.05% and salicylic acid 6 % conventional lotion at 12 hours (16.16% vs. 4.50%), along with greater cumulative permeation, higher dermal retention and lower unabsorbed surface residue, indicating improved skin delivery efficiency. Clobetasol Propionate was below the limit of detection in the receptor phase for both formulations throughout the study period, which is attributed to the low applied dose and analytical sensitivity and therefore reflects absence of measurable permeation under the experimental conditions rather than complete lack of transdermal transport. Overall, clobetasol 0.05% and salicylic acid 6 % nano lotion demonstrated superior dermal targeting and enhanced skin penetration and retention of active ingredients compared with clobetasol 0.05% and salicylic acid 6% conventional lotion, supporting its improved topical delivery performance.

Funding: No funding sources
Conflict of interest: None declared
Ethical approval: The study was approved by the Institutional Ethics Committee

REFERENCES

1. Prausnitz MR, Langer R. Transdermal drug delivery. Nat Biotechnol. 2008;26:1261-8.
2. Bouwstra JA, Honeywell-Nguyen PL. Skin structure and mode of action of vesicles. Adv Drug Deliv Rev. 2002;54(1):41-55.
3. US National Library of Medicine. Clobetasol propionate lotion, 0.05%: prescribing information. DailyMed. Available at: https://www.dailymed.nlm.nih.gov/dailymed/fda. Accessed on 21 March 2026.
4. Dogra S, Mahajan R. Topical therapy in psoriasis. Indian J Dermatol. 2016;61:643-8.
5. Prow TW, Grice JE, Lin LL, Faye R, Butler M, Becker W, et al. Nanoparticles and microparticles

for skin drug delivery. Adv Drug Deliv Rev. 2011;63:470-91.
6. Nastiti CMRR, Ponto T, Abd E, Grice JE, Benson HAE, Roberts MS. Topical nano and microemulsions for skin delivery. Pharmaceutics. 2017;9:37.
7. Organisation for Economic Co-operation and Development. Test No. 428: skin absorption: in vitro method. OECD Guidelines for the Testing of Chemicals, Section 4. Paris: OECD Publishing. 2004.
8. Simon GA, Maibach HI. The pig as an experimental animal model of percutaneous permeation in man: qualitative and quantitative observations—an overview. Skin Pharmacol Appl Skin Physiol. 2000;13:229-34.
9. Bolzinger MA, Briançon S, Chevalier Y. Nanoparticles through the skin: managing conflicting results of inorganic and organic particles in cosmetics and pharmaceutics. Wiley Interdiscip Rev Nanomed Nanobiotechnol. 2011;3:463-78.
10. Wester RC, Melendres J, Sedik L, Maibach H, Riviere JE. Percutaneous absorption of salicylic acid, theophylline, 2,4-dimethylamine, diethyl hexyl phthalic acid and p-aminobenzoic acid in the isolated perfused porcine skin flap compared to man in vivo. Toxicol Appl Pharmacol. 1998;151:159-65.
11. Otto A, du Plessis J, Wiechers JW. Formulation effects of topical emulsions on transdermal and dermal delivery. Int J Cosmet Sci. 2009;31:1-19.
12. Supe SM, Takudage PJ. Methods for evaluating penetration of drug into the skin: a review. Skin Res Technol. 2021;27:299-308.

Cite this article as: Sangle P, Sasane M, Barve S, Das S, Pinto N. In vitro comparative evaluation of skin permeation and dermal retention of clobetasol 0.05% and salicylic acid 6 % nano lotion and clobetasol 0.05% and salicylic acid 6 % conventional lotion using excised swine skin and Franz diffusion cells. Int J Res Dermatol 2026;12:633-9.