Use of Drug Kinetics in Dermis to Predict *in Vivo* Blood Concentration After Topical Application

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Purpose. To use the drug kinetics in dermis to predict the *in vivo* blood concentration after topical administration.

Methods. A two-step pharmacokinetic model was established. The first step was to calculate the drug input rate or flux from the skin to the systemic circulation using the drug kinetic parameters in dermis. These parameters include (a) distance over which the drug concentration declines by 50%, (b) drug concentration at the epidermal-dermal junction, and (c) minimal plateauing drug concentration in the muscle layer. These parameters were experimentally determined from the drug concentration-tissue depth profiles in the dermis, after the application of a topical dose of ddI (200 mg/kg) to rats. The second step was to use the drug input rate together with the systemic disposition pharmacokinetics of ddI in rats to predict the plasma concentration-time profiles. The model-predicted plasma concentration-time profiles were compared with the observed profiles, to determine the validity of the proposed pharmacokinetic model.

Results. The observed steady state concentration (C_{SS}) in individual animals (n = 6) deviated from the predicted values by 3 to 55% with 3 of 6 rats showing a <15% deviation. The mean observed C_{SS} of all animals deviated from the mean predicted values by less than 15%.

Conclusions. The close agreement between the observed and the model-predicted drug concentrations indicates that the systemic drug input can be calculated from the drug kinetics in the dermis.

KEY WORDS: 2′,3′-dideoxyinosine; distributed model; cutaneous absorption; prediction of *in vivo* plasma concentration; animal study.

INTRODUCTION

The skin is a heterogeneous tissue which consists of three distinct layers: the subcutaneous fat, the dermis, and the outer most epidermis layer which can be further divided into the stratum corneum and the viable epidermis (1). Guy et al (2) proposed a linear pharmacokinetic model, in which the drug transport through skin is described as a process of drug partitioning and diffusion from the transdermal device across stratum corneum, viable epidermis and dermis to reach the systemic circulation. This model considers the dermis and viable epidermis as a homogeneous tissue compartment, and has been applied to predict *in vivo* plasma concentrations for transdermal delivery of clonidine, nitroglycerin, estradiol, scopolamine, and timolol (2).

The skin receives one third of the total blood circulation. The dermis constitutes the majority of the skin mass, contains a dense blood and lymphatic vasculatures, and is the site of drug absorption into the systemic circulation (1). A reduction of blood flow by a prior intravenous nicotine dose altered the absorption of a topical nicotine dose: the time to reach the maximum concentration was increased by 60%, and the maximum concentration and area under the plasma concentration-time curve was reduced by 16% and 19%, respectively (3). Variation in cutaneous blood flow within its physiologic range has been reported to have a major effect on the determination of flux and total absorption of caffeine and benzoic acid across human and nude rat skin in vivo (4). Riviere and Williams used simulation data to show that absorption of malathion increases with the capillary volume (5). Singh and Roberts, using tissue distribution profiles of radioactive microsphere in skin to measure the skin blood flow, demonstrated that blood flow can affect tissue distribution and absorption of radioactive microsphere in skin (6). Because of the demonstrated importance of dermal blood flow, inclusion of drug removal by blood perfusing the dermis in the diffusion model such as the one proposed by Guy et al. (2) is likely to improve the prediction of in vivo plasma concentrations.

A distributed model for drug penetration in capillary perfused tissue was first proposed by Dedrick and coworkers (7). This model assumes drug transfer in the interstitial space by diffusion and drug removal by capillary drainage. Omission of drug removal by perfusing blood in dermis reduces this model to a simple diffusion model. The validity of the distributed model in capillary perfused tissue was first verified by our studies which examine the penetration of mitomycin C in the well-perfused dog bladders (8), and the penetration of 2′,3′-dideoxyinosine (ddI) in rat dermis (9). The present study is to determine if the drug kinetics in the dermis can be used to derive the flux from skin to systemic circulation and hence to predict the *in vivo* systemic drug concentration. In this study, the rat was used as the animal model and ddI was used as the model drug.

MATERIALS AND METHODS

Chemicals

ddI (Lot # 234-b-1) was provided by the National Institute of Health (Bethesda, MD). The internal standard, ftorafur (N^1 -(2-tetrahydrofuranyl)-5-fluorouracil), was a gift from the Mead Johnson Research Laboratory (Evansville, Indiana). All high pressure liquid chromatographic (HPLC) solvents and reagents were of analytical grade and purchased from Fisher Scientific Company (Cincinnati, Ohio). The purities of ddI and ftorafur, analyzed by HPLC, were 99%.

Animal Protocol

Our previous study demonstrated a nearly identical cutaneous absorption of ddI in rats with high and low follicular density, indicating that ddI was mainly absorbed by transcellular and/or paracellular routes. The present study used rats with high follicular density, i.e., female Fisher rats.

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These rats, 5-6 months old, were purchased from Charles River Breeding Laboratories, Kingston, New Jersey. An indwelling catheter was implanted in the right jugular vein under ether anesthesia one day before an experiment. Each animal was given a ddI dose of 200 mg/kg, or about 40 mg. The ddI dose was prepared on the day of experiment. The drug was dissolved in 200 μl saline and dispersed in 1 g o/w ointment (Unibase). The ddI dose was applied to a 4×2 cm^2 area on the dorsal interscapular surface as described previously (9). The area adjacent to the application region was covered by masking tape to limit the spread of the applied dose. Serial blood samples were taken through the jugular catheter before dosing and at regular intervals for up to six hr. After the last blood sample was taken, the rat was euthanized and the skin was rinsed with 1 ml methanol twice to remove the residual dose. The skin corresponding to the application area was then removed and immediately frozen.

Sample Analysis

The skin tissue samples were analyzed using previously described procedures (10). Briefly, the frozen skin was trimmed to 1.5×1.5 cm^2, and then cut by cryotome (Reichert American Optical, Buffalo, New York) into 50 μm thick sections, which is the optimal thickness for cutting. Two sections were combined and weighed. The weighed tissue samples were combined with 1 ml of acetonitrile and 400 μl of ftorafur in water (4 μg/ml), vortexed, and homogenized with a rotor/stator type homogenizer (Biospec Products Inc., Bartlesville, Oklahoma). After centrifugation, 1 ml of the clear supernatant was transferred and evaporated to 50 μl. Plasma samples were extracted using solid phase extraction. The skin and plasma extracts were analyzed by HPLC as described previously, using a reversed phase μBondapak C_{18} column (waters Associates, Milford, Massachusetts) and an aqueous mobile phase containing 10 mM sodium phosphate buffer, pH 6.9, and 4% acetonitrile (9).

Frozen section of whole skin was examined microscopically. The thickness of stratum corneum, viable epidermis, dermis were determined to be 8 ± 2 μm, 12 ± 2 μm, and 1450 ± 50 μm (mean±S.D., n = 6). The calculation of drug flux required only the drug concentration within the dermis including the drug concentration in the interface between epidermis and dermis (see below), and did not require the measurement of drug concentration in stratum corneum and epidermis. Hence, the first 50 to 100 μm of skin sections containing the stratum corneum and epidermis were discarded, whereas the sections corresponding to the dermis were analyzed.

Analysis of ddI Tissue Concentration-Depth Profiles Using the Distributed Model

In the distributed model (7), the decline of drug concentrations with respect to the penetrated tissue depth is described by the equation:

$$C_x = (C_0 - C_b)\, e^{\frac{-0.693}{w_{1/2}}\cdot(x-20)} + C_b,$$
$$\text{where } w_{1/2} = 0.693\sqrt{\frac{(pa+q)D}{paq}} \qquad (1)$$

where C_0 is the concentration at the beginning of the capillary perfused tissue, which in the rat skin is the epidermal-dermal junction (about 20 μm deep by microscopic examination); C_b is the drug concentration in the deepest tissue layer; C_x is the concentration at distance x into the capillary tissue; $w_{1/2}$ is the thickness of the tissue over which the concentration declines by one half. As depicted, $w_{1/2}$ is a function of p and a (microscopic permeability coefficient and surface area of the capillaries), D (diffusivity of the drug) and q (capillary blood flow rate per unit tissue volume). Changes in these parameters are reflected by changes in $w_{1/2}$.

The analysis of the tissue concentration-depth profiles by equation 1, using nonlinear least squares estimation of SAS/STAT software (SAS Institute Inc., Cary, North Carolina), gave the three parameters C_0, C_b and $w_{1/2}$.

For a simple diffusion process, the tissue concentration-depth profiles are described by the following equation:

$$C_x = C_0 - k_0 x \qquad (2)$$

where C_0 and C_x are defined as before, and k_0 is the zero order rate constant describing the decline of tissue concentration with depth.

The statistical criteria used to compare the goodness-of-fit by equations 1 and 2 were the Akaike Information Criterion (AIC), Schwartz criterion (SC), and F-test (10).

Calculation of ddI Flux from Skin to Systemic Circulation

The ddI flux from skin to systemic circulation is via its absorption into the perfusing capillaries, and can be derived from the distributed model as follows:

$$Flux = D(C_0 - C_b)\frac{0.693}{w_{1/2}}\left(1 - e^{\frac{-0.693}{w_{1/2}}(l_0-20)}\right)\frac{A}{BW} \qquad (3)$$

where D, C_0, C_b and $w_{1/2}$ are defined as before, A is the effective surface area of ddI transport through the dermis, which is equal to the application area. BW is the rat body weight, which averaged 220 ± 10 g (mean±S.D., n = 6). l_0 is the thickness of the dermal layer.

ddI has a MW of 236 and a pK_a of 9.0. The diffusivity of small water soluble molecules in a homogeneous aqueous system at 37°C, D_{37^0}, was calculated as follows (11):

$$D_{37^0} = 10.18(MW)^{-0.45}\times10^{-5}(cm^2\text{sec}^{-1}) \qquad (4)$$

The relationship between the D_{37^0} and D in a heterogeneous tissue is described by the following empirical equation (12).

$$D = k\, D_{37^0} \qquad (5)$$

where k is a constant, and varies within a narrow range of 0.03 - 0.05 for water soluble compounds in rat diaphragm (12). The k value for sucrose which, similar to ddI, contains a sugar moiety and is water soluble, is 0.036. Assuming that the diffusion of ddI in rat dermis approximates the diffusion of sucrose in rat diaphragm, the D of ddI in dermis was calculated using equations 5 and 6, and a k value of 0.036.

Note that C_0, C_b, $w_{1/2}$, BW, A and l_0, were experimentally determined, whereas D was calculated using empirical equations. The D term is more likely to be error-prone than the other parameters.

Prediction of ddI Plasma Concentration-Time Profiles Using Cutaneous Absorption Kinetic Data and Disposition Kinetic Data

We previously showed that the disposition of ddI is adequately described by a two compartment model (13). Because the ddI plasma concentration-time profiles showed a pseudo steady state condition from 1 to 4 hr (see Results). We used the following equation to calculate the ddI plasma concentration-time profiles (14):

$$C_p = flux \times \frac{k_{21} - \alpha}{V_1(\alpha - \beta)\alpha}(e^{-at} - 1) - flux \times \frac{k_{21} - \beta}{V_1(\alpha - \beta)\beta}(e^{-\beta t} - 1) \quad (6)$$

where ddI flux is as calculated from equation 3, C_p is plasma concentration, and t is the time during which a topical dose is applied to skin. The present study used only topical administration and did not use intravenous injection. Therefore, pharmacokinetic parameters depicted in equation 6 needed to calculate the C_p-time profiles were obtained from data of a previous study. The six sets of pharmacokinetic parameters, V_1 (apparent volume of distribution), k_{21} (intercompartmental transfer rate constant), α and β (macro constants) were obtained from six rats that were given an intravenous radio-labelled tracer ddI dose concomitantly administered with the topical dose of 200 mg/kg unlabeled ddI (13). The values of these parameters ranged from 152 to 726 ml/kg for V_1, 0.0223 to 0.0319 min^{-1} for k_{21}, 0.1611 to 0.2096 min^{-1} for α, and 0.01813 to 0.02451 min^{-1} for β. To generate the lower and upper boundaries, and the average C_p-time profiles, we used these six sets of parameters together with the ddI flux from the dermis from individual animals determined in the present study. This exercise resulted in six predicted plasma concentration-time profiles for each animal. The profiles that showed the highest and lowest values were considered as the upper and lower boundaries of the predicted values. The average of the six profiles gave the mean predicted plasma concentration-time profiles for each animal.

Systemic bioavailability of the topical dose was calculated as follows:

$$Bioavailability = \frac{AUC_{0\text{-}360\ min} \times Clearance}{Dose} \quad (7)$$

where $AUC_{0\text{-}360\ min}$ is the area under the plasma concentration-time curve from time zero to the last sampling time of 360 min.

RESULTS

Mass Balance

Washing the dose application area with methanol at the end of the experiment recovered 44.5±5.7% of the dose. The fraction of dose found in dermis was 24.5±4.6%. The calculated systemic bioavailability was 20.2±4.4%. Hence, about 90% of the applied dose has been accounted for, implying that about 10% of the applied dose remained in the stratum corneum and viable epidermis at 360 min. Note that the bioavailability calculated using $AUC_{0\text{-}360\ min}$ is a conservative estimate because the drug concentration was at a steady state at 360 min.

ddI Concentration-Depth Profiles in Dermis

Figure 1 shows the mean ddI tissue concentration-dermis depth profiles in six rats. In all six rats, the tissue concentration declined exponentially from 100 μm to about 1,000 μm, and then declined at a slower rate. This was in agreement with our previously reported data (9) and with the distributed model which predicts a biphasic concentration decline with respect to tissue depth (7). Analysis of the tissue concentration-depth profiles by the distributed model and the simple diffusion model (Figure 1) showed a better fit for the distributed model as indicated by the high F value, the lower AIC and SC values and the lower weighed square sum of error (data not shown).

ddI Flux from Skin to Systemic Circulation

Table I outlines the tissue pharmacokinetic parameters and the ddI flux in individual rats. The data showed a relatively constant C_0 with a maximum fariation of <67%, indicating a relatively uniform drug partitioning across dhe stratum corneum. The $w_{1/2}$ showed a 2.1 fold range, the C_b a 3 fold range and the ddI flux a 2.3 fold range. $w_{1/2}$ describes the rate of drug removal by capillaries, and is dependent on the product of the capillary permeability and surface area, pa, and the blood flow q. Changes in pa and q will alter the drug removal by capillaries, the slope in the concentration decline across the dermis and, hence, the $w_{1/2}$. The variation in the $w_{1/2}$ values and the resulting calculated flux may be due to the variability in pa and q in individual animals.

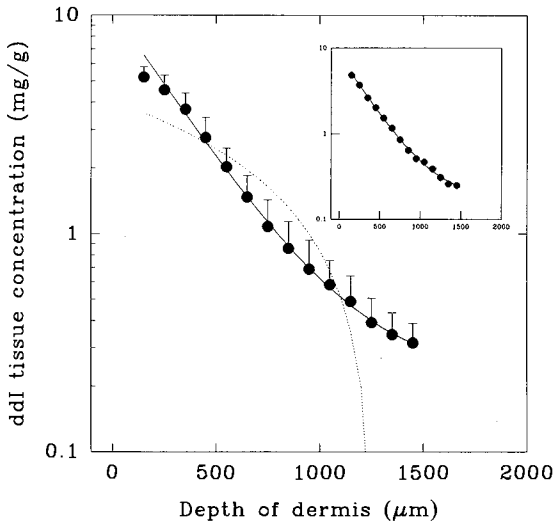

Fig. 1. ddI tissue concentration-dermis depth profiles in rats given a topical dose of 200 mg/kg. Data points with bars are mean + one S.D. of the experimentally determined ddI tissue concentrations of six rats. Solid lines represent the best fit lines according to the distributed model and dashed lines the simple diffusion model. Inset shows the ddI concentration-dermis depth profile in a representative animal.

Table I. Drug Kinetics in the Dermis

Rat #	C_0 (mg/g)	C_b (mg/g)	$w_{1/2}$ (μm)	Flux (μg/min-kg)
1	6.88	0.20	212.1	146.7
2	6.72	0.38	168.1	181.2
3	7.69	0.30	225.1	160.1
4	5.56	0.25	260.5	97.5
5	4.61	0.21	236.2	102.4
6	5.51	0.13	360.5	79.5
Mean ± S.D.	6.16 ± 1.00	0.25 ± 0.08	244.7 ± 64.9	127.9 ± 40.4

Concentrations of ddI in the dermal tissue as a function of depth were analyzed using equation 1 to obtain the values of half width ($w_{1/2}$), tissue equilibrating concentration (C_b) and the extrapolated concentration at the epidermal-dermal junction (C_0). The flux of ddI from skin to systemic circulation was calculated using equation 3.

Predicted ddI Plasma Concentration-Time Profiles Using Drug Kinetic Data in Dermis

The different flux of ddI from skin to systemic circulation in individual animals resulted in different predicted plasma concentration-time profiles. Figures 2A and 2B show the upper and lower limits, and the average of the predicted plasma profiles in two animals, respectively. As indicated below, these are the two animals that showed the greatest and the least deviations among the predicted and observed concentrations. The highest and lowest predicted steady state concentrations in individual animals ranged from 1.51 to 3.25 μg/ml and from 0.66 to 1.42 μg/ml, respectively. The average predicted steady state plasma concentrations in individual animals ranged from 1.03 to 2.30 μg/ml.

The mean of the average predicted profiles of the six rats is shown in Figure 3. The mean predicted steady state plasma concentrations was 1.66 μg/ml.

Observed ddI Plasma Concentration-Time Profiles

Figure 2A and 2B also show the observed profiles in two individual rats and Figure 3 the mean observed profiles of all six rats. The ddI concentration was maintained at a steady state for at least 3 hr, after which time the concentration decreased by about 11%. The observed steady state plasma concentration in 6 rats ranged from 1.15 to 1.91 μg/ml, and the mean value was 1.44 μg/ml.

Comparison of Predicted and Observed ddI Plasma Concentration-Time Profiles

Two comparisons of the observed and predicted concentrations were performed, i.e., individual profiles for each of six animals, and the mean profile for the whole group. For individual animals, the observed steady state concentrations were within the upper and lower limits of the predicted values, with one rat showing the maximum deviation from the average predicted value by less than 55%, two rats showing a 25-30% deviation, and 3 of 6 rats showing a <15% deviation. Data shown in Figure 2A are from one rat where the predicted steady state concentration showed the least deviation (i.e., 3%), whereas Figure 2B shows the data from another rat which had the greatest deviation (i.e., 55%). Therefore, a comparison of data in Figure 2A and 2B gives an appreciation of the best and the worst scenario of the goodness-of-fit between the predicted and the observed values.

A comparison of the mean observed profiles with the mean of the average predicted profiles in the six rats shows a 15% lower value for the observed steady state concentra-

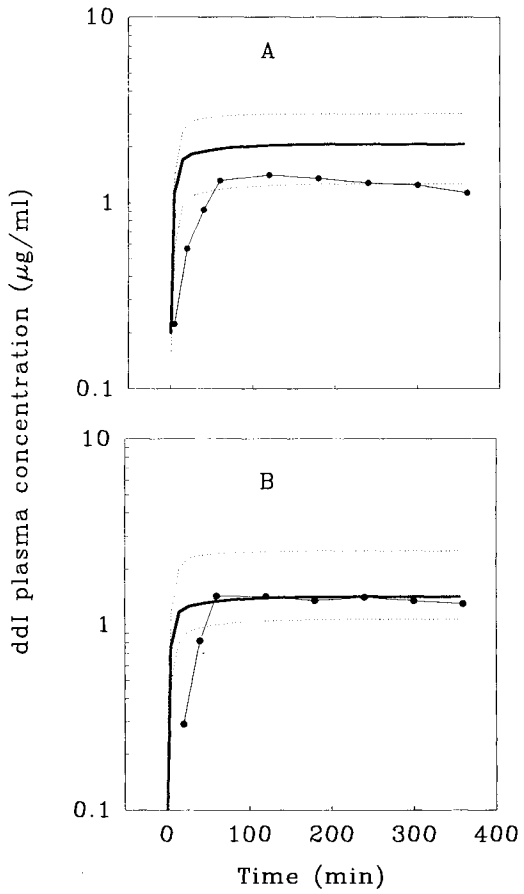

Fig. 2. ddI plasma concentration-time profiles. Data points connected by thin solid lines are the experimentally determined ddI plasma concentrations. Dashed lines represent the lower and upper boundaries of the predicted profiles, and the heavy solid lines represent the average profiles. The two rats that gave the greatest and the least deviation among the predicted and the observed steady state concentration are shown in top and bottom panels, respectively.

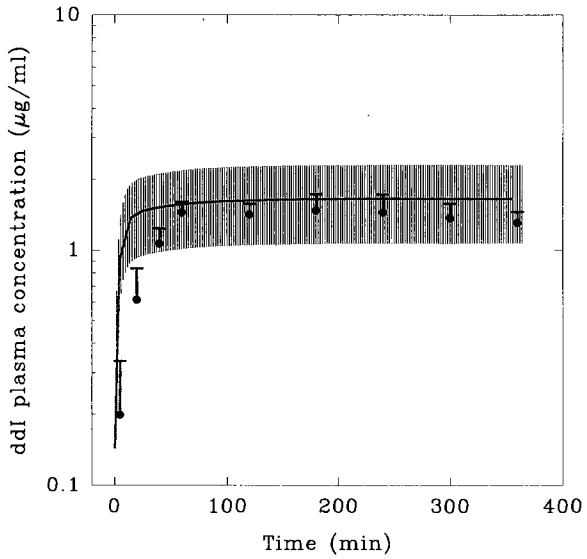

Fig. 3. Comparison of the mean of the predicted ddI plasma concentration-time profiles and the mean of the observed profiles. Data points with bars are mean + one S.D. of the observed profiles of six rats. Solid line is the mean of the average predicted profiles of six rats and shaded area represents boundaries of the mean of the predicted profiles.

tions (Figure 3). The model predicted that the steady state would be reached between 35-50 min, whereas the experimental data showed that the steady state was reached at about 60 min.

DISCUSSION

In Vivo Cutaneous Absorption Studies

Most studies of cutaneous drug absorption have been done using in vitro models such as the skin flap model where the skin is mounted on a diffusion cell and the drug transfer from the donor chamber to the receiver chamber is measured. While these studies are useful to measure the permeability of intact skin, they have two deficiencies. Firstly, because there is no blood supply, the effect of blood flow on drug absorption kinetics cannot be evaluated. Secondly, *in vitro* diffusion studies are done under sink condition where the drug concentration in the receiver chamber approaches zero and/or is insignificant compared to the concentration in the donor chamber. An analogous situation under *in vivo* conditions is an insignificant drug concentration at the epidermis/dermis interface. This would require that the drug, once diffused across the stratum corneum, is rapidly and completely removed by capillaries. Data of the present study demonstrated that C_0 at the epidermis/dermis interface was about 6 mg/g, or 20% of the concentration in the ointment. The high and significant C_0 concentration invalidates the sink condition assumption used in *in vitro* studies. This problem is more pronounced when absorption enhancers are used, because C_0 will increase due to the reduced barrier function of the stratum corneum.

Validity of the Proposed Model

In the previously described model used to predict the *in vivo* plasma concentration after a topical application, the three layers of the skin, i.e. stratum corneum, epidermis and dermis, are considered as a homogeneous absorption barrier; the drug penetration in these layers is described by a simple diffusion process resulting in a linear drug concentration decline in dermis (2). There is evidence that the drug kinetics in the dermis do not follow the zero order diffusion model. Schaefer and Zesch (15) and our laboratory (9) have shown an exponential concentration decline in the dermis for vitamin A and ddI, respectively. This is further confirmed by the data in the present study. The exponential decline indicates first order drug removal processes in tissue, as described in the distributed model (7). The distributed model includes drug transport and absorption by perfusing blood as well as drug diffusion, and is likely to be more physiologically relevant compared to the simple diffusion model.

Data of the present study show that the model-predicted steady state plasma concentrations are within 15% of the observed values. The close agreement between the observed and the model-predicted drug concentration-time profiles indicates that the drug input from the skin to systemic circulation can be accurately calculated from drug kinetics in the dermis, and further supports the distributed model.

In all animals, the observed rate to reach the steady state was slower than the predicted values. A similar finding was reported by Guy et al. (2). In our model, the flux used in equation 8 was calculated using the assumption of an instantaneous drug input to the systemic circulation, and does not take into account the time required to establish the steady state concentration gradient in the skin. Accordingly, the model can be refined by incorporating a function that describes the establishment of a steady state drug flux from stratum corneum to the systemic circulation and, in the case of significant depletion of the dose due to absorption, by incorporating a mass balance term to account for dose depletion.

Summary

In summary, data of the present study highlight the validity of the distributed model in describing cutaneous drug absorption and the need of using *in vivo* models to study the absorption kinetics. We have also described a model of drug absorption in dermis which incorporates drug removal by perfusing blood and depicts the mathematical relationship between the blood flow to skin, drug diffusion in interstitial space, capillary permeability and surface area, and the drug input rate. The physiological relevance of the model should facilitate studies of cutaneous drug absorption, especially evaluations of the effects of altering the regional blood flow, physicochemical properties of drug, and/or capillary permeability. Further studies are needed to establish the quantitative relationship between blood flow and the rate and extent of drug absorption.

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REFERENCES

1. A. C. Williams and B. W. Barry. Skin absorption enhancer. *Critical Rev. Therap. Drug Carrier Systems* 9:305–353 (1992).
2. R. H. Guy and J. Hadgraft. Mathematical models of percutaneous absorption. In R. L. Bronaugh and H. I. Maibach (eds.), *Percutaneous absorption*, Marcel Dekker, Inc. New York, 1989, pp. 13-26.
3. N. L. Benowitz, P. Jacob, P. Olsson, and C.-J. Johansson. Intravenous nicotine retards transdermal absorption of nicotine: Evidence of blood flow-limited percutaneous absorption. *Clin. Pharmacol. Ther.* 52:223-230, 1992.
4. L. K. Pershing, S. Huether, R. L. Conklin, and G. G. Krueger. Cutaneous blood flow and percutaneous absorption: a quantitative analysis using a laser doppler velocimeter and a blood flow meter. *J. Invest. Dermatol.* 92:355-359 (1989).
5. J. E. Riviere and P. L. Williams. Pharmacokinetic implication of changing blood flow in skin. *J. Pharm. Sci.* 81:601-602 (1992).
6. P. Singh and M. S. Roberts. Blood flow measurements in skin and underlying tissues by microsphere method: application to dermal pharmacokinetics of polar nonelectrolytes. *J. Pharm. Sci.* 82:873-879 (1993).
7. M. F. Flessner, R. L. Dedrick, and J. S. Schultz. A distributed model of peritoneal-plasma transport analysis of experimental data in the rat. *Am. J. Physiol.* 241:F413-F424 (1985).
8. M. G. Wientjes, J. T. Dalton, R. A. Badalment, J. R. Drago, and J. L.-S. Au. Bladder wall penetration of intravesical mitomycin C in dogs. *Cancer Res.* 51:4347-4354 (1991).
9. E. Gupta, M. G. Wientjes, and J. L.-S. Au. Penetration kinetics of 2′,3′-dideoxyinosine in dermis is described by the distributed model. *Pharm. Res.* 11:809-815 (1994).
10. B. P. Imbimbo, P. Martinelli, M. Rocchettit, G. Ferrari, G. Bassotti, and E. Imbimbo. Efficiency of different criteria for selecting pharmacokinetic multiexponential equations. *Biopharm. Drug Disp.* 12:139-147 (1991).
11. J. S. Schultz and W. Armstrong. Permeability of interstitial space of muscle (rat diaphragm) to solutes of different molecular weights. *J. Pharm. Sci.* 67:696-705 (1978).
12. E. M. Renkin. Multiple pathways of capillary permeability. *Circ. Res.* 41:735-751 (1977).
13. E. Mukherji. Delivery and pharmacodynamics of dideoxynucleosides. *Doctoral Dissertation.* Ohio State University, Columbus, Ohio (1993).
14. M. Gibaldi and D. Perrier. *Pharmacokinetics*, Marcel Dekker, Inc., New York, 1975.
15. H. Schaefer and A. Zesch. Penetration of vitamin A into human skin. *Acta. Derm. Venereol.* 74(c):50-55 (1975).