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Mathematical models for skin toxicology

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Abstract

Introduction: Our skin is daily exposed to substances many of which are neutral and safe while others are potentially harmful. In order to estimate of the degree of toxicity and damage to skin tissues when exposed to harmful substances skin toxicology studies are required. If these studies are coupled with suitably designed mathematical models it can provide a powerful tool which allows appropriate interpretation of data. This work reviews mathematical models that can be employed in skin toxicology studies.

Areas covered: Two types of mathematical models and their suitability to assessing skin toxicology are covered in this review. The first type is focused on predicting penetration rate through skin from solute's physicochemical properties, whereas the second type models transport processes in skin layers using appropriate equations with the specific aim to predict the concentration of a given solute in viable skin tissues.

Expert opinion: Mathematical models are an important tool for accurate valuation of skin toxicity experiments, estimation of skin toxicity and developing new formulations for skin disease therapy. Comprehensive mathematical models of drug transport in skin, especially those based on more physiologically detailed mechanistic considerations of transport processes, are required to further enhance their role in assessing skin toxicology.

Keywords: dermal drug transport, skin toxicology, mathematical modelling, QSAR

Highlights:

- Skin toxicology is an important area of research which benefits from mathematical modelling.
- Two types of mathematical models: predicting penetration rate through stratum corneum from physicochemical properties and describing transport in skin layers are reviewed.

- The emphasis in this work is on simpler models that can predict concentration in viable skin layers which is central in determining skin toxicology.
- Further experimental and mathematical modelling efforts are required to assess penetration rate from realistic composite vehicles.
- Better elucidation of skin physiology is necessary to improve applicability of current mathematical models to assessing skin toxicology.
- Mathematical models that describe penetration though diseased and compromised skin need to be developed.

1. Introduction

Skin is the largest organ of the human body with large potential to be exposed to environmental toxins and given the prevalence of skin disease (e.g. approximately one-third of US population has one or more significant skin condition [1] that requires treatment) this makes the toxicology of skin an important area of research. Last 30 years have seen a significant progress in available experimental approaches to skin toxicology. An important step was the development by Riviere's group of the isolated perfused porcine skin flap (IPPSF) model [2, 3] which allows *in vitro* cutaneous pharmacology and toxicology studies in physiologically relevant setting with animal skin which best approximates human skin in terms of it permeability properties [4]. This experimental model was further developed by the group to study pharmacokinetics of various solutes [5] characterization of skin toxicity [6] evaluation of protective effects of various compounds [7] and study skin diseases mediated by integrins [8]. The use of human in vitro skin models in combination with cultures of keratinocytes [9] to assess dermal risks as an alternative to animal models have been proposed and assessed [10]. Another significant development is the use of reconstructed human epidermis as an alternative to excised human and animal skin in toxicology studies [11]. *In vivo* human studies using non-invasive techniques, such as fluorescent drug concentration measurements [12], could be practical in some toxicology studies. This variety of experimental approaches increases the role of mathematical analysis and models in skin toxicology.

Mathematical models of epidermal and dermal transport processes are important for the estimation of dermal exposure to drugs which is needed for assessing their toxicity. These models can potentially aid directly by providing information on the rate of drug penetration

through the skin and on the dermal concentration of drugs. The models are also useful in deeper analysis of experimental data, often allowing reduction in the number of experiments

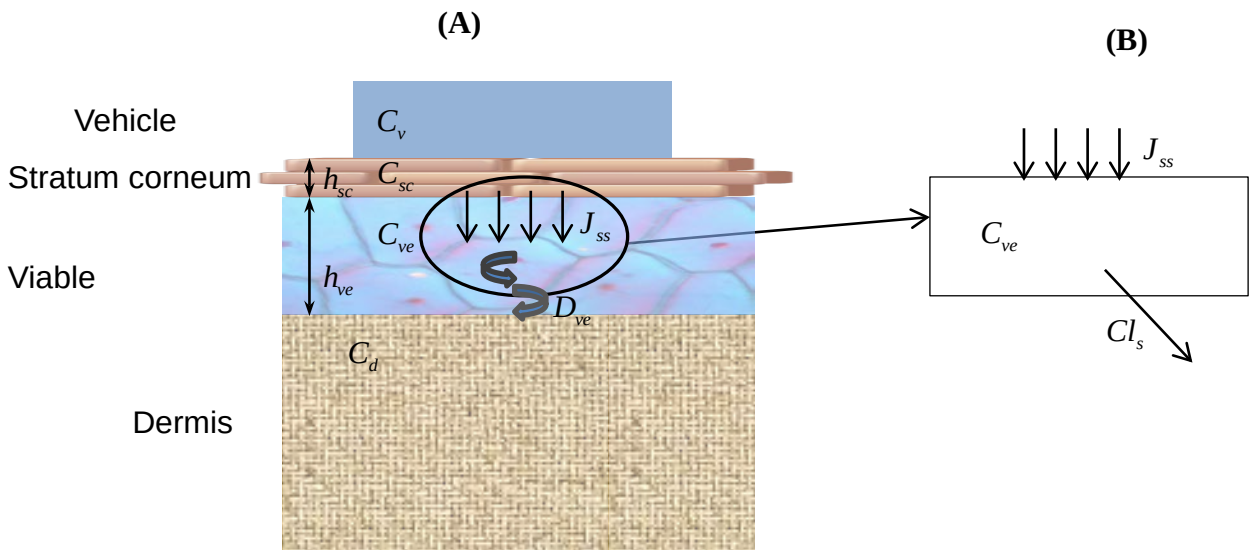

Fig 1. Schematic representation of skin layers (A) and simplified representation of the viable epidermis as a compartment (B).

and helping to interpret the *in vitro* experiments relative to the ones performed under *in vivo* conditions. It is therefore somewhat surprising that Web of Science search of terms: skin, toxicology and mathematical modelling in "Title/Keywords/Abstract" returned no references, it is perhaps due to an association of the mathematical modelling of skin absorption mostly with the transdermal drug delivery area. It seems appropriate, therefore, to compile a review on mathematical models that can aid to the assessment or convey a better understanding of the toxicology of skin. This task is helped by resent reviews on mathematical modelling of drug transport processes in skin [13, 14] and skin permeability [15]. In this work predicting drug penetration rate through skin will be first discussed and reviewed as the penetration rate is the key determinant of concentration of the drug in skin tissues. The concentration in viable layers of skin in turn is central in elucidating skin toxicity. The mathematical modelling of transport processes in deeper skin layers are reviewed with emphasis towards determining the concentration of drugs in viable skin layers.

2. Predicting permeation rate through skin from drug's physicochemical properties

When skin is exposed to a drug or solute (e.g. a cream/ointment is applied or environmental exposure to toxins happens, see Fig. 1A) partitioning into the stratum corneum (SC) and diffusion in it occur that results in flux through the SC, which in turn increases concentration

of drug in the viable epidermis. This concentration is crucial in determining skin toxicity. The process of transport though the SC is time dependent at first and is described by diffusion equation (see next section). In many cases the concentration in the SC and flux through the SC become time independent, or, in mathematical terms, the steady state is reached. This steady state transport process through the SC is often most relevant to assessing skin toxicology (as is determines the maximum concentration in the viable epidermis) and an important parameter is the steady state flux of solute through the skin: [16]

$$J_{ss} = \frac{D_{sc}\Delta C_{sc}}{h_{sc}} \tag{1}$$

where D_{sc} is the diffusion coefficient in the SC, h_{sc} is it's thickness and ΔC_{sc} is the concentration difference between outer and inner SC layers. Generally, due to blood clearance in the dermis, the concentration in the inner layer of the SC is much less than in the outer layer and

$$\Delta C_{sc} \approx C_{sc} = K_{sc} C_v \tag{2}$$

where C_{sc} is the concentration in the outer SC layer, K_{sc} is the partition coefficient between the SC and the vehicle and C_v is the drug concentration in the vehicle. Equation (1) can be expressed then as:

$$J_{ss} = \frac{K_{sc} D_{sc}}{h_{sc}} C_v = k_p C_v \tag{3}$$

where k_p is the permeability coefficient of the SC. The equation (3) for the steady-state flux applies for times t greater than the lag time (practically three lag times, where $lag = h_{sc}^2 / (6 D_{sc})$).

The important parameter for systemic toxicology is the total amount of solute penetrated (Q), as this determines systemic concentration of the drug. The steady-state approximation of the total amount of solute penetrated at time t after the application of the vehicle with concentration C_v to the area of skin (A) is given by: [17]

$$Q(t) = k_p A C_v (t - lag) \tag{4}$$

This equation could be used to estimate systemic absorption of solute for the case of large vehicle and long exposure time ($t \geq 3\ lag$).

The most relevant parameter for skin toxicology is arguably the maximum concentration in the viable layers of skin. This concentration occurs in the top layer of the viable epidermis (C_{ve}). More complex mathematical models that describe concentration in skin layers under the SC will be presented in the next section, but it is sufficient here to consider simple compartmental approach (see Fig 1B) to describe this concentration in the viable layers of skin. Assuming a constant clearance of drug from skin (Cl_s) yields [18, 19]:

$$C_{ve} = \frac{k_p A C_v}{Cl_s + k_p A K_{ve}/K_{sc}} = \frac{A J_{ss}}{Cl_s + k_p A K_{ve}/K_{sc}} \tag{5}$$

where K_{ve} is the partition coefficient between the viable epidermis and the vehicle. The blood clearance in the skin is generally much faster than penetration through the SC ($Cl_s \gg k_p A$) and equation (5) can be simplified to:

$$C_{ve} = \frac{k_p A C_v}{Cl_s} = \frac{A J_{ss}}{Cl_s} \tag{6}$$

It follows from this equation that the concentration in the viable layers of skin is directly proportional to the permeability coefficient or the steady state flux through the SC of a given drug. The maximum possible concentration in the viable epidermis $C_{ve}^{\max}$ is reached when concentration in the vehicle reaches its solubility limit (S_v):

$$C_{ve}^{\max} = \frac{k_p A S_v}{Cl_s} = \frac{A J_{\max}}{Cl_s} \tag{7}$$

This equation demonstrates why predicting the permeability coefficient or maximum flux from drug's physicochemical properties is an important area of skin toxicology.

There are significant uncertainties in predicting skin permeability (represented here in cm/h) from aqueous solution through human SC using drug physicochemical properties and Potts and Guy regression [20]

$$\log k_p = 0.71 \log K_{oct} - 0.0061 MW - 2.74, \qquad r^2 = 0.69, \quad n = 93 \tag{8}$$

remains generally acceptable for over two decades. In equation (8) *MW* is molecular weight and K_{oct} is octanol/water partitioning coefficient. The quantities K_{sc}, D_{sc}, and therefore k_p represents the average of a large range of relations and interactions that can influence how a molecule partitions and diffuses through the SC. Different molecules take different routes through the SC namely, transcellular, intercellular and transappendageal routes, as well as

different pathways through the intercellular lipids [15, 16, 21, 22]. Finding one equation through which to mathematically model such diverse phenomena generally leads to an over-simplification, but this at least allows us to draw general conclusions and provides some tool to predict the rate of penetration through the skin without doing the experiments.

Other significant development in quantitative structure-activity relationship (QSAR) for skin penetration was the use of molecular volume (*MV*, in cm^3/mole) instead of molecular weight and addition of the melting point (*Mp*, in ^oC) of the solute as the predictive parameters [23]:

$$\log k_p = 0.820 \log K_{oct} - 0.00933MV - 0.0387Mp - 2.355, \qquad r^2 = 0.885, \quad n = 60 \quad (9)$$

Although the regression (9) appears to be significantly more accurate than (8) ($r^2 = 0.89$ vs $r^2 = 0.69$) it needs to be noted that some of this was achieved by reduced number of solutes considered in the regression (*n*=60 vs *n*=93). Another factor in determining whether to use equation (8) or (9) is the ease with which the predictive parameters in equations can be obtained. Molecular weight in equation (8) is more readily available compared to the molecular volume and the melting point in equation (9).

A solvatochromic approach based on solute structure parameters such as an excess molar refraction (R_2), the dipolarity/polarizability (π_2^H), the effective hydrogen-bond acidity ($\sum \alpha_2^H$) and basicity ($\sum \beta_2^H$) and the McGowan characteristic volume (V_x) has been developed [24, 25]:

$$\log k_p = 0.437R_2 - 0.410\pi_2^H - 1.631\sum \alpha_2^H - 3.286\sum \beta_2^H + 2.012V_x - 1.685, \quad r^2 = 0.957, \quad n = 47 \quad (10)$$

While the quality of the regression is good ($r^2 = 0.96$), the above equation contains 5 descriptors (as compared with only 2 in Potts and Guy regression (8)) and is based on only 47 solutes. Solvatochromic approach was reconsidered and it was concluded that the coefficients of $\sum \alpha_2^H$ and $\sum \beta_2^H$ strongly depend on the classes of solutes included into the regression [26]. This would not be expected if hydrogen-bond acidity and basicity reflected the fundamental determinants of the skin permeability coefficient as suggested in [24].

Other QSARs for skin permeability coefficient generally represent a combination of solvatochromic approach combined with inclusion of experimental predictive parameters

such octanol/water partitioning coefficient. For comprehensive review of such QSARs reader should refer to reference [27].

A significant shortcoming of QSAR based on the skin permeability coefficient is that this coefficient strongly depends on the solubility of a solute in a given vehicle through its dependence on K_{sc}. For equations (8), (9) and (10) regression coefficients are as a result strongly influenced by solute's aqueous solubilities (S_{aq}) [28, 29]. This solubility makes a significant contribution to the regression parameters. In most cases, k_p from aqueous vehicle is even dominated by S_{aq}, as exemplified by a comparison of permeability data for methanol and octanol: k_p and K_{sc} values for these solutes are 0.0005 cm/h and 0.6; 0.052 cm/h and 50 respectively [16]. This two orders of magnitude difference, in both k_p and K_{sc}, deceptively suggests that octanol is much more soluble in SC and is a better transdermal permeant than methanol. In fact, using equation $K_{sc} = S_{sc}/S_{aq}$ (where S_{sc} is the solubility of solute in SC) and S_{aq} values of 31 mmol/mL and 0.0041 mmol/mL for methanol and octanol respectively [28] one gets 18.6 mmol/mL for methanol solubility in SC and only 0.21 mmol/mL for octanol solubility in SC. Therefore octanol is two orders of magnitude less soluble in SC than methanol. This higher SC solubility now suggests that methanol is a better permeant. Indeed, the maximum flux ($\boldsymbol{J}_{\max}$), that is the flux obtained for drug at saturation in the vehicle, of methanol (even without self-enhancement) is much higher than the maximum flux for octanol (15.5 μmol/cm^2/h vs. 0.21 μmol/cm^2/h). This misrepresentative nature of the permeability coefficient was the principal reason to request a paradigm change in topical delivery literature [29]. It was suggested that $J_{\max}$ must be used instead of permeability coefficient when reporting and discussing skin permeation. Note that equation (6) for viable epidermis concentration can be easily rewritten in terms of $\boldsymbol{J}_{\max}$:

$$C_{ve} = \frac{k_p A C_v}{Cl_s} = \frac{A J_{\max}}{Cl_s}\frac{C_v}{S_v} \tag{11}$$

where S_v is the solubility of the drug in the vehicle.

Saturation of drug in the vehicle leads to saturation in the top layers of SC. Thus permeation from any saturated vehicle produces the same (maximum) flux $\boldsymbol{J}_{\max}$ independent of the vehicle, providing the vehicle does not affect the skin. Using regression analysis for a data

set of 87 compounds delivered from aqueous solution it was found that molecular weight is the principle determinant of the maximum flux (in mole $cm^{-2}h^{-1}$) for a solute [28]:

$$\log J_{\max} = -0.0190MW - 3.90, \qquad r^2 = 0.847, \quad n = 87 \tag{12}$$

Only marginal improvement was achieved when other factors such as solubility in octanol or melting point were taken into account. For a larger data set of 277 compounds delivered from various vehicles, the regression equation was

$$\log J_{\max} = -0.0141MW - 4.52, \qquad r^2 = 0.668, \quad n = 277 \tag{13}$$

These results for maximum flux regression are substantially different to the result of the regression for the k_p described by equation (8), as $\log K_{oct}$ was not found to be a significant predictor of the maximum flux. It is possible that $\log K_{oct}$ is not significant in the regression (13) due to the overwhelming dependence of $J_{\max}$ on the molecular weight. This dependence was negated by considering the $J_{\max}$ of similar molecular size solutes and it was demonstrated that a non-linear parabolic dependence on $\log K_{oct}$ existed for these solutes [30].

It is important to note that $J_{\max}$ is not always independent of the vehicle used. For many vehicles its component solvents can partition into skin lipids, potentially changing solubility and the rate of diffusion and therefore changing the maximum flux. Recently the field of predicting skin permeability from complex vehicles was reviewed and it was concluded that although the use of QSPR models similar to one in equation (10) has some merit for mixtures and complex systems, caution must be taken when using these models outside of the domain of chemical properties of solutes and solvents for which they were formulated [31].

A number of more sophisticated numerical methods (which are often not as easy to apply as the regressions presented here) have also been employed in order to determine the most suitable presentation of linear QSARs, as well as the use of other methods such as principle component analysis and neural networks – see Ref. [32] and references therein for a recent review.

3. Mathematical modelling of transport processes in skin layers

Transport of solutes in skin layers is usually modelled by the diffusion equation:[33]

$$\frac{\partial C(x,t)}{\partial t} = D\frac{\partial^2 C(x,t)}{\partial x^2} \tag{14}$$

where $C(x,t)$ is the concentration in the skin layer at depth x and time t and D is the diffusion coefficient in the skin layer. While D changes from SC to viable epidermis to dermis, the form of equation (14) is generally unaffected, except for the clearance term added in the dermal layer to account for blood circulation (see equation (22)). Specifics of drug application and interactions with other skin layers are described by the appropriate choice of boundary and initial conditions. As an example let us consider the initial and boundary conditions for infinite (that is no or negligible depletion) well stirred vehicle application to the SC. As there is no drug initially present in the SC the condition at t=0 is:

$$C_{sc}(x,0) = 0 \tag{15}$$

The boundary conditions for the SC are formulated for the outer SC layer in contact with the vehicle (x=0) and the inner layer in contact with the viable epidermis (sink condition is assumed at $x=h_{sc}$):

$$C_{sc}(0,t) = K_{sc}C_v, \quad C_{sc}(h_{sc},t) = 0 \tag{16}$$

Equations (16) are the simplest form of boundary conditions for the SC. Other more complex forms of boundary conditions arise when finite vehicle is applied [34, 35] or when sink condition is not justified [36]. Sink boundary condition describes very rapid removal of solute from the inner surface of the SC by fast diffusion through the VE and absorption of the solute by blood in the dermis, so that the concentration in the outer SC layer is much larger than that in the inner layer ($C_{sc}(0,t) \gg C_{sc}(h_{sc},t)$). This assumption of very rapid removal adequately describes transport kinetics for many solutes and experimental and physiological conditions. It needs to be noted though that the sink condition could be invalid, for example, in the case of a very low solubility of a solute in VE or receptor compartment.

Equation (14) with the initial and boundary conditions (15) and (16) can be solved using Laplace transformation: (detailed account of the solution process was recently given in [37], where numerical approaches, advantages and limitations of the Laplace transform technique for modelling skin transport were also given)

$$\hat{C}_{sc}(x,s) = \frac{K_{sc}C_v}{s}\frac{\sinh\left(\sqrt{st_d}\left(h_{sc}-x\right)/h_{sc}\right)}{\sinh\left(\sqrt{st_d}\right)} \tag{17}$$

where t_d ($=h_{sc}^2/D_{sc}$) is the characteristic time of diffusion, a circumflex over a function (^) denotes the Laplace transform and s is the Laplace variable. Equation (17) is similar (with different notations and boundary conditions for x=0 and x=h_{sc} interchanged) to that presented in [38] for the case of heat conduction in solids which is also governed by the diffusion equation. The flux of solute (J) and the total amount of solute absorbed (Q) through the SC to the viable epidermis can be obtained from equation (17):

$$\hat{J}(s) = -D_{sc}\left.\frac{\partial \hat{C}_{sc}}{\partial x}\right|_{x=h_{sc}} = \frac{K_{sc}D_{sc}C_v}{h_{sc}s}\frac{\sqrt{st_d}}{\sinh\left(\sqrt{st_d}\right)} = \frac{J_{\max}}{s}\frac{C_v}{S_v}\frac{\sqrt{st_d}}{\sinh\left(\sqrt{st_d}\right)} \tag{18}$$

$$\hat{Q}(s) = A\frac{\hat{J}(s)}{s} = \frac{AK_{sc}D_{sc}C_v}{h_{sc}s^2}\frac{\sqrt{st_d}}{\sinh\left(\sqrt{st_d}\right)} = \frac{AJ_{\max}}{s^2}\frac{C_v}{S_v}\frac{\sqrt{st_d}}{\sinh\left(\sqrt{st_d}\right)} \tag{19}$$

The attractiveness of Laplace domain solutions is enhanced by the existence of standard nonlinear regression programs such as MULTI FILT, MINIM and SCIENTIST, that enable fast analysis of experimental data using Laplace domain solutions directly and avoid computational complexities associated with infinite series solutions, especially those involving solving transcendental equations. A simple algorithm for numerical inversion of Laplace domain solutions to time domain is presented in [37].

The use of the diffusion equation (14) for modelling transport in the SC can be considered a minimalistic approach where SC's complexity is replaced with a homogeneous membrane. In reality the SC has a microstructure that is often referred to as "brick and mortar". A recent review of models that directly incorporate key features of the SC microstructure is given in [13, 15]. While these models are useful in deeper understanding of transport through the SC, it needs to be stressed that the large number of parameters required by these models confounds their applicability for direct analysis of experimental data.

In a general case a two-layered diffusion problem with viable epidermis (VE) and SC layers as in Refs. [39, 40] needs to be considered to determine concentration in the VE which is most relevant to skin toxicology. This approach is generally complicated mathematically and is not necessary to assess transport in the VE when it does not contribute significantly to overall skin barrier, which is often the case. Here we assume that the flux from the SC to the VE is determined by the SC alone. In this case the concentration in the VE can be calculated considering diffusion equation (14) initial condition $C_{ve}(x,0)=0$ and appropriate boundary

conditions for this layer. Again, for simplicity sink boundary condition can be assumed now for dermal-epidermal junction ($C_{ve}(h_{ve},t)=0$, here x is measured from SC/VE boundary). For the boundary between VE and SC the appropriate boundary condition in this case is:

$$-D_{ve}\left.\frac{\partial C_{ve}}{\partial x}\right|_{x=0}=J(t)$$

where $J(t)$ is the flux from the SC to VE, determined for example by equation (18). As the diffusion rate in viable tissues is usually much faster than in the SC (eg $D_{ve}\gg D_{sc}$) characteristic time of diffusion in VE ($t_{dve}=h_{ve}^2/D_{ve}$) is expected to be much less than that for the SC ($t_{dve}\ll t_d$) and concentration in the VE can be approximated by:

$$\hat{C}_{ve}(x,s)=\hat{J}(s)\frac{h_{ve}}{D_{ve}}\left(1-\frac{x}{h_{ve}}\right) \tag{20}$$

which for the steady state further simplifies to:

$$C_{ve}^{ss}(x)=J_{\max}\frac{C_v}{S_v}\frac{h_{ve}}{D_{ve}}\left(1-\frac{x}{h_{ve}}\right) \tag{21}$$

This concentration reaches its maximum at the top of VE (x=0) and can be determined as:

$$C_{ve}^{\max}=J_{\max}\frac{C_v}{S_v}\frac{h_{ve}}{D_{ve}} \tag{22}$$

We stress again that a more complex solution is required when the diffusion time in VE is not negligible compared to that of SC. This situation was considered in [39] and solved in the Laplace domain. Later a similar two-phases-in-series model was considered and analytical solutions were obtained together with numerical simulations [40]. Perhaps, solution of VE using the two-layer diffusional transport model is most justified when metabolism of the drug in the VE is present. This case was considered and equations for drug and metabolite amount exiting the VE into receptor phase or dermis were obtained [41]. This solution has been recently discussed and presented using notations similar to this work in [13].

Although equation (22) can offer valuable insight, there appears little experimental data for VE diffusion coefficient. Thus prediction of D_{ve} remains to be based on a theoretical approach such as in [40]. In this work a method was developed that took into account limited permeability of the VE and recommended to approximate permeability of the VE as $k_p^{ve}=2.6/\sqrt{MW}$ (cm/h). Therefore, as $k_p^{ve}=K_{ve}D_{ve}/h_{ve}$, and assuming that K_{ve} is close to

unity (which is justified in the absence of significant binding in VE and hydrophilic vehicle) then $h_{ve}/D_{ve} \approx 0.4\sqrt{MW}$ (h/cm) can be used in equations (20) to (22).

For modelling dermal drug transport the distributed elimination model is often used. It has first been suggested for peritoneum [42]. Later concentration-depth profiles of 2',3'-dideoxyinosine in the dermis of a rat were modelled using this model [43]. The model was used to describe the concentration depth profiles of solutes in the dermis after application of solutes to skin with removed epidermis [44]. In [45] the distributed elimination model was applied together with modelling of SC and VE to re-analyse the data in [46]. The distributed elimination model accounts for blood clearance in the dermis by the elimination term added to the diffusion equation:

$$\frac{\partial C_d}{\partial t} = D_d \frac{\partial^2 C_d}{\partial x^2} - k_e C_d \tag{23}$$

where C_d is concentration in the dermis at depth x (here from VE dermis boundary) at time t, D_d is the effective molecular diffusion coefficient in the dermis and k_e is the elimination rate from the dermis due to blood clearance. The boundary condition for the upper layer of dermis is $D_d \frac{\partial C_d}{\partial x}\Big|_{x=0} = -J(t)$ where $J(t)$ is the flux from the VE to dermis, which in the absence of metabolism is the same as the flux from SC to VE. Assuming zero (or negligible) concentration in the very deep dermal layers ($\lim_{x\to\infty} C_d(x,t) = 0$) together with initial condition of no solute originally present in the dermis ($C_d(x,0) = 0$), equation (23) can be easily solved in the Laplace domain: [47]

$$\hat{C}_d(x,s) = \frac{\hat{J}(s)}{s\sqrt{(s+k_e)D_d}} \exp\left(-x\sqrt{(s+k_e)/D_d}\right) \tag{24}$$

the steady state dermal concentration is defined by:

$$C_d^{ss}(x) = J_{\max} \frac{C_v}{S_v} \frac{1}{\sqrt{k_e D_d}} \exp\left(-x\sqrt{k_e/D_d}\right) \tag{25}$$

and as expected the maximum dermal concentration is achieved in the top layer of the dermis (x=0):

$$C_d^{\max} = J_{\max} \frac{C_v}{S_v} \frac{1}{\sqrt{k_e D_d}} \tag{26}$$

Equation (25) can be directly applied to experimental dermal concentration-distance profiles to obtain transport parameter $\sqrt{k_e/D_d}$, providing that the steady-state profile has formed. This approach was used in [48] combined with theoretical analysis of partitioning, diffusivity and clearance rate of 26 solutes in mammalian dermis.

It is important to note that equations (20) to (22) for concentration in VE are only valid when sink boundary condition can be assumed for dermal-epidermal junction. It is therefore essential for these equations to be valid that dermal concentration is much less than VE concentration, that is $K_d C_d^{\max} \ll K_{ve} C_{ve}^{\max}$, where K_d and K_{ve} are partition coefficients between dermis/vehicle and VE/vehicle respectively and the concentrations are defined by equations (26) and (22) respectively. When this is not the case the boundary condition for dermal-epidermal junction becomes $C_{ve}(h_{ve},t) = C_d^{\max} K_{ve}/K_d$ and equation (21) for the VE concentration has to be replaced by:

$$C_{ve}^{ss}(x) = J_{\max}\frac{C_v}{S_v}\frac{h_{ve}}{D_{ve}}\left(1-\frac{x}{h_{ve}}\right)+\frac{K_{ve}}{K_d}C_d^{\max} \tag{27}$$

As a result the maximum concentration in VE will be:

$$C_{ve}^{\max} = J_{\max}\frac{C_v}{S_v}\left[\frac{h_{ve}}{D_{ve}}+\frac{K_{ve}}{K_d}\frac{1}{\sqrt{k_e D_d}}\right] \tag{28}$$

In deriving equations for concentrations in epidermis and dermis it was implicitly assumed that the flux though the SC is uniform. This is clearly not the case when appendageal transport though the SC is not negligible or the SC integrity is compromised through disease or mechanical/chemical damage. In extreme cases of SC barrier damage the concentration in the top VE layer (or in some of it areas under damaged SC barrier) will be at equilibrium with the applied vehicle, that is $C_{ve}^{\max} = K_{ve} C_v$.

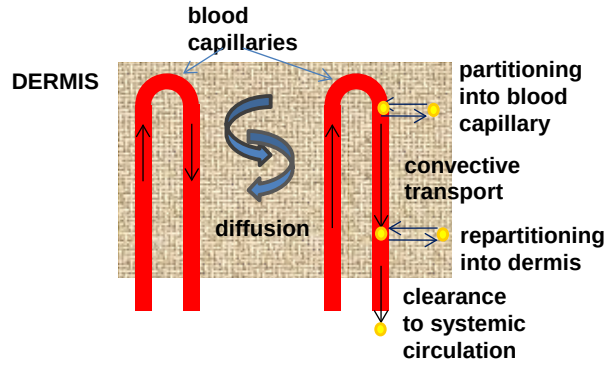

Fig 2. Schematics of drug transport in dermis.

In [47] literature human biopsy data from Refs [49-53] were mathematically modelled and it was established that for many solutes molecular diffusion in

dermal tissue cannot explain the data. Similar conclusion was made after the analysis of published human microdialysis data [54]. It was concluded that partitioning of solute into blood capillaries of the dermis and its subsequent convective transport to deeper layers and partitioning back into the tissue (Fig 2) could significantly contribute to the spatial transport of the solute. As blood in the tissue capillaries can flow in all possible directions it was concluded that these successive repartitioning and convective travels of the solute molecules were analogues to a random walk process, which often is referred to as dispersion [47]. Lymphatic flow could also be a contributing factor to this random walk process. In order to recognise these facts D_d in equations above needs to be replaced by total dispersion coefficient (D_t): [47]

$$D_t = D_v + D_d \tag{29}$$

where D_v is the contribution to the transport processes from blood and/or lymphatics and is referred to as dispersion coefficient. If this dispersion coefficient contributes significantly to the overall transport of solute, D_v will be much greater than molecular diffusion coefficient (D_d). At the condition when the transport is dominated by molecular diffusion, then $D_t \approx D_d$. In [47] for 5 out of 6 solutes D_t was dominated by the dispersion transport and was about 5×10^{-6} cm^2s^{-1}. It is likely that D_t depends on the rate of dermal blood flow. Dispersion coefficient was previously used in modelling of liver clearance [55] and the dispersion coefficient for the liver model was later related to blood velocities in liver sinusoids and other physiological parameters [56] – a similar relationship remains to be established for the dermal transport model.

4. Expert opinion

Arguably the largest effort in mathematical modelling relevant to skin toxicology so far was in predicting permeation rate through skin from drug's physicochemical properties in order to predict systemic absorption of drugs. In terms of toxicity studies such modelling is more relevant to assessing environmental exposure and systemic toxicity associated with long term skin exposure to aqueous solutes, which is important, but does not cover all aspects of skin toxicity. As evident from section 2 of this work a significant progress has been achieved so far in predicting permeation from large (often referred to as infinite) aqueous vehicles. This progress is in part due to an abundance of appropriate literature experimental data owing to relative simplicity of the experiments. Another reason is that constant diffusion coefficient and solubility of solutes in SC can be assumed during experiments with infinite aqueous

vehicles. This constancy simplifies mathematical modelling. Further progress in this area is likely to be related to using larger datasets of skin permeation rates and using solutes physicochemical parameters calculated from more sophisticated molecular simulation software. A significant challenge in further improvement of predicting permeation rate through SC is very significant interindividual and intraindividual variability of skin permeability. Variability of data obtained from different laboratories [57] also poses significant challenges to further refining of the prediction.

Experimental work and modelling for application of infinite aqueous vehicles to skin can be considered a low hanging fruit compared to experimental and theoretical work required for predicting skin permeation and toxicity resulting from realistic drug applications to skin. This is reflected in relative scarcity of studies in the literature investigating shorter exposures to solutes as well as solutes applied in various composite solvents. These studies are more relevant to assessing skin toxicity associated with occupational exposure and better reflect the use of dermal drugs. Composite solvents can alter SC transport properties, such as solubility and diffusion coefficient, in time dependant and non-linear fashion presenting significant mathematical challenges. Skin absorption studies conducted with composite solvents as vehicles are challenging due to a sheer number of possible combinations of vehicle compositions possible and the volume of work required, as well as difficulty in interpreting the results of such studies. These modelling and experimental challenges will need to be resolved to provide useful prediction of drug permeation from more realistic finite vehicles that are practically used in dermal treatments and transdermal drug delivery. These studies constitute an important and challenging area of future research effort.

In the area of mathematical modelling of solutes transport through skin the emphasis is usually on modelling SC as it is the main barrier to solute permeation. In this review priority was given to models which predict drug concentration in the viable layers of skin, as this is most relevant to assessing skin toxicology. To date the mathematical modelling of transport processes in viable epidermis and dermis has been limited to few studies that assume uniform drug penetration and only consider intact SC. Therefore more effort is required to develop more detailed mathematical models of drug transport in deeper skin layers. More explicit interpretation of skin physiology (e.g. binding to various skin components, blood flow and drug transport associated with it and drug metabolism) is required to further advance current mathematical models. Modelling diseased and compromised skin barrier is another

significant challenge in skin absorption studies. A simple act of shaving significantly increases the permeability of SC, which can be easily felt when applying ethanol based aftershave solutions (which are perhaps mostly obsolete now). Rubbing of skin on tough fabric and various other physical actions on SC can potentially exacerbate occupational transdermal exposure to toxins and increase potential for skin toxicity. There are now many techniques such as ultrasound, microneedles, laser and heat oblation and abrasive treatments that are applied intentionally to dramatically increase the permeability of skin. Considering skin in diseased or damaged state adds an extra dimension and is thus a significant challenge to mathematical modelling, but is necessary to further enhance the role of mathematical modelling in helping to assess skin toxicology.

Declaration of interest

Author has no conflicts of interest regarding this review.

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