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THEORETICAL ASPECTS OF METABOLISM IN THE EPIDERMIS

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SUMMARY

Mathematical expressions have been derived to show the effect of metabolism within the epidermis. Particular emphasis has been given to substances such as the corticosteroids which are known to form a reservoir in the skin. Any compound which resides in a depot for any length of time is more liable to metabolic change and the results confirm this quantitatively. Metabolism both in the upper and lower layers of the epidermis has been considered and the relative effects of these described.

INTRODUCTION

The corticosteroids are known to form a reservoir which is located in the stratum corneum. There is evidence that suggests that the residence time of the depot may be as long as 14 days or more (McKenzie et al., 1962; Vickers, 1963; Winkelmann, 1969). Since these compounds remain in this section of the skin for a long time it is possible that they will undergo some metabolic change. Malkinson et al. and Hsia et al. have shown that hydrocortisone is actively metabolized by the skin. It is unclear in which region metabolism occurs, although Ando et al. (1977) suggest that there is a gradual increase in metabolic activity as the basal layer is approached. The analysis by these authors is given where there is no metabolism in the stratum corneum and a uniform distribution of enzyme activity in the viable epidermis.

More recent calculations (Fox et al., 1979) have produced computer profiles to show the variation of prodrug, drug and metabolite concentrations within the dermis and epidermis. The advantage of this type of analysis is that more complex kinetic mechanisms may be postulated. For example, in their publication Michaelis–Menten kinetics were considered. However, the authors did not consider metabolism within the stratum corneum and although stratum corneum is a dead tissue, metabolic enzymes may still be present. This could be caused, in the case of healthy skin, by the continual rapid turnover

of the cells which will transport some of the enzymes upwards with them. Alternatively, in diseased conditions some of the living cells penetrate the stratum corneum. The living cells will contain enzymes and metabolic changes are then possible in the higher regions of the epidermis.

It has been well established that metabolism occurs in the lower regions of the epidermis but in this area the residence time of a molecule is very much shorter than that in the depot. The degree of metabolism in the two areas will thus be a balance between the rate constants involved and the length of time that the drug remains in the regions. In this paper the relative importance of metabolism in the two regions will be discussed by means of a mathematical model.

The model will be essentially that described in an earlier publication (Hadgraft, 1979b). This model will be developed to show the effects of metabolism and the different physicochemical parameters on the overall mass transfer of an active drug across the epidermis. It will show how the extent of drug transformation differs in the two regions of the epidermis: (a) the stratum corneum; and (b) the viable epidermis.

THE MODEL

As in the previous theoretical descriptions of percutaneous absorption (Albery and Hadgraft, 1979; Hadgraft, 1979b), an idealized physical model is used to simplify the mathematical concepts. Two routes of penetration are considered, firstly when the drug diffuses straight through the cells (a diffusional path length, ℓ_T) and secondly when the substrate passes through the channels surrounding the cells (path length, ℓ_I). Typical values of ℓ_T and ℓ_I are respectively, 25 μm and 350 μm (Hadgraft, 1979b).

	Stratum corneum	Viable epidermis	Capillaries
Diffusion coefficient	D_s	D_a	
Concentration	$C_{s,0}$ (at $t = 0$)	C_a	0
Partition coefficient		$K = \frac{C_s}{C_a}$ (At $\chi = \chi' = 0$ and zero flux)	
Distance	$\ell_s \xleftarrow{x'} 0$	$0 \xrightarrow{x} \ell_a$	
Normalized distance	$1 \xleftarrow{\chi'=x'/\ell_s} 0$	$0 \xrightarrow{\chi=x/\ell_a} 1$	
Removal rate by capillaries			$\xrightarrow{k_c}$
Metabolic rates	k'_m	k_m	
Area	$\propto A$	A	A

SCHEME 1

The mathematical model is shown schematically in Scheme 1. In this scheme the rate of removal of the drug is given by 3 first order rate constants. First order constants have been chosen in order that simple analytic functions are produced. No attempt has been made to use Michaelis–Menten kinetics as this has been discussed previously and

requires computer analysis (Fox et al., 1979). The removal by the capillaries at the dermal–epidermal junction is described by the first order rate constant, k_c. This is related to the total amount (M_t) of drug removed at time t by

$$M_t = A\ell_a \int_0^t k_c c_{a(\chi=1)}\, dt \tag{1}$$

where ℓ_ϵ is the thickness of the viable epidermis and $c_{a(\chi=1)}$ is the drug concentration at the capillary interface. Secondly, there are the two metabolic rates k'_m and k_m which describe, respectively, the first order rates of metabolism in the stratum corneum and the viable epidermis.

The following normalized variables are used to simplify the solution of the differential equations generated to describe the overall transport process.

$$u = c/c_{s,o} \tag{2}$$

where c is the concentration in the skin and $c_{s,o}$ is the concentration of drug in the stratum corneum at time t equals zero. As in the previous description (Hadgraft, 1979b), it is assumed that the drug is left in contact with the skin for some period. During this period the drug concentration in the epidermis builds up to a maximum, $c_{s,o}$. At this point the drug in contact with the skin surface is removed and the equations derived predict the rate of disappearance of the drug stored in the stratum corneum. Expressions for the rates of uptake have already been derived (Albery and Hadgraft, 1979).

The other normalized parameters are defined below.

$$\lambda_I = \ell_a/\ell_I,\ \lambda_T = \ell_a/\ell_T \tag{3}$$

$$p = D_s\ell_a^2/D_a\ell_s^2 \tag{4}$$

where D_s and D_a are respectively the diffusion coefficients of the substrate in the stratum corneum and the viable epidermis.

$$\tau = D_a t/\ell_a^2 \tag{5}$$

$$\omega = k_c\ell_a^2/D_a \tag{6}$$

$$\kappa'_m = k'_m\ell_a^2/D_a \tag{7}$$

$$\kappa_m = k_m\ell_a^2/D_a \tag{8}$$

The assumptions that will be made are that diffusion occurs in one dimension and that the diffusion coefficients are independent of concentration. It is then possible to describe the transport processes in terms of the following differential equations. In the stratum

corneum,

$$\frac{\partial u}{\partial \tau} = p\left(\frac{\partial^2 u}{\partial \chi'^2}\right) - \kappa'_m u \tag{9}$$

and in the viable epidermis

$$\frac{\partial u}{\partial \tau} = \left(\frac{\partial^2 u}{\partial \chi^2}\right) - \kappa_m u \tag{10}$$

In a previous publication (Hadgraft, 1979b), it has been shown that slow interfacial transfer at the stratum corneum–viable epidermis junction is insignificant. Thus the following boundary conditions apply.
At the stratum corneum–viable epidermis interface:

$$\psi\left(\frac{\partial u}{\partial \chi'}\right)_0 = -\left(\frac{\partial u}{\partial \chi}\right)_0 \tag{11}$$

where $\psi = \alpha p/\lambda$.
The term ψ describes the effect of the area fraction (α) and the path length differences (λ) caused by the two different routes of penetration. It also allows for the diffusion coefficient and path length differences between the stratum corneum and the viable epidermis. At the dermal–epidermal junction:

$$\left(\frac{\partial u}{\partial \chi}\right)_1 = -\omega u_{a,1} \tag{12}$$

Thus in terms of the dimensionless variables, the amount of drug diffused in time t is given by

$$M_t = c_{s,0} A \ell_a \int_0^\tau \omega u_{a,1}\, d\tau \tag{13}$$

In order to calculate M_t the differential equations (9) and (10) are solved with the boundary conditions,

$$\chi' = 1, (\partial u/\partial \chi') = 0 \tag{14}$$

$$\tau = 0,\ u_s = 1 \tag{15}$$

$$\tau = 0,\ u_a = 0 \tag{16}$$

The first condition, 14, arises since there is no supply of drug on the outside surface of the skin. Condition 15 shows the solution is valid for the case where a finite reservoir of

drug has been established in the stratum corneum and, condition 16, that insufficient time has elapsed for significant transfer into the viable epidermis.

The differential equations are solved using Laplace transforms. In the stratum corneum, Eqn. (9) is transformed to give:

$$s\bar{u}_s - 1 = p\left(\frac{\partial^2 \bar{u}_s}{\partial \chi'^2}\right) - \kappa'_m \bar{u}_s \tag{17}$$

which has the general solution;

$$\bar{u}_s = A\cosh\{(s+\kappa'_m)^{\frac{1}{2}}p^{-\frac{1}{2}}\chi'\} + B\sinh\{(s+\kappa'_m)^{\frac{1}{2}}p^{-\frac{1}{2}}\chi'\} + (s+\kappa'_m)^{-1} \tag{18}$$

Using the boundary condition, 14, the coefficients A and B may be eliminated to give:

$$\bar{u}_{s,\chi'=0} = (s+\kappa'_m)^{-1} - p^{\frac{1}{2}}(s+\kappa'_m)^{-\frac{1}{2}}\left(\frac{\partial \bar{u}_s}{\partial \chi'}\right)_0 \operatorname{cotanh}\{(s+\kappa'_m)^{\frac{1}{2}}p^{-\frac{1}{2}}\} \tag{19}$$

In the viable epidermis the same treatment is adopted:

$$s\bar{u}_a = \left(\frac{\partial^2 \bar{u}_a}{\partial \chi^2}\right) - \kappa_m \bar{u}_a \tag{20}$$

and this equation has the general solution:

$$\bar{u}_a = A'\cosh\{(s+\kappa_m)^{\frac{1}{2}}\chi\} + B'\sinh\{(s+\kappa_m)^{\frac{1}{2}}\chi\} \tag{21}$$

Using methods previously described (Hadgraft, 1979a) the coefficients A′ and B′ may be eliminated to show how the concentration at the inner interface varies in Laplace time:

$$\bar{u}_{a,\chi=1} = -\omega^{-1}\left(\frac{\partial \bar{u}a}{\partial \chi}\right)_0 [\cosh\{(s+\kappa_m)^{\frac{1}{2}}\}(1+(s+\kappa_m)^{\frac{1}{2}}\omega^{-1}\tanh\{(s+\kappa_m)^{\frac{1}{2}}\})]^{-1} \tag{22}$$

and at the other interface:

$$\bar{u}_{a,\chi=0} = \bar{u}_{a,\chi=1}(\cosh\{(s+\kappa_m)^{\frac{1}{2}}\})^{-1} - (s+\kappa_m)^{-\frac{1}{2}}\left(\frac{\partial \bar{u}_a}{\partial \chi}\right)_0 \tanh\{(s+\kappa_m)^{\frac{1}{2}}\} \tag{23}$$

Since interfacial transfer is considered to be very rapid and the diffusional processes are comparatively slow, there will be local equilibrium established at the viable epidermis–stratum corneum junction and,

$$K\bar{u}_{a,\chi=0} = \bar{u}_{s,\chi'=0} \tag{24}$$

combining Eqns. 11, 19 and 24

$$\bar{u}_{a,\chi=0} = K^{-1}(s+\kappa'_m)^{-1} + p^{\frac{1}{2}}(s+\kappa'_m)^{-\frac{1}{2}}K^{-1}\psi^{-1}\left(\frac{\partial \bar{u}_a}{\partial \chi}\right)_0 \operatorname{cotanh}\{(s+\kappa'_m)^{\frac{1}{2}}p^{-\frac{1}{2}}\} \tag{25}$$

It is then possible to eliminate the terms in $(\partial\bar{u}_a/\partial\chi)_0$ to give an expression which shows how $\bar{u}_{a,\chi=1}$ alone varies with the Laplace time variable s.

$$\bar{u}_{a,\chi=1} = F(s)$$

where

$$F(s) = [p^{\frac{1}{2}}(s+\kappa'_m)^{\frac{1}{2}}\psi^{-1}\ \text{cotanh}\{(s+\kappa'_m)^{\frac{1}{2}}p^{-\frac{1}{2}}\}\ (\omega\cosh\{(s+\kappa_m)^{\frac{1}{2}}\} + (s+\kappa_m)^{\frac{1}{2}}\sinh\{(s+\kappa_m)^{\frac{1}{2}}\}) + K(s+\kappa'_m)\,\omega(s+\kappa_m)^{-\frac{1}{2}}\sinh\{(s+\kappa_m)^{\frac{1}{2}}\} + K(s+\kappa'_m)\cosh\{(s+\kappa_m)^{\frac{1}{2}}\}]^{-1} \quad (26)$$

The total amount of active drug that has diffused to the inner interface at time t is then given by application of Eqn. 13.

$$M_t = C_{s,0}A\ell_a\omega\ L^{-1}F(s)/s \quad (27)$$

It is not possible to obtain a simple expression from the inversion of Eqn. 26. However, several valid approximations may be made in order to simplify the equation.

Metabolism will only be significant in long periods of time, ie. $\tau > 1$ and $s < 1$. This condition implies that the amount of metabolism occurring in the time taken to establish steady-state diffusion across the viable epidermis (~40 min) is negligible. Few data have been produced on the rates of metabolism in the epidermis but for diflucortolone valerate (DFV) Täuber and Toda (1976) have shown that 80–90% DFV was still identifiable after 7 h application. This would correspond to a κ value of ~0.1 and thus the approximation that $\kappa < 1$ can also be used. Consequently $(s+\kappa_m)^{1/2} < 1$ and the hyperbolic terms in Eqn. 26 can be approximated to give:

$$F(s) = \left[s\left(\frac{\lambda}{\alpha\omega} + K + \frac{K}{\omega}\right) + \frac{\lambda}{\alpha} + \frac{\lambda\kappa_m}{\alpha\omega} + K\kappa'_m + \frac{K\kappa'_m}{\omega}\right]^{-1} \quad (28)$$

The way in which M_t varies with τ is then found by substituting Eqn. 28 into Eqn. 27 and inverting the Laplace transform (Abramowitz and Stegun, 1970):

$$M_t = C_{s,0}A\ell_a F(\tau) \quad (29)$$

where

$$F(\tau) = \left[\frac{1}{\left(\frac{\lambda}{\alpha} + \frac{\kappa_m}{\alpha\omega} + K\kappa'_m + \frac{K\kappa'_m}{\omega}\right)}\left[1 - \exp\left\{\frac{-\left(\frac{\lambda}{\alpha} + \frac{\kappa_m\lambda}{\alpha\omega} + K\kappa'_m + \frac{K\kappa'_m}{\omega}\right)\tau}{\left(\frac{\lambda}{\alpha\omega} + K + \frac{K}{\omega}\right)}\right\}\right]\right] \quad (30)$$

In terms of the initial unmetabolized drug stored in the stratum corneum (M_∞) which is

also the quantity of drug plus its metabolites which will be released into the capillaries after an infinite period

$$M_t = \frac{\lambda}{\alpha} M_\infty F(\tau) \tag{31}$$

This equation is similar in form to that previously derived (Hadgraft, 1979b) which relates the reservoir capacity of the skin to several basic physicochemical parameters. Using Eqn. 31 and the normalized parameters discussed below, it is possible to see how the metabolic processes affect rates of transfer into the capillary system.

Estimates of the normalized parameters given in Eqns. 3 and 6 have been made in an earlier publication (Hadgraft, 1979b). For the intercellular route, λ_I is equal to 0.4 and for the transcellular route, λ_T is equal to 6. This means that a drug molecule diffuses approximately 15 times further in the intercellular channels than in the corresponding transcellular route. Also the intercellular channels occupy a much smaller area, $\alpha = 7 \times 10^{-3}$, The calculation of the rate of substrate removal by the capillaries has been discussed previously (Hadgraft, 1979b) and is based on a first order rate constant of 10^{-5} s^{-1}. This combined with the length of the viable epidermis (150 μm) and the diffusion coefficient of a steroid in this medium (10^{-11} $m^2 s^{-1}$) gives a value for ω of 2×10^{-2}.

Different values of κ_m, κ'_m and K will be used to show the effect of these fundamental constants in the overall active drug concentration reaching the capillary system.

TRANSCELLULAR ROUTE

Fig. 1A shows how a drug is released into the capillary network as a function of its partition coefficient. The larger the oil/water partition coefficient, the longer the drug remains in the lipoidal regions of the stratum corneum. The profile demonstrates the overall drug release over a period of approximately 10 days ($\tau = 350$). It will be noted that for strongly lipophilic materials (K = 100) only one-third of the total drug concentration has been eliminated from the cutaneous region. This fact has been discussed

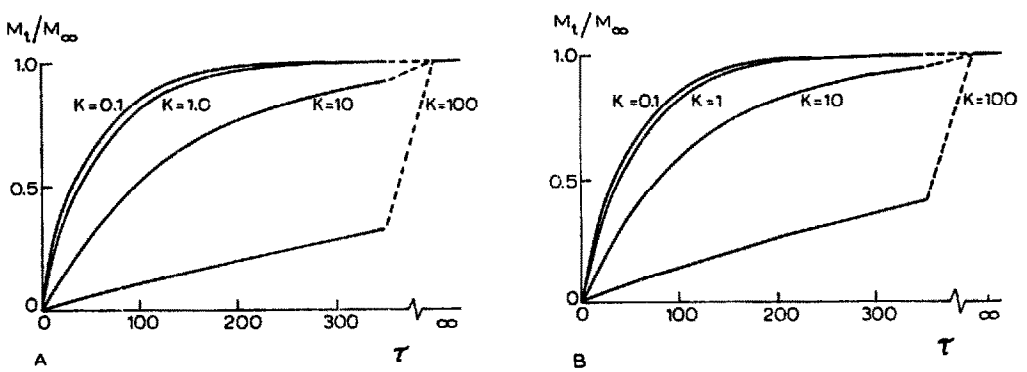

Fig. 1. A: release profiles showing the effect of partition coefficient on the transcellular route with no metabolic processes. B: as in Fig. 1A but for the intercellular route.

previously (Hadgraft, 1979b) and is possibly one of the causes of the reservoir that is formed after the application of topical corticosteroids.

Since the substances do remain in the epidermis for fairly extended periods it is possible that metabolism could occur and produce inactive products. Fig. 2A shows this effect as a function of partition coefficient. In this figure the metabolism occurs solely in the stratum corneum and a first order rate constant for metabolism of 4.4×10^{-5} s^{-1} is assumed ($\kappa'_m = 0.1$). The different curves depicted show that, for substances which are lipophilic, appreciable metabolism occurs. For K = 100, after infinite time, only a very small fraction of the active drug reaches the capillary system. As the partition coefficient decreases the drug has less tendency to reside in the lipophilic regions of the stratum corneum and the degree of metabolism decreases. The curves also show that all the active drug is released into the capillary network over a comparatively short period of time. For K = 100, all the active drug that is going to reach the epidermal–dermal boundary has done so when $\tau \sim 8$ (approximately 5 h) and for K = 1 most of the active drug is transported by $\tau \sim 100$ (approximately 60 h). Thus if this degree of metabolism occurs in the stratum corneum the existence of a drug reservoir which remains active for 15 or 20 days would not be expected.

The next case considered (Fig. 3A) is where there is no metabolism in the stratum corneum but a significant amount in the viable epidermis. For the calculations a first order rate constant of 4.4×10^{-5} s^{-1} ($\kappa_m = 0.1$) has been chosen to provide comparison with the data given in Fig. 2A. The graph shows the effect of partition coefficient with this fixed rate of metabolism. As would be expected in the case of lipophilic materials the effect of this metabolic process is not as great as for metabolism in the stratum corneum. This is reflected in the two curves for K = 100 in Figs. 2A and 3A. It is interesting to note that the combination of diffusion coefficient in the viable epidermis taken with $\kappa_m = 0.1$ produces curves which have the same limit at $\tau \to \infty$. Using these conditions only one sixth of the original active drug reaches the dermal–epidermal junction irrespective of partition coefficient.

The following two figures (Figs. 4A and 5A) show the effect of varying the metabolic rate constant whilst maintaining a fixed drug partition coefficient, arbitrarily chosen to

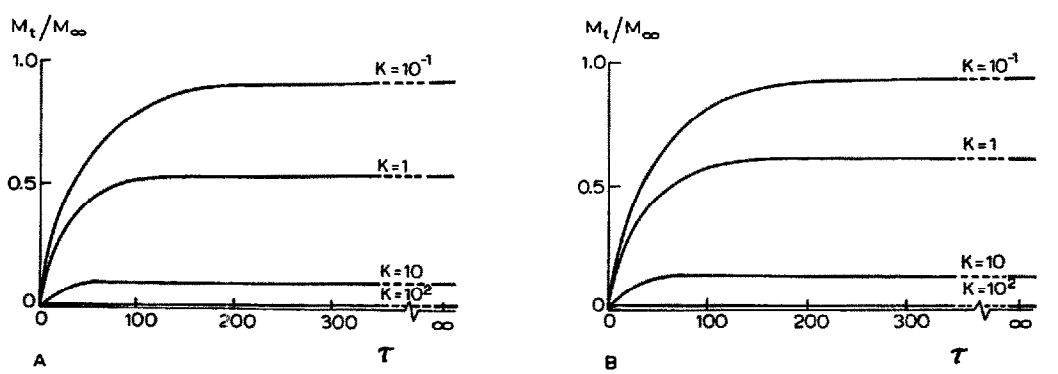

Fig. 2. A: release profile showing the effect of partition coefficient with a fixed metabolic rate in the stratum corneum ($\kappa'_m = 0.1$) for the transcellular route. B: as Fig. 2A but for the intercellular route.

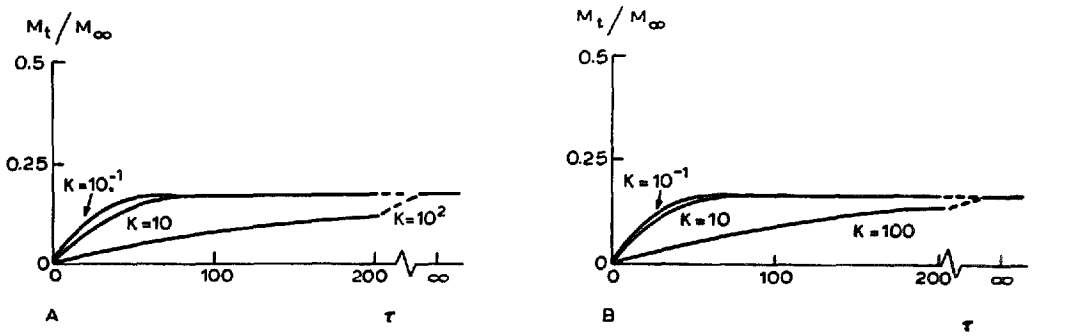

Fig. 3. A: release profile showing the effect of partition coefficient with a fixed metabolic rate in the viable epidermis ($\kappa_m = 0.1$) for the transcellular route. B: as Fig. 3A but for the intercellular route.

be 100. For comparison the curves are included that show the cases where there is no metabolic transformation. The rate constants chosen cover the range 4.4×10^{-8} s^{-1} to 4.4×10^{-5} s^{-1} ($\kappa'_m = \kappa_m = 10^{-3}$–$10^{-1}$). As the rate constant gets progressively faster the amount of drug reaching the capillary network decreases. For $\kappa'_m = 10^{-3}$, 46% of the total starting material will be metabolized but it will also be seen that just over a quarter of the drug will still be present in the stratum corneum after 9 days and the reservoir effect may still be observed. By increasing the rate constant by a factor of 10 only 10% of drug will reach the dermal–epidermal boundary and most of this will have arrived at that site within 4 days. It would be unlikely that a depot effect would be seen under these conditions.

Fig. 5A shows the same profiles for the case where metabolism is occurring in the viable epidermis. It is immediately apparent that the metabolic fate of the drug is far less pronounced. For the slowest rate constant, $\kappa_m = 10^{-3}$, there is very little metabolism and up to 9 days the release profile is coincident with the curve for no metabolism. Only a small difference is seen after infinite time when the figure shows that 95% of the original drug arrives in the blood stream; this should be compared with the 54% that arrives under similar conditions shown in Fig. 4A. Even increasing the rate constant by an order of

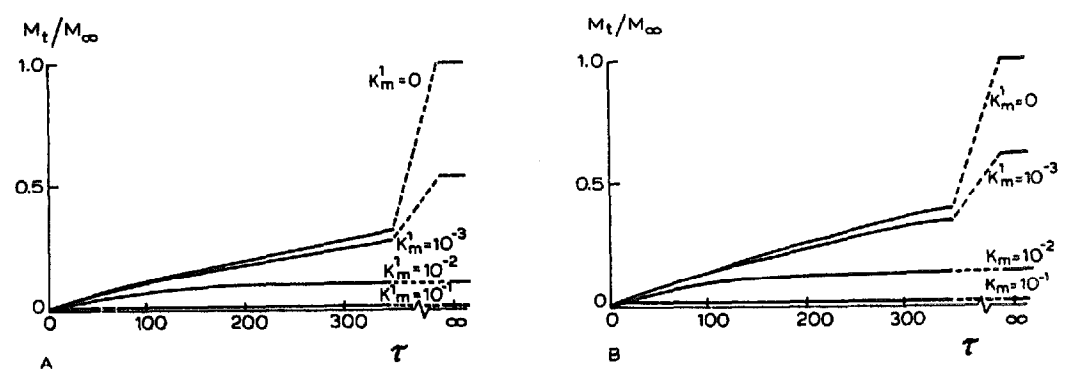

Fig. 4. A: release profile showing the effect of metabolic rate constant in the stratum corneum (the partition coefficient K = 100) for the transcellular route. B: as Fig. 4A but for the intercellular route.

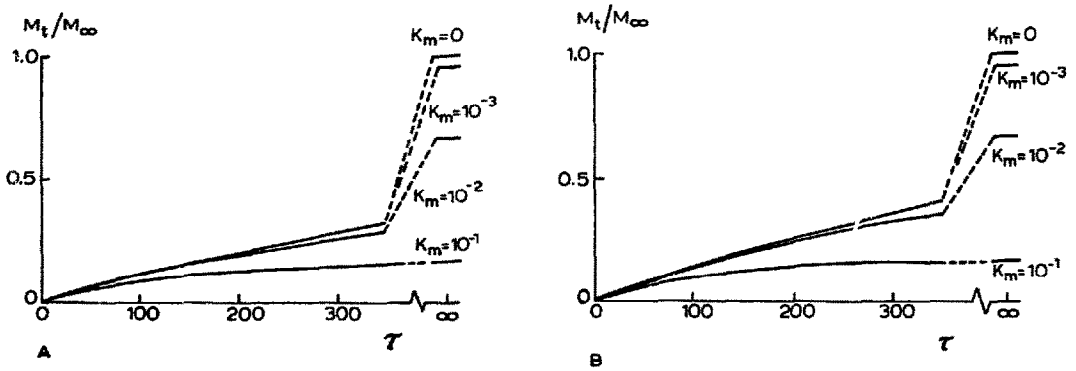

Fig. 5. A: release profile showing the effect of metabolic rate constant in the viable epidermis (the partition coefficient K = 100) for the transcellular route. B: as Fig. 5A but for the intercellular route.

magnitude has little effect over a 9 day period and for both of these cases a significant reservoir persists in the stratum corneum. For the fastest rate constant, $\kappa_m = 0.1$, significant metabolism is occurring and it seems unlikely that the depot would manifest itself for these conditions.

INTERCELLULAR ROUTE

The same profiles have been calculated for the route where the drug diffuses round the dead cells in the stratum corneum via the intercellular channels. In a previous publication (Hadgraft, 1979b) it has been shown that the stratum corneum/water partition coefficients measured and quoted in the literature assume that the whole of the skin sample is involved in the distribution process. If transport is via the channels and the drug is only stored in these regions the quoted partition coefficients have to be corrected by a volume correction factor γ.

$$K_{channels}/K_{measured} = V_{total}/V_{channels} = \gamma$$

γ is typically the order of 7 and this factor has been included in all the profiles labelled 'B'. The curves are thus derived from the formula given by Eqn. 30 but substituting γK for K.

The profiles given in Figs. 1B–5B all exhibit similar properties to their counterparts Figs. 1A–5A and the discussion of the effects applies equally to both routes. The only difference between the two sets of curves is that at a given time the amount of drug that has reached the capillary network is slightly larger for the intercellular route. This is demonstrated by comparing Figs. 2A and 2B; for K = 1 the amount of drug released after an infinite time lapse is 54% for the transcellular route and 62% for the intercellular route. Where metabolism occurs in the viable epidermis the route has no effect on the total amount of drug reaching the capillary network at $\tau \to \infty$ (Figs. 5A and 5B). However, for a given time interval (e.g. $\tau = 300$, $\kappa_m = 10^{-2}$) 26% has been released for the transcellular route and 32% for the intercellular route.

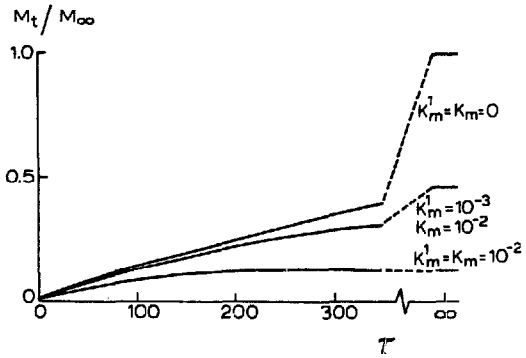

Fig. 6. Release profile showing the effect of metabolic rate constants in both the stratum corneum and the viable epidermis for the intercellular route (K = 100).

Fig. 6B shows that Eqn. 30 can be used to predict release curves assuming metabolism occurs in both the stratum corneum and the viable epidermis. If $\kappa_m = \kappa'_m = 10^{-2}$ it is seen that the metabolism in the viable epidermis contributes a negligible amount. For the slower rate constants, eg. $\kappa'_m = 10^{-3}$ the release profiles can be modified by a faster rate process in the viable epidermis (eg. $\kappa_m = 10^{-2}$).

In any calculation it is thus necessary to balance the different factors involved to see where the major effects exist. More work needs to be conducted to quantify the metabolic rate constants and to find the exact location of the enzymes both in intact skin and in the diseased state. With judicial drug design it should be possible to modify drug structure so its metabolism to inactive materials is minimized or by using the profiles described above to tailor a drug so that it is metabolized to an active substance at its site of action.

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