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Topical delivery of caffeine from some commercial formulations

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Abstract

Permeation of caffeine through human skin and artificial membranes (mounted in modified Franz type diffusion cells) was evaluated, either from saturated solutions or from commercially available topical formulations (all containing 3% caffeine). Data interpretation of the caffeine diffusion through human skin does not implicate transfer through pores despite caffeine being a relatively polar molecule. No correlation was found between transfer though the synthetic membranes (cellulose acetate impregnated with isopropyl myristate and silicone rubber soaked in isopropyl myristate) and that observed through skin. The synthetic membranes can be used for assessing product performance in quality assurance but will give little indication of its performance in vivo. The study investigated the percutaneous permeation of caffeine through human skin in order to obtain a mechanistic interpretation of its route of permeation. Synthetic membranes were also examined to determine if they could be used as models for human skin. Different commercial formulations investigated to determine the significance of enhancement strategies.

Keywords: Percutaneous absorption; Topical delivery; Caffeine; Formulation effects

1. Introduction

Caffeine is found in a number of commercial preparations and we therefore decided to examine its permeation through excised human skin, cellulose acetate and silicone membranes. In order to obtain a mechanistic evaluation we followed diffusion from a saturated aqueous solution followed by a series of commercial preparations, the exact composition of which were not fully known. It is well recognized that the flux of a permeant is related to its applied concentration and its inherent permeability (k_p) through the skin. The permeability of a material from an aqueous

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environment can be estimated from its octanol water partition coefficient (K_{oct}) and its molecular weight (MW). The following equation (Potts and Guy, 1992) gives the permeability coefficient:

$$\log(k_p/\mathrm{cm\cdot h^{-1}}) = -2.7 + /71 \log K_{oct} - 0.0061MW \quad (1)$$

The log [octanol water partition coefficient] can be predicted from numerous software algorithms. Advanced Chemistry Development Labs. (Toronto, Canada) software gives a predicted value of -0.07 and, in its Sangster literature database, provides a recommended measured value also of -0.07. As such caffeine is a rather hydrophilic permeant and may be expected to permeate through any polar channels if they exist in the skin. The calculated value for k_p is therefore 1.16×10^{-4} cm/h. Since caffeine has relatively high water solubility a reasonable flux through skin is anticipated.

Measuring the absolute bioavailability of topical formulations is difficult but inferences can be made from in vitro experiments using human skin. If permeation enhancement strategies have been used in the formulation these should manifest themselves by producing significant deviations from k_p, either estimated as above, or, preferably measured using an aqueous solution of the permeant. The data produced during in vitro experiments can be analyzed to show how the barrier function of the skin has been modulated.

The release characteristics of topical products can be assessed for quality assurance by using membranes other than skin (Guy and Hadgraft, 1990). However a synthetic membrane has yet to be identified which has diffusional characteristics which can be directly correlated to human skin. The usual types of membranes employed are those with porous characteristics, e.g. cellulose acetate or homogeneous permeable polymers such as silicone.

In this publication we have conducted a systematic study on the various types of membranes and correlated the results where possible.

2. Materials and methods

Caffeine was obtained from Boehringer Ingelheim. Somma lotion (Natura, Brazil), Elancyl gel (Vichy, France) and Cellactia lotion (Pierre Fabre, France) were the commercial products investigated. Diffusion experiments were conducted in all glass 'Franz' type diffusion cells with an active surface area of 1 cm^2 and a receptor phase volume of 4 ml. The continuously stirred receptor medium was isotonic phosphate buffered saline (pH 7.4) prepared from materials of analytical grade (Merck, Germany). The receptor compartment was thermostated to 37°C. The membranes used were 0.2 μm cellulose acetate (Gelman Sciences); thickness 110 μm, and silicone (Technical Products Inc.); thickness 127 μm. Prior to the diffusion experiments the membranes were soaked in isopropyl myristate (Merck, Germany) for 12 h. Excised human upper leg skin was heat separated using conventional techniques to produce epidermal membranes.

In the experiments with saturated solutions, 1 ml was applied to the donor compartment. Where the products were tested 100 mg was applied to the surface of the membrane.

The diffusion experiments (four replicates) were conducted over a 24-h period with samples removed, in general, for analysis at 1, 2, 4, 6, 8, 10, 12 and 24 h. The samples were analyzed for caffeine using HPLC (Injector, Merck Hitachi AS-2000A, Detector, Merck Hitachi L-4000UV, Integrator, Merck Hitachi D-7500, Pump, Merck Hitachi L-6000A). A reverse phase column was used (Merck Lichrospher 100RP 18, 125×4 mm, 5 μm), a flow rate of 1 ml/min and UV detection at 273 nm. The retention time was approximately 4 min.

3. Data analysis

The skin experiments were analyzed by fitting (Easyplot, Spiral software) the first five terms of the diffusion equation in Eq. (2) to the data generated during the first 6 h. This time period was chosen so that non steady state diffusion was dominant which provides more confidence in sep-

arating the data into partition and diffusion effects.

$$u = c_{app}\left(\alpha t - \frac{1}{6} - \frac{2}{\pi^2}\sum_{n=1}^{\infty}\exp(-n^2\pi\beta t)\right) \quad (2)$$

where u is the concentration in the receptor phase at time t, c_{app} is the applied concentration, α and β are the two unknowns $\alpha = Kh$ and $\beta = D/h^2$, K is the partition coefficient between the skin and the applied formulation and D is the diffusion coefficient in the skin.

The product $\alpha\beta$ is equal to the permeability coefficient k_p.

The data can therefore be deconvoluted to provide information about how the formulations affect the permeability barrier of the skin. α shows the effect on partitioning and β the effect on diffusion properties. For the analysis, the individual diffusion experiments each provided estimates of α and β from which mean values were calculated.

4. Results and discussion

4.1. Saturated solutions

The solubility of caffeine was found to be 25.82 mg/ml at 32°C. This temperature was chosen since it reflects the temperature found within the donor phase of the Franz type cells. The simplest experiments were those with a saturated solution of caffeine. The results for transfer through epidermis are shown in Fig. 1.

The results in Fig. 2 show the transfer across a silicone membrane pre-soaked in isopropyl myristate.

The other synthetic membrane examined was cellulose acetate impregnated with isopropyl myristate. The data for this are shown in Fig. 3.

It is immediately apparent that the diffusion through epidermal tissue is significantly slower than through the synthetic membranes. The variability in permeability is larger for the epidermis, as expected, and is comparable to that described in the literature. The steady state flux for epidermal tissue and silicone can be estimated by linear regression through the data points obtained from

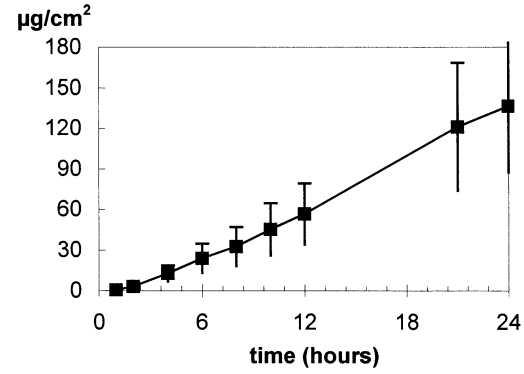

Fig. 1. The transfer of caffeine from a saturated solution through human epidermis ($n = 4$). The error bars show S.D.

8 to 24 h when steady state has been achieved. The values produced are, respectively: 6.65 ± 0.16 and 57.55 ± 1.13 µg/cm²/h. Using the solubility of 25.8 mg/ml, these values can be converted to permeability coefficients (cm/h) of, respectively: 2.58×10^{-4} and 2.23×10^{-3}. The former compares favourably with the value predicted by the Potts and Guy equation (Eq. (1)) of 1.16×10^{-4}.

Fig. 3 shows that the permeation of caffeine through the cellulose acetate membrane is very rapid, there is no detectable lag-time and during 24 h over one third of the applied caffeine has diffused through the membrane. The depletion in concentration reduces the driving force for diffusion and the flux noticeably decreases with time after 5 h. The concentration profile changes expo-

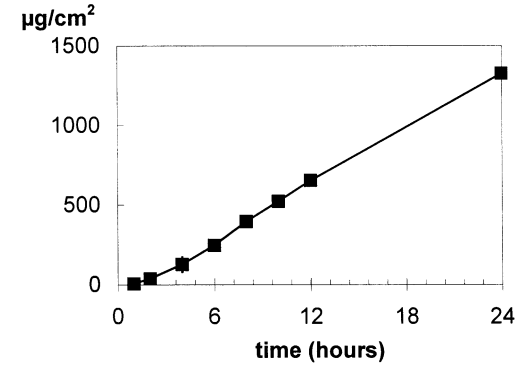

Fig. 2. The transfer of caffeine from a saturated solution through a silicone membrane pre-soaked in isopropyl myristate ($n = 4$). The error bars show S.D.

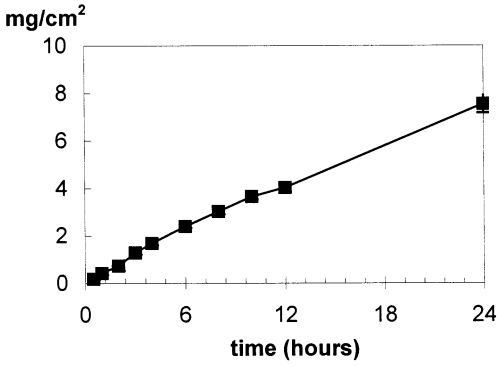

Fig. 3. The transfer of caffeine from a saturated solution through a cellulose acetate membrane impregnated with isopropyl myristate ($n = 4$). The error bars show S.D.

nentially with time with a rate constant 0.029 h^{-1} and a predicted plateau of 13.9 mg cm^{-2}.

The data from Figs. 1 and 2 can be analyzed in more detail using Eq. (2) and the time points up to 6 h which contain non-steady state diffusion information. Table 1 shows the values of α, β and $\alpha\beta$ ($= k_p$) obtained for epidermal membranes and silicone.

The value of k_p obtained from this method compares favourably with that given above using the steady state flux.

Considering the silicone membrane first, further information can be obtained since the thickness of the membrane is known (127 μm). Using $\alpha = Kh$ and $\beta = D/h^2$, estimates of K and D can be obtained:

$K = 2.6$ and $D = 4.6 \times 10^{-9}$ cm^2/s.

Similar calculations are not possible for the epidermal membrane. The rate-limiting barrier is in the stratum corneum and there is still debate about the route of penetration through this domain. If diffusion is straight through the cells the pathlength is approximately 15 μm; for transfer through the intercellular channels, the pathlength has been estimated to be between 350 (Albery and Hadgraft, 1979) and 880 μm (Potts and Francoeur, 1991). It is possible to calculate K_{sc} and D using various values for the pathlength and, as examples, two have been chosen in Table 2, a short pathlength, 15 μm, the thickness of the stratum corneum and 500 μm, a tortuous pathway between the two quoted ranges above.

Table 2
Values for partition between the stratum corneum and water and diffusion within the stratum corneum assuming diffusional pathlengths of 15 and 500 μm

	$h = 15$ μm	$h = 500$ μm
K_{sc}	1.79	0.05
D (cm^2/s)	5.5×10^{-11}	6.2×10^{-8}

Considering the partition coefficients, either value seems reasonable. The value for $h = 15$ μm suggests that the stratum corneum is more lipophilic than octanol but not as lipophilic as silicone soaked in isopropyl myristate. The second value suggests that stratum corneum is not as lipophilic as octanol. The partition coefficient for compounds between stratum corneum and water (K_{sc}) can be estimated (Roberts et al., 1996):

$$\log K_{sc} = -0.024 + 0.59 \log K_{oct} \quad (3)$$

This formula shows that the stratum corneum is not as lipophilic as octanol, which was explained by Roberts et al. (1996) as the solute only being partially desolvated, when it moved from water into the stratum corneum.

Table 1
Diffusion and partition data derived from Figs. 1 and 2 using Eq. (2)[a]

Membrane	α	β	$\alpha\beta = k_p$ (cm/h)
Epidermis	0.0025 ± 0.00098	0.089 ± 0.007	$2.21 \times 10^{-4} \pm 9.6 \times 10^{-5}$
Silicone	0.033 ± 0.021	0.102 ± 0.061	0.0025 ± 0.0004

[a] Values quoted are the mean from four replicates and the S.D.

Table 3
Diffusion and partition data for caffeine through skin from the different preparations derived from Fig. 4 using Eq. (2)[a]

Preparation	α	β	$\alpha\beta = k_p$ (cm/h)
Cellactia	0.0015 ± 0.0005	0.144 ± 0.048	$1.98 \times 10^{-4} \pm 3.27 \times 10^{-5}$
Elancyl	0.011 ± 0.004	0.0998 ± 0.013	$1.07 \times 10^{-3} \pm 0.0034$
Somma	0.0015 ± 0.0009	0.092 ± 0.038	$1.15 \times 10^{-4} \pm 3.12 \times 10^{-5}$

[a] Values quoted are the mean from four replicates and the S.D.

It is more instructive to consider the values of D. The value obtained for $h = 15$ μm seems very low for an aqueous shunt pore. The value of D for the longer pathlength compares favourably with that quoted by Potts and Francoeur (1991) of 1.5×10^{-7} cm^2/s for water in stratum corneum.

The data generated in this analysis for caffeine are consistent with a tortuous path through the intercellular channels of the stratum corneum.

4.2. Formulations

The permeation of caffeine from the different commercial formulations through epidermal membranes is shown in Fig. 4.

It is apparent that there is a significant difference between Elancyl and the other two preparations. Data analysis should indicate how this is achieved. The values of α and β obtained for the preparations are shown in Table 3.

Analysis of variance shows that there is no significant difference for α,β or $\alpha\beta$ between a saturated solution of caffeine, Cellactia and Somma. The two latter formulations do not appear to possess any excipients that enhance the permeation of caffeine through the skin. There is a significant difference for α, and for $\alpha\beta$ between Elancyl and the other preparations including the saturated solution. No difference was apparent for β.

Excipients in the Elancyl preparation appear to increase the partitioning of the caffeine into the stratum corneum by a factor of 4 to 7. They do not appear to alter the diffusional barrier of the skin. Fig. 4 shows that the permeation rate from Elancyl decreases after 12 h, the reasons for this are not fully understood but may be a result of either:

1. loss of the enhancer from the stratum corneum;
2. inability of the formulation to provide caffeine to the skin surface, this may be a result of slow diffusion of the caffeine through the formulation. In the in vitro experiment, the formulation is dosed at 100 mg/cm^2, which results in quite a thick application.

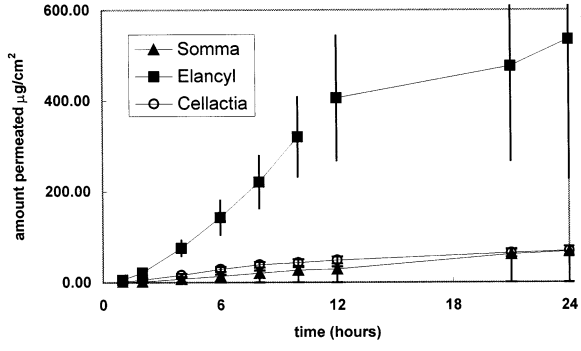

Fig. 4. The permeation of caffeine from three commercial formulations through human epidermal membranes ($n = 4$ ± S.D.).

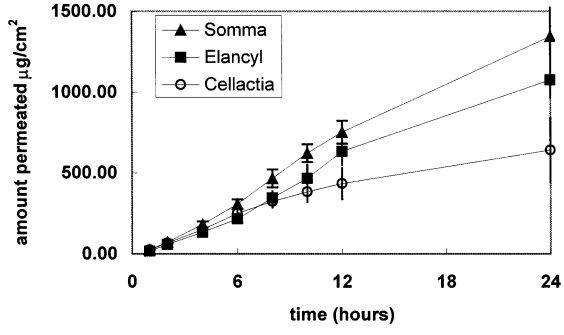

Fig. 5. The permeation of caffeine from three commercial formulations through silicone impregnated with isopropyl myristate ($n = 4$ ± S.D.).

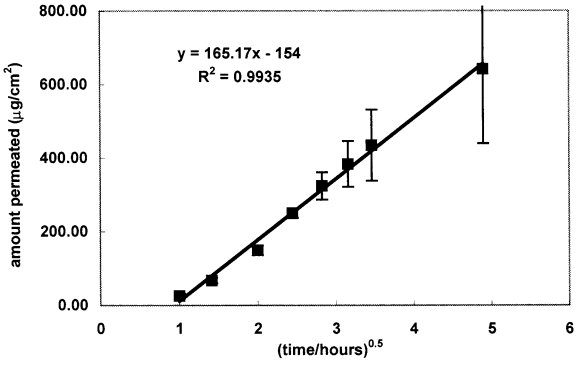

Fig. 6. The relationship between amount of caffeine permeated through a silicone membrane impregnated with isopropyl myristate and the square root of time.

The permeation of caffeine from the different commercial formulations through silicone impregnated with isopropyl myristate is shown in Fig. 5.

An analysis of variance on the 24-h data points shows that, for the silicone membrane there is no significant difference between a saturated solution and Somma or Elancyl. This suggests that the preparation provides caffeine to the membrane surface at a rapid rate and that, although only 3% caffeine is present, it is at or above its saturated solubility in the formulation. For Cellactia, the levels permeated are significantly lower, this is a result of the drug being present in the formulation at a sub-saturated level or because diffusion through the formulation is slower than penetration through the membrane. If the latter is the case, the data should follow a linear relationship with the square root of time (Guy and Hadgraft, 1990). Fig. 6 shows that transformation of the data does give a linear relationship indicative of control from diffusion in the formulation.

A comparison between the epidermal permeation and silicone permeation shows no correlation. This synthetic membrane cannot be used predictively to estimate the efficiency of formulations on skin.

Penetration of the caffeine through the cellulose membrane is even more rapid; this was also apparent for the saturated solution of caffeine (Fig. 3). Only two of the formulations were examined, the data for these are shown in Fig. 7.

The data reflect depletion of the driving concentration in the donor compartment and as such are fit very well using a simple first order equation. The associated rate constants are 0.31 and 0.25 h^{-1} for, respectively, Elancyl and Cellactia. The difference between the two gives an indication that diffusion through the cellulose acetate is faster for the Elancyl than Cellactia but again no direct correlation can be made between these data and those for skin. The difference between the equilibrium values is indicative of the way in which the caffeine partitions between the formulation and the aqueous environment of the receptor phase.

5. Conclusions

The data for caffeine penetration through the skin can be analyzed to show the relative effects of partition and diffusion. In the straightforward experiments, saturated aqueous solutions, the values suggest that there is no evidence for caffeine diffusing through aqueous pores in the skin. The value of the permeability coefficient obtained is comparable to that predicted by the Potts and Guy Eq. (1) which also does not indicate any aqueous pore mechanism in percutaneous penetration.

The use of synthetic membranes is useful in assessments of batch-to-batch variation in quality assurance but gives no indication of how a formulation will behave when it is used on skin.

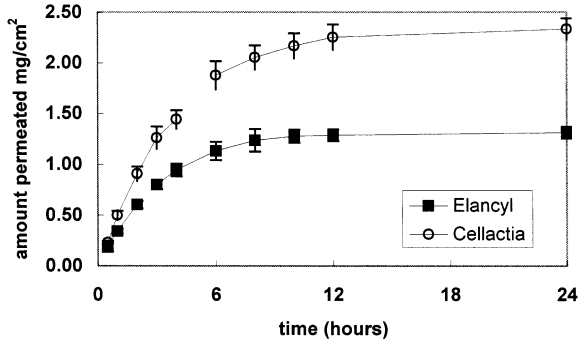

Fig. 7. The permeation of caffeine from Elancyl ($n = 4$) and Cellactia ($n = 3$) through a cellulose acetate membrane impregnated with isopropyl myristate.

The transport through silicone impregnated with isopropyl myristate more rapid than through epidermis. The diffusion coefficient determined is reasonable for a membrane such as silicone. The diffusion coefficient of caffeine through a tortuous intercellular channel is of the same order of magnitude as that determined for water permeation in skin.

The fastest permeation was noted for a cellulose acetate membrane impregnated with isopropyl myristate. This could be anticipated since diffusion is through the liquid medium that isopropyl myristate provides in the porous nature of the membrane. Transport from the topical formulations in contact with the impregnated membrane is much greater than the saturated solution. This may be a result of the isopropyl myristate being abstracted by solvents in the formulations. This is always a concern when using this type of solid supported liquid membrane. When the contacting fluid is water, there is no problem. Often however, pharmaceutical formulations contain solvents and surfactants that can destroy the integrity of this type of membrane. Caution has to be given to the experimental design in this approach.

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