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Prediction of Skin Permeability of Drugs: Comparison of Human and Hairless Rat Skin

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Abstract—Relationships between skin permeability and physicochemical properties of drugs were examined to establish a predictive method for the steady-state permeation rate of drugs through human skin. Human skin permeation properties fell into two categories: one in which the permeability coefficient is correlated to the partition coefficient, revealed with lipophilic drugs; and the other in which the permeability coefficients are almost constant, shown with hydrophilic drugs. The stratum corneum, the main barrier in skin, could be considered as a membrane with two parallel permeation pathways: lipid and pore pathways, and an equation for predicting the steady-state permeation rate of drugs was derived. The skin permeabilities of drugs for man were compared with those for hairless rat. The species difference in skin permeability found was suggested to be due to the difference in skin permeation pathways, since lipid content and water uptake of the stratum corneum varied between human and hairless rat skin.

In the present study, we have attempted to establish a predictive method for the steady-state permeation rate of drugs through human skin. This method is based on an assumption that the stratum corneum, the main barrier in skin (Blank 1965), is a membrane having two parallel permeation pathways: lipid and pore pathways. Excised human skin and several drugs were used, and the relationships between the skin permeability from aqueous suspensions and physicochemical properties of drugs were examined. To characterize human skin permeability more thoroughly, the permeation properties and nature of the stratum corneum, such as lipid content and water uptake, of human skin were compared with those of hairless rat skin.

Materials and Methods

Materials

Drugs used in this study and their physicochemical properties are shown in Table 1. Their sources were as previously reported by Hatanaka et al (1990).

Skin membrane preparation

Hairless rat skin was freshly excised from the abdomen of male WBN/ILA-Ht rats (Life Science Research Center, Josai University, Saitama, Japan) 6–7 weeks of age.

Human skin was obtained following unrelated surgical operations (Department of Surgery, School of Medicine, Kitasato University, Kanagawa, Japan); sources were the chest of 37–75 year old female patients. The human skin samples were stored at −20°C until used. Before use in the permeation experiment, the dermis side was washed with distilled water. It was confirmed that the treatment would not affect the skin permeability of drugs to be tested.

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Skin permeation procedure

Skin permeation experiments were performed according to the method of Okumura et al (1989). A diffusion cell was used which consisted of two half-cells with a water jacket connected to a water bath at 37°C. Each half-cell had a volume of 2·0 mL and an effective area of 0·87 cm^2. The dermis side of the skin was in contact with the receiver compartment and the stratum corneum with the donor compartment. The receiver compartment of each cell was filled with distilled water and the donor compartment with the drug suspension in distilled water (at 2–10 times the amount required for saturation), to assure constant thermodynamic activity throughout the experiment. Both compartments were stirred with a star-head bar driven by a constant-speed synchronous motor (MC-301, Scinics, Tokyo) at 1200 rev min^{-1}. Each experiment lasted for 10 h to achieve a steady-state-permeation. A sink condition was always maintained in the receiver compartment.

Isolation of stratum corneum

The stratum corneum of hairless rat and human skin was isolated by trypsin treatment (Knutson et al 1985). The dermis side of whole skin was placed on a filter paper saturated with 1% (w/v) trypsin solution (10000 ATEE units mL^{-1} in phosphate buffered saline (PBS), at pH 7·4) at 37°C in a sealed Petri dish for 8 h. At the end of this period, the stratum corneum was carefully separated from the viable epidermis and rinsed thoroughly with distilled water. The stratum corneum samples were dried for 24 h and stored in a desiccator at room temperature (about 20°C) until used.

Measurements of lipid content and water uptake

Lipid content and water uptake in the stratum corneum were measured by the method of Raykar et al (1988) with slight modification. Preweighed dry stratum corneum samples (about 10 mg) were placed in 10 mL screw-cap glass tubes containing 7 mL of 2:1 chloroform/methanol mixture and shaken for 20 h at room temperature. At the end of this

period, the delipidized stratum corneum samples were removed, rinsed with fresh chloroform/methanol mixture, and dried to a constant weight. The lipid content of the stratum corneum was determined by its change in weight after solvent extraction.

Accurately weighed stratum corneum samples (about 10 mg) were placed in glass tubes containing 7 mL of water and equilibrated at 37°C for 72 h. At the end of this period, the samples were gently blotted to remove excess water, and immediately weighed. Water uptake was calculated by weight change in the stratum corneum.

Statistics

Statistical significance was evaluated by a paired *t*-test.

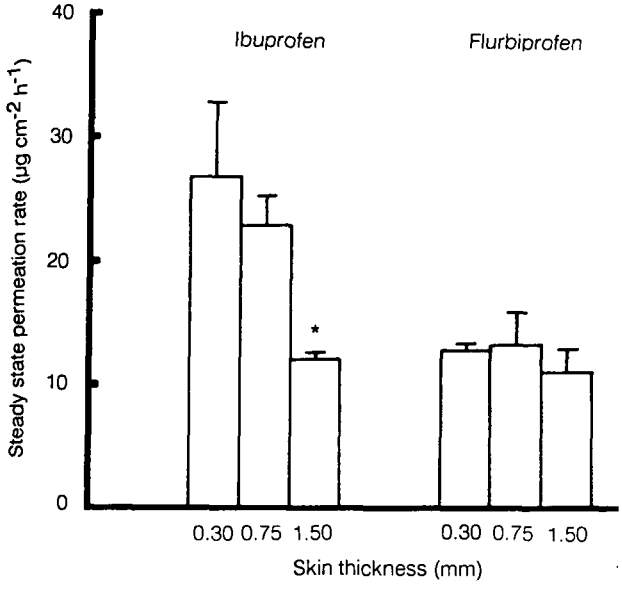

FIG. 1. Permeability of highly lipophilic drugs through full-thickness (about 1·5 mm thick) and split-thickness (about 300 and 750 μm thick) skin. Each column represents the mean ± s.e. of three experiments. *$P < 0.05$ compared with 300 μm.

Results

Influence of dermis barrier on overall human skin permeability of drugs

In the in-vitro permeation experiment using human skin, the resistance of dermis to the overall skin permeation of drug can not be ignored. It is important to note that the dermis of human skin is thicker than that of hairless rat skin (Sato et al 1991). Permeability of highly lipophilic drugs, such as ibuprofen and flurbiprofen, through full-thickness skin (about 1·5 mm thick) was compared with that through split-thickness skin (about 300 and 750 μm thick) (Bronaugh & Stewart 1984). Fig. 1 shows the influence of dermis on the permeability of ibuprofen and flurbiprofen through human skin. Although the steady-state permeation rate increased by cutting the dermis side, significant difference was not found in the values for split-thickness skin between 300 and 750 μm thick for either drug. The following experiments were carried out using the split-thickness skin (about 750 μm thick).

Relationships between human skin permeability and physicochemical properties of drugs

The permeation profiles of drugs through human skin are shown in Fig. 2. The steady-state permeation rate and permeability coefficient of each drug was calculated, and the contribution of physicochemical parameters of each to the skin permeability were evaluated.

Skin permeation of a drug is determined by two potential mechanisms, solution-diffusion and pore penetration, both dependent on the nature of the skin barrier, the stratum corneum (Blank 1965). If the stratum corneum is regarded as a solution-diffusion membrane, the steady-state permeation rate of drug (dQ/dt) is mathematically expressed by equation 1 based on Fick's first law:

$$dQ/dt = D_m K C_v/L \quad (1)$$

where D_m, K and C_v are respective to the diffusion coefficient of drug in the stratum corneum, the stratum corneum/vehicle partition coefficient of the drug and the solubility of the drug

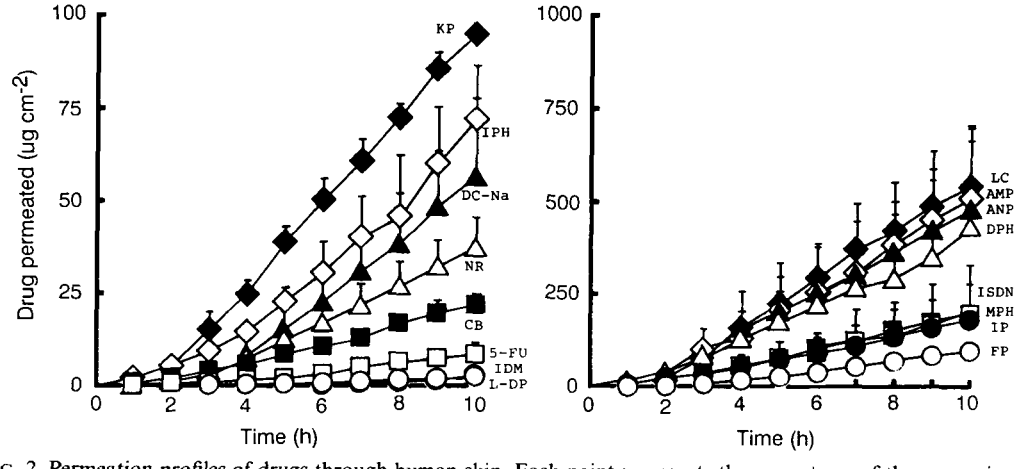

FIG. 2. Permeation profiles of drugs through human skin. Each point represents the mean ± s.e. of three experiments. Abbreviations for drug names in the figure are as follows: IP, ibuprofen; FP, flurbiprofen; IDM, indomethacin; KP, ketoprofen; LC, lignocaine; ISDN, isosorbide dinitrate; CB, cyclobarbitone; AMP, aminopyrine; 5-FU, 5-fluorouracil; DC-Na, diclofenac sodium; NR, nicorandil; ANP, antipyrine; MPH, morphine hydrochloride; IPH, isoprenaline hydrochloride; DPH, dopamine hydrochloride; L-DP, levodopa.

Table 1. Physicochemical parameters of drugs.

	Mol. wt	Solubility in water[a] (mg mL^{-1})	Distribution coefficient[b]	Melting point (°C)	Activity of crystal (g mL^{-1})	Solubility in isooctane[c] (μg mL^{-1})
Ibuprofen	206·27	0·0430	3·94	74·0	6·74	4·58
Flurbiprofen	244·27	0·0277	3·86	113·0	2·31	3·12
Indomethacin	357·81	0·0111	3·19	158·5	0·473	0·126
Ketoprofen	254·29	0·185	3·11	93·0	3·84	2·38
Lignocaine	234·33	3·03	2·37	65·0	17·3	6·25
Isosorbide dinitrate	236·14	1·34	1·34	69·0	7·69	3·29
Cyclobarbitone	236·26	3·07	0·873	171·0	6·71	1·16
Aminopyrine	231·29	55·9	0·497	102·5	1·86	3·76
5-Fluorouracil	130·08	17·1	−0·860	282·6[e]	—	−3·03
Diclofenac sodium	296·15[d]	32·0	−0·962	284·0[e]	—	−1·81
Nicorandil	211·17	39·6	−1·02	85·0	5·73	0·721
Antipyrine	188·23	816	−1·55	105·5	2·97	2·78
Morphine hydrochloride	339·39[d]	82·5	−2·53	200·0[e]	—	−2·97
Isoprenaline hydrochloride	211·24[d]	345	−2·69	165·7[e]	—	−2·16
Dopamine hydrochloride	153·18[d]	520	−3·40	243·6[e]	—	−2·70
Levodopa	197·00	5	−4·70	294·0[e]	—	−3·43

[a] Solubility in water at 37°C. [b]Logarithm of octanol/water distribution coefficient at 37°C.
[c] Logarithm of solubility in isooctane at 37°C. [d]Mol. wt of corresponding un-ionized form.
[e] Decomposition temperature.

in the vehicle, and L is the stratum corneum thickness. L can be considered to be almost the same for each skin sample. D_m can also be regarded to be constant because molecular weights of the drugs used in this study are similar (Table 1) (Baker 1987). Thus, only K and C_v are the practical factors influencing dQ/dt. On the other hand, if the stratum corneum is assumed to be a porous membrane, where a drug diffuses through the membrane pores, equation 1 must be modified as follows:

$$dQ/dt = D_v \varepsilon C_v/\tau L \quad (2)$$

where D_v is the diffusion coefficient of a drug in the vehicle, and ε, τ and L are porosity, tortuosity and thickness of the stratum corneum, respectively. In practice, only C_v becomes an influencing factor on dQ/dt because D_v, ε, τ and L can be considered to be almost the same for each experiment (Baker 1987). In Fig. 3, dQ/dt is plotted against the mol. wt, solubility in water (C_w), and octanol/water distribution coefficient (K_{ow}) of drugs; these are considered indicators of the diffusion coefficient in the stratum corneum and vehicle (D_m and D_v), solubility in the vehicle (C_v), and stratum corneum/vehicle partition coefficient (K), respectively (Hatanaka et al 1990). dQ/dt had a tendency to relate to C_w and a high correlation coefficient ($r = 0{\cdot}894$, $P < 0{\cdot}01$) was obtained only for hydrophilic drugs ($\log K_{ow} < 0$). For lipophilic drugs ($\log K_{ow} \geqq 0$), dQ/dt was also dependent on the mol. wt ($r = -0{\cdot}771$, $P < 0{\cdot}05$). In contrast, dQ/dt was independent of K_{ow} for both hydrophilic and lipophilic drugs.

From a thermodynamic point of view, equations 1 and 2 can be transformed to equations 3 and 4, respectively (Higuchi 1960):

$$dQ/dt = D_m a_v/\gamma_m L \quad (3)$$

$$dQ/dt = D_v \varepsilon a_v/\gamma_v \tau L \quad (4)$$

where a_v is activity of drug in the vehicle, and γ_m and γ_v are activity coefficients of drug in the stratum corneum and

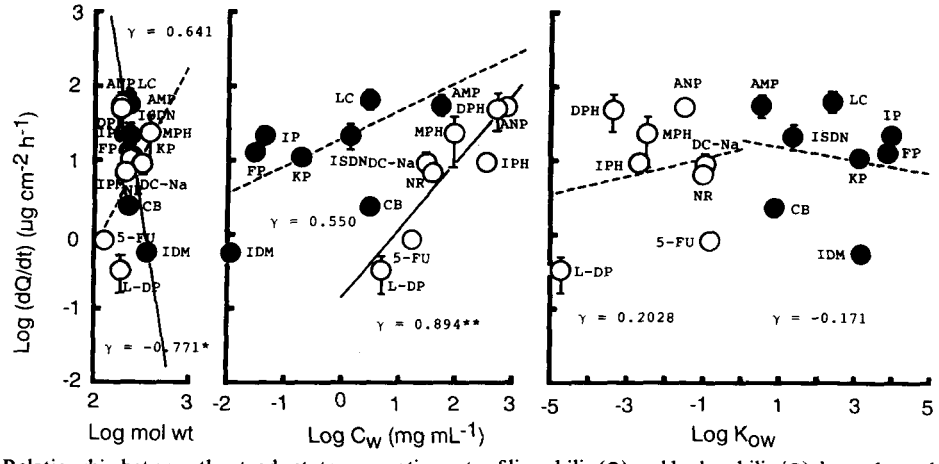

FIG. 3. Relationship between the steady state permeation rate of lipophilic (●) and hydrophilic (○) drugs through human skin and several parameters influencing their permeabilities. Each point represents the mean ± s.e. of three experiments. $*P < 0{\cdot}05$, $**P < 0{\cdot}01$ compared with zero. See Fig. 2 for the key to the abbreviations of the different drug names.

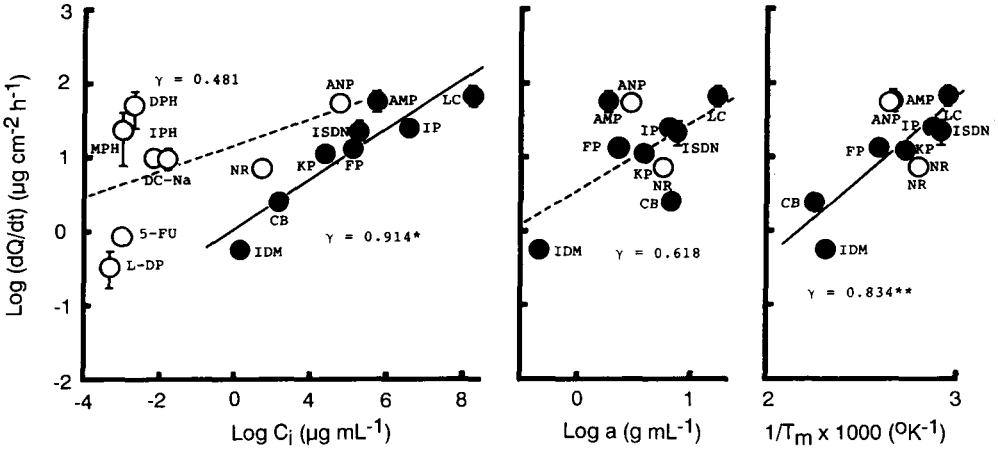

FIG. 4. Relationship between the steady-state permeation rate through human skin and the activity of lipophilic (●) and hydrophilic (○) drugs. Each point represents the mean ± s.e. of three experiments. $*P<0.05$, $**P<0.01$ compared with zero. See Fig. 2 for the key to the abbreviations of the different drug names.

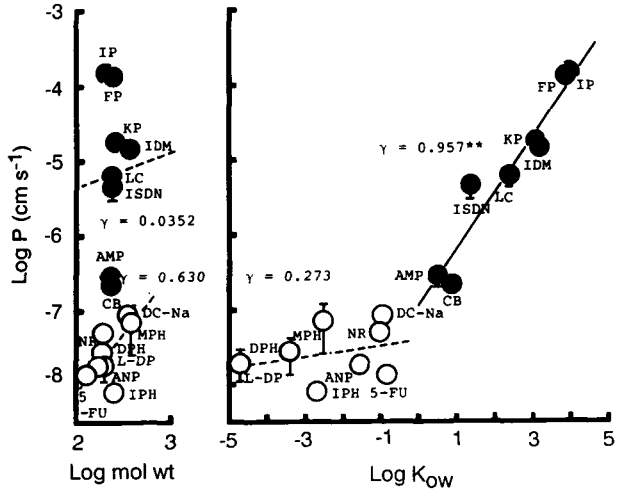

FIG. 5. Relationship between the permeability coefficient of lipophilic (●) and hydrophilic (○) drugs and several parameters influencing their permeabilities. Each point represents the mean ± s.e. of three experiments. $*P<0.05$, $**P<0.01$ compared with zero. See Fig. 2 for the key to the abbreviations of the different drug names.

vehicle, respectively. The relationship between dQ/dt and a_v was then examined (Fig. 4). The solubility of drug in isooctane (C_i), activity of drug in a crystalline state (a) and reciprocal of melting point ($1/T_m$) were employed as indicators for a_v (Rytting et al 1972; Hatanaka et al 1990). Drugs decomposing before melting were not included in this analysis. For lipophilic drugs, although the correlation between dQ/dt and a was low ($\gamma=0.618$, $P>0.05$), linear relations were found between dQ/dt and the other two parameters ($\gamma=0.914$, $P<0.01$ for C_i; $\gamma=0.818$, $P<0.01$ for $1/T_m$). In contrast, dQ/dt was independent of C_i for hydrophilic drugs.

Based on equations 1 and 2, the permeability coefficient of a drug (P) is given by equations 5 and 6, respectively:

$$P=D_m\,K/L \quad (5)$$

$$P=D_v\,\varepsilon/\tau\,L \quad (6)$$

Due to the constancy of D_m, D_v, ε, τ and L in this study, if the stratum corneum is a solution-diffusion membrane, P should increase with increasing K. Similarly, if the stratum corneum is a porous membrane, P should be independent of drug lipophilicity. The relationship between P and physicochemical parameters is shown in Fig. 5. P was little influenced by mol. wt of either lipophilic or hydrophilic drugs. The relationship between P and K_{ow}, however, was linear for lipophilic drugs ($\gamma=0.957$, $P<0.01$), whereas P for hydrophilic drugs was almost constant (about 2.0×10^{-8} cm s^{-1}) and independent of K_{ow}.

Prediction of human skin permeability

The preceding results indicate that human skin permeation of lipophilic and hydrophilic drugs can be described by the solution-diffusion and pore theories, respectively. Therefore, it is suggested that at least two permeation pathways exist in the stratum corneum of human skin as in hairless rat skin (Hatanaka et al 1990) (Fig. 6). The permeability coefficient for such a membrane can be expressed as the total of the permeability coefficients for the two pathways:

$$P=(1-\varepsilon)\,D_L\,K_{LV}/\tau_L\,L+D_V\,\varepsilon/\tau_P\,L \quad (7)$$

where D_L is the diffusion coefficient of the drug in the lipid pathway, K_{LV} is the lipid pathway/vehicle partition coefficient of drug, and τ_L and τ_P are the tortuosities of the lipid and the pore pathways, respectively. Assuming that the

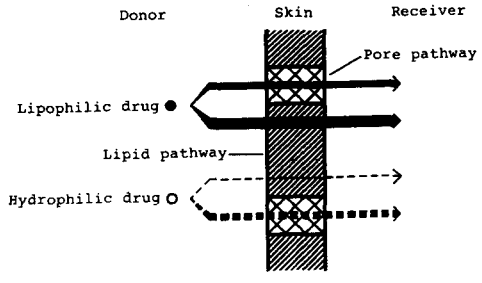

FIG. 6. Permeation model of lipophilic (●) and hydrophilic (○) drugs through human skin.

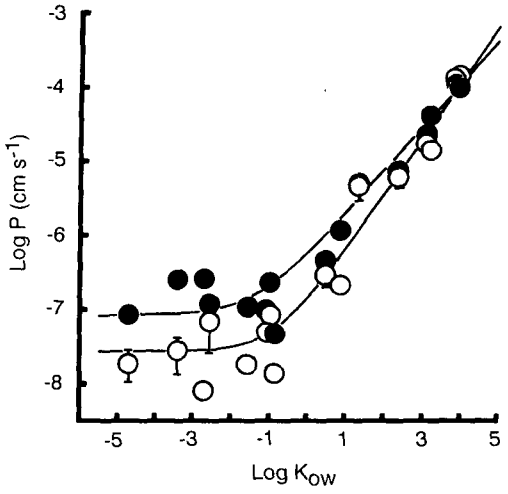

FIG. 7. Relationships between permeability coefficient and octanol/water distribution coefficient of drugs in human (○) and hairless rat (●) skin. Solid lines represent the estimated curves, $P\ (cm\ s^{-1}) = 1{\cdot}17 \times 10^{-7}\ K_{ow}^{0{\cdot}751} + 2{\cdot}73 \times 10^{-8}$ in man, $P(cm\ s^{-1}) = 4{\cdot}78 \times 10^{-7}\ K_{ow}^{0{\cdot}589} + 8{\cdot}33 \times 10^{-8}$ in hairless rat.

Table 2. Comparison of lipid content and water uptake of stratum corneum of human and hairless rat skin.

	Lipid content (%)	Water uptake (mg mL^{-1})
Human chest skin	44·5 ± 1·17	7·08 ± 0·260
Rat abdominal skin	41·2 ± 0·927	8·32 ± 1·51

Each value represents the mean ± s.e. of four tissues.

differences of D_L, D_V, τ_L, τ_P, and L among skin samples and drugs are negligible, and that K_{LV} is calculated from K_{ow} by a linear free energy relationship (Leo & Hansch 1971), equation 7 can be reduced to equation 8:

$$P = A\ K_{ow}^{B} + C \quad (8)$$

where A, B and C are constants. A, B and C are calculated from the skin permeation data in Fig. 5. The final equation becomes:

$$P\ (cm\ s^{-1}) = 1{\cdot}17 \times 10^{-7}\ K_{ow}^{0{\cdot}751} + 2{\cdot}73 \times 10^{-8} \quad (9)$$

Fig. 7 indicates that the relationship between P for human skin and K_{ow} can be described by equation 9, and therefore that dQ/dt for human skin can be predicted from K_{ow} and C_w based on equation 9.

Species difference between hairless rat and man in skin permeability of drugs

Fig. 7 shows the relationship between P and K_{ow} for human and hairless rat skin with the fitted curves (Hatanaka et al 1990). In man, P values of lipophilic drugs were slightly higher than those in hairless rat, whereas those of hydrophilic drugs were remarkably lower.

To clarify the reasons for such a species difference, we looked at the difference in permeation pathways between human and hairless rat skin. The lipid content and water uptake of the stratum corneum for human chest skin were compared with those for hairless rat abdominal skin (Table 2). The lipid content of human stratum corneum was higher than hairless rat, while the water uptake for human skin was lower than hairless rat skin, although no significant differences were found ($P > 0{\cdot}05$).

Discussion

In a previous study, we established a method for predicting hairless rat skin permeability of drugs (Hatanaka et al 1990); however, it is known that such rodent skins do not always have identical permeation properties to those of human skin (Bartek et al 1972; Wester & Noonan 1980; Walker et al 1983). Therefore, we felt our predictive method should be checked by a permeation study using human skin.

At the beginning of this study, an experimental method for human skin permeation was established. In rodent, influence of the dermis on drug permeability, which is usually observed only in an in-vitro skin permeation experiment, can be ignored, because the diffusion in the dermis permeation is relatively high (Flynn et al 1981). However, the dermis of human skin is thick compared with rodent skin (about 2·5 and 0·84 mm, respectively (Sato et al 1991)), and so may act as a significant additional barrier to the permeation of drugs, especially highly lipophilic drugs (Scheuplein & Blank 1973; Bronaugh & Stewart 1984; Scott et al 1986). Skelly et al (1987) proposed that human skin should be used in the form of thinly dermatomed sections ($\leq 0{\cdot}5$ mm thick) or epidermal sections isolated by gentle heat or other methods for in-vitro percutaneous absorption studies. Thus, permeability of highly lipophilic drugs through full-thickness skin was compared with that through split-thickness skin. As shown by our results (Fig. 1), the steady-state permeation rate of ibuprofen and flurbiprofen increased when the dermis side was cut. However, there was no significant difference in the values between 300 and 750 μm thick, suggesting that the permeability through the dermis was too high to affect the total permeability when the dermis was cut to a thickness of 750 μm.

In man, the relationship between skin permeability and lipophilicity of a drug could be roughly classified into the same two types as shown in hairless rat (Hatanaka et al 1990). The stratum corneum could be assumed to be a membrane with a lipid pathway, the main route for the lipophilic drug permeation, which can be described by the solution-diffusion theory, and a pore pathway, the main route for hydrophilic drugs. These assumptions allowed estimation of an equation for predicting human skin permeability of drugs.

We defined the lipophilic and hydrophilic drugs as drugs with $\log K_{ow} \geq 0$ and $\log K_{ow} < 0$, respectively. Some electrolytes such as non-steroidal anti-inflammatory drugs appeared in both classes. Such drugs may dissociate according to the pK_a of the drug and pH of the vehicle. When the donor vehicle contains un-ionized and ionized forms, an apparent or average skin permeability is obtained. Table 3 shows the pK_a of drugs and the pH of aqueous suspension (donor solution) in this study. Hydrophilic drugs with the exception of 5-fluorouracil and antipyrine exist as ionized forms in the aqueous suspension, while lipophilic drugs with the exception of flurbiprofen, indomethacin, ketoprofen and

Table 3. The pK_a of drugs and pH of the aqueous suspension.

	pH	pK_a			
Ibuprofen	4·44	5·20[a]			
Flurbiprofen	4·70	3·73[b]			
Indomethacin	5·15	4·50[a]			
Ketoprofen	3·72	3·90[b]			
Lignocaine	6·82	7·86[a]			
Cyclobarbitone	3·58	7·50[a]			
Aminopyrine	7·94	5·00[a]			
5-Fluorouracil	4·66	8·00	13·0[a]		
Diclofenac sodium	7·96	4·00[c]			
Antipyrine	7·60	1·50[a]			
Morphine hydrochloride	4·22	7·87	9·85[a]		
Isoprenaline hydrochloride	2·75	8·57	10·1	12·0[a]	
Dopamine hydrochloride	3·26	8·74	10·3[b]		
Levodopa	5·42	2·31	8·71	9·74	13·4[a]

[a] Newton & Kluza (1978), [b]Horioka & Fukumuro (1979), [c] Adeyeye & Li (1990).

lignocaine are almost completely un-ionized. We confirmed previously that permeation rates of indomethacin from aqueous suspensions did not change over the pH range 2–5·5 in spite of different concentrations of the ionized form. Similar results were reported by Chiang et al (1991), and may be explained by the fact that the permeability coefficient of the un-ionized form is about 100-fold that of the ionized form.

Although the same skin permeation model was applicable, the skin permeation potential of each drug in man was different from that in hairless rat. In man, permeability of liphophilic drugs was slightly higher than that in hairless rat, whereas that of hydrophilic drugs was remarkably lower than that in hairless rat. Similar results were obtained by Durrheim et al (1980) who compared permeation of alkanols in human epidermis and hairless mouse skin. Bronaugh et al (1982) also pointed out that rat skin was not a good model for human skin for hydrophilic compounds such as urea, but was for lipophilic drugs such as benzoic acid and acetylsalicylic acid. In contrast, Walker et al (1983) reported that skin permeability of paraquat in man was significantly lower than in hairless rat and mice, although the species difference for skin permeability of water was small. They were unable to evaluate accurately the permeability coefficient of paraquat, however, because they used full-thickness human skin. To clarify the reasons for such a species difference, various characteristics of the stratum corneum in each species must be known. Sato et al (1991) indicated that the skin surface lipids have good correlation with the permeability of nicorandil. They also showed that each layer of stratum corneum was thin and the keratinocytes were relatively loose in hairless rat. Also, it was reported that the composition of stratum corneum lipids varied among species (Wertz & Downing 1989). Such a difference may be an important factor in species difference. Further, we noted the ratio of permeation pathways. Lipid content and water uptake were used as indicators of the ratio of lipid and pore pathways, respectively, in the stratum corneum of human and rat skin. Although significant differences were not obtained ($P > 0·05$), the lipid content was slightly higher in man than in hairless rat, and the water uptake was lower. Differences of this type in the ratio of permeation pathways may be associated with species differences in skin permeation.

References

Adeyeye, C. M., Li, P.K. (1990) Diclofenac sodium. In: Florey, K. (ed.) Analytical Profiles of Drug Substances vol. 19 Academic Press, New York, pp 123–144

Baker, R. W. (1987) Controlled Release of Biologically Active Agents. John Wiley & Sons, New York

Bartek, M. J., LaBudde, J. A., Maibach, H. I. (1972) Skin permeability in vivo: comparison in rat, rabbit, pig and man. J. Invest. Dermatol. 58: 114–123

Blank, I. H. (1965) Cutaneous barriers. Ibid. 45: 249–256

Bronaugh, R. L., Stewart, R. F. (1984) Methods for in vitro percutaneous absorption studies III: hydrophobic compounds. J. Pharm. Sci. 73: 1255–1258

Bronaugh, R. L., Stewart, R. F., Congdon, E. R., Giles, A. L. (1982) Methods for in vitro percutaneous absorption studies II: animal models for human skin. Toxicol. Appl. Pharmacol. 62: 481–488

Chiang, C. H., Lai, J. S., Yang, K. H. (1991) The effects of pH and chemical enhancers on the percutaneous absorption of indomethacin. Drug Dev. Ind. Pharm. 17: 91–111

Durrheim, H., Flynn, G. L., Higuchi, W. I., Behl, C. R. (1980) Permeation of hairless mouse skin I: experimental methods and comparison with human epidermal permeation by alkanols. J. Pharm. Sci. 69: 781–786

Flynn, G. L., Durrheim, H., Higuchi, W. I. (1981) Permeation of hairless mouse skin II: membrane sectioning techniques and influence on alkanol permeabilities. Ibid. 70: 52–56

Hatanaka, T., Inuma, M., Sugibayashi, K., Morimoto, Y. (1990) Prediction of the skin permeability of drugs I: comparison with artificial membrane. Chem. Pharm. Bull. 38: 3452–3459

Higuchi, T. (1960) Physical chemical analysis of percutaneous absorption process from creams and ointments. J. Soc. Cosmet. Chem. 11: 85–97

Horioka, M., Fukumoro, K. (1979) Manual for Newly Developed Medicine. Japanese Drug Information Service, Tokyo

Knutson, K., Potts, R. O., Guzek, D. B., Golden, G. M., Mckie, J. E., Lambert, W. J., Higuchi, W. I. (1985) Macro and molecular physical-chemical considerations in understanding drug transport in the stratum corneum. J. Contr. Rel. 2: 67–87

Leo, A., Hansch, C. (1971) Linear free-energy relationships between partitioning solvent systems. J. Org. Chem. 36: 1539–1544

Okumura, M., Sugibayashi, K., Ogawa, K., Morimoto, Y. (1989) Skin permeability of water-soluble drugs. Chem. Pharm. Bull. 37: 1404–1406

Newton, D. W., Kluza, R. B. (1978) pK_a value of medical compounds in pharmacy practice. Drug Intell. Clin. Pharm. 12: 546–554

Raykar, P. V., Fung, M., Anderson, B. D. (1988) The role of protein and lipid domains in the uptake of solutes by human stratum corneum. Pharm. Res. 5: 140–150

Rytting, J. H., Davis, S. S., Higuchi, T. (1972) Suggested thermodynamic standard state for comparing drug molecules in structure-activity studies. J. Pharm. Sci. 61: 816–818

Sato, K., Sugibayashi, K., Morimoto, Y. (1991) Species differences in percutaneous absorption of nicorandil. Ibid. 80: 104–107

Scheuplein, R. J., Blank, I. H. (1973) Mechanism of percutaneous absorption IV: penetration of nonelectrolytes (alcohols) from aqueous solutions and from pure liquids. J. Invest. Dermatol. 60: 286–296

Scott, R. C., Walker, M., Dugard, P. H. (1986) In vitro percutaneous absorption experiments: a technique for the production of intact epidermal membranes from rat skin. J. Soc. Cosmet. Chem. 37: 35–41

Skelly, J. P., Shah, V. P., Maibach, H. I., Guy, R. H., Wester, R. C., Flynn, G., Yacobi, A. (1987) FDA and AAPS report of the workshop on principles and practices of in vitro percutaneous penetration studies: relevance to bioavailability and bioequivalence. Pharm. Res. 4: 265–267

Walker, M., Dugard, P. H., Scott, R. C. (1983) In vitro percutaneous absorption studies: a comparison of human and laboratory species. Hum. Toxicol. 2: 561–562

Wertz, P. W., Downing, D. T. (1989) Stratum corneum: biological and biochemical considerations. In: Hadgraft, J., Guy, R. H. (eds) Transdermal Drug Delivery. Marcel Dekker. New York, pp 1–22

Wester, R. C., Noonan, P. K. (1980) Relevance of animal models for percutaneous absorption. Int. J. Pharm. 7: 99–110