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Prediction of skin permeability of drugs. II. Development of composite membrane as a skin alternative

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Summary

Artificial composite membranes composed of silicone and pHEMA (poly(2-hydroxyethyl methacrylate)) were developed as an alternative for skin membranes. The structure of the composite membranes was designed based on a model simulation of drug permeation properties. Composite membrane permeabilities for a wide range of drugs with diverse physicochemical properties were measured and compared with those of excised hairless rat skin. A reasonable correlation was found between the calculated and observed permeability coefficients, and between the observed values for the composite and excised skin membranes. It is suggested that human skin permeability of drugs may be predicted by using slightly modified composite membranes.

Introduction

In vitro and in vivo permeation experiments using human and animal skin have generally been employed in the screening of drug candidates for transdermal drug delivery (Bronaugh et al., 1985). However, the procedures are expensive and time-consuming, and must meet ethical standards. Because of the problems encountered with the studies, a simple method to predict accurately the skin permeability of drugs is required. Some attempts have already been made utilizing the physicochemical properties of drugs (Higuchi and Davis, 1970; Sloan et al., 1986; Rougier et al., 1987). In our previous report, we offered an equation for prediction of the skin permeability of drugs from their octanol/water partition coefficients (Hatanaka et al., 1990). There are few reports on artificial membranes as a skin alternative (Nacht and Yeung, 1985; Hadgraft and Ridout, 1987, 1988).

Generally, artificial membranes have several advantages over biological membranes: they are more reproducible, easily manufactured and their composition is readily controlled. However, no artificial membrane mimicking the characteristics of skin permeability has been yet developed. Past efforts focused primarily on homogeneous solution-diffusion membranes in spite of the heterogeneity of the skin.

In the present study, we developed a compos-

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ite membrane which mimics the barrier property of hairless rat skin. It consists of a polydimethylsiloxane (silicone) membrane, a typical solution-diffusion membrane, and a poly(2-hydroxyethyl methacrylate) (pHEMA) membrane, a typical porous membrane. The structure of the composite membrane was designed based on a model simulation of drug permeation properties, and the permeabilities of drugs with a variety of physicochemical properties were compared with hairless rat skin permeability.

Materials and Methods

Materials

Indomethacin, isosorbide dinitrate and flurbiprofen were gifts from Toko Pharmaceutical Industries Co. (Tokyo, Japan) and ibuprofen and nicorandil were from Nisshin Flour Milling Co. (Tokyo). Ketoprofen, dopamine hydrochloride and diclofenac sodium were kindly supplied by Nissan Chemical Industries Co. (Tokyo), Nikken Chemicals Co. (Tokyo) and Hamari Pharmaceutical Industries Co. (Osaka, Japan), respectively. Lidocaine, antipyrine, 5-fluorouracil and cyclobarbital were obtained from Tokyo Kasei Kogyo Co. (Tokyo) and aminopyrine and β-estradiol were from Wako Pure Chemical Industrial Co. (Osaka); dl-isoproterenol hydrochloride and l-dopa were from Sigma Chemical Co. (St. Louis, MO, U.S.A). Morphine hydrochloride was purchased from Takeda Yakuhin Industries Co. (Osaka). All these drugs were used without further purification. 2-Hydroxyethyl methacrylate and azobis(isobutyronitrile) were obtained from Tokyo Kasei Kogyo Co. Other chemicals and solvents were of reagent grade and obtained commercially.

Composite membrane preparation

Non-reinforced polydimethylsiloxane sheeting (Silastic®, Dow Corning, Midland, MI, U.S.A) was used as a silicone membrane. pHEMA membrane used in this study was prepared by a radical polymerization of 2-hydroxyethyl methacrylate with azobis(isobutyronitrile) as an initiator as follows (Hatanaka et al., 1990). The initiator (20 mg) was mixed with the monomer (20 g) and the resulting monomer solution was effervesced with N_2 for 15 min. The polymerization of the monomer solution was carried out in a silicone mold ($250 \times 250 \times 0.5$ mm) at 60 °C for 24 h to make pHEMA membranes. After complete polymerization, the pHEMA membrane was repeatedly soaked in water until use. The thickness of these membranes was measured by a dial thickness gauge (Peacook, Ozaki, Tokyo). Each membrane was punched with stainless-steel pipes having appropriate hole size (i.d. 0.50 mm, o.d. 0.95 mm for silicone membrane; i.d. 2.25 mm, o.d. 2.90 mm for pHEMA membrane). The resulting pore size was confirmed under a stereoscopic microscope (model SZ-Tr, Olympus, Tokyo) with an objective micrometer. A sheet of silicone membrane was piled on a sheet of pHEMA membrane without overlapping the pores of each membrane. The distance between both holes was about 5 mm. The adhesion of silicone and pHEMA membranes was carried out using a mixture of silicone adhesive (X7-2920 Sensitive Adhesive, Dow Corning) and silicone oil (360 n Medical Fluid 350 cs, Dow Corning) (9 : 1 ratio). Further information on the membrane is given in Results and Discussion.

Membrane permeation procedure

Membrane permeation experiments were performed according to the method of Okumura et al. (1989). The silicone side of the composite membrane was in contact with the donor compartment and the pHEMA side with the receiver compartment. The receiver compartment of each system was filled with distilled water and the donor compartment with a drug suspension in distilled water (at 2–10 times higher concentration than the solubility of each drug) to ensure constant thermodynamic activity throughout the course of the experiment. At appropriate times, a 0.1–1.0 ml sample was withdrawn from the receiver compartment and the same volume of fresh distilled water was added to keep the volume constant. Drug concentrations in the samples were analyzed by HPLC (Hatanaka et al., 1990) and the steady-state permeation rate and permeability coefficient were calculated. At the end of the

membrane permeation experiment (8 h after it began), the adhesive integrity of the composite membrane was checked by adding 25 μl of brilliant blue aqueous solution (about 1 mg/ml) to the donor compartment and monitoring its leakage into receiver compartment. No leakage was found.

Results and Discussion

Design of composite membrane

The logarithm of the octanol/water partition coefficient of model drugs (log K_{ow}), and permeability coefficient through the silicone membrane (P_S), pHEMA membrane (P_H) and excised hair-

TABLE 1

The logarithm of octanol/water partition coefficient of model drugs (log K_{ow}), and permeability coefficient through the silicone membrane (P_S), pHEMA membrane (P_H) and excised hairless rat skin (P_R)

	log K_{ow} [a]	P_S (cm/s) [b]	P_H (cm/s) [b]	P_R (cm/s) [b]
β-Estradiol	4.40	7.28×10^{-5}	4.54×10^{-5}	2.70×10^{-5}
		(−4.14)	(−4.34)	(−4.57)
Ibuprofen	3.94	3.51×10^{-3}	4.54×10^{-5}	1.02×10^{-4}
		(−2.45)	(−4.34)	(−3.99)
Flurbiprofen	3.86	7.34×10^{-4}	2.69×10^{-5}	1.14×10^{-4}
		(−3.13)	(−4.57)	(−3.94)
Indomethacin	3.19	1.75×10^{-5}	5.54×10^{-6}	4.25×10^{-5}
		(−4.76)	(−5.26)	(−4.37)
Ketoprofen	3.11	2.87×10^{-5}	1.88×10^{-5}	2.41×10^{-5}
		(−4.54)	(−4.72)	(−4.62)
Lidocaine	2.34	2.03×10^{-4}	2.98×10^{-6}	7.43×10^{-6}
		(−3.69)	(−5.53)	(−5.13)
Isosorbide dinitrate	1.34	4.76×10^{-5}	1.51×10^{-5}	4.94×10^{-6}
		(−4.32)	(−4.82)	(−5.31)
Cyclobarbital	0.873	7.48×10^{-7}	4.41×10^{-6}	1.15×10^{-6}
		(−6.13)	(−5.36)	(−5.94)
Aminopyrine	0.497	1.92×10^{-7}	1.14×10^{-5}	4.82×10^{-7}
		(−5.72)	(−4.94)	(−6.32)
5-Fluorouracil	−0.860	1.20×10^{-9}	1.33×10^{-5}	4.80×10^{-8}
		(−8.92)	(−4.88)	(−7.32)
Diclofenac sodium	−0.962	7.05×10^{-8}	5.68×10^{-5}	2.32×10^{-7}
		(−7.15)	(−4.25)	(−6.64)
Nicorandil	−1.02	2.38×10^{-7}	7.55×10^{-6}	9.80×10^{-8}
		(−6.62)	(−5.12)	(−7.01)
Antipyrine	−1.55	3.83×10^{-8}	6.11×10^{-6}	1.09×10^{-7}
		(−7.42)	(−5.21)	(−6.96)
Morphine hydrochloride	−2.53	–	3.09×10^{-6}	1.28×10^{-7}
			(−5.51)	(−6.92)
Isoproterenol hydrochloride	−2.69	–	1.14×10^{-5}	2.54×10^{-7}
			(−4.94)	(−6.59)
Dopamine hydrochloride	−3.40	5.48×10^{-12}	5.78×10^{-5}	2.57×10^{-7}
		(−11.3)	(−4.24)	(−6.59)
Levodopa	−4.70	–	4.54×10^{-5}	8.60×10^{-8}
			(−4.34)	(−7.07)

[a] Mean of three experiments.

[b] Mean of 3–6 experiments. Values in parentheses indicate logarithms.

less rat skin (P_R) reported previously are listed in Table 1 (Hatanaka et al., 1990). The permeability coefficient of drugs for the silicone membrane was dependent on their octanol/water partition coefficient and the relationship could be approximated as follows:

$$P_S = 1.82 \times 10^{-7} K_{ow}^{0.904} \text{ (cm/s)} \quad (1)$$

For the pHEMA membrane, a typical porous membrane, the permeability coefficient was almost constant independent of the partition coefficient:

$$P_H = 2.22 \times 10^{-5} \text{ (cm/s)} \quad (2)$$

On the other hand, the skin permeation properties could be classified into two types: one was the case of lipophilic drugs where the permeability coefficient correlated with the partition coefficient, similarly to silicone membranes, the other being that of hydrophilic drugs where the permeability coefficients were almost constant, similarly to pHEMA membranes. These results might be due to closely comparable diffusion coefficients of drug in these membranes, attributable to the similar molecular weight of the drugs used in this experiment (Baker and Lonsdale, 1974). Based on these data, composite membranes composed of silicone and pHEMA membranes and mimicking the permeability characteristics of hairless rat skin were designed.

Fig. 1 shows four models of the composite membranes. Models 1 and 2 represent the composite membranes where silicone and pHEMA membranes stand in parallel and series, respectively. Models 3 and 4 describe the parallel-type composite membranes with pores on only the silicone membrane and both membranes, respectively. The total permeability coefficient of drug (P) through a parallel-type composite membrane (model 1) is mathematically expressed as:

$$P = (1-f)P_S + fP_H \quad (3)$$

where P_S and P_H are the permeability coefficients of drugs for silicone and pHEMA membranes, respectively, and f denotes an area fraction of the pHEMA membrane. In the series-type composite membrane (model 2), the total permeability coefficient of a drug is generally given by:

$$1/P = 1/P_S + 1/P_H \quad (4)$$

The total permeability coefficient of drugs for Models 3 and 4 can be described using Eqns 5 and 6, respectively:

$$P = f'P_H + (1-f')P_H P_S/(P_H + P_S) \quad (5)$$

$$P = f''P_S + f'P_H - (1-f'-f'')P_H P_S / (P_H + P_S) \quad (6)$$

where f' and f'' are the area fractions of a pore on silicone and pHEMA membranes, respectively. Drug permeation properties for various composite membranes were simulated by the

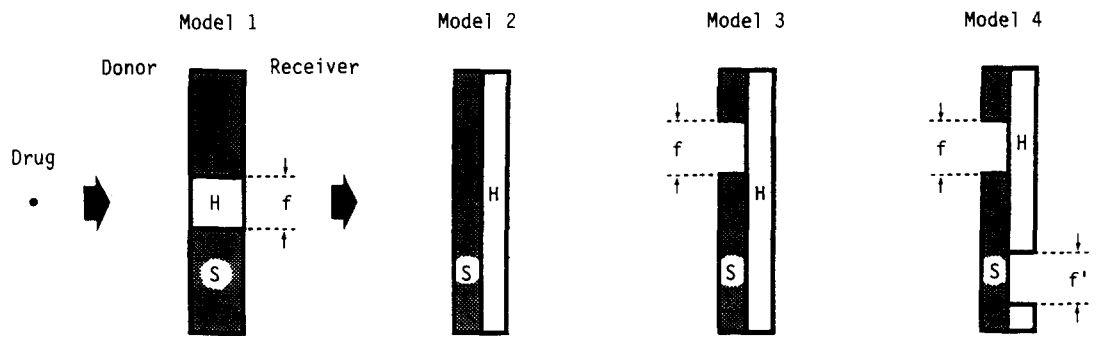

Fig. 1. Composite membrane model for simulating skin permeability of drugs. S, silicone membrane; H, pHEMA membrane; f, f', area fraction.

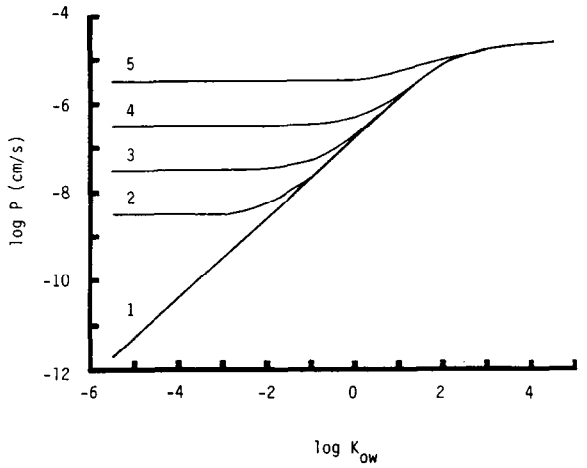

Fig. 2. Effect of pore size of silicone membrane on the permeation of a drug through the composite membrane (models 2 and 3). Thickness of silicone and pHEMA membranes is 150 and 500 μm, respectively, and area fraction of a pore on silicone membrane is 0 (1), 0.0136 (2), 0.136 (3), 1.36 (4) and 13.6% (5).

above equations (Eqns 4–6). P_S and P_H values used in the simulation were calculated based on Eqns 1 and 2, respectively, with correction for the membrane thickness (P_S and P_H values increase with decrease in membrane thickness). Because of difficulty in the preparation technique, model 1 was excluded as a candidate for composite membranes as a skin alternative.

Fig. 2 shows simulation curves for drug permeation profiles of a series-type composite membrane composed of a 150 μm silicone membrane and a 500 μm pHEMA membrane. In a composite membrane without a pore (model 2), the permeability coefficient of drugs increased with increase in their octanol/water partition coefficient, reaching a plateau for highly lipophilic drugs; the profile is different from that of skin. The silicone membrane is thought to be the major rate-limiting component to hydrophilic drugs, and the pHEMA membrane provides more significant resistance against lipophilic drug permeation through the composite membrane. To lower the diffusional resistance of a silicone membrane against only hydrophilic drugs, a composite membrane with a pore on the silicone membrane (model 3) was investigated. The pore provided a constant permeability coefficient for these drugs, and the value could be made equal to that of skin by modifying the area fraction of the pore.

The effect of silicone membrane thickness on the permeability coefficients of drugs was also evaluated (Fig. 3A). Although the permeability

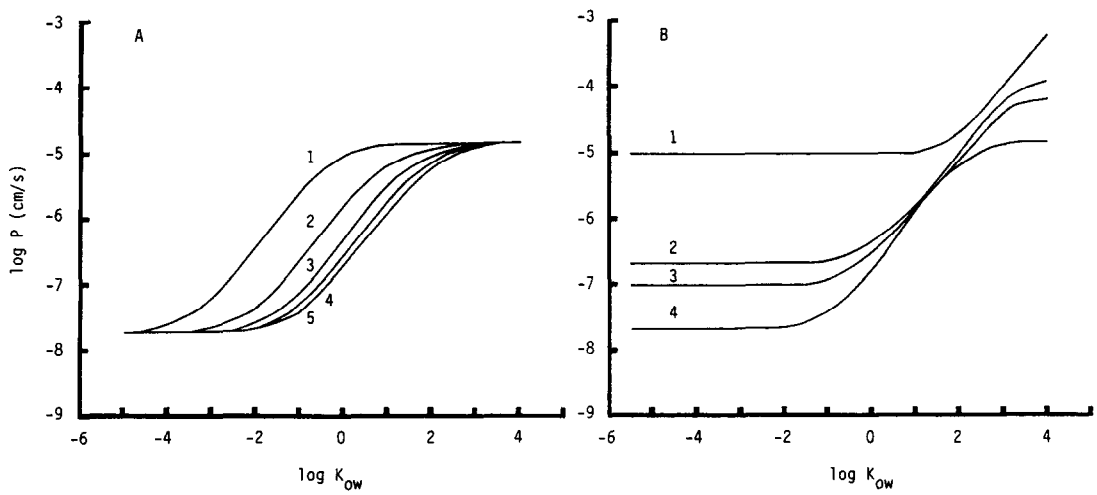

Fig. 3. Effect of thickness of silicone (A) and pHEMA (B) membrane on the permeation of drugs through the composite membrane (model 3). (A) Thickness of pHEMA membrane is 500 μm, area fraction of a pore on silicone membrane is 0.136%, and thickness of silicone membrane is 1 (1), 15 (2), 50 (3), 100 (4) and 150 μm (5). (B) Thickness of silicone membrane is 150 μm, area fraction of a pore on silicone membrane is 0.136%, and thickness of pHEMA membrane is 1 (1), 50 (2), 100 (3) and 500 μm (4).

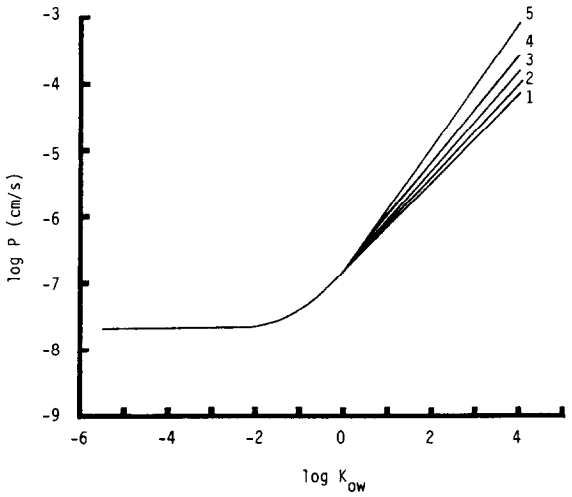

Fig. 4. Effect of pore size of pHEMA membrane on the permeation of drugs through the composite membrane (model 4). Thickness of silicone and pHEMA membranes 150 and 500 μm, respectively, area fraction of a pore on silicone membrane is 0.136%, and area fraction of a pore on pHEMA membrane is 5.22 (1), 10.4 (2), 20.9 (3), 52.2 (4) and 99.9% (5).

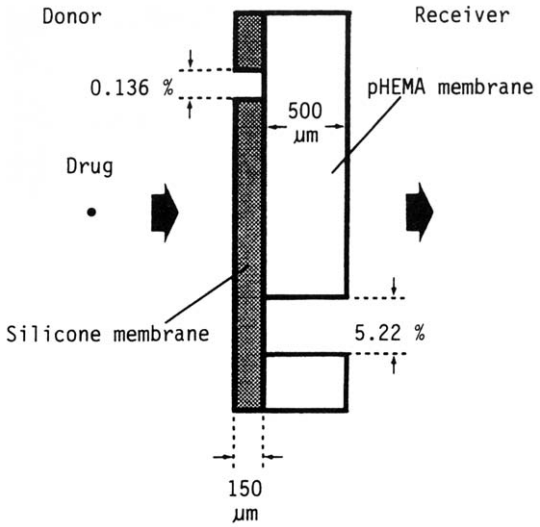

Fig. 5. Composite membrane model for simulating skin permeability of a drug.

coefficients of amphiphilic drugs increased with thinning of the silicone membrane, those of highly lipophilic and hydrophilic drugs were unchanged. This may be due to the facts that the permeability of highly lipophilic drugs through the pHEMA membrane is lower than that through the silicone membrane and that the total permeability of highly hydrophilic drugs is controlled only by the area fraction of the pore and permeability through the pHEMA membrane.

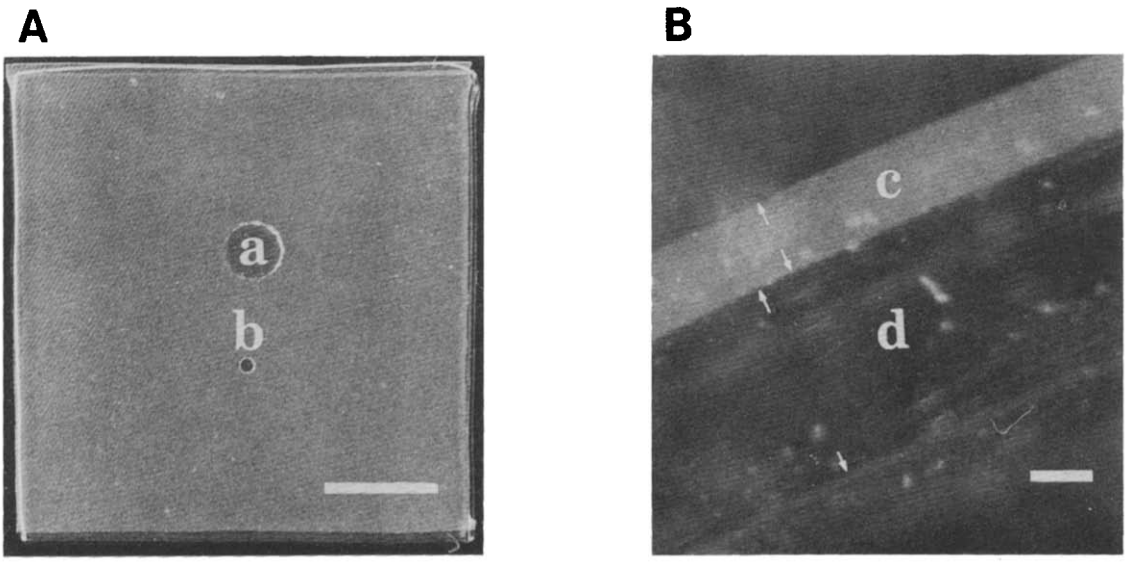

Fig. 6. Photograph of composite membrane and its cross-section for simulating skin permeability of a drug. (A) Top view (silicone membrane side up): a, hole on pHEMA membrane; b, hole on the silicone membrane; Bar = 5 mm. (B) Cross-section (micrograph): c, silicone membrane; d, pHEMA membrane; Bar = 100 μm.

Likewise, the effect of the pHEMA membrane thickness on the permeability coefficient of drugs was examined in order to lower the diffusional resistance of the composite membrane against lipophilic drugs alone (Fig. 3B). As the pHEMA membrane thickness became thinner, the permeability coefficients of both lipophilic and hydrophilic drugs increased, since the pHEMA membrane is a diffusional barrier against both types of drugs.

Subsequently, a composite membrane with pores on both silicone and pHEMA membranes was considered (model 4). Fig. 4 indicates that the pore on the pHEMA membrane provided linearity for the relationship between the permeability and partition coefficients of lipophilic drugs, similar to the case of skin, and that the slope was a function of the area fraction of the pore on the pHEMA membrane.

Preparation and evaluation of composite membrane

Fig. 5 shows the final structure of the composite membrane which mimics the drug permeation property of hairless rat skin. The composite membrane was then prepared and evaluated for drug permeability. Photographs of the composite membrane and its cross-section are shown in Fig 6A and B, respectively. The observed permeability coefficient of drugs for the composite and excised hairless rat skin membrane is shown in Fig. 7, together with its calculated simulation curve based on the model in Fig. 5. The results in Fig. 7 indicate that the final composite membrane should show a barrier property similar to the hairless rat skin barrier and that this property can be controlled by changing the structure of the membrane, namely, the thickness and/or area fraction of the pore, because of the good agreement between the observed and calculated permeability coefficients of drugs. A small discrepancy might be due to the fact that the effective area of pores on the silicone and pHEMA membranes was larger than that calculated. Therefore, a composite membrane as a human skin alternative can be prepared by a slight modification of such membrane structures. These membranes should be useful for the prediction of human skin permeability of drugs and for screening of drug candidates for transdermal drug delivery.

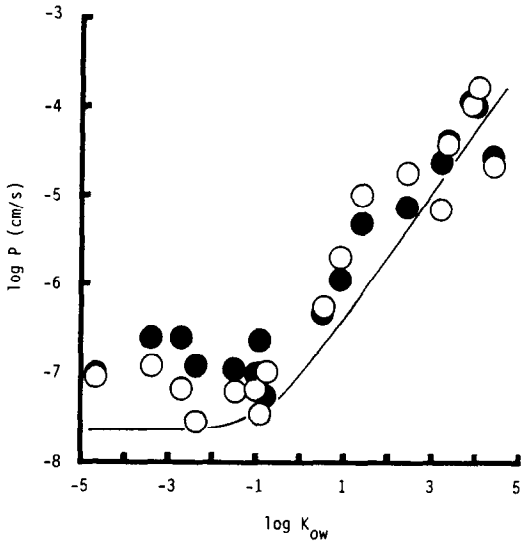

Fig. 7. The observed and calculated permeability coefficients of drugs for composite and excised hairless rat skin membrane. (○) Observed values for excised hairless rat skin; (●) observed values for composite membrane; (———) calculated values for composite membrane.

References

Baker, R.W. and Lonsdale, H.K., Controlled release: Mechanisms and rates. In Tanquary, A.C. and Lacey, R.E. (Eds) *Controlled Release of Biologically Active Agents*, Plenum, New York, 1974, pp. 15–72.

Bronaugh, R.L. and Maibach, H.I., *Percutaneous Absorption*, Dekker, New York, 1985, pp. 245–466.

Hadgraft, J. and Ridout, G., Development of model membranes for percutaneous absorption measurements. I: Isopropyl myristate. *Int. J. Pharm.*, 39 (1987) 149–156.

Hadgraft, J. and Ridout, G., Development of model membranes for percutaneous absorption measurements. II: Dipalmitoyl phosphatidylcholine, linoleic acid and tetradecane. *Int. J. Pharm.*, 42 (1988) 97–104.

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

Higuchi, T. and Davis, S.S., Thermodynamic analysis of structure activity relationships of drugs: Prediction of optimal structure. *J. Pharm. Sci.*, 59 (1970) 1376–1383.

Nacht, S. and Yeung, D., Artificial membrane and skin permeability. In Bronaugh, R.L. and Maibach, H.I. (Eds) *Percutaneous Absorption*, Dekker, New York, 1985, pp. 373–386.

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

Rougier, A., Lotte, C. and Maibach, H.I., In vivo percutaneous penetration of some organic compounds related to anatomic site in human: Prediction assessment by the stripping method. *J. Pharm. Sci.*, 76 (1987) 451–454.

Sloan, K.B., Koch, S.A.M., Siver, K.G. and Flowers, F.P., Use of solubility parameters of drug and vehicle to predict flux through skin. *J. Invest. Dermatol.*, 87 (1986) 244–252.