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Ion-pair Formation as a Strategy to Enhance Topical Delivery of Salicylic Acid

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Abstract

An in-vitro study was carried out to determine the possibility of improving the efficiency of transdermal delivery of salicylate through human epidermis by ion-pair formers (alkylamines and quaternary ammonium ions). Further, the relationship between the physicochemical properties of the counter-ions and salicylate flux was examined.

It was found that flux can be related to the conductivity associated with the penetrant solution, molecular size of the counter-ion and lipophilicity expressed as either octanol/water partition coefficient of the ion pairs or the carbon chain-length of the counter-ions.

Equations have been developed to predict salicylate flux from these physicochemical parameters.

The therapeutic efficacy of a drug following its topical application, mainly depends on its ability to penetrate the skin at an extent sufficient to elicit a pharmacological response. The permeation of ionic or hydrophilic molecules through biological or lipoidal membranes is often low. Unfavourable hydrophilic/lipophilic balance of these molecules could contribute towards their poor transepidermal bioavailability (Langguth & Mutschler 1987). Among the strategies developed to maximise the flux of drugs through the stratum corneum is the use of penetration enhancers (Yu et al 1988; Michniak et al 1995; Suh & Jun 1996; Wang et al 1997). Most of the penetration enhancers are skin irritants and allergens, thus prompting the search for safer approaches in maximising drug delivery through the skin. Although chemical modification of drug molecules (the prodrug concept) has been employed for increasing lipophilicity, the pharmacological effect and enzymatic bioconversion of these derivatives must be confirmed. Moreover, the resultant derivatives are considered as new drugs by regulatory agencies and have to be submitted to rigorous toxicological studies. Lipophilisation of drug candidates without modification of their chemical structure would thus be ideal (Sesaki et al 1992). Irwin et al (1969) were among the first to test the ion-pair hypothesis for the lipophilisation of an ionic drug (isopropamide) using trichloroacetate as the counter-ion. The results of this study indicated that the rate and efficiency of gastrointestinal absorption of isopropamide were increased by ion-pair formation. Many researchers are now exploiting the concept of ion pairs as a means of increasing the relative lipophilicity of hydrophilic ionised molecules for skin permeability (Grant & Higuchi 1990; Matschiner et al 1995; Quintanar-Guerrero et al 1997; Kadono et al 1998). These authors attributed any increase in drug permeability by ion-pair formation to an increase in the partition coefficient.

Although the relevance of ion-pair formation to the membrane uptake of ionised molecules has been discussed, very little systematic study has been attempted (Ruifrok & Meijer 1981) to examine how and why ion pairs could be employed in this role. The formation of ion pairs between ions of opposite electrical charge is both solute and environment dependent (Tomlinson et al 1982).

In this study we examined the effect of ion-pair formation between salicylic acid and alkyl amines on salicylate flux through human epidermis. An attempt was also then made to relate changes in epidermal flux to the physiochemical properties of the counter-ions.

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Materials and Methods

Materials

Salicylic acid, diethylamine, dipropylamine, triethylamine, triethanolamine, tripropylamine, tripentylamine, trihexylamine, trioctylamine, tridodecylamine, tetraethylammonium chloride, tetrabutylammonium chloride, tetrapentylammonium bromide, tetraoctylammonium bromide, propylene glycol and isopropyl myristate were purchased from Sigma Chemical Co. (Sydney, Australia). Methanol (HPLC grade) was obtained from BDH chemicals (Kilsyth, Victoria, Australia) and ^{14}C-salicylic acid was purchased from New England Nuclear, USA. All other reagents were of analytical grade and were used as received.

A liquid scintillation counter (Tri-carb 4000 series, United Technologies Packard, USA) was used to determine radioactivity in the samples. Ultraviolet spectra were obtained using a UNICON 810/820 spectrophotometer.

Partitioning experiments

Octanol was the liquid representative of skin lipids while phosphate buffer (pH 5·0) was the aqueous phase. The aqueous phase had been pre-saturated with octanol by equilibration overnight before the experiments. The samples were dissolved in 10 mL of the phase in which they were most soluble. A trace amount of ^{14}C-salicylic acid was added to the cold salicylic acid solution. Equimolar concentrations of salicylic acid and the respective amine were used in this study. The distribution coefficients were determined by equilibrating 2 mL of both phases by end-to-end mixing at room temperature ($\approx$25°C) for 16 h. The phases were then separated by centrifugation and drug concentrations in both phases determined by scintillation counting.

Conductivity measurements

The specific conductance of drug solutions (salicylic acid or equimolar concentrations of salicylic acid and the amines in ethanol–propylene glycol ((2 : 1 v/v) vehicle) used in permeation studies, was measured at room temperature (approx. 25°C) with a conductivity meter (Radiometer, Copenhagen, Model CDM80). Specific conductance, k, was measured by direct reading of the conductivity meter and given by:

$$k = (d/a)G \quad (1)$$

where d and a denote the distance between the electrodes and the area of the electrode respectively; d/a is thus the cell constant and G is conductivity in reciprocal ohms. The unit of specific conductance is siemens per cm (S cm^{-1}).

Preparation of isolated human epidermis

Human skin was obtained from the abdominal region of female patients undergoing cosmetic surgery. The subcutaneous fat was carefully trimmed off and the full-thickness skin washed with de-ionised distilled water. The epidermis was separated from the dermis by the heat method (Kligman & Christophers 1963). The full-thickness skin freed of subcutaneous fat was immersed in de-ionised distilled water at 60°C for 3 min. The epidermis was gently peeled off with the thumb. The isolated epidermis was dried between filter papers and kept frozen until required.

Permeation experiments

In-vitro permeation studies across isolated human epidermis were carried out in pyrex glass Franz-type diffusion cells. The membrane was immersed in de-ionised distilled water for 1 h before use. A thin film of lubricant was spread on the lapped glass surfaces of the half cells to ensure water-tight glass-to-membrane seals. Isolated human epidermis, supported on gauze, was mounted between the diffusion cells and the assembly held in place with a plastic clamp. The diffusion unit was then immersed in a water bath at 37°C. Penetration occurred through a cross-sectional area of 1·13 cm^2. The receptor cell had a capacity of about 3·5 mL. Phosphate buffer, pH 7·4 containing 25% ethanol was the receptor fluid. The donor phase was a solution of salicylic acid or equimolar concentrations of salicylic acid/amine counter-ion in ethanol–propylene glycol (2 : 1 v/v). Salicylic acid was spiked with trace amounts of ^{14}C-salicylic acid. After equilibration with the buffer, 1·0 mL of the donor solution was added into the donor cell while buffer was introduced into the receptor cell. The receptor compartment was continually stirred by a magnetic stirrer driven by an external magnet. At sampling times, the receptor-cell content was withdrawn and replaced with drug-free buffer kept at 37°C. The concentration of salicylate in the receptor solution was analysed by liquid scintillation counting. The permeation data represent three to six determinations.

Sample treatment and analysis

Salicylate concentrations in both the aqueous and lipid phases collected after partition experiments and in the receptor phase obtained during in-vitro epidermal permeation studies were determined by

liquid scintillation counting. A 100-μL portion of each sample was mixed with 5 mL of liquid scintillation fluid (Ultima Gold, Packard, USA), and counted for radioactivity for 2 min on the liquid scintillation counter. The partition coefficient (P) data represent an average of three determinations and are expressed as:

$$P = (\text{Counts in lipid phase})/(\text{Counts in aqueous phase}) \quad (2)$$

The amount of salicylate permeating through the epidermis during a sampling interval was calculated based on the measured receptor-phase concentration and volume. The cumulative amount of salicylate permeating per unit area vs time was plotted for each diffusion cell. The flux (J) was calculated from the slope of the linear portion of the plot. The permeability coefficient (k_p) was obtained by dividing J with the initial drug concentration in the donor phase. The enhancement ratio, described as the permeation rate from the vehicle with amine divided by the same parameter from control (vehicle without amine adjuvant), was used to evaluate the potential enhancement effect of the counter-ions. The control was assigned a value of 1·00.

Data analysis

All data are reported as mean ± s.e.m., except where indicated. Differences were tested for statistical significance using analysis of variance and paired t-tests. Significance was accepted at the 0·05 level of probability. Minitab statistical software (Minitab Inc., PA) on a Macintosh LC475 computer was used to perform multiple stepwise regressions.

Table 1. Physicochemical properties of the amine counter-ions.

Counter-ions	Chain length (N)	Molecular weight (Da)	Molal volume ($cm^3\ mol^{-1}$)[a]
Diethylamine	2	73·14	75·6
Dipropylamine	3	101·19	108·0
Triethylamine	2	101·20	108·0
Triethanolamine	2	149·19	114·9
Tripropylamine	3	143·27	156·6
Tripentylamine	5	227·44	253·8
Trihexylamine	6	269·52	302·4
Trioctylamine	8	353·68	399·6
Tridodecylamine	12	522·00	592·5
Tetraethylammonium	2	130·20	143·5
Tetrabutylammonium	4	242·40	273·1
Tetrapentylammonium	5	298·50	337·9
Tetraoctylammonium	8	466·80	532·3

[a]Estimated using the method of Yalkowsky & Zografi (1972).

Results and Discussion

The physicochemical properties of the amine counter-ions are shown in Table 1. The molecular volume (MV) of the amines was estimated from partial molal volumes of the fragments comprising the solute (Yalkowsky & Zografi 1972).

Figure 1 shows the penetration profile of salicylate in the presence of secondary amines. The effect of secondary amines on the in-vitro penetration of salicylate through human epidermis is shown in Table 2. Diethylamine had no significant effect on either the flux or the permeability coefficient of salicylic acid. There was approximately a 2-fold enhancement of these parameters by dipropylamine. Permeation enhancement of salicylate

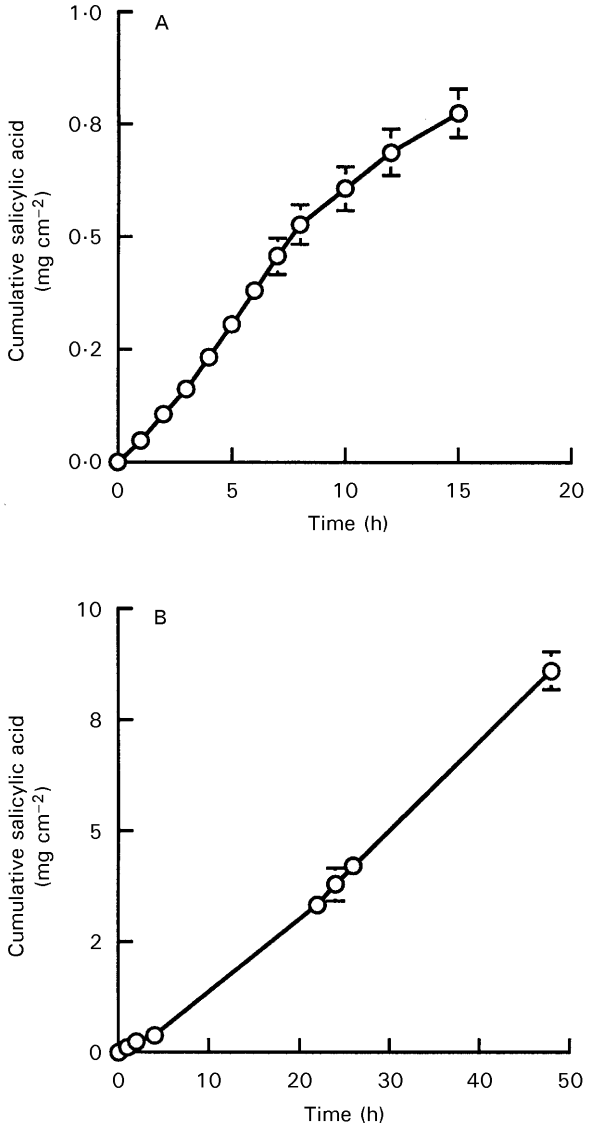

Figure 1. Penetration profile of salicylate across human epidermis in the presence of secondary amines. A: diethylamine; B: dipropylamine.

Table 2. Effect of secondary amines on the flux (J) and permeability coefficient (k_p) of salicylic acid through human epidermis.

	J[a] ($mg\,cm^{-2}\,h^{-1}$) $\times 10^{-2}$	k_p ($cm\,h^{-1}$) $\times 10^{-4}$	Enhancement ratio
Salicylic acid	$8{\cdot}9 \pm 1{\cdot}20$	$8{\cdot}9 \pm 1{\cdot}20$	1·0
Salicylic acid with:			
Diethylamine	$7{\cdot}4 \pm 0{\cdot}01$	$7{\cdot}4 \pm 0{\cdot}01$	0·83
Dipropylamine	$20{\cdot}1 \pm 0{\cdot}05$	$20{\cdot}1 \pm 0{\cdot}05$	2·26

[a]Equimolar concentrations of salicylic acid and amine in ethanol–propylene glycol (2 : 1).

observed with secondary amines could not be conclusively discussed due to the small number of this group of counter-ions used in the study.

The penetration profiles of salicylate in the absence or presence of tertiary amines are shown in Figure 2. The summary of the skin permeation data for salicylic acid and the effects of tertiary amines are presented in Table 3. The flux and permeability coefficient of salicylic acid across human epidermis were enhanced by tertiary amines. It was hypothesised that an increase in drug permeability by ion-pair formation can be generally attributed to an increase in the partition coefficient with a resultant decrease in the conductivity of the donor solution. This assumption was supported by the partition coefficient results in the presence of amines with a chain length > 3. The results from the diffusion cell experiments show that ion-pair formation with tertiary amines is significantly more effective (1·4–4·8 times) at delivering salicylate through human epidermis. There is also a good correlation between lipid solubility, expressed as the partition coefficient, and the relative ability of the members of this series of amine to deliver salicylate (flux and permeability coefficient) through the membrane. The enhancement of both the flux and permeability coefficient of salicylic acid by tertiary amines was found to be related to the alkyl chain length of the counter-ions. The longer the alkyl chain length, the greater the enhancement. This is to be expected since lipophilicity increases with increasing carbon chain-length, resulting in the formation of a more fat-soluble ion pair. Tomlinson et al (1982) have shown that in the concentration region where only ion pairs and free ions exist, an increase in the quaternary ammonium ion chain-length results in an increase in the flux of cromoglycate ions across a polyamide membrane. The relationship between the improved cromoglycate flux and quaternary ammonium chain-length was, however, dependent on the initial pairing-ion concentration.

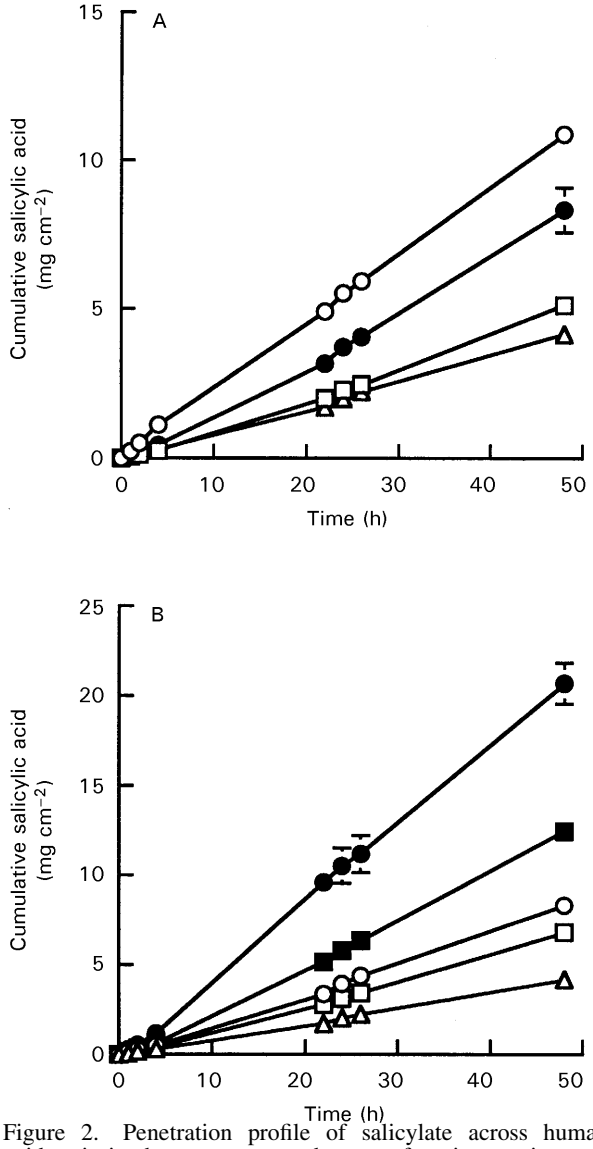

Figure 2. Penetration profile of salicylate across human epidermis in the presence or absence of tertiary amines. A. △, salicylic acid alone; ○, triethylamine; □, tripropylamine; ●, tripentylamine. B. △, salicylic acid alone; ○, triethanolamine; □, trihexylamine; ■, trioctylamine; ●, tridodecylamine.

The conductivity of a solution is proportional to the current that is transported through it. The conductivity of a solution containing ionic species is largely dependent on the population of ions present. The ion-pairing process involves a degree of charge neutralisation and its formation could thus be observed as a net reduction in the electrical conductivity of the solution in which it is occurring. The conductivity of pure media was negligibly small in all cases ($0{\cdot}23 \pm 0{\cdot}006$, $0{\cdot}33 \pm 0{\cdot}025$ and $0{\cdot}43 \pm 0{\cdot}02\,\mu S\,cm^{-1}$ for ethanol, propylene glycol and ethanol–propylene glycol (2 : 1), respectively). It is noteworthy that the highest conductivity was generally measured for salicylic acid in the vehicle. Conductivity measurements provided evidence of

Table 3. Effect of tertiary amines on partition coefficient (P), conductivity, flux (J) and permeability coefficient (k_p) of salicylic acid through human epidermis.

	P[a]	log P	J[b] ($mg\,cm^{-2}\,h^{-1}$) $\times 10^{-2}$	k_p ($cm\,h^{-1}$) $\times 10^{-3}$	Enhancement ratio	Conductivity[c] ($mS\,cm^{-1}$)
Salicylic acid	47·23 ± 0·827	1·675	8·90 ± 1·20	0·890 ± 0·120	1·00	2·03 ± 0·06
Salicylic acid with:						
Triethylamine	0·360 ± 0·003	−0·439	15·40 ± 3·85	1·540 ± 0·385	1·73	1·77 ± 0·06
Triethanolamine	0·007 ± 0·004	−1·131	11·90 ± 1·23	1·190 ± 0·123	1·34	1·53 ± 0·00
Tripropylamine	3·180 ± 0·036	0·502	18·50 ± 2·26	1·850 ± 0·226	2·08	1·35 ± 0·06
Tripentylamine	109·77 ± 11·37	2·040	19·50 ± 3·63	1·950 ± 0·363	2·19	0·90 ± 0·00
Trihexylamine	152·17 ± 26·81	2·182	22·60 ± 1·14	2·260 ± 0·114	2·54	0·80 ± 0·00
Trioctylamine	140·58 ± 16·33	2·148	27·90 ± 3·98	2·790 ± 0·398	3·13	0·60 ± 0·00
Tridodecylamine	140·66 ± 17·23	2·148	42·70 ± 2·04	4·270 ± 0·204	4·80	0·30 ± 0·00

[a]Partitioning in octanol–phosphate buffer (pH 5·0). [b]Diffusion from ethanol–propylene glycol (2 : 1) containing salicylic acid or equimolar salicylic acid and amine. [c]Measurements carried out in ethanol–propylene glycol (2 : 1) containing salicylic acid or equimolar salicylic acid and amine.

ion-pair formation in the presence of tertiary amines. Lipophilicity increased with increasing carbon chain-length resulting in the formation of more stable ion pairs. The net electrical charge of the ionic species would thus be reduced and hence the reduction in the conductivity of the medium. The results of the conductivity measurement were generally in agreement with the partition coefficient data.

Figure 3 shows the permeation profiles of salicylate in the presence of quaternary ammonium compounds. Results presented in Table 4 show that quaternary ammonium compounds did not have consistent effect on either the permeation parameters or conductivity of the model drug. The relatively lower flux, permeability coefficient and hence smaller enhancement ratios obtained with the use of quaternary ammonium compounds could possibly be due to sterically or thermodynamically induced hindrance in the formation of ion pairs. This results in larger distances and smaller attractive forces between the anion and cation. The bulky ions with four alkyl groups at the nitrogen cannot come into such close contact with the salicylate ion as do ions with fewer alkyls. Gustavii (1967) came to a similar conclusion while studying the complexes formed between amines, quaternary ammonium ions and picrate. Neubert & Dittrich (1989) studied the effect of a series of lipophilic counter-ions on the transport of ampicillin across dodecanol collodion membranes. Among the counter-ions they studied, only dodecylsulphate significantly increased the rate of transport of ampicillin. These authors attributed the observed relative lack of effect on the transport of ampicillin with the other counter-ions to steric hindrance effects. Barker & Hadgraft (1981) found that morpholines were ineffective as carriers for methyl orange across a membrane filter impregnated with isopropyl myristate. They attributed this observation to steric hindrance of the N-centre. Further, Lee & Kim (1987) found that the transport of a series of drugs through a silicon rubber membrane was ion-pair-size dependent.

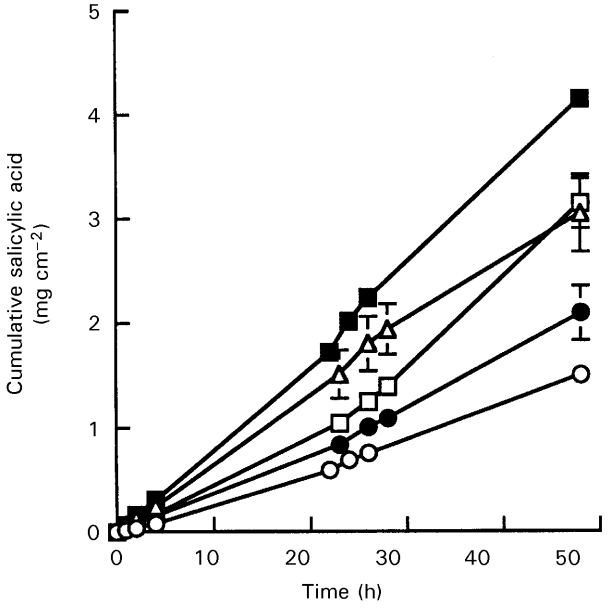

Figure 3. Penetration profile of salicylate across human epidermis in the presence or absence of quaternary ammonium cations. ■, salicylic acid alone; ○, tetraethylamine; △, tetrabutylamine; ●, tetrapentylamine; □, tetraoctylamine.

Results from our study, combined with those from a similar study using primary alkylamines as counter-ions (unpublished data), show that the amine counter-ions affect salicylate penetration through human epidermis in the following order:

Table 4. Effect of quaternary ammonium compounds on the flux (J), conductivity and permeability coefficient (k_p) of salicylic acid through human epidermis.

	J[a] ($mg\,cm^{-2}\,h^{-1}$) $\times 10^{-2}$	k_p ($cm\,h^{-1}$) $\times 10^{-3}$	Enhancement ratio	Conductvity[b] ($mS\,cm^{-1}$)
Salicylic acid	8·90 ± 1·20	0·890 ± 0·120	1·00	2·03 ± 0·06
Salicylic acid with:				
Tetraethylammonium	8·30 ± 0·28	0·830 ± 0·028	0·93	3·30 ± 0·00
Tetrabutylammonium	5·60 ± 0·99	0·560 ± 0·099	0·63	1·95 ± 10·06
Tetrapentylammonium	4·90 ± 1·00	0·490 ± 0·100	0·55	2·30 ± 0·00
Tetraoctylammonium	6·90 ± 1·36	0·690 ± 0·136	0·78	1·70 ± 0·00

[a]Diffusion from ethanol–propylene glycol (2 : 1) containing salicylic acid or equimolar salicylic acid and amine. [b]Measurements carried out in ethanol–propylene glycol (2 : 1) containing salicylic acid or equimolar salicylic acid and amine.

quaternary < primary < secondary < tertiary. The nature of the salicylate anion used in this study excludes the formation of multiple hydrogen-bonded ion pairs with either the primary or secondary amines. The carboxyl of salicylate has a low hydrogen-accepting ability due to intramolecular hydrogen bonding thus explaining the higher stability of tertiary alkylamine salicylate compared with primary or secondary. The higher the stability of the ion pair, the greater its lipophilicity and thus the better its chance of penetrating through lipid membranes. This result is consistent with the findings of Schröder-Nielsen (1974, 1976), who studied the extraction of ion pairs formed between alkylammonium cations and sulphonate and carboxylate anions, and found the following order of extraction ability of alkylammonium ions: quaternary < primary < secondary < tertiary with chloroform as organic phase. Introduction of a methylene group onto the nitrogen gave rise to a higher increase of the extraction constant than an additional methylene group in an alkyl chain. Schröder-Nielsen (1974, 1976) attributed the higher extractability of primary relative to quaternary ammonium ion pairs to a lower polarity of the former due to hydrogen bonding, thus favouring solvation in the organic phase. This is in accordance with an earlier observation for picrates using methylene chloride as organic phase (Gustavii 1967). Fung & Ow (1972) studied the functional group contribution in ion-pair extraction of tricyclic amine antidepressants. They compared the tertiary amines with their corresponding secondary amines to obtain the group contribution value of the N-substituted methyl moiety. Although they found that this value was highly anion dependent, the extraction of the tertiary amines was always more favourable than that of the secondary amines.

Patterns are beginning to emerge that allow prediction of a chemical's likely tissue penetration, based on physicochemical or merely structural data, with an increasing accumulation of percutaneous penetration data, particularly in-vitro (Roberts et al 1990, 1998; Kasting et al 1992; Yoshida & Roberts 1992; Lai & Roberts 1998a). Multiple stepwise regression was thus applied to interrelate the flux (J) of the ion pairs through human epidermis to their other physical properties and those of the counter-ions. The data generated in the previous study with primary amines and those from this study were used in the regression analysis. Combinations of normal and logarithmic forms of parameters such as partition coefficient (P) of salicylate in the presence or absence of amines, conductivity (k) of the donor solution, alkyl chain length (N), molecular weight (MW) and molecular volume (MV) of the amine counter-ions were examined to determine the best predictor of log J. The relationships between log J and conductivity, k were:

$$\log J = -0{\cdot}275 - 0{\cdot}42\,k \quad (r^2 = 0{\cdot}809, n = 23) \tag{3}$$

$$\log J = -0{\cdot}785 - 0{\cdot}987 \log k \quad (r^2 = 0{\cdot}766, n = 23) \tag{4}$$

Equations 3 and 4 show that solute conductivity is a significant determinant of salicylate flux (J). The importance of conductivity as a determinant of solute transport had previously been recognised by Gangarosa et al (1978) and Yoshida & Roberts (1994, 1995). Kamath & Gangarosa (1995) examined the relationship of various solutes and their iontophoretic transport and suggested that the transport of these solutes correlated with pK_a on the basis that pK_a is a predictor of both their conductivity and ionised state. Yoshida & Roberts (1994, 1995) developed the conductivity model, whereby ion conductivity was used as a predictor of iontophoretic flux. The conductivity model was based on the prediction of solute flux by measuring

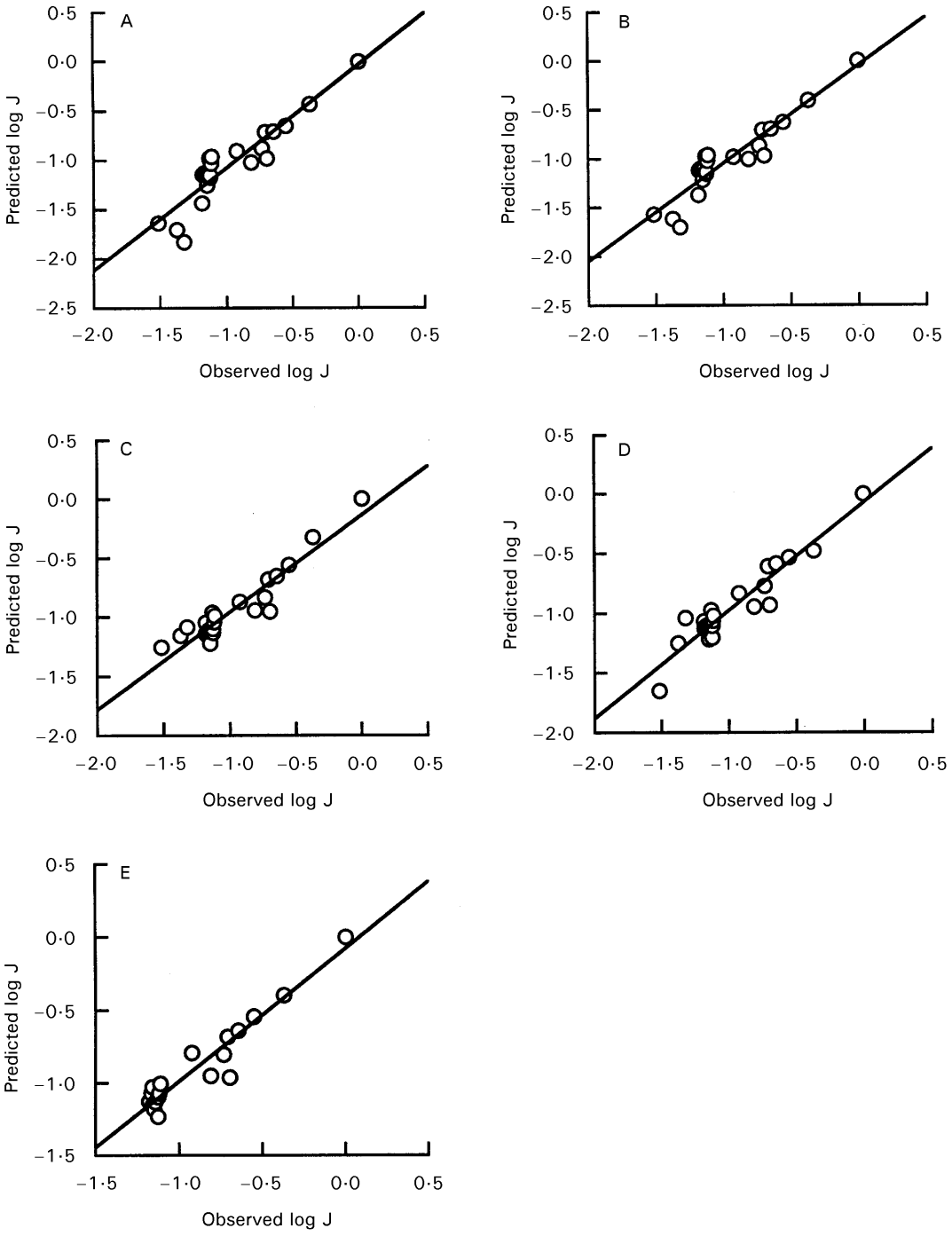

Figure 4. Predicted log J vs observed log J according to equation 5 (A), equation 6 (B), equation 8 (C), equation 9 (D) or equation 10 (E).

the specific conductance of solutes in de-ionised distilled water and in donor buffer solutions.

The effect of the inclusion of additional physical and chemical properties of the penetrating molecules on the relationship between flux and conductivity (equations 3 and 4) were also investigated. Comparison of the values of the validated coefficient of determination (r^2) serve as an indicator of such effects.

$$\log J = -0{\cdot}338 - 1{\cdot}876 \log k - 0{\cdot}002\ MW \quad (r^2 = 0{\cdot}823, n = 23) \tag{5}$$

$$\log J = -0{\cdot}302 - 2{\cdot}02 \log k - 0{\cdot}002\ MV \quad (r^2 = 0{\cdot}837, n = 23) \tag{6}$$

The relationship between flux and conductivity (equation 4) was improved by the inclusion of

either the molecular weight (equation 5) or molecular volume (equation 6) of the counter-ions. The coefficient of determination (r^2) increased from 0·766 for equation 4, to 0·823 and 0·837 for equations 5 and 6, respectively.

There was minimal covariance (< 4·0%) between log k and molecular weight or volume. The importance of the permeant molecular size to the penetration through various biological membranes has been pointed out by several researchers (Itoh et al 1990; Takahashi et al 1993). Anderson & Raykar (1989) observed a steep dependence of the diffusion constant on molecular weight of substituted *p*-cresols and hydrocortisone esters. Equations 5 and 6 are analogous to the free-volume form of the model for epidermal iontophoretic transport of solutes derived by Lai & Roberts (1998b). The free-volume model suggests that solutes move through a matrix by jumping between holes (free volumes) formed in the matrix by the movement or orientation of the molecules making up the matrix and that the progress of the solute is thus determined by its size in relation to the average free volume in the matrix (Yoshida & Roberts 1992). Lai & Roberts (1998b) reported that the logarithm of the iontophoretic permeability coefficient (log k_p) for local anaesthetics was directly related to the log ionic mobility (log μ) and molecular weight or volume. The permeability coefficient, k_p, may be thought of as normalised flux. The mobility, μ, of the diffusing species is directly proportional to conductivity, k, and is described by equation 7:

$$\mu = k/zFC \tag{7}$$

where C is the solute concentration used in the determination of conductivity, F the Faraday's constant ($9{\cdot}648 \times 10^4\,C\,mol^{-1}$) and z is the valency and equals 1 for all species. Equivalent solute concentrations were used in this study, thus making the product of z, C and F a constant. The slope of 0·002 for both molecular weight and molecular volume obtained in this study can be considered consistent with that reported by Lai & Roberts (1998b), Roberts et al (1990) and Yoshida & Roberts (1992), at similar receptor pH of 7·4 and a range of other solutes. The slopes of 1·876 and 2·02 obtained for log k in equations 5 and 6 are almost identical to those obtained by Lai & Roberts (1998b).

The relationship between flux and conductivity (equations 3 and 4) were also improved by the inclusion of N and log P as shown by equations 8–10:

$$\log J = -0{\cdot}604 - 1{\cdot}156 \log k - 0{\cdot}027\ N \quad (r^2 = 0{\cdot}876, n = 23) \tag{8}$$

$$\log J = -0{\cdot}091 - 0{\cdot}46\ k - 0{\cdot}021\ N \quad (r^2 = 0{\cdot}875, n = 23) \tag{9}$$

$$\log J = -0{\cdot}122 - 0{\cdot}485\ k - 0{\cdot}061 \log P \quad (r^2 = 0{\cdot}850, n = 19) \tag{10}$$

Both carbon chain-length (N) in equations 8 and 9 and partition coefficient (P) in equation 9 are measures of lipophilicity. The penetration of a compound through the stratum corneum is thus expected to be dependent on its lipophilicity. The relationship between skin permeability and solute lipophilicity has been reported (Scheuplein & Blank 1973; Kasting et al 1992; Potts & Guy 1992; Roberts & Walters 1998). Adjei et al (1993) stated that the formation of a peptidic ion pair results in the burying of the charges involved and the alteration of physical properties such as lipophilicity.

Figure 4 shows the relationship between predicted log J and experimentally observed log J using equations 5, 6 and 8–10. The straight lines obtained suggest perfect correlation between predicted and observed log J in each case. The plots show that 84·5, 87·1, 89·2, 89·8 and 91·3% of the data were accounted for by the variables in equations 5, 6 and 8–10, respectively. The logarithm of conductivity (log k) and size of solute as defined by molecular weight or molecular volume, conductivity in the form of k, or log k and size, as defined by carbon chain-length (N), are good predictors of log J. The highest correlation was obtained with equation 10, suggesting that the best predictors of log J are conductivity, k and the logarithm of octanol/water partition coefficient, log P.

In conclusion, this work suggests that the ability of ion-pair formation to influence the behaviour of drugs depends strongly on the physicochemical properties of both the drugs and counter-ions. The amines that imparted highly different hydrophobic properties to the parent drug upon ion-pair formation, improved the bioavailability of the hydrophilic ionisable salicylate. The results from this study indicate that it is possible to improve the efficiency of transdermal penetration of ionised salicylate through human epidermis by the selection of appropriate ion-pair formers.

Results also show that the flux of salicylate through human epidermis was controlled by the conductivity associated with the penetrant, molecular weight or molecular volume and lipophilicity

(expressed as either octanol/water partition coefficient of the ion pairs or the carbon chain-length of the amine counter-ions). Equations have been developed to predict the flux from the conductivity of the donor solution and the other physicochemical properties of the ion pairs or counter-ions. The equations relating flux to conductivity of the donor solution and molecular weight or molecular volume of the amine counter-ions are consistent with the free-volume model.

An objective assessment of the performance of amines as counter-ions in this study could have been obscured by the comparison of equal concentrations rather than equivalent thermodynamic activities of their respective formulations.

The prospect of transdermal drug delivery in biomedical applications is undoubtedly bright. Although the role of ion-pair formation in drug penetration through membranes is still largely controversial, the result of this study suggest that the successful development of the ion-pair approach capable of enhancing the skin permeability of highly ionised drugs will widen the scope of transdermal drug delivery.

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