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Prediction of Cathodal Iontophoretic Transport of Various Anions Across Excised Skin from Different Vehicles Using Conductivity Measurements

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Abstract

Solute concentration, buffer concentration, applied pH and buffer constituents affect the cathodal iontophoresis of salicylate, benzoate and butyrate across excised skin. Experiments were conducted in which the iontophoretic flux of salicylate was measured across excised human skin with variations in salicylate concentration, donor solution pH, buffer concentrations and buffer consitutents. The conductivity of these solutes and of solutions described in studies on the iontophoresis of benzoate and butyrate were then measured.

The observed variations in salicylate, benzoate and butyrate fluxes across a range of conditions were found to be related to the ratio of specific conductance of the solutes in deionized distilled water to that in the buffer solution containing the solutes.

The present results suggest that conductivity of solutes in vehicle solutions is one means of predicting the iontophoretic flux of solutes from different vehicle compositions.

Iontophoresis can be defined as the movement of charged and uncharged solutes across a tissue under an electrical potential gradient (Roberts et al 1989). Iontophoresis has been shown to be effective in the enhancement of transdermal delivery of many drugs. Although many factors influencing iontophoretic transport have been reported (Bellantone et al 1986; Burnette & Marrero 1986; Siddiqui et al 1989; Wearley et al 1989), there is an obvious lack of mechanistic information which precisely quantifies the ionic competition between the solute and other ions. It appears that iontophoretic transport of a solute shows a pronounced and complicated dependence on extraneous ions, including buffer ions, about which some factors still remain unclear. We have recently reviewed the determinants of a solute's iontophoretic transport in terms of structure-transport relationships (Yoshida & Roberts 1992). Other studies have examined the role of the type of current system (Numajiri et al 1993) and penetration enhancers (Hirvonen et al 1993) on the transdermal iontophoresis of model solutes.

Whilst a number of buffer constituents are used in iontophoretic transdermal studies, their contribution to the overall flux of a solute is poorly defined. We have previously suggested that the iontophoretic flux of a solute will be reduced when other ions are present and proposed a simple competing-ion model (Roberts et al 1989). The key components in this model were the transport number and concentration of the component ions. It was also suggested that the more mobile sodium and chloride ions could dominate ionic transport across the skin. As a consequence, the iontophoretic flux of the solute of interest could be low. Hence, the ionic composition of a selected vehicle affects the effective transdermal delivery of a given solute. We have suggested that conductivity of cations in solution may be used to predict the iontophoretic flux of cations from various vehicles during anodal iontophoretic conditions (Yoshida & Roberts 1994). Gangarosa et al (1980) have previously demonstrated that measurement of the specific conductance of several compounds could be used to assess likely candidates for iontophoresis. Kislalioglu et al (1992) have also reported that the permeability coefficient of erythromycin is linearly related to the solution conductivity.

In this study, the effect of the ionic constituents of donor solutions on the cathodal iontophoresis of salicylate across excised human skin in-vitro was quantified. The iontophoretic flux of salicylate was then related to its conductivity in various vehicles of differing buffer composition, buffer concentration and pH. The approach was validated using reported iontophoretic data for benzoate and butyrate together with measured conductivities in the vehicles used for the studies.

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Experimental Section

Materials

All solutions were made using deionized distilled water and were stored at 4°C until used. Radiolabelled salicylic acid ([^{14}C], 56 mCi mmol^{-1}) was obtained from Amersham Australia Pty Ltd, stored in ethanol, and diluted with the appropriate buffer before each experiment. Radiolabelled sodium chloride ([^{22}Na]NaCl, 0·32 μCi mg^{-1}) and water ([^{3}H]H_2O, 1 mCi g^{-1}) were also obtained from Amersham Australia Pty, Ltd. All buffer salts used were analytical grade. HEPES (*N*-2-hydroxyethylpiperazine-*N'*-2-ethanesulphonic

acid), tricine (*N*-tris (hydroxymethyl) methylglycine), Tris-HCl (triethanolamine-hydrochloride), disodium hydrogen orthophosphate and sodium dihydrogen orthophosphate were used in the preparation of buffers. BCS (biodegradable counting scintillant, NBCS.101, Amersham) was used as the scintillation fluid.

Buffer preparation

Transport studies were carried out using isotonic 20 mM HEPES buffer (osmolarity 286–300 mmol kg^{-1}, pH 7·4) in the receptor compartment, unless otherwise stated. The following solutions were used to examine the role of ionic constituents in iontophoretic transport: deionized distilled water, HEPES buffer (20, 50, 100, 250 and 500 mM), tricine buffer (20 and 50 mM), Tris-HCl (50 mM) and phosphate buffer (50 mM), the pH being adjusted with either 1 M NaOH or 1 M HCl except for phosphate buffer. All buffer solutions were degassed before use by sonicating at 40°C, to avoid any possible bubble formation on the skin at the epidermal side during iontophoresis studies.

Iontophoresis studies

Full-thickness human cadaver skin was kindly provided by the Department of Pathology, Princess Alexandra Hospital, Queensland. The skin, obtained from the abdominal region, was dermatomed to < 0·5 mm thickness using a Zimmer Electrodermatome (model 901, USA) and stored at −20°C. Before use, the skin was allowed to thaw by immersing it in deionized distilled water at room temperature for 1 h and was then equilibrated with isotonic HEPES buffer for 1 h at 37°C.

Excised human skin was then mounted between two half-diffusion cells, with the epidermis facing the donor compartment, supported by a polyethylene ring. A thin film of silicon lubricant was spread on the lapped glass surfaces of the cell to provide a water-tight seal and the cells were clamped. The surface area of epidermis exposed to the solution was 0·785 cm^2. The diffusion cells were immersed in a water bath maintained at 37 ± 0·5°C by a constant temperature bath heating system (Haaka E1). The half-cell volume was 1·0 mL and both compartments were stirred at 100 rev min^{-1} with Teflon coated fleas using external magnets. Platinum wires (99·99% purity; 2·5 cm × 1 mm) were used as electrodes and were positioned 1 cm from either side of the skin. In cathodal iontophoresis, the cathode (−) was placed on the epidermal side and the anode (+) on the dermal side, whereas in anodal iontophoresis, the electrodes were reversed. The constant current required in the iontophoretic studies was generated by a constant current source which was custom made (Department of Physical Sciences, Princess Alexandra Hospital) and showed a variability of < 0·5% over a 5-h period of continuous use.

Samples were withdrawn from the donor solution after each diffusion study. The receptor volume (1·0 mL) was removed at designated times (0, 10, 20, 30, 40, 60, 80, 100, 120 min) over the duration of the experiment and immediately replaced with fresh solution. The pH in the donor compartment was also measured after the diffusion studies. All diffusion studies were carried out at least in triplicate and results are expressed as mean ± s.d.

Factors influencing iontophoresis

In anodal and cathodal iontophoresis, radiolabelled salicylic acid was added to the donor solutions to achieve the desired specific activity. A current density of 0·38 mA cm^{-2} was applied for all experiments, unless otherwise specified. The donor solution was 50 mM HEPES buffer (pH 7·4) containing 1 mM salicylic acid, while the receptor solution was isotonic 20 mM HEPES buffer (pH 7·4).

In cathodal iontophoresis, the backward flux of sodium ions and water was also studied by adding radiotracer ([^{22}Na]NaCl and [^{3}H]H_2O) solutions to the receptor compartment (isotonic 20 mM HEPES buffer pH 7·4). The donor solution contained 1 mM unlabelled salicylic acid and 50 mM HEPES buffer (pH 7·4). At each of the designated time points, donor solution was replaced with fresh solution (50 mM HEPES + 1 mM salicylic acid).

The effect of varying salicylic acid concentration in the donor solution on its flux was investigated in cathodal iontophoresis at a current density of 0·38 mA cm^{-2}. The concentration of salicylic acid in the donor solution was varied from 1·0 to 10 mM in 50 mM HEPES buffer (pH 7·4), while the receptor solution was isotonic 20 mM HEPES buffer (pH 7.4).

The effect of the pH of the donor solution was studied at the following pH values: 6·1, 6·7, 7·4 and 8·0 (50 mM HEPES buffer). In each case, the pH was adjusted with 1 M NaOH. The donor solution contained 1 mM of salicylic acid and a current density of 0·38 mA cm^{-2} was applied for all experiments. The effect of HEPES buffer concentration (0–500 mM) in the donor solution was also examined. The concentration of salicylic acid was 1 mM and all studies were carried out with a current density of 0·38 mA cm^{-2} at pH 7·4.

The effect of donor buffer constituents for iontophoresis were examined using the following buffer constituents: 50 mM HEPES, 50 mM tricine, 50 mM Tris-HCl and 50 mM phosphate buffer. The ionic strength of buffer was 0·05 for HEPES and tricine, 0·03 for Tris-HCl and 0·13 for phosphate buffer. The concentration of salicylic acid was 1 mM and all studies were carried out with a current density of 0·38 mA cm^{-2} at pH 7·4. The receptor solution was isotonic 20 mM HEPES buffer.

The specific conductance of each solution studied was measured at 37°C before the experiment. To define the contribution of salicylate conductivity to the total conductivity of the applied solution (buffer containing salicylic acid), the specific conductance of salicylate solutions without the buffer was also measured.

Analysis

Samples (200 μL) containing [^{14}C]salicylic acid or [^{22}Na]sodium and [^{3}H]H_2O were mixed with 5·0 mL BCS and counted in a liquid scintillation counter (model MINAX. TRI-CARB 4000 series, United Technologies Packard).

The cumulative quantities of the compounds appearing in the receptor compartment (or donor compartment) were plotted against time. The steady-state flux was calculated from the slope of the linear portion of the plot. No leakage or adsorption to the glass cells of [^{14}C]salicylic acid was observed during experiments conducted for 5 h in the

absence of skin. The stability of salicylic acid was confirmed by HPLC analysis (Rumble et al 1981). More than 95% of intact salicylic acid was recovered after iontophoresis at 0·38 mA cm^{-2} for 5 h. No peaks other than salicylic acid were found in the resultant chromatograms.

Specific conductance

The specific conductance of the various buffer solutions containing salicylic acid was measured by a conductivity meter (Radiometer, Copenhagen, model CDM-8). The intrinsic conductivity of salicylic acid was measured in deionized distilled water to estimate the contribution of salicylic acid conductivity to the overall conductivity of the donor solution. Specific conductance, k, was measured by direct reading of the conductivity meter and given by:

$$k = \frac{d}{a}G \quad (1)$$

where d is the distance between the electrodes and a is the area of the electrodes, d/a corresponds to the cell constant and G is the conductivity in reciprocal ohms. The unit of specific conductance is S cm^{-1}. The specific conductance of deionized distilled water used for all buffer solutions was 0·44–0·65 μS cm^{-1}.

Statistics

The statistical significance was evaluated by the two-tailed Student's *t*-test. The 0·05 level of probability was used as the lowest level of significance.

Theoretical Section

The flux of an ion (J_i) can be defined as a function of its transport number and is proportional to the current density (Schultz 1980):

$$J_i = \frac{t_i I_T}{z_i F} \quad (2)$$

where t_i is the transport number of an ion, I_T is the total current density, z_i is the charge of ion, and F is the Faraday constant.

The transport number (t_i) of a given ion (i) is a function of its concentration (c_i) and ionic mobility in the solution (u_i) relative to the n other ions present as defined by equation 3:

$$t_i = \frac{I_i}{I} = \frac{Fc_i u_i z_i}{F\sum_{i=0}^{n} c_i u_i z_i} \quad (3)$$

The specific conductance of an ion in solution (k_i.s) is also a function of the molar concentration and ionic mobility of that ion in solution (Smedley 1980):

$$k_{i.s} = Fc_i u_i z_i \quad (4)$$

$$\frac{k_{i.s}}{k_s} = \frac{Fc_i u_i z_i}{F\sum_{i=0}^{n} c_i u_i z_i} \quad (5)$$

where k_s is the total specific conductance in a solution. The specific conductance of ions in a membrane is related to that in a solution by the volume fraction of the skin (Bruggemann 1935). Hence, both the total specific conductance of a membrane (k_m) and the specific conductance of an ion in a membrane ($k_{i.m}$) can be expressed as:

$$k_m = \phi k_s \quad (6)$$

and

$$k_{i.m} = \phi k_{i.s} \quad (7)$$

In reality, the relationship between $k_{i.m}$ and $k_{i.s}$ is also likely to be affected by the apparent zeta potential arising from fixed charged groups in a membrane pore. It is therefore assumed that a proportionality constant (A) defines the relationship between k_m, $k_{i.m}$, k_s and $k_{i.s}$:

$$\frac{k_{i.m}}{k_m} = A\frac{k_{i.s}}{k_s} \quad (8)$$

The transport number t_i in a membrane can be related to the specific conductance $k_{i.s}$ in a solution (from eqns 3, 5 and 8) by:

$$t_i = A\frac{k_{i.s}}{k_s} \quad (9)$$

Substituting equation 9 into equation 2 allows the flux of an ion to be expressed in the form:

$$J_i = A\frac{k_{i.s}}{k_s}\frac{I_t}{z_i F} \quad (10)$$

equations 9 and 10 are limited in their application in that the specific conductance of an ion ($k_{i.s}$) cannot be directly measured in the buffer solution. We have therefore assumed that the specific conductance of an ion ($k_{i.s}$) is proportional to the specific conductance of an ion in deionized distilled water (k_w). The iontophoretic flux of an ion (J_i) can then be expressed in the form:

$$J_i = B\frac{k_w}{k_s}\frac{I_t}{z_i F} \quad (11)$$

where B is the proportionality constant, A $k_{i.s}/k_w$. Thus, the flux of an ion is related directly to the specific conductance of solute in deionized-distilled water and inversely related to the specific conductance of the solution (Yoshida & Roberts 1994). In practice, the receptor solution composition remains constant and contributes approximately 60% of the total current for cathodal iontophoresis (Burnette & Ongpipattanakul 1987; Roberts et al 1989). The flux of anions from the donor solution is therefore approximately defined by the conductivity of the donor solution ($k_{s.d}$) according to equation 12:

$$J_i = 0{\cdot}4 \times B\frac{k_w}{k_{s.d.}}\frac{I_t}{z_i F} \quad (12)$$

Results and Discussion

Anodal and cathodal salicylic acid iontophoresis

Table 1 shows the steady-state flux of salicylic acid after passive, anodal and cathodal iontophoresis at 0·38 mA cm^{-2}. The flux of salicylic acid after cathodal iontophoresis was 34·6-fold that of passive transport (i.e. without iontophoresis). The anodal salicylic acid iontophoretic flux was not significantly different from the passive flux. The significant cathodal iontophoretic transport of salicylic acid is consistent with the negative charge of

Table 1. Anodal and cathodal salicylic acid iontophoresis at 0·38 mA cm^{-2} across excised human skin.

	n[a]	Flux (nmol cm^{-2} h^{-1})	Enhancement ratio[b]	pH after 2 h
Passive	3	1·96 ± 0·07	—	7·4 ± 0·1
Anodal	3	2·46 ± 0·80	1·3	5·3 ± 0·3
Cathodal	6	67·75 ± 3·18	34·6	8·3 ± 0·2

[a] The number of experiments. [b] Iontophoretic flux/passive flux.

salicylate at pH 7·4. Both anodal and cathodal iontophoresis with platinum electrodes were associated with pH changes, the pH of the donor solution in cathodal iontophoresis increasing from 7·4 to 8·3 over 2 h at 0·38 mA cm^{-2} current density whereas the pH decreased from 7·4 to 5·3 over 2 h during anodal iontophoresis. These pH changes are most likely due to electrochemical reactions associated with use of inert electrodes, whereby either oxidation (anode) or reduction (cathode) of water occurs in the donor compartment.

The contribution of water flow to the flux of salicylate was estimated from the observed ^{3}H water fluxes. At a salicylic acid concentration of 1 mM, the amount of salicylic acid carried by water in cathodal iontophoresis was calculated as 2 nmol cm^{-2} h^{-1}, or approximately 3% of the overall flux of salicylate. This is similar in magnitude to that reported in earlier studies (Yoshida & Roberts 1992). No detectable transport of Na^+ from donor to the receptor compartment was observed, whereas a steady transport number (t_{Na}) of 0·62 was reached after 40 min in transport of Na^+ from receptor to donor. This transport number for Na^+ is similar to that reported by Burnette & Ongpipattanakul (1987) and serves as the basis for the anion transport number used in equation 12.

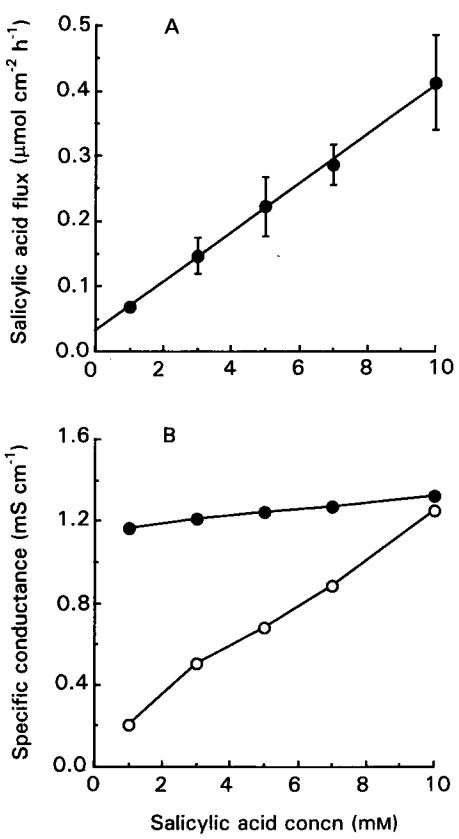

FIG. 1. A. Salicylic acid concentration in the donor compartment and cathodal iontophoresis at 0·38 mA cm^{-2}. Donor solution contained 50 mM HEPES buffer (pH 7·4) with salicylic acid varying 1–10 mM. B. Specific conductance vs salicylic acid concentration. Salicylic acid in deionized distilled water, salicylic acid in 50 mM HEPES buffer at pH 7·4.

Solute concentration

Fig. 1A shows that the flux of salicylic acid was proportional to applied salicylic acid concentration during cathodal iontophoresis ($r^2 = 0·998$) with the flux of salicylic acid increasing 6-fold for a 10-fold change in salicylic acid donor concentration. Bellantone et al (1986) also reported that benzoate flux increased 1·7-fold with a 20-fold increase in benzoate concentration during cathodal iontophoresis. The change in salicylic acid flux parallels the change in solution conductance as shown in Fig. 1B for different concentrations of salicylic acid in deionized distilled water and in buffer solutions. The specific conductance also differed approximately 6-fold in deionized distilled water between 1 and 10 mM salicylic acid. Hence, the iontophoretic flux of salicylic acid appears to be proportional to the specific conductance of salicylic acid in water. The most likely explanation for this effect is a decline in the voltage differential across the membrane due to an increase in membrane conductance/solution conductance in a constant current system. Accordingly, at higher conductances, the increase in solute flux is less than anticipated. It seems unlikely that such an effect reflects either a reduction in the number or the mobility of conducting ions at high solute concentrations as a result of increased association between ions (Kohlrausch's law (Niebergall 1980)).

pH in the donor compartment

Fig. 2A shows the fluxes of salicylic acid as a function of the pH in the donor compartment. At pH 6·0, the salicylic acid flux was approximately 9-fold that measured at pH 8·0, whereas water flux was independent of pH. The iontophoretic flux for a given solute may change with pH due to altered ionization of the solute, counterion action of groups in the membrane pathway, or resultant alterations in solute/membrane conductivity. Siddiqui et al (1985) reported that changes in the iontophoretic fluxes of lidocaine with pH appeared to be attributed to its state of ionization. The observed changes in iontophoretic flux of salicylic acid is, however, unlikely to be affected by ionization since salicylic

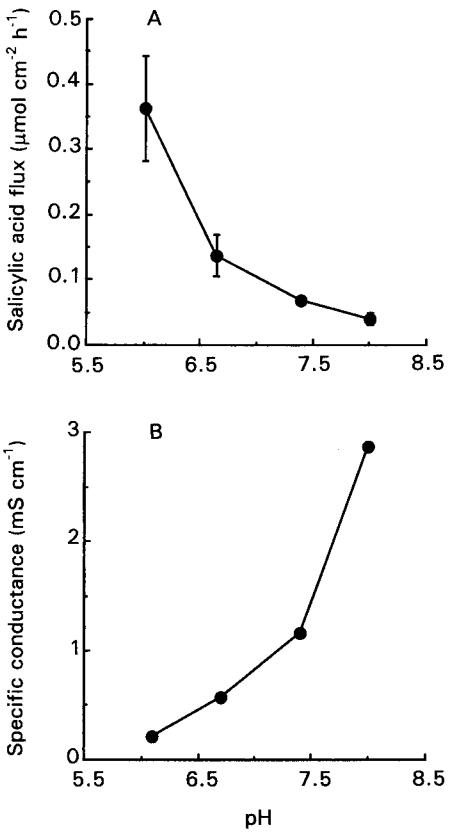

FIG. 2. A. Effect of pH in the donor compartment on salicylic acid cathodal iontophoresis at 0·38 mA cm^{-2}. Donor solution contained 1 mM salicylic acid and 50 mM HEPES buffer. B. Specific conductance vs pH solution applied.

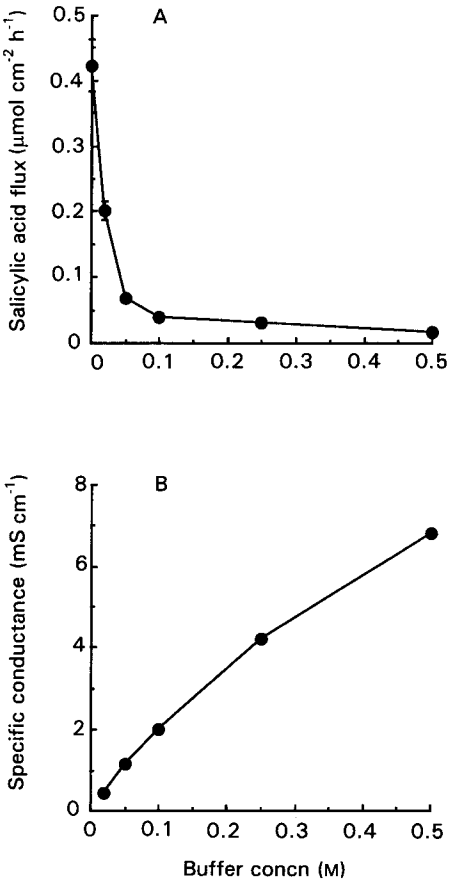

FIG. 3. A. Effect of buffer concentration in the donor compartment on salicylic acid cathodal iontophoresis at 0·38 mA cm^{-2}. Donor solution contained 1 mM salicylic acid and HEPES buffer varying 0–500 mM (pH 7·4). B. Specific conductance vs buffer concentration in the donor compartment.

acid is almost completely ionized in the pH range 6·0–8·0 (pK_a of salicylic acid is 3·0).

Decreased salicylate flux with increasing pH may be due to a change in the ionization of membrane groups with pH. The isoelectric point of the stratum corneum is at pH 3–4, therefore, the stratum corneum will become more negatively charged as the pH of the donor solution increases. Such an explanation was offered by Wearley et al (1989). At the pH range studied (pH 4–6), verapamil flux slightly increased with the increase in pH, even though the degree of ionization of verapamil is approximately constant in this pH range. Although the observed difference in salicylate flux could be due to changes in the charge on skin proteins, this dependence is unlikely.

The third mechanism accounting for the pH dependence is a change in salicylate iontophoretic flux as a consequence of an altered ionic composition of buffer constituents used to obtain a given pH. Such a change should be quantifiable by variations in conductivity with pH. Fig. 2B shows that the specific conductance of a buffer solution increases with an increase in buffer pH. The observed specific conductance difference is 13·6-fold which can be related to the amount of NaOH added to adjust the pH of the buffer. The flux of salicylate across the membrane appears to be inversely proportional to the specific conductance of the buffer in the donor compartment. The highest salicylate flux was obtained at pH 6·0, where the buffer is at its lowest

Table 2. Effect of buffer constituents on salicylic acid cathodal iontophoresis at 0·38 mA cm^{-2} across excised human skin.

Buffer	Salicylic acid flux[a] (μmol cm^{-2} h^{-1})	Specific conductance (mS cm^{-1})	Ionic strength	Buffer capacity
Tricine	0·151 ± 0·040	0·53	0·05	0·02
HEPES	0·068 ± 0·003	1·16	0·05	0·03
Tris-HCl	0·024 ± 0·001	3·52	0·02	0·01
Phosphate	0·014 ± 0·007	7·29	0·13	0·03

[a] Mean ± s.d.

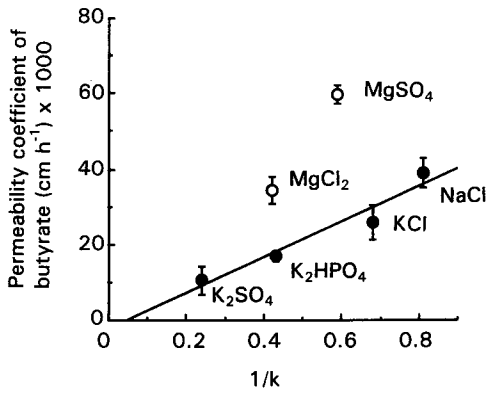

FIG. 4. Relationship between the permeability coefficient of butyrate (data from Del Terzo et al (1989)) and the specific conductance of the diffusion medium.

conductivity. The salicylate and buffer constituents compete in contributing to the total ion current. This effect can thus be minimized when the conductivity of buffer is at its lowest. The data reported by Burnette & Marrero (1986) can also be accounted for by the hypothesized relationship between the specific conductance and pH effect. The flux of thyrotropin hormone (TRH) at pH 8, where TRH is 98% unionized, was found to be 2–3-fold that at pH 4, where TRH is approximately 99% ionized (pK_a of TRH is 6.2). It is of note that two buffer solutions used in their study had different Na concentrations (pH 4; 83 mM Na_2HPO_4, 0·59 mM citric acid and 427 mM NaCl, pH 8; 185 mM Na_2HPO_4, 2·8 mM citric acid and 41 mM NaCl). Based on the data shown in Fig. 2, one could speculate that varying Na content could have affected TRH transport. The specific conductances of the buffers used by Burnette & Marrero (1986) were therefore measured and found to be 40·9 and 21·4 mS cm^{-1} at pH 4 and 8, respectively. The approximately 2-fold difference in the buffer specific conductances may account for the higher flux of TRH at pH 8.

Buffer concentration in the donor compartment

Fig. 3A shows that salicylate iontophoretic flux was reduced when the HEPES buffer concentration in the donor compartment was increased from 0 to 500 mM. The specific conductance of the solution also increased with an increase in the HEPES buffer concentration (Fig. 3B). These results suggest that the flux of salicylate is inversely proportional to the specific conductance of the buffer used. As the buffer ion concentration increases, competition amongst solute and buffer ions for iontophoretic transport becomes more pronounced and therefore a decreased solute flux is observed. A minimal buffer concentration is necessary in practice, however, in order to maintain solute ionization and to avoid significant pH shifts.

Buffer constituents

Given the dependence of the iontophoretic flux of a given solute on buffer concentration, the choice of buffer itself can also be an important determinant of solute transport. Table

Table 3. Specific conductances of applied solutions and salicylic acid fluxes during cathodal iontophoresis (current density, 0·38 mA cm^{-2}) across excised human skin.

Experimental conditions	k_w[a] (mS cm^{-2})	$k_{s.d}$[b] (mS cm^{-2})	Ratio ($k_{i.w}/k_{s.d}$)	Salicylic acid flux (μmol cm^{-2} h^{-1})
Salicylic acid[c] 1 mM	0·20	0·20	1·00	0·423 ± 0·039
Salicylic acid[c] concn (+ HEPES)				
1 mM	0·20	1·16	0·17	0·068 ± 0·003
3 mM	0·50	1·21	0·41	0·146 ± 0·027
5 mM	0·68	1·24	0·55	0·222 ± 0·047
7 mM	0·88	1·27	0·69	0·286 ± 0·030
10 mM	1·25	1·32	0·95	0·407 ± 0·073
Buffer concn (+ salicylic acid 1 mM)				
20 mM	0·20	0·44	0·45	0·201 ± 0·014
50 mM	0·20	1·16	0·17	0·068 ± 0·003
100 mM	0·20	2·01	0·10	0·038 ± 0·001
250 mM	0·20	4·20	0·05	0·030 ± 0·001
500 mM	0·20	6·81	0·03	0·016 ± 0·004
pH (+ salicylic acid 1 mM)				
8·0	0·20	2·86	0·07	0·040 ± 0·009
7·4	0·20	1·16	0·17	0·068 ± 0·003
6·7	0·20	0·56	0·36	0·136 ± 0·032
6·1	0·20	0·21	0·94	0·362 ± 0·088
50 mM Buffers (+ salicylic acid 1 mM)				
HEPES	0·20	1·16	0·17	0·068 ± 0·003
Tricine	0·20	0·53	0·38	0·151 ± 0·040
Tris-HCl	0·20	3·52	0·06	0·024 ± 0·001
Phosphate	0·20	7·29	0·03	0·014 ± 0·007
20 mM Tricine	0·20	0·25	0·80	0·321 ± 0·073

[a] k_w was measured when salicylic acid alone was dissolved in deionized distilled water. [b] $k_{s.d}$ was measured when salicylic acid was dissolved in the donor solution. [c] Maximum.

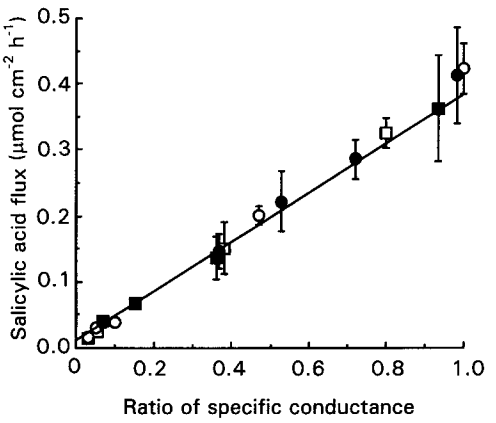

FIG. 5. Relationship between salicylic acid flux and the ratio of specific conductance in cathodal iontophoresis at 0·38 mA cm^{-2}. Salicylic acid concentration, buffer concentration, pH in the donor compartment, buffer constituents.

2 shows the iontophoretic flux of salicylic acid from 50 mM tricine, HEPES, Tris-HCl and phosphate buffers at pH 7·4. The flux of salicylic acid was highest with the tricine buffer, approximately 10-fold that with phosphate buffer. It was observed that no relationship existed between ionic strength and the iontophoretic flux of salicylic acid.

Table 2 also shows that the zwitterionic amino acid buffers, tricine and HEPES, have a relatively low specific conductance. The differing fluxes of salicylic acid from these buffers may be accounted for by the low ionic mobilities of tricine and HEPES relative to Tris-HCl and phosphate buffers. It should also be noted that the amount of salt ions with a smaller molecular size, e.g. OH^-, Cl^- in a buffer may be important in allowing effective transport of a given solute. The higher specific conductance of HEPES buffer relative to tricine buffer reflects a larger salt ion concentration, associated with the addition of NaOH to adjust the buffer pH. The specific conductance of tricine and HEPES without NaOH is only 21·4 and 24·8 μS cm^{-1}, respectively. Each of the buffer ions has an independent ionic mobility in an electric field, and hence ions compete with a given solute ion in a different magnitude in the rate of its transport.

DelTerzo et al (1989) demonstrated the effect of competitive ions in butyrate cathodal iontophoresis. The

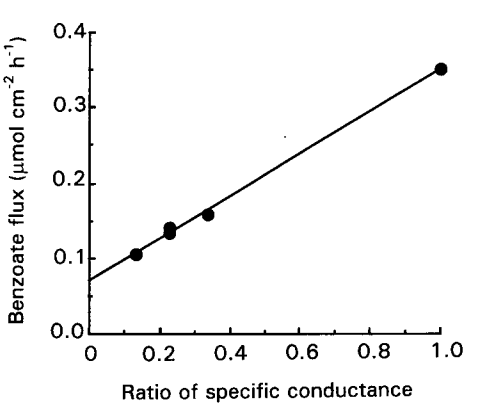

FIG. 6. Relationship between benzoate flux (data from Bellantone et al (1986)) and the ratio of specific conductance in cathodal iontophoresis at 0·1 mA.

permeability coefficient of butyrate ions was studied in the presence of various electrolyte solutions (e.g. NaCl, K_2SO_4, $MgCl_2$). Their results suggested that no relationship existed between the electrolyte type and butyrate transport. We therefore attempted to define the relationship between the permeability coefficient of butyrate and the conductivity of electrolytes, by independently measuring the specific conductance of each solution used by DelTerzo et al (1989). Fig. 4 shows that the permeability coefficient of butyrate is linearly related to the reciprocal specific conductance for the monovalent cationic solutions studied, in accordance with equation 12. The specific conductance of these solutions thus appears to provide an estimate of ionic competition to solute flux in iontophoretic transport. Although the effect of an electrolyte solution containing a divalent cation is not clear, due to there being only two data points, it is suggested that the divalent magnesium ion may have a different effect on butyrate iontophoretic transport, in that multivalent cations could bind to a fixed negative site in the skin pore (Burnette & Ongpipattanakul 1987), and therefore increase the transport of a negatively charged solute.

Relationship between salicylic acid flux and specific conductance

Table 3 summarizes the fluxes and specific conductances obtained in this study. According to equation 12, the ratio of specific conductance ($k_{i.w}/k_{s.d}$) can be used to estimate ion competition between a given solute and buffer salts in iontophoretic transport, and could then be related to the observed fluxes of salicylic acid (Fig. 5). The results for specific conductance were expressed as mean values. An excellent correlation ($r^2 = 0{\cdot}996$) was obtained for a direct relationship between salicylic acid flux and the ratio of specific conductance under the various experimental conditions studied—varying salicylic acid concentrations, buffer concentrations, pH values and buffer constituents. The strength of this regression strongly suggests that measurement of specific conductance is a useful method for estimating the complex ionic competition that occurs during the process of iontophoretic transport, and that the specific conductance is a major factor affecting iontophoretic delivery rate.

The flux-conductivity relationships reported in this study have obvious implications in terms of analysis of data resulting from iontophoretic experiments conducted under different conditions. For instance, Bellantone et al (1986) studied the effect of NaCl concentrations (0–20 mg mL^{-1}) on benzoate flux in cathodal iontophoresis at 0·1 mA, and observed that benzoate flux decreased nonlinearly with an increase of NaCl concentrations. The specific conductances of benzoate solutions employed in their study were therefore determined and plotted against the reported benzoate fluxes. The result shown in Fig. 6 shows that a linear relationship is evident between the flux of benzoate and the ratio of specific conductance. These ionic competitive data can thus be interpreted by the specific conductance theory as shown in Fig. 5.

Conclusions

The complex ionic competition in iontophoresis can be adequately defined by measuring the specific conductances of solute in deionized distilled water and the buffer solution.

An excellent relationship between the iontophoretic flux of various anions and the ratio of specific conductance ($k_w/k_{s.d}$) under a variety of conditions was observed. The present study suggests that conductivity may be an important predictor of the iontophoretic flux of a given solute under various experimental conditions.

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