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Calculation of electrophoretic mobility in ternary solvent electrolyte systems

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Abstract

Electrophoretic mobility of salmeterol and phenylpropanolamine in capillary electrophoresis has been determined using acetate buffer containing different concentrations of water, methanol and acetonitrile. Maximum electrophoretic mobilities for salmeterol and phenylpropanolamine have been observed at water–methanol–acetonitrile (5:50:45, v/v) and (3:60:37, v/v), respectively, while minimum mobilities of both compounds occurred at methanol–acetonitrile (30:70, v/v). The generated experimental data have been used to evaluate a mathematical model to compute the electrophoretic mobility of the analytes. The proposed model reproduced the mobility data with mean percentage deviations within 1–4%.

Keywords: Salmeterol; Phenylpropanolamine; Electrophoretic mobility; Mathematical model

1. Introduction

Salmeterol is a long-acting beta (β) adrenergic drug which is used to manage nocturnal asthmatic crisis. It contains a basic functional group, formulated as xinafoate salt and delivered to the lung. For assay of the drug by capillary electrophoresis (CE), because of its poor aqueous solubility [1], mixed aqueous–organic modifier buffer systems are more favourable as analytical media for CE assay. Phenylpropanolamine is an alfa (α) adrenergic drug, which is regularly used in nasal decongestant formulations. Both drugs are chosen as model drugs in this work.

Mixed aqueous–non-aqueous and non-aqueous buffer systems have been used in many validated CE methods. The main advantages of using these mixed buffer systems are to improve the selectivity, efficiency and resolution of a separation during method development [2–4]. Although binary mixed solvent buffer systems can be appropriate in many separations, they are not able to solve every problem and the analyst must add the third solvent. Ternary mixed running buffers have been

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employed in validated capillary electrophoretic methods [5,6]. These mixtures possess different characteristics in comparison with aqueous or a non-aqueous buffer. The main causes of changes to the electrophoretic behaviour of the ions in mixed solvent running buffers are: (a) different pK_a values of electrolyte and analyte, (b) different viscosities, (c) different dielectric constants, (d) different electroosmotic behaviour and (e) different conductivity. Generally, it is a combination of these effects, which is responsible for any changes in electrophoretic mobility and also separation efficiency of the system. In establishing the condition for a separation, a trial and error approach is often used by the analyst to optimise the concentrations of organic modifiers in running buffer. However, this procedure can take a long time and is therefore costly. In a recent paper [7], mathematical models have been proposed to correlate/predict the electrophoretic mobility in binary solvent electrolyte systems. Using such equations, the trial and error approach can be replaced by a more rational method development and also the generated experimental data can be evaluated to determine possible outliers when re-determination is required. An extended form of one of the models was presented here to compute the electrophoretic mobility in ternary solvent systems. The applicability of the proposed model to real data has been investigated using the generated experimental mobility data of salmeterol and phenylpropanolamine in water–methanol–acetonitrile mixtures.

2. Theoretical treatment

Fu and Lucy [8] developed nonlinear expressions for correlating/predicting the absolute mobility of monoamines. The average percentage deviation (APD) in the best-proposed model by the authors for correlating the mobility of 34 aliphatic amines is 3.7%. The authors also employed this model for predicting the mobility of 20 other monoamines, where APD was 5.1%. The most accurate model deduced from models developed by Fu and Lucy is:

$$\mu_0 = \frac{6.3 \times 10^{-3}}{W^{0.620} + 0.22H - 0.175S} \quad (1)$$

where μ_0 denotes the absolute mobility (mobility in infinite dilution), W the molecular weight of the monoamines, H the mean water of hydration calculated using McGowan's fragment addition method and S the empirical shape index which is defined as:

$$S = \frac{(N_L + 1)(B + 1)}{(N_S + 1)} \quad (2)$$

where N_L is the number of carbon atoms in the longest chain, B the number of side chains on the amine and N_S the average number of carbon atoms in the side chains.

Lucy and coworkers [9] then developed an equation based on the Max Born model for correlating the mobility of analytes with respect to molar volume (V) and acid/base dissociation constant. Max Born's model is:

$$\mu_0 = \frac{q}{f_h + f_{dl}} \quad (3)$$

where q is the charge on the analytes, f_h the hydrodynamic friction and f_{dl} the dielectric friction. The model proposed by Lucy and coworkers [9] for correlating the absolute mobility of acids or bases is formulated as:

$$\mu_0 = \frac{C_0}{V^{C_1} + C_2\, pK} \quad (4)$$

where C_0, C_1 and C_2 are the model constants and pK the dissociation constant (pK_a for acids and pK_b for bases). The calculated values of the constants for acids and bases (by using a least-squares analysis) are very close to each other. The obtained percentage error for 34 amines and 15 aliphatic acids are 4.1 and 3.7%, respectively, based on correlative equations.

Absolute mobility is also influenced by ionic strength of the background electrolyte and is represented by Pitts equation [10]:

$$\mu = \mu_0 - Aq\left(\frac{\sqrt{I}}{1 + 2.4\sqrt{I}}\right) \quad (5)$$

where A is a constant and I denotes the ionic

strength of the background electrolyte. There have also been reports of other mathematical equations to correlate electrophoretic mobility. These include Liang et al. [11] who employed topological indices for describing mobility behaviour of flavonoids.

In our earlier work [7], a solution model has been proposed to compute the electrophoretic mobility of analytes in binary solvent electrolyte systems. It showed accurate results for correlating the mobility of 57 analytes in water–organic modifier mixtures. The model is:

$$\ln \mu_m = f_1 \ln \mu_1 + f_2 \ln \mu_2 + f_1 f_2 [A_0 + A_1(f_1 - f_2)] \quad (6)$$

where μ is the electrophoretic mobility, subscripts m, 1 and 2 refer to mixed solvent, solvents 1 and 2, respectively, f the volume fraction of the solvent in the mixed solvent system and A_0, A_1 the model constants calculated by a least-squares analysis. When electrophoretic mobility in pure solvent 2 electrolyte system (μ_2) is not available or not applicable (for example, insolubility of buffering agents in pure solvent 2), a modified version of the model can be employed:

$$\ln \mu_m = f_1 \ln \mu_1 + C f_2 + f_1 f_2 [B_0 + B_1(f_1 - f_2)] \quad (7)$$

where C, B_0 and B_1 are the model constants. The theoretical value of C is equal to $\ln \mu_2$. An extended form of the model is applicable for ternary solvent mixtures:

$$\begin{aligned} \ln \mu_m = {} & f_1 \ln \mu_1 + f_2 \ln \mu_2 + K f_3 \\ & + f_1 f_2 [M_0 + M_1(f_1 - f_2)] \\ & + f_1 f_3 [M_0' + M_1'(f_1 - f_2)] \\ & + f_2 f_3 [M_0'' + M_1''(f_2 - f_3)] \end{aligned} \quad (8)$$

in which subscript 3 refers to solvent 3, and K, M_0, M_1, M_0', M_1', M_0'' and M_1'' are the model constants calculated by fitting $(\ln(\mu_m - f_1) \ln(\mu_1 - f_2) \ln \mu_2)$ against f_3, $f_1 f_2$, $f_1 f_2 (f_1 - f_2)$, $f_1 f_3$, $f_1 f_3 (f_1 - f_3)$, $f_2 f_3$ and $f_2 f_3 (f_2 - f_3)$ using a no intercept least-squares analysis.

3. Experimental

3.1. Instrumentation

All experiments were performed using a programmable CE instrument (model P/ACE 5510, Beckman Instruments, High Wycombe, UK) and a 75 μm i.d. × 37 cm length (30 cm to detector) fused silica capillary at 25 °C. Samples were injected by pressure mode for 1 s and analytes were detected by UV detection at 214 nm. The applied voltage was 24 kV. The CE instrument was interfaced with a microcomputer using Gold software (version 1.0) for data collection and analysis.

3.2. Chemicals

The analytes used were phenylpropanolamine hydrochloride and mesityl oxide purchased from Aldrich Chemical Company (Dorset, UK). Salmeterol xinafoate was a gift from Glaxo Wellcome (Ware, UK). Methanol, sodium acetate and glacial acetic acid were purchased from BDH (Poole, UK) and acetonitrile from Riedel-de-Haen (Germany). Deionised water was used in preparing the buffer and sample solutions.

3.3. Method

A stock aqueous acetate buffer was prepared by dissolving 3.28 g sodium acetate and 3.8 ml glacial acetic acid in a 100-ml volumetric flask in pure water and/or methanol. The running buffers with binary and/or ternary solvents were prepared by mixing appropriate volumes of the stock buffer, deionised water, methanol and acetonitrile. The buffers were unadjusted for pH in this work. We ensured that all our buffers contained the same volume of glacial acetic acid (0.38 ml per 100 ml) and sodium acetate (0.328 g per 100 ml). The sample solutions were prepared by dissolution in diluted running buffer solutions. Mesityl oxide was added to the sample solutions as a neutral marker.

3.4. Electrophoretic procedure

When a new capillary was used, the capillary was washed with sodium hydroxide solution (1 M,

30 min), deionised water (30 min) and running buffer (30 min). The experiments were performed after pre-washing with sodium hydroxide solution (0.1 M) for 1 min and with running buffer for 2 min. All measurements were repeated at least three times. Each sample was injected for 2 s.

3.5. Computational analysis

The electrophoretic mobility of analytes was calculated by

$$\mu = \frac{L_t L_d}{E}\left(\frac{1}{t_m} - \frac{1}{t_0}\right) \quad (9)$$

where L_t and L_d are the total capillary length and length to detector window in metres, respectively, E the applied voltage and t_m, t_0 the migration times for the analytes, and the electroosmotic flow is in seconds.

The accuracy of the calculated mobilities was then examined with respect to APDs, which were computed from the expression

$$\mathrm{APD} = \frac{100}{N}\sum_{1}^{N}\left(\frac{|\text{calculated} - \text{observed}|}{\text{observed}}\right) \quad (10)$$

where N is the number of experimental data points in each set. The mean of APD is then calculated as an overall accuracy criterion. All calculations were carried out using the statistical package for social sciences (SPSS) in a Windows environment.

4. Results and discussion

Table 1 shows the average electrophoretic mobilities and their standard deviations of salmeterol and phenylpropanolamine in different concentrations of water, methanol and acetonitrile in the solvent mixture. Minimum and maximum mobilities of salmeterol have been observed at water–methanol–acetonitrile (0:30:70, v/v) and at water–methanol–acetonitrile (5:50:45, v/v), respectively. The corresponding values for phenylpropanolamine are at water–methanol–acetonitrile (0:30:70, v/v) and at water–methanol–acetonitrile (3:60:37, v/v). Figs. 1 and 2 illustrate the mobility of the analytes studied in a three-dimensional plot. A nonlinear relationship exists between $\ln \mu_m$ and the solvent composition. The electrophoretic mobility at pure aqueous and methanolic running buffers has been reported here. The pure acetonitrile running buffer showed solubility problem with sodium acetate and has been ignored in this work.

The experimental mobility data (Table 1) for salmeterol have been fitted to Eq. (8) and the trained model obtained is:

$$\begin{aligned}\ln \mu_m = {} & 2.57f_1 + 2.66f_2 + 0.47f_3 \\ & + f_1f_2[-2.05 + 1.23(f_1 - f_2)] \\ & + f_1f_3[4.18 - 5.09(f_1 - f_2)] \\ & + f_2f_3[4.12 - 1.21(f_2 - f_3)]\end{aligned} \quad (11)$$

$R = 0.998$, $F = 821$, $N = 29$, $\mathrm{APD} = 4.17\%$ and $P < 0.0005$.

For phenylpropanolamine:

$$\begin{aligned}\ln \mu_m = {} & 3.17f_1 + 3.16f_2 + 1.33f_3 \\ & + f_1f_2[-1.77 + 0.94(f_1 - f_2)] \\ & + f_1f_3[3.74 - 4.41(f_1 - f_2)] \\ & + f_2f_3[3.41 - 0.70(f_2 - f_3)]\end{aligned} \quad (12)$$

$R = 0.999$, $F = 1815$, $N = 29$, $\mathrm{APD} = 3.46\%$ and $P < 0.0005$.

In these calculations, the robustness of the relationship between $\ln \mu_m$ and the volume fractions of the solvents is indicated by higher F-value and lower P-values. The produced APDs lie in experimental uncertainty. Therefore, by using the proposed model, it is possible to correlate the electrophoretic mobility data in ternary solvent mixtures. The mean APD for two sets studied is 3.82%, which is an acceptable error range where the relative standard deviation between repeated experiments is $\sim 4\%$. In order to provide improved accuracy, more curve-fitting parameters may be included in the model. The general form of the model is:

Table 1
The experimental electrophoretic mobility (10^{-9} m^2 V^{-1} s^{-1}) data for salmeterol and phenylpropanolamine in different solvent compositions and their standard deviations

No.	Volume fraction			Salmeterol		Phenylpropanolamine	
	Water	Methanol	Acetonitrile	Mean[a]	Standard deviation	Mean[a]	Standard deviation
1	0.00	0.30	0.70	7.43	0.18	13.12	0.08
2	0.00	0.50	0.50	13.31	0.06	22.59	0.24
3	0.00	0.70	0.30	15.26	0.24	25.74	0.22
4	0.00	0.90	0.10	15.12	0.05	25.19	0.07
5	0.00	1.00	0.00	14.23	0.21	23.66	0.34
6	0.03	0.60	0.37	15.93	0.11	26.35	0.03
7	0.05	0.50	0.45	16.17	0.07	25.92	0.25
8	0.07	0.53	0.40	14.27	0.11	24.00	0.28
9	0.10	0.45	0.45	15.02	0.00	24.75	0.00
10	0.10	0.47	0.43	14.57	0.13	24.39	0.18
11	0.10	0.60	0.30	14.19	0.06	23.82	0.11
12	0.17	0.33	0.50	14.90	0.00	24.87	0.00
13	0.20	0.40	0.40	13.28	0.06	23.14	0.04
14	0.20	0.47	0.33	12.95	0.22	22.60	0.27
15	0.23	0.20	0.57	14.48	0.00	24.82	0.00
16	0.30	0.35	0.35	12.82	0.08	22.30	0.04
17	0.30	0.70	0.00	8.58	0.09	15.82	0.09
18	0.33	0.33	0.33	12.67	0.05	22.35	0.12
19	0.47	0.20	0.33	12.61	0.12	22.82	0.27
20	0.50	0.25	0.25	11.51	0.05	20.88	0.10
21	0.50	0.33	0.17	10.06	0.04	18.50	0.07
22	0.50	0.50	0.00	8.29	0.03	15.53	0.07
23	0.70	0.15	0.15	11.25	0.06	20.62	0.06
24	0.70	0.20	0.10	10.46	0.03	19.14	0.07
25	0.70	0.30	0.00	9.52	0.10	17.56	0.13
26	0.90	0.05	0.05	12.25	0.07	22.36	0.15
27	0.90	0.07	0.03	12.00	0.13	21.87	0.18
28	0.90	0.10	0.00	11.78	0.01	21.35	0.12
29	1.00	0.00	0.00	13.04	0.02	23.81	0.20

[a] The experiments were carried out at least in triplicate with a 37 cm (30 cm effective length) × 75 μm i.d. fused silica capillary. The electrolyte was 106 mM sodium acetate buffer containing different concentrations of the organic modifiers, applied voltage: 24 kV, temperature: 25 °C and the wavelength: 214 nm.

$$\ln \mu_m = f_1 \ln \mu_1 + f_2 \ln \mu_2 + Kf_3 + f_1 f_2 \times \sum_{i=0}^{n} B_i (f_1 - f_2)^i + f_1 f_3 \sum_{i=0}^{n} B_i' (f_1 - f_3)^i + f_2 f_3 \sum_{i=0}^{n} B_i'' (f_2 - f_3)^i \quad (13)$$

where B_i, B_i' and B_i'' are the binary solvent interaction terms and the numerical value of n can be considered from 1 to 3. The resultant APD for $n = 2$, where two generated experimental data sets are used, is 2.49%. It is also possible to include ternary interaction terms and obtain:

$$\ln \mu_m = f_1 \ln \mu_1 + f_2 \ln \mu_2 + Kf_3 + f_1 f_2 \times \sum_{i=0}^{n} B_i (f_1 - f_2)^i + f_1 f_3 \sum_{i=0}^{n} B_i' (f_1 - f_3)^i + f_2 f_3 \sum_{i=0}^{n} B_i'' (f_2 - f_3)^i + f_1 f_2 f_3 \times \sum_{i=0}^{n} T_i (f_1 - f_2 - f_3)^i \quad (14)$$

where T_i is ternary solvent interaction term. When

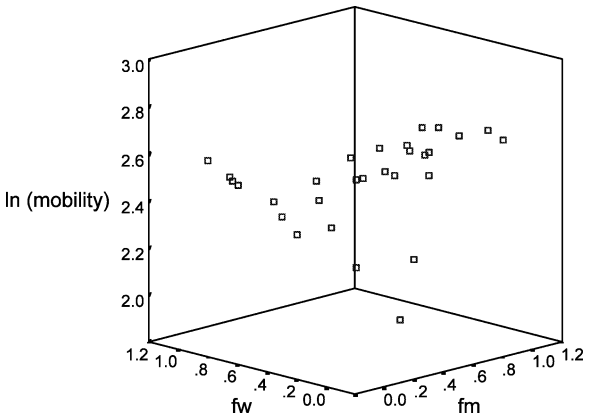

Fig. 1. Logarithm of electrophoretic mobility for the salmeterol in acetate buffer (106 mM) at different volume fractions of water (f_w) and methanol (f_m), fused silica capillary 75 μm i.d. × 37 cm (30 cm), applied voltage: 24 kV, temperature: 25 °C and wavelength: 214 nm.

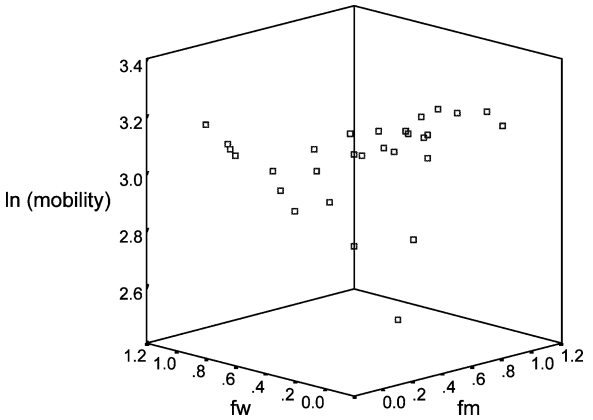

Fig. 2. Logarithm of electrophoretic mobility for phenylpropanolamine in acetate buffer (106 mM) at different volume fractions of water (f_w) and methanol (f_m), fused silica capillary 75 μm i.d. × 37 cm (30 cm), applied voltage: 24 kV, temperature: 25 °C and wavelength: 214 nm.

the electrophoretic mobility data for salmeterol and phenylpropanolamine are fitted to Eq. (14), the APD values are 1.68 and 1.14%, respectively, and the mean APD value is 1.41%.

As a general conclusion, the proposed model showed accurate results to calculate electrophoretic mobility in ternary mixed solvent running buffers. In terms of the model's general use, it is proposed that the model could be employed to optimise the solvent composition in method development in CE, where using mixed solvent systems is required to overcome the analytical problem. By using this mathematical procedure, it would be possible to replace a trial and error approach with a rational method development, thus saving time and reducing cost.

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