



ELSEVIER

Journal of Chromatography A, 829 (1998) 65–80

JOURNAL OF
CHROMATOGRAPHY A

Critical comparison of retention models for optimisation of the separation of anions in ion chromatography

I. Non-suppressed anion chromatography using phthalate eluents and three different stationary phases

John E. Madden, Paul R. Haddad*

School of Chemistry, University of Tasmania, G.P.O. Box 252-75, Hobart, Tasmania 7001, Australia

Received 7 July 1998; received in revised form 22 September 1998; accepted 25 September 1998

Abstract

A series of mathematical models describing analyte retention behaviour in non-suppressed ion chromatography of anions has been compared in order to assess their suitability for inclusion in computer optimisation software for determining the optimal eluent composition for a desired separation. The series of models comprised the linear solvent strength model (using both the dominant equilibrium approach and the competing ion effective charge approach), the dual eluent species model, the Kuwamoto model, the extended dual eluent species model and the multiple species eluent/analyte model, together with a new empirical model, the end points model. An extensive set of experimental retention data obtained for 15 anions (acetate, fluoride, iodate, bromate, chloride, nitrite, bromide, chlorate, nitrate, iodide, oxalate, sulfate, sulfite, thiosulfate and phosphate) on three columns (Waters IC Pak A, Hamilton PRP-X100 and Vydac 302 IC) using phthalate eluents of varying concentration and pH was used to evaluate the ability of each model to predict retention factors. Statistical comparison of the predicted retention factors with those obtained experimentally showed that the performance of the theoretical models improved with the complexity of the model, but none of the theoretical models could give sufficiently reliable prediction of retention factors (especially for divalent analyte ions) for the model to be used in optimisation software. However, the empirical end points model (in which a linear relationship is assumed between $\log k'$ and $\log [\text{eluent}]$, but the slope of the relationship is determined empirically) gave satisfactory performance, with correlation coefficients for all analytes of 0.9953, 0.9840 and 0.9919 for the Hamilton PRP-X100, Vydac 302 IC and Waters IC Pak A columns, respectively. © 1998 Elsevier Science B.V. All rights reserved.

Keywords: Retention models; Anions; Phthalate

1. Introduction

The development of computer-assisted optimisation routines in ion chromatography (IC) can be

achieved in a number of ways, including simplex, factorial design and interpretive methods. An interpretive optimisation method requires knowledge of the system being optimised (usually in the form of a retention model providing a mathematical relationship for calculation of the capacity factor of an analyte under differing eluent conditions) and uses

*Corresponding author. Fax: +61 3 62262858, E-mail: paul.haddad@utas.edu.au

this information to predict results. The first step in the development of a computer based interpretive optimisation method is normally the selection of a suitable retention model so that retention times can be predicted under a range of eluent conditions. Subsequent steps involve the analysis of these predicted retention times, leading ultimately to the selection of the optimal eluent composition for the desired separation.

Two types of retention model can be identified, namely theoretical and empirical. A theoretical model is derived totally from theory and invariably requires knowledge (or estimation) of a range of parameters relating to the analyte, the stationary phase, and the eluent before calculation of the capacity factor of the analyte is possible. These parameters are usually estimated by performing some preliminary experiments in which the capacity factors of desired analytes are measured under controlled conditions. Once the relevant parameters are known, a theoretical model enables, in principle, the calculation of capacity factors for all possible eluents and stationary phases. However, in practice it is more common for the relevant parameters to be determined only for a single stationary phase and a single type of eluent, and to then use the retention model to predict the effects of varying the composition (typically the concentration of competing ion and the pH) of the eluent. Empirical models concentrate on predicting the manner in which retention changes when the eluent composition is varied between two or more known values. That is, an empirical model is concerned with the observed effects of changes in eluent composition, rather than the underlying theoretical explanation for these changes. Application of an empirical model usually begins with the measurement of retention for the analytes of interest using eluents of known composition, followed by interpolation of retention behaviour at intermediate eluent compositions using the model.

A number of important factors must be considered when developing an optimisation method, including ease of use, robustness, accuracy, precision and speed. With most theoretical retention models, an improvement in the accuracy and precision is often accompanied by a decrease in the ruggedness, but empirical models tend not to suffer from this problem.

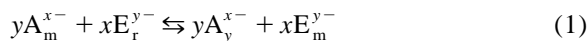
In this paper, seven theoretical IC retention models have been chosen from the literature, and one empirical model has been developed. These models have been applied to a set of previously published retention data in order to assess their suitability for inclusion in an interpretive computer-assisted optimisation method. In forthcoming papers in this series, these models will be applied to other types of IC in an attempt to cover the widest range of analyte and eluent systems possible in isocratic IC separations.

2. Theory

The seven theoretical models will be discussed in order of increasing complexity, with the empirical model being considered last. All models are expressed for use with a phthalate eluent between pH 4 and 6.

2.1. Linear solvent strength model, multiple eluent approaches

The linear solvent strength model is the simplest and earliest of the theoretical models [1–3]. It is based on the equilibrium shown in Eq. (1) which depicts the ion-exchange displacement of an eluent ion E^{y-} from the resin phase (denoted by the subscript r) of an anion-exchange material by an analyte A^{x-} initially present in the mobile phase (denoted by the subscript m). The outcome of this equilibrium is the binding of the analyte by the stationary phase.



From this equation and invoking a number of basic chromatographic principles, the following model can be derived:

$$\log k'_A = \frac{1}{y} \log (K_{A,E}) + \frac{x}{y} \log \left(\frac{Q}{y} \right) + \log \left(\frac{w}{V_m} \right) - \frac{x}{y} \log [E_m^{y-}] \quad (2)$$

where, k'_A is the capacity factor of the analyte, $K_{A,E}$ is the ion-exchange selectivity constant between the analyte and the eluent competing ion, Q is the effective ion-exchange capacity of the stationary

phase, w is the mass of the stationary phase and V_m is the volume of the eluent phase.

This equation predicts a linear relationship between the logarithm of the capacity factor and the logarithm of the eluent concentration. Furthermore, the slope of this linear relationship is equal to the negative ratio of analyte to eluent charge. In this form, the model can only be used for the prediction of retention times for an eluent which contains a single type of competing ion. However the model can be extended to predict retention times in systems using multiple eluent competing anions using two approaches, namely the dominant equilibrium approach and the competing ion effective charge approach [4].

2.1.1. Dominant equilibrium approach

The dominant equilibrium approach assumes that the most highly charged competing anion in the eluent is solely responsible for the elution of the analyte, even when relatively large amounts of less charged competing ions are present. Thus, for the case of eluents containing phthalic acid (H_2P), there are two ionised forms of the eluent which can act as competing ions (HP^- and P^{2-}), of which only P^{2-} is considered to exert an influence on the retention of analytes. Eq. (2) becomes:

$$\log k'_A = C_1 - \frac{x}{y_{\max}} \log [P^{2-}] \quad (3)$$

where C_1 is a constant which can be determined experimentally.

2.1.2. Competing ion effective charge approach

The effective charge approach assumes that all charged competing ions are responsible for the elution of the analyte, in proportion to their charges. The charge on the competing ion (y) in Eq. (2) is equated to the “effective charge” of the eluent which is the sum of the mole fractions of eluent species present multiplied by their charge. Thus for phthalate, the equation becomes:

$$\log k'_A = C_1 + C_2 \log \left(\frac{1}{y_{\text{eff}}} \right) - \frac{x}{y_{\text{eff}}} \log P_T \quad (4)$$

where C_1 and C_2 are constants that need to be determined experimentally, P_T is the total eluent concentration and the subscript eff denotes effective charge.

y_{eff} can be calculated if the acid dissociation constants for the eluent, K_{a1} and K_{a2} , are known.

$$y_{\text{eff}} = \alpha_1 + 2\alpha_2$$

$$\alpha_1 = \frac{[HP^-]}{P_T} = \frac{K_{a1}[H^+]}{[H^+]^2 + K_{a1}[H^+] + K_{a1}K_{a2}} \quad (5)$$

$$\alpha_2 = \frac{[P^{2-}]}{P_T} = \frac{K_{a1}K_{a2}}{[H^+]^2 + K_{a1}[H^+] + K_{a1}K_{a2}}$$

where, α_1 is the mole fraction of singly charged species in the eluent and α_2 is the mole fraction of doubly charged species in the eluent.

2.2. Hoover model

The Hoover model [5] was the earliest model to present a more complex treatment than the linear solvent strength model. Although this model was not used directly in this study, it has great importance as some of the following models are derivatives of the Hoover model, or draw from its concepts. The Hoover model is similar to the linear solvent strength model competing ion effective charge approach in that it takes into consideration the influence of all eluent species on analyte retention, but differs in that each eluent species can have a different, experimentally determined ion-exchange selectivity coefficient. The model gives the following two equations for the case of a phthalate eluent:

$$k'_{A^-} = \frac{w}{V_m} \frac{K_{A,HP}}{K_{P,HP}} \frac{([HP^-]_m + K_{OH,HP}[OH^-]_m)}{4[P^{2-}]_m} \left[\sqrt{1+x} - 1 \right] \quad (6)$$

$$k'_{A^{2-}} = \frac{w}{V_m} \frac{K_{A,HP}}{(K_{P,HP})^2} \frac{([HP^-]_m + K_{OH,HP}[OH^-]_m)^2}{8[P^{2-}]_m^2} \left[1 + \frac{x}{2} - \sqrt{1+x} \right] \quad (7)$$

where,

$$x = \frac{8QK_{P,HP}[P^{2-}]_m}{([HP^-]_m + K_{OH,HP}[OH^-]_m)^2}$$

and k'_{A^-} and $k'_{A^{2-}}$ are the capacity factors for mono-

and divalently charged analytes, respectively. It should be noted that the effect of hydroxide ion in the eluent is also considered.

This model contains three ion-exchange selectivity coefficients that must be derived experimentally, $K_{A,HP}$, $K_{P,HP}$ and $K_{OH,HP}$. To assist solution, the model can be linearised to determine these parameters.

2.3. Dual eluent species model

Jenke and Pagenkopf introduced this model (which is a modification of the Hoover model) in 1984 [6–8]. In this model the effect of hydroxide is assumed to be zero on the basis that hydroxide concentrations are usually very low and hydroxide is a very weak competing ion in anion-exchange systems. The model is given by the following equation:

$$k'_A = \frac{V_m K_{A,HP}}{w} \left(\frac{[H^+]}{4K_{P,HP}K_{a_2}} \right)^x \left[\left(1 + \frac{8QK_{P,HP}K_{a_2} \left(1 + \frac{K_{a_2}}{[H^+]} \right)^{1/2}}{(1 - \alpha_{H_2P})P_T[H^+]} \right)^{1/2} - 1 \right]^x \quad (8)$$

where,

$$\alpha_{H_2P} = \frac{[H^+]^2_m}{K_{a_1}K_{a_2} + K_{a_1}[H^+]_m + [H^+]^2_m}$$

This model simplifies the Hoover model by reducing the number of parameters that need to be determined experimentally from three to two, ($K_{P,HP}$ and $K_{A,HP}$). The model is linear with respect to $K_{A,HP}$, which can be solved mathematically for a given $K_{P,HP}$, but $K_{P,HP}$ must be solved through a numerical process of iterative minimisation.

2.4. Kuwamoto model

In 1989 Kuwamoto et al. [9] simplified the Hoover equation further by eliminating the ion-exchange selectivity constants and introducing elution system coefficients. Effects on analyte elution by hydroxide are disregarded. The two elution system coefficients,

C_1 and C_2 , can be determined mathematically. The model is given by the following equation:

$$\frac{1}{P_T} = C_1 \left\{ \frac{[H^+]_m}{(K_{a_2} + [H^+]_m)} \right\} (k'_A)^{1/x} + C_2 \left\{ \frac{K_{a_2}}{(K_{a_2} + [H^+]_m)} \right\} (k'_A)^{2/x} \quad (9)$$

For use as a retention model, the equation needs to be solved for k'_A , and becomes;

$$(k'_A)^{1/x} = \frac{-b^2 \pm \sqrt{b^2 + 4a \frac{1}{P_T}}}{2a} \quad (10)$$

where,

$$a = C_2 \left\{ \frac{K_{a_2}}{(K_{a_2} + [H^+]_m)} \right\} \text{ and}$$

$$b = C_1 \left\{ \frac{[H^+]_m}{(K_{a_2} + [H^+]_m)} \right\}$$

At high pH values, where elution is caused mainly by the doubly charged form of the eluent, P^{2-} , elution by the monovalent eluting species can be neglected and C_1 is assumed to be zero, allowing the value of C_2 to be determined. Once C_2 is known, solving for C_1 is straightforward. The disadvantage of this type of model is that specific experiments must be carried out to facilitate the solution of the elution coefficients, namely that an experiment must be conducted under conditions where the C_2 term can be determined.

2.5. Extended dual eluent species model

In 1994 Jenke extended his original model to allow for changes in analyte charge at different values of the pH of the eluent and an empirical correction term, $10^{[a(e)+b]}$, was also added [10]. The model is given by the following equation:

$$k' = (10^{[a(e)+b]}) \left(\frac{[H^+]}{4K_{P,HP}K_{a_2}} \right)^e \left[\left(1 + \frac{8QK_{P,HP}K_{a_2} \left(1 + \frac{K_{a_2}}{[H^+]} \right)^{1/2}}{(P_T)[H^+]} \right)^{1/2} - 1 \right]^e \quad (11)$$

where, $e = \alpha_1 + 2\alpha_2$ and

$$\alpha_1 = \frac{K_{a_1a}[\text{H}^+]}{K_{a_1a}K_{a_2a} + K_{a_1a}[\text{H}^+] + [\text{H}^+]^2}$$

$$\alpha_2 = \frac{K_{a_1a}K_{a_2a}}{K_{a_1a}K_{a_2a} + K_{a_1a}[\text{H}^+] + [\text{H}^+]^2}$$

where K_{a_1a} and K_{a_2a} are the first and second acid dissociation constants for the analyte.

This model requires an increase in the amount of knowledge of the chromatographic system compared with the original Jenke and Pagenkopf model, in that the extended model requires knowledge of the acid dissociation constants for all analytes, or alternatively knowledge that a particular analyte does not change its charge within the pH range being studied.

2.6. Multiple species eluent/analyte model

The multiple species eluent/analyte model was presented by Hajós et al. in 1995 [11] and is the most complex of the theoretical models in that it takes into consideration the interaction of all eluent species, including hydroxide, with all analyte species, as well as the effects of varying eluent pH. The model is given by the following equation for the case of a triprotic analyte, H_3A :

$$k'_{\text{A}+\text{HA}+\text{H}_2\text{A}} = \frac{w}{V_m} \left\{ K_{\text{A,HP}} \left(\frac{\sqrt{p^2 + q - p}}{4K_{\text{P,HP}}[\text{P}^{2-}]} \right)^3 \Phi_{\text{A}} \right. \\ \left. + K_{\text{HA,HP}} \left(\frac{\sqrt{p^2 + q - p}}{4K_{\text{P,HP}}[\text{P}^{2-}]} \right)^2 \Phi_{\text{HA}} \right. \\ \left. + K_{\text{H}_2\text{A,HP}} \left(\frac{\sqrt{p^2 + q - p}}{4K_{\text{P,HP}}[\text{P}^{2-}]} \right) \Phi_{\text{H}_2\text{A}} \right\} \quad (12)$$

where, $p = [\text{HP}^-] + K_{\text{OH,HP}}[\text{OH}^-]$ and $q = 8K_{\text{P,HP}}Q[\text{P}^{2-}]$ and Φ_{A} , Φ_{HA} and $\Phi_{\text{H}_2\text{A}}$ denote the partial molar fractions of deprotonated and partially protonated forms of the analyte H_3A . For the case of a phthalate eluent at acidic pH, $[\text{OH}^-]$ is so low that any contribution from hydroxide can be neglected, thus the term $K_{\text{OH,HP}}$ can be disregarded and p and q can be simplified to:

$$p = [\text{HP}^-] \quad (13)$$

$$q = 8K_{\text{P,HP}}Q[\text{P}^{2-}] \quad (14)$$

2.7. Empirical end points model

This empirical model was derived from the linear solvent strength model, but modified to allow purely empirical measurement of the slope rather than to rely on the theoretical value. The following algorithm gives the model;

$$\log k'_A = (f_1 + f_2P_T) + (f_3 + f_4P_T) \log [\text{P}^{2-}] \quad (15)$$

The four chromatographic constants, f_{1-4} can be solved using a set of simultaneous equations which employ experimental retention data for known eluent conditions.

3. Experimental

3.1. Retention data

Statistical analysis of the performance of the various retention models was carried out using tabulated retention data derived from a previous publication [12]. These data had been collected using phthalate eluents on three different columns, a Hamilton PRP-X100 polystyrene–divinylbenzene column, a Vydac 302 IC 4.6 silica column, and a Waters IC Pak A polymethacrylate column. The eluents comprised sodium phthalate solutions at concentrations of 1.0, 2.0 and 4.0 mM, and pH values of 4.0, 5.0 and 6.0.

The analyte anions studied were fluoride, chloride, bromide, iodide, chlorate, bromate, iodate, nitrite, nitrate sulfite, sulfate, thiosulfate, orthophosphate, acetate and oxalate.

All calculations were performed using Microsoft Excel 97 on a Pentium MMX 200 computer with 64 MB of SDRAM, running Windows NT workstation v4.0.

3.2. Methods for solution of models

3.2.1. Linear solvent strength model; dominant equilibrium approach

Only one experimental data point was required to

solve the parameter C_1 in this model, which could be calculated by rearranging Eq. (3). For all three columns, values for k'_A and P^{2-} were obtained from the total eluent concentration of 1 mM and pH 4.0, see Table 1.

3.2.2. Linear solvent strength model; effective charge approach

Two experimental data points were required to solve the parameters for this model. The eluents used needed to have different pH so that y_{eff} varied, but eluent concentration was kept the same for the two experiments to simplify the solution of the model parameters. The following equations were used to calculate C_1 and C_2 :

$$C_2 = \left[\log \left(\frac{k'_{A1}}{k'_{A2}} \right) + \left(\frac{x}{y_{\text{eff1}}} - \frac{x}{y_{\text{eff2}}} \right) \log (P_T) \right] / \log \left(\frac{y_{\text{eff2}}}{y_{\text{eff1}}} \right) \quad (16)$$

$$C_1 = \log k'_{A1} - C_2 \log \left(\frac{1}{y_{\text{eff1}}} \right) + \frac{x}{y_{\text{eff1}}} \log (P_T) \quad (17)$$

For all three columns, values for k'_A and P^{2-} were obtained from the experimental data with a total eluent concentration of 1.0 mM and pH values 4.0 and 6.0, see Table 1.

3.2.3. Dual eluent species model

Two experimental data points were required to determine the ion exchange selectivity constants for this model. These experiments could be any combination of total eluent concentration or pH, but it is desirable that the two data points used should have the maximum and minimum values for eluent concentration and pH. $K_{A,HP}$ was determined by rearranging Eq. (8) and solving for k'_A using an arbitrary value of $K_{P,HP}$. $K_{P,HP}$ was then solved numerically by altering its value until the value for k'_A matched the second experimentally determined value. Thus both ion-exchange selectivity constants were found.

Table 1
Experimental data points used to solve each retention model

Model	Exp. No.	[Eluent] (mM)	pH
Dominant equilibrium	1	1.0	4.0
Effective charge	1	1.0	4.0
	2	1.0	6.0
Dual eluent species	1	1.0	4.0
	2	4.0	6.0
Kuwamoto	1	1.0	4.0
	2	1.0	6.0
Extended dual eluent species	1	1.0	4.0
	2	4.0	4.0
	3	1.0	6.0
Multiple species eluent (singly or doubly charged)	1	1.0	4.0
	2	4.0	6.0
Multiple species eluent (partially doubly charged)	1	1.0	4.0
	2	1.0	6.0
	3	4.0	4.0
End points model	1	1.0	4.0
	2	1.0	6.0
	3	4.0	4.0
	4	4.0	6.0

Choice of the arbitrary value of $K_{P,HP}$ was found to be critical in order to find a solution.

For all three columns, values for k'_A and P^{2-} were obtained from the experimental data using a total eluent concentration of 1.0 mM and pH 4.0, and a total eluent concentration of 4.0 mM and pH 6.0, see Table 1. In some circumstances solutions for these values could not be found, so alternative values for the latter experiment were used.

3.2.4. Kuwamoto model

Two experimental data points were required to determine the elution system coefficients for this model. These data points needed to be chosen carefully and for the first data point, a low pH was required to ensure that elution due to the divalent eluent species could be neglected. For the second data point a higher pH was required to ensure that elution by both eluent species would occur. The elution system coefficients were calculated as follows:

$$C_2 = \frac{1}{P_T} \left\{ \frac{(K_{a_2} [H^+]_{m1})}{K_{a_2}} \right\} (k'_{A1})^{-2/x} \quad (18)$$

$$C_1 = \left(\frac{1}{P_T} - C_2 \left\{ \frac{K_{a_2}}{(K_{a_2} + [H^+]_{m2})} \right\} (k'_{A2})^{2/x} \right) \times \left\{ \frac{(K_{a_2} [H^+]_{m2})}{[H^+]_{m2}} \right\} (k'_{A2})^{-1/x} \quad (19)$$

For all three columns, C_2 was calculated using values for k'_A and P_T from the experimental data points using a total eluent concentration of 1.0 mM and pH 4.0. C_1 was calculated using experimental data points with a total eluent concentration of 1.0 mM and pH 6.0, see Table 1.

3.2.5. Extended dual eluent species model

The majority of the analytes included in the study did not alter their charge over the pH range used, so the standard dual eluent species model was used for their solution. For those analytes that did show variation of charge in the pH range 4.0–6.0, three experimental data points were required to determine the ion-exchange selectivity constant and the two empirical constants, a and b .

First, the ion-exchange selectivity constant and the empirical relationship ($10^{[a(e)+b]}$) were determined using two experiments of identical pH. This ensured that e remained constant so the empirical relationship would also remain constant. The solution was achieved in a similar way to the dual eluent species model, however the empirical relationship, ($10^{[a(e)+b]}$) was solved by rearrangement of Eq. (11), while $K_{P,HP}$ was determined using the iterative process described for the dual eluent species model.

To solve for the empirical constants, a and b , a third experimental data point was used, having an identical value of P_T to the first experiment, but a different pH. Thus a different value for ($10^{[a(e)+b]}$) was found and because e varied between the two experiments, a and b could be calculated:

$$a = \log \left[\frac{(10^{[a(e)+b]})_1}{(10^{[a(e)+b]})_3} \right] / (e_1 - e_3) \quad (20)$$

$$b = \log (10^{[a(e)+b]})_1 - a(e_1) \quad (21)$$

For all three columns, the selectivity constant and the empirical relationship were calculated using values for k'_A obtained from the experimental data points with $P_T = 1.0$ and 4.0 mM, and pH 4.0. To solve for the empirical constants a third value of k'_A was taken from the experimental data point with $P_T = 1.0$ mM and pH 6.0, see Table 1.

3.2.6. Multiple species eluent/analyte model

Solutions for this model varied in complexity depending on the charge and speciation of the analyte. The methods used for solution of this model will be broken into three parts dealing with each of the three possibilities for this model. The fourth and fifth possibilities, namely a partially triply charged anion, and a triply charged anion, will not be covered as none of the anions studied fell into these categories at the pH values studied here.

3.2.6.1. Singly or partially singly charged analyte anion

For this case the multiple species eluent/analyte model simplifies to:

$$k'_A = \frac{wK_{A,HP}}{V_m} \times \left(\frac{\sqrt{[\text{HP}^-]^2 + 8K_{P,HP}Q[\text{P}^{2-}]_m} - [\text{HP}^-]}{4K_{P,HP}[\text{P}^{2-}]_m} \right) \times \frac{[\text{A}^-]_m}{A_T} \quad (22)$$

Only two experimental data points are required to solve this model. $K_{A,HP}$ can be determined by rearranging Eq. (22), $K_{P,HP}$ can be calculated using the iterative process described for the dual eluent species model.

For all three columns the ion-exchange selectivity constants were calculated using values of k'_A , $[\text{HP}^-]$, $[\text{P}^{2-}]$ and $[\text{A}^-]/A_T$ obtained from the experimental data points with $P_T=1.0$ mM and pH 4.0, and $P_T=4.0$ and pH 6.0, see Table 1.

3.2.6.2. Doubly charged analyte anion

For this case the multiple species eluent/analyte model simplifies to:

$$k'_A = \frac{wK_{A,HP}}{V_m} \left(\frac{\sqrt{[\text{HP}^-]^2 + 8K_{P,HP}Q[\text{P}^{2-}]_m} - [\text{HP}^-]}{4K_{P,HP}[\text{P}^{2-}]_m} \right)^2 \quad (23)$$

Again, only two experiments are required to solve this model. The entire process for solution is identical to that used for a singly or partially singly charged anion, but rearranging Eq. (23) for $K_{A,HP}$.

3.2.6.3. Partially doubly charged analyte anion

For this case the multiple species eluent/analyte model simplifies to:

$$k'_A = \frac{wK_{A,HP}}{V_m} \left(\frac{\sqrt{[\text{HP}^-]_m^2 + 8K_{P,HP}Q[\text{P}^{2-}]_m} - [\text{HP}^-]_m}{4K_{P,HP}[\text{P}^{2-}]_m} \right)^2 \times \frac{[\text{A}^{2-}]_m}{A_T} + \frac{wK_{HA,HP}}{V_m} \times \left(\frac{\sqrt{[\text{HP}^-]_m^2 + 8K_{P,HP}Q[\text{P}^{2-}]_m} - [\text{HP}^-]_m}{4K_{P,HP}[\text{P}^{2-}]_m} \right)^2$$

$$\times \frac{[\text{HA}^-]_m}{A_T} \quad (24)$$

Solution of the two ion-exchange selectivity constants $K_{A,HP}$ and $K_{HA,HP}$ can be achieved using two experimental data points. The following equations are used:

$$K_{HA,HP} = \frac{V_m}{w} \times \frac{\left[\frac{k'_{A1} - k'_{A2}}{\text{factor}_1^2} \frac{[\text{A}^{2-}]_{m1}}{A_T} - \frac{A_T}{[\text{A}^{2-}]_{m2}} \right]}{\left[\frac{[\text{HA}^-]_{m1}}{\text{factor}_1 A_T} - \text{factor}_2 \frac{[\text{HA}^-]_{m2}}{A_T} - \frac{\text{factor}_1^2}{\text{factor}_2^2} \frac{[\text{A}^{2-}]_{m1}}{A_T} - \frac{A_T}{[\text{A}^{2-}]_{m2}} \right]} \quad (25)$$

$$K_{A,HP} = \frac{V_m}{w} \times \left[k'_{A1} - \frac{wK_{HA,HP}}{V_m} \text{factor}_1 \frac{[\text{HA}^-]_{m1}}{A_T} \right] \times \frac{1}{\text{factor}_1^2} \frac{A_T}{[\text{A}^{2-}]_{m1}} \quad (26)$$

where,

$$\text{factor}_n = \left(\frac{\sqrt{[\text{HP}^-]_{mn}^2 + 8K_{P,HP}Q[\text{P}^{2-}]_{mn}} - [\text{HP}^-]_{mn}}{4K_{P,HP}[\text{P}^{2-}]_{mn}} \right)$$

A third experimental data point is then used to solve for $K_{P,HP}$ using the iterative minimisation process described for the dual eluent species model.

For all three columns the ion-exchange selectivity constants, $K_{A,HP}$ and $K_{HA,HP}$, were calculated using values of k'_A , $[\text{HP}^-]$, $[\text{P}^{2-}]$, $[\text{HA}^-]/A_T$ and $[\text{A}^{2-}]/A_T$ obtained from the experimental data points with $P_T=1.0$ mM and pH=4.0 and 6.0. $K_{P,HP}$ was calculated from a third experimental data point using values of k'_A , $[\text{HP}^-]$, $[\text{P}^{2-}]$, $[\text{HA}^-]/A_T$ and $[\text{A}^{2-}]/A_T$ obtained at $P_T=4.0$ mM and pH=4.0, see Table 1.

Like the dual eluent species model, choice of the arbitrary value for $K_{P,HP}$ was found to be critical in order to find a solution to the model. For some experiments values for $K_{P,HP}$ could not be found at all and had to be estimated.

3.2.7. End points model

Four experimental data points were required to

solve this model. As the name suggests, these data points needed to lie at the four end points of the desired search area of eluent compositions. Since the experimental data available had been obtained by varying both the eluent concentration and pH, the experimental data points were broken into two sets, namely those with low eluent concentration and those with high eluent concentration. Thus the model can be split into two empirical equations;

$$\log k'_A = C_{11} + C_{21} \log [P^{2-}]_{m1} \quad (27)$$

for low eluent concentrations,

$$\log k'_A = C_{12} + C_{22} \log [P^{2-}]_{m1} \quad (28)$$

for high eluent concentrations.

These chromatographic constants can then be solved:

$$\begin{aligned} C_{21} &= \log\left(\frac{k'_{A1}}{k'_{A2}}\right) / \log\left(\frac{[P^{2-}]_{m1}}{[P^{2-}]_{m2}}\right) \\ C_{11} &= \log(k'_{A1}) - C_{21} \log [P^{2-}]_{m1} \\ C_{22} &= \log\left(\frac{k'_{A3}}{k'_{A4}}\right) / \log\left(\frac{[P^{2-}]_{m3}}{[P^{2-}]_{m4}}\right) \\ C_{12} &= \log(k'_{A3}) - C_{22} \log [P^{2-}]_{m3} \end{aligned} \quad (29)$$

These can be related back to the original end points model;

$$C_1 = f_1 + f_2 P_T \quad (30)$$

$$C_2 = f_3 + f_4 P_T \quad (31)$$

Thus the values for four chromatographic constants f_{1-4} can be solved;

$$\begin{aligned} f_2 &= \frac{(C_{11} - C_{12})}{(P_{T1} - P_{T2})} \\ f_1 &= C_{11} - f_2 P_{T1} \\ f_4 &= \frac{(C_{21} - C_{22})}{(P_{T1} - P_{T2})} \\ f_3 &= C_{21} - f_4 P_{T2} \end{aligned} \quad (32)$$

For all three columns, values for k'_A and other chromatographic conditions were obtained from the experimental data points with total eluent concentration of 1.0 and 4.0 mM and pH values 4.0 and 6.0, see Table 1.

4. Results and discussion

4.1. Prediction of retention times using the models

Once the various parameters for each model had been found, the models were used to predict retention data for all 15 analytes on each of the three columns using each of the eluent compositions in the data set. This gave a total of 45 predicted retention times for each model. Table 2 shows a typical set of results for sulfate on the Hamilton PRP-X100 col-

Table 2

Experimentally measured retention times (E) and predicted retention times from the seven retention models: dominant equilibrium approach (DE), effective charge approach (EC), dual eluent species model (DES), Kuwamoto model (K), extended dual eluent species model (EDS), multiple species eluent/analyte model (MSE) and end points model (End) for sulfate on the Hamilton PRP-X100 column

Eluent composition		Retention time (min)							
pH	P_T	E	DE	EC	DES	K	EDS	MSE	End
4.00	1.00	39.37	39.37	39.37	39.37	39.37	39.37	39.37	39.37
	2.00	14.07	21.17	10.36	13.21	13.29	15.12	13.21	17.57
	4.00	5.58	11.40	3.130	4.448	4.494	5.580	4.448	5.580
5.00	1.00	14.15	5.526	13.01	14.57	15.41	15.85	14.57	14.09
	2.00	6.29	3.303	4.966	6.764	7.196	7.881	6.764	7.348
	4.00	3.04	2.172	2.272	3.265	3.477	4.049	3.265	3.292
6.00	1.00	8.35	2.317	8.350	7.462	7.742	7.208	7.462	8.350
	2.00	4.30	1.584	4.320	4.099	4.245	4.025	4.099	4.676
	4.00	2.42	1.217	2.453	2.420	2.497	2.420	2.420	2.420

Phthalate eluents with a total concentration of P_T mM were used.

umn. The measured retention times are shown for each eluent composition, as well as those predicted by each of the seven models.

To identify the “best” model the residuals, that is the differences, d_i , between the experimental and calculated values of the retention times [13], must be analysed statistically. Because retention times for each experiment vary by such a large amount, it is better to calculate the percentage normalised difference ($\%d_i$) between the measured and predicted retention times for a particular ion using the following equation:

$$\% d_i = \frac{t_{R \text{ act}} - t_{R \text{ pred}}}{t_{R \text{ act}} + t_{R \text{ pred}}} \cdot 2 \cdot \frac{100}{1} \quad (33)$$

where, $t_{R \text{ act}}$ is the experimentally determined retention time and $t_{R \text{ pred}}$ is the retention time predicted by the retention model.

Table 3 shows the values of $\%d_i$ for the data given in Table 2, i.e., for the retention of sulfate on the Hamilton PRP-X100 column.

If a model represents the data adequately, the normalised percentage differences should possess characteristics that agree with, or at least do not refute, the basic assumption that the differences are

randomly distributed about the $t_{R \text{ pred}}$ values. Systematic departures from randomness indicate that the model is not satisfactory. A study of the signs and magnitude of the normalised percentage differences can aid further in the analysis of the data.

If the values of $\%d_i$ are now averaged for a particular ion and a particular stationary phase (to give $\%d_{\text{av}}$), graphical representations of the performance of each model can be obtained, as shown in Figs. 1–3 for the Hamilton PRP-X100, Vydac 302 IC 4.6 and Waters IC Pak A columns, respectively. These values should be zero, or as close to zero as possible. Calculation of $\%d_{\text{av}}$ includes the reference points on which the predictions for each model were made. This was considered necessary in that the ability of a model to return accurate data close to its reference points is an important consideration when carrying out an interpretive optimisation. It was also noted that some models that relied on non-linear calculations to solve their parameters did not return normalised percentage differences of zero at all reference points used. A model that requires use of a large number of reference points to make a prediction will gain an advantage simply because it will have a large number of points with a normalised percentage difference at or close to zero.

Table 3

Normalised percentage differences ($\% d_i$) between the measured and predicted retention times for the seven retention models: dominant equilibrium approach (DE), effective charge approach (EC), dual eluent species model (DES), Kuwamoto model (K), extended Jenke model (EDS), multiple species eluent/analyte model (MSE) and end points model (End) for sulfate on the Hamilton PRP-X100 column

Mobile phase		Normalised % Difference ($\%d_i$)						
pH	P_T	Model						
		DE	EC	DES	K	EDS	MSE	End
4.00	1.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	2.00	-40.30	30.37	6.30	5.70	-7.19	6.30	-22.12
	4.00	-68.55	56.26	22.58	21.56	0.00	22.58	0.00
5.00	1.00	87.66	8.39	-2.92	-8.53	-11.33	-2.92	0.42
	2.00	62.27	23.53	-7.26	-13.44	-22.45	-7.26	-15.52
	4.00	33.31	28.92	-7.14	-13.41	-28.47	-7.14	-7.96
6.00	1.00	113.1	0.00	11.23	7.56	14.68	11.23	0.00
	2.00	92.32	-0.46	4.79	1.29	6.61	4.79	-8.38
	4.00	66.15	-1.35	0.00	-3.13	0.00	0.00	0.00
Average ($\%d_{\text{av}}$)		53.133	16.189	3.067	-0.265	-5.349	3.067	-5.952

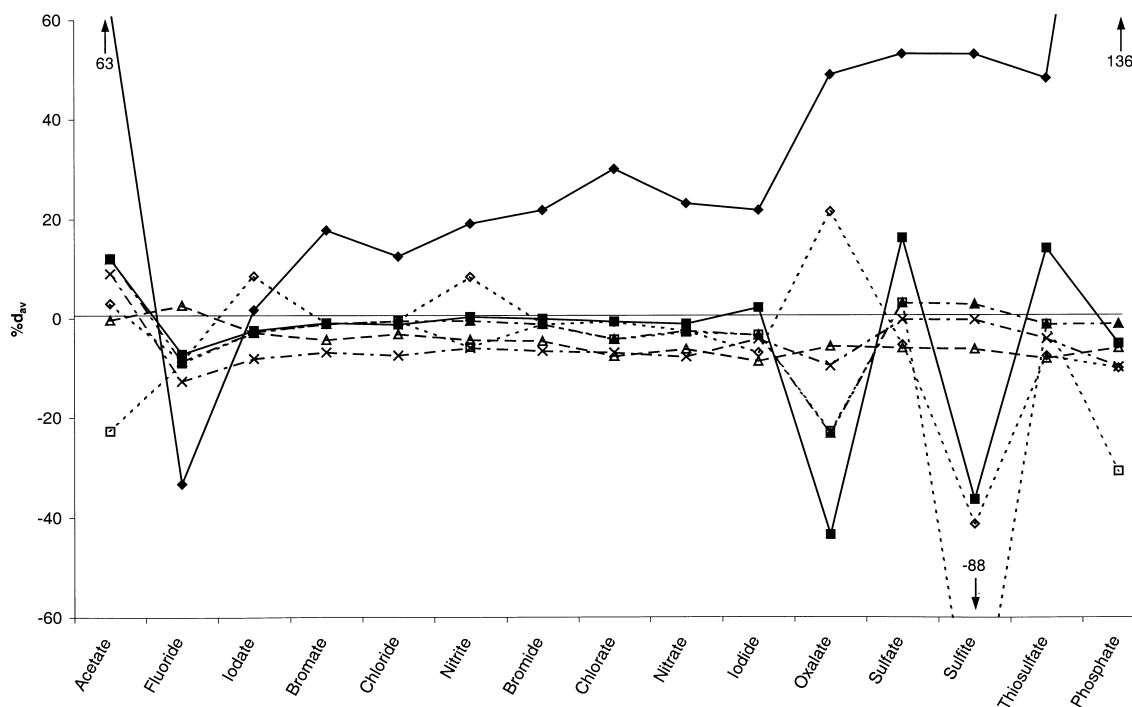


Fig. 1. Average of the normalised percentage differences on the Hamilton PRP-X100 column. ◆, Dominant equilibrium approach; ■, effective charge approach; ▲, dual eluent species model; ×, Kuwamoto model; ◇, extended dual eluent species model; □, multiple species eluent/analyte model; △, end points model.

4.2. Overall results for all analytes and stationary phases

Data similar to Tables 2 and 3 could be generated for each of the analytes using each of the columns. However, the volume of these data necessitates the use of some statistical approach in order that trends may be identified.

The accuracy of the models for all analytes using various phthalate concentrations and pH values in the eluent can be assessed by the global average of the percentage normalised differences ($%d_{\text{glob}}$) achievable for all analytes using each model and each stationary phase, which should be zero. The precision for each model at various phthalate concentrations and pH values for all analytes is given by the standard deviation of the percent normalised differences [$s_{d(\%d)}$], which should be zero. An alternative statistical approach for an overall analysis of each

model is the calculation of the correlation coefficient [14] for each stationary phase and all analytes and eluent compositions using the following equation.

$$\text{Corr. coeff.} = \frac{\sum_{x=1}^n t_{R \text{ act}_x} t_{R \text{ pred}_x}}{\sqrt{\sum_{x=1}^n (t_{R \text{ act}_x})^2 \sum_{x=1}^n (t_{R \text{ pred}_x})^2}} \quad (34)$$

where n is the number of data pairs being compared.

This value demonstrates the ability of the retention model to accurately return retention factors that are the same as those acquired experimentally. The closer the value of the correlation coefficient is to unity, the closer the retention model results are to the experimentally determined results.

Data for $%d_{\text{glob}}$, $s_{d(\%d)}$ and the correlation coefficient (r) for each model on each stationary phase are given in Table 4. The data are presented in three

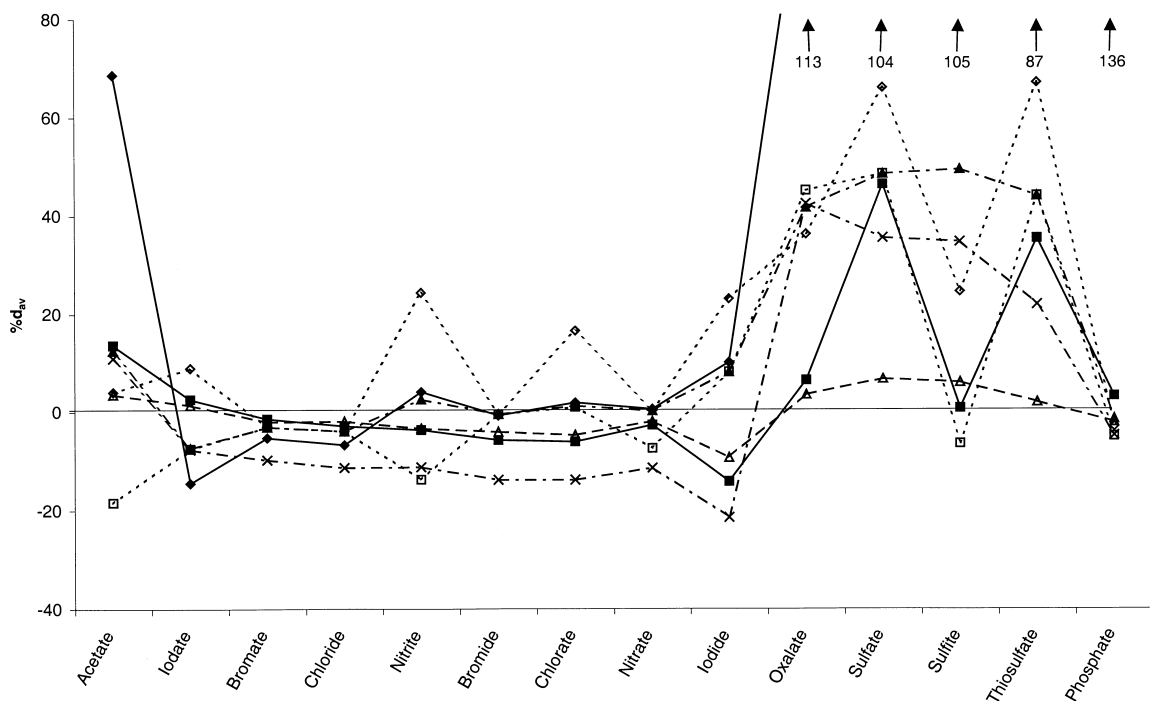


Fig. 2. Average of the normalised percentage differences on the Vydac 302 IC 4.6 column. \blacklozenge , Dominant equilibrium approach; \blacksquare , effective charge approach; \blacktriangle , dual eluent species model; \times , Kuwamoto model; \diamond , extended dual eluent species model; \square , multiple species eluent/analyte model; \triangle , end points model.

ways that reflect three different approaches to the optimisation of eluent composition in IC. In the first, the experimental and predicted data points used to assess the model are confined to those in which the pH is constant at 4.0 and the eluent concentration is varied, whilst in the second the data points are those in which the eluent concentration is constant at 1.0 mM and the pH is varied. The third approach, labelled as two-dimensional, is where all the data points covering variations in both the eluent concentration and pH are included.

The overall performance of each model can be best demonstrated using the correlation coefficient between the predicted and experimental retention times over all stationary phases. This is shown in Fig. 4 as a graph of the correlation coefficient for each analyte as well as the overall performance of each model for all analytes.

There are several other important factors that must be considered when comparing the utility and performance of the retention models and these factors

are summarised in Table 5. The number of experiments required to solve the model should be as small as possible in order to maximise the speed of any optimisation routine using that model. In terms of the complexity of finding a solution, the models fall into two categories: those that have a linear solution and those that must be solved using a process of iterative minimisation. Whilst the latter approach is more time consuming and demanding of computing power, all of the models proved to be relatively straightforward to solve with the exception of the multiple species eluent/analyte model which required the simultaneous determination of two or more constants at higher pH values when hydroxide concentration in the eluent became significant. Furthermore, the choice of the starting point for the iterative minimisation process has a large influence on the chances of finding a solution and the time taken for the process to be completed. The amount of knowledge of the system required to find a solution is also a significant consideration. Most of the models require knowledge

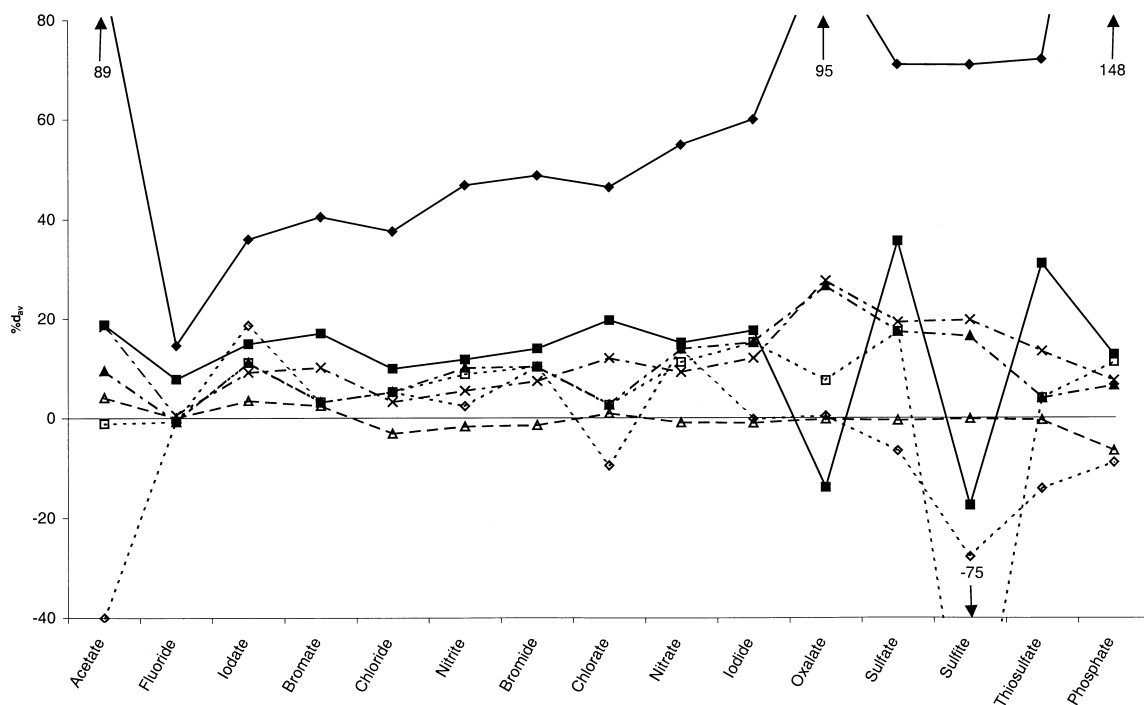


Fig. 3. Average of the normalised percentage differences on the Waters IC Pak A Column. \blacklozenge , Dominant equilibrium approach; \blacksquare , effective charge approach; \blacktriangle , dual eluent species model; \times , Kuwamoto model; \diamond , extended dual eluent species model; \square , multiple species eluent/analyte model; \triangle , end points model.

of the acid dissociation constant(s) of the eluent and some also require knowledge of the acid dissociation constant(s) for each analyte. The reliability of the model is dependent on the quality of these input data, but since the number of eluents suitable for non-suppressed IC of anions is quite limited, the acid dissociation constants can be stored in a database within the software. On the other hand the number of analytes that can be separated using non-suppressed IC is quite large and the requirement for acid dissociation constants for analytes may impose a restriction on the applicability of some models.

4.3. Summary of the performance of the retention models

Figs. 1–4 and Table 4 provide information on which the performance of the retention models can be assessed. Several trends are evident.

(i) The dominant equilibrium model was consistently the worst performer and gave particularly

poor results when applied to analytes with a charge of greater than -1 .

(ii) The remaining models gave reasonable performance for singly charged analytes, but showed erratic behaviour for analytes of higher charge. The exception was the end points model, which showed consistent performance for all analytes.

(iii) The ability of a particular model to predict retention times was dependent to some extent on the type of stationary phase used, and in particular the type of material used to support the functional group. For singly charged analytes, all models except for the dominant equilibrium approach gave negative errors on the polystyrene–divinylbenzene and silica-based stationary phases, whilst for the polymethacrylate stationary phase, positive errors were generally observed. Since all of the theoretical models consider only electrostatic effects leading to ion-exchange retention, the presence of other retention mechanisms (such as adsorption effects between the analyte and the unfunctionalised portions

Table 4

Global average normalised percentage differences ($%d_{\text{glob}}$), standard deviation of the percent normalised differences ($s_{d(\%d)}$) and correlation coefficient (r) between predicted and experimental data for all columns and retention models given for three different types of optimisation techniques

Model	Hamilton PRP-X100			Vydac 302 IC 4.6			Waters IC Pak A		
	$%d_{\text{glob}}$	$s_{d(\%d)}$	r	$%d_{\text{glob}}$	$s_{d(\%d)}$	r	$%d_{\text{glob}}$	$s_{d(\%d)}$	r
<i>Constant pH</i>									
Dominant equilibrium	-25.98	35.64	0.98050	-3.37	36.34	0.96653	-2.49	26.82	0.99742
Effective charge	3.37	25.69	0.99071	210.92	36.19	0.91012	21.78	26.15	0.99169
Dual eluent species	-1.37	11.76	0.99962	70.99	49.56	0.90561	21.88	230.93	0.99078
Kuwamoto	-0.15	11.34	0.99944	16.65	45.41	0.90283	20.64	22.87	0.99118
Extended dual eluent	-2.88	20.06	0.99567	6.77	41.56	0.91994	-1.67	30.75	0.99954
Multiple species	-7.14	16.76	0.99627	0.46	38.14	0.93462	13.35	18.65	0.99500
End points	-4.28	7.45	0.99737	-3.30	11.58	0.99194	-1.07	4.46	0.99991
<i>Constant total eluent concentration</i>									
Dominant equilibrium	56.86	51.28	0.97676	56.64	62.20	0.89719	67.37	57.46	0.94888
Effective charge	-0.63	7.16	0.99811	2.89	12.33	0.98696	6.04	11.22	0.99679
Dual eluent species	50.90	110.99	0.99672	24.44	21.21	0.97537	2.24	11.02	0.99782
Kuwamoto	-1.40	7.30	0.99843	2.52	8.79	0.99531	5.01	6.76	0.99796
Extended dual eluent	4.45	23.75	0.98394	24.12	33.51	0.93456	-4.01	23.03	0.98812
Multiple species	-2.43	22.46	0.96341	13.44	25.12	0.97290	0.38	19.37	0.97129
End points	-1.35	4.50	0.99960	2.20	9.44	0.99275	4.33	7.43	0.99846
<i>Two-dimensional (overall performance)</i>									
Dominant equilibrium	35.27	69.61	0.71109	43.21	69.79	0.86037	62.00	600.92	0.63566
Effective charge	-3.28	21.63	0.94778	5.11	31.82	0.89549	12.72	23.05	0.96575
Dual eluent species	-2.00	13.08	0.95285	13.70	34.27	0.91320	10.07	18.89	0.97234
Kuwamoto	-5.39	12.24	0.99340	2.87	33.57	0.89020	11.59	17.50	0.97963
Extended dual eluent	-3.13	210.91	0.82179	18.62	39.44	0.86994	-3.55	26.34	0.95454
Multiple species	-12.81	31.43	0.83703	5.76	320.91	0.92570	2.03	30.54	0.84768
End points	-4.74	8.65	0.99527	-0.44	11.34	0.98399	-0.34	9.72	0.99190

The lowest result in each category is shown in bold face for convenience.

of the stationary phase) or the occurrence of factors influencing the ion-exchange process (such as steric effects between the analyte and the functional group on the ion exchanger) will influence the predictive ability of the models.

(iv) The six theoretical models gave improved accuracy and precision as their complexity increased. However, no theoretical model gave reliable predictions of retention times for all analytes, presumably due to the inability of such models to consider secondary contributions to analyte retention, such as those mentioned in (iii) above. On the other hand, the empirical approach exemplified by the end points model was applicable to all analytes and stationary phases. This was particularly evident when predictions were made under conditions where both the

eluent concentration and pH were varied (i.e., for a two-dimensional optimisation, see Table 4).

Taking into account the factors outlined in Table 5, together with the data on the accuracy, precision and correlation coefficient of each model discussed earlier, it can be seen that the end points model offers ease of numerical solution and requires minimal input data, while at the same time providing the most reliable prediction of retention times. The only disadvantage of this method is that it requires the greatest number of initial experiments when a two-dimensional optimisation is to be performed. A further advantage of the end points model when used as a basis for optimisation is that its accuracy can be improved by iteration during the optimisation process. This is demonstrated in Fig. 5 which shows an

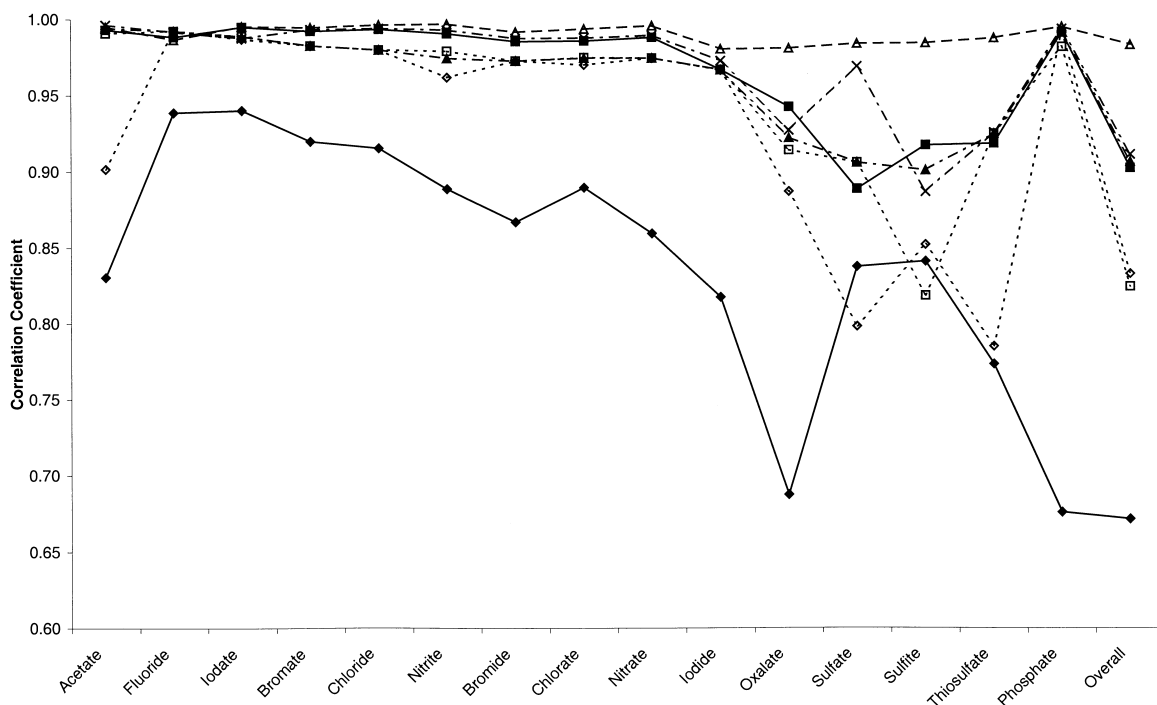


Fig. 4. Correlation coefficients between the predicted and actual retention times given for each analyte separately and combined overall, on all columns. ♦, Dominant equilibrium approach; ■, effective charge approach; ▲, dual eluent species model; ×, Kuwamoto model; ◇, extended dual eluent species model; □, multiple species eluent/analyte model; △, end points model.

optimisation based on four initial experiments, a–d, using mobile phase conditions situated at the corners of the search area. After prediction of an initial optimum at point e, an experiment is conducted using these conditions and the measured retention

times compared to those predicted by the model. If the agreement between measured and predicted retention times is less than that desired, the optimisation is repeated using point e as a new experimental value, which results in the initial search area

Table 5

Other important factors that must be considered when choosing a retention model for optimisation

Model	No. of experimental data points needed	Method used to solve model	Knowledge required to solve model
Dominant equilibrium	1	Linear solution	Analyte and eluent charge
Effective charge	2 (1) ^a	Linear solution	Analyte charge and pK_a of eluent
Dual eluent species	2	Iterative minimisation	Analyte charge and pK_a of eluent
Kuwamoto	2	Linear solution, but restricted pH selection	Analyte charge and pK_a of eluent
Extended dual eluent species	3 (2) ^b	Iterative minimisation	pK_a of eluent and analyte
Multiple eluent species	2 or 3	Iterative minimisation	pK_a of eluent and analyte
End points	4 (2) ^a	Linear solution	pK_a of eluent

^a For one-dimensional experiment.

^b Simplifies to the dual eluent species model for some analytes.

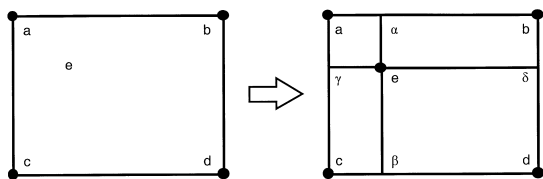


Fig. 5. Diagram of how an extra experiment can be added to the End Points Model to break the experiment into a series of smaller end points models.

being subdivided into smaller search areas. The boundary points that have not been acquired experimentally can be found by calculating their retention times from the original End Points Model, a–d. The advantage of this approach is that the boundary points used by the retention model are now closer together, which should result in improved accuracy of the model. The iterative process can be continued until the agreement between the measured and predicted retention times for the optimal conditions is within desired limits.

5. Conclusions

The retention characteristics of a number of anions on a series of ion chromatographic columns were studied as a function of changing mobile phase concentration and pH. The retention characteristics were compared statistically with those predicted by a number of retention models found in the literature. A new retention model, based on the linear solvent strength model and empirical measurements was also developed, called the end points model. The models were studied to find their suitability for use in computer assisted interpretive optimisation of anion chromatography. All the models studied were found to mimic the behaviour of all species examined and on all columns used with varying effectiveness. A

number of the models were found to have limitations that eliminated them from consideration, leaving only a few models suitable for comparison. Of the remaining models the end points model was found to be the most suitable due to its high degree of accuracy and precision, low level of knowledge requirements and excellent ruggedness.

Acknowledgements

Financial support from the Dionex Corporation is gratefully acknowledged.

References

- [1] D.T. Gjerde, G. Schmuckler, J.S. Fritz, *J. Chromatogr.* 12 (1980) 35.
- [2] P.R. Hadda, C.E. Cowie, *J. Chromatogr.* 303 (1984) 321.
- [3] M.J. van Os, J. Slanina, C.L. de Ligny, W.E. Hammers, J. Agerdenbos, *Anal. Chim. Acta* 1 (1982) 73.
- [4] P.R. Haddad, P.E. Jackson, *Ion Chromatography: Principles and Applications*, Elsevier, Amsterdam, 1990, Ch. 5.
- [5] T.B. Hoover, *Sep. Sci. Technol.* 17 (1982) 295–305.
- [6] D.R. Jenke, G.K. Pagenkopf, *Anal. Chem.* 56 (1984) 85.
- [7] D.R. Jenke, G.K. Pagenkopf, *Anal. Chem.* 56 (1984) 88.
- [8] D.R. Jenke, *Anal. Chem.* 56 (1984) 2674.
- [9] M. Maruo, N. Hirayama, T. Kuwamoto, *J. Chromatogr.* 481 (1989) 315–322.
- [10] D.R. Jenke, *Anal. Chem.* 66 (1994) 4466–4470.
- [11] P. Hajós, O. Horváth, V. Denke, *Anal. Chem.* 67 (1995) 434–441.
- [12] A.D. Sosimenko, P.R. Haddad, *J. Chromatogr.* 546 (1991) 37–59.
- [13] M. Meloun, J. Havel, E. Högföldt, *Computation of Solution Equilibria: A Guide to Computational Methods in Extraction, Potentiometry and Spectrophotometry*, Ellis Horwood, Chichester, 1985, pp. 101–108.
- [14] C.H. Goulden, *Methods of Statistical Analysis*, Wiley, New York, 1939, Ch. VII.