

**MODELING OF ION-EXCLUSION AND VACANCY
ION-EXCLUSION CHROMATOGRAPHY
ON A STRONGLY ACIDIC CATION-EXCHANGE RESIN
IN THE H⁺ FORM**

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ABSTRACT

Ion-exclusion chromatography (IEC) can sometimes be used for separation of the weak acids using pure water as mobile phase. Characteristic leading (i.e. frontally tailing) peaks are obtained and retention depends on the concentration of solutes. It was recently shown that this order could be reversed. In vacant ion exclusion chromatography (vIEC) sample flows as mobile phase and pure water is injected as the sample. Symmetrical peaks are obtained; this is believed to be because of self-buffering of the solutes in the sample. The aim of the work discussed in this paper was to describe the mechanism of retention in IEC and vIEC by using the modified equilibrium–dispersive (ED) model. It was found that the retention times and peak shapes predicted by the derived equations are in good agreement with experimental data. These equations also predict new features of vIEC, and these were confirmed experimentally. It was shown that in vIEC, in contrast with IEC, symmetrical peaks are obtained even for a single analyte.

INTRODUCTION

Ion exclusion chromatography (IEC) is a chromatographic technique used for separation of partially ionized molecules [1–5]. When the pure ion-exclusion mechanism of retention is involved, retention volumes of medium-strength electrolytes are proportional to their dissociation constants. Strong and weak electrolytes are separated, the first at the beginning

of the elution order and the latter at the end. As with other chromatographic techniques, the name 'IEC' is derived from the predominant mechanism of retention, i.e. ion exclusion. Other mechanisms (e.g. adsorption on the resin, the screening effect, or size exclusion) enable separation of sugars or alcohols [6–8]. The dead volume (volume of mobile phase) and the inner volume of the chromatographic column (the total volume occupied by the water in a column) can be determined from the dependence of retention volumes on dissociation constants [9]. The retention mechanism in IEC has been described using the so-called global thermodynamic equations and by computational modeling of the column, using the Craig method [1].

The characteristic feature of IEC is the same charge on the dissociated functional groups of the ion-exchange resin and on the solute. Negatively charged ions (e.g. dissociated acidic compounds) are therefore separated on cation-exchange resins with anionic functional groups, usually sulfonic acid groups. Although the same column can be used for both IEC and ion-exchange chromatography, true ion-exchange reactions are not involved in IEC. For the specific requirements of IEC a large ion-exchange capacity is preferred. An increase in this capacity can be achieved by increasing the column dimensions or the concentration of functional groups, or by using strong exchangers. Water molecules accumulate, as hydration spheres, around the dissociated functional groups of the support. Contained in the resin pores and in the hydration spheres, they are immobilized, forming a stationary phase. Neutral, uncharged molecules penetrate into the resin whereas similarly charged co-ions are repelled by the presence of the dissociated functional groups immobilized in the resin skeleton. Similarly to the Donnan membrane equilibrium, the hydrated resin network behaves as a semi-permeable membrane between the stationary and the mobile phases. With the exception of the covalently bound functional groups, all other species are freely exchanged through this hypothetical membrane.

In IEC dilute sulfuric acid is frequently used as mobile phase. The small amount of sulfuric acid prevents peak tailing, which is frequently observed when pure water is used. Another method of avoiding tailing is to employ vacancy ion exclusion chromatography (vIEC). In this technique a mixture of analytes is used as mobile phase and pure water is injected as the sample [10–14]. In the same way as in IEC, in vIEC the main retention mechanism, based on exclusion of ions, can be coupled with complementary adsorption of an analyte by the resin skeleton.

The aim of this paper is to describe the mechanism of retention in IEC and vIEC, using the equilibrium–dispersive (ED) model. The effect on

retention of two factors in particular, the dissociation constant and the concentration of the solute, will be discussed. The chromatographic system was chosen in such a way as to avoid adsorption of analytes on resin skeleton.

THEORY

Retention of Acids in Ion-Exclusion Chromatography with Very Dilute Solutions

To model peak retention in analytical chromatography, it generally suffices to develop an equation for the retention factor, k .

In the simplest model of ion-exclusion chromatography, when analyte adsorption can be neglected and assuming total exclusion of co-ions from the water occluded in the adsorbent pores, the retention factor can be expressed by the equation [15]:

$$k = \frac{t_r - t_0}{t_0} = \frac{C_{S,HR} V_S}{C_{HR} V_m + C_{R^-} V_m} \quad (1)$$

where V_m is the volume of the mobile phase (the so-called dead volume), V_S is the volume of liquid inside the resin in the column (the stationary liquid phase), C_{HR} is the concentration of acid in the mobile phase, $C_{S,HR}$ is the concentration of the acid in the stationary liquid phase, C_{R^-} is the concentration of ion R^- in mobile phase, t_r is the retention time, and t_0 is the dead time (i.e. the elution time of species totally excluded from adsorbent pores).

For a wider discussion it is convenient to define several variables [16,17] including the external porosity (bulk porosity, voidage), $\varepsilon_e = V_m/V_k$, where V_k is the column volume, the particle porosity (internal porosity of the particles), $\varepsilon_p = V_S/V_a$, where V_a is the adsorbent volume (the sum of the skeleton resin volume and the stationary phase), and the total porosity

$$\varepsilon_t = \frac{\text{total volume occupied by eluate in column}}{\text{column volume}} .$$

It is easy to verify that the following relationship $\varepsilon_t = \varepsilon_e + (1 - \varepsilon_e)\varepsilon_p$ is valid. Taking into the account definition of external porosity, the dead time can be expressed by the equation:

$$t_0 = \frac{L\varepsilon_e}{u} \quad (2)$$

where u is the superficial velocity. Introducing definition of porosity to eq. (1), the expression for retention time can be written as:

$$t_r = t_0 + t_0 \frac{C_{S,HR}}{C_{HR} + C_{R^-}} \frac{\varepsilon_p(1 - \varepsilon_e)}{\varepsilon_e} = t_0 \left(1 + \frac{C_{S,HR}}{C_{HR} + C_{R^-}} \frac{\varepsilon_t - \varepsilon_e}{\varepsilon_e} \right) \quad (3)$$

It is assumed that equilibrium between HR and R^- in the mobile phase is established immediately, i.e.:

$$C_{HR} K_a = C_{R^-} C_{H^+} \quad (4)$$

and that resistance mass transfer of the acid from the bulk phase to the resin can be ignored, i.e.:

$$C_{S,HR} = C_{HR} \quad (5)$$

Then, taking into the account eqs (2), (4), and (5), eq. (3), can be rewritten as:

$$t_r = \frac{L}{u} \left(\varepsilon_e + \frac{\varepsilon_t - \varepsilon_e}{1 + \frac{K_a}{C_{H^+}}} \right) \quad (6)$$

The retention times of peaks in IEC can be calculated from eq. (6), in which the dependency of retention time on acid concentration is clearly visible. The error in the calculation of retention time by use of this equation increases with increasing acid concentration and it cannot therefore be used for calculation of retention for high concentrations of analytes in IEC and vIEC, because of changes in ion concentrations in the course of migration of the acid peaks or acid vacancies along the column. To calculate the retention time, an appropriate mass transfer equation must be solved.

The Mass Transfer Model

Retention time in chromatography depends mainly on the thermodynamics of adsorption, whereas the shape of the peak profile is a function of dispersion and resistance to mass transfer in the bulk phase and on the adsorbent. In IEC the retention is also a function of external and the particle porosity. The general rate model takes all these factors into account [16–18]. When mass-transfer resistance is not extremely low, the lumped pore-diffusion model (POR) is recommended [18]. Finally, when mass-

transfer resistance is negligible, the equilibrium–dispersive (ED) [17] model is used. In this work, a suitable ED model was used for investigation of IEC. Adsorption of the analyte in the resin was ignored. It was convenient to develop the ED model used in this study from the POR model.

The POR model applied to ion-exclusion chromatography enables definition of the mass transfer equation for the acid (HR) in the mobile phase and in the adsorbent particles, and for the R^- ions in the mobile phase only – assuming the co-ions are totally excluded from the adsorbent pores (i.e. from the stationary volume). Mass transfer for the acid in the mobile phase is given by:

$$\varepsilon_e \frac{\partial C_{HR}}{\partial t} + u \frac{\partial C_{HR}}{\partial z} = \varepsilon_e D_L \frac{\partial^2 C_{HR}}{\partial z^2} - (1 - \varepsilon_e) k_{0,HR} a_p (C_{HR} - C_{p,HR}) - \varepsilon_e r_{HR} \quad (7)$$

where D_L is the dispersion coefficient, $a_p = 3/R_p$, R_p is the particle diameter, k_0 is the overall mass-transfer coefficient, and r_{HR} characterizes the dissociation reaction:

$$r_{HR} = k_a \left(C_{HR} - \frac{1}{K_a} C_{R^-} C_{H^+} \right) = k_a \left(C_{HR} - \frac{1}{K_a} C_{R^-}^2 \right) \quad (8)$$

where k_a is the dissociation rate constant, and the difference between the concentrations of the R^- and H^+ ions was ignored. It is usually assumed that the dissociation reaction is infinitely fast. For the sake of convenience, for numerical solution of this model a finite rate of dissociation was assumed. It should, however, be noted that for high enough values of k_a the solution obtained for a finite value of the rate constant approximates that with for an infinite rate constant with negligible error only. In this work, the k_a was chosen in such way that the growth of its value did not affect the simulated peak profiles. The mass balance for R^- ions in the mobile phase is given by:

$$\varepsilon_e \frac{\partial C_{R^-}}{\partial t} + u \frac{\partial C_{R^-}}{\partial z} = \varepsilon_e D_L \frac{\partial^2 C_{R^-}}{\partial z^2} + \varepsilon_e r_{HR} \quad (9)$$

and the mass balance for HR in water occluded in the adsorbent's pores (i.e. in the stationary volume), ignoring adsorption in the resin is given by:

$$\varepsilon_p \frac{\partial C_{S,HR}}{\partial t} = k_{0,HR} a_p (C_{HR} - C_{S,HR}) \quad (10)$$

It was also assumed, that mass transfer resistances could be neglected. With this assumption, concentrations of acid in the two water phases were equal: $C_{HR} = C_{S,HR}$. After elimination of the mass transfer terms from eqs (7) and (10) it is easy to obtain the equilibrium–dispersive model (eqs 11 and 12) used in this work, remembering that $\varepsilon_t = \varepsilon_e + (1 - \varepsilon_e)\varepsilon_p$:

$$\frac{\partial C_{HR}}{\partial t} + \frac{u}{\varepsilon_t} \frac{\partial C_{HR}}{\partial z} = D_L \frac{\varepsilon_e}{\varepsilon_t} \frac{\partial^2 C_{HR}}{\partial z^2} - \frac{\varepsilon_e}{\varepsilon_t} r_{HR} \quad (11)$$

$$\frac{\partial C_{R^-}}{\partial t} + \frac{u}{\varepsilon_e} \frac{\partial C_{R^-}}{\partial z} = D_L \frac{\partial^2 C_{R^-}}{\partial z^2} + r_{HR} \quad (12)$$

Model expressed by eqs. (11) and (12) were solved assuming the following initial and boundary conditions:

- *For IEC*

For $t = 0$ and $z > 0$, $C_{HR} = C_{R^-} = 0$

For $0 < t < t_{injection}$ and $z = 0$, the inlet concentration of the non-dissociated acid and ion was calculated from eq. (4) and from the relationship $C_{HR} + C_{R^-} = C_{inlet}$;

For $t > t_{injection}$ and $z = 0$, $C_{HR} = C_{R^-} = 0$

For $t > 0$ and $z = L$ it was assumed that: $\frac{\partial C_{HR}}{\partial z} = \frac{\partial C_{R^-}}{\partial z} = 0$

- *For vIEC*

For $t = 0$, $t > t_{injection}$ and $z > 0$, the concentration of the non-dissociated acid and ion was calculated from eq. (4) and from the relationship $C_{HR} + C_{R^-} = C_{inlet}$

For $0 < t < t_{injection}$ and $z = 0$ $C_{HR} = C_{R^-} = 0$

For $t > 0$ and $z = L$ it was assumed that $\frac{\partial C_{HR}}{\partial z} = \frac{\partial C_{R^-}}{\partial z} = 0$

The model expressed by eqs (11) and (12) were solved using the orthogonal collocation method on finite elements [19,20]. From these models and after the next simplification, the retention time formula given by eq. (6) can also be developed. Assuming that the dissociation kinetics are infinitely fast, and neglecting the dispersion terms and adding eqs (11) and (12), the following formula is obtained:

$$\varepsilon_t \frac{\partial C_{HR}}{\partial t} + \varepsilon_e \frac{\partial C_{R^-}}{\partial t} + u \frac{\partial C_{HR}}{\partial z} + u \frac{\partial C_{R^-}}{\partial z} = 0 \quad (13)$$

or, taking into the account that, $\partial C_{R^-} = \frac{C_{H^+}}{K_a} \partial C_{HR}$ we have:

$$\left(\varepsilon_t \frac{C_{H^+}}{K_a} + \varepsilon_e \right) \frac{\partial C_{R^-}}{\partial t} + u \left(1 + \frac{C_{H^+}}{K_a} \right) \frac{\partial C_{R^-}}{\partial z} = 0 \quad (14)$$

and, finally:

$$\frac{\partial C_{R^-}}{\partial t} + \frac{u \left(1 + \frac{C_{H^+}}{K_a} \right)}{\left(\varepsilon_t \frac{C_{H^+}}{K_a} + \varepsilon_e \right)} \frac{\partial C_{R^-}}{\partial z} = 0 \quad (15)$$

The second term before the partial derivative in eq. (15) represents the velocity of migration of the part of the peak in which concentration is C_{H^+} . Taking this fact into account, eq. (6) can be obtained.

EXPERIMENTAL

Instrumentation and Procedure

Ion separation was performed with Tosoh (Tokyo, Japan) Model IC-2001 ion chromatograph equipped with vacuum degasser, pump, column oven, auto-sampler, and conductivity detector. IEC and vIEC experiments were performed on a polystyrene–divinylbenzene-based strongly acidic cation-exchange resin column in the H^+ -form (Tosoh TSKgel SCX; 150 mm \times 6 mm i.d., 5 μ m particle size, pore diameter 60 Å, 1.5 mequiv mL^{-1} cation-exchange capacity). The experimental conditions were: column temperature 40°C, flow rate 0.5 $mL\ min^{-1}$, and injection volume 30 μ L. The separation column was equilibrated until stable mobile phase background conductivity was obtained (i.e. after ca. 60 min).

Reagents

All reagents were of the analytical reagent grade from Wako (Osaka, Japan). Standard solutions were prepared in distilled and deionized

water. Appropriate amounts of analyte samples at a concentration of 0.1 M were diluted with water, as necessary.

Ion-Exclusion Chromatography

The mobile phase was distilled and deionized water. Injection sample: 1 mM H₂SO₄ as an indicator of the dead volume (V_m) and 1 mM NaHCO₃ as an indicator of the sum of dead volume and the volume of the mobile phase contained within the pores of the resin beads (inner column volume = $V_s + V_m$) [21]. The standard samples injected into the separation column were 0.005, 0.05, 0.25, 0.5, 1.0, 2.0, 2.5 and 5.0 mM oxalic acid, and 0.5, 1.0, 2.5, 5.0 and 8.0 mM acetic acid.

Vacancy Ion-Exclusion Chromatography

The mobile phase was 0.005, 0.05, 0.25, 0.5, 1.0, 2.0, 2.5, or 5.0 mM oxalic acid, or 0.5, 1.0, 2.5, 5.0, or 8.0 mM acetic acid. The sample injected was the distilled and deionized water.

Model Data

For the calculations relating to IEC and vIEC dissociation constants, K_a , of 4.266×10^{-2} and 2.95×10^{-5} were used for oxalic and acetic acids, respectively [22]. Numerical values of u , ε_t , ε_e , and D_L necessary for solution of the ED model expressed by eqs (11) and (12), were calculated on the basis of the above mentioned experimental conditions. The superficial velocity was $1.768 \text{ cm min}^{-1}$ and the injection time was 0.06 min. External porosity, ε_e , calculated from the retention of H₂SO₄ was 0.253. To calculate this from the measured retention volume of sulfuric acid (1.172 mL) the numerical value of the external porosity, an extra volume corresponding to the dead time and equal to 0.0981 mL was subtracted. In a similar way, the total porosity ($\varepsilon_t = 0.876$) was calculated from the retention of NaHCO₃ [21]. The measured NaHCO₃ retention volume was 3.72 mL.

To develop the equilibrium–dispersive (ED) model, mass-transfer resistances were ignored. In practice, however, mass transfer resistances always exist and in the ED model they are indirectly taken into the account by formally regarding the dispersion coefficient (D_L) as the apparent dispersion coefficient [17], which includes the dispersion and the mass-transfer resistances. The apparent dispersion coefficient, calculated by use of the formula $N = uL/(\varepsilon_t D_L)/2$ [17] was $D_L = 0.0076 \text{ cm}^2 \text{ min}^{-1}$, where the number of the theoretical plates (N) was estimated from the H₂SO₄ peak, using the half bandwidth method (N was approx. 2000).

It should be noted that, in contrast with oxalic and acetic acids, sulfuric acid does not penetrate inside the adsorbent particles, so the number of the theoretical plates can be overestimated by use of the first two acids. In this study, however, this error was neglected.

RESULTS AND DISCUSSION

Ion-Exclusion Chromatography

Experimental and calculated retention times of the peak apexes are presented in Table I. The experimental retention times were calculated by subtracting the extra volume dead time (which was equal to 0.196 min) from the measured time. To calculate theoretical retention times and peak

Table I

The retention times of oxalic acid and acetic acid obtained in IEC

| Concentration of sample injected (mM) | Oxalic acid | | Acetic acid | |
|---------------------------------------|-----------------------------------|--|-----------------------------------|--|
| | Experimental retention time (min) | Calculated retention time (min) for $\varepsilon_t = 0.70$ | Experimental retention time (min) | Calculated retention time (min) for $\varepsilon_t = 0.70$ |
| 0.005 | 2.16 | 2.17 | – | – |
| 0.05 | 2.22 | 2.18 | – | – |
| 0.25 | 2.28 | 2.20 | – | – |
| 0.5 | 2.31 | 2.22 | 4.30 | 4.45 |
| 1.0 | 2.35 | 2.26 | 4.71 | 4.89 |
| 2.0 | 2.37 | 2.32 | – | – |
| 2.5 | 2.40 | 2.36 | 5.23 | 5.32 |
| 5.0 | 2.44 | 2.43 | 5.53 | 5.56 |
| 8.0 | – | – | 5.67 | 5.67 |

profiles the model given by eqs (11) and (12) was solved. The retention time predicted for acetic acid was, however, ca. 1 minute longer than that measured experimentally. This error can result from an inaccurate value of the external or total porosity. Incorrect calculation of the external porosity was virtually impossible under our experimental conditions, because completely dissociated strong sulfuric acid is repelled from the adsorbent. Use of NaHCO_3 for calculation of total porosity can also be imperfect. It seems that NaHCO_3 can slightly be adsorbed and, as a result, the calculated total

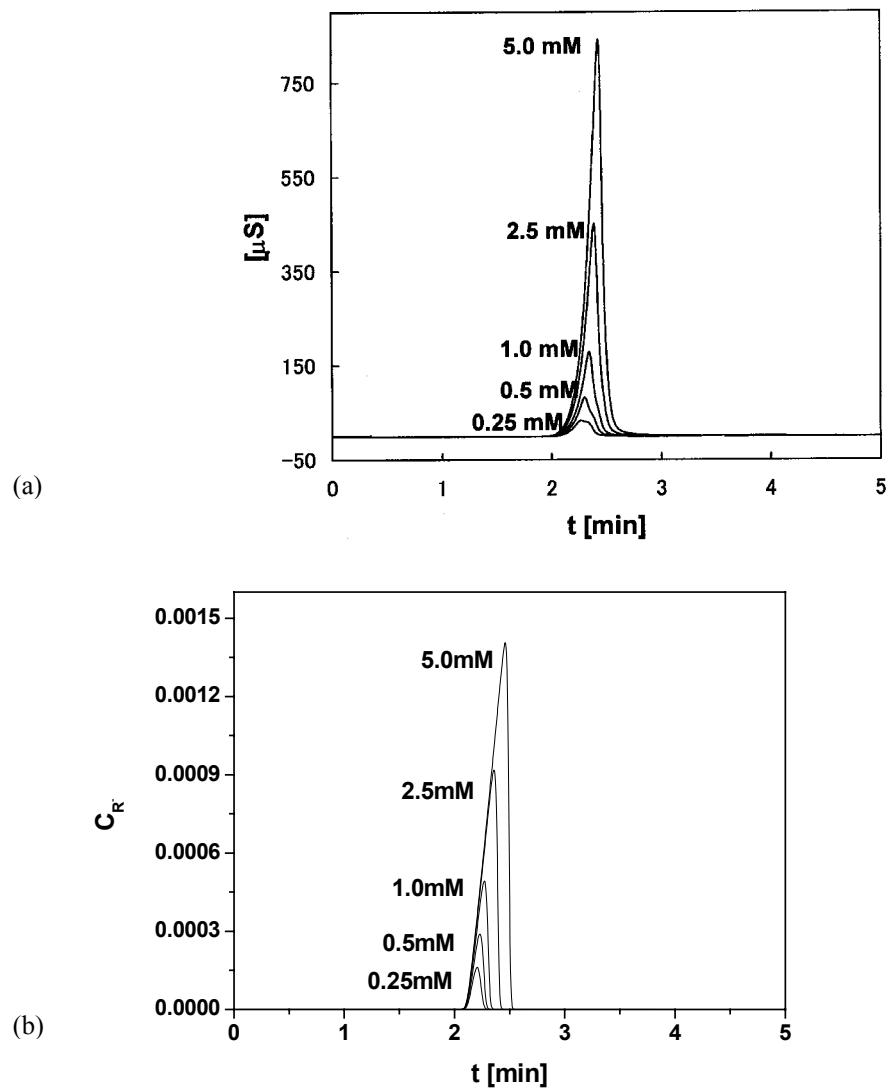


Fig. 1

Comparison of (a) experimental peak profiles and (b) theoretical peak profiles for oxalic acid in IEC. The inlet concentrations (c_{inlet}) were 0.25, 0.5, 1.0, 2.5, and 5.0 mM

porosity can be overestimated. The total porosity was finally estimated by comparison of experimental and the theoretical retention times of acetic acid for its inlet concentration of 8 mM. The estimated value ($\varepsilon_t = 0.70$) was employed in all the subsequent calculations.

From comparison of the retention times given in Table I for both relatively strong oxalic acid and relatively weak acetic acid it is apparent that the maximum error of prediction of the retention times was always less than 4%. Very good agreement was also obtained between experimental and the simulated peak profiles (Figs 1 and 2).

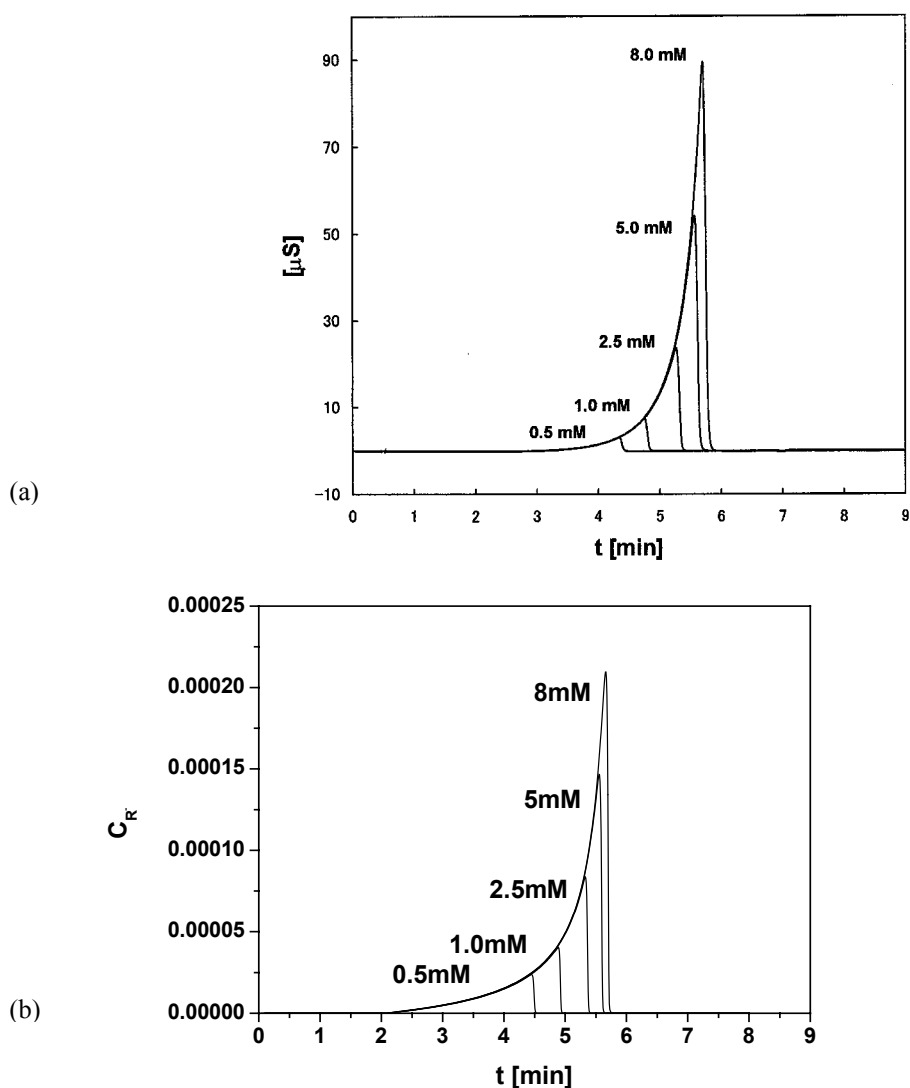


Fig. 2

Comparison of (a) experimental peak profiles and (b) theoretical peak profiles for acetic acid in IEC. The inlet concentrations (c_{inlet}) were 0.5, 1.0, 2.5, 5.0, and 8.0 mM

Vacancy Ion-Exclusion Chromatography

Experimental and the calculated retention times of the peak apexes are presented in Table II. The experimental retention time was corrected by subtracting the extra volume dead time from the measured time. Relative differences between experimental and theoretical retention times for acetic acid are similar to those observed for IEC. For oxalic acid, the relative calculation error of the retention time has increased to 10%. It seems, however, that this increase in the error is partly because of experimental error (for example, compare the experimental retention data for oxalic acid concentrations of 1 and 2 mM).

Table II

Retention times of oxalic and acetic acids obtained in vIEC, and the pH of the mobile phases

| Concentration of sample injected (mM) | Oxalic acid | | | Acetic acid | | |
|---------------------------------------|-------------|-----------------------------------|---|-------------|-----------------------------------|---|
| | pH | Experimental retention time (min) | Calculated retention time (min) for $\epsilon_t = 0.70$ | pH | Experimental retention time (min) | Calculated retention time (min) for $\epsilon_t = 0.70$ |
| 0.005 | 5.08 | 2.20 | 2.17 | – | – | – |
| 0.05 | 4.16 | 2.36 | 2.18 | – | – | – |
| 0.25 | 3.56 | 2.42 | 2.19 | – | – | – |
| 0.50 | 3.28 | 2.46 | 2.21 | 4.10 | 5.71 | 5.44 |
| 1.0 | 3.00 | 2.52 | 2.26 | 3.96 | 5.82 | 5.58 |
| 2.0 | 2.71 | 2.51 | 2.36 | – | – | – |
| 2.5 | 2.66 | 2.64 | 2.41 | 3.71 | 5.91 | 5.72 |
| 5.0 | 2.36 | 2.64 | 2.64 | 3.56 | 5.97 | 5.79 |
| 8.0 | – | – | – | 3.45 | 5.98 | 5.83 |

The theoretical peak profiles obtained for the two acids, presented in Figs 3 and 4, are again in good agreement with experimental results.

CONCLUSION

In this paper a simple equilibrium–dispersive (ED) model is proposed for prediction of retention for two non-adsorbed organic acids in the ion-exclusion chromatography (IEC) and in vacancy ion-exclusion chromatography (vIEC). The model was successfully tested by use of a stron-

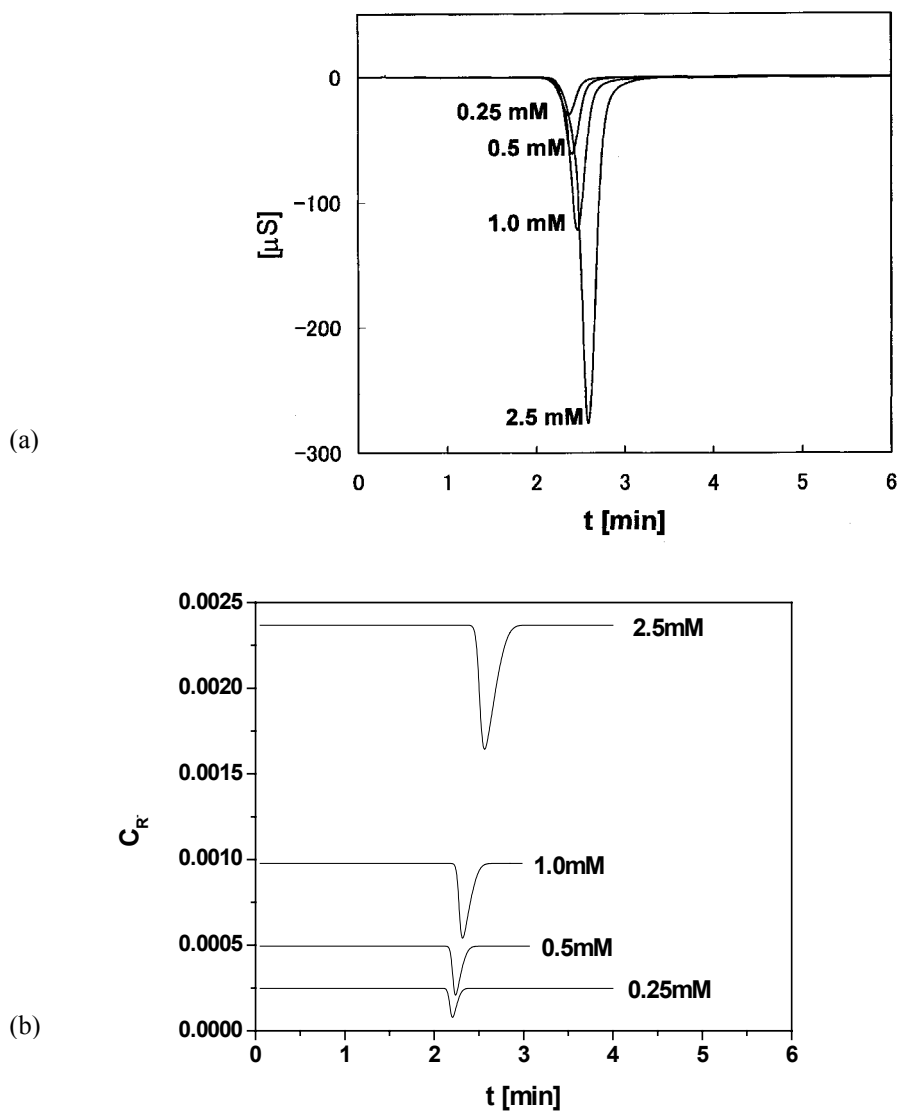


Fig. 3

Comparison of (a) experimental peak profiles and (b) theoretical peak profiles for oxalic acid in vIEC. The mobile phase concentrations (C_{inlet}) were 0.25, 0.5, 1.0, 2.5, and 5.0 mM

gly acidic ion-exchange resin, in the H^+ form, characterized by a small pore diameter, 60 Å. It was found that retention values and peak shapes predicted by use of the derived equations were in good agreement with experi-

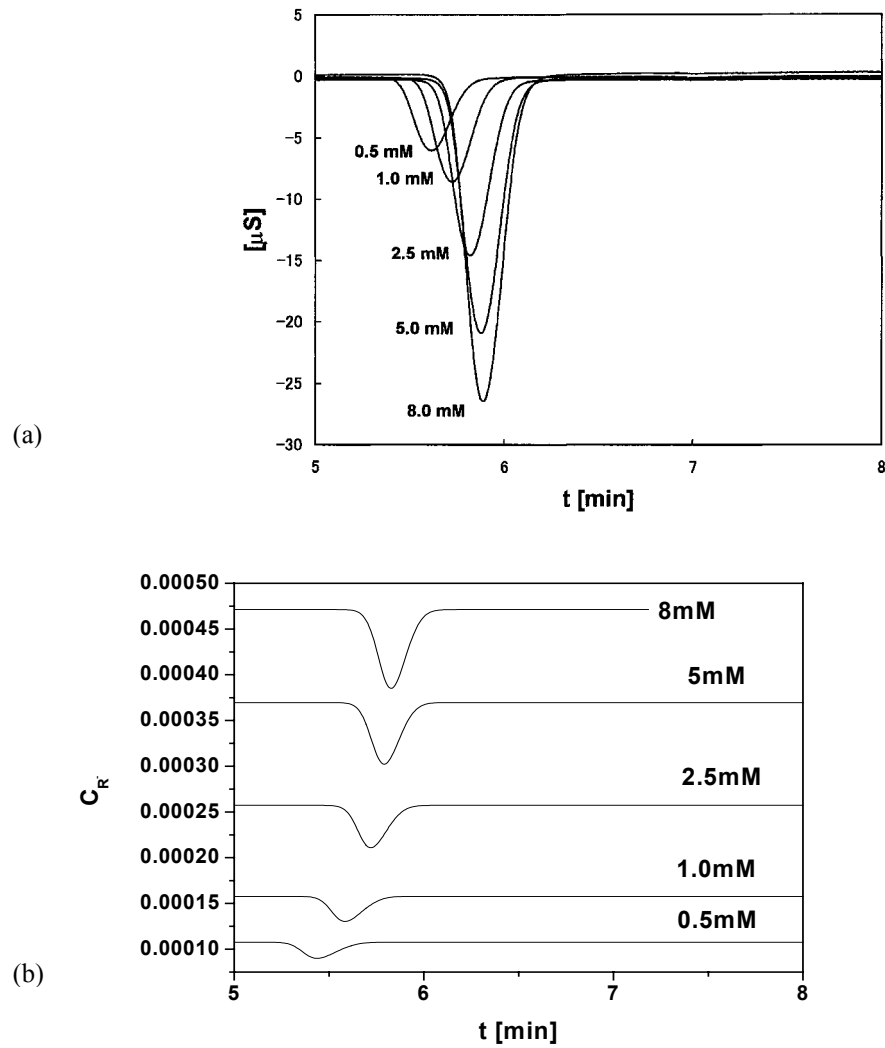


Fig. 4

Comparison of (a) experimental peak profiles and (b) theoretical peak profiles for acetic acid in vIEC. The mobile phase concentrations (c_{inlet}) were 0.5, 1.0, 2.5, 5.0, and 8.0 mM

mental data for a wide range of concentration of the analytes investigated. These equations also predict new features of vIEC which were confirmed experimentally. For example it was found that in vIEC, in contrast with IEC, symmetrical peaks were obtained even for a single solute. The model

can be directly extended to examples in which several analytes are separated and in which the analytes are adsorbed on the resin. Modeling these particular examples will be the subject of our next paper.

REFERENCES

- [1] B.K. Głód, *Neurochem. Res.*, **22**, 1237 (1997)
- [2] B.K. Głód, *Acta Chromatogr.*, **12**, 122 (2002)
- [3] K. Tanaka and J.S. Fritz, *J. Chromatogr.*, **361**, 151 (1986)
- [4] K. Tanaka, M. Mori, Q. Xu, M.I.H. Helaleh, M. Ikedo, H. Taoda, W. Hu, K. Hasebe, J.S. Fritz, and P.R. Haddad, *J. Chromatogr. A*, **997**, 127 (2003)
- [5] K. Ito, Y. Takayama, M. Ikedo, M. Mori, H. Taoda, Q. Xu, W. Hu, H. Sunahara, T. Hayashi, S. Sato, T. Hirokawa, and K. Tanaka, *J. Chromatogr. A*, **1039**, 141 (2004)
- [6] G. Iwinski and D.R. Jenke, *J. Chromatogr.*, **392**, 397 (1987)
- [7] K. Tanaka and J.S. Fritz, *J. Chromatogr.*, **409**, 271 (1987)
- [8] K. Tanaka, K. Ohta, J.S. Fritz, Y.-S. Lee, and S-B. Shim, *J. Chromatogr. A*, **706**, 385 (1995)
- [9] B.K. Głód and W. Kamala, *J. Chromatogr.*, **366**, 39 (1986)
- [10] K. Tanaka, M.-Y. Ding, M.I.H. Helaleh, H. Taoda, H. Takahashi, W. Hu, K. Hasebe, P.R. Haddad, J.S. Fritz, and C. Sarzanini, *J. Chromatogr. A*, **956**, 209 (2002)
- [11] K. Tanaka, M.-Y. Ding, H. Takahashi, M.I.H. Helaleh, H. Taoda, W. Hu, K. Hasebe, P.R. Haddad, M. Mori, J.S. Fritz, and C. Sarzanini, *Anal. Chim. Acta*, **474**, 31 (2002)
- [12] M.I.H. Helaleh, K. Tanaka, M. Mori, Q. Xu, H. Taoda, M.-Y. Ding, W. Hu, K. Hasebe, and P.R. Haddad, *J. Chromatogr. A*, **997**, 133 (2003)
- [13] M.I.H. Helaleh, K. Tanaka, M. Mori, Q. Xu, H. Taoda, M.-Y. Ding, W. Hu, K. Hasebe, and P.R. Haddad, *J. Chromatogr. A*, **997**, 139 (2003)
- [14] M. Mori, M.I.H. Helaleh, Q. Xu, W. Hu, M. Ikedo, H. Taoda, and K. Tanaka, *J. Chromatogr. A*, **1039**, 129 (2004)
- [15] K.L. Ng, B. Paull, P.R. Haddad, and K. Tanaka, *J. Chromatogr. A*, **850**, 17 (1999)
- [16] M. Suzuki, *Adsorption Engineering*, Elsevier, Amsterdam, 1990

- [17] G. Guiochon, S.G. Shirazi, and A. Katti, *Fundamentals of Preparative and Nonlinear Chromatography*, Academic Press, Boston, MA, 1994
- [18] K. Kaczmarski and D. Antos, *J. Chromatogr. A*, **756**, 73 (1996)
- [19] A.J. Berninger, R.D. Whitley, X. Zhang, and N.-H.L. Wang, *Comput. Chem. Eng.*, **15**, 749 (1991)
- [20] K. Kaczmarski, G. Storti, M. Mazzotti, and M. Morbidelli, *Comput. Chem. Eng.*, **21**, 641 (1997)
- [21] K. Tanaka and P.R. Haddad, *Encyclopedia of Separation Science, III Ion Exclusion Chromatography*, Academic Press, London, 2000, pp. 3193–3201
- [22] G. Kortün, W. Vogel, and K. Andrussov (Ed.) *Dissociation Constants of Organic Acids in Aqueous Solution*, Butterworths, London, 1961