

Modeling of the influence of the experimental conditions on the separation of the components of a binary mixture in isocratic overloaded elution preparative chromatography

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Abstract

The elution band profiles of the components of a binary mixture were calculated for systematic variations of the experimental conditions. Three values of the separation factor were used, 1.8, 1.3 and 1.1. The column efficiency was 3000 theoretical plates, except for $\alpha=1.1$, in which case efficiencies of both 3000 and 5000 plates were used. The relative compositions of 3:1, 1:1, and 1:3 were selected. The values of the loading factor were chosen to illustrate the increasing degrees of interference and band interaction which take place with increasing column loading. In each case, the cutting volumes for the production of each component at a 98% purity were calculated and tabulated. The influence of the experimental conditions on the recovery yield is discussed.

Keywords: Preparative chromatography; Optimization; Elution profiles; Computer simulation; Thermodynamic parameters

1. Introduction

Fast computers and the increasing pressure of environmental regulations are progressively changing the way experimental research is conducted in chemistry. Fast computers provide the possibility to use sophisticated models accounting for all the main features of a detailed representation of the way in which scientists or engineers figure out the interactions between the experimental conditions and the observed results in the phenomena they studied. The programming of complex models and the run of these programs to calculate the results of hundreds of experiments can be done rapidly and inexpensively.

Accordingly, it is no longer necessary to simplify excessively the models used in order to permit the calculation of the numerical solutions. In many fields of engineering, computer experiments which differ minimally from the conditions of actual practice can be carried out and their results used confidently for learning, training, or for optimization procedures.

These experiments are all the easier to perform and instructive because it is possible to adjust separately the value of each parameter as desired, while it would be nearly impossible to find the chemicals having the required combination of properties. From these results, the scientist can select the few experiments whose results will provide the most informative test of the validity of the model used or will validate the conclusions of the study. Thus, only these few selected experiments have to be run. This allows a considerable reduction of the amount of

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chemicals needed to acquire the desired information, chemicals which, in almost all cases, are eventually sent to waste.

The procedure improves considerably the knowledge we can accumulate regarding complex natural phenomena within a limited period of time and limited means. The best experiments can be selected, the number of experiments needed is markedly decreased, and the amount of wastes generated is greatly reduced. Physics and engineering are farther advanced on this evolutionary trend than chemists. It should always be remembered, however, that whatever a computer prints, draws or says has been willed by the human who programmed it. The validity of the program has to be demonstrated by the agreement between calculations and experimental results. In the case in point the proofs are now abundant. They have been discussed recently [1].

We present here an example of application of computer engineering by investigating the changes of the elution band profiles and of the band interference caused by modifications of the amount of sample, of the relative composition of the feed, and of the relative retention or separation factor of the feed components in preparative chromatography. The interest of this application stems from the considerable difficulties experienced in optimizing the experimental conditions under which a separation or purification is carried out. Because of the intricacies of the dependence of the production rate on these parameters, empirical procedures of optimization are tedious, long, costly, and cause the waste of important amounts of solvents and valuable chemicals without any guarantee of convergence towards the true optimum. By contrast, the use of a proper modeling of the separation process, of physico-chemical parameters which can be measured easily on small, analytical size columns, and of computer calculations permits the rapid derivation of approximate values of the optimum parameters. The model allows also the calculation of separations taking place under a given set of experimental conditions. Chemists can train with such programs as they would do on a real process, change systematically the values of the parameters, do it one by one as it would be impossible in actual experiments, and perform separations under conditions too costly or dangerous for actual training. A good understanding

of the behavior of separation units can be acquired rapidly, at low cost.

Band profiles of single- and multi-component samples have been studied experimentally and theoretically for over fifty years. The publications of Bohart and Adams [2], Wicke [3,4], Martin and Synge [5], Glueckauf [6,7], Lapidus and Amundson [8], Van Deemter et al. [9], Rhee et al. [10,11], and Helfferich and Klein [12] have marked the progressive development of our understanding of chromatography, non-linear or linear. A review of this theoretical work and of the experimental results which validate it is available [1]. The goal of this paper is to present the results of systematic calculations performed in order to illustrate the effects of systematic variations of the main experimental parameters on the chromatograms of binary mixtures.

2. Theory

2.1. Model of non-linear chromatography

The elution band profiles of the two components of a binary mixture are obtained as solutions of the system of the two mass balance equations of chromatography for these compounds

$$\frac{\partial C_i}{\partial t} + F \frac{\partial q_i}{\partial t} + u \frac{\partial C_i}{\partial z} = D_a \frac{\partial^2 C_i}{\partial z^2} \quad i = 1, 2 \quad (1)$$

where C_i and q_i are the mobile and stationary phase concentration of component i at equilibrium, at time t and position z , respectively, F is the phase ratio, related to the total column porosity, ϵ_T , by

$$F = \frac{1 - \epsilon_T}{\epsilon_T} \quad (2)$$

u is the mobile phase velocity, and D_a is the apparent axial dispersion coefficient (supposed to be the same for both components).

Several models of non-linear chromatography are available to solve the system of Eq. 1 [1]. They differ by the nature of the relationships between the component concentrations in the mobile and stationary phases which are used for that purpose. The model used in this work is the equilibrium-dispersive model [1], the simplest model providing for a finite

column efficiency. It assumes that there is constant equilibrium between the stationary and the mobile phase and that the mass transfer resistance can be accounted for by using an appropriate value for the apparent axial dispersion coefficient. The validity of this assumption has been demonstrated in all cases in which the column efficiency was moderate or high. In this model, the relationship between the concentrations in the mobile and stationary phases are provided by the competitive adsorption isotherms of the two components.

The simplest multicomponent isotherm model is the Langmuir isotherm

$$q_i = \frac{a_i C_i}{1 + \sum_{j=1}^2 b_j C_j} \quad j=1, 2 \quad (3)$$

where a_i and b_i are numerical coefficients characterizing each component. The ratio a_i/b_i is the saturation capacity, q_s , of the stationary phase, or stationary phase concentration in equilibrium with a mobile phase having an infinitely large concentration. It must be the same for the two components¹ [1,13]. Otherwise, the isotherm model is not thermodynamically consistent and, in practice, does not account well for the competitive adsorption behavior of two components which, independently, follow the single-component Langmuir isotherm derived from Eq. 3. [1]. More complex isotherm models are available if needed [1]. While the band profiles depend obviously on the isotherm model used, however, the qualitative conclusions derived from calculations carried out with different isotherm models remain the same. This justifies the use of the Langmuir model in this work.

The ratio of the coefficients a_i and a_j for the two components is their separation factor, $\alpha_{i,j}$ (i.e., $\alpha_{i,j} = a_i/a_j$). In binary mixtures, $\alpha_{i,j}$ is replaced by α . Effective separation of the two components requires that the separation factor be larger than unity. The difficulty of the separation increases approximately

in proportion to $(\alpha-1)^2$ [14]. Thus, the separation with $\alpha=1.1$ is considerably more difficult than the one with $\alpha=1.8$.

Finally, the amount of feed injected into the column is characterized by the loading factor L_f , defined as the ratio of the amount of a component injected to the column saturation capacity

$$L_f = \frac{M_{inj}}{(1 - \epsilon_T)V_c q_s} \quad (4)$$

where M_{inj} is the total amount of the sample (mg) injected, V_c is the column geometrical volume (ml) and q_s is the saturation capacity.

The initial condition for the solution of the model is the column containing the mobile phase in equilibrium with the stationary phase but none of the components. The boundary condition is the injection of a rectangular pulse of feed, at the same flow-rate as the mobile phase. The pulse width is t_p , its volume is $t_p u V_c/L$ (L , column length) and the concentration of the two components in the feed are C_1^0 and C_2^0 . We consider below three typical mixtures, with values of C_1^0/C_2^0 equal to 1/3, 1/1, and 3/1, respectively.

2.2. Numerical solution of the model

The model described above has no analytical, closed-form solutions. However, numerical solutions are easily calculated. We used for this purpose a finite difference algorithm. The forward-backward scheme selected has the advantage of being very fast [1]. It is written to calculate numerical solutions of Eq. 1 with $D_a=0$. The proper amount of axial dispersion is obtained by adjusting the numerical diffusion accordingly. This is achieved by setting equal to $H=L/N$ (H , column HETP, N , plate number; note that $H=2 D_a/u$), the differential element of length in the integration process, while the differential increment of time is set equal to $2H(1+k'_0)/u$. This selection ensures that the Courant number of the problem is equal to 2 and the calculation process is stable [1]. Implicit in this selection is the assumption that the mass transfer kinetics, characterized by the number of theoretical plates of the column, is independent of the concentration.

All calculations were carried out assuming a 25-

¹This is true provided that the saturation capacities are expressed in moles, not in weight units. However, in chromatography, concentrations are more often given in mass of solute per unit volume of solution, a more practical unit. In this work, concentrations are given in mg/ml.

cm long column with an inner diameter 0.46 cm. The flow-rate was 1.0 ml/min and the phase ratio 0.25. The total feed volume injected was 1.0 ml. The isotherm parameters for the first component were chosen as $a_1=12.0$ and $b_1=0.024$ ml/mg, corresponding to a saturation capacity q_s of 500 mg/ml. In most cases, a column having a fair efficiency, $N=3000$ theoretical plates, was used. When the separation was difficult ($\alpha=1.1$), a more efficient column ($N=5000$ theoretical plates) was chosen.

3. Results and discussion

The results of our calculations are presented in Figs. 1–8 and discussed below. In all the figures, the concentrations have been divided by the loading factor to normalize them and permit an easy comparison of the profiles obtained for widely different feed sizes. Note that, with this scale, the maximum height of the band decreases when the band broadens. It is clear on all figures that when the loading factor is increased, the maximum height of the first band decreases first (because the band broadens) and then increases when band interference takes place (because of the displacement effect). The maximum height of the second band decreases constantly with increasing loading factor because the tag-along effect² [1] contributes to enhance band broadening.

3.1. Influence of the loading factor on the chromatogram obtained for a given mixture

In each series of these calculations whose results are presented in Figs. 1–4, the column efficiency, N , the mixture composition, and the separation factor, α , are kept constant. The calculations were carried out successively for three different values of the separation factor (1.8 in Fig. 1; 1.3 in Fig. 2; 1.1 in Fig. 3 and Fig. 4) and for three different values of the relative composition of the feed $C_1^0/C_2^0=3:1$ in Fig. 10 and Fig. 2a, 3a and 4a; 1:1 in Fig. 1b, 2b, 3b

and 4b; and 1:3 in Fig. 1c, 2c, 3c and 4c). C_i^0 is the feed concentration of component i .

3.1.1. Separation factor $\alpha=1.8$

Because of the large values of the separation factor, the separation is easy to achieve. Under analytical conditions (i.e., when the product bC , or the sum $b_1C_1+b_2C_2$, in Eq. 3 is negligible compared to unity), a column efficiency of 3000 plates would afford a 5.3 resolution between the two bands. The calculations were made for four rather large values of the loading factors: $L_f=50, 20, 10$ and 5%. The chromatograms obtained are shown in Fig. 1. There is no significant band interference for the lowest value of L_f and the bands are still well resolved for the two intermediate values. However, the tag-along effect is clearly illustrated not only by the chromatogram at a loading factor of 50% but also by those at loading factors of 20 and 10% in Fig. 1a and, to a lesser degree, 1b. In these last two cases, the flat top of the second band results from the interference between the two bands in the first part of their migration along the column. If we compare Fig. 1a, b, and c, we see a decreasing intensity of the tag-along effect with increasing relative concentration of the second component and a parallel increase of the intensity of the displacement effect.

The profile of the top of the second band exhibits an incomplete recovery from a tag-along type of interaction with the first one in all four chromatograms in Fig. 1a which exhibit a flat (last two profiles) or a round top; the recovery is complete in the first chromatogram in Fig. 1b and in the first two in Fig. 1c. Similarly, the width of the rear plateau of the second band of the chromatogram at a loading factor of 50% decreases from Fig. 1a to 1b while in Fig. 1c the plateau has disappeared to be replaced by an inflection point. By contrast, the height of the first component band decreases only slightly from Fig. 1a to 1c, while the amount injected decreases by a factor 3. The band width does not decrease in the same proportion either. However, it adopts the L-shape characteristic of a strong displacement effect [1]. The shock layer of negative amplitude on the rear of the first band at a loading factor of 50% is very slight in Fig. 1a, becomes significant in Fig. 1b and of major importance in Fig. 1c.

The position of the cut-points and the recovery

²This effect is due to the excess of molecules of the less retained compound crowding out of the solution the molecules of the more retained component and reducing their retention (see Eq. 3 for q_2 , with a large value of C_1 and a small value of C_2).

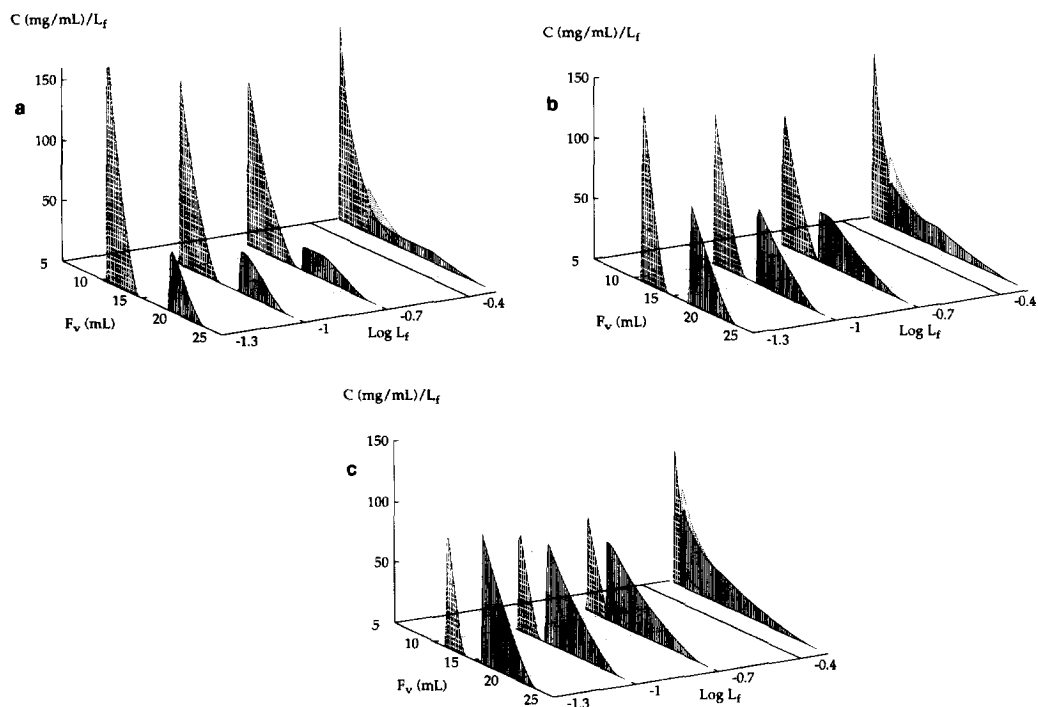


Fig. 1. Calculated band profiles for a binary mixture. Column efficiency, $N=3000$, separation factor, $\alpha=1.8$, concentration ratio, C_1^0/C_2^0 , 3:1 (a), 1:1 (b), and 1:3 (c). Isotherm coefficients: $a_1=12.0$, $b_1=0.024$ ml/mg; $a_2=21.6$, $b_2=0.0432$ ml/mg. X-axis: flow volume (ml), product of retention time and flow-rate. Y-axis: logarithm of the sample loading factor. Z-axis: mobile phase concentration normalized by sample loading factor. Dashed-line: first component. Solid-line: second component. Dotted-line: sum of the 1st and 2nd components.

yield were calculated for the production of 98% pure fractions. The results are listed in Table 1. Only when the column is severely overloaded ($L_f=50\%$), is the separation incomplete. The recovery yield achieved in these cases is around 2/3. When L_f is decreased to 20%, the separation becomes complete.

3.1.2. Separation factor $\alpha=1.3$

This separation is more difficult than the previous one but typical of many encountered in preparative chromatography. Still, under analytical conditions, a resolution of 2.6 between the two bands would be obtained. However, the difficulty of the separation prevents the use of values of the loading factor as high as in the previous case. Since the resolution between the two bands was very poor for a 20% loading factor, we show in Fig. 2a–c the chromatograms calculated for the following values of the loading factor: 10, 5, 2, 1 and 0.5%. In each case, the

calculations were made for the same relative composition of the binary mixture as in Fig. 1. The values of the cut-point and the recovery yield calculated for the collection of 98% pure fractions of each component are listed in Table 2.

Because the values of the loading factor are lower than in Fig. 1, the intensity of the non-linear effects is lower and the band profiles appear more diffuse than in these previous chromatograms. Note that for the chromatograms calculated at a loading factor of 0.5%, the profile of the band of the less concentrated component appears close to Gaussian. The profiles of the other bands and of the two bands of the chromatogram at a loading factor of 1% appear as slightly overloaded. Nevertheless, the characteristic features of column overload and band interferences can be noticed in the chromatograms corresponding to the three highest values of the loading factor in the three figures. Their intensity is high at a loading factor of 10% for which case the band resolution is

Table 1

Cutting volumes and recovery yields calculated for a 98% purity of each component; column efficiency $N=3000$ and separation factor $\alpha=1.8$

C_1^0/C_2^0	L_f (%)	Cutting volume ^a No. 1 (ml)	Recovery yield No. 1 (%)	Cutting volume ^a No. 2 (ml)	Recovery yield No. 2 (%)
3:1	50	9.44	91.04	11.97	63.16
	20	14.87	100.00	12.74	100.00
	10	17.19	100.00	13.50	100.00
	5	18.65	100.00	13.97	100.00
1:1	50	7.65	83.38	10.21	62.37
	20	12.68	100.00	11.44	100.00
	10	15.27	100.00	12.72	100.00
	5	17.17	100.00	13.49	100.00
1:3	50	6.61	77.20	8.34	69.43
	20	11.08	99.93	10.32	100.00
	10	13.88	100.00	12.04	100.00
	5	16.08	100.00	13.08	100.00

^aAll cutting volumes and recovery yields for a 98% purity.

poor. The resolution is excellent at a loading factor of 5% and exceeds 'touching band' conditions at a loading factor of 2% (see also Table 2).

3.1.3. Separation factor $\alpha=1.1$, $N=3000$ plates

This separation is difficult. With this value of the separation factor, a resolution of only 1.0 can be

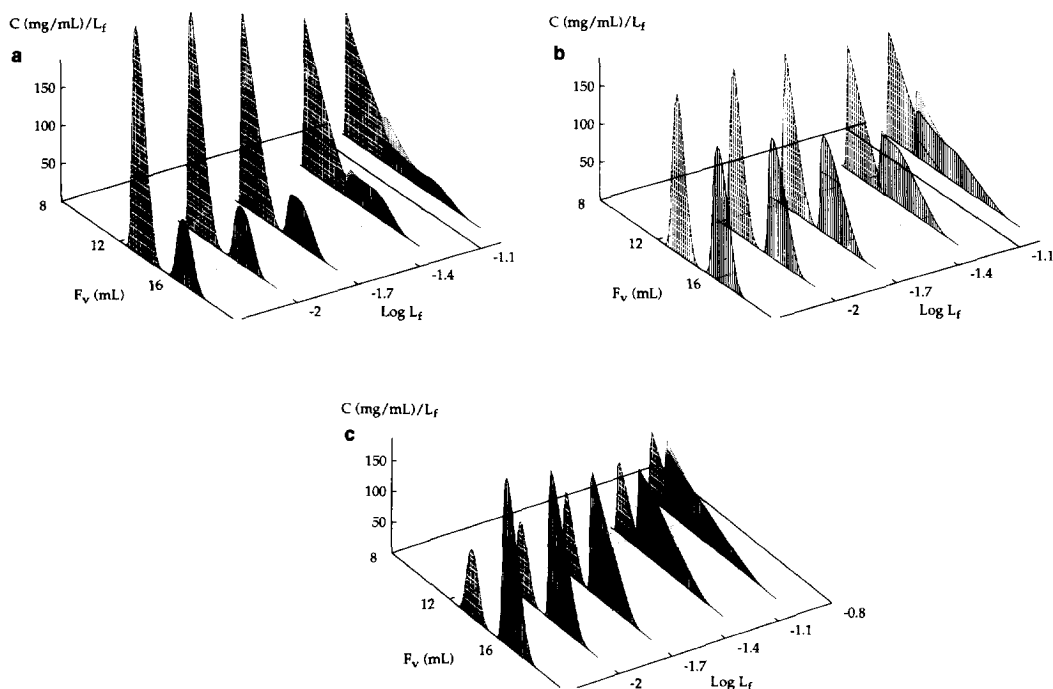


Fig. 2. Calculated band profiles for a binary mixture. Same conditions as in Fig. 1, except separation factor, $\alpha=1.3$, $a_2=15.6$, $b_2=0.0312$ ml/mg.

Table 2
Cutting volumes and recovery yields calculated for a 98% purity of each component; column efficiency $N=3000$ and separation factor $\alpha=1.3$

C_1^0/C_2^0	L_r (%)	Cutting volume ^a No. 1 (ml)	Recovery yield No. 1 (%)	Cutting volume ^a No. 2 (ml)	Recovery yield No. 2 (%)
3:1	10	12.16	91.30	13.54	59.08
	5	13.91	99.66	13.69	97.29
	2	15.22	100.00	14.17	100.00
	1	15.71	100.00	14.39	100.00
	0.5	15.98	100.00	14.52	100.00
1:1	10	11.46	88.51	12.58	67.26
	5	13.25	99.26	12.98	100.00
	2	14.76	100.00	13.80	100.00
	11	15.39	100.00	14.13	100.00
	0.5	15.73	100.00	14.33	100.00
1:3	10	10.91	86.63	11.34	86.37
	5	12.71	98.95	12.39	100.00
	2	14.32	100.00	13.48	100.00
	1	15.10	100.00	13.91	100.00
	0.5	15.55	100.00	14.12	100.00

obtained under analytical conditions. Although it has been shown that high production rates can be obtained under such conditions, their use is possible only as far as a recovery yield for each pass of between 0.5 and 0.7 is acceptable [1]. Recycling of the intermediate, mixed zone becomes necessary and should be integrated in the process [15,16]. For illustrative purposes, the values of the loading factor used for the calculations were kept relatively low, in order to achieve some visible degree of separation. The values selected were 5, 2, 1, 0.5, 0.2 and 0.1%. In each case, the same three values of the relative composition of the feed were used as in the previous cases. The chromatograms calculated are shown in Fig. 3a, b, and c. The cutting volumes³ and recovery yields calculated for the collection of 98% pure fractions of each component are listed in Table 3.

As noted earlier, the chromatograms obtained at low loading factors have Gaussian or nearly Gaussian band profiles. Accordingly, band interference appears to be similar to what is encountered in analytical chromatography at loading factors below 0.1%. However, tag-along effects are clearly notice-

able in Fig. 3a and displacement effects in Fig. 3c at loading factors of 1% and above. The recovery yield (Table 3) is always well below 100%. Values of 85 to 87% can be achieved with a loading factor of 0.2% for the major component, whether eluted first or second. At high loading factors, the recovery yield is much higher for the first component than for the second, whatever the relative composition.

3.1.4. Separation factor $\alpha=1.1$, $N=5000$ plates

The same calculations were performed assuming a column with a higher efficiency, 5000 plates, instead of 3000, thus providing a resolution of 1.3 between the analytical bands. Although it would be easy to perform calculations with any arbitrarily high value of the column efficiency, we selected a realistic value which is not too difficult to achieve in practice. The results are shown in Fig. 4 for the same mixture compositions as before. The position of the cut-points and the value of the recovery yield for the collection of 98% pure fractions of each individual component are listed in Table 4.

Compared to those shown in Fig. 3, the separations exhibited in Fig. 4 are markedly improved. For example, the bands are higher in all chromatograms; non-linear effects, including tag-along (Fig. 4a) and displacement (Fig. 4c) effects become easy to recognize in the chromatogram calculated at a

³Volumes of eluent at which the collection of fractions is stopped (first component) or started (second component) to prepare these components at a required degree of purity (here 98%).

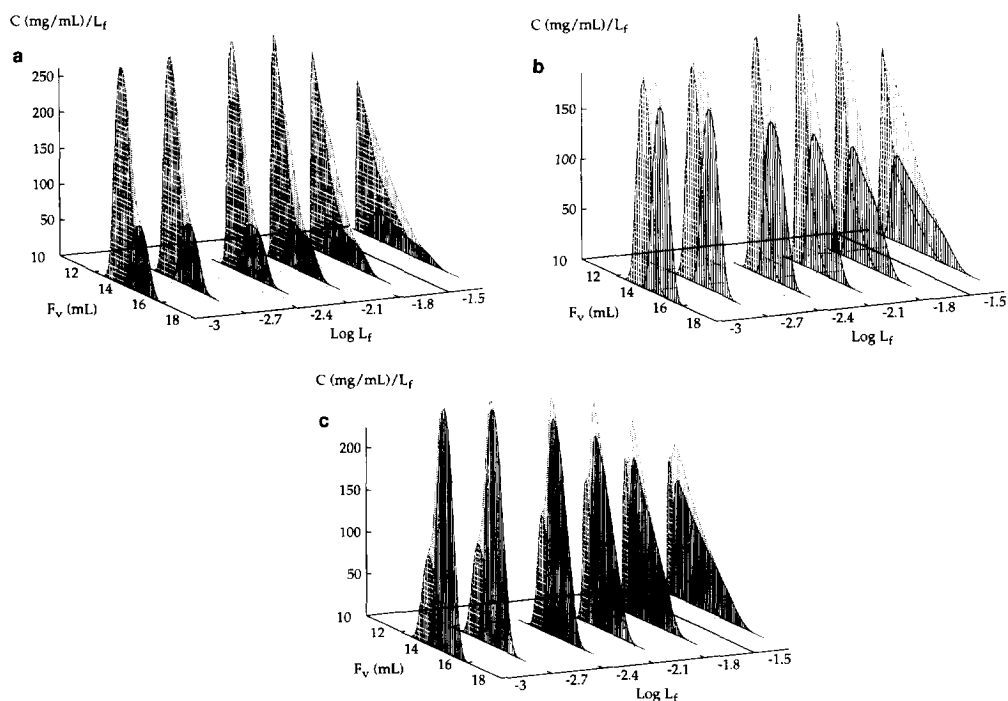


Fig. 3. Calculated band profiles for a binary mixture. Same conditions as in Fig. 1, except separation factor, $\alpha = 1.1$, $a_2 = 13.2$, $b_2 = 0.0264$ ml/mg.

Table 3

Cutting volumes and recovery yields calculated for a 98% purity of each component; column efficiency $N = 3000$ and separation factor $\alpha = 1.1$

C_1^0/C_2^0	L_f (%)	Cutting volume No. 1 (ml)	Recovery yield No. 1 (%)	Cutting volume No. 2 (ml)	Recovery yield No. 2 (%)
3:1	5	11.32	47.74	14.39	11.08
	2	12.73	65.27	14.44	22.67
	1	13.39	75.00	14.50	33.73
	0.5	13.79	81.26	14.57	44.16
	0.2	14.06	84.95	14.64	52.59
	0.1	14.15	86.03	14.68	55.44
1:1	5	11.10	39.69	13.96	14.98
	2	12.54	57.30	14.05	30.87
	1	13.22	66.76	14.16	46.53
	0.5	13.64	71.95	14.30	59.08
	0.2	13.89	72.98	14.44	67.68
	0.1	13.97	73.04	14.49	70.56
1:3	5	10.96	27.86	13.34	23.23
	2	12.43	48.68	13.50	47.58
	1	13.15	59.31	13.70	69.74
	0.5	13.56	64.04	13.96	82.14
	0.2	13.78	61.83	14.19	86.18
	0.1	13.84	60.76	14.27	86.89

Table 4
Cutting volumes and recovery yields calculated for a 98% purity of each component; column efficiency $N=5000$ and separation factor $\alpha=1.1$

C_1^0/C_2^0	L_f (%)	Cutting volume No. 1 (ml)	Recovery yield No. 1 (%)	Cutting volume No. 2 (ml)	Recovery yield No. 2 (%)
3:1	5	11.38	51.82	14.22	13.60
	2	12.78	69.54	14.28	27.68
	1	13.44	79.72	14.34	41.41
	0.5	13.84	86.13	14.41	54.05
	0.2	14.11	90.10	14.50	64.12
	0.1	14.21	91.29	14.54	67.71
1:1	5	11.18	47.10	13.80	17.40
	2	12.61	65.22	13.90	35.88
	1	13.30	75.06	14.02	53.86
	0.5	13.71	80.19	14.17	68.12
	0.2	13.97	82.26	14.32	77.54
	0.1	14.06	82.57	14.39	79.91
1:3	5	11.04	42.29	13.21	25.46
	2	12.50	61.70	13.37	52.46
	1	13.22	72.57	13.60	76.01
	0.5	13.65	76.82	13.87	89.34
	0.2	13.89	75.18	14.11	92.64
	0.1	13.96	73.96	14.21	92.57

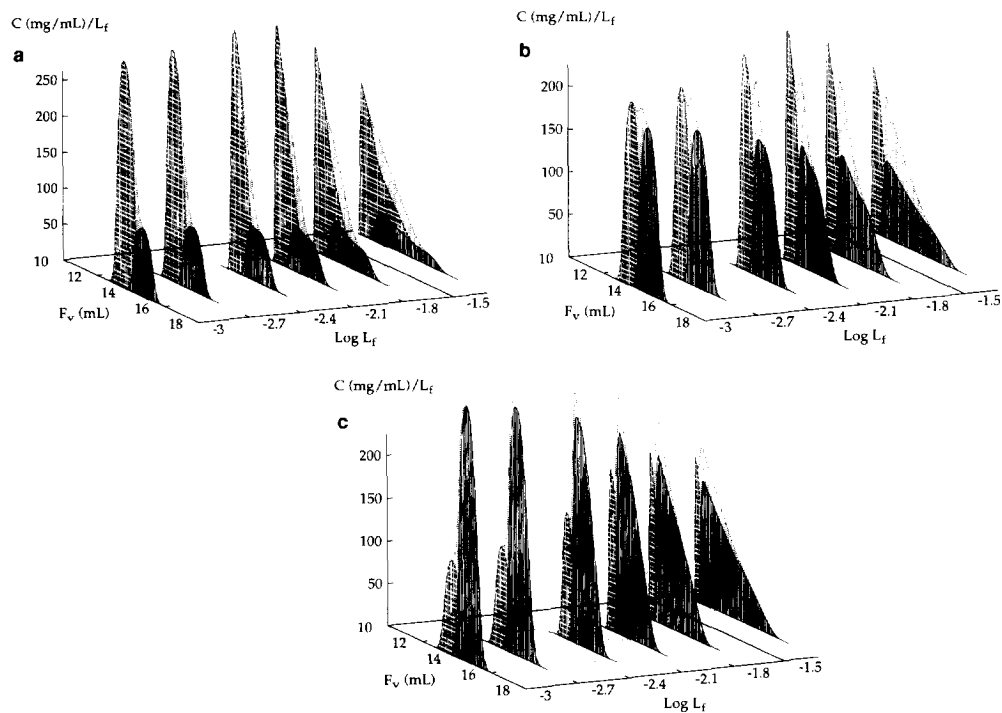


Fig. 4. Calculated band profiles for a binary mixture. Same conditions as in Fig. 3, except column efficiency $N=5000$.

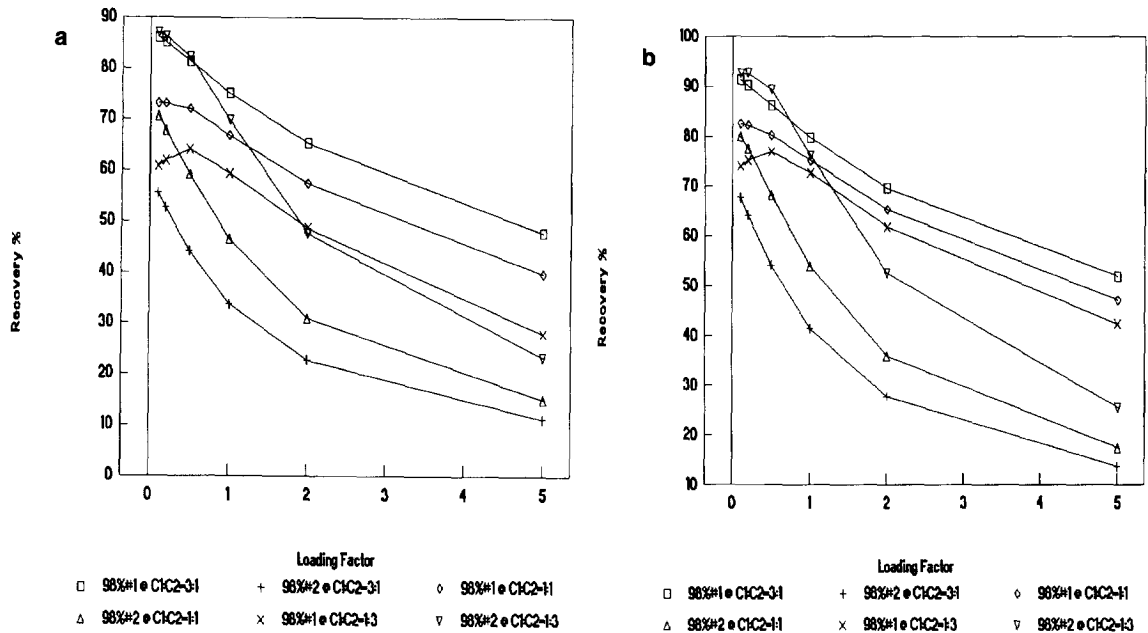


Fig. 5. (a) Plot of the recovery yield of the two components versus the total loading factor. Same experimental conditions as in Fig. 3. (b) Plot of the recovery yield of the two components versus the total loading factor. Same experimental conditions as in Fig. 4.

loading factor of 0.5%; their influence is much sharper for the chromatograms calculated at higher loading factors; and the values of the recovery yield are closer to unity, as shown by a comparison of Table 3 and Table 4. Thus, a comparison of Fig. 3a, b, and c with Fig. 4a, b, and c, respectively, illustrates the advantage associated with the use of

columns having a higher efficiency, provided that this improvement is not achieved by using a longer column, but by having a better packed column or one made with smaller particles.

Plots of the recovery yields versus the loading factor are given in Fig. 5a ($N=3000$ plates) and b ($N=5000$ plates). They illustrate the improvement in

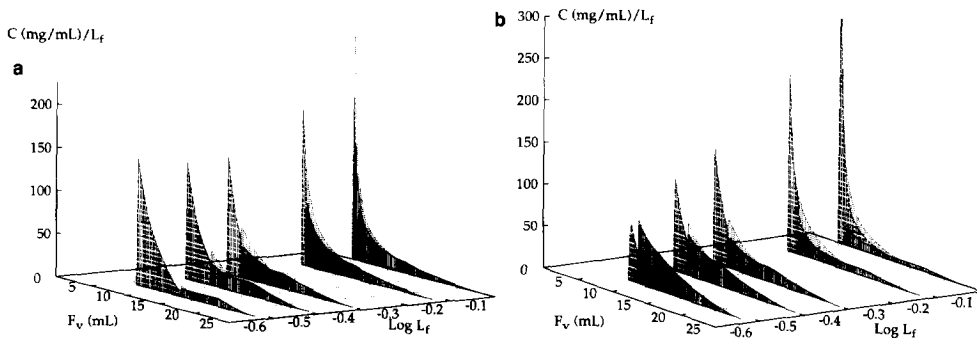


Fig. 6. (a) Calculated band profiles for a binary mixture. Same conditions as in Fig. 1 except first component concentration, $C_1^0=104$ mg/ml and first to second component concentration ratio, C_1^0/C_2^0 ; profile No. 1, 1:3 ($L_f=100\%$); No. 2, 1:2 ($L_f=75\%$); No. 3, 1:1 ($L_f=50\%$); No. 4, 1:0.6 ($L_f=40\%$); and No. 5, 1:0.2 ($L_f=30\%$). (b) Calculated band profiles for a binary mixture. Same conditions as in (a) except second component concentration $C_2^0=104$ mg/ml and first to second component concentration ratio, C_1^0/C_2^0 ; profile No. 1, 3:1 ($L_f=100\%$); No. 2, 2:1 ($L_f=75\%$); No. 3, 1:1 ($L_f=50\%$); No. 4, 0.6:1 ($L_f=40\%$); and No. 5, 0.2:1 ($L_f=30\%$).

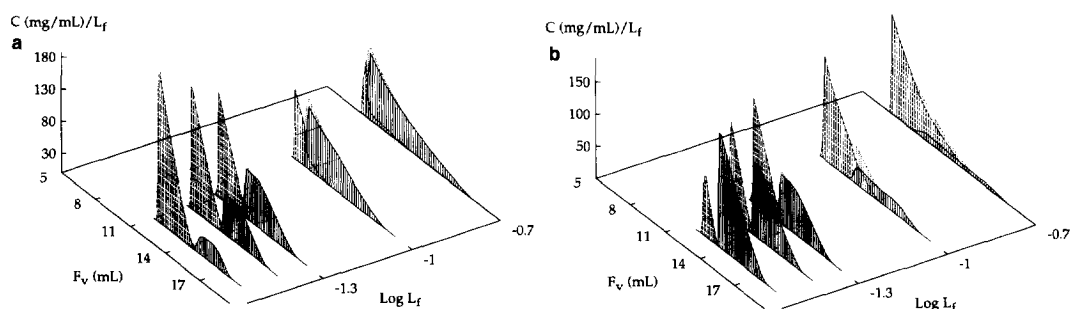


Fig. 7. (a) Calculated band profiles for a binary mixture. Same conditions as in Fig. 6a except separation factor $\alpha=1.3$, first component concentration $C_1^0=10.4$ mg/ml, and first to second component concentration ratio, C_1^0/C_2^0 : profile No. 1, 1:7 ($L_r=20\%$); No. 2, 1:3 ($L_r=10\%$); No. 3, 1:1 ($L_r=5\%$); No. 4, 1:0.6 ($L_r=4\%$); and No. 5, 1:0.2 ($L_r=3\%$). (b) Calculated band profiles for a binary mixture. Same conditions as in Fig. 6b except separation factor, $\alpha=1.3$, second component concentration, $C_2^0=10.4$ mg/mL, and first to second component concentration ratio, C_1^0/C_2^0 : profile No. 1, 7:1 ($L_r=20\%$); No. 2, 3:1 ($L_r=10\%$); No. 3, 1:1 ($L_r=5\%$); No. 4, 0.6:1 ($L_r=4\%$); and No. 5, 0.2:1 ($L_r=3\%$).

recovery yield achieved by reversing the elution order, using moderate values of the loading factor and a high column efficiency.

3.2. Influence of the relative composition of the feed

In the previous section, we studied the influence of the loading factor at constant feed composition. In this section, we keep constant the amount injected of one of the two components and increase the loading factor by increasing only the amount of the other component in the feed. In the corresponding figures,

the area of the band of the compound whose injected amount is kept constant is inversely proportional to the loading factor. The profile of this band changes, reflecting the changing interaction with the other component band. The study is done with the same values of the separation factor and the column efficiency as those used above.

3.2.1. Separation factor $\alpha=1.8$

Fig. 6a shows the band profiles obtained when the loading factor of the first component is held constant at 25% and the loading factor for the second component is increased from 5% to 75% (thus, the

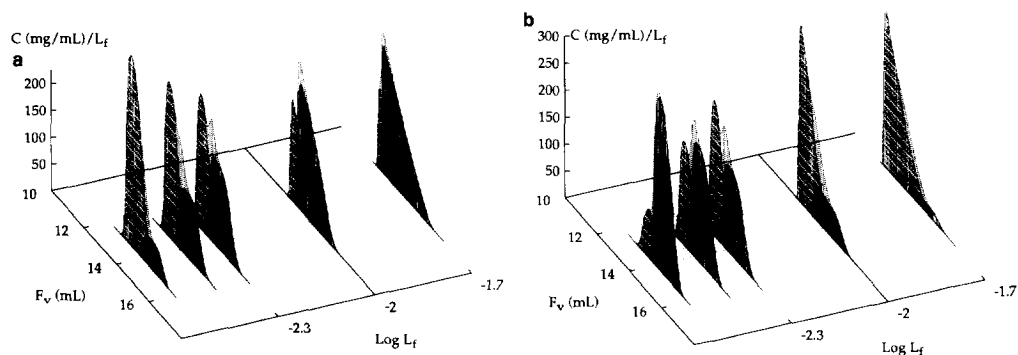


Fig. 8. Calculated band profiles for a binary mixture. Same conditions as in Fig. 6a except column efficiency $N=5000$, separation factor $\alpha=1.1$, first component concentration $C_1^0=1.04$ mg/ml, and first to second component concentration ratio, C_1^0/C_2^0 ratio: profile No. 1, 1:7 ($L_r=2\%$); No. 2, 1:3 ($L_r=1\%$); No. 3, 1:1 ($L_r=0.5\%$); No. 4, 1:0.6 ($L_r=0.4\%$); and No. 5, 1:0.2 ($L_r=0.3\%$). (b) Calculated band profiles for a binary mixture. Same conditions as in (a) except second component concentration, $C_2^0=1.04$ mg/ml and first to second component concentration ratio, C_1^0/C_2^0 ratio: profile No. 1, 7:1 ($L_r=2\%$); No. 2, 3:1 ($L_r=1\%$); No. 3, 1:1 ($L_r=0.5\%$); No. 4, 0.6:1 ($L_r=0.4\%$) and No. 5, 0.2:1 ($L_r=0.3\%$).

total loading factor increases from 30 to 100%). The height of the first component band increases (it more than doubles although its area decreases three fold) while its width and its retention time decrease. These changes illustrate the progressive evolution from a situation in which the tag-along effect is dominant (e.g., the two chromatograms at low loading factors of 30% and 40% in Fig. 6a) to one in which the displacement effect controls the band profiles (the two chromatograms at high loading factors, 75 and 100%). In the last case, the fronts of the two bands nearly coelute and there is hardly any separation (see Table 5, the recovery yields are very low in this case).

Fig. 6b illustrates the converse case in which the amount of the second component injected is kept constant ($L_{r,2}=25\%$) while that of the first component is increased from a small value ($L_{r,1}=5\%$), in which case the band of the second component in large excess displaces the one of the first component to a large value ($L_{r,1}=75\%$), in which case the second component band tags along the band of the first component. As a consequence, the height of the band decreases and its width increases when the loading factor of the first component is increased, an effect which is in the opposite direction to the influence of an increase in the amount of second component in the feed, as illustrated in Fig. 6a. The recovery yields are generally, but not always, better than in the converse case.

3.2.2. Separation factor $\alpha=1.3$

The results obtained in this case, illustrated in Fig. 7a and b, are similar to those obtained in the previous case and in the next one. The value of the constant loading factor is 2.5%. The loading factor of the other component increases from 0.5 to 17.5%, thus providing a total loading factor increasing from 3 to 20% and a relative concentration decreasing from 83% to 16.7%. At low concentrations of the second component, the band of the first one is eluted as if this compound were pure and there is no band interference. When the concentration of the second component increases, the profile of the first band remains the same at first, the only effect on the profiles being the tag-along effect of the second band and a shorter retention time of the first band front. Displacement becomes significant when the concentration of the second component becomes significant compared to that of the first, i.e., with $L_{r,2}=7.5\%$ (fourth profile). Eventually, the displacement of the first component becomes the overwhelming effect, as illustrated in the rightmost chromatogram in Fig. 7a.

Increasing the concentration of the first component (Fig. 7b) causes the rapid increase of the tag-along effect illustrated first by the flat top of the band e.g., second and third profiles ($L_{r,1}=1.5$ and 2.5%, respectively), then by the long intermediate plateau. Total recovery is achieved for values of the total loading factors below 5% (Table 6).

Table 5

Cutting volumes and recovery yields calculated for a 98% purity of each component; column efficiency $N=3000$, separation factor $\alpha=1.8$, and C_1^0 or $C_2^0=104$ mg/ml

	C_1^0/C_2^0	L_r (%)	Cutting volume No. 1 (ml)	Recovery yield No. 1 (%)	Cutting volume No. 2 (ml)	Recovery yield No. 2 (%)
$C_1^0=104$ mg/ml	1:3	100	4.35	19.72	8.28	35.34
	1:2	75	5.22	60.55	9.00	44.04
or	1:1	50	7.65	83.38	10.21	62.37
$L_{r,1}=25\%$	1:0.6	40	9.73	93.64	11.08	76.98
	1:0.2	30	13.93	100.00	12.74	99.81
$C_2^0=104$ mg/ml	3:1	100	5.36	61.99	11.91	32.54
	2:1	75	6.36	72.30	11.30	42.49
or	1:1	50	7.65	83.38	10.21	62.37
$L_{r,2}=25\%$	0.6:1	40	8.20	88.41	9.39	79.18
	0.2:1	30	8.70	93.73	8.51	100.00

Table 6

Cutting volumes and recovery yields calculated for a 98% purity of each component; column efficiency $N=3000$, separation factor $\alpha=1.3$, and C_1^0 or $C_2^0=10.4$ mg/ml

	C_1^0/C_2^0	L_f (%)	Cutting volume No. 1 (ml)	Recovery yield No. 1 (%)	Cutting volume No. 2 (ml)	Recovery yield No. 2 (%)
$C_1^0=10.4$ mg/ml	1:7	20	8.51	61.81	9.78	68.06
	1:3	10	10.91	86.63	11.34	86.37
	or 1:1	5	13.25	99.26	12.98	100.00
$L_{f,1}=2.5\%$	1:0.6	4	14.00	99.93	13.52	99.99
	1:0.2	3	15.04	100.00	14.18	99.87
$C_2^0=10.4$ mg/ml	7:1	20	10.59	80.42	14.00	30.26
	3:1	10	12.16	91.30	13.54	59.08
	or 1:1	5	13.25	99.26	12.98	100.00
$L_{f,2}=2.5\%$	0.6:1	4	13.45	99.87	12.97	100.00
	0.2:1	3	13.54	99.97	12.96	100.00

3.2.3. Separation factor, $\alpha=1.1$

In this case, only the results obtained with the higher efficiency of 5000 plates are reported in Fig. 8a and b. The value of the constant loading factor is 0.25% while the loading factor of the other component is increased from 0.05 to 1.75%. As in previous cases, the intensity of the displacement effect increases with increasing values of the total loading factor and of the concentration of the second component (Fig. 8a). Similarly, the intensity of the tag-along effect increases with increasing amount of the first component.

With the value selected for the column efficiency,

it is not possible to achieve total separation nor a 100% recovery yield (Table 7). Reasonable values can be obtained at low loading factors.

4. Conclusions

The band profiles, the degree of separation between the bands of two components, the production rate and the recovery yield, all depend on the loading factor and the parameters of the competitive isotherms. In turn, the loading factor, a dimensionless number is a function of both the sample size and the

Table 7

Cutting volumes and recovery yields calculated for a 98% purity of each component; column efficiency $N=5000$, separation factor $\alpha=1.1$, and C_1^0 or $C_2^0=1.04$ mg/ml

	C_1^0/C_2^0	L_f (%)	Cutting volume No. 1 (ml)	Recovery yield No. 1 (%)	Cutting volume No. 2 (ml)	Recovery yield No. 2 (%)
$C_1^0=1.04$ mg/ml	1:7	2	12.44	52.84	12.81	80.04
	1:3	1	13.22	72.57	13.60	76.01
	or 1:1	0.5	13.71	80.19	14.17	68.12
$L_{f,1}=0.25\%$	1:0.6	0.4	13.85	83.72	14.33	64.07
	1:0.2	0.3	14.12	92.81	14.54	55.44
$C_2^0=1.04$ mg/ml	7:1	2	13.04	77.86	14.47	23.75
	3:1	1	13.44	79.72	14.34	41.41
	or 1:1	0.5	13.71	80.19	14.17	68.12
$L_{f,2}=0.25\%$	0.6:1	0.4	13.76	79.01	14.09	80.31
	0.2:1	0.3	13.80	74.00	13.92	97.30

column saturation capacity. Thus, this latter parameter is extremely important when selecting the stationary phase for a preparative separation, although it is often and wrongly ignored. Clearly, a high separation factor is needed but careful attention should be paid to the value of the saturation capacity of the phases available since, everything else being equal, the production rate is proportional to this capacity.

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References

- [1] G. Guiochon, S. Golshan Shirazi and A.M. Katti, *Fundamentals of Preparative and Nonlinear Chromatography*, Academic Press, Boston, MA, 1994.
- [2] G.S. Bohart and E.Q. Adams, *J. Am. Chem. Soc.*, 42 (1920) 523.
- [3] E. Wicke, *Kolloid Z.*, 86 (1939) 295.
- [4] E. Wicke, *Kolloid Z.*, 90 (1940) 156.
- [5] A.J.P. Martin and R.L.M. Synge, *Biochem. J.*, 35 (1941) 1358.
- [6] E. Glueckauf, *Proc. Roy. Soc. (London)*, A186 (1946) 35.
- [7] J.I. Coates and E. Glueckauf, *J. Chem. Soc.*, 1947, 1309.
- [8] L. Lapidus and N.R. Amundson, *J. Phys. Chem.*, 56 (1952) 984.
- [9] J.J. Van Deemter, F.J. Zuiderweg and A. Klinkenberg, *Chem. Eng. Sci.*, 5 (1956) 271.
- [10] H.-K. Rhee, R. Aris and N.R. Amundson, *Trans. Roy. Soc. (London)*, A267 (1970) 419.
- [11] H.-K. Rhee, R. Aris and N.R. Amundson, *First-Order Partial Differential Equations. II Theory and Application of Hyperbolic Systems of Quasilinear Equations*, Prentice Hall, Englewood Cliffs, NJ, 1989.
- [12] F. Helfferich and G. Klein, *Multicomponent Chromatography. A Theory of Interference*, Marcel Dekker, New York, NY, 1970.
- [13] M.D. LeVan and T. Vermeulen, *J. Phys. Chem.*, 85 (1981) 3247.
- [14] S. Golshan-Shirazi and G. Guiochon, *J. Chromatogr.*, 523 (1990) 1.
- [15] A. Seidel-Morgenstern and G. Guiochon, *AIChE J.*, 39 (1993) 809.
- [16] F. Charton, M. Bailly and G. Guiochon, *J. Chromatogr. A*, 687 (1994) 13.