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Optimization of the experimental conditions and the column design parameters in displacement chromatography

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ABSTRACT

Optimization of the experimental conditions (sample size, reduced mobile phase velocity and displacer concentration) and the column design parameters (column length and average particle size) was performed for the maximum production rate of either component of a binary mixture. The displacement chromatograms were calculated by means of the equilibrium-dispersive model of chromatography, assuming competitive Langmuir isotherms. Binary mixtures of separation factor 1.2, 1.5 and 1.8 and relative concentrations 3:1 and 1:3 were studied. For the systems studied, no optimum displacer concentration was found, the maximum production rates being achieved at the highest displacer concentration allowed by its solubility. The optimum column characteristics depend strongly on the separation factor and on the mixture composition. The maximum production rates were achieved at a much lower value of the retention factor than usually chosen in displacement chromatography. The optimum value of the retention factor of the less retained component was found to be between 1.2 and 2.0, depending slightly on the mixture composition and more strongly on the separation factor.

INTRODUCTION

Displacement chromatography was introduced by Tiselius [1] in 1942. Its theory in the case of Langmuir isotherms was developed soon afterwards by Glückauf [2], later by Helfferich and Klein [3] and subsequently by Rhee and Amundson [4], all these workers using the ideal model of chromatography, *i.e.*, assuming infinite column efficiency. After a period of intensive use during the Manhattan project [5], the method fell into oblivion as a separation technique for 30 years, until it was reintroduced by Horváth *et al.* [6].

There is currently strong interest in displacement chromatography [7–9] because it can deliver purified fractions which are much more concentrated than overloaded elution [1–10], although it is more difficult to use. The question of which one of these two modes of chromatography permits the highest production rate remains controversial because they have never been optimized simultaneously for the same separation problem. In a recent study, Katti *et al.* [11] compared on a theoretical basis the performance of displacement and overloaded elution chromatography, using the same column. The maximum production rates of the two modes were similar. Overloaded elution gives a better recovery yield, whereas the concentration of the fraction collected was one to two orders of magnitude higher in the displacement mode.

In this work, we applied to displacement chromatography the multi-parameter optimization method

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previously developed and applied to the study of the optimization of the experimental conditions in overloaded elution chromatography [12]. This method uses the numerical solution of the equilibrium-dispersive model of chromatography developed by Guiochon and co-workers [13–15]. Previously, Katti and Guiochon [16] have calculated band profiles in displacement chromatography by means of the same model and solution and discussed some of their properties.

In displacement chromatography, first the sample is introduced into the column equilibrated with the carrier. Then a solution of the displacer, a compound which is more strongly adsorbed on the stationary phase than any sample component, is pumped continuously into the column. The steep front of the displacer moves at a constant velocity, proportional to the carrier velocity and to the column capacity for the displacer at the concentration used. Being the most strongly retained component, the displacer pushes all the sample components in front of its band.

When a steady state is achieved, all the component bands travel at the same velocity, forming the isotachic train [2–4,6]. These bands are nearly rectangular and, unless the amount of a component is very small, the maximum concentration of the band does not depend on the size injected, but on the displacer concentration and on the adsorption isotherms of the component and the displacer.

THEORY

The band profiles of the two components of the binary mixture and that of the displacer were calculated using the method first developed by Rouchon *et al.* [13] and employed by Katti and Guiochon [16] for the calculation of the band profiles in displacement chromatography. This approach is based on the numerical solution of the equilibrium-dispersive model of chromatography. The mass balance equation and an isotherm model are written for each component of the sample and for the displacer. No mass balance is needed for the inert carrier [17]. The individual band profiles are obtained as numerical solutions of this system of equations, calculated using a finite difference method [18].

When the band profiles have been calculated, an integration routine allows the determination of the cutting times at a given purity for the fractions of either component. The amount produced, the recovery yield and the production rate are then calculated.

Column properties

The following assumptions were made for the characteristics of the chromatographic system in the calculations. The total column porosity was $\varepsilon = 0.8$, the viscosity of the mobile phase was $\eta = 1$ cP and the molecular diffusivity of each compound in the mobile phase was $D_m = 1 \cdot 10^{-5}$ cm^2/s. The column inner diameter was 4.6 mm. A maximum inlet pressure of 125 atm was allowed. The sample was injected as a rectangular plug, with a volume $V_p = 0.5$ ml. The values given for the loading factor (L_f) refer to the total loading of both components, and were calculated as

$$L_f = \frac{V_p}{(1-\varepsilon)\, s\, L}\left(\frac{C_1^0}{q_{1,s}} + \frac{C_2^0}{q_{2,s}}\right) \tag{1}$$

where C_1^0 and C_2^0 are the concentrations of the first and the second component in the injected sample, respectively, $q_{1,s}$ and $q_{2,s}$ are the column saturation capacities of the two components, respectively, s is the column cross-sectional area and L is the column length.

The column efficiency was calculated using the Knox plate-height equation [19]:

$$h = \frac{2}{\nu} + \nu^{1/3} + \frac{\nu}{10} \tag{2}$$

where $h = H/d_p$ is the reduced plate height, H is the actual height equivalent to a theoretical plate, $\nu = ud_p/D_m$ is the reduced mobile phase velocity and u is the actual mobile phase velocity.

In the first part of the calculations, we optimized the operating parameters for an arbitrary column, with a length $L = 25$ cm, packed with particles of average diameter $d_p = 20$ μm. In this instance, only the reduced mobile phase velocity, the sample loading factor and the displacer concentration were optimized. In the second part of the calculations, the column length and the average particle diameter were also optimized. Throughout this work a maximum displacer solubility of 300 mg/ml was assumed.

The required purity of the collected fractions was 99%, allowing 1% of the second component as an impurity in the collected fraction of the first component, and 0.5% of the first component and 0.5% of the displacer as impurities in the collected fraction of the second component.

The same general properties (column porosity, mobile phase viscosity, molecular diffusivity, maximum inlet pressure, column saturation capacities, column cross-sectional area, coefficients of the plate-height equation and required purity) were assumed in our previous work, investigating the optimum conditions for maximum production rate in the overloaded elution mode [12].

Equilibrium isotherms

We assumed that the isotherms of the two components of the sample and of the displacer are given by the competitive Langmuir isotherm model [20]:

$$q_i = \frac{a_i C_i}{1 + b_1 C_1 + b_2 C_2 + b_3 C_3} \qquad i = 1, 2, 3 \qquad (3)$$

where q_i and C_i are the concentrations of component i at equilibrium in the stationary and the mobile phase, respectively, and a_i and b_i are numerical coefficients. As the Langmuir model is valid only if the column saturation capacities are identical for all components [21], the numerical parameters were chosen accordingly, with $a_1 = k'_1/0.25$, $a_2 = \alpha a_1$, $a_3 = \alpha a_2$, $b_1 = k'_1/325$ ml/mg, $b_2 = \alpha b_1$ and $b_3 = \alpha b_2$, where k'_1 is the retention factor of the first component at infinite dilution and α is the separation factor between subsequently eluted components. The separation factor for the less retained and the more retained components of the sample is assumed to be equal to the separation factor of the more retained sample component and the displacer. With these parameters, the saturation capacity of all compounds is 1300 mg/ml.

Definitions

The production rate was defined as the amount of the purified component produced per unit cross-sectional area and per unit time, in mg/cm^2 · s. The

TABLE I

OPTIMUM OPERATING CONDITIONS FOR A GIVEN COLUMN[a]

α	Compound			Production rate (mg/cm^2 · s)	Yield (%)	v	L_f (%)	C_d (mg/ml)
	C_1/C_2	No.	k'_1					
1.2	3:1	1	2.7	1.430	35.9	113	10.0	103
1.2	3:1	2	2.5	0.085	29.7	42	6.7	41
1.2	1:3	1	3.1	0.196	43.1	57	8.8	57
1.2	1:3	2	3.6	0.709	43.9	75	7.8	75
1.5	3:1	1	1.8	14.38	53.8	353	20.9	187
1.5	3:1	2	1.9	1.63	44.6	232	12.5	40
1.5	1:3	1	2.0	2.14	48.5	335	11.6	91
1.5	1:3	2	1.9	8.20	63.5	230	16.4	112
1.5	3:1	1	1.9	14.36	60.0	303	20.7	188
1.5	3:1	2	1.9	1.47	60.0	158	14.0	73
1.5	1:3	1	2.0	1.99	60.0	255	12.2	99
1.5	3:1	1	2.3	8.69	90.0	153	16.2	154
1.5	3:1	2	2.0	0.98	90.0	153	8.2	28
1.5	1:3	1	2.2	1.53	90.0	155	9.9	80
1.5	1:3	2	2.0	5.51	90.0	178	14.7	178
1.8	3:1	1	1.4	35.60	63.0	371	34.5	276
1.8	3:1	2	1.6	4.49	60.9	266	21.0	81
1.8	1:3	1	1.6	5.48	62.5	293	23.3	171
1.8	1:3	2	1.4	19.22	65.1	242	24.1	176

[a] $L = 25$ cm; $d_p = 20$ μm.

cycle time, *i.e.*, the time between two consecutive injections, was defined as the breakthrough time of the displacer front. No provsion is made for the regeneration of the column.

Calculation procedures

The modified simplex algorithm extended by Dose [22] to handle boundary problems was used in this work. The convergence of the simplex was checked by the standard deviation of the coordinates of the simplex vertices. The convergence was achieved when the standard deviation for all the parameters to be optimized decreased below 0.1%. The optimum separation conditions were calculated successively for the less and the more retained components. Binary mixtures of relative concentrations 1:3 and 3:1 were studied at three values of the separation factor ($\alpha = 1.2$, 1.5 and 1.8).

RESULTS AND DISCUSSION

Optimization of the experimental conditions for a given column

In the first part of this work the column design parameters (column length and particle size) were not changed, only the experimental conditions (sample loading factor, reduced velocity of the mobile phase and concentration of the displacer) were optimized simultaneously, for a given column. The optimum values of the parameters, the production rate and the corresponding value of the recovery yield are given in Table I.

Influence of the retention factor. As the separations that use displacement chromatography are usually carried out at high values of the retention factor, the optimization procedure was re-run several times, using decreasing values of the retention factor, starting with $k'_1 = 6$. This procedure was repeated with different combinations of the relative mixture composition and the separation factor. In Fig. 1, the production rates of both components are plotted against k'_1 for a separation factor of $\alpha = 1.8$. The optimum value of the retention factor, k'_1, was always found to be between 1.4 and 1.6 for either mixture composition studied, irrespective of the component for which the production rate is optimized. The production rates at the optimum retention factor are 48–67% higher than at $k'_1 = 6$, which is a significant increase. However, when the retention factor is decreased below the optimum value, the production rate drops rapidly.

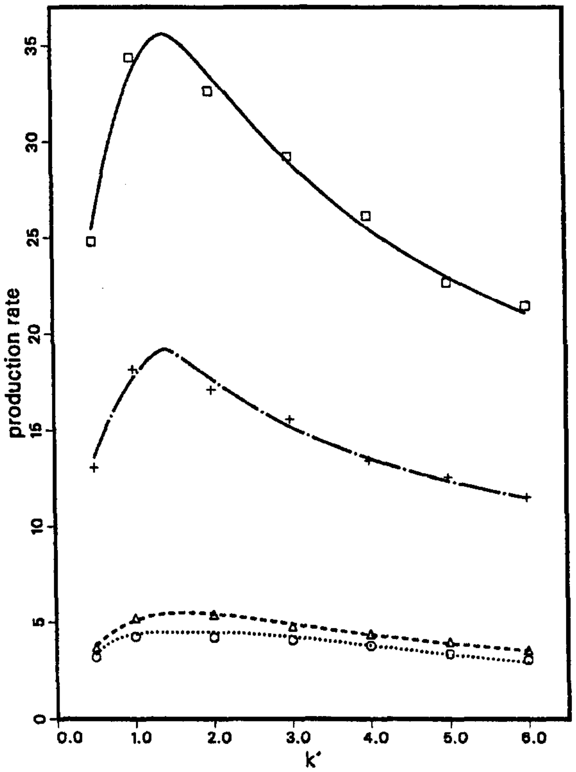

Fig. 1. Plot of the maximum production rates of either component *versus* the retention factor of the less retained component, $\alpha = 1.8$. (□) First and (○) second component of a 3:1 mixture; (△) first and (+) second component of a 1:3 mixture.

The same trend is observed at all values of the separation factor. For example, in Fig. 2a and b the maximum production rates corresponding to different yield constraints and mixture compositions are plotted *versus* k'_1 at $\alpha = 1.5$. The optimum value of k'_1 is now close to 2.0, and the gain in production rate on decreasing k'_1 from 6 to the optimum retention factor is less important than it was at $\alpha = 1.8$, but it is still 28–38%. At $\alpha = 1.2$ (Fig. 3), the optimum value of k'_1 is still higher, 2.4–3.6, and the gain in production rate associated with the decrease in k'_1 from 6 to the optimum value is only 16–25%.

Influence of a constraint on the recovery yield. Without any constraints, the recovery yield corresponding to the maximum production rate at $\alpha = 1.5$ was between 45 and 64% (Table I), depending on

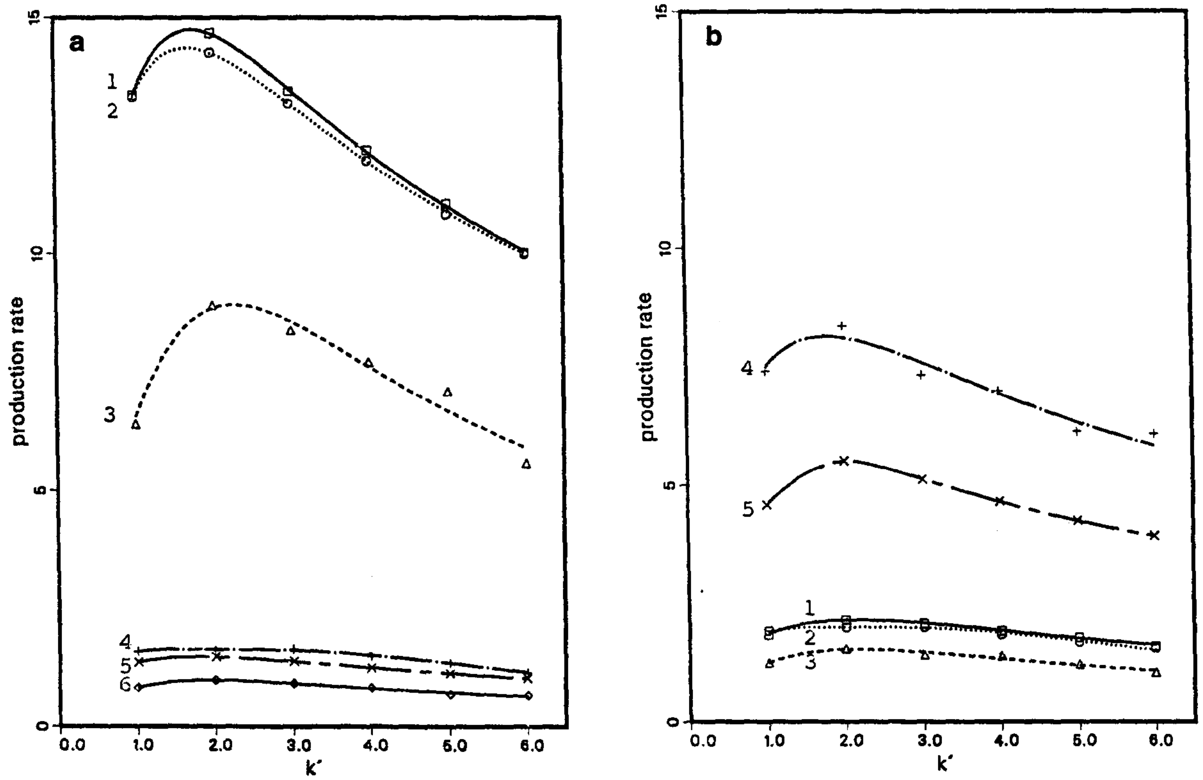

Fig. 2. Plot of the maximum production rates of either component *versus* the retention factor of the less retained component and influence of a recovery yield constraint, $\alpha = 1.5$. (a) Feed composition 3:1. 1 = First component without yield constraint; 2 = first component, 60% recovery yield; 3 = first component, 90% recovery yield; 4 = second component without yield constraint; 5 = second component, 60% recovery yield; 6 = second component, 90% recovery yield. (b) Feed composition 1:3. 1 = First component without yield constraint; 2 = first component, 60% recovery yield; 3 = first component, 90% recovery yield; 4 = second component without yield constraint; 5 = second component, 90% recovery yield.

the mixture composition and on whether the production rate of the first or the second component was optimized. The effect of a recovery yield constraint on the production rate was studied, setting minimum yield requirements of 60% and 90%.

The recovery yield achieved when no constraints are applied does not change significantly with the retention factor as long as this factor is higher than its optimum value, but decreases rapidly with decreasing retention factor if the separation is carried out at k' values smaller than the optimum. The effect of a recovery yield constraint on the production rate is shown in Fig. 2a and b ($\alpha = 1.5$). Since in this instance the recovery yield without constraint is only slightly less than 60% (or, even in one instance, is higher), there is no real change in the production rate when setting the constraint to 60%. When a minimum recovery yield of 90% is required, however, the production rate drops by about 30–40% compared with the global optimum. The optimum value of k'_1 is slightly higher when a recovery yield constraint is applied.

Influence of the separation factor. The optimization was carried out for three values of the separation factor, 1.2, 1.5 and 1.8 (Table I). As noted above, the optimum value of the retention factor increases with decreasing separation factor.

The production rate increases enormously when the separation factor is increased from 1.2 to 1.5. In three cases, a 10–11-fold increase was observed, while the gain is 17-fold for the purification of the second component of a 3:1 mixture. These gains are

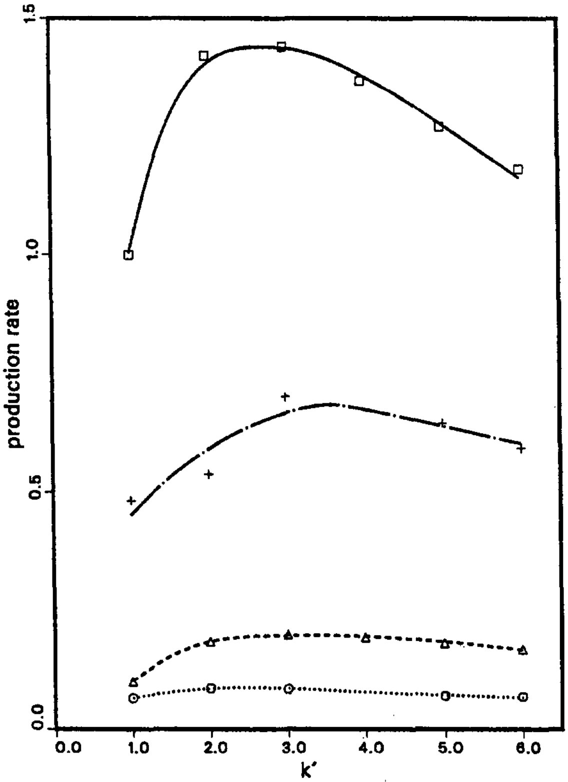

Fig. 3. Same as Fig. 1, except $\alpha = 1.2$.

achieved by large increases in both the optimum reduced velocity and optimum sample size. In the purification of the second component of a 3:1 mixture, a minor component is eluted between a main component and the displacer. For this separation at $\alpha = 1.2$ a very low reduced velocity ($\nu = 42$) is required to obtain the second component of the sample at a desired degree of purity, and the recovery yield is only 30%. At $\alpha = 1.5$, the optimum velocity is six times higher, the optimum sample size is doubled and the recovery yield is increased by 50% (relative).

The increase in production rate resulting from an increase in the separation factor from 1.5 to 1.8 is less significant, the improvement being by a factor of only *ca.* 2.3–2.75. These values are consistent with a production rate approximately proportional to $(1 - \alpha)^3$. The optimum experimental conditions are summarized in Table I.

Simultaneous optimization of the column design parameters and the operating conditions

The simultaneous optimization of the experimental conditions and the column design parameters leads to a five-dimensional maximum search. However, by utilizing the effects of the displacer concentration and the column length, as will be shown below, the problem can be reduced to a three-dimensional optimization. The optimum values of the parameters are given in Table II.

TABLE II

OPTIMUM VALUES OF THE PARAMETERS OF OPERATING CONDITIONS AND COLUMN DESIGN

α	Compound			Production rate (mg/cm^2 · s)	Yield (%)	ν	L_f (%)	d_p (μm)	d_p^2/L (μm^2/cm)	N
	C_1/C_2	No.	k'_1							
1.2	3:1	1	2.0	4.57	58.4	64	14.00	13.7	3.75	3 509
1.2	3:1	2	1.8	0.648	59.8	14	12.69	8.1	1.32	15 774
1.2	1:3	1	1.5	1.011	36.0	27	23.81	10.2	2.08	8 578
1.2	1:3	2	1.9	3.15	67.1	30	11.33	10.7	2.29	7 610
1.5	3:1	1	1.8	21.59	71.7	207	27.50	20.2	8.19	934
1.5	3:1	2	1.7	4.07	67.0	57	30.00	13.2	3.48	3 997
1.5	1:3	1	1.4	5.18	64.6	106	29.49	16.2	5.24	2 007
1.5	1:3	2	1.6	15.22	71.0	112	23.64	16.5	5.44	1 896
1.8	3:1	1	1.5	40.80	74.0	320	38.80	23.5	11.00	544
1.8	3:1	2	1.4	9.01	66.9	111	43.90	16.4	5.35	1 993
1.8	1:3	1	1.2	9.28	75.1	167	36.20	18.8	7.09	1 197
1.8	1:3	2	1.3	28.14	74.5	183	30.52	19.5	7.63	1 064

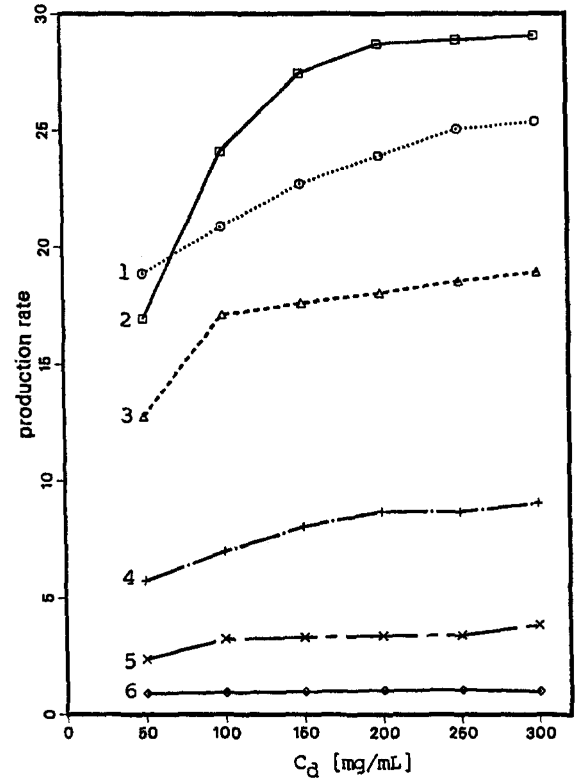

Fig. 4. Effect of the displacer concentration on the production rate of either component. 1 = Second component of a 1:3 mixture, $\alpha = 1.8$, $k'_1 = 1$; 2 = first component of a 3:1 mixture, $\alpha = 1.8$, $k'_1 = 6$; 3 = second component of a 1:3 mixture, $\alpha = 1.8$, $k'_1 = 6$; 4 = first component of a 1:3 mixture, $\alpha = 1.8$, $k'_1 = 1$; 5 = first component of a 3:1 mixture, $\alpha = 1.2$, $k'_1 = 6$; 6 = first component of a 1:3 mixture, $\alpha = 1.2$, $k'_1 = 1$.

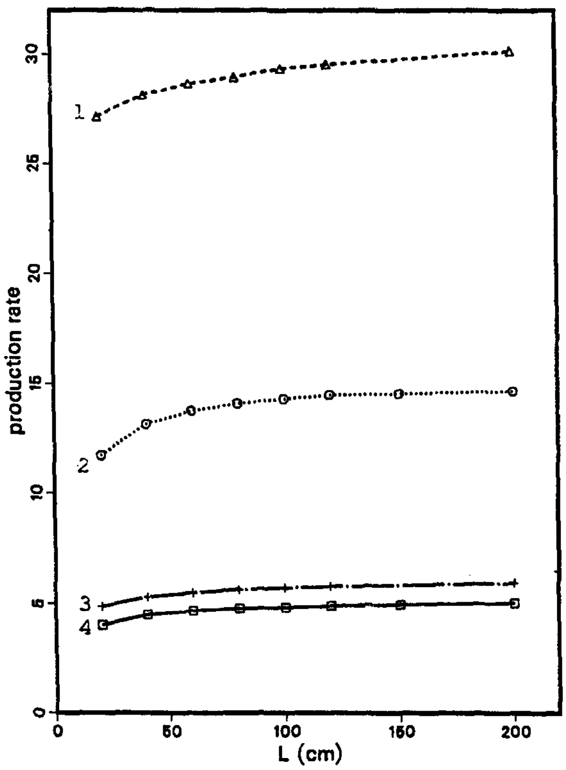

Fig. 5. Effect of the column length on the production rate. 1 = First component of a 3:1 mixture, $\alpha = 1.8$, $k'_1 = 6$; 2 = second component of a 1:3 mixture, $\alpha = 1.5$, $k'_1 = 2$; 3 = first component of a 1:3 mixture, $\alpha = 1.8$, $k'_1 = 6$; 4 = first component of a 3:1 mixture, $\alpha = 1.2$, $k'_1 = 2$.

Influence of the displacer concentration. To understand the effect of the displacer concentration on the production rate, we fixed the value of this parameter and optimized the others. However, we found that the higher the displacer concentration, the higher is the production rate. As the displacer concentration range was limited by a supposed solubility at 300 mg/ml, the production rate maximum was found at this concentration. The optimum values of the column design parameters, of course, change with varying displacer concentration, but the resulting recovery yield was independent of the displacer concentration. In Fig. 4 the effect of the displacer concentration on the production rate is shown for a few selected but typical sets of values of the retention factor, the separation factor and the mixture composition.

Consequently, in all subsequent calculations, a constant displacer concentration (300 mg/ml) was applied.

Influence of column length. The next calculations were carried out to optimize the average particle diameter, the sample size and the reduced velocity for a given column length. Different columns having lengths between 10 and 200 cm were considered, but no optimum value was found, as the longer the column the higher was the production rate of either component. The optimum average particle size increases with increasing column length, keeping the ratio d_p^2/L unchanged. It was found that, at the same time as the production rate, the recovery yield increases slightly with increasing column length. In

Fig. 5, some examples of the variation of the production rate with the column length are given.

As no optimum column length was found, we chose for further calculations a column of constant length (50 cm), and only the other column design parameter, *i.e.*, the average particle diameter, was optimized. This length is in agreement with the current practice. We see in Fig. 5 that the production rate gain associated with the use of a longer column is only marginal. With this restriction, of course, we shall not find the global optimum conditions for separation. However, the aim of this work was to draw qualitative information regarding the best practical conditions under which preparative separations should be conducted.

Optimum column design parameters. The analysis of the solution of the ideal model of chromatography demonstrates that neither the column length nor the average particle size has optimum values, but that there is an optimum value for the ratio d_p^2/L at which the maximum production rate will be achieved [23]. This result was essentially confirmed by numerical optimization of the overloaded elution mode based on the equilibrium-dispersive model of chromatography [12]. In the latter instance, only a very flat maximum was found when optimizing both the column length and the particle size.

The same phenomenon was observed for the displacement mode in this work. As reported above, when the experimental conditions are optimized for a particular column length, as shown in Fig. 5, the optimum particle size depends on the column length, and the ratio d_p^2/L remains virtually constant. The optimum values of the d_p^2/L ratios obtained for the same conditions as discussed in Fig. 5 are plotted *versus* the column length in Fig. 6. We see that the optimum value of the ratio changes very slowly with the column length. For this reason, we shall use also

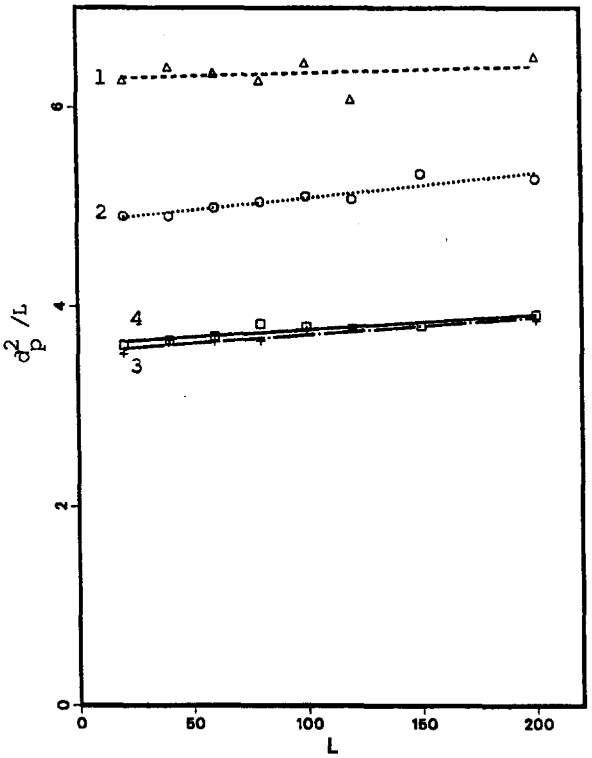

Fig. 6. Optimum value of d_p^2/L *versus* the column length. The lines correspond to the same conditions as in Fig. 5.

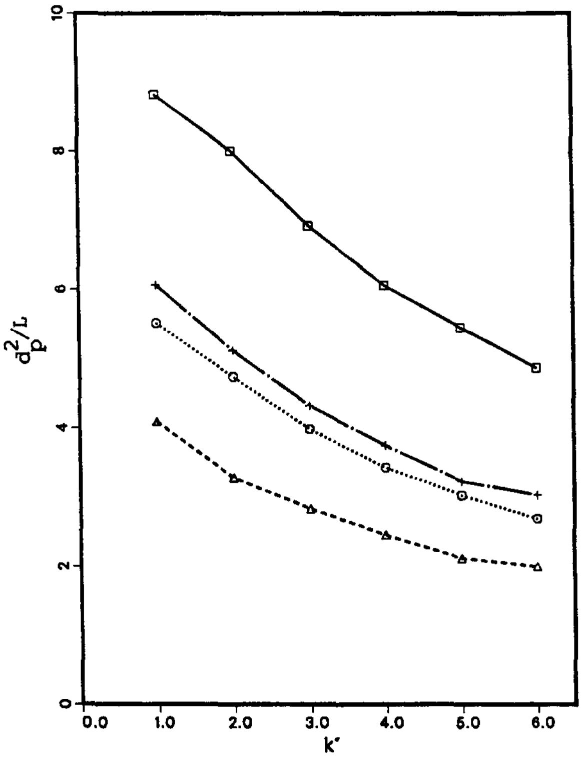

Fig. 7. Optimum value of d_p^2/L *versus* the retention factor of the less retained component; $\alpha = 1.5$. (□) First and (△) second component of a 3:1 mixture; (○) first and (+) second component of a 1:3 mixture.

in the displacement mode the ratio d_p^2/L for characterizing the column dimensions.

The optimum ratio d_p^2/L decreases rapidly with increasing retention factor, k'_1. In Fig. 7, the optimum value of d_p^2/L is plotted against k'_1 for some selected cases.

For the column used in the first part of this work ($L = 25$ cm, $d_p = 20$ μm), $d_p^2/L = 16$. For all the mixture compositions and the separation factors studied in this work, the optimum value of d_p^2/L is much smaller than 16. The closer the separation factor is to unity, the smaller is the optimum particle size for a column of given length (see Table II), because a higher column efficiency is required to separate the components of a mixture when the separation factor approaches 1.0.

Small particles are needed to purify the more retained component of a 3:1 mixture, *i.e.*, when a component at a relatively low concentration is eluted between the less retained component and the displacer. In this instance, the purity of the collected fraction is strongly influenced by the separation from the bands on both sides of the band of interest. For this reason, the production rate of the second component in a 3:1 mixture with $\alpha = 1.2$ increases about 7.6 times on switching from the standard column (Table I) to the optimum column (Table II).

In contrast, the easiest task is the purification of the less retained component from the same 3:1 mixture. In this instance the presence of both the second component of the sample and the displacer band tends to concentrate the first component band. The improvement of the prouction rate observed on replacing the standard column (Table I) by the optimum column (Table II) is 3.2-fold at $\alpha = 1.2$.

The difficulty with the other two cases studied, the purification of either the less or the more retained components from a 1:3 mixture, is intermediate

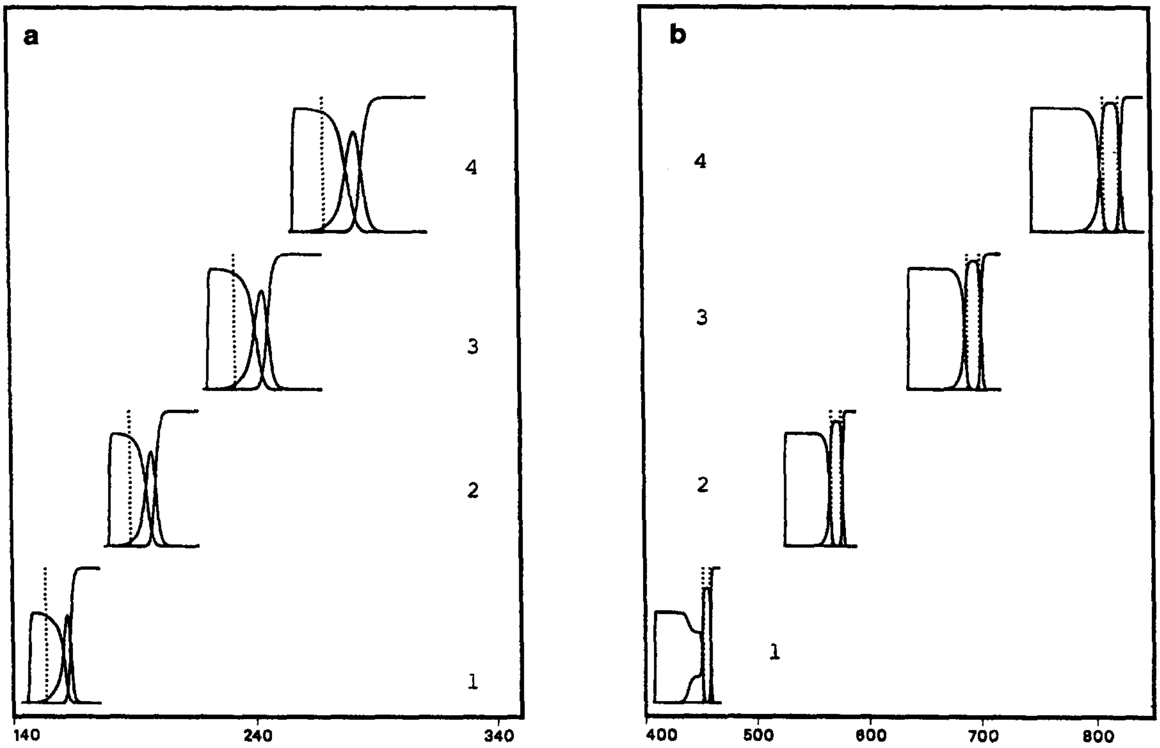

Fig. 8. Optimum chromatograms corresponding to the maximum production rate of (a) the less retained and (b) the more retained component in a mixture of relative concentration 3:1; the separation factor is $\alpha = 1.2$. Values of the retention factor: (1) $k'_1 = 1.0$; (2) $k'_1 = 2.0$; (3) $k'_1 = 3.0$; (4) $k'_1 = 4.0$. The vertical dotted lines indicate the cut points for fraction collection.

between the two situations described above. Nearly identical column characteristics are required (Table II).

Influence of mixture composition. As we have seen in the previous discussions, the relative concentration of the components of the mixture and whether the production rate of the less or the more retained component is optimized have a strong influence on the optimum column design.

In Fig. 8, two series of calculated chromatograms are plotted, corresponding to the maximum production rate of the less (Fig. 8a) and the more (Fig. 8b) retained components of a 3:1 binary mixture, with $\alpha = 1.2$, and for different values of the retention factor. In Fig. 8a, the band of the less retained component has a very steep front, and high concentration fractions can be collected. However, in this instance the column efficiency is poor, the second component band is broad and no pure second component at all can be recovered.

When the production of the second component is optimized (Fig. 8b), a much smaller reduced velocity (Table II) allows a much higher resolution between the second component band and those of the first component and the displacer, permitting the collection of 99% pure fractions of the second component, but the cut times on the two sides of the band are very close to one another.

Optimum value of the retention factor. At low values of the separation factor, the optimum retention factor at which the maximum production rate is reached is lower when using the optimum column (Table II) than with the standard column (Table I). For example, at $\alpha = 1.2$, the optimum retention factor, k'_1 is between 1.5 and 2.0 when all the column design parameters are optimized (Fig. 9), compared with the optimum value of k'_1 between 2.5 and 3.6 that was obtained for the column studied in Table I

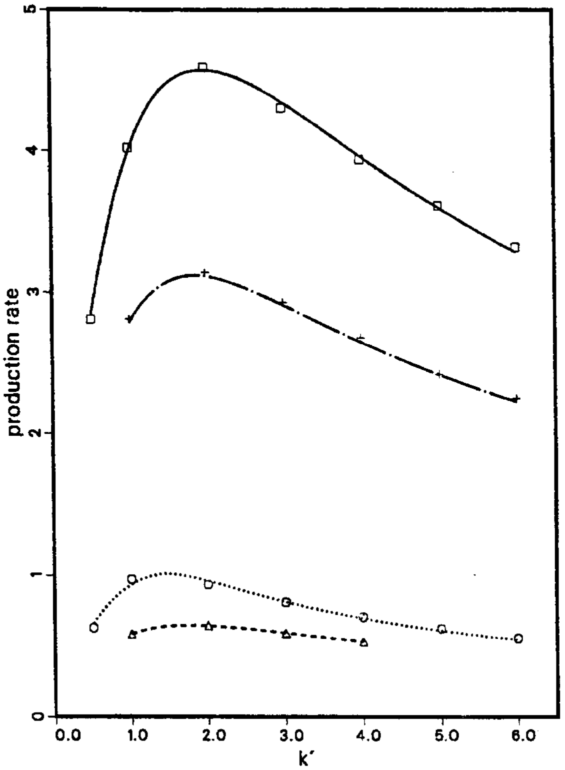

Fig. 9. Plots of the maximum production rate of either component *versus* the retention factor of the less retained component; $\alpha = 1.2$. (□) First and (△) second component of a 3:1 mixture; (○) first and (+) second component of a 1:3 mixture.

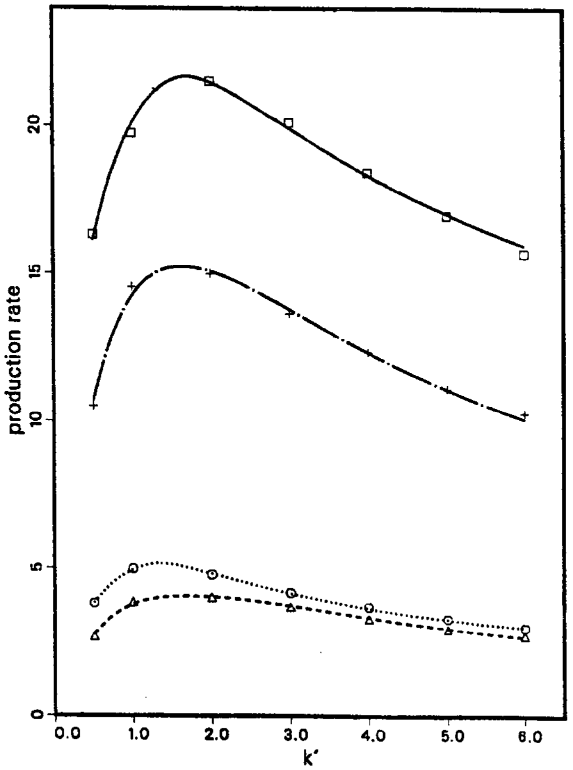

Fig. 10. Same as Fig. 9, except $\alpha = 1.5$.

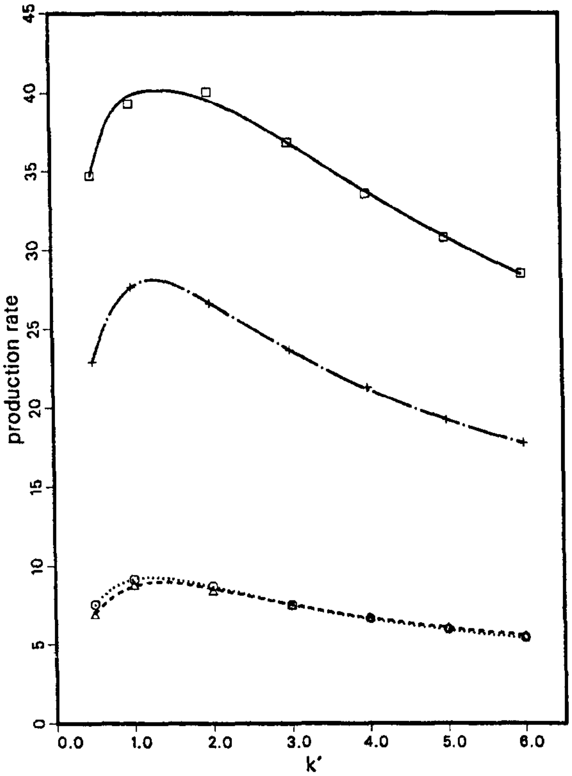

Fig. 11. Same as Fig. 9, except $\alpha = 1.8$.

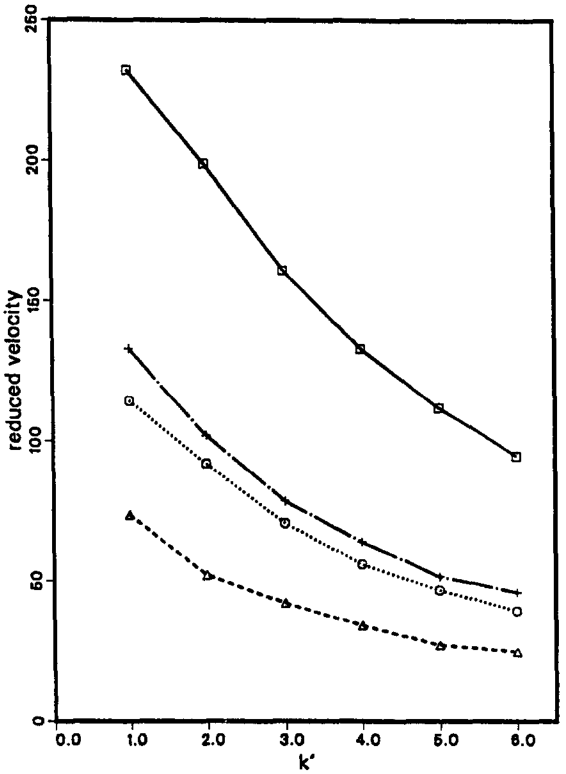

Fig. 12. Plot of the optimum mobile phase reduced velocity *versus* the retention factor of the first component for an optimum designed column. Same conditions as in Fig. 7.

and Figs. 1–4. At higher values of the separation factor, the value of the optimum retention factor is nearly independent of whether or not the column design parameters are optimized (Figs. 10 and 11). The optimum conditions are summarized in Table II.

The optimum sample size increases with increasing retention factor (as can be seen in Fig. 8a and b), but so does the cycle time, and it is the ratio of the sample size to the cycle time, multiplied by the recovery yield, which determines the production rate.

Optimum mobile phase velocity. When we optimized both the column characteristics and the experimental conditions for the separation, the optimum reduced velocity of the mobile phase was always determined by the maximum allowed inlet pressure. This confirmed previous theoretical results [23] that the maximum production rate for a given separation increases with increasing maximum allowed inlet pressure.

In contrast, in the first part of this work, when we optimized the operating conditions for a particular column, the maximum inlet pressure was never reached. Interestingly, as is seen in Fig. 12, the optimum reduced velocity decreases with increasing retention factor. The behavior of both the reduced velocity and the ratio d_p^2/L is just the opposite of what was found when optimizing the production rate in the overloaded elution mode [11].

As a consequence of this behavior, the column efficiency required to perform the separation at the optimum retention factor, is smaller than the efficiency needed when the separation is carried out at a retention factor higher than the optimum value. The dependence of the required efficiency on the retention factor and the influence of the mixture composition are illustrated in Fig. 13.

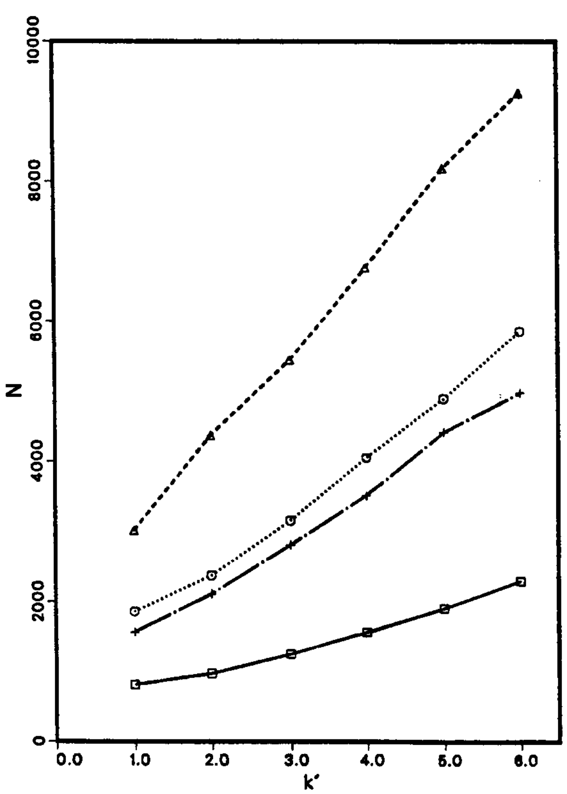

Fig. 13. Plot of the column efficiency required to perform the separation. Same conditions as in Fig. 7.

CONCLUSIONS

A few general rules can be derived from the results of this work, in spite of the fact that numerical solutions are always specific to a set of particular experimental conditions.

First, there always exists an optimum value for the retention factor. This optimum value is relatively low, much lower than the retention factors currently used in practice. It depends slightly on the mixture composition and on the component for which the production rate is to be maximized. The dependence of the optimum value of k' on the separation factor is more significant. The optimum retention factor is higher if the column design parameters are not optimum. The optimum sample size, and hence the throughput, increase with increasing retention factor, but the optimum conditions at higher k'_1 demand a more efficient column and the cycle time also increases.

Second, in the instances studied, no optimum displacer concentration was found. The more concentrated the displacer, the higher is the production rate. Third, no optimum column length was detected. The production rate of either component increases with increasing column length. However, there is always an optimum value for the ratio d_p^2/L. As long as this ratio is kept constant, the production rate does not change significantly.

Finally, the maximum production rate can be several times higher when the column having the optimum characteristics is chosen compared with a standard column operated under optimum conditions. This incentive to optimize the column design is most important when optimizing the production rate of the more retained component from a feed where its concentration is lower than that of the less retained component.

In all instances, the optimum conditions were found to be such that the isotachic train is not yet formed, but nearly so. Probably because the characteristics of our standard column were relatively closer to those of the optimum column, the chromatograms were also closer to the isotachic train than in our previous study, as in this case the fractions of both components could be collected at very high concentrations.

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