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Effect of feed concentration on the preparative separation of systems having reversed selectivity

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Abstract

A theoretical study of the effect of feed concentration on the preparative performance of liquid chromatography has been performed for systems having solute-concentration-dependent selectivity. The investigations were based on the numerical simulations of a detailed rate-equation model and the local equilibrium distribution between the fluid and adsorbed phases is assumed to follow a quadratic isotherm. The elimination and the utilisation of the selectivity reversal have been discussed in terms of the optimisation of the preparative performance.

1. Introduction

The establishment of a comprehensive theoretical framework relating the preparative performance of liquid chromatography to the competitive adsorption isotherm, the column parameters and the operating conditions is an important subject which has been actively studied in recent years. Considerable effort has been made to investigate the effects of various factors using numerical or analytical approaches. Felinger and Guiochon [1] have shown that the maximum production rates were obtained for very low values of the retention factor of the first component, of the order of 0.3–0.5. Ghodbane and Guiochon [2] investigated the influence of the relative retention on the column loading capacity and observed that the optimal production rate increases with an increase in the relative retention. By combining an analytical solution of the ideal model of chromatography with the classical expressions of band-broadening effects due to the finite efficiency, Golshan-Shirazi and Guiochon [3] derived expressions for the dependence of the production rate of the second solute of a binary mixture on the relative retention. The effect of mobile phase flow-rate on production rate and recovery in overloaded chromatographic column was also investigated from a theoretical standpoint [4].

Among the issues that have been addressed, one of the simplest, yet of the greatest practical importance, is how the feed concentration affects the preparative performance of liquid chromatography. Knox and Pyper [5] developed an equation for calculating the conditions for the optimum throughput, assuming no competitive adsorption and 100% yield and 100% purity. They concluded that concentration overloading provides the greatest throughput. The numerical

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simulations carried out by Katti and Guiochon [6] have demonstrated that low-volume, concentrated samples give higher production rates. Using the Craig model, CRAIGSM, to simulate the chromatogram of a binary mixture, Cretier et al. [7] confirmed the existence of the optimum injection concentrations corresponding to the maximum recovered amount of the solute of interest. They also found that the optimum injection concentration is an increasing function of column efficiency. Although the investigations published so far [6–9] have shown that increasing feed concentration is beneficial to the improvement of the preparative performance of liquid chromatography, these studies were basically limited to the cases having Langmuir competitive isotherms, in which the separation factor is a constant.

The Langmuir competitive isotherm is a good first-order approximation of the adsorption behaviour for similar compounds and especially for closely related isomers. In some practical situations, however, compounds would undergo strong sorbate–sorbate interactions in the stationary phase, violating the basic assumption of Langmuir isotherms. Depending on the comparative strengths of the interactions among the molecules of the same species and those between species, deviation from the adsorption behaviour of the Langmuir isotherm may be expected. It has been known that the selectivity in some chromatographic systems depends on the solute concentration, and selectivity reversal may occur in the operational concentration range [10]. The reversal of the elution order of *cis*- and *trans*-androsterone with increasing sample size has been observed [11]. The Langmuir competitive isotherm fails to explain the phenomena of peak reversal, since it assumes a constant separation factor. It has been demonstrated that the LeVan–Vermeulen isotherm can predict the inversion of the elution order of the components of binary mixtures at larger sample sizes [12]. A new isotherm with uneven saturation capacities, induced either by size exclusion or by racemic discrimination of the active sites toward the solutes, was also used to simulate the phenomena of peak reversals [13]. It has been recognised that the inversion of elution order is usually a consequence of selectivity reversal at high concentration. However, the peak reversal in elution occurs only when the sample size is so large that the concentrated sample is not diluted too much during migration inside the column. Otherwise, the sample will be quickly diluted, and the reversal may not occur at all.

Although there have been some studies directed to the phenomena of selectivity reversal at high concentrations, the effect of feed concentration on the preparative performance of liquid chromatography has not yet been well addressed for systems having solute-concentration-dependent selectivity. This work is devoted to deal with this effect by means of the simulations based on a general rate-equation model proposed previously [14].

2. Description of simulated cases

2.1. Equilibrium isotherm

The quadratic isotherm [15,16], suggested by statistical thermodynamics, was selected as the isotherm model for the simulations in this work:

$$q_1 = \frac{A_1C_1 + A_{12}C_1C_2}{1 + B_1C_1 + B_2C_2 + B_{12}C_1C_2} \tag{1a}$$

$$q_2 = \frac{A_2C_2 + A_{21}C_1C_2}{1 + B_1C_1 + B_2C_2 + B_{21}C_1C_2} \tag{1b}$$

where solute 1 is the early-eluted component at a very small sample size, and solute 2 is the late-eluted component. q_i and C_i are the concentrations of the component i at equilibrium in the stationary and mobile phases, respectively. All concentration units are in mg/ml. A_i, B_i, A_{ij} and B_{ij} are the isotherm parameters, and $B_{12} = B_{21}$. According to the physical meaning of these parameters, A_i, B_i and B_{ij} must be positive, and A_{ij} could be positive or negative. This isotherm defines a solute-concentration-dependent selectivity as follows:

$$S_f = \frac{q_2/C_2}{q_1/C_1} = \frac{A_2 + A_{21}C_1}{A_1 + A_{12}C_2} \tag{2}$$

Depending on the parameter values in Eq. 2, a binary system described by the quadratic isotherm could be with or without selectivity reversal. This work will focus attention to cases having selectivity reversal. By setting S_f (in Eq. 2) equal to 1, the rearrangement of the equation gives

$$C_1 = \frac{A_1 - A_2}{A_{21}} + \frac{A_{12}}{A_{21}} \cdot C_2 \tag{3}$$

This equation defines a boundary which divides the concentration range into two zones. One is for $S_f > 1$, and the other for $S_f < 1$. Although the sign of both A_{12} and A_{21} could be positive or negative, the combination of negative A_{12} and positive A_{21} would produce negative C_1 from Eq. 3 for any positive C_2. This case is physically unrealistic and will be not considered in this work. There are three physically possible cases in terms of the sign combinations of A_{12} and A_{21}. The isotherm parameters for these three cases (i.e. cases 1, 2 and 3) are presented in Table 1. These parameter values were chosen with reference to some experimental data [17]. Lines of C_1 versus C_2 have been plotted as shown in Fig. 1. It is clear by an examination of Fig. 1 that the separation factor is changed from $S_f < 1$ to $S_f > 1$ when the initial high feed concentrations, at which S_f is less than 1, are diluted to a certain extent.

The choice of the quadratic isotherm was based on the considerations that this isotherm model not only defines a solute-concentration-dependent selectivity but also has the theoretical rigour of statistical thermodynamics. In the comparison of various isotherm models for prediction of competitive adsorption data [17], it has been shown that the quadratic isotherm with seven floating parameters gives excellent fit of the experimental data. Because of the seven adjustable parameters, the quadratic isotherm could be fitted to adsorption data of most practical mixtures better than others. The theoretical soundness and empirical adaptability of the selected isotherm would ensure that the choice of the quadratic isotherm is more reasonable than others for the objective in this work.

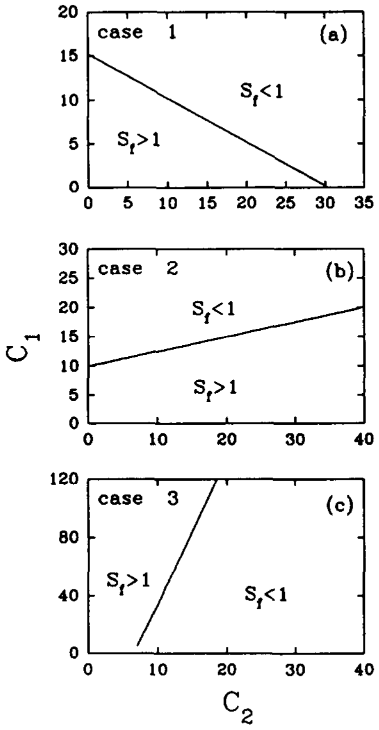

Fig. 1. Concentration zones giving $S_f > 1$ or $S_f < 1$.

2.2. Simulation model and conditions

The aim of this work is to study the effect of feed concentration on the preparative separation for systems with selectivity reversal by means of

Table 1
Parameters of quadratic isotherm

Case	A_1	A_2	B_1	B_2	A_{12}	A_{21}	B_{12}
1	2.0	2.76	0.04	0.065	0.025	−0.05	0.01
2	2.0	2.80	0.04	0.065	−0.02	−0.08	0.01
3	2.2	2.86	0.03	0.06	0.1	0.01	0.01

numerical simulations of a model. The feed concentration studied may be relatively high. Despite the numerous applications of lumped parameter models, the lumped kinetic coefficients in these models have to be solute-concentration dependent in order to accurately predict elution profiles for cases with high feed concentration and large sample size [18]. Because of this limitation, a simple lumped kinetic model with constant (i.e. solute-concentration independent) lumped parameters may be not sufficient for studies on selectivity reversal at high feed concentration. Therefore, a detailed rate-equation model, in which parameters, such as pore diffusivity, film mass transfer coefficient and axial diffusion coefficient, are basically independent of solute concentration, was used in the simulations of this work. The details of the model formulation and the numerical solution procedure based on the method of orthogonal collocation on finite elements were reported elsewhere [14].

The column for the simulations in this chapter was chosen as follows: column length $L = 25$ cm; average particle diameter $d_p = 10$ μm; void fraction of the packed bed $\epsilon_b = 0.5$; particle porosity $\epsilon_p = 0.4$. The superficial velocity, v_f, is $1.2 \cdot 10^{-4}$ m/s, which is equivalent to a reduced velocity $(v_f d_p / \epsilon_b D_{AB})$ of 40. The corresponding pressure drop of the column at this velocity is about 20 atm (1 atm = 101 325 Pa). The physical properties of the system are assumed to be as follows: the viscosity of the mobile phase was $\mu = 1.3$ cP; the solvent density was 1 g/ml. The molecular diffusivity of solute in bulk phase was $D_{AB} = 6 \cdot 10^{-11}$ m^2/s [19] and the pore diffusivity of solutes was $D_p = 8 \cdot 10^{-12}$ m^2/s. The Peclet number (*Pe*), accounting axial dispersion, and the film mass transfer coefficient (k_f), estimated by the correlations in literature [20,21], were 10 000 and $2 \cdot 10^{-5}$ m/s, respectively.

3. Results and discussion

The simulations of elution profiles were carried out for various feed compositions and concentrations by the numerical solutions of the dynamic model, using the model parameters in the previous section. The production rate and recovery yield were then determined from the simulated elution profiles. The definitions of production rate, recovery yield and load factor are the same as those in a previous report [14]. The product purity was set to be 99% for all the simulations.

The production rates (*P*) and recovery yields (*Y*) of case 1 under different feed concentrations were plotted against load factor (L_f) in Figs. 2 and 3 for feed composition 1:1. It was shown in these figures that the use of a properly diluted feed solution would permit an improvement in both production rate and the corresponding recovery yield. The maximum production rates of both solutes were increased more than 25%, when the feed concentrations were reduced from 40/40 to 5/5. However, the over-diluted feed solution (i.e. the feed concentrations are 1/1) does not enjoy this improvement. This is because the production rate depends not only on the separation factor but also on the column ad-

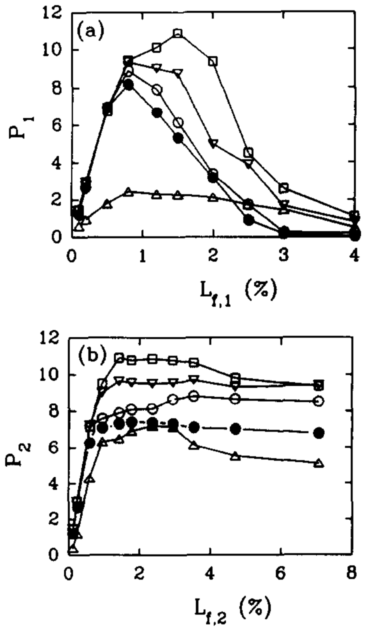

Fig. 2. Production rate versus load factor for case 1. Feed concentrations (C_1/C_2): ● = 40/40; ○ = 20/20; ▽ = 10/10; □ = 5/5; △ = 1/1.

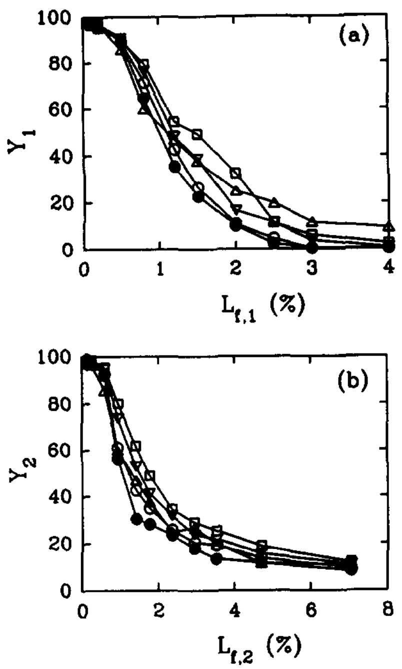

Fig. 3. Recovery yield versus load factor for case 1. Feed concentrations (C_1/C_2): ● = 40/40; ○ = 20/20; ▽ = 10/10; □ = 5/5; △ = 1/1.

sorption capacity which is in equilibrium with the feed concentration. It is suggested from these results that the production rates under very high recovery yield (i.e. corresponding low load factor) are not sensitive to the feed concentrations. The recovery yields corresponding to the maximum production rates are not very high. If this moderate recovery yield is acceptable for a given separation, the dilution of concentrated feed solution would be a worthwhile practice at least for a system such as case 1. The improvement on the preparative performance by the dilution of concentrated feed solution can be explained by the elimination of selectivity reversal. The separation factor for case 1 at the feed concentrations of 40/40 is less than 1, which means a selectivity reversal. As a consequence of the selectivity reversal, the solutes would undergo a certain degree of elution inversion, depending on the sample size. When the feed concentrations of both solutes are reduced to 5, the separation factor is always larger than 1 in the injection as well as in the whole process of elution, and the separation of the solute bands, therefore, consistently proceeds.

The elution profiles at load factors of 0.5 and 1.5% for the two different feed concentrations were shown in Fig. 4. It can be seen by comparing the elution profiles in Fig. 4a that the feed concentration has no significant influence on the touching-band separation. The difference in the retention times of different feed concentrations is due to the longer injection duration for the lower feed concentration at a fixed injection amount. The results in Figs. 4a and b imply that the selectivity reversal arising from high feed concentrations does not affect the separation with the requirement of very high recovery yield. The reason for this is that a small volume of sample solution can be quickly diluted due to the smoothing effect of axial dispersion and mass transfer resistance. On the contrary, the feed concentration does affect the separation at the load factor of 1.5% as shown in Fig. 4b. The elution profiles in the figure indicated that the separation under a lower feed concentration is

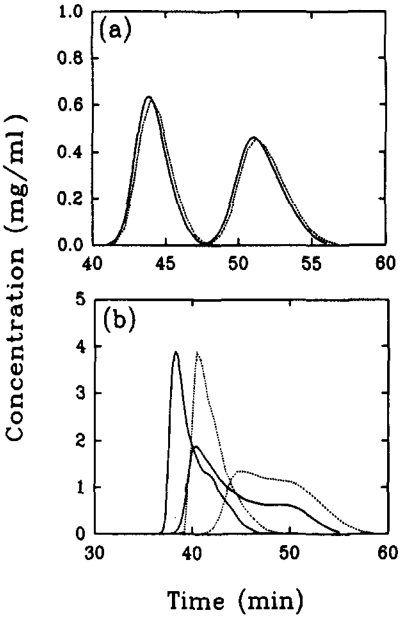

Fig. 4. Elution profiles for different feed concentrations for case 1. Feed concentration (C_1/C_2): solid lines, 40/40; dotted lines, 5/5. Sample size: (a) $L_{f,i} = 0.5\%$; (b) $L_{f,i} = 1.5\%$.

better than that for a high feed concentration. Although the elution profiles in Fig. 4b show no peak reversal, the elution order of the fronts of solute bands was indeed reversed to some extent during the injection and the early stage of elution, and then back to normal due to the dilution of band concentrations. It is the transient selectivity reversal that negatively influence the band separation. Fig. 4b also illustrates that there is no much difference on the peak heights for a given load factor, even though the feed concentrations were significantly different. This feature is particularly useful in practice, since the product concentrations are not significantly decreased by the proper dilution of feed solution.

The production rates and recovery yields of case 1 were presented in Figs. 5 and 6 for feed composition 5:1, and in Figs. 7 and 8 for feed composition 1:5. The production rates for diluted feed solutions at favourable sample sizes are always higher than those for higher feed concentrations, no matter what feed composition is involved. The corresponding recovery yields are also increased with a decrease in feed concentration. The separation factor corresponding the initial feed concentrations (i.e. 40/8 or 8/40) is changed from $S_f < 1$ to $S_f > 1$ after dilution to certain extent. Obviously, the elimination of selectivity reversal is also responsible for the improvement on the preparative performance for feed mixtures with compositions other than 1:1.

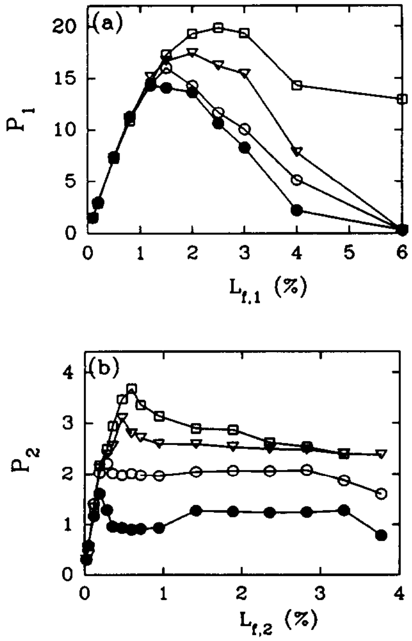

Fig. 5. Production rate versus load factor for case 1. Feed concentrations (C_1/C_2): ● = 40/8; ○ = 20/4; ▽ = 10/2; □ = 5/1.

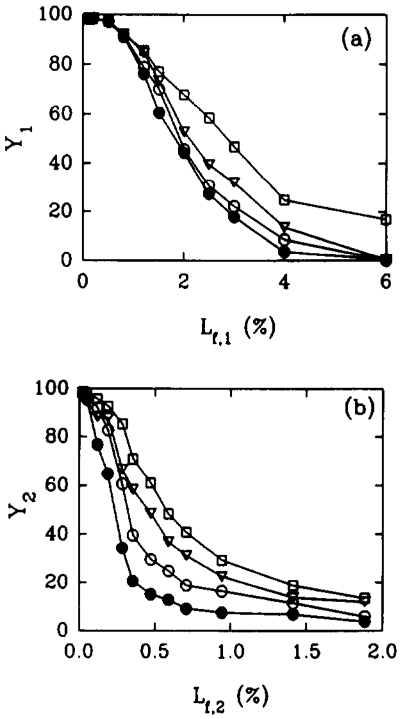

Fig. 6. Recovery yield versus load factor for case 1. Feed concentrations (C_1/C_2): ● = 40/8; ○ = 20/4; ▽ = 10/2; □ = 5/1.

To investigate the influence of the isotherm parameters, simulations were performed for cases 2 and 3. Figs. 9–12 show the production rates and recovery yields as functions of load factor for these two cases. The feed compositions and concentrations corresponding to the filled symbols in these figures were selected such that the separation factors are less than 1. By the stepwise dilution of the feed solution, the maximum production rates and the corresponding recovery yield can be improved. Similar to the results of case 1, the feed concentration has no effect on the separation having a high recovery

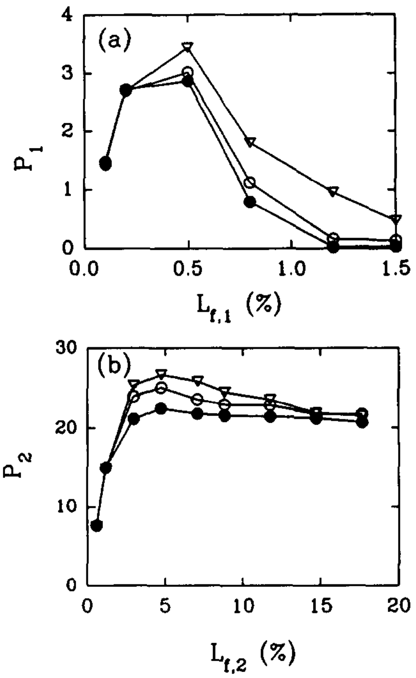

Fig. 7. Production rate versus load factor for case 1. Feed concentrations (C_1/C_2): ● = 8/40; ○ = 4/20; ▽ = 2/10.

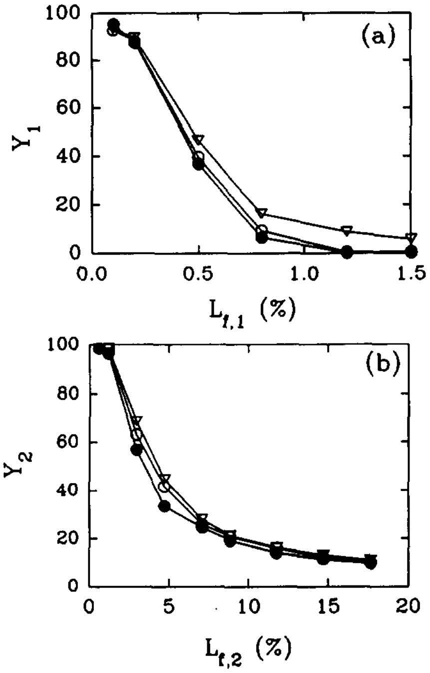

Fig. 8. Recovery yield versus load factor for case 1. Feed concentrations (C_1/C_2): ● = 8/40; ○ = 4/20; ▽ = 2/10.

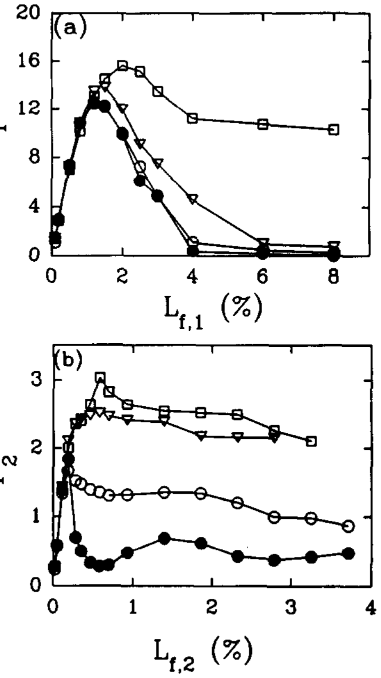

Fig. 9. Production rate versus load factor for case 2. Feed concentrations (C_1/C_2): ● = 40/8; ○ = 20/4; ▽ = 10/2; □ = 5/1.

yield. Simulations were also carried out to study the effect of the dilution of high feed concentrations which are located in the zones of $S_f > 1$, and the results show that the dilution for these situations have negative effects. Undoubtedly, the dilution of feed solution with high concentration would be an advantage only if the selectivity determined by the feed concentrations is reversed.

So far, we have demonstrated that the elimination of selectivity reversal by the dilution can improve the production rate and recovery yield at appropriate sample sizes. On the other hand, the selectivity reversal can be utilised to increase the production rate of solute 2, but the corresponding recovery yield is low. Fig. 13 shows the inversion of elution order observed at large sample sizes. When the inversion of elution order occurs, the band front of solute 1 emerges later than solute 2, while the rear boundary of the band for solute 1 is eluted out earlier than solute 2. As shown in the figure, the concentration

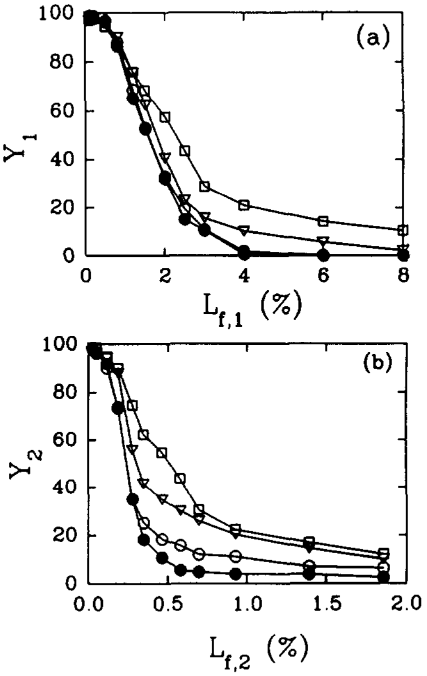

Fig. 10. Recovery yield versus load factor for case 2. Feed concentrations (C_1/C_2): ● = 40/8; ○ = 20/4; ▽ = 10/2; □ = 5/1.

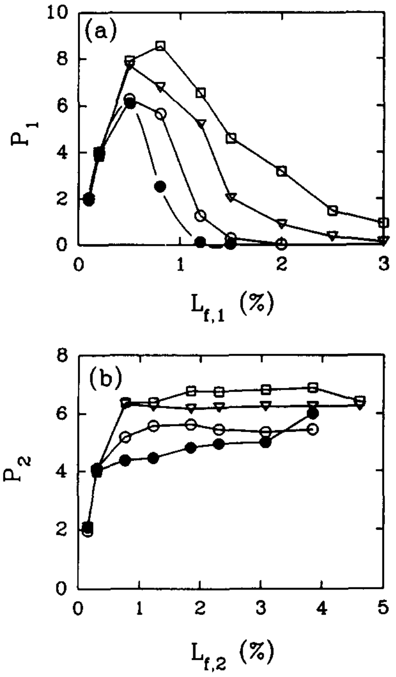

Fig. 11. Production rate versus load factor for case 3. Feed concentrations (C_1/C_2): ● = 40/40; ○ = 20/20; ▽ = 10/10; □ = 5/5.

band of solute 1 is completely embraced by the band of solute 2, and the recovery of pure solute 1 from the elution fractions is, therefore, impossible. However, the elution fractions containing solute 2 with high purity can be collected from the band front and the tailing part. If the selective reversal is maintained sufficiently long by injecting a large sample, the reversal displacement, i.e, solute 2 displaced by solute 1, would occur. As a result, a peak containing nearly pure solute 2 is formed before the band front of solute 1. Intuitively, the smaller the separation factor calculated at the feed concentrations, the stronger the reversed displacement effect. As shown in Fig. 13, the concentration of solute 2 in the first peak is even higher than that in feed solution due to the effect of reversed displacement.

Figs. 14 and 15 show the production rate and recovery yield of solute 2 obtained under the conditions of peak reversal. It is seen that a tremendous increase in the production rate is achieved by the reversal displacement, although the recovery yield remains low. If most of the injected sample can be recycled without causing significant problems, the utilisation of the effect of reversed displacement would provide a means to considerably increase the production rate of solute 2.

The simulations reported in this work are based on the quadratic isotherm which is derived from the statistical thermodynamics. A practical system may obey an isotherm other than the quadratic isotherm. However, the qualitative rules developed in this work could be reasonably extended to systems with other isotherms, as long as the phenomena of selectivity reversal takes place at certain high feed concentrations. Finally, it should be pointed out that the validity of the simulation results and the qualitative rules need no experimental confirmation, since the simulations were based on first principles.

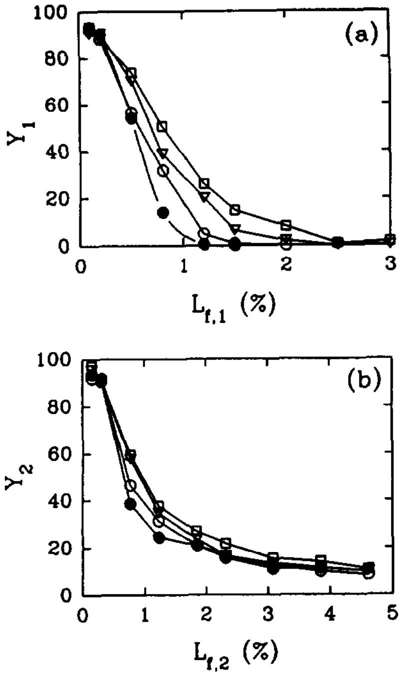

Fig. 12. Recovery yield versus load factor for case 3. Feed concentrations (C_1/C_2): ● = 40/40; ○ = 20/20; ▽ = 10/10; □ = 5/5.

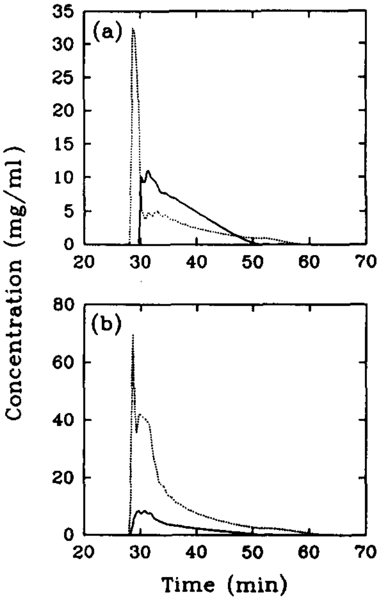

Fig. 13. Elution profiles with peak reversal. (a) Case 3; feed concentrations (C_1/C_2): 40/40; $L_{f,2}$ = 12%. (b) Case 1; feed concentrations (C_1/C_2): 8/40; $L_{f,2}$ = 40%. Solid lines = solute 1; dotted lines = solute 2.

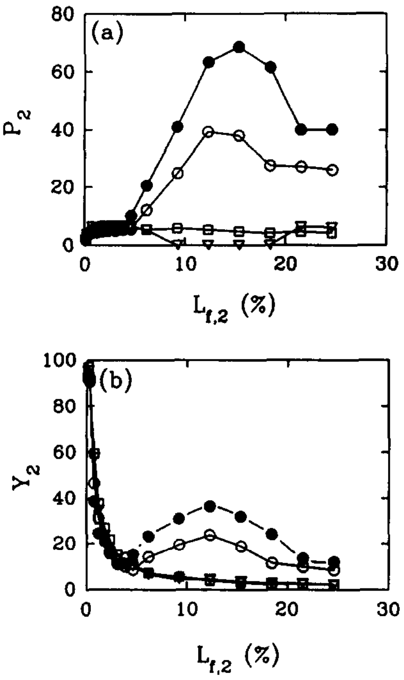

Fig. 14. Production rate and recovery yield of solute 2 for case 3. Feed concentrations (C_1/C_2): ● = 40/40; ○ = 20/20; ▽ = 10/10; □ = 5/5.

4. Conclusions

It has been demonstrated that the elimination of selectivity reversal, arising from high feed concentrations, by the dilution of feed solution would improve the preparative performance, especially for separations with a medium recovery yield. Cautions are also needed to avoid over-dilution, which gives low production rate. For separations with the requirement of very high recovery yield, the feed solution can be directly injected without the need of dilution, because the feed concentration has no effect on the touching-band separations. In the situation where the recycle of the major fraction of an injected sample is technically and economically feasible, the reversed displacement effect, taking place at large sample sizes, should be implemented in order to maximise the production rate of solute 2, i.e. the late-eluted component at a very small sample size.

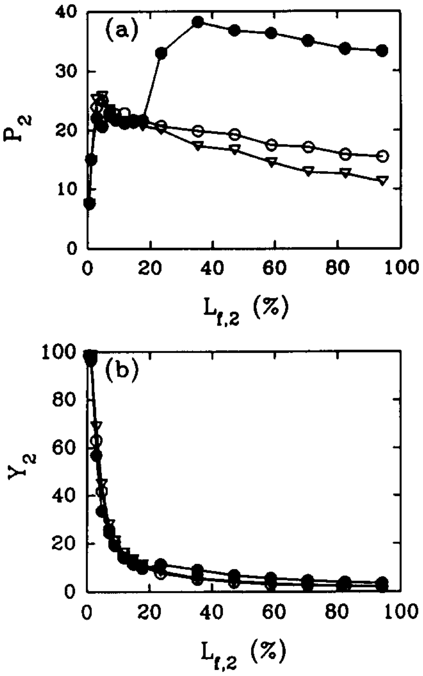

Fig. 15. Production rate and recovery yield of solute 2 for case 1. Feed concentrations (C_1/C_2): ● = 8/40; ○ = 4/20; ▽ = 2/10.

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