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Optimization of the experimental conditions in preparative liquid chromatography with touching bands

SADRODDIN GOLSHAN-SHIRAZI and GEORGES GUIOCHON*

**Department of Chemistry, University of Tennessee, Knoxville, TN 37996-1600 and Division of Analytical Chemistry, Oak Ridge National Laboratory, Oak Ridge, TN 37831-6120 (U.S.A.)*

ABSTRACT

Relationships are derived between the sample size permitting the elution of the components of a binary mixture as two touching bands and the parameters characterizing the separation and the column performance. These equations take into account the competitive interactions of the mixture components. They permit the determination of the optimum experimental conditions for maximum production rate with touching band elution.

The production rate increases monotonically with increasing inlet pressure available, provided that the column of optimum characteristics is used. For a given column, there is an optimum flow velocity and hence an optimum inlet pressure. Because of the competition between the mixture components for interaction with the stationary phase, the production rate under touching band conditions can be larger (under conditions of a predominant displacement effect) or lower (under conditions of a predominant tag-along effect) than the value derived when this competition is ignored.

INTRODUCTION

The optimization of experimental conditions for preparative chromatography is an important subject which has been actively studied in the recent years¹. Experimentalists have tried to generalize their observations^{2–5}. Various theoretical approaches have been used to derive either qualitative rules^{6–10} or quantitative procedures for the calculation of optimum values of the parameters of a chromatographic separation^{6–8,10}. Considerable confusion and some level of controversy are still understandably observed in an area where even the objectives are not always clarified. Depending on the type of work conducted, very different strategies are possible¹¹. The preparation of small amounts of purified material needed for further investigations of its properties cannot be carried out following the same methods and principles as the industrial production of large amounts of pure compounds to be sold as drugs or fine chemicals. The time spent in developing and optimizing the separation

is a major cost in the former instance and must itself be minimized. More subtle problems are still poorly understood. The optimum conditions for maximum production rate are very different for the first and the second component of a pair of components, especially if their relative concentration is far from unity¹⁰. Finally, specific requirements regarding the recovery yield and the fraction purity may affect considerably the outcome of the optimization process.

We have discussed recently the optimization of the experimental conditions for the purification of the second component of a binary mixture^{7,8}. We took into account the competition between the two components for interaction with the stationary phase and the finite column efficiency. We also allowed slight band overlap by considering the cases when the required purity of the prepared product can be 99% or lower and accepting low recovery yields (e.g., 60%). We showed that a considerable increase in the production rate is permitted by the use of high-performance columns at high reduced flow velocities^{8,10,12}.

Nevertheless, there is still great interest among practitioners in the "touching bands" case⁶. This permits the achievement of a recovery yield close to 100% and at the same time the production of nearly 100% pure compounds. The determination of the cutting time is straightforward and all the information required for the operation of the chromatograph is available from the detector signal. No separate on-line analysis of the eluate is necessary for the proper operation of the instrument. The influence of minor fluctuations in column temperature, sample size, mobile phase velocity and possibly composition has little consequence on the degree of band interference, so the control of the chromatograph is much simpler.

Knox and Pyper⁶ discussed the optimization of the experimental parameters in the touching band case. They made two restrictive assumptions: first they assumed that both band profiles are right triangles and second they neglected the competition between the two components and assumed that their elution is independent of each other. These assumptions are necessary for a simple, general solution of the problem. The extent to which they affect the end result has never been investigated. Recently, Snyder and Cox¹³ reformulated the equations derived by Knox and Pyper⁶, multiplying the number of non-independent equations and obfuscating the issue without contributing to the solution.

The purpose of this paper is the investigation of the solution of the optimization problem in the case of touching bands for components having a Langmuir competitive isotherm and a separate assessment of the errors introduced by the two assumptions made by Knox and Pyper⁶. The problem is investigated first with the ideal model, for which an exact solution can be derived under close form¹⁴. Then corrections are introduced into the ideal model solution to account for the effect of the finite column efficiency, as was done in previous papers^{6,8}.

This paper is not intended to be a critique of the landmark paper by Knox and Pyper⁶, but rather an extension of it. Within the limits of the assumptions they made, their results were correct and they provided an excellent first-order approximation. They were aware that neglecting the competitive interactions between the mixture components could not provide a better result⁶. Similarly, this paper attempts to take competitive interactions into account, but for the lack of a better model uses the Langmuir competitive isotherms. This isotherm model itself is only a first approximation, convenient for simulations, but which fits accurately the equilibrium data of few

systems^{15,16}. Accordingly, our results provide a second-order approximation, not exact values of the optimum experimental conditions. The accuracy of our predictions depends on the degree of agreement between the experimental isotherms and the competitive Langmuir model.

THEORY

Touching bands with the ideal model

The ideal model of chromatography neglects the axial dispersion and assumes that the mass transfers between phases are infinitely fast, *i.e.*, it assumes that the column efficiency is infinite, whatever the mobile phase velocity. Hence the only parameter to optimize is the loading factor, which is independent of the column length. In this section, we first derive the close-form equations giving the loading factor for which touching bands is achieved, in three successive cases: (i) in the general case, (ii) when competition between the two components is ignored and (iii) when competition is ignored and the elution profiles are right triangles.

The fundamental equations

We derive the equation giving the loading factor for touching bands in the general case, then introduce the simplifications brought by the assumptions made by Knox and Pyper⁶.

In the general case. We assume for the sake of simplicity that the equilibrium of the two components in the chromatographic system used is correctly described by binary Langmuir isotherms:

$$q_i = \frac{a_i C_i}{1 + b_1 C_1 + b_2 C_2} \quad (1)$$

where q_i is the amount of compound i adsorbed at equilibrium with a mobile phase where the concentrations of the components 1 and 2 are C_1 and C_2 , respectively. a_i and b_i are numerical coefficients characteristic of the compounds and the chromatographic system which can be derived from the pure compound isotherms.

We have shown previously¹⁴ that the loading factor, $L_{f,2}$, for the second component (*i.e.*, the ratio of the amount of second component in the sample injected to the column saturation capacity for this compound) which corresponds to exact separation of the two component bands, *i.e.*, to touching bands, is given by the following equation:

$$L_{f,2} = \frac{\left(\frac{\alpha - 1}{\alpha}\right)^2}{(1 + b_1 r_1 / b_2)} \quad (2)$$

where α is the relative retention of the two compounds ($\alpha = a_2/a_1 = k'_{0,2}/k'_{0,1}$) and r_1 is the positive root of the characteristic equation of the problem (ref. 8, eqn. II-3). In

practice, r_1 almost always is nearly equal to $C_{0,1}/C_{0,2}$, the ratio of the concentrations of the two components in the feed, so the loading factor for touching bands is given by

$$L_{f,2} = \frac{\left(\frac{\alpha - 1}{\alpha}\right)^2}{\left(1 + \frac{q_{s,2}C_{0,1}}{\alpha q_{s,1}C_{0,2}}\right)} = \frac{\left(\frac{\alpha - 1}{\alpha}\right)^2}{\left(1 + \frac{L_{f,1}}{\alpha L_{f,2}}\right)} \quad (3a)$$

where $q_{s,i} = a_i/b_i$, is the specific column saturation capacity. Solving eqn. 3a for $L_{f,2}$ gives

$$L_{f,2} = \left(\frac{\alpha - 1}{\alpha}\right)^2 - \frac{L_{f,1}}{\alpha} \quad (3b)$$

where $L_{f,1}$ is the loading factor for the first component. Depending on the relative concentration of the two components (*i.e.*, $C_{0,1}/C_{0,2}$), the loading factor for the second component corresponding to touching bands in the ideal model decreases from $[(\alpha - 1)/\alpha]^2$ (when $C_{0,1}$ is close to 0) to 0 (when $C_{0,2}$ is close to 0). The corresponding values of the loading factor of the first component are 0 and $(\alpha - 1)^2/\alpha$, respectively.

When the competition is ignored. In this instance we have two independent bands, the equilibrium isotherms of each compound being given by a simple one-component Langmuir isotherm. The condition for touching bands is that the band profile of the lesser retained component ends when the second component front is eluted. The first component band ends at the time $t_{e,1}$, given by¹⁷

$$t_{e,1} = t_p + t_{R0,1} \quad (4a)$$

where t_p is the width of the injection pulse, $t_{R0,1}$ and $t_{R0,2}$ are the limiting retention times of the two compounds at infinite dilution and t_0 is the hold-up time of the column. The retention time of the front of the second component¹⁷, $t_{R,2}$, is

$$t_{R,2} = t_p + t_0 + (t_{R0,2} - t_0)(1 - \sqrt{L_{f,2}})^2 \quad (4b)$$

The condition for touching bands is obtained by writing $t_{e,1} = t_{R,2}$. This gives the loading factor value for the second component:

$$L_{f,2} = \left(\frac{\sqrt{\alpha} - 1}{\sqrt{\alpha}}\right)^2 \quad (5)$$

When the competition is ignored and the band profiles are right triangles. We have shown that right triangles are the asymptotic solution of the ideal model in the case of Langmuir isotherms¹⁷. The exact solution of the ideal model for a parabolic isotherm is also a right triangle profile¹⁸. This parabolic isotherm is the two-term Taylor expansion of the Langmuir isotherm. In this case, the retention time of the band front is given by¹⁸

$$t_{R,2} = t_p + t_0 + (t_{R0,2} - t_0)(1 - 2\sqrt{L_{f,2}}) \quad (6)$$

Writing that this retention time for the second component is equal to the elution time of the end of the first component band gives the loading factor of the second component corresponding to touching bands in this assumption:

$$L_{f,2} = \left(\frac{\alpha - 1}{\alpha} \right)^2 / 4 \quad (7)$$

This equation is equivalent to that derived by Knox and Pyper⁶.

Production rate and throughput. It is remarkable that both eqns. 5 and 7 are independent of the concentration of the first component in the mixture, whereas the exact eqn. 3 depends on the composition of the binary mixture.

If we assume that the cycle time is equal to the corrected retention time of the second component, $t_c = t_{R0,2} - t_0$, we obtain the following general equations for the production rate of the two components, Pr_1 and Pr_2 , respectively:

$$Pr_2 / (1 - \varepsilon) S = q_{s,2} u L_{f,2} / k'_{0,2} \quad (8)$$

and

$$Pr_1 / (1 - \varepsilon) S = q_{s,1} u L_{f,1} / k'_{0,2} \quad (9)$$

where ε is the packing porosity, S the geometrical column cross-sectional area and u the mobile phase linear velocity. The throughput is given by

$$T / (1 - \varepsilon) S = u (q_{s,1} L_{f,1} + q_{s,2} L_{f,2}) / k'_{0,2} \quad (10)$$

In the case when a different definition is chosen for the cycle time, the results of eqns. 8–10 should be multiplied by $(t_{R0,2} - t_0) / t_c$. The following discussions are based on the use of the production rate for the second component. Obviously, in the case of touching bands the production rate for the first component of a binary mixture is the product of the production rate for the second multiplied by the relative concentration of the two compounds.

Consequence of the first assumption made by Knox and Pyper

As we have shown in a previous discussion, a right triangle is the asymptotic solution of the ideal model¹⁷. It is a good approximation of the band profile obtained with this model for a Langmuir isotherm when the loading factor is less than 1%. We have also shown that the elution profile obtained for real columns (*i.e.*, with a finite efficiency) can be approximated by right triangles for still larger values of the loading factor, in fact as long as $bC_{\text{Max}} \leq 0.1$ (ref. 18).

We can expect that for low values of the relative retention, α , the value of the loading factor corresponding to touching bands is small and this first approximation has no serious influence on the result. At higher values of α , in contrast, the consequence could be more significant. Figs. 1 and 2 illustrate this effect. On both figures are plotted the individual elution profiles obtained with the loading factors given by eqn. 5 (no competition, correct Langmuir profiles) and by eqn. 7 (Knox and Pyper value, no competition, right triangles). Fig. 1 corresponds to $\alpha = 1.20$ and Fig.

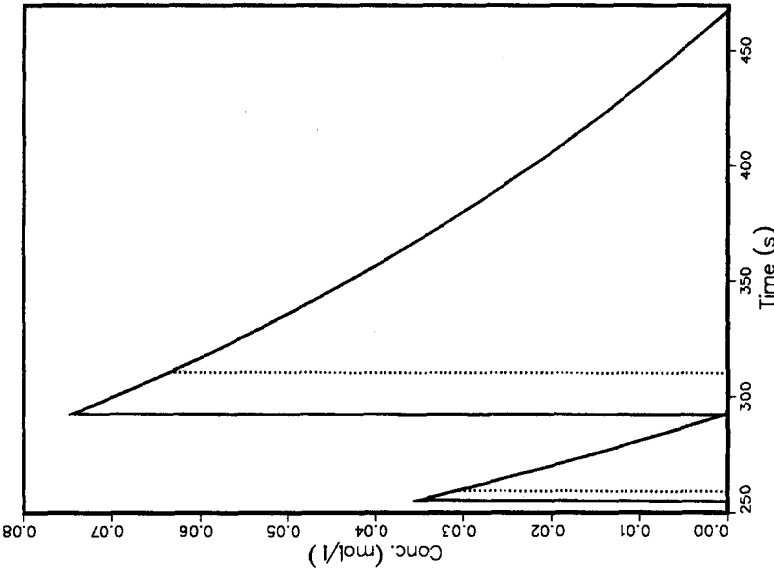


Fig. 1. Individual elution profiles of the bands of two components with non-competitive adsorption isotherms. Feed composition, 1:9; $K'_{0,1} = 6$; $\alpha = 1.2$; isotherm coefficients, $b_1 = 2.4$, $b_2 = \alpha b_1$; column length, 25 cm; mobile phase velocity, 0.6 cm/s. Solid lines: exact solution of the ideal model in the touching band case for two independent Langmuir isotherms. Dotted lines: profile calculated assuming the profiles are right triangles. Sample size from eqn. 5 (solid line) or eqn. 7 (dotted line).

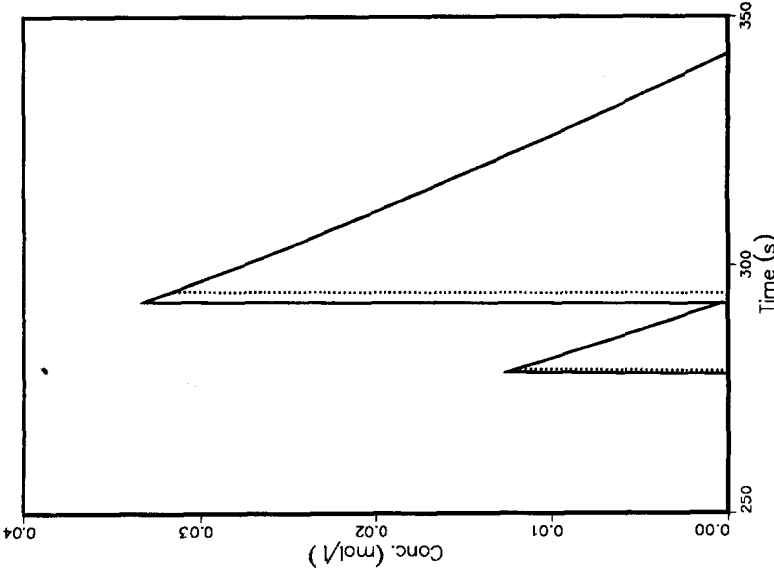


Fig. 2. Individual elution profiles of the bands of two components with non-competitive adsorption isotherms. Conditions as in Fig. 1, except $\alpha = 1.7$.

2 to $\alpha = 1.70$. In the former instance the differences are small but in the latter they are fairly important. This shows that the assumption that the band profiles are right triangles is reasonably satisfactory if the relative retention is close to unity (difficult separations). If the relative retention is large, the assumption may contribute to some extent to the large difference between the production rates predicted by the Langmuir competitive and the parabolic non-competitive models.

Consequence of the second assumption made by Knox and Pyper

A much more profound and important source of error comes from the assumption of an independent behavior of the two bands during their migration. In non-linear chromatography, the competition between the different components of the mixture cannot be neglected^{4,19,20}. As long as the column is overloaded, *i.e.*, behaves non-linearly for one component, it behaves similarly for all the components whose bands interfere with the band of that compound. In agreement with the independence assumption, the loading factor for touching bands (eqns. 5 and 7) and the corresponding values of the production rates for the two compounds (eqns. 8 and 9) do not depend on the composition of the feed. This is not realistic, and eqn. 3 shows that, on the contrary, the loading factor does depend strongly on this composition when the competition is taken into account.

Comparing eqns. 3 and 7 shows that the values of the loading factors they predict are equal only for $L_{f,1}/\alpha L_{f,2} = b_1 C_{0,1}/b_2 C_{0,2} = 3$. When the ratio $L_{f,1}/\alpha L_{f,2}$ is smaller than 3 (dominant displacement effect), the loading factor corresponding to touching bands (eqn. 3) is larger than predicted by the Knox and Pyper model (eqn. 7), whereas the reverse is true for values of that ratio larger than 3 (dominant tag-along affect). If we inject the amount derived from eqn. 7, we obtain well resolved bands in the former instance and overlapping bands in the latter.

Figs. 3–5 show two chromatograms each. They have been obtained for three different feed compositions, under the same simulated experimental conditions with the sample sizes predicted by eqns. 3 and 7, respectively. In all instances, the band profiles are obtained as the analytical solutions of the ideal model for binary Langmuir isotherms¹⁴. Fig. 3 is obtained with a relative feed composition such that $L_{f,1}/\alpha L_{f,2}$ is smaller than 3. As expected, the sample size predicted by eqn. 3 is the larger and the sample size predicted by eqn. 7 leads to well resolved bands (resolution 1.2), whereas the nearly four times larger sample size which is predicted by eqn. 3 leads to touching bands. A strong displacement effect is observed. The rear profile of the first component is convex upward whereas the rear profile of the second band is convex downward, as is conventional with Langmuir isotherms, for single components. For Fig. 4, the feed composition is such that $L_{f,1}/\alpha L_{f,2}$ is equal to 3 and the two sample sizes are equal. Note, however, that the profile predicted by the ideal model is different from the right triangle assumed by Knox and Pyper⁶. For Fig. 5, the ratio $L_{f,1}/\alpha L_{f,2}$ is larger than 3. Now the sample amount predicted by eqn. 7 is too large. The chromatogram obtained corresponds to strongly overlapping bands. To obtain touching bands, the smaller sample given by eqn. 3 should be used. It is nearly half the one predicted by eqn. 7. The column is still strongly overloaded and a marked tag-along effect is observed.

The previous discussion concerns only ideal chromatography. With real columns, the elution bands are wider and shorter than predicted by the ideal model. The concentration shock is damped by the finite rates of the axial diffusion and the

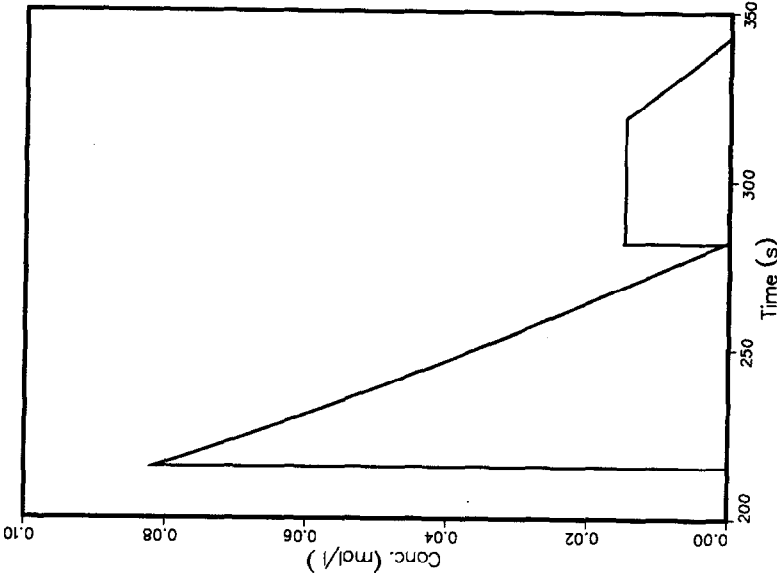


Fig. 3. Individual elution profiles of the bands of two components with Langmuir competitive adsorption isotherms. Conditions as in Fig. 1. Solid lines: elution profiles calculated with the ideal model using the sample size predicted by eqn. 3. Dotted lines: elution profiles calculated following the method of Knox and Pyper⁶, with the sample size given by eqn. 7. The production rate predicted by the non-competitive model is too low.

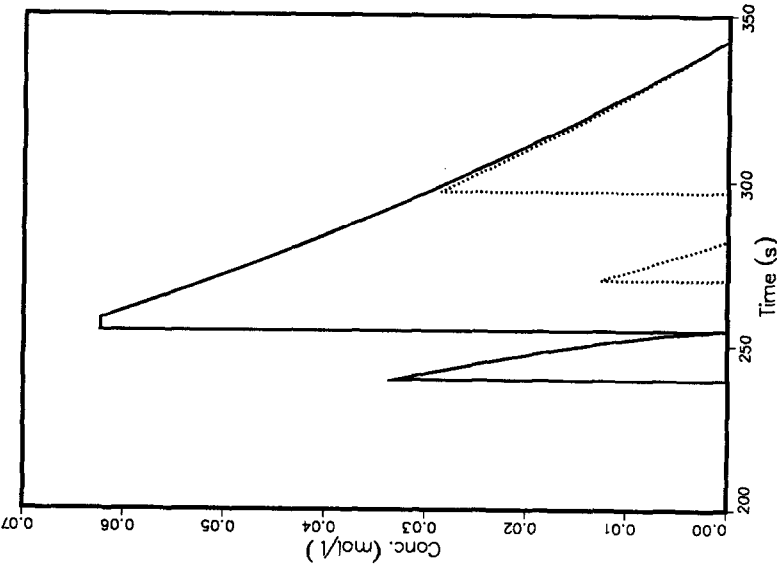


Fig. 4. Individual elution profiles of the bands of two components with Langmuir competitive adsorption isotherms. Conditions as in Fig. 3, except feed composition = 3.6:1 ($L_{j,1}/\alpha L_{j,2} = 3$). The production rates predicted by the competitive and the non-competitive models are equal.

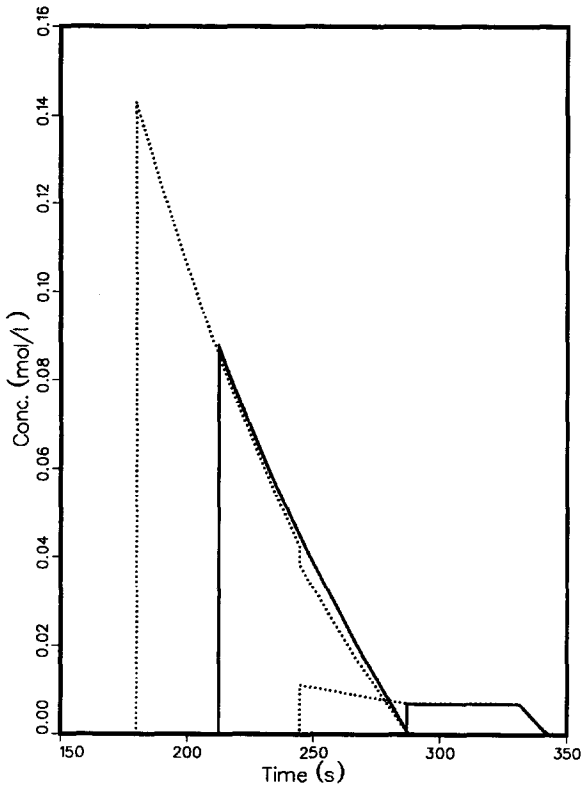


Fig. 5. Individual elution profiles of the bands of two components with Langmuir competitive adsorption isotherms. Conditions as in Fig. 3, except feed composition = 9:1. The production rate predicted by the non-competitive model is too high.

mass-transfer kinetics, so a shock layer is observed instead. In order to achieve touching bands, the sample should be smaller than predicted by eqn. 3. The calculation of the sample corresponding to touching bands for an actual column is discussed in the next section.

Touching bands in the case of a real column

In this section we derive a close-form expression of the sample size corresponding to touching bands in the case of an actual column, with a finite efficiency. This value is a function of the column efficiency and, accordingly, of the column characteristics, length, packing particle size and packing quality, and of the flow velocity. In the following section we discuss the optimization of the experimental conditions.

In a previous paper we showed that the maximum production rate is achieved when the column is operated at a high mobile phase flow velocity, so that the cycle time is short⁸. The column efficiency is then low and the relative band width is large. The effect of the band width on the degree of band interference is significant. Similarly, in the touching band case we must take into account the effect of column efficiency on the

band profile. A further difficulty is in the definition of the touching band condition. With the ideal model, the elution of a band begins and ends at well defined times. Before and after, the concentration of the compound in the mobile phase is zero. There is no such thing with real columns: the beginning and end of a band elution depend on the sensitivity of the detection. We shall neglect these effects, however, and write that the difference between the elution times of the rear of the second and first bands is equal to the total band width of the second component.

Derivation of the band width of the second component

The effect of the finite column efficiency on the band broadening during its elution can be accounted for by following the procedure first suggested by Haarhoff and Van der Linde²¹ and later used by Poppe and Kraak²² and Knox and Pyper⁶. We assume that the two contributions to the elution band width with which we are concerned, the thermodynamics (*i.e.*, the non-linear behavior of the phase equilibrium) and the kinetics (finite rates of mass transfer between phases and of axial dispersion) can be treated as two independent contributions. Therefore, their variances are additive and we have

$$\sigma_{\text{tot}}^2 = \sigma_{\text{th}}^2 + \sigma_{\text{kin}}^2 \quad (11)$$

where $\sigma_{\text{tot}}^2 = w_{\text{tot}}^2/16$ is the variance of the elution profile, $\sigma_{\text{th}}^2 = w_{\text{th}}^2/16$ the variance contribution due to the non-linear behavior of the isotherm and $\sigma_{\text{kin}}^2 = w_{\text{kin}}^2/16$ the variance contribution of the mass-transfer kinetics and of the axial dispersion, where the w s are the corresponding baseline band widths. By definition of the column height equivalent to a theoretical plate, H_{tot} , we have

$$H_{\text{tot}} = H_{\text{th}} + H_{\text{kin}} \quad (12)$$

The kinetic contribution is given by

$$w_{\text{kin}}^2 = \frac{16}{N_0} \left(\frac{k'_{0,2} + 1}{k'_{0,2}} \right)^2 (t_{R0,2} - t_0)^2 \quad (13)$$

where N_0 is the column plate number under linear conditions (in practice, for a very small size sample), $k'_{0,2}$ is the column capacity factor at infinite dilution (*i.e.*, $k'_{0,2} = a_2F$, where F is the phase ratio) and $t_{R0,2}$ and t_0 are the limiting retention time at infinite dilution of the second component and the hold-up time, respectively.

The thermodynamic contribution is calculated from the band width of the solution of the ideal model¹⁴. In this work we are interested in the width of the second component band. There are two cases. The ideal model solution shows that the band of the second component has completely recovered from its interaction with the first component band and achieved the same profile as if the same amount of second component was injected alone when the loading factor is equal to or smaller than $L_{f,2,p}$ ¹⁴, as given by

$$L_{f,2,p} = \left(\frac{\frac{\alpha - 1}{\alpha}}{1 + \frac{b_1 r_1}{b_2}} \right)^2 = \left(\frac{\alpha - 1}{\alpha + \frac{q_{s,2} C_{0,1}}{q_{s,1} C_{0,2}}} \right)^2 = \left(\frac{\frac{\alpha - 1}{\alpha}}{1 + \frac{L_{f,1}}{\alpha L_{f,2}}} \right)^2 \quad (14)$$

When the loading factor is between the values given by eqns. 3 and 14, we still have resolved bands, but the band profile of the second component has not had time to recover from the deformation resulting from the interaction between the two component bands. Consequently, a different procedure must be used to calculate the band width of the second component, depending on whether the loading factor is smaller or larger than $L_{f,2,p}$.

Loading factor of the second component for touching band when $L_{f,2} < L_{f,2,p}$

In this case, the profile of the second component band is the same as if it is injected pure. Its width is given by the following equation^{17,23}:

$$w_{th} = (2\sqrt{L_{f,2}} - L_{f,2})(t_{R0,2} - t_0) \quad (15)$$

The total band width of the second component band in the touching band case is equal to the difference between the retention times of the rear of the first band and the rear of the second band:

$$w_{tot} = t_{R0,2} + t_p - t_{e,1} \quad (16)$$

In order to calculate the loading factor corresponding to touching bands, we must know the retention time of the rear of the first component band, $t_{e,1}$. It can be derived from eqn. 63 in ref. 14 by writing that $C_1 = 0$ in this equation. We obtain

$$t_{e,1} = t_p + t_{R0,1} - \frac{L_{f,2}}{(\alpha - 1)}(t_{R0,2} - t_0) \quad (17)$$

This equation demonstrates the consequence of band interactions in the column. Although we are looking for conditions under which the two bands are just resolved when they elute, they have interacted during their entire migration. If there were to be no interaction, the retention time of the end of the first band would be $t_e = t_p + t_{R0,1}$. Eqn. 17 shows that the band is actually eluted earlier. The decrease in the retention time is directly proportional to the loading factor for the second component.

Combining eqns. 16 and 17 gives

$$w_{tot} = (t_{R0,2} - t_0) \left(\frac{\alpha - 1}{\alpha} + \frac{L_{f,2}}{\alpha - 1} \right) \quad (18)$$

Eqn. 18 shows that the second component band width corresponding to touching bands and, accordingly, the apparent column efficiency, are a function of the sample size. This is the major difference from the simplified approach by Knox and Pyper⁶, which assumed that the values of w_{tot} and N_{tot} corresponding to touching band are constant, independent of the amount of second component injected.

Combination of eqns. 11, 13, 15 and 18 gives

$$(2\sqrt{L_{f,2}} - L_{f,2})^2 + \frac{16}{N_0} \left(\frac{k'_{0,2} + 1}{k'_{0,2}} \right)^2 = \left(\frac{\alpha - 1}{\alpha} + \frac{L_{f,2}}{\alpha - 1} \right)^2 \quad (19)$$

Solving eqn. 19 for $L_{f,2}$ and putting the solution into eqn. 8 gives the production rate of the second component.

Simplified equations

Several simplifications are possible. We can assume that the loading factor is relatively small and neglect $L_{f,2}$ in comparison with $\sqrt{L_{f,2}}$ in eqn. 19. This amounts to assuming that the band profile of the second component is a right triangle, as did Knox and Pyper⁶, but still conserves the competitive interaction between the two components which is introduced by the competitive Langmuir isotherms. In this case, it is easy to solve the modified eqn. 19 and to obtain

$$L_{f,2} = (\alpha - 1)^2 \left\{ 2 - \frac{1}{\alpha} - \sqrt{4 \left(\frac{\alpha - 1}{\alpha} \right)^2 + \frac{16}{N_0} \left[\frac{k'_{0,2} + 1}{k'_{0,2}(\alpha - 1)} \right]^2} \right\} \quad (20)$$

Inserting the value of $L_{f,2}$ into eqn. 8 gives the production rate:

$$\frac{Pr_2}{(1 - \varepsilon)S} = \frac{q_{s,2}u(\alpha - 1)^2}{k'_{0,2}} \left\{ 2 - \frac{1}{\alpha} - \sqrt{4 \left(\frac{\alpha - 1}{\alpha} \right)^2 + \frac{16}{N_0} \left[\frac{k'_{0,2} + 1}{k'_{0,2}(\alpha - 1)} \right]^2} \right\} \quad (21)$$

If, in addition, we assume that the competitive interaction between the two components of the mixture is negligible, we ignore the term $L_{f,2}/(\alpha - 1)$ on the right-hand side of eqn. 19 and $L_{f,2}$ compared with its square root on the left-hand side. We then obtain

$$L_{f,2} = \left[\left(\frac{\alpha - 1}{\alpha} \right)^2 - \frac{16 \left(\frac{k'_{0,2} + 1}{k'_{0,2}} \right)^2}{N_0} \right] / 4 \quad (22)$$

Combination of eqns. 8 and 22 gives the production rate:

$$\frac{Pr_2}{(1 - \varepsilon)S} = \frac{q_{s,2}u}{4k'_{0,2}} \left[\left(\frac{\alpha - 1}{\alpha} \right)^2 - \frac{16 \left(\frac{k'_{0,2} + 1}{k'_{0,2}} \right)^2}{N_0} \right] \quad (23)$$

This equation is the same as that given by Knox and Pyper⁶ (combination of their eqns. 40, 47, 63, 67) in the case when the cycle time is equal to $t_{R0,2} - t_0$.

Loading factor of the second component for touching band when $L_{f,2} > L_{f,2,p}$

Eqn. 19 is valid only as long as the loading factor obtained is lower than the threshold value (or equal to it), $L_{f,2,p}$ derived from eqn. 14. In contrast, when $L_{f,2}$ is larger than $L_{f,2,p}$, a plateau remains on the top of the second elution band, at least within the ideal model approximation. This plateau is more or less eroded by the axial diffusion and the finite rate of the mass-transfer processes. In this instance, neither the first nor the second component band profiles are identical with the band profile obtained with a pure component sample. The first band is modified by the

displacement effect and the second has not yet recovered from the tag-along effect.

Because the concentration of the second component is constant during elution of the plateau, we can assume, as a first approximation, that the finite column efficiency does not affect the contribution of the length of this plateau to the band width⁸. Hence, the total band width in this instance should be calculated as

$$w_{\text{tot}} = w_{\text{plateau}} + \sqrt{w_{\text{kin}}^2 + (w_{\text{th}} - w_{\text{plateau}})^2} \quad (24)$$

where w_{plateau} is the width of the plateau (see below). As we have shown elsewhere⁸, we have

$$w_{\text{th}} - w_{\text{plateau}} = (2\sqrt{L_{f,2,p}} - L_{f,2,p})(t_{R0,2} - t_0) \quad (25)$$

where $L_{f,2,p}$ is given by eqn. 14 and w_{tot} for touching bands is given by eqn. 18.

The width of the plateau can be calculated by writing that the area of the second component band is equal to the area injected in the column, *i.e.*, that mass is conserved. Thus,

$$C_2^B w_{\text{plateau}} + \int_{t_B}^{t_{R0,2} + t_p} C_2 dt = L_{f,2}(t_{R0,2} - t_0)/b_2 \quad (26)$$

where C_2^B , t_B and C_2 are given by eqns. 28, 48 and 47, respectively, in ref. 14. The first term on the left-hand side of eqn. 26 is the area under the plateau, the second term, the area under the continuous profile, the right-hand side of eqn. 26 is the area $C_{0,2}t_p$ of the rectangular pulse of second component injected (see eqn. 42, ref. 14). The calculation gives

$$w_{\text{plateau}} = \left(\frac{1 + \frac{\alpha b_1 r_1}{b_2}}{\alpha - 1} \right) \left[L_{f,2} - \left(\frac{\frac{\alpha - 1}{\alpha}}{1 + \frac{b_1 r_1}{b_2}} \right)^2 \right] (t_{R0,2} - t_0) \quad (27)$$

Combination of eqns. 13, 18, 24, 25 and 27 gives

$$L_{f,2} = \frac{\left(\frac{\alpha - 1}{\alpha} \right)^2}{X(1 + X)^2} \left[X^2 + 3X + \frac{\alpha + 1}{\alpha} - \sqrt{\left(2X + \frac{\alpha + 1}{\alpha} \right)^2 + \frac{16 \left(\frac{k'_{0,2} + 1}{k'_{0,2}} \right)^2 (1 + X)^4}{N_0 \left(\frac{\alpha - 1}{\alpha} \right)^2}} \right] \quad (28)$$

with

$$X = \frac{q_{s,2}C_{0,1}}{\alpha q_{s,1}C_{0,2}} = \frac{L_{f,1}}{\alpha L_{f,2}} \quad (28a)$$

Eqn. 28 gives the loading factor for touching bands for values of the loading factor between those given by eqns. 3 and 14. The production rate is then calculated using eqn. 8.

Optimization of the experimental conditions

The optimization requires the use of a plate-height equation to relate the column efficiency to the mobile phase flow velocity. There are two possibilities at this stage. We can use a very simplified equation which assumes that the column is operated at a high reduced flow velocity. We have shown previously that the optimum flow velocity for maximum production is very high⁸. In this instance, we can also solve the optimization problem in close form and give a detailed, classical analysis of the effect of each parameter. Alternatively, we can use the classical Knox equation²⁴ and solve the problem numerically. These two approaches are discussed below.

Optimization with a simple plate-height equation

We know that the linear mobile phase velocity, u , is related to the column design and operating parameters by the following equation:

$$u = \frac{k_0 \Delta P d_p^2}{\eta L} \quad (29)$$

where k_0 is the column specific permeability (of the order of $1 \cdot 10^{-3}$), ΔP is the pressure drop between the column inlet and outlet, d_p is the average particle size of the packing material used, η is the mobile phase viscosity and L is the column length.

As the column is operated at a high reduced mobile phase velocity, we assume that the height equivalent to a theoretical plate height (HETP) of the column, is given by the simplified equation⁶:

$$h = Cv \quad (30)$$

where h is the reduced plate height ($h = H/d_p$) and v is the reduced velocity ($v = ud_p/D_m$). The limiting column efficiency at infinite sample dilution (i.e., L/H_{kin}) is therefore

$$N_0 = \frac{\eta D_m}{k_0 \Delta P C (d_p^2/L)^2} \quad (31)$$

As u and N_0 depend on the ratio d_p^2/L , not separately on the column length and the particle size, so does the production rate. As long as eqn. 30 is valid, in both the touching band and the overlapping band cases⁸, long columns packed with coarse

particles may give the same production rate as short columns packed with fine particles, if they have the same value of the ratio d_p^2/L . Provided that the column length can be adjusted easily, a variety of packing materials of different sizes can be used (within reason). The existence of an optimum particle size at constant length has been demonstrated experimentally in a recent paper⁵, in agreement with the results of a previous study¹⁹.

Depending on the value of the optimum sample size, we have two different cases. When the optimum loading factor is less than $L_{f,2,p}$, we should calculate it using eqn. 19. When the optimum loading factor is greater than $L_{f,2,p}$, eqn. 19 is no longer valid, and we must use eqn. 28. This condition is awkward to use. We show later that in practice eqn. 19 must be used to calculate the optimum loading factor for touching band when $L_{f,1}/\alpha L_{f,2}$ is lower than 0.4 (and the displacement effect is dominant) whereas eqn. 28 must be used when $L_{f,1}/\alpha L_{f,2}$ is larger than 0.4 (and the tag-along effect predominates). We discuss successively the cases when $L_{f,1}/\alpha L_{f,2}$ is smaller and larger than 0.4.

Optimization with a simple plate-height equation: displacement effect dominant

Combining eqns. 8, 19, 29 and 31 gives the production rate. Differentiation with respect to d_p^2/L permits the calculation of the optimum column configuration for a certain value of the pressure drop available. The equations could not be solved in close form, but the numerical calculation is easy.

If we assume that the band profiles are right triangles, as we have shown above that this assumption is reasonable when the relative retention α is close to 1 and the loading factor is small, we perform the same calculation as above, but using eqns. 20 and 21 instead of eqn. 19. Then it is easy to solve the equation giving the optimum value of the ratio d_p^2/L . We obtain

$$\left(\frac{d_p^2}{L}\right)_{\text{opt}} = \left(\frac{\alpha - 1}{\alpha}\right) \frac{\sqrt{(\alpha - 1/2)\sqrt{\alpha^2 - \alpha + 1/36} - (\alpha^2 - \alpha - 1/12)}}{4\left(\frac{k'_{0,2} + 1}{k'_{0,2}}\right)\sqrt{\frac{2k_0\Delta PC}{3D_m\eta}}} \quad (32)$$

Combination of eqns. 20, 31 and 32 gives the optimum value of the loading factor for touching bands:

$$L_{f,2,\text{opt}} = 3\left[\frac{(\alpha - 1)^2}{2\alpha}\right](\alpha - 1/2 - \sqrt{\alpha^2 - \alpha + 1/36}) \quad (33)$$

The value of the production rate is obtained by combining eqns. 21, 29, 32 and 33:

$$\frac{Pr_2}{(1 - \varepsilon)S} = \left[\frac{3q_{s,2}(\alpha - 1)^3}{8\alpha^2(1 + k'_{0,2})}\right]\sqrt{\frac{3k_0\Delta PD_m}{2\eta C}}(\alpha - 1/2 - \sqrt{\alpha^2 - \alpha + 1/36}) \times \sqrt{(\alpha - 1/2)\sqrt{\alpha^2 - \alpha + 1/36} - (\alpha^2 - \alpha - 1/12)} \quad (34)$$

Finally, if we ignore the competitive interactions between the two components of the

mixture and assume right triangle band profiles, we can use the value of the loading factor given by eqn. 22. A calculation similar to those made using eqn. 19 or 20 and described above gives in this case the following optimum value of the ratio d_p^2/L :

$$\left(\frac{d_p^2}{L}\right)_{opt} = \frac{\frac{\alpha - 1}{\alpha}}{4\left(\frac{k'_{0,2} + 1}{k'_{0,2}}\right)\sqrt{\frac{3k_0\Delta PC}{D_m\eta}}} \tag{35}$$

and the optimum value of the loading factor is

$$L_{f,2,opt} = \frac{1}{6}\left(\frac{\alpha - 1}{\alpha}\right)^2 \tag{36}$$

Inserting eqn. 35 into eqn. 31 gives the optimum column efficiency:

$$N_{0,opt} = 48\left(\frac{\alpha}{\alpha - 1}\right)^2\left(\frac{1 + k'_{0,2}}{k'_{0,2}}\right)^2 \tag{37}$$

and the maximum production rate is

$$\frac{Pr_2}{(1 - \epsilon)S} = \frac{q_{s,2}}{24(1 + k'_{0,2})}\left(\frac{\alpha - 1}{\alpha}\right)^3\sqrt{\frac{k_0\Delta PD_m}{3\eta C}} \tag{38}$$

which are equivalent to the equations derived by Knox and Pyper⁶.

Optimization with a simple plate-height equation: tag-along effect dominant

The optimum loading factor is now higher than the threshold ($L_{f,2,p}$) given by eqn. 14 and, as we show later, the ratio $L_{f,1}/(\alpha L_{f,2})$ is larger than 0.4. The value of the optimum loading factor for touching bands is given by eqn. 28. Eqns. 29, 30 and 31 still give the mobile phase flow velocity, the column plate height and the plate number of the column at infinite sample dilution, respectively. Combining eqns. 8, 28, 29 and 31 gives the production rate. Writing that the differential of the production rate with respect to d_p^2/L is zero gives the optimum value of this ratio:

$$\left(\frac{d_p^2}{L}\right)_{opt} = \frac{\frac{\alpha - 1}{\alpha}\left(X^2 + 3X + \frac{\alpha + 1}{\alpha}\right)}{8\left(\frac{k'_{0,2} + 1}{k'_{0,2}}\right)(1 + X)^2\sqrt{\frac{k_0\Delta PC}{D_m\eta}}} \times$$

$$\sqrt{\frac{1}{2} - \frac{2\left(2X + \frac{\alpha + 1}{\alpha}\right)^2}{\left(X^2 + 3X + \frac{\alpha + 1}{\alpha}\right)^2}} + \sqrt{\frac{1}{4} + \frac{2\left(2X + \frac{\alpha + 1}{\alpha}\right)^2}{\left(X^2 + 3X + \frac{\alpha + 1}{\alpha}\right)^2}} \tag{39}$$

where X is given by eqn. 28a. When X becomes large, the square-root term on the right-hand side of eqn. 39 is almost equal to unity and the equation simplifies considerably:

$$\left(\frac{d_p^2}{L}\right)_{\text{opt}} = \frac{\frac{\alpha - 1}{\alpha}}{8 \left(\frac{k'_{0,2} + 1}{k'_{0,2}}\right) \sqrt{\frac{k_0 \Delta PC}{D_m \eta}}} \quad (39a)$$

This result is now independent of the feed composition.

Inserting the optimum value of d_p^2/L given by eqn. 39 into eqn. 31 gives the optimum limiting column efficiency (at negligible sample size). Combination of these equations with eqn. 28 gives the optimum sample size:

$$L_{f,2,\text{opt}} = \left(\frac{\alpha - 1}{\alpha}\right)^2 \frac{X^2 + 3X + \frac{\alpha + 1}{\alpha}}{4X(1 + X)^2} \left[3 - \sqrt{1 + 8 \left(\frac{2X + \frac{\alpha + 1}{\alpha}}{X^2 + 3X + \frac{\alpha + 1}{\alpha}}\right)^2} \right] \quad (40)$$

In the case when X is large, the optimum column efficiency and loading factor are

$$N_{0,\text{opt}} = 64 \left(\frac{\alpha}{\alpha - 1}\right)^2 \left(\frac{1 + k'_{0,2}}{k'_{0,2}}\right) \quad (40a)$$

$$L_{f,2,\text{opt}} = \frac{1}{2X} \left(\frac{\alpha - 1}{\alpha}\right)^2 \quad (40b)$$

This value is equal to that predicted by the non-competitive model for $X = 3$, lower when X is larger than 3 and larger when X is smaller than 3.

Finally, combination of eqns. 8, 29, 39 and 40 gives the maximum production rate which can be achieved with a certain value of the inlet pressure.

Optimization with the Knox plate-height equation

More accurate results could be obtained using the classical Knox plate-height equation²⁴:

$$h = \frac{L}{d_p N_0} = \frac{B}{v} + Av^{1/3} + Cv \quad (41)$$

It is easy to calculate numerically the optimum values of the column length and particle size, flow velocity and sample size for any problem, but a close-form solution cannot be derived.

RESULTS AND DISCUSSION

We have performed a number of calculations in the two cases when either the displacement or the tag-along effect predominates, using the simplified and the more accurate plate-height equations, and we report the results in Tables I–IV, discussed in the following sections. We then compare these results with previously published data.

Optimization with a simple plate-height equation: displacement effect dominant

In this instance, the optimum sample size is given by eqn. 19. We see that, as assumed by Knox and Pyper⁶, the optimum values of the sample size and of the limiting column efficiency do not depend on the composition of the feed.

Table I compares the results derived from our present approach (eqns. 8, 19, 29 and 31) and those calculated using the model derived by Knox and Pyper⁶ (eqns. 35–38). The optimum values of the limiting column efficiency, of the value of the ratio d_p^2/L , of the mobile phase flow velocity and of the loading factor are given together with the maximum production rate that can be achieved. A maximum value of the inlet pressure of 200 atm has been assumed. The comparison between the two sets of results shows little difference between the optimum values of the ratio d_p^2/L , usually 10–20% larger with the Knox and Pyper assumptions than with our more exact approach. The difference in the values of the optimum limiting column efficiency is greater; our approach requires a column efficiency roughly 35% larger than that of Knox and Pyper⁶. Correspondingly, the optimum value of the mobile phase velocity is lower with

TABLE I

OPTIMUM VALUE OF THE RATIO d_p^2/L AND MAXIMUM PRODUCTION RATE UNDER TOUCHING BAND CONDITIONS

$$\text{First case: } \frac{q_{s,2}C_{0,1}}{\alpha q_{s,1}C_{0,2}} = \frac{L_{f,1}}{\alpha L_{f,2}} < 0.4. \quad h = Cv.$$

α	N_0	d_p^2/L $\times 10^8$ (cm)	u (cm/s)	$L_{f,2}$ (%)	$Pr_2/(1-\varepsilon)S$ [$\mu\text{mol}/(\text{cm}^2 \text{ s})$]	Pr_2^a/Pr_2^b
1.1 ^a	10500	2.2	0.44	0.41	2.72	
1.1 ^b	7700	2.55	0.51	0.138	1.06	2.57
1.2 ^a	3100	4.0	0.8	1.37	15.2	
1.2 ^b	2260	4.7	0.94	0.46	6.1	2.51
1.3 ^a	1600	5.55	1.1	2.62	37.4	
1.3 ^b	1150	6.6	1.3	0.89	15.0	2.30
1.5 ^a	760	8.1	1.6	5.41	97.6	
1.5 ^b	550	9.55	1.9	1.88	39.8	2.45
1.7 ^a	530	9.75	1.95	8.45	161.6	
1.7 ^b	350	12.0	2.4	2.83	66.8	2.42

^a Calculated with the simple HETP equation $h = Cv$, using the model described in this work. Conditions: $C = 0.1$; $k'_{0,1} = 6$, $\Delta P = 200$ atm, $D_m = 1 \cdot 10^{-5}$ cm²/s, $\eta = 1$ cP, $q_{s,2} = 10$. The cycle time is given by $t_c = t_{R0,2} - t_0$.

^b Calculated with the same plate-height equation, using the model of Knox and Pyper⁶, which ignores the competition and assumes right triangular band profiles.

our results, by *ca.* 15–20%. However, the major difference between the two sets of results is the loading factor. Knox and Pyper neglect the band interaction. Because of the importance of the displacement effect, a nearly three times larger sample size is needed to achieve touching bands when we take into account the competitive interaction between the two components of the mixture (Table I). The end result is a production rate which is *ca.* 2.5 times larger than that predicted by the non-competitive model⁶.

If we ignore the competition between the mixture components, the optimum values of d_0^2/L , N_0 and $L_{f,2,opt}$ and the maximum production rate are proportional to $(\alpha - 1)/\alpha$, $[(\alpha - 1)/\alpha]^{-2}$, $[(\alpha - 1)/\alpha]^2$, and $[(\alpha - 1)/\alpha]^3$, respectively (see eqns. 35, 36, 37 and 38). When we take the competitive interaction between the two components into account, the exact dependence of these parameters on the relative retention is more complicated. The analysis of the data in Table I shows, however, that in practice the optimum sample size is given by

$$L_{f,2} = 0.5 \left(\frac{\alpha - 1}{\alpha} \right)^2 \quad (42)$$

instead of $L_{f,2} = 0.166 [(\alpha - 1)/\alpha]^2$, as predicted by the approximate eqn. 37⁶. This important increase is not surprising. The first component band is pushed forward by the second band. Eqn. 17 shows the reduction in the retention time of the rear of that band, proportional to the amount of second component injected. Accordingly, in order to reach touching band conditions, we need a wider second component band, hence a larger sample size.

Further, we see that the limiting efficiency required, N_0 , is equal to four times the efficiency, N^* , necessary for the separation of the mixture with a resolution of 1 under linear (*i.e.*, analytical) conditions, instead of three times as predicted by the non-competitive model of Knox and Pyper⁶. In other words, the column should afford the separation of a very small sample with a resolution of 2.0, not 1.7.

As the optimum sample size for touching bands under a dominant displacement effect is given by eqn. 42, the threshold condition, $L_{f,2} < L_{f,2,p}$ where $L_{f,2,p}$ is given by eqn. 14, can be rewritten as

$$L_{f,2,opt} \leq L_{f,2,p} \quad (43a)$$

or, by combining eqns. 14 and 42:

$$0.5 \left(\frac{\alpha - 1}{\alpha} \right)^2 \leq \frac{\left(\frac{\alpha - 1}{\alpha} \right)^2}{\left(1 + \frac{L_{f,1}}{\alpha L_{f,2}} \right)^2} \quad (43b)$$

which gives

$$\frac{L_{f,1}}{\alpha L_{f,2}} \leq \sqrt{2} - 1 \quad (43c)$$

which is the condition mentioned above ($L_{f,1}/\alpha L_{f,2} < 0.4$). When condition 43c is not satisfied, eqn. 28 or 40 should be used instead of eqn. 19 for the optimization.

Optimization with a simple plate-height equation: tag-along effect dominant

In this case, the optimum sample size is not given by eqn. 19 but by eqn. 40. The tag-along effect dominates, so the second component band is spread over a wide range of retention times. In the ideal model approximation it exhibits a plateau whose width depends on the loading factor (see eqn. 27). The second component individual elution profile is different from the profile observed for the same amount of pure second component. Especially noteworthy is the fact that, in contrast to what happens in the previous instance, the optimum conditions depend on the feed composition.

Calculations were performed using our approach (eqns. 8, 29, 31, 39 and 40) and that of Knox and Pyper⁶ for binary mixtures of different compositions and the same relative retention, $\alpha = 1.20$. The results are reported in Table II. As in the previous instance, the differences between the optimum values of the limiting column efficiency, the ratio d_p^2/L and the mobile phase flow velocity are relatively small. Compared with the optimum values predicted by Knox and Pyper, the optimum values of the column

TABLE II

OPTIMUM VALUE OF d_p^2/L AND MAXIMUM PRODUCTION RATE UNDER TOUCHING BAND CONDITIONS

$$\text{Second case: } \frac{q_{s,2}C_{0,1}}{\alpha q_{s,1}C_{0,2}} = \frac{L_{f,1}}{\alpha L_{f,2}} > 0.4^a. \quad h = Cv.$$

Feed composition	N_0	d_p^2/L $\times 10^8$ (cm)	u (cm/s)	$L_{f,2}$ (%)	$Pr_2/(1-\epsilon)S$ [$\mu\text{mol}/(\text{cm}^2 \text{ s})$]	Pr_2^a/Pr_2^b
1:1 ^a	2100	4.9	0.98	0.98	13.2	2.17
3:1 ^a	2000	5.0	1.0	0.49	6.8	1.12
9:1 ^a	2470	4.5	0.9	0.189	2.36	0.39
For comparison:						
Conditions in ref. 6 ^b	2260	4.7	0.94	0.46	6.08	n.a.
This work, with $L_{f,1}/\alpha L_{f,2} < 0.4^a$	3100	4.0	0.8	1.37	15.2	2.50
Knox and Pyper column with: $L_{f,1}/\alpha L_{f,2} < 0.4^c$	2260	4.7	0.94			Pr_2^a/Pr_2^c
				1.14	14.8	1.03
1:1 ^c				1.0	13.2	1.0
3:1 ^c				0.51	6.76	1.00
9:1 ^c				0.18	2.36	1.00

^a Values calculated with the Langmuir competitive adsorption model, using the simplified simple HETP equation $h = Cv$. Same conditions as for Table I, except $\alpha = 1.20$.

^b Values calculated using the non-competitive model and the simplified HETP equation and assuming right triangular band profiles. The same results are obtained, regardless of the mixture composition.

^c Values calculated for a column designed with the approach suggested by Knox and Pyper⁶ ($d_p^2/L = 4.7$) and using the same velocity ($u = 0.94$) but the much larger sample size predicted by the competitive model.

efficiency range between -10 and $+40\%$, those of the ratio d_p^2/L between -15 and $+7\%$ and those of the mobile phase velocity between -15 and $+7\%$. On the other hand, the loading factor required to achieve touching bands decreases rapidly with decreasing concentration of the second component. The sample sizes and production rates predicted by the two methods are nearly equal for a 3:1 mixture. For mixtures richer in the second component, the sample size and the production rate are higher than those predicted by the Knox and Pyper model, because of the displacement effect, whereas for mixtures richer in the first component, the production rate is lower, because of the tag-along effect.

The optimization of the column efficiency, of its conformation and of the mobile phase velocity are not very critical and nearly identical results are obtained with the columns which have either the optimum characteristics calculated in this work or those calculated after Knox and Pyper, provided that they are both operated with the optimum sample size as calculated in this work. This is illustrated by the comparison between the data at the top and bottom of Table II.

Optimization with the Knox plate-height equation

Numerical calculations were carried out using the Knox plate-height equation with conventional values of the coefficients, $A = 1$, $B = 2$ and $C = 0.1$, which corresponds to an average quality packing material packed properly into a chromatographic column (minimum reduced plate height 2.40; optimum analytical reduced velocity 2.7). The optimization procedure is as follows. Values of the column length and particle size are selected and the mobile phase velocity is calculated (eqn. 29). The

TABLE III

OPTIMUM VALUE OF COLUMN LENGTH AND MAXIMUM PRODUCTION RATE UNDER TOUCHING BAND CONDITIONS

$$\text{First case: } \frac{q_{s,2}C_{0,1}}{\alpha q_{s,1}C_{0,2}} = \frac{L_{f,1}}{\alpha L_{f,2}} < 0.4. \quad h = 2/v + v^{0.33} + 0.1v.$$

α	N_0	L (cm)	v	$L_{f,2}$ (%)	$Pr_2/(1 - \epsilon)S$ [$\mu\text{mol}/(\text{cm}^2 \text{ s})$]	Pr_2^a/Pr_2^b
1.1 ^a	8880	59.	34	0.375	1.93	
1.1 ^b	6850	50.5	39.5	0.129	0.775	2.49
1.2 ^a	2750	29.6	67.5	1.28	12.0	
1.2 ^b	2050	25.2	79.0	0.443	4.88	2.46
1.3 ^a	1480	20.9	96	2.51	30.8	
1.3 ^b	1070	17.4	115	0.85	12.6	2.44
1.5 ^a	700	13.8	145	51.8	83.5	
1.5 ^b	500	11.5	174	1.79	34.6	2.41
1.7 ^a	450	10.9	183	7.84	141.1	
1.7 ^b	320	9.0	222	2.72	59.4	2.38

^a Values calculated using the Langmuir competitive adsorption model and the general Knox HETP equation. Same parameters as for Table I except particle size $d_p = 10 \mu\text{m}$. The cycle time is $t_c = t_{R0,2} - t_0$.

^b Values calculated using the non-competitive model, assuming that the band profiles are right triangles and using the general Knox HETP equation.

TABLE IV

OPTIMUM VALUE OF THE COLUMN LENGTH AND MAXIMUM PRODUCTION RATE UNDER TOUCHING BAND CONDITIONS

$$\text{Second case: } \frac{q_{s,2}C_{0,1}}{\alpha q_{s,1}C_{0,2}} = \frac{L_{f,1}}{\alpha L_{f,2}} > 0.4. \quad h = 2/v + v^{0.33} + 0.1v.$$

Feed composition	N_0	L (cm)	v	$L_{f,2}$ (%)	$Pr_2/(1-\varepsilon)S$ [$\mu\text{mol}/(\text{cm}^2 \text{ s})$]
1:1 ^a	1920	24.2	82.5	0.94	10.7
3:1 ^a	1810	23.4	77.0	0.46	5.5
9:1 ^a	2180	26.0	85.5	0.18	1.9
For comparison:					
$L_{f,1}/\alpha L_{f,2}^* < 0.4$ (this work)	2750	29.6	67.5	1.28	12.0
Knox ^b	2060	25.2	79.0	0.44	4.9

^a Values calculated with the Langmuir competitive adsorption model, using the general Knox HETP equation ($h = 2/v + v^{0.33} + 0.1v$). Same conditions as for Table II, except particle size $d_p = 10 \mu\text{m}$.

^b Values calculated with the non-competitive model, assuming right triangle band profiles and using the general Knox HETP equation. The values are constant, regardless of the feed composition.

reduced plate height is then derived (eqn. 41) and the plate number introduced in eqn. 19 or in eqn. 28, depending on whether $L_{f,1}/\alpha L_{f,2}$ is smaller or larger than 0.4. This gives the sample size. Inserting the sample size and the velocity in eqn. 8 gives the production rate. The calculation is repeated for successive values of the column length at constant particle size to determine the optimum column length giving the maximum production rate. Alternately, the particle size could be varied at constant column length or the calculation could be made for the columns currently available in the laboratory to find which will give the highest production rate. Although this procedure of optimization is generally not correct, it gives a result which is satisfactory in practice, because particles are available in only a small number of sizes and the variation of the production rate near its optimum is fairly flat. The same procedure is followed for the calculation of the optimum conditions within the frame of the Knox and Pyper model, but eqn. 22 is used to calculate the sample size.

Tables III and IV summarize the data obtained. Optimum values of the limiting column efficiency, the column length, the reduced flow velocity and the loading factors are given in Table III for different values of the relative retention between 1.10 and 1.70 for binary mixtures having a composition such that $L_{f,1}/(\alpha L_{f,2})$ is smaller than 0.40. We have assumed the particle size to be $10 \mu\text{m}$. Also given in Table III is the maximum production rate. Comparison between the exact values and those derived from the Knox and Pyper model again shows fairly small differences between the optimum values of the limiting column efficiency (30–40%), the column length (10–20%) and the reduced velocity (15–20%). The loading factor, on the other hand, is 2.5–3 times larger than that predicted by the Knox and Pyper model and the production rate is about 2.20 times higher. As expected, comparison between the results in Tables I and III shows general agreement for the optimum limiting column efficiency and loading factor and for the maximum production rate at high values of α when the mobile phase velocity is very high. The difference increases, however, with decreasing values of α and

TABLE V

OPTIMUM VALUE OF d_p^2/L AND MAXIMUM VALUE OF THE PRODUCTION RATE UNDER OVERLAPPING BAND CONDITIONS^a FOR DIFFERENT FEED COMPOSITIONS

Feed composition	N_0	d_p^2/L $\times 10^8$ (cm)	u (cm/s)	$L_{f,2}^b$ (%)	$Pr_2/(1 - \epsilon)S$ [$\mu\text{mol}/(\text{cm}^2 \text{ s})$]
1:9	380	11.5	2.3	4.8	91.6
1:3	490	10.1	2.0	3.05	52.0
1:1	630	8.9	1.8	1.822	26.6
3:1	910	7.4	1.5	0.875	10.2
9:1	1200	6.5	1.3	0.343	3.1
Knox ^c	2260	4.7	0.94	0.46	6.1

^a Values calculated with the competitive model and the simple HETP equation $h = Cv$, with $C = 0.1$. Same conditions as for Table II except required product purity, 99%. $\alpha = 1.2$; $q_{s,2} = q_{s,1} = 10$. In all instances the recovery yield is about 60%.

^b Values calculated from the equation $L_{f,2} = \frac{1}{(1 + b_1 r_1 / b_2)} \left(\frac{1 - 1/\alpha}{1 - x} \right)^2$, where $x = \left(\frac{1 - Pu_2}{Pu_2 \alpha r_1} \right)^{1/2}$, Pu_2 being the required purity of the product. This equation is derived in ref. 7.

^c Values calculated with the non-competitive model, assuming right triangle band profiles and a simple HETP equation⁶. The results are the same, regardless of the feed composition.

for $\alpha = 1.10$, the production rate predicted is 40% less with the correct plate-height equation (eqn. 41) than with the simplified eqn. 30. A significant difference between the results obtained with the two plate height equations is expected in this case: α is low, a relatively high efficiency is needed, leading to a moderate reduced velocity (Table III). Obviously, the difference between eqns. 30 and 41 is larger at low flow velocities.

Table IV reports similar data calculated for binary mixtures of different compositions, such that $L_{f,1}/(\alpha L_{f,2})$ is larger than 0.4, for different compositions and $\alpha = 1.20$. As with the simplified plate-height equation, the production rate predicted by the Knox and Pyper model is lower than that given by the more exact approach when the relative concentration of the second component exceeds about 0.25 and is too large when the relative concentration of the second component is smaller than 0.25. The production rates are about 20% lower than those calculated with the simplified plate-height equation and reported in Table II, because of the influence of the two neglected terms of the plate-height equation.

Comparison with previous results

Depending on the required degree of purity of the prepared products and the desired recovery yield, there are two extreme strategies and a variety of possible compromises between them^{2,5}. The first strategy is to achieve a total recovery yield (ca. 100%). In this instance the touching band approach as discussed in this paper must be used. The second strategy consists into looking for the maximum possible production rate without concern for the value of the recovery yield. We have previously discussed this strategy which can give recovery yields as low as 60%^{8,12}. For example, data have been calculated and are reported in Table V illustrating the advantages and inconveniences of this strategy. A purity of 99% of the product was required and no condition was placed on the recovery yield.

TABLE VI

OPTIMUM VALUE OF d_p^2/L AND MAXIMUM PRODUCTION RATE UNDER OVERLAPPING BAND CONDITIONS^a FOR DIFFERENT VALUES OF α

α	N_0	d_p^2/L $\times 10^8$ (cm)	u (cm/s)	$L_{f,2}$ (%) ^b	$Pr_2/(1-\epsilon)S$ [$\mu\text{mol}/(\text{cm}^2 \text{ s})$]
1.1	1150	6.6	1.3	1.47	17.9
1.2	380	11.5	2.3	4.80	91.6
1.3	240	14.4	2.9	9.0	205.2
1.5	145	18.5	3.7	18.0	461.2
1.7	105	21.8	4.4	26.6	679.2

^a Values calculated with the competitive model, using the simple HETP equation $h = Cv$, with $C = 0.1$. Same conditions as for Table I, except required product purity, 99%, relative feed composition, 1:9, $q_{s,2} = q_{s,1} = 10$. In all instances the recovery yield is about 60%.

^b Values calculated from the equation $L_{f,2} = \frac{1}{(1 + b_1 r_1 / b_2)} \left(\frac{1 - 1/\alpha}{1 - x} \right)^2$, where $x = \left(\frac{1 - Pu_2}{Pu_2 \alpha r_1} \right)^{1/2}$.

Comparison between the results in Tables II and V shows that when the relative concentration of the first component is small, the production rate is much greater when the overlapping-band approach is used, the column is overloaded and the individual elution bands exhibit a degree of overlap than when the touching band strategy is used. As shown in Table V, for a 1:9 mixture the production rate is six times larger with overlapping bands than with touching bands (and fifteen times larger than that predicted by the model of Knox and Pyper⁶). The ratio of the production rate with overlapping bands to the production rate with touching bands decreases with decreasing relative concentration of the second component. It is still three times for a 1:3 mixture, two times for a 1:1 mixture, 1.5 times for a 3:1 mixture and only 1.3 times for a 9:1 mixture. In all instances the yield is *ca.* 60%.

The comparative advantage of using the first or the second strategy depends very much on the composition of the feed. If the second compound is in large excess, the

TABLE VII

OPTIMUM VALUE OF THE COLUMN LENGTH AND MAXIMUM PRODUCTION RATE UNDER OVERLAPPING BAND CONDITIONS^a FOR DIFFERENT VALUES OF α

α	N_0	L (cm)	v	$L_{f,2}$ (%) ^b	$Pr_2/(1-\epsilon)S$ [$\mu\text{mol}/(\text{cm}^2 \text{ s})$]
1.1	1000	16	120	1.47	15.0
1.2	360	9.6	210	4.80	81
1.3	220	7.3	275	9.0	186
1.5	130	5.6	357	18.0	425
1.7	95	4.7	425	26.6	630

^a Values calculated with the competitive model, using the Knox general HETP equation, $h = 2/v + v^{0.33} + 0.1v$, and the same conditions as for Table VI, except the particle size, $d_p = 10 \mu\text{m}$. In all instances the recovery yield is about 60%.

^b Values calculated from the equation $L_{f,2} = \frac{1}{(1 + b_1 r_1 / b_2)} \left(\frac{1 - 1/\alpha}{1 - x} \right)^2$, where $x = \left(\frac{1 - Pu_2}{Pu_2 \alpha r_1} \right)^{1/2}$.

overlapping band approach permits, compared with the touching band approach, a much higher production rate, for which the penalty is a decrease in yield. This advantage, however, decreases rapidly with increasing relative concentration of the first compound. Then it is reasonable to adopt an intermediate approach, where the degree of band overlapping accepted decreases with decreasing concentration of the second component, with a parallel increase in the recovery yield.

Finally, Tables VI and VII give the optimal experimental conditions in the case of overlapping band⁸ for a series of 1/9 mixtures with different values of the selectivity, α , using either the simple plate height equation (eqn. 30) or the more general Knox plate height equation (eqn. 41), respectively. Comparison of these two sets of data with the data in Tables I and III show that when the ratio of the component concentrations in the feed, $C_{0,1}/C_{0,2}$, is small, the overlapping-band strategy has the following advantages over the touching-band strategy: a lower optimum column efficiency, a larger optimum ratio d_p^2/L , a larger optimum reduced velocity, and a higher production rate of the second component. The production rate increases rapidly with increasing value of the selectivity, nearly in proportion to $\left(\frac{\alpha-1}{\alpha}\right)^3$, in both the touching-band case (Table III) and the overlapping-band case (Table VI).

CONCLUSION

As our previous discussion of the overlapping band strategy has shown, the maximum production rate with the touching band strategy increases indefinitely with increasing permissible column pressure drop, provided that the column of optimum geometry is used. For a given column, in contrast, there is an optimum pressure drop as there is an optimum flow velocity. The maximum production rate is usually achieved at very high values of the reduced flow velocity, much in excess of that which corresponds to the maximum column efficiency^{8,12}. We note, however, that the optimum velocity is lower for touching bands than for overlapping bands.

For values of the ratio $L_{f,1}/(\alpha L_{f,2})$ less than 0.4, the optimum sample size is independent of the feed composition and the sample size should be such that $L_{f,2} = 0.5[(\alpha - 1)/\alpha]^2$, *i.e.*, approximately three times larger than that predicted by the simple model of Knox and Pyper⁶, which neglects the interaction between the two components of the mixture during the separation. In this instance, the analytical resolution under the conditions selected for the separation should be 2, larger than the value of 1.7 suggested by Knox and Pyper. For values of the ratio $L_{f,1}/(\alpha L_{f,2})$ between 0.4 and 3, the sample size depends markedly on the feed composition but is still larger than the value predicted by the Knox and Pyper model. Finally, for values of $L_{f,1}/(\alpha L_{f,2})$ larger than 3, the sample size should be less than that predicted by Knox and Pyper. It depends strongly on the composition of the mixture and decreases rapidly with increasing concentration of the first component. In preparative chromatography a system eluting the main component first and the impurities later should be avoided as much as possible.

The use of the overlapping strategy entails a recovery yield of the order of 60% but permits a considerable increase in production rate for both components when the displacement effect predominates, a result which has already been observed experimentally and is well documented^{4,19,20}. The gain in production rate decreases rapidly

with decreasing concentration of the second component in a binary mixture. The overlapping band strategy has no special advantage for the production rate of the second component when this compound is the minor one.

The quantitative accuracy of the present results depends on the extent to which the equilibrium isotherms of the components of the mixture are approximated by the competitive Langmuir model. On the other hand, the accuracy of our results is considerably better than those given by models, computerized or not, which do not take the competitive interactions of the mixture components into account properly⁹. As shown above, the production rate predicted by these models can be three times too low or too high, depending on the composition of the feed.

SYMBOLS

A	Coefficient in the Knox plate-height equation
a_i	Numerical coefficient in the competitive Langmuir isotherm
B	Coefficient in the Knox plate-height equation
b_i	Numerical coefficient in the competitive Langmuir isotherm
C	Coefficient of mass-transfer resistance in the plate-height equation
C_i	Mobile phase concentration of component i
C_{Max}	Maximum concentration of a compound in the eluent
$C_{0,i}$	Concentration of component i in the feed
C_2	Concentration of the second component during its elution. In eqn. 26, concentration of the second component in the last zone, where it is pure
C_2^B	Concentration of the elution plateau of the second component
D_m	Molecular diffusion coefficient of the mixture components
d_p	Average particle size of the packing material used
H_{kin}	HETP of the elution profile of a Dirac pulse under linear conditions
H_{th}	HETP of the elution profile of a Dirac pulse predicted by the ideal model under non-linear conditions
H_{tot}	HETP of the elution profile of a Dirac pulse under non-linear conditions
h	Reduced plate height ($h = H/d_p$)
k_0	Column specific permeability (of the order of $1 \cdot 10^{-3}$)
$k'_{0,i}$	Column retention factor or capacity factor of component i under linear conditions; $k'_{0,i} = (t_{R0,i} - t_0)/t_0$
L	Column length
$L_{f,i}$	Loading factor for component i
$L_{f,2,p}$	Loading factor for the second component corresponding to the elution of a band profile for which the tag-along effect has just disappeared
N_0	Column plate number under linear conditions (in practice, for a very small size sample)
opt	The subscript opt represents the optimum value of a parameter
ΔP	Pressure drop between the column inlet and outlet
Pr_i	Production rate of component i
q_i	Amount of compound i adsorbed at equilibrium with the mobile phase
$q_{s,i}$	Specific column saturation capacity for component i ; $q_{s,i} = a_i/b_i$
r_1	Positive root of the characteristic equation of the problem (ref. 8, eqn. II-3). In practice, $r_1 \approx C_{0,1}/C_{0,2}$

S	Geometrical column cross-sectional area
T	Total feed throughput
t_B	Elution time of the end of the plateau of the second component
t_c	Cycle time
$t_{e,1}$	Elution time of the rear of the first component band
t_0	Hold-up time of the column
t_p	Width of the injection pulse
$t_{R,i}$	Retention time of the front of component i
$t_{R0,i}$	Limiting retention time of component i at infinite dilution
u	Mobile phase linear velocity
w_{kin}	Baseline band width of the elution profile of a Dirac pulse under linear conditions
$w_{plateau}$	Width of the elution plateau of the second component; $w_{plateau} = t_{R,2} - t_B$
w_{th}	Baseline band width of the elution profile of a Dirac pulse predicted by the ideal model under non-linear conditions
w_{tot}	Total baseline band width of the elution profile of a Dirac pulse under non-linear conditions
X	Dummy parameter in eqns. 28 and 39–40
α	Relative retention of the two compounds ($\alpha = a_2/a_1$)
ε	Packing porosity
η	Mobile phase viscosity
v	Reduced velocity ($v = ud_p/D_m$)
σ_{kin}^2	Variance contribution of the mass-transfer kinetics and the axial dispersion
σ_{th}^2	Variance contribution due to the non-linear behavior of the isotherm (thermodynamics)
σ_{tot}^2	Variance of the elution profile

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