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Calculation of programmed temperature gas chromatography characteristics from isothermal data V. Prediction of peak asymmetries and resolution characteristics

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Abstract

A previously described procedure for the calculation of peak widths in programmed temperature gas chromatograms from corresponding isothermal widths has been adapted to the analogous calculation of peak asymmetries. Comparison with experimental data is satisfactory. Composite predictions of retention times, widths and asymmetries are then used to predict (a) the forms of complete chromatograms comprising close peaks, (b) resolution characteristics of such chromatograms and (c) optimum programmed temperature conditions.

1. Introduction

Previous papers in this series [1–4] have been concerned with (a) prediction of retention times, elution temperatures, retention indices, equivalent temperatures and peak widths in programmed-temperature gas chromatography (PTGC) from experimental retention times obtained under isothermal conditions and (b) comparison of theory and experiment, the latter restricted to the single linear temperature–time ramp (although the theory presented covered more general situations). The present paper extends predictions and experimental comparisons to PTGC peak asymmetry. The predictive procedure used is quite similar to one successfully used previously [4] for peak width. This procedure was based upon the subtraction of two calculated "retention times", one for a point on the ascending section of a peak, e.g. at half height, and the other for the point at the same height on the descending section. The extension to asymmetry was simply to include the predicted peak maximum, i.e. the true retention time, as a third time. If now one assumes a two-parameter function for peak shape, the composite calculation of retention times, peak widths and peak asymmetries then allows the prediction of overall forms of mixture chromatograms, in particular those where components have similar retention times. Associated with this prediction is the

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calculation of various measures of peak-pair and overall resolution. Variation of PTGC conditions (here only initial column temperature and rate of temperature increase) then allows prediction of optimum resolution conditions, subject to a not excessive time for the chromatographic experiment. Similar optimizations have been described by other authors previously [5].

2. Peak asymmetry and shape

Chromatographic peaks have commonly been treated as either gaussian (for which there is reasonable theoretical support) or isosceles-triangular in shape. Such shapes are symmetrical and each is fully described by two parameters, such as (maximum) height and width at half-height (or, in place of the latter, standard deviation). In practice however, there always exists some degree of asymmetry —either fronting or tailing— in a peak and further parameters are then necessary to describe the shape. Asymmetry is known to arise from several causes, e.g. proportionality failure of mobile and stationary phase concentrations, column overloading, or slow injection. In the present work, it has been assumed that instrumental/operator effects on width and asymmetry are minimized and essentially constant, i.e. that band shape is solely an analyte/column/temperature characteristic. A commonly used function for an asymmetric peak is the exponentially modified gaussian [6]. This is a convolution integral, characterized by three parameters: height, standard deviation and exponential decay constant. This means that a single asymmetry parameter is sufficient to define deviations from symmetry and this has been assumed in the present work. However, a simpler three-parameter function has been used in simulated chromatograms, viz. a bi-gaussian. This uses one gaussian for the ascent and a second one for the descent. Its disadvantage is a discontinuity of curvature at the peak maximum, but generally this has not been observable in our simulations. For the purpose of the resolution characteristics considered here, the choice of shape function is in fact of no relevance.

Peak asymmetry A, at a particular fractional height, is conventionally defined as the ratio b/a, where b is the time difference between the fractional height point on the trailing edge and the peak maximum (t_R) and a is the time difference between the peak maximum (t_R) and the corresponding point on the leading edge. (For a bi-gaussian, A should be independent of fractional height.) Width w is, of course, equal to $a+b$ so that (w,A) and (a,b) statements are readily interconvertible. A Pascal programme was written for use on an Amdahl 5890 mainframe to calculate PTGC retention times, peak widths and asymmetries from corresponding isothermal data together with information on the dependence of column dead time upon temperature. The latter was either (a) in the form of data pairs to be fitted to a least squares linear relationship of dead time to thermodynamic temperature or its square root or (b) as previously determined coefficients for one of these relationships; for the predictions reported in the present paper, only the square root option was used. As previously indicated, the calculation was essentially that of three "retention times" for each compound, the true retention time and the two fractional height times, one on each edge. As in the previous programme for the calculation of width [4], there was a choice of polynomial order for the logarithm of capacity factor ($\ln k'$) against the reciprocal of thermodynamic temperature (T^{-1}). In the previous programme, the value was calculated for all orders up to 4 *within the single run* but, for the present programme, a single order (up to three) was selected in the data file. This programme handled up to twenty compounds and all single temperature–time ramp combinations of initial temperatures (333.2 to 393.2 K in 10.0 K steps) and heating rates (1.0 to 15.0 K min^{-1} in 1.0 K min^{-1} steps) within one run and also calculated resolution characteristics (see below) for a chromatogram of a mixture of the compounds. The chosen characteristics are independent of relative abundance and detector response factors.

3. Comparison of calculated and observed asymmetries

The experimental procedure for obtaining chromatograms using capillary columns under both isothermal and programmed temperature conditions has been previously described [2,4], as has a description of the computational procedure for displaying replicate details of individual peaks (isothermal or programmed temperature) and associated width information at various fractional heights [4]. The FORTRAN programme used for this purpose also computes and displays half-height asymmetry for the replicate runs (see Fig. 2 of ref. [4] for an example). Average values may then be compared with predictions. Table 1 presents a typical comparison for methyl carboxylate esters with an SE-30 column, a single temperature–time ramp starting at 373.2 K, and employing both first- and second-order polynomials for ln k' against T^{-1}. Isothermal data for 393.2 to 433.2 K (in 10.0 K steps) were used in the predictions. Quality of agreement varies but is generally satisfactory, bearing in mind the given standard errors of the observations and assumed —but not calculated in the present work— errors of similar magnitude for the predictions.

4. Resolution characteristics

The ideal chromatogram is one for which individual peaks of analytical interest are cleanly separated from one another, i.e. are well resolved. The optimization of experimental conditions to further this objective in the various kinds of chromatographic experiment has been extensively covered previously. A summary (to 1986) has been provided by Schoenmakers [7]. Quantitative resolution characteristics are of two kinds: (a) for adjacent pairs of peaks —"elementary criteria", and (b) for a complete chromatogram. Some of these criteria (i) depend upon relative peak area [i.e. on the product of relative quantity (mass or amount of substance) and

Table 1
Comparison of experiment and half-height asymmetry predictions

Compound	Asymmetry					
	Heating rate = 5 K min^{-1}		Heating rate = 10 K min^{-1}		Heating rate = 15 K min^{-1}	
	Predicted[a]	Observed[b]	Predicted[a]	Observed[b]	Predicted[a]	Observed[b]
Methyl octanoate	0.73 0.88	0.85 (0.10)	0.83 0.90	0.93 (0.07)	0.89 0.93	0.91 (0.07)
Methyl nonanoate	0.86 0.94	0.74 (0.06)	0.89 0.92	0.83 (0.09)	0.91 0.95	0.86 (0.12)
Methyl decanoate	0.85 0.86	0.73 (0.11)	0.88 0.90	0.84 (0.11)	0.91 0.95	0.92 (0.09)
Methyl undecanoate	0.84 0.84	0.67 (0.10)	0.85 0.83	0.71 (0.06)	0.85 0.83	0.92 (0.16)
Methyl dodecanoate	0.80 0.84	0.76 (0.14)	0.87 0.99	0.83 (0.14)	0.91 1.14	0.83 (0.14)

Methyl carboxylic esters (SE-30 column), single-ramp temperature programme, initial temperature 373.2 K.
[a] First line of each pair: first order in ln k' vs. T^{-1}. Second line of each pair: second order in ln k' vs. T^{-1}.
[b] Values in parentheses: estimated standard errors of mean based upon six replicates.

Table 2
Typical section of output from width/asymmetry predictive computer programme

Name	Retention time/s	Width/s	Asymmetry	R_s	*APR*	*POF*/s	*APOF*/s
Diethyl malonate	356.40	2.194	0.800				
				0.660	0.645	−1.273	−1.359
Octan-1-ol	358.88	2.217	0.731				
				1.781	1.731	2.945	2.837
Acetophenone	365.59	2.222	0.649				

Single-ramp temperature programme, SE-54 column. First-order polynomial for log k' vs. T^{-1}. Heating rate = 5.0 K min^{-1}, initial temperature = 353.2 K. Resolution product = 1.176, asymmetric peak resolution product = 1.118, normalized resolution product = 0.789, normalized asymmetric peak resolution product = 0.791, sum of peak overlap functions = 1.672 s, sum of asymmetric peak overlap functions = 1.477 s, highest retention time (analysis time) = 365.6 s. The following "modified" values include only resolutions < 1.50 or peak overlap functions < 0.00 s: modified resolution product = 0.990, modified asymmetric peak resolution product = 0.968, modified sum of peak overlap functions = − 1.273 s, modified sum of asymmetric peak overlap functions = −1.359.

Table 3
Elementary optimization criteria for mixture/column of Table 2 for selected single-ramp programmed-temperature conditions

Initial temperature/K	Parameter	Heating rate/K min^{-1}						
		2.0	4.0	6.0	8.0	10.0	12.0	14.0
343.2	$R_{s,12}$	0.17	0.15	0.48	0.72	0.92	1.08	1.22
	$R_{s,23}$	0.32	0.76	1.42	1.91	2.31	2.64	2.92
	APR_{12}	0.17	0.15	0.47	0.71	0.89	1.05	1.17
	APR_{23}	0.32	0.75	1.38	1.85	2.23	2.54	2.80
	POF_{12}/s	−5.55	−3.98	−1.92	−0.85	−0.21	0.19	0.47
	POF_{23}/s	−4.52	−1.11	1.55	2.83	3.52	3.90	4.12
	$APOF_{12}$/s	−5.53	−4.03	−1.99	−0.92	−0.29	0.11	0.39
	$APOF_{23}$/s	−4.55	−1.18	1.46	2.74	3.42	3.81	4.03
363.2	$R_{s,12}$	0.65	0.90	1.10	1.27	1.40	1.52	1.62
	$R_{s,23}$	1.75	2.27	2.67	3.00	3.28	3.52	3.73
	APR_{12}	0.63	0.88	1.07	1.22	1.35	1.45	1.55
	APR_{23}	1.70	2.19	2.57	2.88	3.14	3.36	3.55
	POF_{12}/s	−1.63	−0.35	0.31	0.70	0.93	1.08	1.18
	POF_{23}/s	3.49	4.63	5.10	5.28	5.32	5.30	5.24
	$APOF_{12}$/s	−1.74	−0.45	0.21	0.60	0.83	0.99	1.09
	$APOF_{23}$/s	3.36	4.51	4.98	5.16	5.21	5.20	5.14
383.2	$R_{s,12}$	1.51	1.65	1.76	1.86	1.95	2.03	2.11
	$R_{s,23}$	3.46	3.74	3.99	4.20	4.39	4.55	4.70
	APR_{12}	1.44	1.57	1.68	1.77	1.85	1.92	1.99
	APR_{23}	3.29	3.55	3.78	3.97	4.14	4.29	4.42
	POF_{12}/s	1.49	1.62	1.69	1.71	1.72	1.71	1.70
	POF_{23}/s	7.33	7.02	6.73	6.46	6.22	6.00	5.80
	$APOF_{12}$/s	1.36	1.50	1.57	1.61	1.62	1.62	1.61
	$APOF_{23}$/s	7.18	6.88	6.60	6.34	6.11	5.90	5.70

First-order polynomial for log k' vs. T^{-1}. The first/second line of each pair is for the (first and second)/(second and third) eluted peaks. N.B. Order of elution varies with conditions.

detector response (in terms of the same quantity)], and/or (ii) involve peak asymmetry [8]. Those used in the present work are independent of relative peak area; some of them ignore asymmetry while others include it. Extension of characteristics of type b are so-called "composite criteria" and include other factors, principally total analysis time. These criteria are based upon the paradigm that good resolution at the expense of excessive analysis time is unacceptable on ergonomic grounds. Such criteria were not used in the present study, however.

Four elementary criteria have been considered here:

(a) *Resolution* [8] (R_s): defined as $2(t_2 - t_1)/(w_1 + w_2)$, where t_1, t_2 are retention (peak maximum) times for adjacent bands ($t_2 > t_1$) and w_1, w_2 are base peak widths, defined for (bi-) gaussians as four times the (mean) standard deviation or equivalently as $\sqrt{(2/\ln 2)}$ times the width at half height; this shape has been assumed here.

(b) *Asymmetric peak resolution* [9] (*APR*): defined as $(t_2 - t_1)/(b_1 + a_2)$, where a, b are asymmetric peak parameters defined above but appropriate to the peak "base".

(c) *Peak overlap function* (*POF*): a new function, with dimension of time, defined as $t_2 - t_1 - (w_1 + w_2)/2$. This may be positive or negative; zero corresponds approximately to a peak pair just resolved, and increasingly positive values to a larger gap between the peaks.

(d) *Asymmetric peak overlap function* (*APOF*): the analogue of c when asymmetry is properly considered, and defined as $t_2 - t_1 - b_1 - a_2$ with a, b as in b above.

Table 4
Complete chromatogram resolution criteria for mixture/column of Table 2 for selected single-ramp programmed-temperature conditions

Initial temperature/K	Parameter[a]	Heating rate/K min^{-1}						
		2.0	4.0	6.0	8.0	10.0	12.0	14.0
343.2	1	0.06	0.12	0.68	1.39	2.13	2.86	3.56
		0.06	0.11	0.65	1.31	1.99	2.66	3.29
	2	0.90	0.55	0.75	0.80	0.82	0.83	0.83
		0.90	0.55	0.76	0.80	0.82	0.83	0.83
	3	−10.1	−5.09	−0.37	1.99	3.31	4.10	4.59
		−10.1	−5.21	0.53	1.82	3.14	3.93	4.42
363.2	1	1.13	2.05	2.95	3.80	4.61	5.36	6.06
		1.08	1.92	2.74	3.51	4.22	4.88	5.50
	2	0.79	0.82	0.83	0.83	0.84	0.84	0.84
		0.79	0.82	0.83	0.84	0.84	0.84	0.85
	3	1.85	4.29	5.41	5.98	6.25	6.38	6.42
		1.62	4.05	5.19	5.76	6.05	6.18	6.23
383.2	1	5.22	6.16	7.03	7.83	8.57	9.26	9.91
		4.75	5.58	6.33	7.01	7.65	8.24	8.79
	2	0.85	0.85	0.85	0.85	0.85	0.85	0.85
		0.85	0.85	0.85	0.85	0.85	0.85	0.86
	3	8.82	8.64	8.42	8.18	7.94	7.71	7.50
		8.54	8.38	8.17	7.95	7.73	7.51	7.31

First-order polynomial for log k' vs. T^{-1}.

[a] 1 = Resolution product (first line), asymmetric peak resolution product (second line); 2 = normalized resolution product (first line), normalized asymmetric peak resolution product (second line); 3 = peak overlap function sum/s (first line), asymmetric peak overlap function sum/s (second line).

These elementary criteria have then been compounded to form ten complete chromatogram parameters as follows:

(e) *Resolution product*: the product of all R_s in the chromatogram —the number of factors in the product is one less than the number of peaks.

(f) *Asymmetric peak resolution product*: as e but using *APR* instead of R_s.

(g) *Normalized resolution product*: as e but using the ratio of $R_s/\langle R_s \rangle$ [$\langle R_s \rangle$ being the (arithmetic) mean of the R_s].

(h) *Normalized asymmetric peak resolution product*: as g but using *APR* instead of R_s.

(i) *Peak overlap function sum*: the sum of *POF* in the chromatogram.

(j) *Asymmetric peak overlap function sum*: as i but using *APOF* instead of *POF*.

(k) *Modified resolution product*: as a but including in the product only those R_s less than a given "critical value". Essentially this is based upon the idea that two peaks are adequately separated when the critical value is reached; beyond this value, the peaks become increasingly separated but this does not improve the analytical value of separating overlapping peaks. A critical value of 1.5 has been used here, but this can be changed in the computer data file.

(l) *Modified asymmetric peak resolution product*: as j but with *APR* instead of R_s; the same critical value has been used.

(m) *Modified peak overlap function sum*: as c but including in the sum only those *POF* less than a given "critical value", specified in the computer programme data file, and chosen here as zero.

(n) *Modified asymmetric peak overlap function sum*: as m but with *APOF* instead of *POF*.

The four elementary (for each adjacent peak pair) and the ten chromatogram parameters are evaluated and printed for each temperature programme in the mainframe Pascal programme evaluating peak width and asymmetry, described above. The further parameter, analysis time equated to the longest retention time was also printed. The programme was tested on a solute mixture [4] of diethyl malonate, octan-1-one and acetophenone on an SE-54 capillary column; elution order varied with temperature programme conditions. Typical predictions are shown in Tables 2 to 4; further information is available elsewhere [10]. The small variation with conditions for the two normalized resolution products (g and h) is interesting and would suggest limiting value of these parameters.

The resolution parameters and the analysis time were also written by the Pascal programme to a file used as input data to a very simple mainframe FORTRAN programme, utilising two GINOSURF routines available in a package on

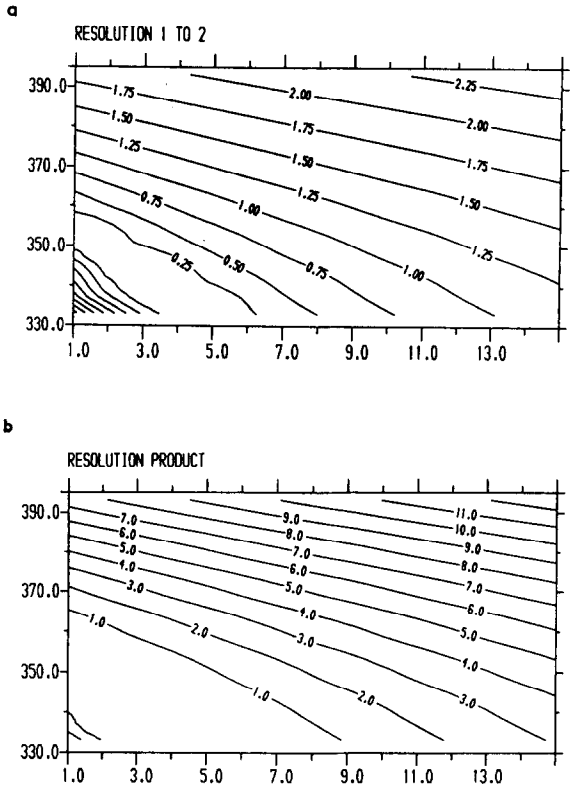

Fig. 1. Contour diagrams showing (a) resolution between peaks for the first two eluted compounds [of (i) diethyl malonate, (ii) octan-1-ol and (iii) acetophenone on an SE-54 column] and (b) resolution product. The abscissae are heating rates/K min^{-1} and the ordinates initial temperatures/K. The contours in the lower left corner ascend in value towards the corner [0.25, 0.50, 0.75, 1.00, 1.25, 1.50, 1.75, 2.00 for (a); 1.0, 2.0 for (b)]. The valley crossing from ca. 350 K min^{-1} to 5 K represent a changeover in order of elution [(ii) then (iii) then (i) in the lower left corner; (i) then (ii) then (iii) elsewhere]. The upper right corner represents good pair and overall resolution/selectivity but at the expense of long analysis time.

the Amdahl 5890 computer. This latter programme produced two kinds of "three-dimensional" plot —(i) isometric projection (with selectable viewing angle), and (ii) contour— of chosen parameters against initial temperature and heating rate. The programme was modified as required to select the chosen parameters and plot types. Fig. 1 shows two examples of contour plots: resolution between the first two eluted compounds and resolution product. These plots

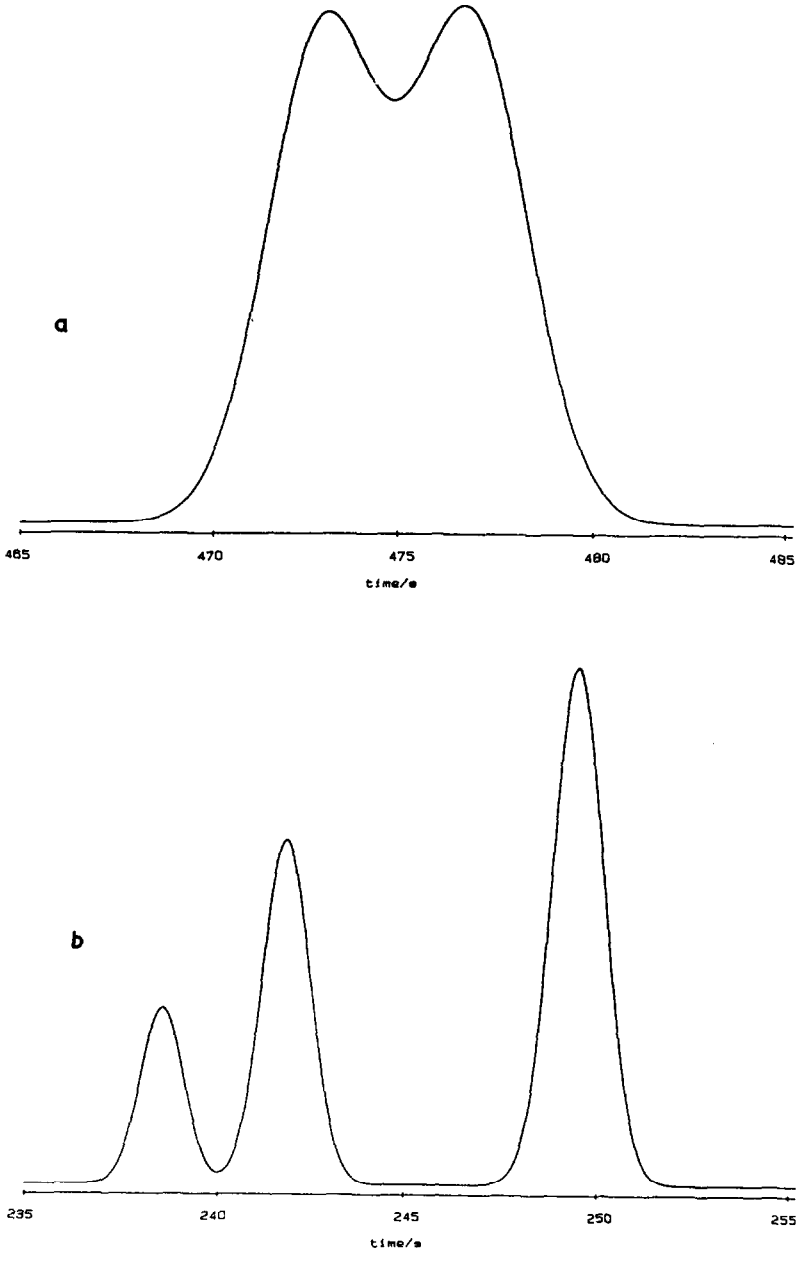

Fig. 2. Two examples of predicted chromatograms [bi-gaussian peaks with areas in ratio 1:2:3 for (i) diethyl malonate, (ii) octan-1-one and (iii) acetophenone on SE-54 column: (a) initial temperature = 343.2 K, heating rate = 4.0 K min^{-1} [peaks for (i) and (ii) on the left are not separated]; (b) initial temperature = 363.2 K, heating rate = 10.0 K min^{-1}.

allow visual selection of optimum/satisfactory programmed temperature conditions.

A third computer programme was written in Turbo Pascal for use on an IBM 8086 microcomputer (with 8087 coprocessor) with Hewlett-Packard 7470A graph plotter connected to the computer through an IEEE 488 card. This programme plots a simulated chromatogram from given relative amounts, response factors, retention times, half-height widths and half-height asymmetries (isothermal or programmed temperature, observed or predicted), and includes a choice of peak shape between (scalene) triangular and bi-gaussian. A procedure for transferring a screen-displayed chromatogram to printer paper (as an alternative or addition to plotting) was incorporated into the programme; this was provided by Dr. M.J. Parrott and was more efficient than the usual "print screen" facility. Fig. 2 shows simulated, temperature programmed, bi-gaussian chromatograms for the test mixture used in the mainframe Pascal programme; the composition is such that peak areas are in the ratio 1:2:3 for diethyl malonate, octan-1-ol and acetophenone, respectively. This provides an alternative procedure for assessing resolution/selectivity quality for programmed conditions without actually performing the experiment.

Copies of the various computer programmes referred to above are available from the authors.

References

[1] E.E. Akhporhonor, S. Le Vent and D.R. Taylor, *J. Chromatogr.*, 405 (1987) 67.
[2] E.E. Akhporhonor, S. Le Vent and D.R. Taylor, *J. Chromatogr.*, 463 (1989) 271.
[3] E.E. Akhporhonor, S. Le Vent and D.R. Taylor, *J. Chromatogr.*, 504 (1990) 269.
[4] T.I. Al-Bajjari, S. Le Vent and D.R. Taylor, *J. Chromatogr. A*, 683 (1994) 367.
[5] D. Repka, J. Krupčík, A. Brunowská, P.A. Leclerq and J.A. Rijks, *J. Chromatogr.*, 463 (1989) 235; and references cited therein.
[6] R.E. Pauls and L.B. Rogers, *Anal. Chem.*, 49 (1977) 625; and references cited therein.
[7] P.J. Schoenmakers, *Optimization of Chromatographic Selectivity* (*Journal of Chromatography Library*, Vol. 35), Elsevier, Amsterdam, 1986.
[8] P.J. Schoenmakers, *Optimization of Chromatographic Selectivity* (*Journal of Chromatography Library*, Vol. 35), Elsevier, Amsterdam, 1986, p. 116.
[9] P.J. Schoenmakers, J.K. Strasters and Á. Bartha, *J. Chromatogr.*, 458 (1988) 355.
[10] T.A. Al-Bajjari, *Ph.D. Thesis*, University of Manchester, Manchester, 1990.