Journal of Chromatography A, 1218 (2011) 5305–5310

Contents lists available at ScienceDirect

Journal of Chromatography A

journal homepage: www.elsevier.com/locate/chroma

Comparing columns for gas chromatography with the two-parameter model for retention prediction

Yasar Thewalim, Ioannis Sadiktsis, Anders Colmsjö*

Stockholm University, Department of Analytical Chemistry, SE-106 91 Stockholm, Sweden

ARTICLE INFO

Article history:
Received 22 March 2011
Received in revised form 20 May 2011
Accepted 23 May 2011
Available online 12 June 2011

Keywords:
Gas chromatography
Prediction
Retention time
PCA score plot
Aged column
Thermodynamic properties

ABSTRACT

The retention times of selected compounds in temperature programmed gas chromatography were predicted using a two-parameter model, on the basis of thermodynamic data obtained from isothermal runs on seven capillary columns, primarily substituted with 5% diphenylsiloxane. The scope for using thermodynamic data obtained from isothermal runs on one column to optimize separation on a different column or a different instrument setup was investigated. Additionally, the predictive utility of thermodynamic data obtained using a DB-5 column that had been in use for three years was compared to that of a new column of the same model. It was found that satisfactory separation could be achieved on one capillary column or instrument setup on the basis of thermodynamic data obtained using a different column or instrument set-up.

1. Introduction

The most common application of gas chromatography is in the separation of target analytes using temperature-programmed runs. Although temperature gradients are less time consuming than isothermal runs, their optimization can be a tedious task [1]. To this end, computational simulation of chromatographic processes can be an extremely powerful tool. Simulations can provide valuable insights into the effects of physical parameters on the retention and separation of compounds in different samples. Moreover, differences between the simulated and experimental results can be used to identify the occurrence of second- and third-order perturbations, thus providing still deeper insights into the chemical interactions that enhance or deteriorate separation in specific systems. Computer simulations can thus considerably reduce the time required for experimental optimization [1]. The data required to perform these simulations can be obtained from a relatively small number of isothermal runs [2,3].

Several models for predicting retention times in temperature-programmed gas chromatography have been presented over the years [1–10]. Research in our laboratories has focused on the development and application of a theoretical and computational procedure for predicting compound retention times that takes thermodynamic data (ΔH and ΔS) obtained under isothermal conditions as its input [1–3]. In this context, ΔH and ΔS refer, respectively, to the change in enthalpy and the change in entropy associated with the transition of the analyte from the mobile phase to the stationary phase.

In addition, comparative studies of the chromatographic behavior of columns produced by different manufacturers and having different dimensions and polarities have been reported, using a diverse set of test compounds [1–3,5,7,11–18]. However, there is no systematic compilation of thermodynamic data for a diverse range of compounds on columns made by different manufacturers but having the same dimensions and equivalent stationary phases.

The aim of this study was to determine whether thermodynamic data for a specific compound obtained on one gas chromatographic column can reliably be used to optimize that compound's separation on a different column, made by a different manufacturer, that has a chemically equivalent stationary phase (*i.e.* the same percentage of phenyl substitution). An additional aim was to investigate the scope for using thermodynamic data obtained using a specific experimental setup to design an optimized separation program for a different experimental setup – for example, would it be possible to use data obtained using a system with the column outlet at ambient temperature and hydrogen as the carrier gas to predict retention times in a mass spectrometric system with helium as the carrier gas and the column outlet to vacuum?

This paper also details studies on the influence of column aging over a period of three years on the measured thermodynamic properties and the reliability of predictions based upon them.

* Corresponding author. Tel.: +46 08 674 71 97; fax: +46 815 63 91.
E-mail address: anders.colmsjo@anchem.su.se (A. Colmsjö).

0021-9673/$ – see front matter © 2011 Elsevier B.V. All rights reserved.
doi:10.1016/j.chroma.2011.05.082

2. Materials and methods

2.1. Chemicals

Six standard solutions were used in the experiments. The *n*-alkane standard contained 15 even carbon number alkanes between C_{12}–C_{40}. The PAH solution contained naphthalene, 1,2-dihydro acenaphthylene, fluorene, acenaphthene, anthracene, phenanthrene, fluoranthene, pyrene, benz[*a*]anthracene, chrysene, benz[*b*]fluoranthene, benz[*k*]fluoranthene, benz[*a*]pyrene, indeno[1,2,3-*cd*]pyrene, benz[*ghi*]perylene and dibenz[*a,h*]anthracene. Grobs' calibration mixture contained decane, 1-octanol, 2,6-dimethylphenol, dodecane, 2,6-dimethylaniline, methyl decanoate, methyl undecanoate, dicyclohexylamine, methyl dodecanoate. The alcohol mixture contained cyclohexanol, 3-chloro-2,2-dimethyl-1-propanol, 3-bromo-2-methyl-1-propanol, 1-octanol, 1-decanol, 1-dodecanol, 1-tetradecanol, 1-hexadecanol, 1-octadecanol, and 1-eicosanol. The amine mixture contained cyclohexylamine, 1-octylamine, 1-decylamine, 1-dodecylamine, 1-tetradecylamine, 1,2-diphenylethylamine, 1-hexadecylamine and 1-octadecylamine. The pesticide mixture contained diazinon, dimethoate, chlorpyrifos, fenitrothion and malathion. Aside from the compounds in Grobs' calibration mixture (Supelco, Bellefonte, PA, USA), all compounds were supplied by Sigma–Aldrich (Schnelldorf, Germany).

Methane gas was used as the column dead time marker.

2.2. Instrumental

Two different instrument setups were used.

Setup 1 was similar to that described in previous publications from our laboratories [1–3], and was used for both isothermal and temperature-programmed experiments with a flame ionization detector (FID). Hydrogen was used as the carrier gas. Four different columns with 5% diphenylsiloxane substitution were investigated: the DB-5 (J&W Scientific Columns from Agilent Technologies), the Factor 4 (Varian), the ZB-5 (Phenomex) and the TR-5 (Themo Scientific). Each column was 30 m long, with an inner diameter of 0.25 mm and a stationary phase film thickness of 0.25 μm. The column dimensions specified by the supplier were used for all calculations. The DB-5 column had been used extensively for three years before the start of this study [1–3]. Among other things, it had been in use for a total of ca. 1000 runs in the analysis of pesticides, heavy PAHs (of M_W up to 302), esters and phosphates. In addition three columns from J&W Scientific Columns, Agilent Technologies, were used for comparative reasons: DB-1 (pure polydimethylsiloxane), DB-17 (50% diphenylsiloxane) and DB-23 (polydimethylsiloxane with 50% dicyanopropylsiloxane substitution).

Setup 2 featured a GC 6890A (Agilent Technologies) equipped with a TSQ 7000 triple quadrupole mass spectrometer (Finnigan MAT, A Subsidiary of Thermo Quest Corp.) and was used for experimental temperature programmed runs. Helium was used as the carrier gas, with a constant flow rate of 1 mL min^{-1}. The compounds were separated on a DB-5 capillary column (J&W Scientific Columns, Agilent Technologies) with a length of 26.2 m long, an inner diameter of 0.25 mm, and a stationary phase film thickness of 0.25 μm.

2.3. Temperatures

The retention times of the 15 *n*-alkanes were measured under isothermal conditions over two runs at temperatures between 60 and 300 °C, with 30° increments. The isothermal retention times of the Grob standard mixture were measured between 40 and 120 °C, with 20° increments. The retention times of the alcohol and amine mixtures were measured under isothermal conditions over two runs at temperatures between 40 and 240 °C, with 20° increments between 40 and 80 °C and 40° increments between 80 and 240 °C. The retention times of the PAHs were measured under isothermal conditions between 60 and 260 °C, with 20° increments between 60 and 100 °C, 40° increments between 100 and 180 °C, 20° increments between 180 and 240 °C, and a 10° increment between 240 and 250 °C. The pesticides' retention times were measured at temperatures between 130 and 170 °C under isothermal conditions, in 20° increments. Three isothermal runs, at different temperatures with double injections, were sufficient to obtain good estimates of ΔH and ΔS and to check their linearity over the temperature range investigated [3]. The GC *Interactive Simulations* [19] software package was used to predict compounds' retention times under specific temperature conditions on the basis of their estimated ΔH and ΔS values. The predicted results were compared to the average retention times observed in two runs using the temperature programs presented in Table 1

2.4. Calculations

Several software packages were used in the study described in this paper. ELDS Professional Win v1.1 (Chromatography Data Systems, Sweden) was used for recording chromatograms and integrating peaks. Microsoft Excel 2000 (Microsoft Corporation) was used for some calculations. The Unscrambler® 6.1 (CAMO AS) was used for Principal Components Analysis (PCA). GC Interactive Simulation, a customized computer program developed in-house and based on Delphi 4.9, was used for chromatographic simulations and for calculating the thermodynamic parameters [19].

3. Results and discussion

Using a series of isothermal runs, the thermodynamic parameters for a number of *n*-alkanes, PAHs, linear and branched alcohols and amines, pesticides and a Grob calibration mixture were determined on several capillary columns. All data were collected at temperatures at which the retention factor, *k*, was greater than one; at higher temperatures (and thus lower values of *k*), the error in the calculated value of *k* becomes increasingly large [1].

Although the columns were supplied by different manufacturers and some differences in their stationary phases are therefore to be expected [20,21], the thermodynamic quantities estimated on the different columns were remarkably similar. For example, the thermodynamic parameters for the Grob calibration mixture on the various columns are shown in Table 2.

This indicates that as long as the columns' stationary phases can be considered to be chemically equivalent (*i.e.* they have the same percentage of phenyl substitution), the analyzed compounds will not differ substantially in their thermodynamic behavior on the stationary phases, regardless of the precise identity of the column.

Principal component analysis (PCA) was used to analyze the thermodynamic data obtained in the course of these studies and that gathered in previously published work [1–3]. PCA facilitates the exploration of patterns in obtained data and can be used to identify correlations between datasets. Fig. 1 shows a matrix representation of the thermodynamic data (ΔH and ΔS values) used to generate the PCA plot. A total of 470 chromatographic runs were used for this PCA plot.

Fig. 2 shows the results of the PCA using the ΔH and ΔS data from all of the 5% diphenylsiloxane substituted columns (including the aged DB-5 column) and those obtained from previous studies using the DB-1 (non-polar), DB-17 (moderately polar) and DB-23 (polar) columns [1–3].

Table 1
Temperature programs used in the experiments.

Mixture	Start temperature	Hold time	Ramp 1	Temperature	Hold time	Ramp 2	Final temperature	Hold time
n-Alkanes	60 °C	2 min	10 °C/min	–	–	–	300 °C	20 min
PAHs	60 °C	2 min	5 °C/min	–	–	–	250 °C	30 min
Alcohols/amines	60 °C	3 min	8 °C/min	–	–	–	240 °C	10 min
Grobs mix	40 °C	3 min	8 °C/min	–	–	–	120 °C	15 min
Pesticides	60 °C	1 min	30 °C/min	120 °C	0 min	15 °C/min	295 °C	5 min

Table 2
Calculated thermodynamic values for standard according to Grob mixture on Factor 4, ZB-5, TR-5 and the old DB-5 capillary columns, determined using isothermal runs within the temperature interval 40–120 °C. ΔH in kJ/mol and ΔS in J/mol K.

	Factor 4	ZB-5	TR-5	DB-5 (new)	DB-5 (old)	Factor 4	ZB-5	TR-5	DB-5 (new)	DB-5 (old)
Compound	ΔH	ΔH	ΔH	ΔH	ΔH	ΔS	ΔS	ΔS	ΔS	ΔS
Decane	−45.34	−44.59	−44.23	−4303	−44.44	−72.95	−70.77	−70.49	−66.57	−71.03
1-Octanol	−49.48	−48.74	−48.49	−46.65	−48.55	−80.27	−78.39	−78.37	−72.66	−78.58
2,6-Dimethylphenol	−48.53	−47.97	−47.87	−51.72	−48.35	−75.31	−74.08	−74.39	−79.25	−72.49
2,6-Dimethylaniline	−49.32	−48.65	−48.51	−45.98	−50.74	−74.09	−72.59	−72.77	−68.70	−76.76
Dodecane	−53.56	−53.39	−53.15	−46.93	−47.70	−83.82	−83.77	−83.86	−68.03	−74.11
Methyl decanoate	−58.41	−55.12	−55.72	−56.16	−56.25	−89.96	−81.25	−83.65	−84.59	−84.97
Dicyclohexylamine	−55.31	−54.12	−54.66	−59.80	−54.03	−76.88	−74.29	−76.47	−89.06	−74.76
Methyl undecanoate	−60.41	−59.23	−59.84	−54.91	−59.27	−89.74	−86.96	−89.42	−76.89	−87.81
Methyl laurate	−64.60	−63.33	−63.93	−62.32	−63.39	−95.58	−92.70	−95.11	−93.26	−93.59

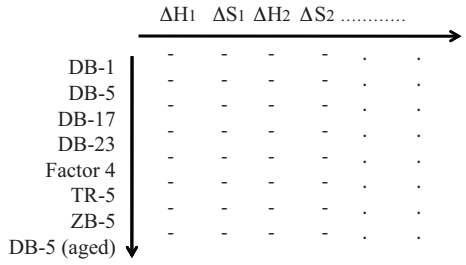

Fig. 1. Matrix representation of coupled chromatographic data. ΔH_n and ΔS_n represents compound *n*.

As expected, the 5% diphenylsiloxane substituted columns all exhibit similar behavior and are well-separated from the DB17 and DB23 columns. However, the encircled area of Fig. 2 shows that there are distinct differences between the 5% diphenylsiloxane-substituted columns. Also, the DB-1 column falls within the 5% diphenylsiloxane-substitution ellipse indication that thermodynamic properties are influenced due to manufacturing variations of the columns in the same degree as the low percentage of substitution.

The figure also shows that the thermodynamic values (*i.e.* ΔH and ΔS) obtained on a new DB-5 column were significantly different to those obtained using a column that had seen three years'

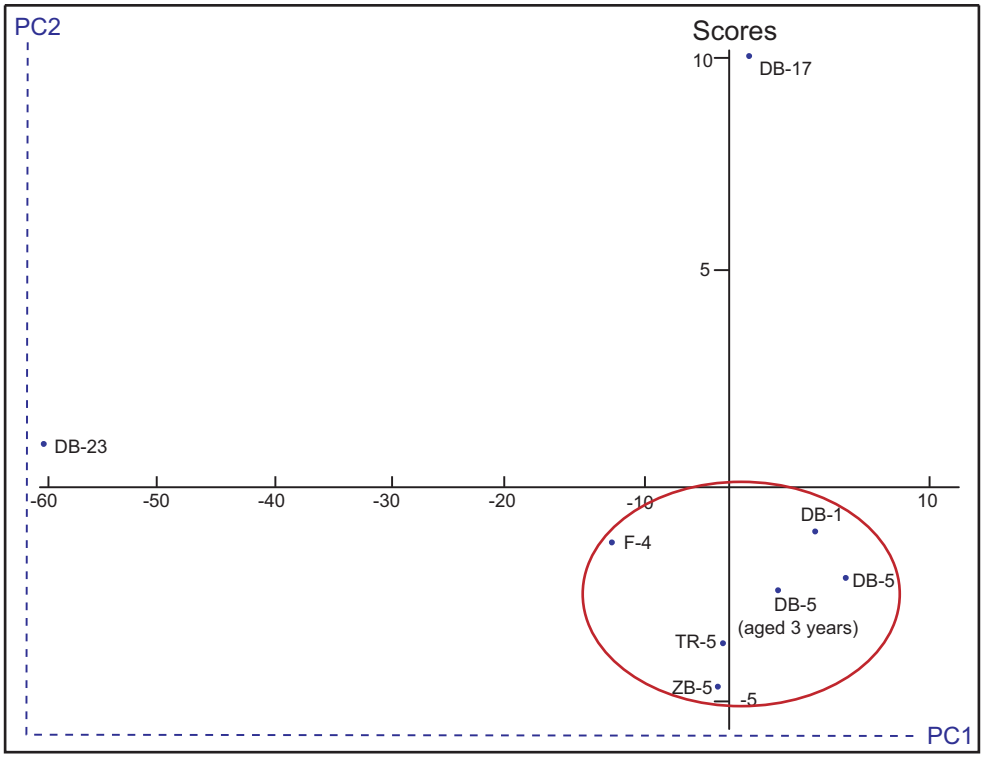

Fig. 2. PCA score plot showing similarities of six columns in the ellipse.

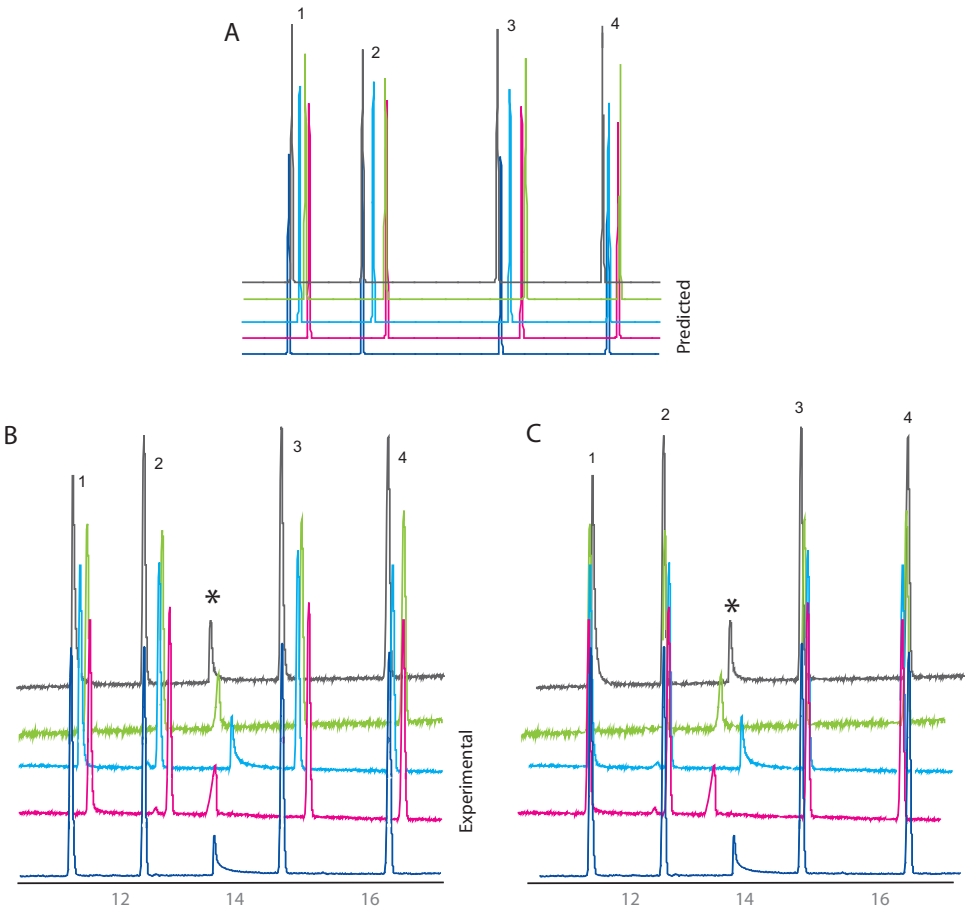

Fig. 3. A region of the predicted (A) and experimental (before peak alignment: B; after peak alignment: C) chromatograms for 1-octanol (1), 2,6-dimethylphenol (2), 2,6-dimethylaniline (3), and dodecane (4) in the Grob mixture, analyzed on the F-4 (red), TR-5 (light blue), ZB-5 (green), DB-5 (dark blue) and aged DB-5 (dark grey) columns. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

extensive usage. Since the column's dimensions, such as its inner diameter and stationary phase thickness should remain unchanged, and thermodynamic parameters are independent of the length of the column, the differences between the ΔH and ΔS values obtained before [2] and after three years usage must primarily reflect modifications of the stationary phase.

Using the two-parameter model described in detail in previous work [1], the retention times of the analytes were predicted using data derived from three isothermal runs. Generally, the predicted retention times were within 1% of the experimentally determined values using the same column. In some cases, it was necessary to perform some peak alignment in order to compare the predicted and experimental data for specific columns. In general, it was only necessary to shift the chromatograms by a couple of seconds or less, and only time corrections were required – there was no need to perform "rubber band" type expansion or contractions.

Fig. 3 shows an expansion of a region of the predicted and simulated chromatograms for a temperature-programmed run on the F-4, TR-5, and ZB-5 columns, and on the new and aged DB-5 columns.

Looking at Fig. 3A and B, it is apparent that after peak alignment, the chromatograms for these compounds are very similar, irrespective of the identity of the column (at least for columns having the same percentage of phenyl substitution). The peaks for 2-ethylcaproitic acid, are indicated with an asterisk in the chromatograms. The acid is retained also by adsorption to the column backbone, the nature of which differs significantly between the columns examined in this study, *i.e.* two retention mechanisms are involved. For the adsorption, the number and character of sites differ largely between the columns giving either leading or tailing peaks.

The relative standard deviation between the experimental retention times obtained on one column and the predicted retention times obtained from isothermal data from another column is within 2% before peak alignment. ANOVA tests indicate that in general, each compound has different ΔH and ΔS values on the 5% diphenylsiloxane substituted columns (at the 5% significance level). This is because one of the compounds exhibits extremely low variance in its retention time on one particular column, which gives rise to differences in spite of the fact that the chromatographic column-to-column variation is less than 2%. Overall, these results imply that thermodynamic data obtained on one column can indeed be used to optimize separations, and that the optimization parameters so obtained can be used to design separations of the same solutes on a different capillary column.

Fig. 4 compares the simulated and experimental chromatograms of five selected pesticides. The predicted retention times (A) were obtained from thermodynamic data derived from isothermal runs performed on a GC-FID instrument. These data were then used to separate the pesticides on a GC–MS instrument equipped with a triple quadrupole with a measured outlet pressure of 16 mTorr at the initial temperature.

Although the instrument and setup used to acquire the experimental chromatogram differed significantly from those used to acquire the thermodynamic data, good agreement between the experimental and predicted retention times was achieved.

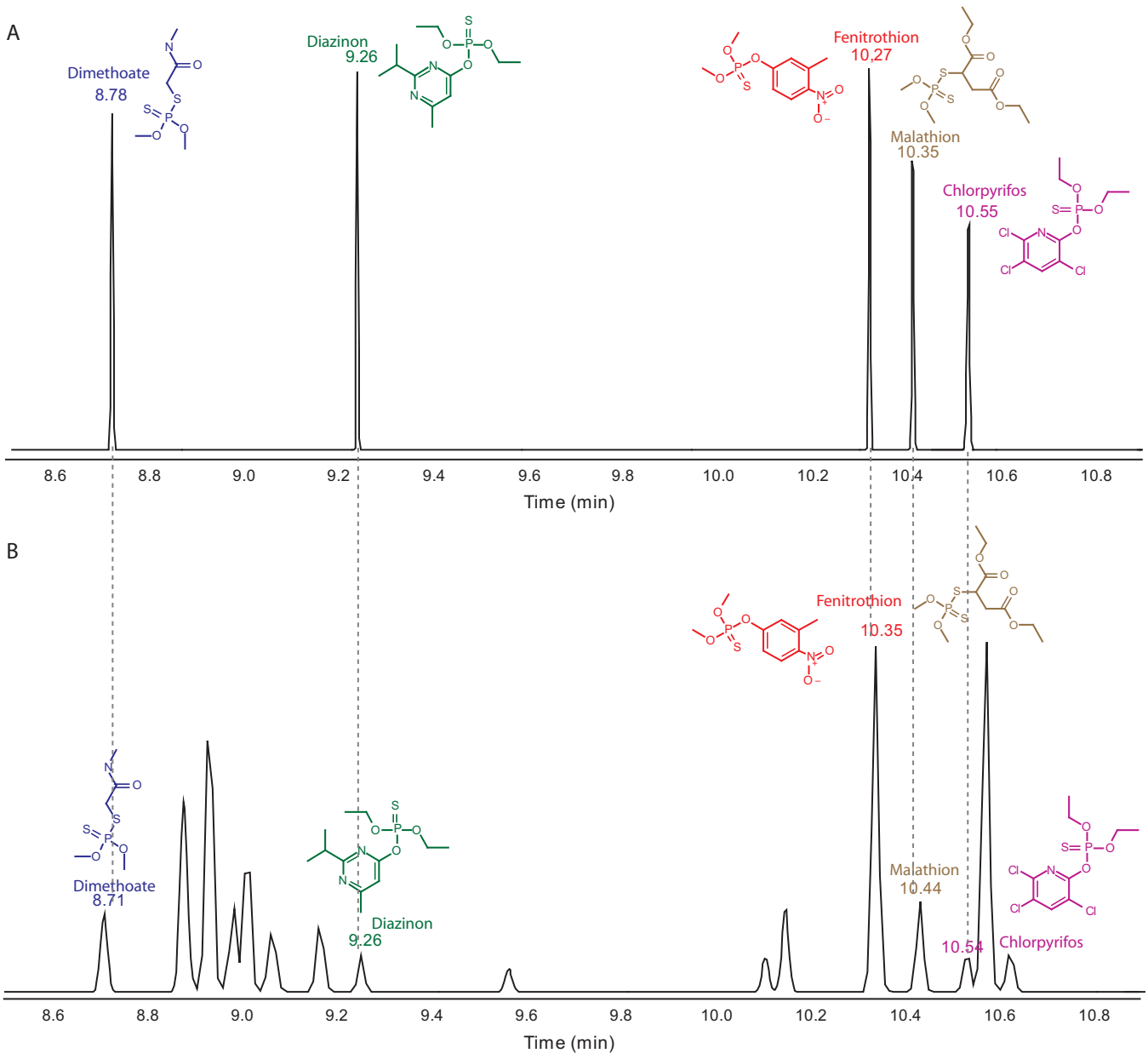

Fig. 4. A chromatogram of five selected pesticides on the DB-5 column using a temperature program of 60 °C for 1 min followed by a 30 °C/min ramp to 120 °C, followed by a 15 °C/min ramp to 295 °C for 5 min. (A) Simulated chromatogram generated using the GC Interactive Simulation software package and data from a GC-FID system, (B) experimental chromatogram obtained on a GC–MS 6890A-TSQ 7000.

The results presented in Fig. 4 show that thermodynamic parameters derived using one instrumental set-up can be used to design and optimize separations using completely different instrumental setups.

4. Conclusions

A two-parameter model that uses the thermodynamic parameters ΔH and ΔS to calculate the retention times of analytes in gas chromatography has been validated on several capillary columns and instruments. Using the model, chromatograms were simulated extremely well. The results obtained show that although the 5% diphenylsiloxane substituted columns were produced by different manufacturers, the analyzed compounds generally experienced similar thermodynamic forces and thus exhibited very similar ΔH and ΔS values on columns with chemically equivalent stationary phases (*i.e.* columns whose stationary phases have the same percentage of phenyl substitution). The thermodynamic data obtained can successfully be used to predict retention times on instruments and setups different to those used to generate the thermodynamic data in the first place.

This study also shows that it is also possible to optimize the separation of complex samples on one GC instrumental set-up using thermodynamic data obtained on a completely different GC instrumental setup.

In summary, the two-parameter model can be used to create a database of ΔH and ΔS values, which can in turn be used to optimize the separation of complex samples on any given column using computer simulation.

References

[1] F. Aldaeus, Y. Thewalim, A. Colmsjö, Anal. Bioanal. Chem. 389 (2007) 941.
[2] Y. Thewalim, F. Aldaeus, A. Colmsjö, Anal. Bioanal. Chem. 393 (2009) 327.

[3] D.W. Grant, M.G. Hollis, J. Chromatogr. 158 (1978) 3.
[4] V. Bartu, S. Wicar, Anal. Chim. Acta 150 (1983) 245.
[5] E.V. Dose, Anal. Chem. 59 (1987) 2414.
[6] E.J. Cavalli, C. Guinchard, J. Chromatogr. Sci. 33 (1995) 370.
[7] H. Snijders, H.-G. Janssen, C. Cramers, J. Chromatogr. A 718 (1995) 339.
[8] S.J. Hawkes, J. Chromatogr. A 746 (1996) 282.
[9] Y. Thewalim, O. Bruno, A. Colmsjö, Anal. Bioanal. Chem. 399 (2011) 1335.
[10] B. Karolat, J. Harynuk, J. Chromatogr. A 1217 (2010) 4862.
[11] E.E. Akporhonor, S. Le Vent, D.R. Taylor, J. Chromatogr. 463 (1989) 271.
[12] D.E. Bautz, J.W. Dolan, W.D. Raddatz, L.R. Snyder, Anal. Chem. 62 (1990) 1560.
[13] G. Castello, P. Moretti, S. Vezzani, J. Chromatogr. 635 (1993) 103.
[14] S. Vezzani, P. Moretti, G. Castello, J. Chromatogr. A 677 (1994) 331.
[15] G. Castello, S. Vezzani, P. Moretti, J. Chromatogr. A 742 (1996) 151.
[16] S. Vezzani, P. Moretti, G. Castello, J. Chromatogr. A 767 (1997) 115.
[17] H. Du, Z. Ring, Y. Briker, P. Arboleda, Catal. Today 98 (2004) 217.
[18] S. Vezani, P. Moreti, G. Castelo, Anal. Chim. Acta 599 (2007) 151.
[19] A. Colmsjö, GC Interactive Simulation. See corresponding author, 2007.
[20] K.J. Hyver, High Resolution Gas Chromatography, Hewlett-Packard, 1989.
[21] J. Bjorklund, P. Tollback, C. Hiarne, et al., J. Chromatogr. A 1041 (2004) 201.