

See discussions, stats, and author profiles for this publication at: <https://www.researchgate.net/publication/231172673>

Characterization and selection of electrolyte systems for isotachophoresis of anions by cluster analysis

ARTICLE *in* ANALYTICAL CHEMISTRY · JANUARY 1988

Impact Factor: 5.64 · DOI: 10.1021/ac00153a005

CITATIONS

5

READS

22

2 AUTHORS:



Ernst Kenndler

University of Vienna

258 PUBLICATIONS **6,362** CITATIONS

SEE PROFILE



Gregor Reich

University of Vienna

12 PUBLICATIONS **90** CITATIONS

SEE PROFILE

column lifetime. As shown in Figure 6 and also in animal pharmacokinetic studies currently in progress, this methodology is proving to be quite useful and will be utilized for pharmacokinetic studies in the clinic and aid in the development of this potent class of compounds for treatment of malaria in man.

The analysis of this class of compounds in biological fluids in the low nanogram per milliliter range is a significant challenge in which little progress has been made in the past. The studies reported in this paper were initiated when it was observed that adequate sensitivity could not be obtained by HPLC with reductive electrochemical detection (14). Unlike the HPLC method in which qinghaosu and any derivatives with the peroxide moiety can be detected and quantitated, this method was specifically designed for the analysis of DQHS after administration of ARTA, or other DQHS derivatives with a carboxyl moiety. Since ARTA and other ester or ether derivatives of DQHS also pyrolyze to the cyclohexane pyrolysis product, a carboxyl moiety permits separation of DQHS from the parent compound by liquid-liquid extraction. Therefore, this method is not useful for the analysis of DQHS formed from artemether or arteether (methyl or ethyl ethers of DQHS). This difficulty represents the major limitation in the general utility of the method for the analysis of DQHS; however, as shown in this paper, the method is useful for the quantitation of DQHS after administration of ARTA. Other studies to find a generally applicable method are in progress in this laboratory. In the interim, this method is now being used to study the pharmacokinetics of DQHS formation and elimination in various animal models.

Registry No. DQHS, 71939-50-9.

LITERATURE CITED

- (1) Fourth Meeting of the Scientific Working Group on the Chemotherapy of Malaria, Beijing, People's Republic of China; WHO Report TDR/CHE-MAL-SWG(4)/QHS/81, 1981.
- (2) Qinghaosu Research Group, *Sci. Sin. (Engl. Transl.)* **1980**, *23*, 380.
- (3) Liu, J.; Ni, M.; Fan, J.; Tu, Y.; Wu, Z.; Qu, Y.; Chou, W. *Acta Chim. Sin.* **1979**, *37*, 129.
- (4) China Cooperative Research Group on Qinghaosu and Its Derivatives as Antimalarials *J. Tradit. Chin. Med.* **1982**, *2*, 3.
- (5) China Cooperative Research Group on Qinghaosu and Its Derivatives as Antimalarials *J. Tradit. Chin. Med.* **1982**, *2*, 17.
- (6) China Cooperative Research Group on Qinghaosu and Its Derivatives as Antimalarials *J. Tradit. Chin. Med.* **1982**, *2*, 45.
- (7) China Cooperative Research Group on Qinghaosu and Its Derivatives as Antimalarials *J. Tradit. Chin. Med.* **1982**, *2*, 31.
- (8) China Cooperative Research Group on Qinghaosu and Its Derivatives as Antimalarials *J. Tradit. Chin. Med.* **1982**, *2*, 125.
- (9) Klayman, D. L. *Science* **1985**, *228*, 1049.
- (10) China Cooperative Research Group on Qinghaosu and Its Derivatives as Antimalarials, *J. Tradit. Chin. Med.* **1982**, *2*, 25.
- (11) Lin, A. J.; Theoharides, A. D.; Klayman, D. L. *Tetrahedron* **1986**, *42*, 2181.
- (12) Paulsen, R.; Valentine, J. L. *Lab. Anim.* **1984**, *13*, 34.
- (13) Desjardins, R. E.; Pamplin, C. L.; von Bredow, J.; Barry, K. G.; Canfield, C. J. *Clin. Pharmacol. Ther. (St. Louis)* **1979**, *26*, 372.
- (14) Zhou, Z. M.; Anders, J. C.; Chung, H.; Theoharides, A. D. *J. Chromatogr. Biomed. Appl.* **1987**, *414*, 77.
- (15) Oichao, Y.; Weizhi, S.; Rei, L.; Jun, G. *J. Tradit. Chin. Med.* **1982**, *2*, 99.
- (16) Li, Z.; Gu, H.; Warhurst, D.; Peters, W. R. *Soc. Trop. Med. Hyg.* **1983**, *77*, 522.

RECEIVED for review May 29, 1987. Accepted September 21, 1987. Zhong Ming Zhou was a visiting scientist sponsored by the UNDP/World Bank/WHO Special Programme for Research and Training in Tropical Diseases.

Characterization and Selection of Electrolyte Systems for Isotachophoresis of Anions by Cluster Analysis

Ernst Kenndler* and Gregor Reich

Institute for Analytical Chemistry, University of Vienna, Waehringerstrasse 38, A-1090 Vienna, Austria

A rational strategy based on cluster analysis is proposed for the selection of leading electrolytes in the identification of anions by isotachophoresis in aqueous solutions. The calculations were carried out by using ionic mobilities or not linearly transformable relative step heights as properties. Linear correlation coefficients and Euclidian distances were used as similarity measures. The data for the computation were taken from a library consisting of the values for 263 anionic compounds in eight electrolyte systems with integer pH values ranging from 3 to 10. The sequence of the similarities of the systems is nearly unaffected by the choice of the properties or that of the measure of resemblance. The results of a hierarchical and a nonhierarchical clustering procedure were used to select combinations of electrolytes, which yield maximum identification power. For binary combinations, a system with pH 3 and one with pH ranging from 6 to 10 were found to be most favorable. For ternary combinations, pH 3, 4, and one from 6 to 10 or 3, 5, and one from 6 to 10 were selected. Additional systems do not essentially increase the identification power.

The selection of an electrolyte system is one of the most important steps in developing a method for the separation or identification of ions by electrophoresis. Until now all

strategies proposed for isotachophoresis were applicable only for a restricted range of separation problems (1-12). They allowed a selection of an electrolyte system for the separation of a known pair of ions: starting from their known ionic mobilities and pK_a values it was possible to calculate whether the considered pair of ions could be separated under the given conditions. This method fails, however, even for the simple case to select electrolyte systems for the identification of analytes from a known large set of substances. In practice this problem was solved by informed trial and error, starting from a first scouting experiment.

On the other hand, chemometric methods (13) were already applied to solve similar problems for other separation techniques like thin-layer chromatography or gas chromatography, where phase systems were characterized and their optimal combinations were selected by cluster analysis (or numerical taxonomy) (14-24). In a previous paper (25), we compared the similarities of aqueous and mixed aqueous-organic electrolyte systems in isotachophoresis by this technique. In the present paper, cluster analysis is used to characterize aqueous buffering electrolytes with different pH values, described for the isotachophoresis of anions, and a rational strategy for the combination of electrolytes is proposed. Furthermore, the effect of the choice of different analyte characters on the result of the clustering procedure is briefly discussed, when the different properties are not linearly transformable.

Table I. Buffering Counterions of the Leading Electrolytes with Different pH Values^a

pH of the leading electrolyte	counterion
3.0	β -alanine
4.0	β -alanine
5.0	creatinine
6.0	histidine
7.0	imidazole
8.0	tris(hydroxymethyl)aminomethane
9.0	2-amino-2-methyl-1,3-propanediol (ammediol)
10.0	ethanolamine

^a The leading ion was chloride in all systems

EXPERIMENTAL SECTION

Relative step heights (R_E values) and ionic mobilities of 263 anions in eight electrolyte systems with different pH values of the leading electrolyte were used for cluster analysis. The solutes were selected from the library published by Hirokawa et al. (26) as described in a previous paper (27). The (calculated) R_E values are related to chloride as the leading ion. The buffering counterions of the different leading electrolytes are presented in Table I. The ionic mobilities were calculated from the R_E values by assuming a mobility of $74.6 \times 10^{-5} \text{ cm}^2 \cdot \text{V}^{-1} \cdot \text{s}^{-1}$ for chloride. The Euclidian distances were calculated from the scaled values of mobilities and R_E values. The computer program used for the cluster analysis was ARTHUR (28).

RESULTS AND DISCUSSION

Taxonomic Units and Properties. Eight leading electrolyte systems with (integer) pH values from 3.0 to 10.0 are chosen as taxonomic units. These systems are symbolized by the numbers corresponding to the particular pH values. The solutes are characterized as usual in isotachophoresis by relative step heights like R_E values or h^{rel} values, defined by

$$R_{E,i} = m_L / m_i \quad (1)$$

and

$$h^{\text{rel}} = (m_L / m_i - 1) / (m_L / m_R - 1) = (R_{E,i} - 1) / (R_{E,R} - 1) \quad (2)$$

where m_L , m_R , and m_i are the ionic mobilities of the leading ion, L, the reference ion, R, and the analyte ion, i, respectively. It can be seen from the equations given above that R_E values can be linearly transformed into h^{rel} and vice versa. This means, that both properties are totally correlated, and the results of cluster analysis based on these characters will be redundant. In contrast, the ionic mobility, which is the most important property in electrophoresis, is not linearly transformable into either the h^{rel} or the R_E values. We have therefore selected the ionic mobilities and the R_E values as properties for the cluster analysis.

Measures of Resemblance. The similarity of the electrolyte systems was expressed as in previous papers (25, 27) by the linear correlation coefficient, r , and the Euclidian distance, d_{kl} . The latter is the distance between two points, representing two electrolyte systems, k and l, in an n -dimensional pattern space. n is the number of characters in the particular systems. In the present work, each electrolyte system is therefore represented by one point in a 263-dimensional space.

When the linear correlation coefficient was used as a measure of resemblance and the ionic mobilities were taken as properties, systems 8 and 9 have the highest and 3 and 10 have the lowest similarity, as can be seen from Table II. Although mobilities are not linearly transformable into R_E values, nearly the same sequence of the similarity parameters is found with the R_E values as properties (27). However, the absolute values of the correlation coefficients differ: it is

Table II. Similarity Matrix of the Electrolyte Systems, Given by the Linear Correlation Coefficients^a

system	correlation coefficient r							
	3	4	5	6	7	8	9	10
3	1.000							
4	0.976	1.000						
5	0.882	0.950	1.000					
6	0.801	0.875	0.975	1.000				
7	0.779	0.845	0.944	0.987	1.000			
8	0.777	0.837	0.928	0.972	0.996	1.000		
9	0.772	0.831	0.921	0.964	0.991	0.998	1.000	
10	0.759	0.819	0.910	0.954	0.982	0.990	0.996	1.000

^a The ionic mobilities of the solutes in the different electrolyte systems were used as the properties.**Table III. Similarity Matrix of the Electrolyte Systems, Given by the Euclidian Distances^a**

system	Euclidian distance, $d_{kl} \times 10$							
	3	4	5	6	7	8	9	10
3	0							
4	2.03	0						
5	7.34	5.54	0					
6	9.66	8.12	3.19	0				
7	9.85	8.49	4.30	1.96	0			
8	9.82	8.55	4.76	2.83	0.97	0		
9	9.87	8.62	4.93	3.12	1.43	0.71	0	
10	9.97	8.74	5.16	3.45	2.02	1.52	0.88	0

^a The scaled R_E values of the solutes were used as the properties.**Table IV. Similarity Matrix of the Electrolyte Systems, Given by the Euclidian Distances**

system	Euclidian distance, $d_{kl} \times 10$							
	3	4	5	6	7	8	9	10
3	0							
4	2.20	0						
5	4.85	3.17	0					
6	6.30	4.99	2.25	0				
7	6.65	5.56	3.35	1.60	0			
8	6.68	5.71	3.80	2.38	0.94	0		
9	6.76	5.81	3.99	2.68	1.37	0.63	0	
10	6.95	6.02	4.25	3.03	1.91	1.39	0.84	0

^a The distances are based on the scaled values of the ionic mobilities of the solutes.

found, that the mobilities show a higher linear correlation than the R_E values in most cases.

In Tables III and IV, the similarities of the electrolyte systems are expressed by their Euclidian distances, based on R_E values and on ionic mobilities. It follows, that the results are in agreement with those obtained by the correlation coefficient: the smallest distances are found for systems 8 and 9, and the largest distances for 3 and 10.

It can be seen that the sequence of the similarity of the systems under consideration is not sensitive to the choice of the taxonomic properties or to the choice of the measure of resemblance.

Clustering Procedure. The electrolyte systems are classified by the aid of two clustering methods: a hierarchical and a nonhierarchical procedure. For both, the Euclidian distances were used as similarity parameters.

The results of the hierarchical clustering procedure are shown in Figure 1. The dendrograms were obtained by an unweighted, average linkage method, using the normalized distances, Δ , calculated from the values in Tables III and IV. It can be seen from both dendrograms, that two main clusters are formed, one consisting of systems 3 and 4 and the other

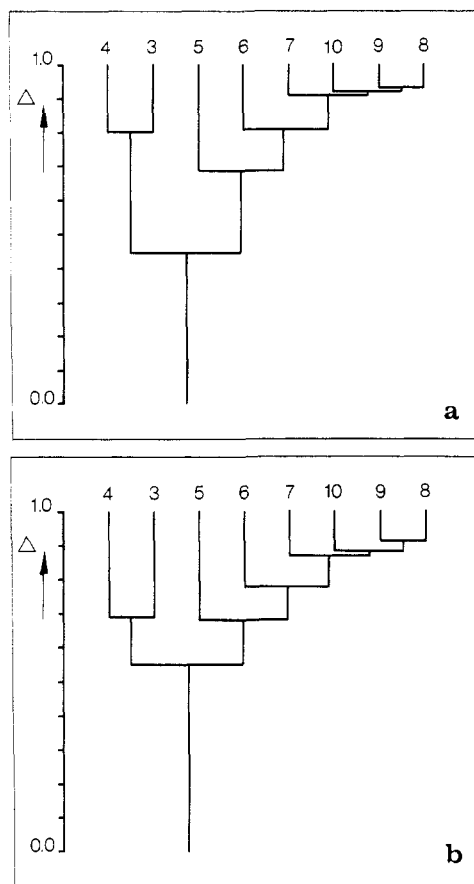


Figure 1. Dendrograms as the results of the hierarchical clustering procedures, based on the data of the Euclidian distances, given in Tables III and IV. The properties used were (a) R_E values and (b) ionic mobilities of the solutes in the different electrolyte systems. Δ = normalized distances.

consisting of the remaining systems with pH values from 5 to 10.

As a second clustering algorithm, the nonhierarchical method of the minimum spanning tree was used. The nonlinear mapping (29) projects multidimensional data to the two-dimensional space, preserving the interpoint distances as correct as possible. The axes of the plots have no known relationship to the data values.

The results of this classification method are presented in Figure 2. In both graphs, the two subgroups separated by the longest branch consist of the same electrolyte systems as found in the dendrograms.

Selection of Leading Electrolyte Systems. The result of the clustering procedures can serve as a tool to select combinations of electrolyte systems that yield maximum identification power. By the aid of the dendrograms shown in Figure 1, two systems will be selected from each of the two main clusters in a first step. As an aid for the selection, the information content (23) calculated from the same isotachopheretic data as applied in the present work can be used. The information content, expressed in bits, is an appropriate measure of the identification power of electrolyte systems. It was found (27), that system 3 has the highest information content of all single systems (about 5.7 bits), and the combination of systems 3 and 10 has the highest information content of all binary combinations (ca. 8.6 bits). These systems are found in the two separated main clusters and are therefore selected as a first choice. The differences in similarities are found to be small for all systems with pH values from 6 to 10. Thus, instead of system 10, another system from this group can be selected without substantial loss of information, which is in agreement with previous results (27). In a second

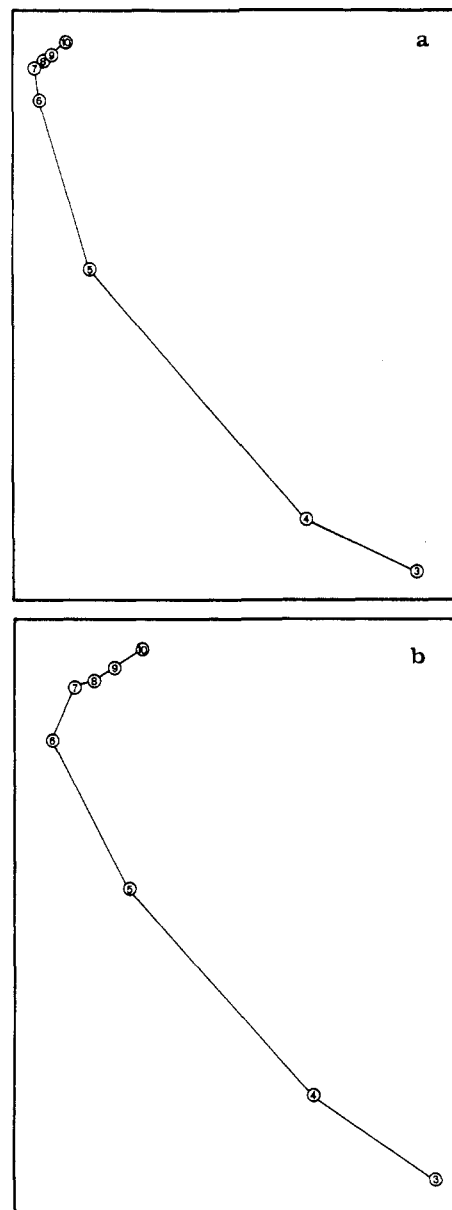


Figure 2. Nonlinear mapping of the data connected by the minimum spanning trees, based on the Euclidian distances given in Tables III and IV. The properties used were (a) R_E values and (b) ionic mobilities of the solutes in the different electrolyte systems.

step, an additional, third system with pH of 4 (or 5) is chosen according to the dendrograms. The application of further systems should not lead to a significant increase of identification power, due to the high similarity of these systems with those already combined.

The selection strategy by the minimum spanning tree, presented in Figure 2, consists of successively breaking the longest branches between the points, by which the systems are represented. This leads to the same two groups of systems as found with the hierarchical method. On the basis of the information content of the different pairs of systems, 3 and 10 are selected too. The high resemblance of systems 6, 7, 8, 9, and 10 clearly can be seen from the structure of the tree. These systems are therefore interchangeable in the combinations without significant loss of information. From the remaining clusters, the longest branch is found between systems 5 and 6. This leads to the selection of three systems: 3, 5, and one from 6 to 10. Again, only short distances between the latter systems are found, indicating only small increase of identification power for additional electrolyte systems. Thus, different types of cluster analysis lead to the same

Table V. Number of Pairs of Ions of the Data Library, Remaining Undifferentiated in the Single Electrolyte Systems and their Binary Combinations^a

electrolyte system	3	4	5	6	7	8	9	10
3	873	249	148	128	125	120	126	123
4		1322	306	229	216	206	211	208
5			2199	778	675	655	636	633
6				2337	1739	1620	1546	1547
7					2344	2057	1976	1980
8						2199	2032	2019
9							2212	2072
10								2157

^a For details, see text.

conclusions. The following leading electrolyte systems are selected as yielding favorable results in the following situations: for binary combinations, 3 and one from 6 to 10; for ternary combinations, 3 and 5 or 3 and 4 and one from 6 to 10.

Evaluation of the Results of the Selection Procedure.

The identification power of the particular leading electrolyte systems and their combinations as discussed above is verified by another approach. It is based on the number of analytes, which can be discriminated in an isotachopherogram under realistic conditions with a universal detector like a potential gradient or electric conductivity detector. This number either can be derived theoretically (27) or can be calculated by using the data of the library mentioned above. For the latter calculation, two analytes are considered to be differentiated when their ionic mobilities differ more than 3% in at least one of the electrolyte systems. The lack in identification power is related to the number of pairs of analytes remaining nondifferentiated, given by the number of pairs with ratios m_i/m_j smaller than 1.03. From the data of the library, matrices of 263×263 elements are constructed, formed by the ratios of the mobilities, m_i/m_j , for each possible pair of analytes, i and j , in the particular electrolyte system, with $m_i > m_j$. The number of pairs of analytes that have ratios smaller than 1.03 in all considered systems is determined. The resulting numbers of nondifferentiated pairs are given in Table V for the single systems and their binary combinations. One can see for the single systems (represented by the diagonal), that system 3 has the lowest number of nondiscriminated pairs of analytes, followed by system 4. All other single systems from 5 to 10 show about the same number of undifferentiated pairs. This result is in agreement with the results obtained with information theory (27).

The selection of system 3 in combination with a second system in the range from 6 to 10, as predicted by cluster analysis, is in accordance with the values of Table V: the number of pairs is reduced to about 120–128. All other combinations of two electrolyte systems lead to a significantly higher number of undifferentiated pairs of analytes.

The calculation for three electrolyte systems, which include system 3, leads to numbers of pairs ranging from 120 (systems 3, 5, 6 and 3, 9, 10) to 112 (3, 4, 8 and 3, 5, 8). The only exception is the combination of 3, 4, and 5, with 138 undiscriminated pairs of analytes.

All other combinations of three systems, not containing system 3, show essentially larger numbers of nondifferentiated pairs of analytes: about 200 pairs for all ternary combinations with system 4, about 600 to 700 for all combinations including system 5 (but without 3 and 4), and between 1600 and 2000 pairs for the remaining ternary combinations (without systems 3, 4, and 5).

However, the minimum number of 112 pairs, obtained for the combination of the three most favorable systems, as mentioned above, is very close to the number of 109 pairs, found as the minimum number of pairs of analytes, when all the eight electrolyte systems under consideration are com-

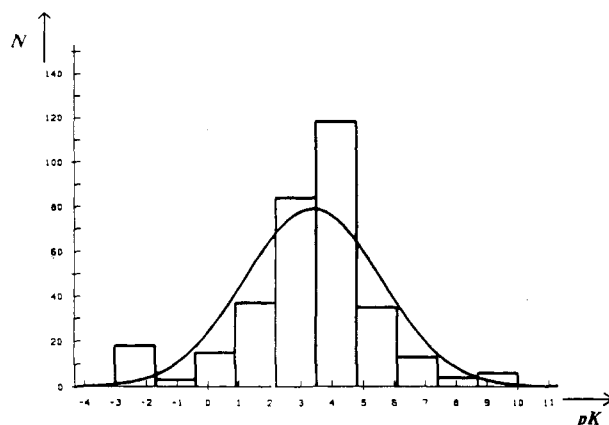


Figure 3. Distribution of the pK_a values of the solutes. Both the histogram and the fitted Gaussian curve are presented. N = number of analytes in the particular class of the histogram.

bined. This number of pairs is almost reached by the combination of the four most favorable systems: with 3, 4, 8, 9 and 3, 5, 8, 9 only 110 pairs of analytes remain undiscriminated, and with the combinations of 3, 4, 5, 8; 3, 4, 6, 8; 3, 4, 8, 10; and 3, 5, 8, 10 only 111 pairs remain.

The minimum value of 109 is already reached by the combination of five instead of eight electrolyte systems, namely with 3, 4, 5, 8, 9 and 3, 4, 6, 8, 9. It can be seen, that, compared with the combination of only two or three favorable systems, the increase in identification power is poor.

In addition it can be seen that the results of the cluster analysis are in agreement with the evaluation given above. It follows that the gain in identification power of isotachopheresis in aqueous solutions obtained by combining different leading electrolyte systems is limited: after the combination of only a few systems, no further substantial information is gained.

The results of the cluster analysis are very plausible, when the distribution of the pK_a values of the analytes is considered. The effect of the pH of the leading electrolyte and that of the sample zone, respectively, on the mobilities is most pronounced in the pH range near the pK_a value, due to the strong change of the degree of dissociation. It can be seen from the distribution of the pK_a values presented in Figure 3 that the maximum number of analytes is found at pK_a range of about 3 to 4. The largest effects on the effective mobilities therefore can be expected at the corresponding pH range of the leading electrolyte. This is in fact the result of both approaches, the cluster analytical one as well as the information theoretical one. It can be seen from Figure 3, that the standard deviation of the Gaussian distribution, fitted to the distribution of the pK_a values, is about 2 pK units. The less pronounced variation of the effective mobilities is expected therefore with electrolyte systems with pH values larger than 6. In fact, the electrolyte systems with pH 6 and higher are found in clusters with high similarity values. Therefore, the results of the cluster analysis to characterize and select leading electrolyte systems are

plausible with regard to the physicochemical behavior of the analytes in the electrolyte systems.

LITERATURE CITED

- (1) Everaerts, F. M.; Routs, R. J. *J. Chromatogr.* **1971**, *58*, 181.
- (2) Beckers, J. L.; Everaerts, F. M. *J. Chromatogr.* **1972**, *68*, 207.
- (3) Everaerts, F. M.; Beckers, J. L.; Verheggen, Th. P. E. M. *Isotachopheresis; Theory, Instrumentation and Applications*; Elsevier, Amsterdam, 1976.
- (4) Mikkers, F. E. P.; Everaerts, F. M.; Peek, J. A. F. *J. Chromatogr.* **1979**, *168*, 293.
- (5) Mikkers, F. E. P.; Everaerts, F. M.; Peek, J. A. F. *J. Chromatogr.* **1979**, *168*, 317.
- (6) Bocek, P.; Lekova, K.; Deml, M.; Janak, J. *J. Chromatogr.* **1976**, *117*, 97.
- (7) Gebauer, P.; Bocek, P. *J. Chromatogr.* **1983**, *267*, 49.
- (8) Bocek, P.; Gebauer, P. *Electrophoresis (Weinheim, Fed. Repub. Ger.)* **1984**, *5*, 338.
- (9) Gebauer, P.; Bocek, P. *J. Chromatogr.* **1985**, *320*, 49.
- (10) Krivankova, L.; Foret, F.; Gebauer, P.; Bocek, P. *J. Chromatogr.* **1987**, *390*, 3.
- (11) Kasicka, V.; Vacik, J.; Prusik, Z. *J. Chromatogr.* **1985**, *320*, 33.
- (12) Mosset, D.; Garell, P.; Desbarres, J.; Rosset, R. *J. Chromatogr.* **1987**, *390*, 69.
- (13) Kaufman, L.; Massart, D. L. In *Chemometrics, Mathematics and Statistics in Chemistry*; Kowalski, B. R., Ed.; Reidel: Dordrecht, Netherlands, 1984; pp 393-402.
- (14) De Clercq, H.; Massart, D. L. *J. Chromatogr.* **1975**, *115*, 1.
- (15) Massart, D. L.; De Clercq, H. *Anal. Chem.* **1974**, *46*, 1988.
- (16) De Clercq, H.; Massart, D. L.; Dryon, L. *J. Pharm. Sci.* **1977**, *66*, 1269.
- (17) Leary, J. J.; Justice, J. B.; Tsuge, S.; Lowry, S. R.; Isenhour, T. L. *J. Chromatogr. Sci.* **1973**, *11*, 201.
- (18) Lowry, S. R.; Tsuge, S.; Leary, J. J.; Isenhour, T. L. *J. Chromatogr. Sci.* **1974**, *12*, 124.
- (19) Haken, J. K.; Wainwright, M. S.; Do Phuong, N. *J. Chromatogr.* **1976**, *117*, 23.
- (20) Eskes, A.; Dupuis, F.; Dijkstra, A.; De Clercq, H.; Massart, D. L. *Anal. Chem.* **1975**, *47*, 2168.
- (21) De Beer, J. O.; Heyndrickx, A. M. *J. Chromatogr.* **1982**, *235*, 337.
- (22) Lowry, S. R.; Ritter, G. L.; Woodruff, H. B.; Isenhour, T. L. *J. Chromatogr. Sci.* **1976**, *14*, 126.
- (23) Massart, D. L.; De Clercq, H.; In *Advances of Chromatography*; Giddings, J. C., Keller, R. A., Eds.; Marcel Dekker: New York, 1978; Vol. 16.
- (24) Huber, J. F. K.; Reich, G. *J. Chromatogr.* **1984**, *294*, 15.
- (25) Kenndler, E.; Jenner, P. *J. Chromatogr.* **1987**, *390*, 169.
- (26) Hirokawa, T.; Nishino, M.; Aoki, N.; Kiso, Y. *J. Chromatogr.* **1983**, *271*, D1.
- (27) Kenndler, E. *Anal. Chim. Acta* **1985**, *173*, 239.
- (28) Duewer, D. L.; Harper, A. M.; Koskinen, A. M.; Fasching, J. L.; Kowalski, B. R. *ARTHUR*, Version Infomatrix: Infomatrix: Seattle, WA.
- (29) Kowalski, B. R.; Bender, C. F. *J. Am. Chem. Soc.* **1973**, *95*, 686.

RECEIVED for review May 26, 1987. Accepted September 14, 1987.

Effects of Dilution of Poly(ethylvinylbenzene-divinylbenzene) Adsorbent on the Adsorption of Aliphatic, Alicyclic, and Aromatic Hydrocarbon Adsorbates from Effective Zero to Finite Surface Coverage

N. M. Djordjevic and R. J. Laub*

Department of Chemistry, San Diego State University, San Diego, California 92182

The chromatographic measurement and systematic interpretation of the solid/gas partition coefficients K_s and related thermodynamic properties of a number of hydrocarbon adsorbates (*n*-pentane through *n*-octane, cyclohexane, methylcyclohexane, benzene, dichloromethane, chloroform, carbon tetrachloride, tetrahydrofuran, thiophene, and acetone) at effective zero surface coverage with a 1:10 admixture of the title adsorbent-inert diluent from 393 to 443 K are reported. Despite a difference of an order of magnitude in the surface areas of bulk and admixed packings, the adsorbate relative retentions were in good agreement with those found in previous work with neat Porapak Q. The heats of adsorption also coincided to within an experimental error of ca. $\pm 5\%$. The GSC technique of elution by characteristic point was then used to derive the finite-concentration adsorption isotherms and isosteric heats of adsorption of *n*-hexane, cyclohexane, benzene, carbon tetrachloride, and acetone adsorbates with "diluted" Porapak Q over the temperature range 393-443 K. All exhibited BET Type IV isotherms, as well as changes in the respective isotherm temperature coefficients. The latter is said to be a consequence of the microporous substructure of this adsorbent.

The preponderance of packed-column gas chromatographic separations is carried out today with a liquid stationary phase

(i.e., GLC), since conventional solid inorganic adsorbents (e.g., silica, alumina) generally give rise to retentions that, relatively speaking, are inordinately long. Also, because of the slow kinetics of mass transfer in gas-solid chromatography (GSC), adsorbate peaks often exhibit considerable band broadening (poor column efficiency). In addition, isotherm curvature even at very low adsorbate surface coverage can result in peak asymmetry, which reduces even further the column efficiency as well as rendering retentions dependent upon the amount of material injected.

Attempts at overcoming some of the drawbacks to analytical GSC have historically included the fabrication of "porous-layer" open-tubular (PLOT) columns, wherein either the column wall is activated or where finely divided adsorbent particles are deposited and made to adhere to it (ref 1 and 2 and references therein). The advantage in either case is said to be improvement in the mass-transfer characteristics of adsorbate/stationary-phase interactions, resulting in turn in higher chromatographic efficiency. However, there is considerable practical skill involved in the fabrication of columns of these types. Moreover, despite recent and considerable progress (3), the number of commercially available PLOT systems comprised of varieties of adsorbents is as yet somewhat limited.

As one alternative to PLOT technology, packed-column GSC efficiency can be improved somewhat, e.g. by using columns of reduced internal diameter. The system efficiency can also be enhanced by dilution by mechanical admixture