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Review

Retention indices in micellar electrokinetic chromatography

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Abstract

The use of retention indices in micellar electrokinetic chromatography (MEKC) is evaluated both from a theoretical and a practical point of view. Fundamental equations for the determination of retention indices in MEKC are described, showing that retention indices are independent of the surfactant concentration. Possibilities as well as limitations of different homologous series as reference standards are described. In addition, the practical application of retention indices for identification, investigation of solute–micelle interactions, characterization and classification of pseudo-stationary phases and determination of solute lipophilicity are discussed. © 1997 Elsevier Science B.V.

Keywords: Retention indices; Reviews; Micellar electrokinetic chromatography

Contents

1. Introduction

With the introduction of micellar electrokinetic chromatography (MEKC) by Terabe et al. [1,2] the application area of capillary electrophoresis (CE)

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was extended from charged species to neutrals. MEKC is a highly efficient separation technique, developed at the crossroads of chromatography and electrophoresis. The separation mechanism of neutral species in MEKC is based on differences in partitioning between the aqueous phase and the micellar phase (chromatographic principle). These two phases are moving with different velocities according to electrokinetic transport phenomena (electrophoretic principle). Besides neutral species, mixtures of charged and neutral compounds can also be separated by MEKC [3]. In this case differences in electrophoretic mobility as well as differences in phase distribution are exploited simultaneously to obtain separation. An important feature of MEKC is its flexibility. The composition of the electrolyte system can easily be changed by rinsing the capillary in order to control migration behaviour and optimize selectivity. In this respect the pseudo-stationary phase plays a key role, since its chemical nature has a major influence on solute–micelle interactions and consequently on the separation process. Various surfactant systems can be used as well as mixed micelles, possessing different solubilization characteristics. However, despite the ease of changing the electrolyte system, proper selection of a suitable micellar pseudo-stationary phase is still a difficult task.

Since the availability of fully automated commercial CE systems during the last decade, MEKC methods are beginning to find routine application, especially in pharmaceutical and biotechnological fields. In these areas separations of complex mixtures are often required, as well as quantitative and qualitative analyses of purified substances for quality control. These analyses require accurate methods for recording migration data and the conformation of peak identities.

In gas chromatography (GC) retention indices have found widespread application for the identification of substances in complex matrices [4–6]. They are considered to express retention with the best reproducibility and precision. In addition, retention indices form the basis of the Rohrschneider–McReynolds system of phase constants for the characterization and classification of stationary phases [4,7–9]. In reversed-phase liquid chromatography (LC) retention indices have found use in identification, characterization of separation systems, including both stationary and mobile phases, and in the investigation of solute-retention relationships [10,11].

Recently, the possibilities of a retention index scale in MEKC have been described by Muijselaar et al. [12] and Ahuja and Foley [13]. In this review various fundamental aspects as well as practical applications of the retention index concept in MEKC are summarized.

2. Retention indices in MEKC

2.1. Fundamental equations

Retention index scales in chromatography are generally based on the Martin equation [14] which states that the partition of an analyte between two phases is an additive effect, directly related to the structure of the analyte and the chemical nature of the two phases. Thus the retention of an analyte, k, is a summation of the retention of a parent compound, k_p, plus the contributions for i individual substituents $\Delta R_M(i)$ [10]:

$$\log k = \log k_p + \sum_i \Delta R_M(i) \quad (1)$$

This means that for a homologous series of reference compounds with an increasing number of methylene groups a systematic increase of log k by ΔR_{CH_2} will occur, which is referred to as the methylene selectivity. Consequently a linear relationship exists between log k and the number of carbon atoms in the homologues, z, according to:

$$\log k = az + b \quad (2)$$

Constant a represents the methylene selectivity. In MEKC a will depend on the way of solubilization of the homologues, i.e. they may be located in the hydrophobic core of the micelles or in the more polar outer region. Constant b is characteristic for the functional group of the homologues and depends on the phase ratio. In addition, both constants a and b depend on the chemical nature of the aqueous and micellar phase.

In 1958 Kováts described the basic principles for a retention index scale in gas–liquid chromatography

[15]. In this concept the retention behaviour of analytes is related to the retention of a homologous series of reference compounds. The members of this series are assigned retention index values equal to 100 times the number of their carbon atoms, i.e. $z \times 100$. The retention index of a specific solute is calculated by the logarithmic interpolation between the two neighbouring members of the homologous series, according to:

$$I = 100z + 100\frac{\log k_S - \log k_z}{\log k_{z+1} - \log k_z} \quad (3)$$

For the calculation of retention indices from migration times in MEKC, the movement of the pseudo-stationary phase has to be taken into account. The retention factor, k, is given by:

$$k = \frac{t_S - t_{EOF}}{t_{EOF}\left(1 - \frac{t_S}{t_{MC}}\right)} \quad (4)$$

where t_S, t_{EOF} and t_{MC} are the migration times of the solute, the electroosmotic flow and the micelles, respectively. Combination of Eqs. (3) and (4) leads to:

$$I = 100z + 100\frac{\log\left(\frac{t_S - t_{EOF}}{t_{MC} - t_S}\right) - \log\left(\frac{t_z - t_{EOF}}{t_{MC} - t_z}\right)}{\log\left(\frac{t_{z+1} - t_{EOF}}{t_{MC} - t_{z+1}}\right) - \log\left(\frac{t_z - t_{EOF}}{t_{MC} - t_z}\right)} \quad (5)$$

In practice, retention indices can be found graphically by interpolation and for compounds migrating faster than the first homologue by extrapolation of the equation:

$$\log k = a\frac{I}{100} + b \quad (6)$$

In principle these equations are derived for neutral species. Recently Ishihama et al. [16,17] demonstrated that retention indices can be equally well applied for charged species if the effective mobility in the aqueous phase is properly taken into account with the calculation.

2.2. Homologous series

Based on the criteria for reference compounds in LC, suggested by Smith [18] and Pacáková and Feltl [4], the following requirements were formulated for a homologous series to be applicable as retention index scale in MEKC [12]:

1. relationship between log k and the number of carbon atoms in the homologues must be linear;
2. lowest homologue should be reasonably polar, in order to obtain a wide scale of retention indices, covering the greater part of the elution window;
3. should contain a strong chromophore to detect them spectrophotometrically, as most CE instruments apply on-column UV detection;
4. should not possess an electrophoretic mobility, i.e. they should be uncharged;
5. should be readily available at reasonable price;
6. should be chemically stable in common electrolyte systems;
7. should not interact with the fused-silica capillary wall.

In Table 1 correlation data of log k versus carbon number reported in literature are listed for various homologous series in different electrolyte systems. Linear relationships according Eq. (2) were obtained in all cases.

Muijselaar et al. [12] applied n-alkylbenzenes and alkylaryl ketones as retention index standards in sodium dodecyl sulphate (SDS), cetyltrimethylammonium bromide (CTAB) and dodecyltrimethylammonium bromide (DTAB) surfactant systems. In Fig. 1 two electrokinetic chromatograms are shown of the n-alkylbenzenes and alkylaryl ketones standards, respectively, in a 50 mM SDS electrolyte system. In contrast to other chromatographic techniques where the standards form a regularly increasing scale of reference peaks across the chromatogram, in MEKC the higher more hydrophobic homologues migrate closer to each other due to the limited elution range [2]. The methylene selectivity, a in Eq. (2), was shown to be not affected by the surfactant concentration as illustrated by the constant slopes of the graphs in Fig. 2. Although the retention of the first homologue (benzene) is too strong to cover the complete elution window, the n-alkylbenzenes were found to be favourable for SDS micellar systems, which is the most widely used surfactant in MEKC. These standards are assumed to be solubilized in the hydrophobic core of the micelles. Consequently specific selective interactions with the polar head

Table 1
Correlation between log k and carbon number in MEKC according to Eq. (2) for different homologous series in various electrolyte systems reported in literature

Homologous series	Electrolyte system	a	b	r	Ref.
Alkylbenzenes, C_6–C_{10}	50 m*M* SDS, 20 m*M* Tris–boric acid (pH 8.5)	0.43	−2.54	0.9998	[12]
	50 m*M* CTAB, 20 m*M* Tris–boric acid (pH 8.5)	0.43	−2.24	0.9991	[12]
	50 m*M* DTAB, 20 m*M* Tris–boric acid (pH 8.5)	0.39	−2.29	0.9997	[12]
	50 m*M* SDS, 20 m*M* NaOH–phosphoric acid (pH 7.0)	0.42	−2.46	0.9998	[a]
	50 m*M* TDS, 20 m*M* Tris–phosphoric acid (pH 7.0)	0.41	−2.40	0.9998	[a]
	50 m*M* SDSo, 20 m*M* NaOH–phosphoric acid (pH 7.0)	0.39	−2.40	0.9998	[a]
	50 m*M* SDS–2 m*M* Brij 35, 20 m*M* NaOH–phosphoric acid (pH 7.0)	0.43	−2.51	0.9998	[a]
	50 m*M* SDS–5 m*M* Brij 35, 20 m*M* NaOH–phosphoric acid (pH 7.0)	0.42	−2.39	0.9998	[a]
	50 m*M* SDS–10 m*M* Brij 35, 20 m*M* NaOH–phosphoric acid (pH 7.0)	0.43	−2.37	0.9998	[a]
	50 m*M* SDS, 0.1 *M* borate–0.05 *M* phosphate (pH 7.0)	0.44	−2.63	0.9995	[19][b]
	50 m*M* SDS, 0.1 *M* borate–0.05 *M* phosphate (pH 7.0), 8% (v/v) MeOH	0.42	−2.54	0.9992	[19][b]
	50 m*M* SDS, 0.1 *M* borate–0.05 *M* phosphate (pH 7.0), 8% (v/v) BuOH	0.37	−1.96	0.9996	[19][b]
	50 m*M* SDS, 0.1 *M* borate–0.05 *M* phosphate (pH 7.0), 8% (v/v) BuOH, 0.82% (w/w) heptane (MEEKC)	0.40	−1.83	0.9987	[19][b]
	100 m*M* SDS, 20 m*M* Tris–boric acid (pH 8.5)	0.42	−2.48	0.9993	[20][b]
Alkylaryl ketones; C_8–C_{12}	50 m*M* SDS, 20 m*M* Tris–boric acid (pH 8.5)	0.35	−2.56	0.9998	[12]
	50 m*M* CTAB, 20 m*M* Tris–boric acid (pH 8.5)	0.39	−2.87	0.9998	[12]
	50 m*M* DTAB, 20 m*M* Tris–boric acid (pH 8.5)	0.34	−2.82	0.9999	[12]
Alkylparabens; C_8–C_{12}	100 m*M* SDS, 20 m*M* Tris–boric acid (pH 8.5)	0.32	−2.24	0.9999	[20][b]
Dansylated amines; C_{16}–C_{18}	25 m*M* SDS, 25 m*M* phosphate–0.625 m*M* borate (pH 8.0)	0.43	−4.90	0.9981	[21][b]
C_{16}–C_{19}	25 m*M* SDS, 25 m*M* phosphate–0.625 m*M* borate (pH 8.0), 10% (v/v) MeOH	0.36	−4.73	0.9999	[21][b]
C_{16}–C_{19}	25 m*M* SDS, 25 m*M* phosphate–0.625 m*M* borate (pH 8.0), 20% (v/v) MeOH	0.33	−4.84	0.9956	[21][b]
C_{18}–C_{24}	25 m*M* SDS, 25 m*M* phosphate–0.625 m*M* borate (pH 8.0), 30% (v/v) MeOH	0.31	−4.86	0.9991	[21][b]

[a] Unpublished results.
[b] Calculated from data reported in these references.

groups of the surfactant will be minimized which is advantageous if retention indices are applied to study solute–micelle interactions or for the classification of micellar pseudo-stationary phases as described in Section 3. However, a disadvantage of *n*-alkylbenzenes compared to alkylaryl ketones is their moderate UV absorbance.

Ishihama et al. [19] extended this reference scale of *n*-alkylbenzenes in microemulsion electrokinetic chromatography (MEEKC) to lower retention index values by including a polar compound (benzaldehyde) with a known I value, measured in the same electrolyte system.

Ahuja and Foley [13] also applied *n*-alkylbenzenes and alkylaryl ketones in SDS surfactant systems containing 15% (v/v) organic modifier. The *n*-alkylbenzenes were found to be too hydrophobic, but they did not include benzene in the homologous series. In addition they applied 1-nitroalkanes in SDS and mixed SDS–Brij 35 and SDS–SB-12 surfactant systems. Linear relationships between log k and z were obtained in all cases. The 1-nitroalkanes were found to be suitable standards for polar compounds.

2.3. Iterative determination of t_{MC}

Bushey and Jorgenson [21,22] used the migration data of a homologous series of dansylated amines for the iterative determination of t_{MC} in electrolyte systems containing different amounts of methanol.

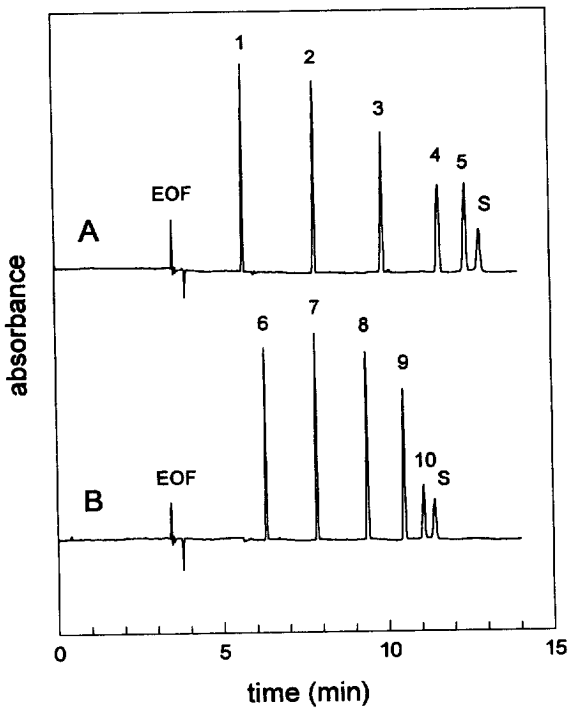

Fig. 1. Electrokinetic chromatograms of the separation of homologous series of (A) *n*-alkylbenzenes and (B) alkylaryl ketones. Reference compounds, (1) benzene, (2) toluene, (3) ethylbenzene, (4) propylbenzene, (5) butylbenzene, (6) acetophenone, (7) propiophenone, (8) butyrophenone, (9) valerophenone and (10) hexanophenone, (S) Sudan III. Electrolyte system, 50 m*M* SDS, 20 m*M* Tris–boric acid (pH 8.5). Reproduced with permission from Ref. [12]. ©1994 American Chemical Society.

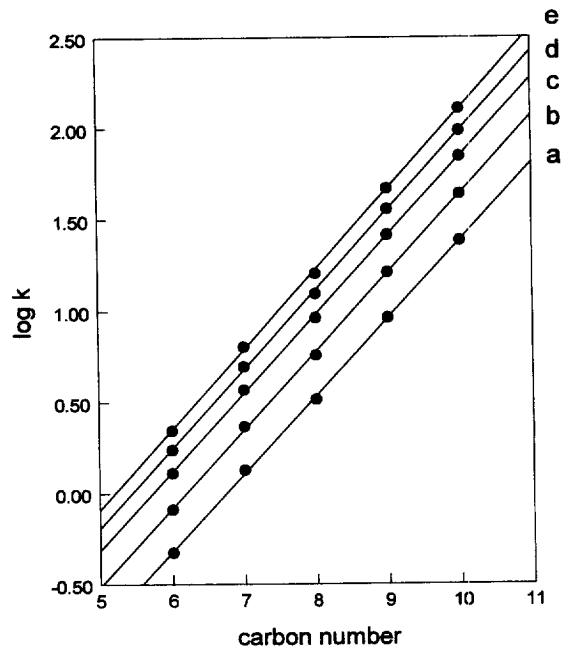

Fig. 2. Relationship between log *k* and *z* for homologous series of *n*-alkylbenzenes in a 20 m*M* Tris–boric acid (pH 8.5) electrolyte system, containing (a) 25, (b) 40, (c) 60, (d) 80 and (e) 100 m*M* SDS. Reproduced with permission from Ref. [12]. ©1994 American Chemical Society.

The methylene selectivity was shown to decrease with increasing methanol concentrations (see Table 1). In this iteration procedure the migration time of the last homologue is used as an estimation for t_{MC}. For the other homologues retention factors are calculated and a linear graph is constructed of log *k* versus *z*. From this graph *k* is determined for the last homologue and with this value a new t_{MC} is calculated using Eq. (4). Applying this t_{MC} value the retention factors of the other homologues are recalculated. This procedure is repeated until the difference in the consecutive calculated t_{MC} values is considered negligible. Since a homologous series is applied as retention index standards, the migration data of this series can be utilized for the calculation of t_{MC} with this iterative method and, in reverse, the constructed linear graph of log *k* versus *z* can be applied for the calculation of retention indices according Eq. (6). Recently, Kuzdzal et al. [20] extended this procedure to the simultaneous determination of t_{EOF} and t_{MC} from the migration data of a homologous series of *n*-alkylbenzenes or alkylparabens.

3. Applications of retention indices in MEKC

Similar to retention index scales in GC and LC [4,10], retention indices in MEKC can be applied for different purposes:

1. reproducible identification parameter
2. investigation of solute–micelle interaction phenomena
3. characterization and classification of pseudo-stationary phases
4. determination of lipophilicity and correlation with biological activity

3.1. Identification

The principle application of retention indices in both GC and LC has been for the identification of analytes [4,10]. By the use of a homologous series as internal standards, retention indices provide a migration parameter which is largely independent of the exact operating conditions. This enables the comparison of experimental results obtained with different batches of electrolyte systems or in different laboratories. For example retention indices for phenol and nitrobenzene have been reported to be 531 and 622 [12] in an electrolyte system of 50 m*M* SDS, 20 m*M* Tris–boric acid (pH 8.5), and 534 and 626 (calculated from retention data in [19]) in an electrolyte system of 50 m*M* SDS, 100 m*M* borate–50 m*M* phosphate (pH 7.0), respectively.

Retention indices were shown to provide a significant improvement in reproducibility compared to retention factors as illustrated in Fig. 3 [12]. The retention indices for the first three compounds were obtained by extrapolation. Their R.S.D. values can be improved by including a polar compound with a known *I* value in the series of reference standards [19].

Often the retention behaviour of analytes and reference standards is influenced more or less to the same extend by fluctuations in the experimental conditions, e.g. differences in organic modifier content, temperature or surfactant concentration. Consequently retention indices are less affected by these fluctuations than retention factors. In Fig. 4 the changes in retention indices and retention factors are shown for four different experimental conditions, including electrolyte systems with organic modifiers and an MEEKC system. Clearly the influence of the composition of the electrolyte system is smaller and more similar for *I* than for *k*.

The dependence of retention indices on temperature was shown to be small ($dI/dT<0.6$) [12]. Taking into account the working temperature range for MEKC experiments (typically 15–60°C) and the

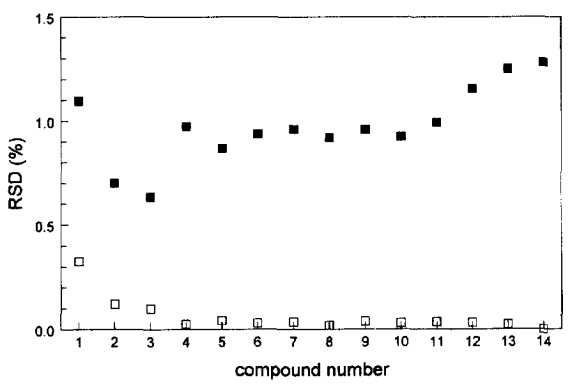

Fig. 3. Relative standard deviations, R.S.D. (%, $n=10$), for (closed symbols) *k* and (open symbols) *I* of 14 aromatic compounds. Data from Ref. [12].

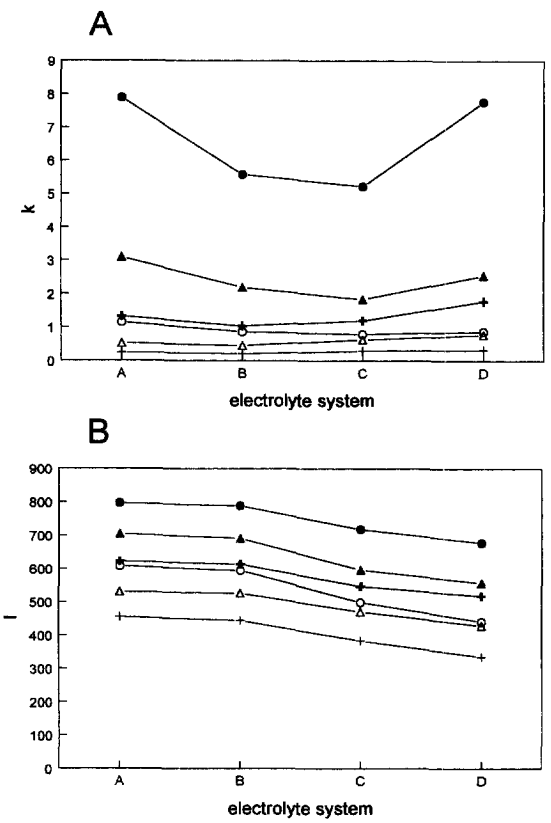

Fig. 4. (A) Retention factors and (B) retention indices for six compounds in four different electrolyte systems. (A) 50 m*M* SDS, 0.1 *M* borate–0.05 *M* phosphate (pH 7.0), (B) 50 m*M* SDS, 0.1 *M* borate–0.05 *M* phosphate (pH 7.0), 8% (v/v) MeOH, (C) 50 m*M* SDS, 0.1 *M* borate–0.05 *M* phosphate (pH 7.0), 8% (v/v) BuOH, (D) 50 m*M* SDS, 0.1 *M* borate–0.05 *M* phosphate (pH 7.0), 8% (v/v) BuOH, 0.82% (w/w) heptane (MEEKC). Data from Ref. [19].

thermoregulation of most commercial CE instruments, the influence of temperature on retention indices will be of minor importance.

Several authors demonstrated that retention indices are effectively independent of the surfactant concentration [12,13,16], as illustrated in Fig. 5. In MEKC the retention factor is related to the distribution coefficient, K, and the phase ratio, β, according to:

$$k\beta = K \tag{7}$$

The phase ratio can be calculated according to:

$$\beta = \frac{V_{AQ}}{V_{MC}} = \frac{1 - v(C_{SF} - CMC)}{v(C_{SF} - CMC)} \tag{8}$$

where V_{AQ} and V_{MC} are the volume of the aqueous and the micellar phase respectively, v is the partial molar volume of the micelles, C_{SF} is the concentration of the surfactant and CMC is the critical micelle concentration. Under practical MEKC conditions the volume of the micellar phase is small compared to the volume of the aqueous phase. Hence the numerator of Eq. (8) equals 1 and the retention factor is linearly related to the surfactant concentration according to:

$$k = Kv(C_{SF} - CMC) \tag{9}$$

Since the retention index is a relative quantity, it is independent of the phase ratio, i.e. independent of the partial molar volume of the micelles, surfactant concentration and critical micelle concentration. This can be seen after combination of Eqs. (3) and (9) which results in:

$$I = 100z + 100\frac{logK_S - logK_z}{logK_{z+1} - logK_z} \tag{10}$$

This is an important advantage compared to retention factors. In MEKC the phase ratio is proportional to the surfactant concentration in the electrolyte system. However, the dissolved amount of surfactant may differ from batch to batch. Ishihama et al. [19] demonstrated an improvement of the reproducibility using retention indices in MEEKC where the preparation of similar batches of electrolyte systems may be troublesome due to evaporation of one of the electrolyte constituents. They reported an R.S.D. (%, $n=5$) for k and I of 20.18 and 0.33, respectively, for nitrobenzene.

3.2. Solute–micelle interactions

Different methods have been employed to study the influence of surfactant structure on solute–micelle interactions and selectivity in MEKC. Terabe

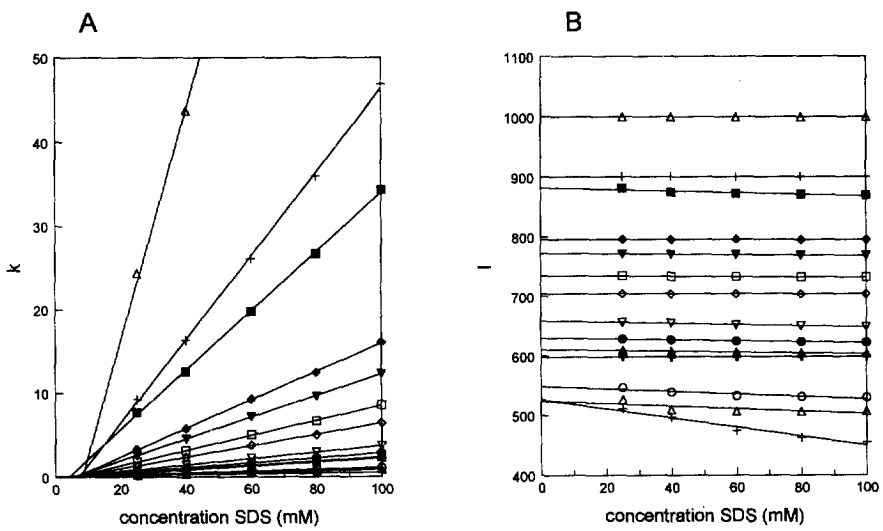

Fig. 5. (A) Retention factors and (B) retention indices versus SDS concentration for 14 aromatic compounds. Electrolyte system, 20 m*M* Tris–boric acid (pH 8.5). Reproduced with permission from Ref. [12]. ©1994 American Chemical Society.

and Okada [23] determined thermodynamic quantities of micellar solubilization. The contribution of the entropy change on selectivity was found to be significant for many compounds. Yang and Khaledi [24,25] applied linear solvation energy relationship (LSER) modelling for the characterization of solute–micelle interactions. They demonstrated that with the applied surfactant systems differences in selectivity are primarily due to their hydrogen bonding characteristics.

Muijselaar et al. [12] applied retention indices to quantify the influence of different surfactant systems on MEKC selectivity. Since retention indices are independent of the phase ratio and less influenced by the composition of the electrolyte system than retention factors (see Section 3.1), they may form the basis of reliable retention comparison in MEKC. The difference in retention indices obtained with two pseudo-stationary phases for a specific solute provides information about the interaction between the characteristic groups of both the solute and the micellar phases. In this way retention indices facilitate the classification of sample compounds in terms of functional group selectivities. This is illustrated in Fig. 6 for an SDS and a mixed SDS–Brij 35 micellar system.

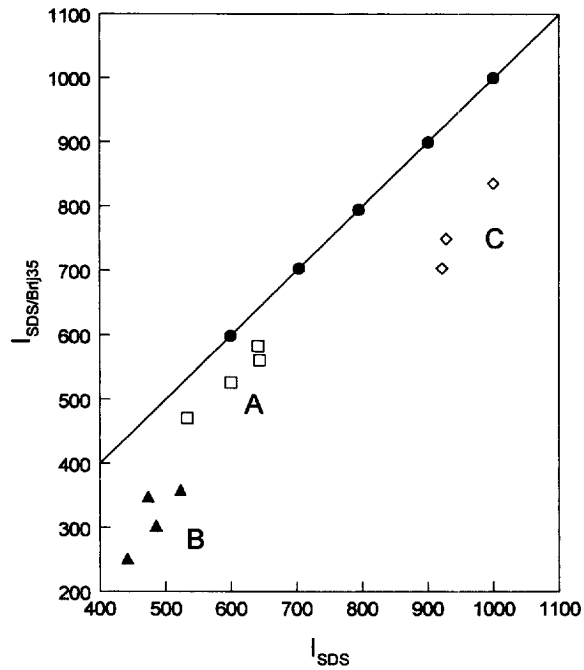

Fig. 6. $I_{SDS-Brij\ 35}$ (50 m*M* SDS + 10 m*M* Brij 35) versus I_{SDS} for (line) *n*-alkylbenzenes, (A) hydrogen bond accepting aromatic compounds, (B) xanthines and (C) corticosteroids. Data from Ref. [27].

3.3. Characterization and classification of pseudo-stationary phases

Analogous to the Rohrschneider–McReynolds scale in GC [4,7–9] and similar methods in LC [10], retention indices can be applied for the characterization of retention properties of pseudo-stationary phases in MEKC. This facilitates the classification of micellar systems according to specific selective chemical interactions. These kind of classifications may be helpful in the fast selection of a suitable micellar system for a given separation problem. Assuming that the individual intermolecular forces that contribute to solute retention are independent, ΔI_{SF} for a specific surfactant system SF can be expressed as [4]:

$$\Delta I_{SF} = I_{SF} - I_{SDS} = \sum_i (A_i X_i) \qquad (11)$$

where A_i and X_i represent factors for specific solute–micelle interactions for the solute and the micellar phase, respectively. The system constants X_i provide a quantitative characterization of different pseudo-stationary phases. Here SDS is chosen as reference system because this is the most widely used surfactant system in MEKC. Recently this approach was applied for the classification of two anionic, three mixed anionic/non-ionic and two cationic micellar systems according to their hydrogen bonding characteristics [26,27]. Acetophenone (a strong hydrogen bond acceptor) and phenol (a strong hydrogen bond donor) were applied as standard compounds in a two parameter model. In Fig. 7 the classification of these pseudo-stationary phases according their hydrogen bond donor strength (X_1) and their hydrogen bond acceptor strength (X_2) are illustrated. The results were shown to be comparable with classifications based on LSER models [27].

3.4. Correlation with n-octanol–water partition coefficients

n-Octanol–water partition coefficients, log P_{OW},

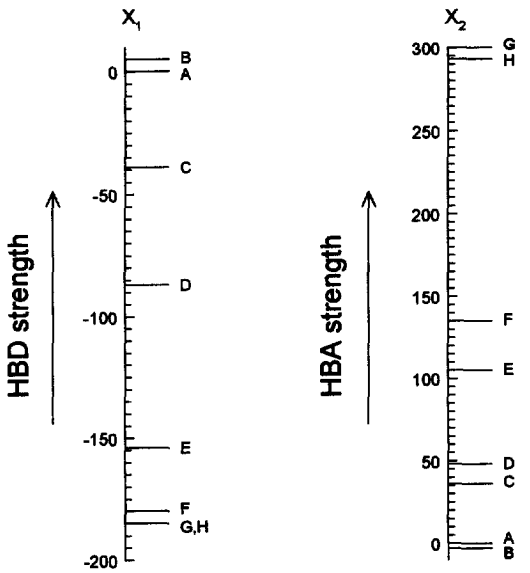

Fig. 7. Classification of eight pseudo-stationary phases according to their hydrogen bond donor (HBD) and hydrogen bond acceptor (HBA) strength, respectively. (A) 50 m*M* SDS, (B) 50 m*M* TDS, (C) 50 m*M* SDSo, (D) 50 m*M* SDS–2 m*M* Brij 35, (E) 50 m*M* SDS–5 m*M* Brij 35, (F) 50 m*M* SDS–10 m*M* Brij 35, (G) 50 m*M* CTAB and (H) 50 m*M* DTAB. Data from Ref. [27].

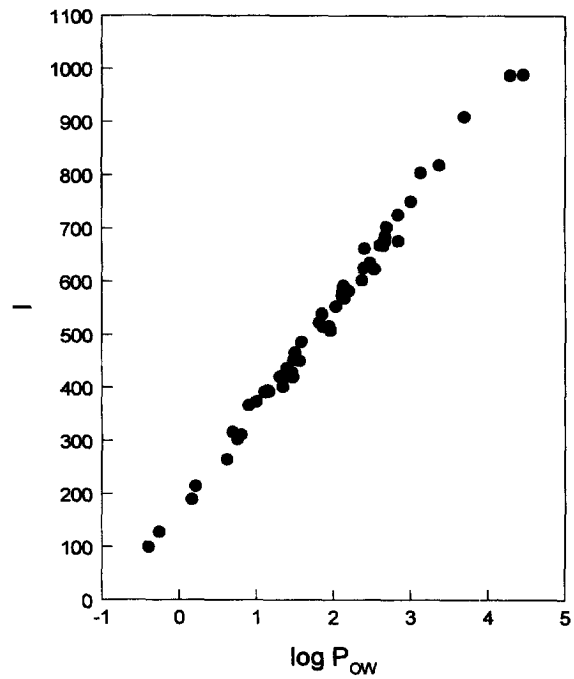

Fig. 8. Correlation between I and log P_{ow} for 53 compounds in MEEKC. Electrolyte system, 50 m*M* SDS, 0.1 *M* borate–0.05 *M* phosphate (pH 7.0), 8% (v/v) BuOH, 0.82% (w/w) heptane. Data from Ref. [19].

are frequently used as parameter for lipophilicity of substances and are applied in various disciplines such as drug design, toxicology and environmental monitoring of pollutants. Recently, several authors paid attention to log P_{ow} screening using MEKC [16,19,28–32]. This microscale separation technique offers some unique advantages such as speed, small sample size, suitability for mixtures and feasibility for automation. In reverse, the existing large data bases of log P_{ow} can be applied for the prediction of retention in MEKC analyses. If the distribution mechanism of the analytes in MEKC follows the same free energy relationship as the distribution in the *n*-octanol–water system, I and log P_{ow} are linearly related according to:

$$I = p\log P_{ow} + q \qquad (12)$$

Yang et al. [32] demonstrated that the type of surfactant has a major effect on the relationship between I and log P_{ow}. From the results of LSER modelling they concluded that these differences may be attributed to different hydrogen bonding characteristics of the micellar systems and the *n*-octanol–water system. Ishihama et al. [19] reported a high correlation ($r = 0.996$) between I and log P_{ow} for 53 aromatic sample compounds possessing different functionalities in MEEKC which is illustrated in Fig. 8.

4. Conclusions

Retention indices form a valuable way of expressing migration data in MEKC. Various homologous series can be applied as reference standards, e.g. *n*-alkylbenzenes, alkylaryl ketones or 1-nitroalkanes. Linear relationships were reported between log k and carbon number for all homologous series. This relationship can be applied for the iterative calculation of t_{MC}.

Analogous to retention index scales in GC and LC, retention indices can serve different purposes in

MEKC. They provide a reproducible identification parameter which is effectively independent of the surfactant concentration. A significant decrease in relative standard deviations was obtained compared to retention factors. In addition, they were shown to be less affected by the experimental conditions than retention factors. Comparison of retention indices obtained with different micellar systems can provide information about solute–micelle interactions and specific selectivities of pseudo-stationary phases in MEKC. Analogous to the Rohrschneider–McReynolds scale in GC, retention indices can be applied for the characterization and classification of pseudo-stationary phases in MEKC. The correlation between retention indices and *n*-octanol–water partition coefficients in MEEKC facilitates the fast determination of solute hydrophobicity with small sample volumes.

5. Symbols and abbreviations

a	constant, methylene selectivity
A	solute factor solute–micelle interaction
b	constant
C_{SF}	concentration surfactant
i	counter
I	retention index
k	retention factor
k_p	retention of parent compound
K	distribution coefficient
l_c	total length capillary
l_d	capillary length from injection to detection
R_M	contribution of substituent to retention
t_S	migration time solute
t_{EOF}	electroosmotic migration time
t_{MC}	migration time micelles
t_z	migration time standard with z carbon atoms
t_{z+1}	migration time standard with $z+1$ carbon atoms
v	partial molar volume micelles
V	voltage/volume
X	micelle factor solute–micelle interaction
z	carbon number
β	phase ratio
μ_{eff}^{ps}	pseudo-effective mobility

Brij 35	polyoxyethylene-23-lauryl ether
BuOH	butanol
CMC	critical micelle concentration
CTAB	cetyltrimethylammonium bromide
DTAB	dodecyltrimethylammonium bromide
GC	gas chromatography
LC	liquid chromatography
LSER	linear solvation energy relationship
MEEKC	microemulsion electrokinetic chromatography
MEKC	micellar electrokinetic chromatography
MeOH	methanol
NaOH	sodium hydroxide
SB-12	N-dodecyl-N, N-dimethylammonium-3-propane-1-sulfonic acid
SDS	sodium dodecyl sulphate
SDSo	sodium dodecyl sulphonate
SF	surfactant system
TDS	tris dodecyl sulphate
TRIS	tris(hydroxymethyl)amminomethane

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