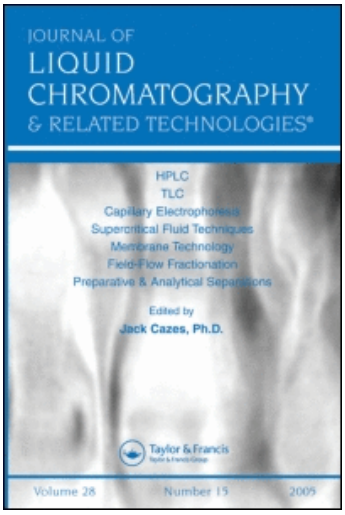

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MICELLAR ELECTROKINETIC CAPILLARY CHROMATOGRAPHY WITH CATIONIC SURFACTANTS

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ABSTRACT

A series of alkyltrimethylammonium chloride and bromide surfactants were evaluated in micellar electrokinetic capillary chromatography (MECC) of urea herbicides, alkylbenzenes and phenylalkylalcohols. The magnitude of the anodal electroosmotic flow obtained with these cationic micellar phases was largely unaffected by the length of the alkyl chain of the surfactant while the migration time of the micelle increased with decreasing the length of the alkyl tail. The net result was an increase in the retention window as the size of the alkyl tail of the surfactant decreased. The breadth of the retention window stayed almost the same when the micelle counterions were changed from chloride to bromide. At constant micellized surfactant concentration, the capacity factors of neutral solutes increased linearly with increasing alkyl chain length of the surfactant, indicating an increase in the hydrophobic phase ratio of the MECC systems. Under this condition, the value of the methylene group selectivity for the homologous solutes was largely unaffected by the length of the surfactant tail. Also, when the micellized surfactant concentration was held constant, the homologous solutes exhibited quasi-homoenergetic retention on the different cationic micellar phases. In addition, when going from a cationic surfactant to an anionic surfactant while keeping the length of the alkyl tail the same, the value of the methylene group selectivity remained unchanged, and the energetic of retention was not affected by the net charge of the micelle. The separation of a mixture of six urea herbicides was best achieved when an MECC system of low hydrophobic phase ratio and wide retention window, such

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as dodecyl- or decyltrimethylammonium chloride (DoTAC or DTAC), was used as the micellar phase. Tetradecyltrimethylammonium chloride (TTAC) micellar phase having medium hydrophobic character and narrower retention window than DoTAC or DTAC, was slightly less effective in separating the urea herbicide mixture. The overall separation of the urea herbicides could be enhanced by the inclusion of small amounts of octyltrimethylammonium chloride (OTAC) surfactant into the TTAC micellar phase. This is because the addition of OTAC to the TTAC micellar phase decreased the capacity factors and increased the breadth of the retention window.

INTRODUCTION

Micellar electrokinetic capillary chromatography (MECC), employing surfactant-rich electrolyte solutions and open tubular fused-silica capillaries, was first introduced in 1984 by Terabe *et al.* (1). MECC, which is a modification of capillary zone electrophoresis (CZE), has extended the utility of CZE to the separation of neutral solutes. Uncharged solutes are separated *via* their differential distribution between a fast moving aqueous phase and a slow moving micellar pseudo-stationary phase, and are eluted within a retention window that extends from the retention time of an unretained solute, t_0, to the retention time of another solute completely solubilized by the micelles, t_{mc}.

Thus far, most MECC separations have utilized aqueous sodium dodecyl sulfate (SDS) as the micellar phase. Other ionic surfactants, such as sodium tetradecyl sulfate (STS), dodecyltrimethylammonium chloride or bromide (DoTAC, DoTAB), cetyltrimethylammonium chloride or bromide (CTAC, CTAB) and bile salts, have been briefly explored. The types of micellar phases and their applications in MECC have been discussed in recent reviews by Terabe (2), Sepaniak *et al.* (3) and Janini and Issaq (4).

To add to the armory of useful surfactants in MECC and to further improve the methodology of the technique, our laboratory very recently introduced and evaluated alkylglucoside-borate micelles. The surface charge density of these new micellar phases can be varied conveniently by changing the borate concentration

and/or the pH of the running electrolyte (5). As a result, the retention window of the alkylglucoside-borate micellar system can be varied systematically over a wide range. These readily tuned features allowed the manipulation of resolution, separation efficiency and peak capacity. Other micellar phases based on the principle of adjustable surface charge density are being investigated in our laboratory, and the results are planned for future papers.

In this paper, our objectives entail the following: (i) to shed light on the energetics of retention of neutral solutes with various micellar phases, (ii) to examine the correlation between solute retention and the hydrophobic character of the micelles, (iii) to illustrate the dependence of the retention window of MECC on the nature of the surfactant and the composition of the aqueous phase and (iv) to provide selected MECC applications pertaining to species of environmental implications. In this regard, a series of cationic surfactants having alkyl chains of various lengths were evaluated over a wide range of conditions with different neutral homologous series and urea herbicides.

EXPERIMENTAL

Instrument and Capillaries

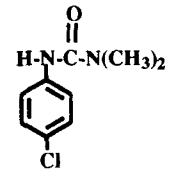
The capillary electrophoresis instrument was assembled in-house from commercially available components. It consisted of a 30-kV dc power supply of dual polarity Model CZE 1000R from Spellman High Voltage Electronics Corp. (Plainview, NY, U.S.A) and a UV-Vis variable wavelength detector Model 204 (Linear Instrument, Reno, NV, U.S.A.) equipped with a cell for on-column detection. Electropherograms were recorded and processed with Multichrom software (V1.8, VG Data Systems LTD, Cheshire, UK) *via* a VAX 4000-200 mini-computer (DEC, Maynard, MA, U.S.A.).

Fused-silica capillaries with an I.D. of 50 μm and O.D. of 363 μm were purchased from Polymicro Technology (Phoenix, AZ, U.S.A.). The total length of the capillary used in this study was either 55 or 80 cm and the corresponding separation distances were 32.5 or 50 cm.

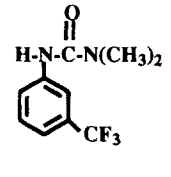
All injections were made by gravity for 10 sec at a differential height of approximately 24 cm between the inlet and the outlet buffer reservoirs. The running voltages were 10-kV for the 55/32.5 cm capillaries and 20-kV for the 80/50 cm capillaries.

Reagents and Materials

All chemicals used in this study were of analytical grade. Salts used in the preparation of electrolyte solutions were obtained from Fisher Scientific (Pittsburgh, PA, U.S.A.) and Mallinchrodt (Saint Louis, MO, U.S.A.). Urea herbicides and ethylbenzene were purchased from Chem Service (West Chester, PA, U.S.A.). The structures of the herbicides are shown below.



Monuron



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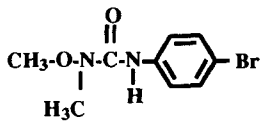
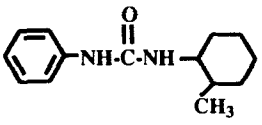
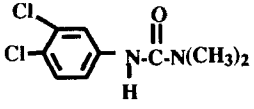


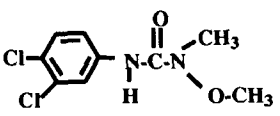
Metobromuron



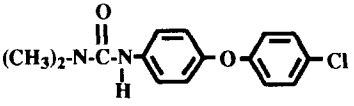
Siduron



Diuron



Linuron



Chloroxuron

Table 1 lists all surfactants used in this study with their critical micelle concentration (CMC) and aggregation number (n_{agg}). Octyltrimethylammonium chloride and bromide (OTAC and OTAB, respectively), decyltrimethylammonium chloride and bromide (DTAC and DTAB, respectively), dodecyltrimethylammonium chloride and bromide (DoTAC and DoTAB, respectively), tetradecyltrimethylammonium chloride (TTAC) and 3-phenyl-1-propanol were obtained from TCI America (Portland, OR, U.S.A.). Cetyltrimethylammonium chloride (CTAC) was from Kodak (Rochester, NY, U.S.A.). Tetradecyl- and cetyltrimethylammonium bromide (TTAB and CTAB, respectively) were from Janssen Chimica (Turnhoutseweg, Belgium). n-Propyl- and n-butylbenzene were from Alfa (Danvers, MA, U.S.A.). Methanol (used to measure t_0) and toluene were purchased from J.T. Baker (Phillipsburgh, NJ, U.S.A.). Benzylalcohol and phenethylalcohol were from Schweizerhall (South Plainfield, NJ, U.S.A.). 4-Phenyl-1-butanol, 5-phenyl-1-pentanol, 6-phenyl-1-hexanol and 7-phenyl-1-heptanol were obtained from Lancaster (Windham, NH, U.S.A.). Sudan III (used to measure t_{mc}) was purchased from Aldrich (Milwaukee, WI, U.S.A.). All solutions were filtered with 0.45 μm PTFE Titan syringe filters (SRI, Somerest, NJ, U.S.A.).

RESULTS AND DISCUSSION

Retention Window

Effect of the Length of the Alkyl Tail of the Surfactant. Fig. 1 illustrates the breadth of the retention window obtained with the alkyltrimethylammonium halide surfactants as a function of the number of carbon atoms in the tail of the surfactant molecule. As can be seen in Fig. 1, the width of the retention window increased with decreasing the length of the alkyl chain of the surfactant (6). The

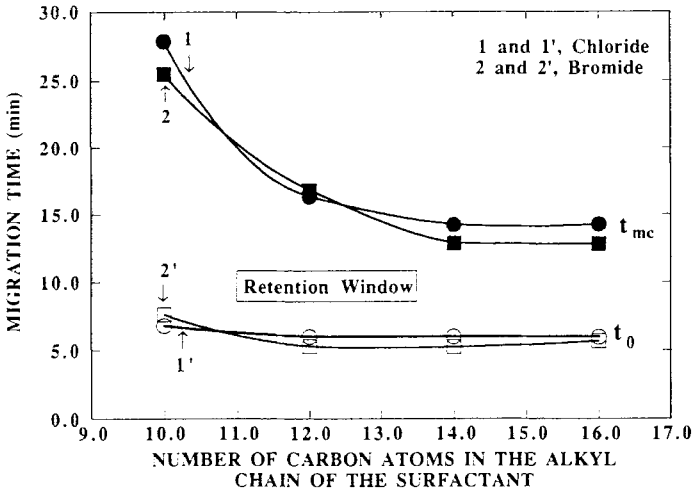


FIGURE 1. Breadth of the retention window as a function of the alkyl chain length of the surfactant molecule obtained with alkyltrimethylammonium bromide and chloride surfactants at constant micellized surfactant concentration. Capillary, fused-silica, 55 cm total length (32.5 cm separation distance) x 50 μm I.D.; running electrolyte, 50 mM phosphate, pH 7.0. In the case of alkyltrimethylammonium chloride surfactants, the running electrolytes contained 143, 102, 86.5 or 83.3 mM DTAC, DoTAC, TTAC or CTAC, respectively; in the case of alkyltrimethylammonium bromide surfactants, the running electrolytes contained 150, 98, 85.6 or 82.9 mM DTAB, DoTAB, TTAB or CTAB, respectively. Running voltage, 10 kV.

migration time of the unretained species t_0 (i.e., the magnitude of the electroosmotic velocity, v_{eo}) was largely unaffected by the length of the alkyl tail. This may indicate that the binding of the various cationic surfactants to the naked wall of the fused-silica capillary occurs to the same extent. In this binding process, it is believed that the surfactant molecules are attracted electrostatically *via* their quaternary ammonium groups to the negatively charged surface silanols, thus forming a primary hydrophobic layer. This tightly bound layer of surfactant molecules "neutralizes" the negative surface charge, and through its nonpolar chains may undergo hydrophobic interaction with the nonpolar tails of other surfactant

molecules, thus leading to the formation of a bilayer (7). In the hydrophobic bilayer, the quaternary ammonium functions of the secondary surfactant layer are oriented toward the aqueous phase. The net charge of the wall becomes positive and, consequently, under the influence of an electric field an anodal electroosmotic flow takes place (i.e., the bulk flow is toward the anode) (7-10). On the other hand, the electrophoretic mobility of the micelle v_{ep}, which is smaller in magnitude than the anodal electroosmotic flow but opposite in direction, increases with decreasing the length of the alkyl tail (i.e., decreasing the size of the micelle). The migration time of the micelle t_{mc} is given by

$$t_{mc} = l/v_{mc} = l/(v_{eo} + v_{ep}) \quad (1)$$

where l is the separation length (i.e., from capillary inlet to detection point) and v_{mc} is the net velocity of the micelle. Since the velocities v_{eo} and v_{ep} are of opposite sign and v_{eo} remains almost unchanged, as v_{ep} increases with decreasing the alkyl chain length of the surfactant, the difference between v_{eo} and v_{ep} decreases and v_{mc} decreases too. Under these conditions, and according to eqn 1, t_{mc} will keep rising, see Fig. 1. The net result is an increase in the retention window as the size of the alkyl tail decreases (6).

As seen in Fig. 1, alkyltrimethylammonium halide surfactants yielded similar retention windows. Thus, by keeping the surfactant tail the same, the micelle counterions can be changed from chloride to bromide without introducing a significant change in the retention window. The *CMC* of an alkyltrimethylammonium salt has been found to increase slightly when the micelle counterions are changed from Br^- to Cl^- (11), see Table 1. This is because the polarizability of bromide ions and, consequently, the extent of their binding to the micelle are slightly higher (11). This means that the aggregation number and, consequently, the size of the micelle remains almost unchanged for the same alkyl

Table 1. Surfactants used in this study. The values of *CMC* and n_{agg} were taken from Ref. 19.

Surfactant	Abbreviation	*CMC* (mM)	n_{agg}
Octyltrimethylammonium chloride	OTAC	-	-
Octyltrimethylammonium bromide	OTAB	140[a]	-
Decyltrimethylammonium chloride	DTAC	61[a]	-
Decyltrimethylammonium bromide	DTAB	68[a]	39[c]
Dodecyltrimethylammonium chloride	DoTAC	20[a]	-
Dodecyltrimethylammonium bromide	DoTAB	16[a]	55[c]
Sodium dodecyl sulfate	SDS	8.2[a]	64[c]
Tetradecyltrimethylammonium chloride	TTAC	4.5[a]	-
Tetradecyltrimethylammonium bromide	TTAB	3.6[a]	70[c]
Cetyltrimethylammonium chloride	CTAC	1.3[b]	-
Cetyltrimethylammonium bromide	CTAB	0.92[a]	89[c]

[a] 25 °C; [b] 30 °C; [c] 20 °C

tail. This would explain the constancy of the retention window when going from micelles with chloride counterions to micelles with bromide counterions.

pH of the Micellar Phase. To further characterize the cationic micellar phases under investigation, the breadth of the retention window was measured with CTAB surfactant over the pH range 4.5 to 9.0 where the silica surface is negatively charged. Under these conditions, the width of the retention window stayed practically unchanged as the pH of the micellar phase was varied between 4.5 and 9.0. Similar behavior could be predicted for the other cationic micellar phases, since the extent of their binding to the capillary surface was almost the same as observed from the preceding set of experiments. These characteristics of the alkyltrimethylammonium salts may prove useful when such surfactants are applied to the simultaneous separations of neutral and ionizable species. With such mixtures, the migration time of neutral solutes would be unaffected by the pH of

the micellar phase, while that of the ionizable species would vary to a large extent, thus allowing the manipulation of the resolution of the system. This type of behavior can not be attained with SDS micellar phase. In fact, at pH below 5.0-6.0, the electroosmotic flow decreases while the electrophoretic mobility of SDS micelles, which is in the opposite direction, remains the same (12). Consequently, the net mobility of the SDS micelles is opposite in direction to the electroosmotic flow (12). Thus, only those solutes that partition into the micelles can be eluted and separated.

Effect of Short Tail Surfactant Additives. Although DTAB and DTAC surfactants afforded the widest retention window (see Fig. 1), they must be used at elevated concentration since their *CMC* is relatively high, see Table 1. These conditions produce relatively high currents and, consequently, would require the use of lower running voltage or longer capillary columns. To provide an adequate retention window without producing excessive currents, an alternative to DTAB or DTAC would be to use cationic surfactants of longer alkyl chains, e.g., TTAC, in the presence of small amounts of short alkyl tail surfactant such as OTAC (i.e., mixed cationic micelles). As can be seen in Fig. 2, the inclusion of small amounts of OTAC surfactant into the TTAC micellar phase at the level of 50-100 mM increased the ratio t_{mc}/t_0 by a factor of 1.4-1.86. In addition, and as will be shown below, the addition of OTAC enhanced the resolution of late eluting peaks. The increase in the breadth of the retention window of TTAC micellar phase upon adding OTAC surfactant may be explained by the fact that a mixed micelle of smaller size was formed and, consequently, the net anodal migration velocity of the micelle decreased as the amount of added OTAC increased. This is contrary to mixed micelles involving ionic and nonionic surfactants (13), whereby a lower surface charge and a larger micelle size were obtained, which lowered the

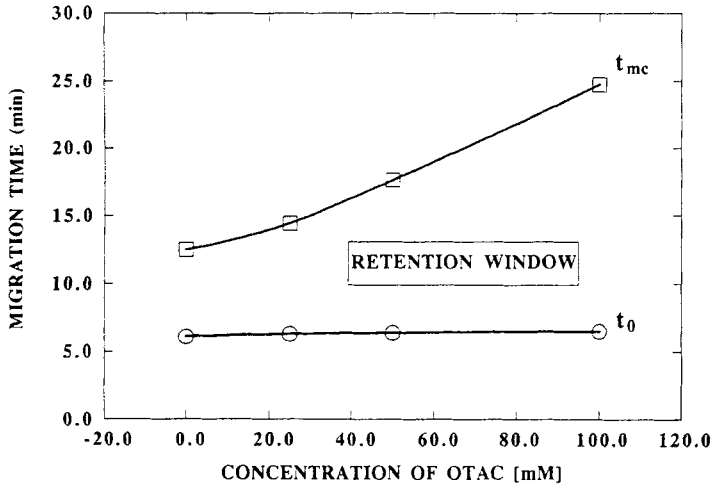


FIGURE 2. Breadth of the retention window as a function of the amount of OTAC in TTAC micellar phase. Capillary, fused-silica, 80 cm total length (50 cm separation distance) x 50 μm I.D.; running electrolytes, 50 mM phosphate, pH 7.0, containing 40 mM TTAC and 0, 25, 50 or 100 mM OTAC. Running voltage, 20 kV.

electrophoretic mobility of the micelles and caused a narrower retention window (13).

Retention Behavior of Neutral Solutes

Correlation Between Capacity Factor and Carbon Number of Homologous Series. The retention behavior of two sets of homologous series, namely phenylalkylalcohols and alkylbenzenes, was examined with the various alkyltrimethylammonium bromide and chloride micellar phases. Typical results are depicted in Fig. 3 in terms of plots of logarithmic capacity factors versus the number of carbon atoms, n_c, in the alkyl chains of the homologues. In all cases, the measurements were carried out at constant micellized surfactant concentration, $[S]-CMC$. As seen in Fig. 3, linear plots were obtained over the range studied. It

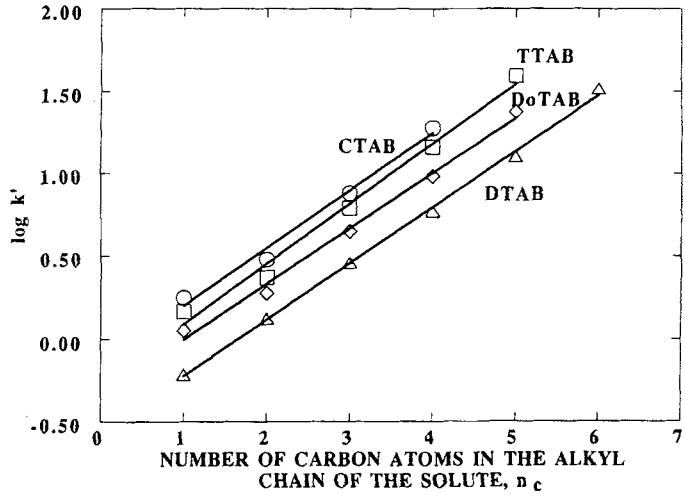


FIGURE 3. Plots of *logk'vs* n_c for phenylalkylalcohol homologous series obtained with alkyltrimethylammonium chloride surfactants at constant micellized surfactant concentration. Running electrolytes, 50 mM phosphate buffer, pH 7.0, containing 150, 98, 85.6 or 82.9 mM DTAB, DoTAB, TTAB or CTAB, respectively. Other conditions are as in Fig. 1.

should be noted that with the exception of DTAC and DTAB, the value of *logk'* of benzylalcohol solute did not align well with the rest of the homologues. This is why the R values ranged between 0.993-0.996 for surfactants having more than ten carbon atoms in their alkyl chains (see Tables 2 and 3).

Due to the stronger hydrophobic character of the alkylbenzene homologous series, these solutes were resolved only with the decyltrimethylammonium micellar phases (i.e., DTAB and DTAC) for n_c up to 4 under the experimental conditions used in this study. Plots of *logk' vs* n_c were linear with an R value of 0.999, see Table 4.

From the above results, the relationship between *logk'* and n_c seems to follow the expression normally found in reversed-phase chromatography (14)

Table 2. Slopes and intercepts of plots of $logk'$ *vs* n_c for phenyalkylalcohol homologous series obtained with the various alkyltrimethylammonium chloride micellar phases as well as with SDS. Capillary, fused-silica, 55 cm total length (32.5 cm separation distance) x 50 μm I.D.; running electrolyte, 50 mM phosphate, pH 7.0, containing 143, 102, 86.5 or 83.3 mM DTAC, DoTAC, TTAC or CTAC, respectively; running voltage, 10 kV. In the case of SDS surfactant the running electrolyte was 50 mM phosphate buffer, pH 9.2, containing 90.2 mM surfactant.

Surfactant	$log\beta$	$log\alpha$	R
DTAC	-0.652	0.318	0.999
DoTAC	-0.313	0.339	0.996
TTAC	-0.162	0.360	0.995
CTAC	-0.159	0.365	0.994
SDS	-0.348	0.346	0.999

Table 3. Slopes and intercepts of plots of $logk'$ *vs* n_c for phenyalkylalcohol homologous series obtained with the various alkyltrimethylammonium bromide micellar phases. Capillary, fused-silica, 55 cm total length (32.5 cm separation distance) x 50 μm I.D.; running electrolyte, 50 mM phosphate, pH 7.0, containing 150, 98, 85.6 or 82.9 mM DTAB, DoTAB, TTAB or CTAB, respectively; running voltage, 10 kV.

Surfactant	$log\beta$	$log\alpha$	R
DTAB	-0.565	0.340	0.999
DoTAB	-0.339	0.336	0.996
TTAB	-0.276	0.365	0.994
CTAB	-0.148	0.348	0.993

Table 4. Slopes and intercepts of plots of $logk'$ *vs* n_c for alkylbenzene homologous series obtained with DTAB and DTAC surfactants. Other experimental conditions are as in Tables 2 and 3.

Surfactant	$log\beta$	$log\alpha$	R
DTAC	0.036	0.342	0.999
DTAB	0.217	0.398	0.999

$$\log k' = (\log\alpha)n_c + \log\beta \qquad (2)$$

where the slope $\log\alpha$ is a measure of methylene or hydrophobic selectivity which characterizes nonspecific interactions, while the intercept $\log\beta$ reflects the specific interactions between the residue of the molecule and the aqueous and micellar phases. This equation implies a constant contribution to the free energy of transfer of the solute between the aqueous phase and the micellar phase with each CH_2 increment in the chain length of the homologue.

As seen in Tables 2, 3 and 4, the $\log\alpha$ values remained almost the same when varying the length of the surfactant tail. The nature of micelle counterion (i.e., chloride or bromide) does not seem to affect the value of the slope. However, as expected, the values of the intercepts increased with the length of the surfactant tail and varied slightly with the nature of the micelle counterions. These findings suggest similar physico-chemical basis for retention on the various cationic micellar phases, and the only difference is the phase ratio.

There are no substantial differences between the $\log\alpha$ values obtained with alkylbenzenes and phenylalkylalcohols with the various surfactants. This may suggest that the contribution by a methylene group to the free energy transfer of the solute between the aqueous and micellar phases is largely independent of the rest of the molecule.

When an anionic surfactant such as SDS was used, the $\log\alpha$ value was virtually the same as that obtained with the cationic surfactants, see Table 2. This indicates that changing the nature of the ionic head group from cationic to anionic while keeping the size of this group approximately the same, does not change the energetics of retention of neutral, hydrophobic compounds. This may mean that the retention of nonpolar compounds is largely due to their interaction with the hydrophobic core of the various micelles.

Comparison of the Energetics of Retention on the Various Surfactants. For two different MECC systems A and B, the logarithmic capacity factors are written as:

$$logk'_A = \phi_A - \Delta G^0_A/2.3RT \quad (3)$$

$$logk'_B = \phi_B - \Delta G^0_B/2.3RT \quad (4)$$

where R,T,ϕ and ΔG^0 are the gas constant, absolute temperature, logarithmic phase ratio and Gibbs free energy, respectively. Upon subtraction, eqns 3 and 4 can be rearranged as:

$$logk'_A = logk'_B + (\phi_A - \phi_B) + (\Delta G^0_B - \Delta G^0_A)/2.3RT \quad (5)$$

If the differences in the Gibbs retention energies of the two micellar systems is zero for all solutes, eqn 5 can be simplified to:

$$logk'_B = logk'_A - \phi_A + \phi_B \quad (6)$$

A plot of $logk'_B$ versus $logk'_A$ should give a straight line with a slope of unity and an intercept equals to the logarithm of the quotient of the phase ratios. In this case, the retention is termed homoenergetic.

On the other hand, if the corresponding Gibbs energies for the two MECC systems are proportional, such as $\Delta G^0_A = \alpha \Delta G^0_B$, where α is a constant , eqn 6 can be combined with eqns 3 and 4 to yield:

$$logk'_A = \alpha logk'_B + \phi_A - \alpha\phi_B \quad (7)$$

Equation 7 shows that when the ratio of the Gibbs retention energies in the two micellar phases is constant, linear $logk'$-$logk'$ plots with slope of α are obtained, and the retention is termed homeoenergetic. Equation 6 is a special case of eqn 7 when α is unity.

The above treatment was originally introduced by Horváth *et al.* (15) to evaluate reversed-phase chromatographic retention data obtained on various nonpolar silica-based stationary phases. The same model was also applied by El Rassi and Horváth (16) for the comparison of the reversed-phase chromatographic

properties of silica-based stationary phases which were designed for ion-exchange and hydrophobic interaction chromatography of nucleic acids. Very recently, the same treatment allowed the comparison of the energetics of retention on various zirconia-based reversed phase packings introduced by Yu and El Rassi (17).

The various surfactants were compared in terms of their energetics of retention using $logk'-logk'$ plots, *cf.* eqn 6. Typical results are depicted in Fig. 4 a and b in terms of plots of $logk'_B$ versus $logk'_A$, which show a linear correlation. The values of the slopes and intercepts are summarized in Tables 5 and 6. As seen in Tables 5 and 6, the slopes are close to unity indicating quasi-homoenergetic micellar systems. With the exception of TTAC, the hydrophobic phase ratio decreased monotonically when going from a C_{10} to C_{16} surfactant. The hydrophobic phase ratios of DTAB and DTAC are less than those of CTAB and CTAC by factors of 0.41 and 0.33, respectively, whereas the hydrophobic phase ratios of DoTAB and DoTAC are less than those of CTAB and CTAC by factors of 0.68 and 0.73, respectively. The hydrophobic phase ratio of TTAB is 0.82 that of CTAB. One interesting point is that the hydrophobic character of SDS is similar to that of a cationic surfactant of same alkyl tail, e.g., DoTAB, see Table 6 and Fig. 4b.

Correlation Between Capacity Factor and Carbon Number of Surfactant. In MECC, the capacity factor k' is given by (18):

$$k' = \varphi K \approx K\vartheta([S]-CMC) \qquad (8)$$

where $\varphi, K, \vartheta, [S]$ and CMC are the phase ratio (i.e., ratio of the volume of the micellar phase to that of the aqueous phase), solute distribution coefficient between micellar and aqueous phases, the partial specific volume of the micelle, the concentration of the surfactant and the critical micellar concentration, respectively. $[S]-CMC$ is the concentration of micellized surfactant.

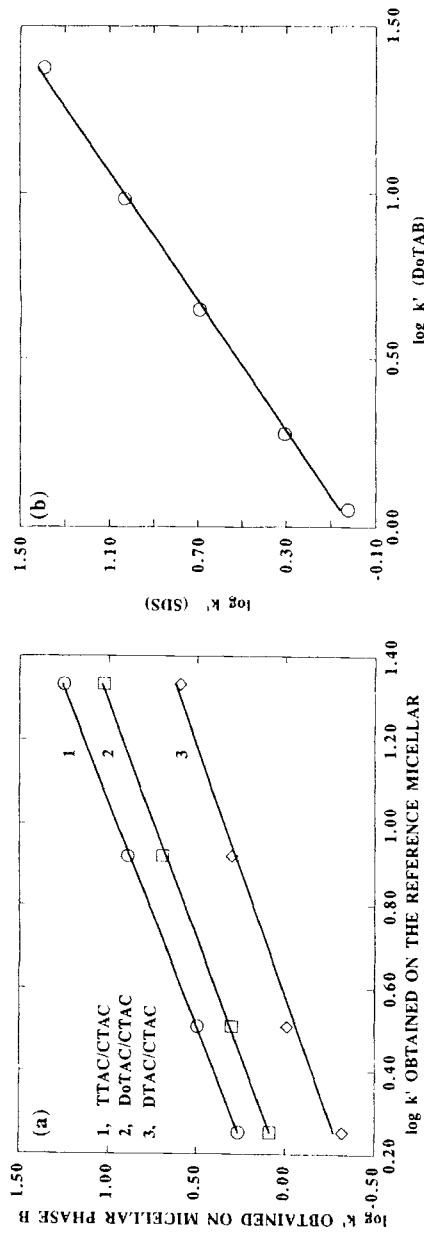


FIGURE 4. (a), Plots of *logk'–logk'* of phenylalkylalcohol homologous series obtained on one micellar phase versus another reference micellar phase at constant micellized surfactant concentration. (b), Plot of *logk'–logk'* of phenylalkylalcohol homologous series obtained on SDS versus DoTAB at constant micellized surfactant concentration. Running electrolyte, 50 mM phosphate buffer, pH 7.0, containing 98 mM DoTAC or pH 9.2, containing 90.2 mM SDS. Other conditions are as in Fig.1.

Table 5. Slopes, intercepts and antilog of intercepts (i.e., quotient of phase ratios φ_B/φ_A) of *logk'–logk'* plots of phenylalkylalcohol homologous series obtained on alkyltrimethylammonium chloride micellar phases. All experimental conditions are as in Table 2.

Micellar phase B/ Micellar Phase A	Slope	Intercept	R	φ_B/φ_A
CTAC/CTAC	1.000	0.000	1.000	1.00
TTAC/CTAC	0.920	0.03358	0.9998	1.08
DoTAC/CTAC	0.880	-0.1359	0.9994	0.73
DTAC/CTAC	0.830	-0.4798	0.9933	0.33

Table 6. Slopes, intercepts and antilog of intercepts (i.e., quotient of phase ratios φ_B/φ_A) of *logk'–logk'* plots of phenylalkylalcohol homologous series obtained on alkyltrimethylammonium bromide as well as SDS micellar phases. All experimental conditions are as in Table 2.

Micellar phase B/ Micellar Phase A	Slope	Intercept	R	φ_B/φ_A
CTAB/CTAB	1.000	0.000	1.000	1.00
TTAB/CTAB	0.979	-0.0846	0.9997	0.82
DoTAB/CTAB	0.906	-0.1639	0.9996	0.68
DTAB/CTAB	0.929	-0.3872	0.9913	0.41
SDS/DoTAB	1.027	0.0045	0.9988	1.01

According to eqn 8, at constant micellized surfactant concentration, the partial specific volume of the micelle ϑ is the parameter that determines the magnitude of the phase ratio φ of the various micellar phases under investigation. In other words, by varying the size of the alkyl tail of the surfactant while keeping $[S]-CMC$ constant, ϑ will vary and the phase ratio φ too. Under these conditions, eqn 8 can be expressed as

$$k' = \varphi K \approx (Constant) K \vartheta \qquad (9)$$

From reported values (19), the aggregation number, n_{agg}, of the surfactants under consideration is a linear function of the number of carbon atoms in the alkyl chain of the surfactant molecule, $n_{c,surf}$, see Fig. 5. This quasi-linear relationship is

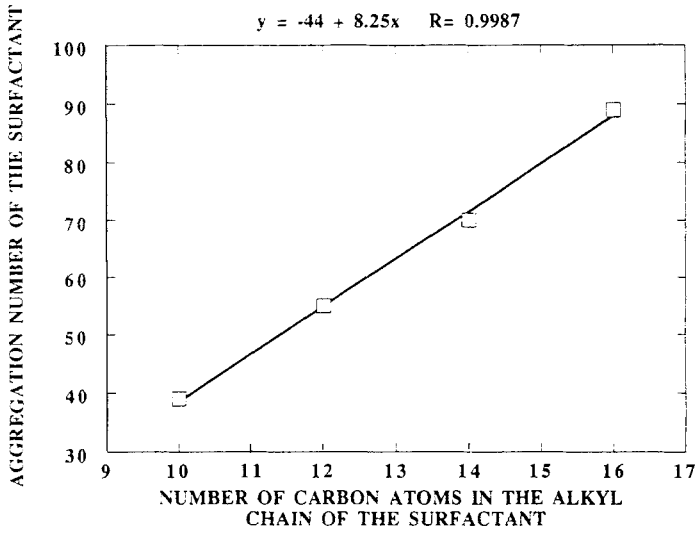


FIGURE 5. Plot of aggregation number versus the number of carbon atoms in the alkyl chain of the surfactant for alkyltrimethylammonium bromide salts. Data taken from Ref. 16.

also found with other ionic surfactants having similar alkyl tails such as sodium alkyl sulfates and sulfonates whereby the R values of plots of n_{agg} vs $n_{c,surf}$ were 0.990 and 0.988, respectively (plots not shown). Literature data on micellization (20) of neutral surfactants having aggregation numbers similar to the cationic surfactants under investigation, reveal that there is a linear correlation between the ϑ and n_{agg} of the surfactant, see Fig. 6. It follows then that the partial specific volume of the micelle would also be a linear function of the carbon number of the alkyl tail of the surfactant. Thus,

$$k' = \varphi K \approx (Constant) K \vartheta \propto K . n_{C,surf} \tag{10}$$

According to eqn 10, at constant micellized surfactant concentration, the nature of the surfactant may affect the capacity factor through its effect on either the phase ratio (i.e., ϑ or $n_{c,surf}$), the partition coefficient K, or both.

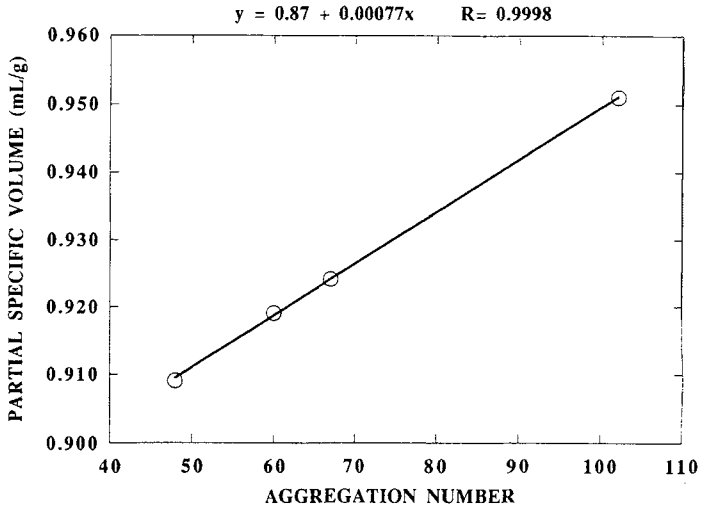


FIGURE 6. Plot of partial specific volume versus the aggregation number of methoxypolyoxyethylene decanoate and dodecanoate surfactants. Data taken from Ref. 17.

Fig. 7a and b shows plots of capacity factors of phenylalkylalcohol homologues and herbicides versus the number of carbon atoms in the surfactant molecule, respectively. As seen in Fig. 7a and b, the plots are quite linear. This may suggest that the capacity factor depends on the size of the alkyl tail of the surfactant, i.e., $n_{c,surf}$, while the distribution coefficient K remains the same. Stated differently, at constant micellized surfactant concentration, the distribution coefficient of a given solute is largely unaffected by the length of the alkyl tail and the major contributor to retention is the phase ratio (i.e., ϑ or the size of the alkyl tail $n_{c,surf}$). These findings corroborate earlier observations by Terabe *et al.* (18), in that the distribution coefficients of a series of neutral solutes were found to be almost the same with sodium dodecyl sulfate and sodium tetradecyl sulfate micellar phases.

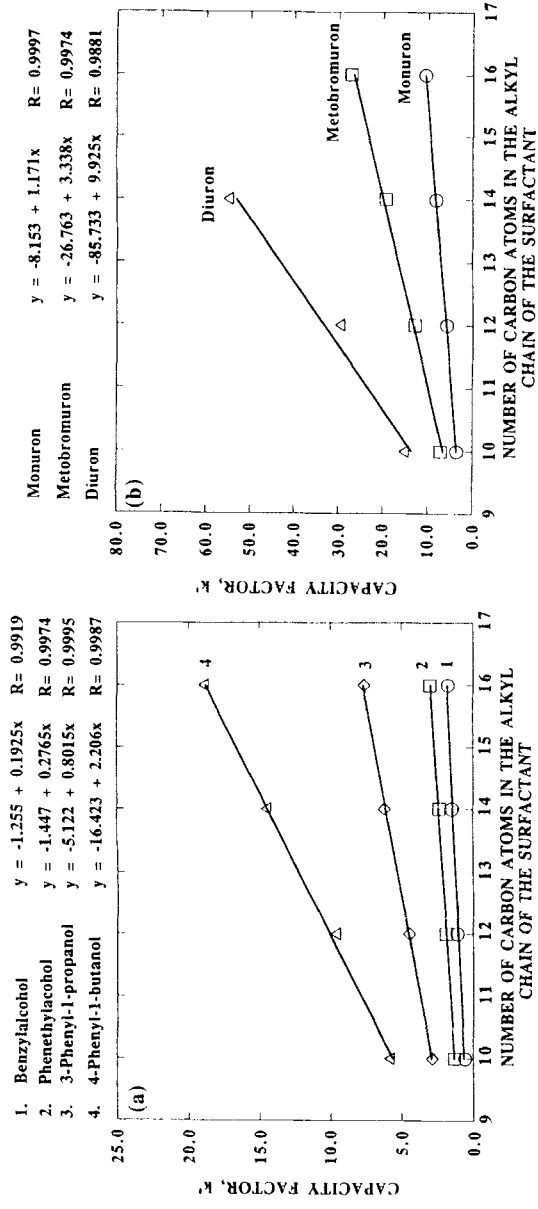


FIGURE 7. Plots of capacity factors of phenylalkylalcohols in (a) and urea herbicides in (b) versus the number of carbon atoms in the alkyl chain of the alkyltrimethylammonium bromide surfactants at constant micellized surfactant concentration. Conditions are as in Fig. 1.

Effect of Short Tail Surfactant Additive. Besides affecting the retention window (see Fig. 2), a short alkyl chain surfactant, such as OTAC, produced a monotonic decrease in the retention of neutral species when added in small amounts to the TTAC micellar phase as illustrated in Fig. 8a and b. This may be attributed to the formation of a mixed micelle of smaller size than that of the TTAC micelle. The correlation between *logk'* and the concentration of OTAC additive is quasi-linear and yields parallel lines for the homologues and the herbicides. This may mean that the addition of OTAC to the TTAC micellar phase does not produce any significant change in selectivity. Instead, the addition of OTAC resulted in enhancing the resolution of late eluting peaks (i.e., very hydrophobic solutes) by decreasing the capacity factors and enlarging the retention window of the MECC system, see below.

Selected Applications

To illustrate the potentials of the various alkyltrimethylammonium halide surfactants in MECC of neutral species of environmental significance, we have selected a series of six urea herbicides, namely monuron, fluometuron, metobromuron, siduron, linuron and chloroxuron, the structures of which are given in the Experimental.

As seen in Fig. 9a, the CTAC surfactant at a concentration of 31.2 mM (i.e., 24 times the CMC) in the running electrolyte allowed the separation of only five herbicides with an average plate count of 120,000 plates/m. At this surfactant concentration, linuron almost co-eluted with chloroxuron. At CTAC concentration below 31.2 mM, the overall separation did not improve and broad peaks were observed. This may be attributed to the fact that the packing density of the capillary column with micelles decreased at lower amount of surfactant in the running electrolyte. This would lead to longer intermicellar distances which would

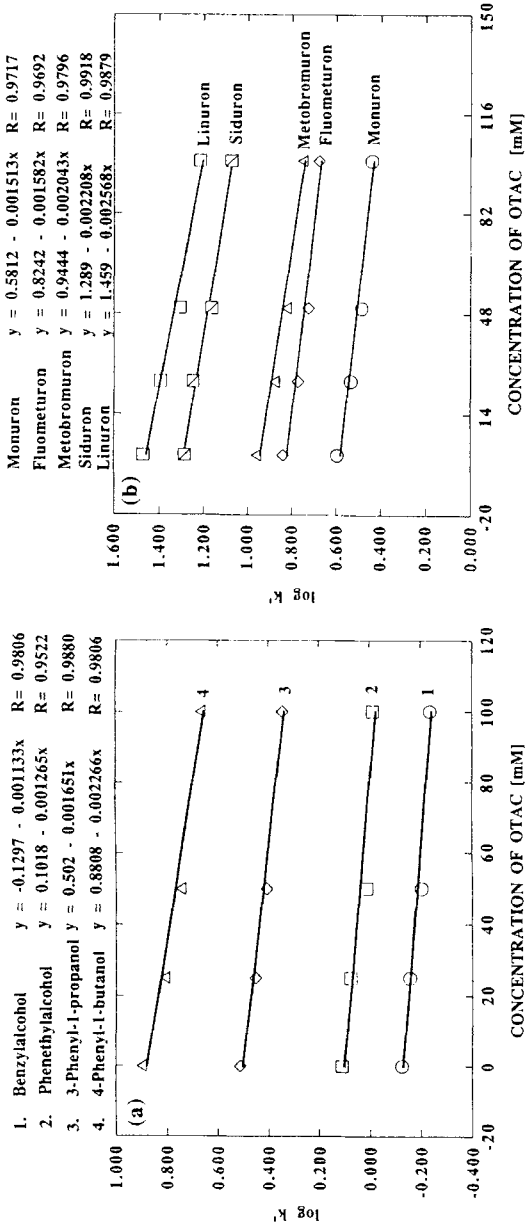


FIGURE 8. Plots of *logk'* of phenylalkylalcohol homologous series in (a) and urea herbicides in (b) versus the concentration of OTAC in TTAC micellar phase. Conditions are as in Fig. 2.

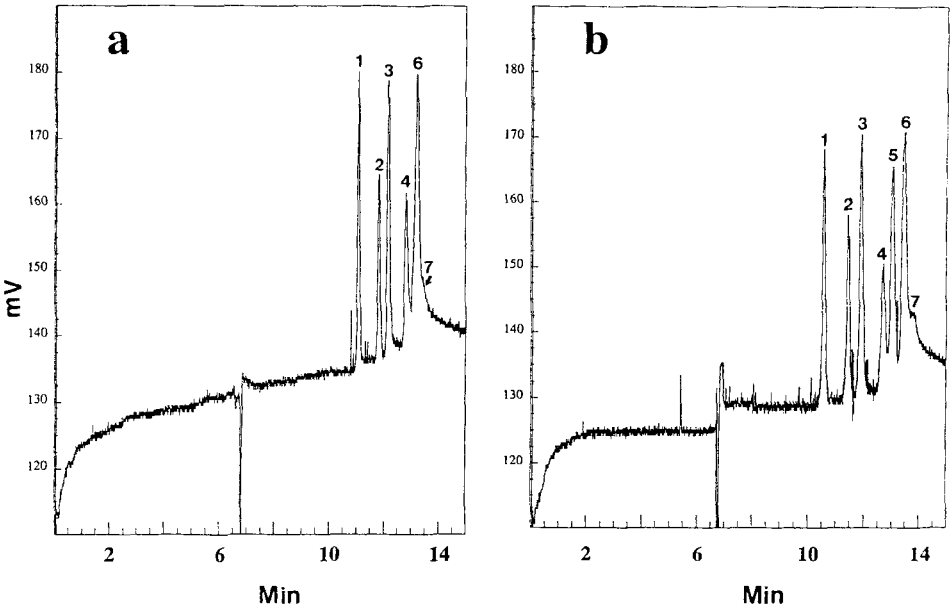


FIGURE 9. Electropherograms of urea herbicides obtained with CTAC in (a) and TTAC in (b). Capillary, fused-silica, 80 cm total length (50 cm separation distance) x 50 μm I.D.; running electrolyte, 50 mM phosphate, pH 7.0, containing 31.2 mM CTAC or 27 mM TTAC; running voltage, 20 kV; current, ca. 27 μA. Solutes: 1, monuron; 2, fluometuron; 3, metobromuron; 4, siduron; 5, linuron; 6, chloroxuron; 7, Sudan III.

give rise to slower mass transfer in the aqueous phase and concomitantly lower separation efficiencies (21).

The TTAC surfactant at a concentration of 27 mM (i.e., 6 times the CMC) in the running electrolyte proved to be useful for the separation of the herbicide mixture, see Fig. 9b. Chloroxuron was slightly resolved from the Sudan III (i.e., the migration time of the micelle), and the separation efficiency was 63% of that obtained with CTAC (N_{av} = 75,600 plates/m). Increasing the concentration of TTAC in the running electrolyte to 40 mM increased the separation efficiency by a factor of 2.65 (N_{av} = 201,000 plates/m), see Fig. 10a. This may be due to

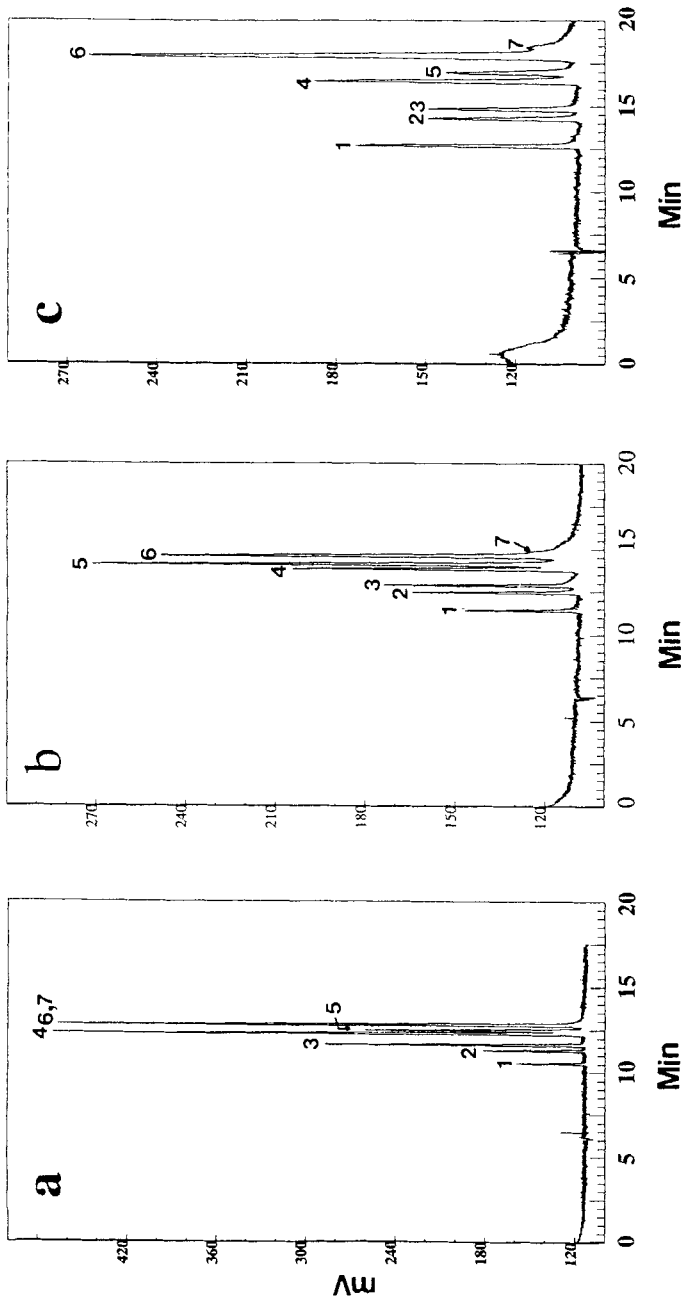


FIGURE 10. Electropherograms of urea herbicides. Running electrolytes, 50 mM phosphate, pH 7.0, containing 40 mM TTAC and 0 or 25 or 50 mM OTAC in (a), (b) and (c), respectively. Currents, 29 μA in (a), 44 μA in (b) and 60 μA in (c). Solutes and other conditions are as in Fig. 9.

shortening the intermicellar distances and, consequently, to faster mass transfer in the aqueous phase. However, at this surfactant concentration (i.e., 40 mM) chloroxuron co-eluted with Sudan III. To provide an adequate retention window, OTAC surfactant was added to the 40 mM TTAC micellar phase at concentrations of 25, 50 or 100 mM. As can be seen in Fig. 10b and c, the addition of 25 or 50 mM OTAC enlarged the retention window and, consequently, the resolution between chloroxuron and Sudan III increased, but at the expense of decreasing the overall separation efficiency by factors of 0.83 (N_{av} = 167,000 plates/m) and 2.48 (N_{av} = 84, 200 plates/m), respectively. By adding 100 mM OTAC to the TTAC micellar phase (results not shown), the chloroxuron peak was completely resolved from the Sudan III peak, but the separation efficiency decreased even further to 47,160 plates/m (i.e., by a factor of 4.26). From these results, the addition of OTAC to the TTAC micellar phase in the concentration range of 25-50 mM seems to provide an adequate retention window (also resolution) with sufficient plate count.

As shown above (*cf.* Fig. 1 and Table 5), DoTAC micellar phase exhibited a wider retention window and a lesser hydrophobic phase ratio than CTAC or TTAC. These two features of the DoTAC micellar phase yielded a better overall separation of the urea herbicide mixture than CTAC or TTAC, see Fig. 11a. The concentration of DoTAC surfactant in the running electrolyte was 80 mM (i.e., 4 times the CMC), and the average plate count was approximately 164,000 plates/m. All peaks were well resolved and the most hydrophobic herbicide (i.e., chloroxuron) was quite separated from sudan III.

Figure 11b displays the separation of the same herbicide mixture with DTAC micellar phase. As seen in Fig. 11b, the DTAC surfactant at a concentration of 170 mM (i.e., ca. 2.8 times the CMC) in the running electrolyte yielded an overall separation that was the best among the various alkyltrimethylammonium

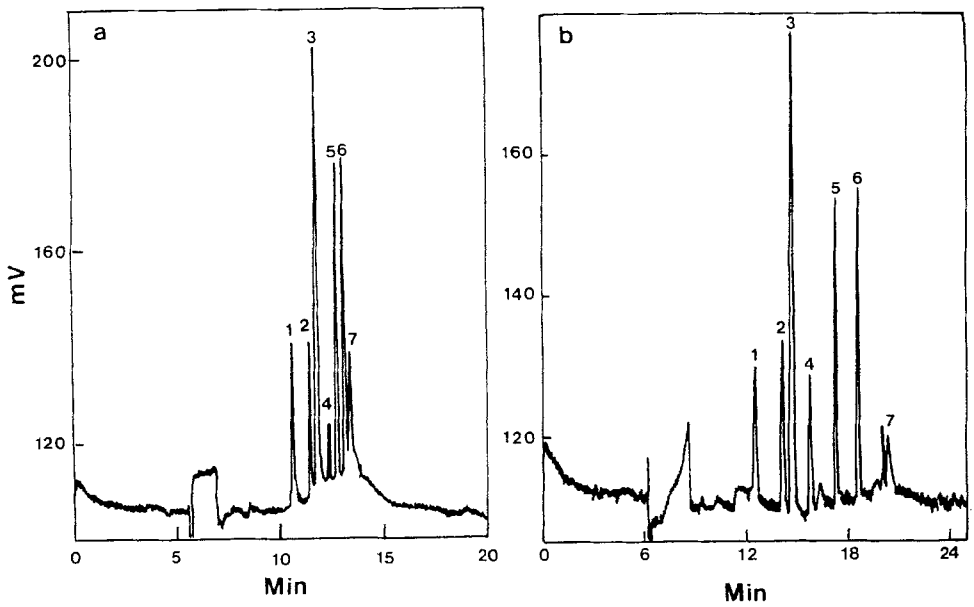


FIGURE 11. Electropherograms of urea herbicides obtained with DoTAC in (a) and DTAC in (b). Running electrolyte, 30 mM phosphate, pH 7.0, containing 80 mM DoTAC or 170 mM DTAC. Running voltages, 20 kV in (a) and 19 kV in (b); current, ca. 38 μA in (a) and 95 μA in (b). Solutes and other conditions are as in Fig. 9.

halide micellar phases. This is not surprising since DTAC afforded the widest retention window and the lowest hydrophobic phase ratio (see Fig. 1 and Table 5). The average plate count was approximately 115, 000 plates/m.

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REFERENCES

1. S. Terabe, K. Otsuka, K. Ichikama, A. Tsuchiya and T. Ando, Anal. Chem., 56, 111, 1984.
2. S. Terabe, Trends Anal. Chem., 8, 129, 1989.
3. M.J. Sepaniak, A.C. Powell and D.F. Swaile, "Fundamentals of Micellar Electrokinetic Capillary Chromatography" in Capillary Electrophoresis, Theory and Practice, P.D. Grossman and J.C. Colburn, eds, Academic Press, NY, 1992, pp. 159-189.
4. G.M. Janini and H.J. Issaq, J. Liq. Chromatogr., 15, 927, 1992.
5. J. Cai and Z. El Rassi, J. Chromatogr., 608, 31, 1992.
6. A.T. Balchunas and M.J. Sepaniak, Anal. Chem., 59, 1466, 1987.
7. A. Emmer, M. Janson, and J. Roeraarde, J. Chromatogr., 547, 544, 1991.
8. K.Otsuka, S. Terabe and T. Ando, J. Chromatogr., 332, 39, 1985.
9. T. Kaneta, S. Tanka and H. Yoshida, J. Chromatogr., 538, 385, 199.
10. X. Huang, J.A. Luckey, M.J. Gordon and R.N. Zare, Anal. Chem., 61, 766, 1989.
11. P. Mukerjee, Adv. Colloid Interface Sci., 1, 241, 1967.
12. K. Otsuka and S. Terabe, J. Microcol. Sep., 1, 150, 1989.
13. H.T. Rasmussen, L.K. Goebel and H.M. McNair, J. Chromatogr., 517, 549, 1990.
14. H. Colin, G. Guiochon, Z. Yun, J.C. Diez-Masa and P.J. Sandra, J. Chromatogr. Sci., 21, 179, 1983.
15. W.R. Melander, J. Stoveken and Cs. Horváth, J. Chromatogr., 199, 35, 1980.
16. Z. El Rassi and Cs. Horváth, Chromatographia, 19, 9, 1984.
17. J. Yu and Z. El Rassi, J. Chromatogr., in press.
18. S. Terabe, K. Otsuka and T. Ando, Anal. Chem., 57, 834, 1985.
19. M.J. Rosen, Surfactants and Interfacial Phenomena, John Wiley & Sons, New York, 2nd edition, 1988, pp.109-168.
20. K. Shinoda, T. Nakagawa, B.-I. Tamamushi and T. Isemura, Colloidal Surfactants, Some Physicochemical Properties, Academic Press, New York, 1963, pp. 114-117.
21. M.J. Sepaniak and R.O Cole, Anal. Chem., 59, 472, 1987.