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Mechanism of Electrokinetic Separations of Hydrophobic Compounds with Sodium Dodecyl Sulfate in Acetonitrile−Water Mixtures

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The effect of the addition of 0−70% (v/v) acetonitrile (AcCN) as organic modifier, into a buffer solution containing 0−60 mmol L^{-1} sodium dodecyl sulfate (SDS), on the electrokinetic behavior of neutral hydrophobic compounds has been studied experimentally. The effective mobilities of the hydrophobic compounds sharply decreased above a certain AcCN concentration, depending to some extent on the SDS concentration. In solutions with up to 40% AcCN, with SDS concentrations below the literature values of the critical micelle concentration, neutral hydrophobic analytes still showed a considerable electrophoretic mobility. The observed mobilities were too high to be attributed to solvophobic interaction of the analytes with single SDS ions. From conductivity measurements, a strong indication was found that increasing the fraction of AcCN in the SDS solution does not lead to a complete disintegration of the micelles. Even with high AcCN concentrations, the mechanism of the separation was found to be a distribution between the aqueous phase and micelle-like SDS aggregates. The influence of the addition of AcCN on the mobilities could be explained as the combined effect of a decrease of the micellar volume fraction, a change of the micellar properties, and a decrease of the distribution constants of the analytes.

Capillary electrophoresis (CE) is a very powerful technique for the separation of charged analytes.[1] With the introduction of micellar electrokinetic chromatography (MEKC),[2] the high separation efficiency of CE became accessible for the separation of neutral compounds. In MEKC, an ionic surfactant is added to the background electrolyte (BGE) in a concentration higher than the so-called critical micelle concentration (cmc). Charged micelles are formed in the solution that migrate differently from the aqueous phase and thus can act as a pseudostationary phase. When an electric field is applied, a separation can be obtained for compounds that distribute differently between the micellar and the aqueous phase.

Over the years, MEKC has developed into one of the most successful electrophoretic separation techniques. The addition of a micelle-forming surfactant to the BGE in CE has even become routine for the separation of (partly) ionized compounds, to give additional selectivity in the separation. The most commonly used surfactant for MEKC is sodium dodecyl sulfate (SDS). It has a fairly low cmc (∼8 mmol L^{-1} in pure water), so that the

conductivity of the BGE can be kept low. The SDS micelles migrate against the electroosmotic flow (EOF) in the capillary, thereby giving a relatively wide elution window for neutral compounds. Although the nucleus of the SDS micelles is strongly hydrophobic, even rather polar compounds are absorbed to some extent and can be separated. On the other hand, for very hydrophobic compounds, MEKC with SDS is often insufficiently selective.[3,4] The hydrophobic compounds tend to be absorbed virtually completely into the micelles; in that case the selectivity is lost because all compounds are migrating with the velocity of the micelles. Several methods have been proposed to expand the application range of MEKC to more hydrophobic compounds: the use of surfactants with a hydrophobicity lower than SDS such as cholic acid or derivatives,[5] or the addition of compounds to the BGE that interact with the analytes in the aqueous phase such as cyclodextrins,[6] can be used to introduce a shift in the distribution equilibrium toward the aqueous phase. Also, a frequently applied procedure is to change the solvent strength of the aqueous phase by the addition of organic modifiers such as methanol,[7−9] AcCN,[7,10,11] or urea to the BGE.[12] Addition of the organic solvents can improve the solubility of the solutes in aqueous solution, but at the same time, it can influence the micellar properties and thereby the mechanism of separation.

The exact mechanism of the micellar electrokinetic separation of hydrophobic compounds in BGEs containing organic solvents is still not clear. While some authors assume that the distribution between an aqueous and a micellar phase is still the underlying principle of the separation,[10] others argue that at higher organic solvent contents the cmc is strongly increased, and micelles are not formed with the surfactant concentrations commonly used. They attribute the separations observed under these conditions to the so-called solvophobic interaction between the analyte molecules and individual surfactant ions.[11,13,14]

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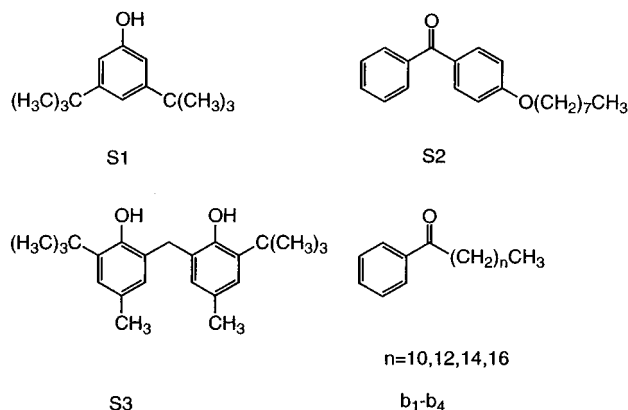(14) Walbroehl, Y.; Jorgenson, J. W. *Anal. Chem.* **1986,** *58,* 479−481.

Figure 1. Structural formulas of the solutes used.

In the work presented in this paper, we have tried to shed some light on this question. The study involved the measurement of the migration behavior of a number of hydrophobic antioxidants and alkyl aryl phenones as a function of the SDS and AcCN concentration. Also, the conductivities of the applied BGE solutions were determined in order to get some information about the micelle formation.

EXPERIMENTAL SECTION

Apparatus. Experiments were performed with a Prince CE injection system (Lauer Labs, Emmen, Netherlands) in combination with an on-column UV detection system (Spectroflow-757, Kratos Analytical, Ramsey, NJ). UV absorption detection was performed at 280 nm for the antioxidants and at 244 nm for the alkyl aryl phenones. Fused-silica capillaries of 50 μm i.d., purchased from Polymicro Technologies (Phoenix, AZ), with a total length of 90 cm and 60 cm to the detector window, were used. The applied voltage was 20 kV. Conductivity measurements were performed with a digital conductometer (Consort K720, Turnhout, Belgium).

Chemicals and Solutions. 3,5 Di-*tert*-butyl phenol (S1) was obtained from Koch Light Labs. (Colnbrook, Bucks., UK); 2-hydroxy-4-(octyloxy)benzophenone (S2), 2,2′-methylenebis(6-*tert*-butyl-4-methylphenol) (S3), and alkyl aryl phenone homologues (b$_1$–b$_4$) were obtained from Aldrich (Milwaukee, WI). Figure 1 shows the structural formulas of these compounds. Stock solutions of the analytes were prepared in pure AcCN and diluted with the BGE before injection. Solute concentrations in each experiment were at or below 10^{-4} mol L^{-1}. Other chemicals obtained from various suppliers were of analytical grade purity.

The BGEs used contained 10 mmol L^{-1} tris(hydroxymethyl)-methylammonium chloride (Tris-HCl) and 8.3 mmol L^{-1} tris-(hydroxymethyl)aminomethane (Tris) (pH = 8 measured in water), 0–60 mmol L^{-1} SDS, and 0–70% (v/v) AcCN. Since AcCN influences the pH, the value of pH = 8 has to be considered as an operational value for the BGE.

Procedures. New capillaries were successively flushed for 20 min with 1 mol L^{-1} KOH, 0.1 mol L^{-1} KOH, water, and finally buffer solution. Before each experiment, the capillary was rinsed with 0.1 mol L^{-1} KOH, water, and buffer solution for 2, 3, and 5 min, respectively. It was found that the outer polyimide coating of the capillary was attacked in BGE solutions containing AcCN, which disturbed the measurements seriously. Therefore, the outer polyimide coating was removed at both ends of the capillary

(over ~2 cm) by means of a heating filament. All running buffer solutions were filtered through 0.45 μm membrane and degassed with helium gas for 3 min prior to use. All measurements were performed at 25 °C.

RESULTS AND DISCUSSION

Effect of SDS and AcCN Addition on the Separation System. In preliminary measurements, it was observed that with a BGE containing only Tris-HCl/Tris buffer, pH = 8, all selected analytes migrated with the electroosmotic velocity. From this finding it can be assumed that these hydrophobic compounds have no electrophoretic mobility. Addition of 15–60 mmol L^{-1} SDS to the BGE appeared to shift the effective mobilities (μ_{eff}) of the solutes considerably. The solutes had all the same effective mobilities (4.0×10^{-8} m^2 V^{-1} s^{-1}), irrespective of the SDS concentration, except with 15 mmol L^{-1} SDS, when the mobility of S1 was slightly lower than that of the other compounds. The investigated SDS concentration range is above the cmc in aqueous solutions (8 mmol L^{-1}). This migration behavior indicates that all solutes are largely distributed into the micellar phase and migrate approximately with the mobility of the micelles. In order to improve the separation, the distribution of the solutes toward the micellar phase has to be decreased. This can be realized by making the BGE solution less polar by adding an organic solvent, such as AcCN, to the BGE. The addition of an organic solvent to the BGE will favor the distribution of the solutes toward the aqueous phase but may also influence the electroosmotic flow and the structure of the micelles. The effects of the addition of an organic solvent was studied with AcCN.

The AcCN content in the BGE has a distinguished effect on the electroosmotic flow. The electroosmotic mobility decreases in a linear fashion with the AcCN content, from 6.77 to 5.03 $\times$ 10^{-8} m^2 V^{-1} s^{-1}, going from 0 to 40% AcCN. The electroosmotic mobility (μ_{eo}) depends on the dielectric constant (ϵ) and the viscosity (η) of the BGE and on the zeta potential (ζ) of the wall according to

$$\mu_{eo} = (\epsilon\epsilon_0/\eta)\zeta \tag{1}$$

where ϵ_0 is the permittivity of free space. According to Cunningham et al.,[15] the ratio ϵ/η for water–AcCN mixtures is fairly constant. Therefore, it can be concluded that a change of the ζ occurs when AcCN is added to the BGE.[10,16]

The width of the peaks in electropherograms and the background noise also appear to be influenced by addition of AcCN. Plate numbers for the test solutes typically increases from 100 000 with 20% AcCN to 200 000 with 40% AcCN. Figure 2 shows two representative electropherograms of the test solutes obtained with 40% (v/v) AcCN and 15 mmol L^{-1} SDS. At higher AcCN contents (50–70%), excessive background noise emerged on the base line after a few minutes from the starting point. Similar observations have been made by Vindevogel and Sandra.[10] The noise generation coincides with a gradual decrease of the electrophoretic current (see Figure 3). The noise generation and the current drop start earlier after the start of the electrophoresis with increasing AcCN content. Thorough degassing of the running buffer solution

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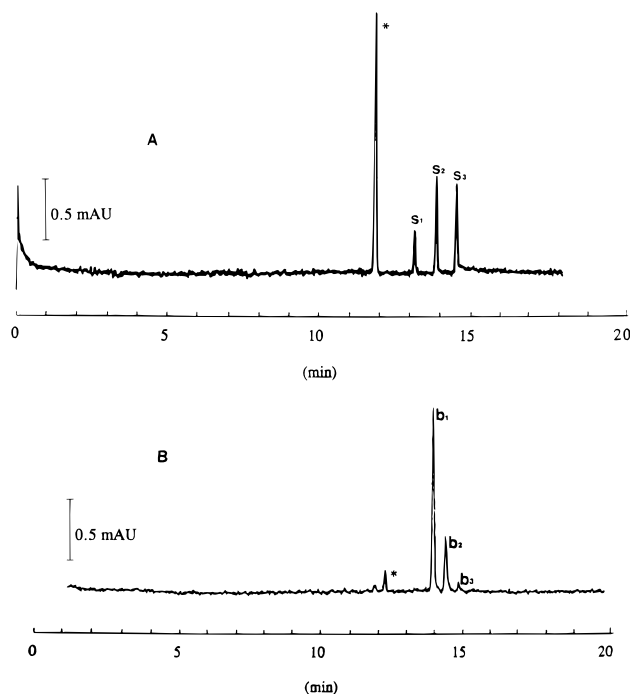
Figure 2. Representative electropherograms of the separation of the hydrophobic antioxidants (A) and alkyl aryl ketones (B). BGE composition: Tris buffer (pH = 8.0), 15 mmol L^{-1} SDS, and 40% (v/v) AcCN. *, acetone peak.
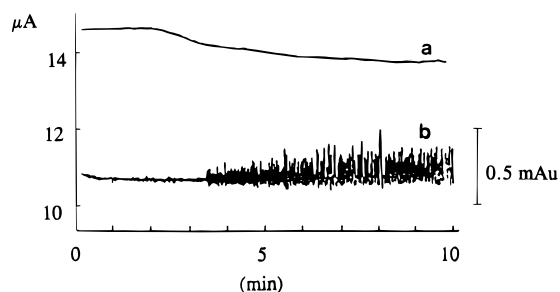

Figure 3. Typical current (a) and detector response (b) obtained when the BGE contains 50−70% (v/v) AcCN and 15−60 mmol L^{-1} SDS.

before the experiments did not help to prevent it. It was noticed that, with solutions containing 50−70% AcCN without SDS, or with 70% AcCN and only 15 mmol L^{-1} SDS, excessive noise and the drop in the current were not observed. Since all solutions were completely clear, it seems that under the influence of the electrophoretic current tiny air bubbles are formed in the solution at higher AcCN contents in the presence of SDS. Pressurizing both electrode reservoirs, as applied in electrochromatography, may give a remedy for these phenomena. Although the excessive noise may prevent the application of a high AcCN content in quantitative work, it was still possible to study the electrokinetic behavior of the model compounds, since the peak migration times could still be established accurately.

Effect of SDS and AcCN on the Effective Mobilities of Hydrophobic Compounds. The effective mobilities of the hydrophobic antioxidants measured at four SDS concentrations as a function of the AcCN content are shown in Figure 4. The same behavior is found with all SDS concentrations. The effective mobilities decrease gradually up to 20% (v/v) AcCN, then decrease sharply in the range 20−40% (v/v) AcCN, then decrease again gradually with increasing AcCN content, and finally become very

small at 70% (v/v). In the same figure it, can be seen that the highest selectivity for the solutes is obtained with 20−40% (v/v) AcCN. Similar behavior was observed with the alkyl aryl ketons as solutes.

The sharp decrease of the effective mobilities in the range 20−40% AcCN indicates that a significant change of the capacity factors occurs. As can be seen from Figure 4, this change is rather sharp at low SDS concentration and slightly more gradual over the AcCN range with increasing SDS concentration. Still, the effect of the SDS concentration is not strong. This is illustrated also in Figure 5, showing the effect of the SDS concentration at different AcCN contents on the effective mobility of dodecanophenone. With up to 20% of AcCN, the effect of the SDS concentration on the mobilities found was small. Even with 30% AcCN, the observed effective mobilities approach a value of the same order of magnitude as the micellar mobility in purely aqueous solutions. Moreover, the mobilities of the test solutes, with different sizes and hydrophobicities, all tend to the same value at higher SDS concentrations. An example of this is shown in Figure 6. For a solution with 30% AcCN (Figure 6a), the effective mobilities all approach a same plateau value at high SDS concentration. With 40% AcCN, such a plateau is not reached within the SDS concentration range experimentally accessible (Figure 6b). However, the same trend of converging mobilities can be discerned. According to the literature, the addition of AcCN strongly increases the cmc.[17,18] On basis of the literature data, it should be assumed that, with SDS concentrations up to 60 mmol L^{-1}, micelles are completely disintegrated in 30% AcCN and only individual SDS ions are present in the solution. Surprisingly, the neutral test solutes used in this study appear to still have a significant effective mobility under these conditions. This indicates that still other separation mechanisms would contribute to the migration of neutral solutes in SDS solutions containing organic solvents.

Solvophobic interaction between the individual SDS ions and the neutral solutes has been advocated to explain this behavior.[11] However, the observed analyte mobilities seem too high to be attributed to a solute−SDS−ion complex. This conclusion is supported by the comparison of the effective mobilities of the individual SDS ions and the observed maximum effective mobilities of the solutes. The mobility of SDS ions was determined by injecting SDS at low sample concentration (2 mmol L^{-1}) in a BGE containing 0−40% (v/v) AcCN. The SDS peak was detected indirectly by using potassium benzoate as the UV-absorbing compound. The results are given in Table 1. When only solvophobic interactions with single SDS ions would have been involved, the effective mobilities of the solutes would be significantly smaller than that of the separate SDS ions. However, the effective mobilities appear to be larger, at least with AcCN contents up to 40%. The experiments described above seem to support the conclusion that SDS micelles are present in 0−40% (v/v) AcCN solutions.

The possible existence of micelles cannot be shown simply with the help of a micellar marker as normally used in MEKC. Such markers (e.g., Sudan III or Yellow OB) have a lower hydrophobicity than some of the analytes studied here. Therefore, in solutions with a high AcCN content, there is no certainty that

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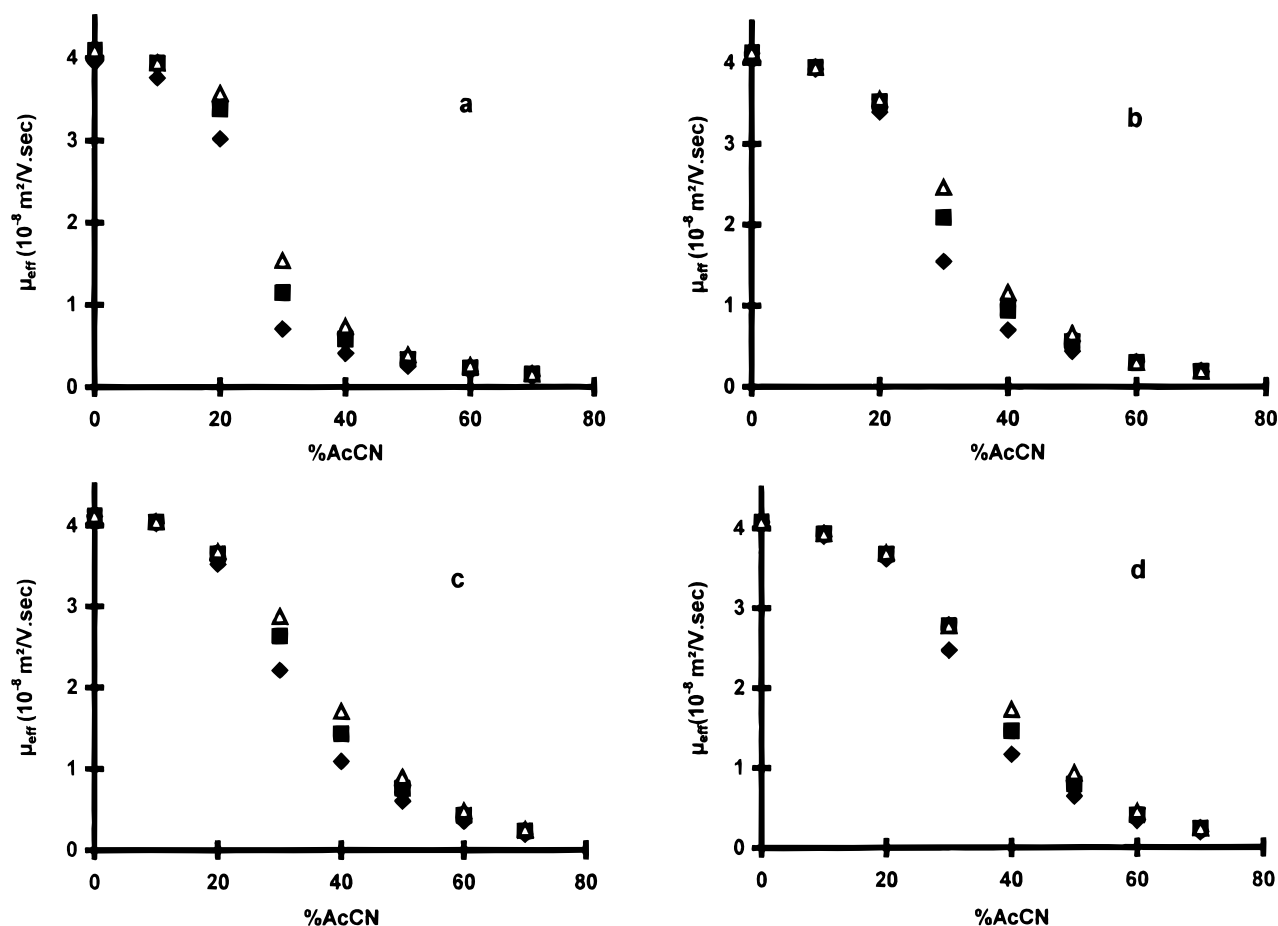(18) Misra, P. K.; Mishra, B. K.; Behra, G. B. *Colloids Surf.* **1991**, *57*, 1−10.

Figure 4. Effect of AcCN on the effective mobilities of the antioxidants S1 (◆), S2 (■) and S3 (△). SDS concentration: (a) 15, (b) 30, (c) 45, and (d) 60 mmol L^{-1}.
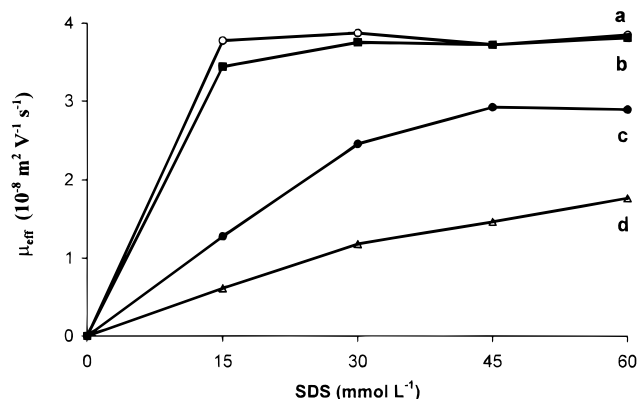

Figure 5. Effect of the SDS concentration on the effective mobility of dodecanephenone (b$_1$) with an AcCN content of 10 (a), 20 (b), 30 (c), and 40% (d).

Figure 6. Effect of the SDS concentration on the effective mobilties of all test compounds, with (A) 30 and (b) 40% (v/v) AcCN in the solvent. Test compounds: S1 (○), S2 (■), S3 (△), b$_1$ (●), b$_2$ (*), b3 (▲) and b4 (□).

they will indicate the mobility of the micellar phase, when such a phase would exist. In order to gain more information about the mechanism of the separations, the conductivities of SDS solutions were determined.

Conductivity of SDS Solutions. For the determination of the cmc of a surfactant in a particular solvent, many methods are available, based on, for instance conventional light scattering,[19] quasielastic light-scattering,[20] static or time-resolved fluorescence,[21]

or conductivity measurements.[22] A common feature of these methods is that the result obtained can strongly depend on the method used. Misra et al.[18] have determined the cmc of SDS in

(19) Mysels K. J.; Princen, L. H. *J. Phys. Chem.* **1959**, *63*, 1696.
(20) Mazer, N. A.; Benedek, G. B.; Carey, M. C. *J. Phys. Chem.* **1976**, *80*, 1075–1084.
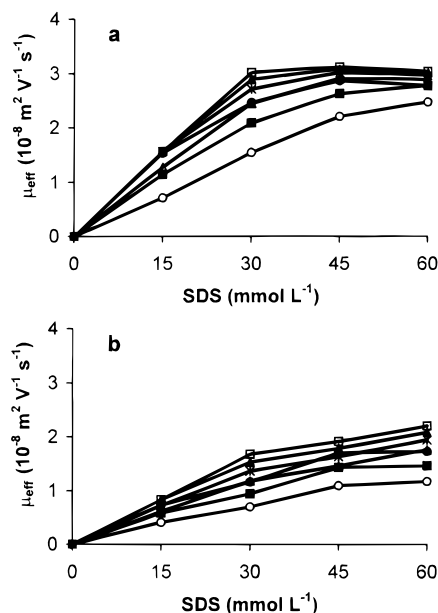(21) Turro, N. J.; Yekta, A. *J. Am. Chem. Soc.* **1978**, *100*, 5951.

(22) Goddard, E. D.; Benson, G. C. *Can. J. Chem.* **1957**, *35*, 986–991.

Table 1. Effective Mobility of Individual SDS Ions (μ_{SDS}) and the Maximum Effective Mobility of Hydrophobic Analytes (μ_{max}) Observed in BGEs Containing AcCN and SDS

% AcCN (v/v)	μ_{SDS}	μ_{max}	% AcCN (v/v)	μ_{SDS}	μ_{max}
0	2.68	4.12	30	2.04	3.12
10	2.14	4.04	40	2.21	2.20
20	2.03	3.81			

Table 2. Assessment of the Critical Micelle Concentration (cmc) of SDS at Various AcCN Contents

% AcCN (v/v)	line intersection [mmol L^{-1}]		cmc (mmol L^{-1})[a]
	water	Tris buffer	
0	7	6	8
10	13	10	17[b]
20	27	32	32
30	48	71	>60[c]

[a] Reference 18. [b]Obtained by interpolation. [c] Obtained by extrapolation.
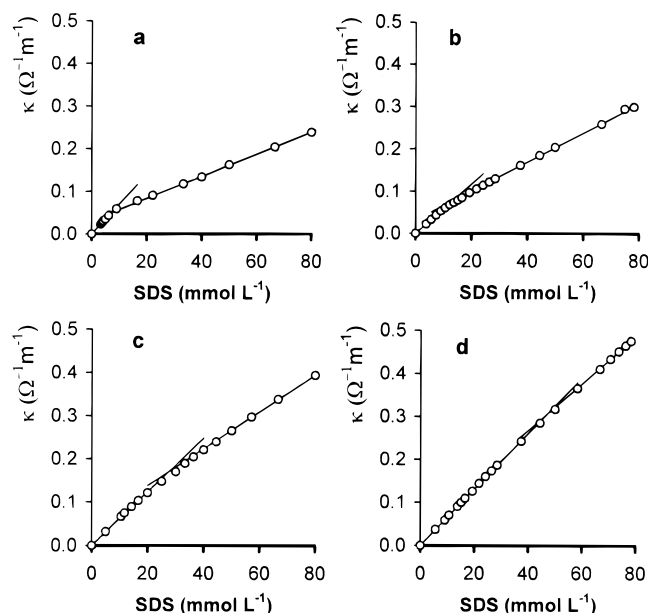

Figure 7. The conductivity of SDS solutions in (a) water, (b) 10% (v/v) AcCN, (c) 20% AcCN, and (d) 30% AcCN.

water−AcCN mixtures by spectroscopic and conductivity measurements, which in this case gave results that agreed well. For the conductivity method, plots were made of the observed solution conductivity against the total SDS concentration with different solvent compositions. For the particular solvent, straight lines were fitted through the experimental points at low and at high SDS concentrations. The SDS concentration at the intersection of these lines was regarded as the cmc.

We have performed similar experiments for aqueous solutions containing 0−40% (v/v) AcCN. The conductivity measurements were carried out with and without addition of Tris buffer (pH = 8) to the solvent, and the SDS concentrations were varied in the range 0−80 mmol L^{-1}. Examples of the observed relations between the conductivity and the SDS concentration, and the straight lines fitted through the data points, are shown in Figure 7. The intersection of the straight line sections at low and high SDS concentrations are given in Table 2. For 30% AcCN, the values given are based on a somewhat arbitrary selection of data points at the low and high side of the SDS range. With 40% AcCN, no clear distinction between a low and a high concentration region could be found. The values for the intersections can be compared with the data for the cmc of SDS as given by Misra et al.[18] Given the uncertainty in the assessment of the straight lines, especially for higher AcCN contents, the agreement of our experiments with the results of Misra et al. (for solutions without buffer) is good. The influence of the buffer addition seems to be small.

When the line intercepts as given in Table 2 are accepted as the cmc values in the strict sense, the conclusion appears to be

that neutral compounds have been shown to have an electrophoretic mobility in solutions where micelles do not exist. This of course supports the solvophobic interaction model proposed by several authors. A closer inspection of the data in Figure 7, however, shows that this conclusion might be premature. The following observations can be made on the experimental results.

(a) The slopes of the curves at the lower end of the SDS concentration scale are all similar. When the observed slopes are corrected for the viscosity differences between various AcCN to water volume ratios, they are even equal within experimental error. This means that, in solvents with up to 40% AcCN, no appreciable ion association is observed in dilute SDS solutions.

(b) For all AcCN contents, experimental conductivity data start to deviate from the lower region straight lines at approximately 5−10 mmol L^{-1} SDS.

(c) With water as solvent, a sharp bend is observed in the conductivity vs SDS concentration curve, which is generally interpreted as the indication of the cmc. With increasing AcCN content, however, the change in the slope becomes more and more gradual.

(d) The magnitude of the change of the slope decreases with increasing AcCN content.

With aqueous solutions, it has amply been shown that the cmc of SDS (and other surfactants) marks a sharp change in the behavior of the surfactant ions. At concentrations below the cmc, all surfactant particles are present as free ions in the solutions. At concentrations above the cmc, the concentration of free SDS ions remains virtually at the cmc value. All further SDS added to the solution contributes to the micellar fraction. SDS micelles in water have a narrow size distribution with a mean aggregation number of ~70. This change in behavior around the cmc is reflected in the change of the differential equivalent conductance Λ_d (the slope of the conductivity vs concentration plot). At low concentrations, the differential equivalent conductance can be written as

$$\Lambda_d = F(\mu_{Na^+} + \mu_{SDS^-}) \qquad (2)$$

where Λ_d is the equivalent conductance of the electrolyte solution, F is the faraday constant, and μ_{Na^+} and μ_{SDS^-} are the (absolute values of the) mobilities of sodium and SDS ions, respectively. The fact that Λ_d is lower above the cmc, even when the mobility of the micelles is higher than that of the individual, free SDS ions, can be explained as the result of the partial neutralization of the micelles by sodium cations:[23]

(23) Hunter, R. J. *Foundations of Colloid Science*; Clarendon Press: Oxford, U.K., 1987; Vol. 1, Chapter 10.

$$nS^- + (n-p)Na^+ \rightarrow M^{p-} \qquad (3)$$
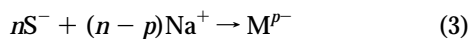
where S is the SDS ion and M is micelle, n the aggregation number, p the effective charge on the micelle, and $(n - p)$ the number of counterions bound in the Stern layer. When a micelle of n SDS ions binds with $(n - p)$ sodium ions, its net negative charge, q, is equal to $(n - p)$. The differential equivalent conductance above the cmc can be written as

$$\Lambda_d = F\left\{\left(1 - \frac{p}{n}\right)\mu_{Na^+} + \frac{q}{n}\mu_{mc}\right\} = F\frac{q}{n}(\mu_{Na^+} + \mu_{mc}) \qquad (4)$$

Since the mobilities referred to in eqs 2 and 4 are known from electrophoretic experiments, the value of q/n for the SDS micelles can be derived from the ratio of the slopes in Figure 7 at high and low concentrations of SDS. For aqueous solutions without buffer, we find a value of 0.30 for q/n. This means that ~70% of the charge of the SDS ions in a micelle is neutralized by sodium ions. For aqueous Tris buffers, the partial neutralization of the micelles by cations found in this way was 63%.

With increasing AcCN content, the slopes in Figure 7 at high SDS concentration increase. When the model valid for micelle formation in pure aqueous solutions is also adopted for AcCN−water mixtures, this would mean that the partial neutralization of the micelles decreases with increasing AcCN content. This seems very unlikely. The dielectric constant of AcCN is lower than that of water; therefore, one would sooner expect an increase of ion association phenomena with increasing AcCN content. A more logical explanation for the increasing slope is that the formation of micelles is less abrupt when the SDS concentration is increased. The formation of micelles is based on very subtle energy balances.[24] Mukerjee[25] showed that the typical SDS behavior in water (an almost constant free SDS concentration above the cmc and a narrow size distribution of the micelles) can be modeled as the result of a very small energetic preference (in the order of 1%) for micelles of a certain size range. It can be imagined that in the presence of AcCN the energy balance changes in favor of the formation of a wider micellar size range. If that is the case, the addition of more SDS to the solution will result in an increase of the volume of the micellar phase and of the average size of the micelles, but also in an increase of the free SDS concentration.

As an illustration, we have interpreted the conductivity data as given in Figure 7 in this way. The increase of the conductivity with the total SDS concentration is seen as originating partly from an increase in free SDS and partly from an increase of the micellar fraction. The value of Λ_d observed at low concentrations was taken for the equivalent conductance of free SDS. For the calculation of the equivalent conductance of the micellar fraction, we estimated the micellar mobility as the maximum mobility observed in the electrophoretic experiments. Furthermore, it was assumed that the value of q/n for the micelles does not depend on the AcCN content of the solution. In Figure 8, the calculated free and micellar fractions of SDS are plotted as a function of the total SDS concentration for various AcCN contents. In water, the sudden appearance of micelles at the cmc is seen in the figure,
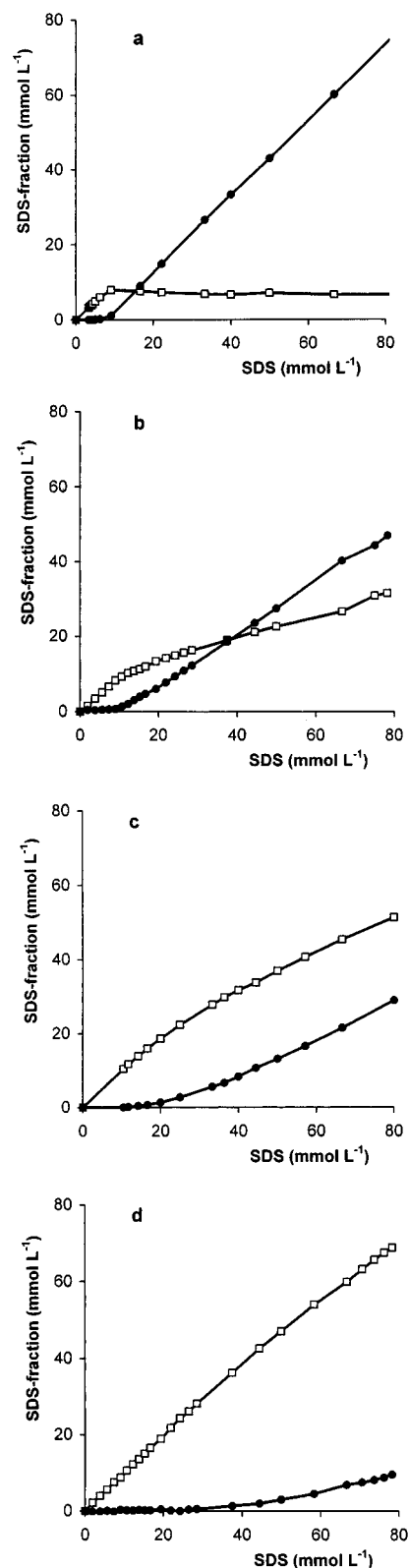
(24) Stigter D.; Overbeek I. T. G. *Proceeding of the Second International Congress on Surface Activity*; 1957; Vol. 1, p 311.
(25) Mukerjee, P. In *Physical Chemistry, Enriching Topics from Colloid and Surface Science*; van Olphen, H., Mysels, K. J., Eds.; IUPAC Commission I.6; Theorex: La Jolla, CA, 1975; Chapter 9.

Figure 8. Calculated free ($\square$) and micellar ($\bullet$) fractions of SDS as a function of the total concentration of SDS at various AcCN contents: (a) 0, (b) 10, (c) 20, and (d) 30%.

with a virtually constant free SDS concentration above the cmc. With AcCN in the solvent, the first micelles also appear at ~10 mmol L^{-1}, but their concentration increases gradually. Although the data shown in Figure 8 have only an indicative value, the results support the previously derived conclusion that even with high AcCN contents SDS can form micelles when its concentration

exceeds ~10 mmol L^{-1}. Therefore, it is questionable to attribute the migration of neutral compounds in SDS solutions containing AcCN by solvophobic interactions between the solutes and individual SDS molecules. On basis of the conductivity measurements, a mechanism based on the distribution of the solutes into smaller micelles is more likely.

CONCLUSIONS

From the electrophoretic experiments conducted in this study it may be concluded that the electrokinetic separation of neutral hydrophobic compounds in SDS solutions with up to 30% AcCN is (still) based on the distribution of the compounds between the solvent and SDS micelles. The mobilities observed are simply too high to be attributed to so-called solvophobic interaction with single, free SDS ions in a 1:1 complexation. It seems also unlikely that the observed mobilities can be ascribed to the formation of higher order complexes with free SDS ions. The mobilities of all compounds tested, which had strongly different sizes and structures, all tended to approach the same maximum value at high SDS concentrations.

With AcCN contents of 40% and higher, the evidence for the existence of SDS micelles is less strong. However, on increasing the AcCN content of the solvent, no abrupt changes of the electromigration behavior of the model compounds have been observed. The results obtained in our experiments appear to indicate that the effects of increasing the AcCN content are better explained as a gradual change of the volume fraction and the properties of the micellar phase than as a sudden disappearance of the micelles. At very high AcCN contents, it will be difficult to find a distinction between micelle-like SDS aggregates and higher order SDS complexes of the analytes as the basis for their electromigration.

The assumed existence of SDS micelles in AcCN−water mixtures, with total SDS concentrations in the range of 15−60 mmol L^{-1}, is in contradiction to the cmc values reported in the literature. However, from conductivity measurements, it appears that the classical micelle formation model is not valid for these solvents. With water as solvent, the cmc marks a sudden change in the solution behavior of SDS. Below the cmc, only free SDS ions are present; SDS added above the cmc is virtually completely present as micelles with a narrow size distribution. With AcCN−water mixtures as solvent, the first micelles are also formed in the solution at SDS concentrations of ~10 mmol L^{-1}. With higher SDS concentrations, the number and possibly also the average size of the micelles gradually increases. The mobility of the micelles in solutions with 10 or 20% AcCN is similar to that in water. It seems to decrease with higher AcCN content. This could be explained as the result of a smaller average size, or of a stronger binding of counterions to the micelle, due to the lower dielectric constant of the solution.

The decrease of the observed mobilities of neutral hydrophobic compounds by the addition of AcCN in a concentration range of 20−40% (v/v) to a SDS solution can be attributed to the combined effect of a decrease of the volume fraction of the micellar phase, a decrease of the average size of the micelles, and a decrease of the distribution constants of the solutes.