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Double-chain surfactant as a new and useful micelle-forming reagent for micellar electrokinetic chromatography

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ABSTRACT

Disodium 5,12-bis(dodecyloxymethyl)-4,7,10,13-tetraoxa-1,16-hexadecanedisulphonate, a surfactant with two ionic groups and two lipophilic chains, was first utilized in micellar electrokinetic chromatography (MEKC). A good linear relationship was obtained between the capacity factors of several model analytes tested and the concentrations of the surfactant. Compared with widely used sodium dodecyl sulphate, this new surfactant exhibited remarkably different selectivity for several substituted naphthalene and benzene derivatives and gave a wider migration time window. MEKC could be performed at surfactant concentrations at most one order of magnitude lower, because of the low critical micelle concentration.

INTRODUCTION

Micellar electrokinetic chromatography (MEKC) [1] was developed for the separation of electrically neutral analytes by electrophoresis with an ionic micellar solution as a separation solution, and has become a method of great importance. The separation principle of MEKC is based on the differential partition of the analyte between the micelle and water. Therefore, selectivity in MEKC is considered to be essentially dependent on the choice of surfactant. Since most analytes interact with micelles at their surfaces, the ionic group is generally more important in terms of selectivity. For instance, for the polar analytes, sodium dodecyl sulphate (SDS) and sodium tetradecyl sulphate exhibit very similar selectivity, but SDS and sodium N-lauroyl-N-methyltaurate yield considerably different selectivity [2]. Corticosteroids cannot be successfully separated with SDS but are completely resolved with sodium cholate [3]. Addition of a second surfactant to form a mixed micelle also affects the selectivity. Mixed micelles are successfully employed in separations of enantiomers [4–6]. The addition of a non-ionic surfactant to an ionic one causes a narrower migration time window because of the lower electrophoretic mobility of the mixed micelle.

Until now, surfactants with one hydrophilic group and one lipophilic chain, especially SDS, have been used widely in MEKC. Recently, synthetic amphipathic compounds with several

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lipophilic chains have attracted much attention as biomimetic functional materials [7]. In our previous papers [8,9], we have shown that amphipathic compounds containing two sulphate or sulphonate groups and two long alkyl chains, which are derived from glycol diglycidyl ethers, have good water solubility and excellent surface-active properties in water. These new types of surfactants have not yet been utilized as micelle-forming reagents in MEKC. Consequently, their use in MEKC is of great interest and is expected to bring about dramatic selectivity changes.

In this paper, preliminary results on the separation behaviour of several model analytes by MEKC using disodium 5,12-bis(dodecyloxymethyl)-4,7,10,13-tetraoxa-1,16-hexadecanedisulphonate (DBTD) as a double-chain surfactant are compared with the results with SDS.

EXPERIMENTAL

Apparatus

An Applied Biosystems Model 270A capillary electrophoresis system (CA, USA) was used with a 72 cm (50 cm from inlet to detector) × 50 μm I.D. fused-silica capillary. On-column UV detection was at 210 nm. The temperature and applied voltage were held constant at 35°C and 15 kV, respectively. Sample solutions ($1.0 \cdot 10^{-2}$% in 10% methanol) were injected by the vacuum technique (12.7 cmHg pressure difference for 0.5 s). For data processing, a Hitachi D-2500 Chromato-Integrator (Hitachi, Japan) was used. All experiments were performed in duplicate to ensure reproducibility.

Reagents

DBTD, whose structure is shown in Fig. 1, was synthesized as reported previously [8]. All other reagents were of analytical-reagent grade and used as received.

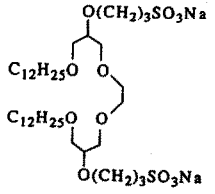

Fig. 1. Structure of a double-chain surfactant, DBTD.

Separation solutions were prepared by dissolving DBTD in a buffer solution of 0.1 *M* sodium dihydrogenphosphate–0.05 *M* sodium borate at pH 7.0. Methanol was used as a marker of the electroosmotic flow and Sudan III as a micelle tracer.

RESULTS AND DISCUSSION

From several double-chain synthetic surfactants bearing two ionic groups, DBTD was chosen in this work because of its low critical micelle concentration (CMC) (0.014 m*M*), low Krafft point (below 0°C) and high purity. On the other hand, SDS has a much higher CMC of 8.1 m*M* and a higher Krafft point of 16°C. It is suggested that MECK may be performed with DBTD at lower concentrations. This will result in low viscosities of separation solutions and in low currents, which is quite favourable for MEKC.

Separation with DBTD

Several benzene and naphthalene derivatives were used as model analytes, and their migration times were measured in the DBTD concentration range of 0–10 m*M* in 0.1 *M* phosphate–0.05 *M* borate buffer at pH 7.0.

Six naphthalene derivatives were baseline separated at DBTD concentrations above 7.5 m*M*. A typical chromatogram is shown in Fig. 2. At DBTD concentrations above 1.0 m*M*, 1- and 2-naphthol could be readily baseline separated. 1,3-Dihydroxybenzene, phenol, nitrobenzene and *p*-nitroaniline were also resolved at DBTD concentrations above 5.0 m*M*.

The capacity factor of a neutral analyte in MEKC can be calculated by the following equation [1].

$$k' = \frac{t_R - t_0}{t_0(1 - t_R/t_{mc})} \quad (1)$$

where t_R, t_0 and t_{mc} are the migration times of the analyte, the solute that does not interact with the micelle (methanol peak) and the micelle (Sudan III peak), respectively. When micellar

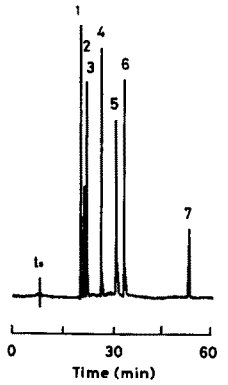

Fig. 2. Separation of naphthalene derivatives with $1.0 \cdot 10^{-2}$ *M* DBTD. Peaks: 1 = 1-naphthalenemethanol; 2 = 1,6-dihydroxynaphthalene; 3 = 1-naphthylamine; 4 = 1-naphthaleneethanol; 5 = 2-naphthol; 6 = 1-naphthol; 7 = Sudan III.

concentrations are low, k' is approximately related to the surfactant concentration by eqn. 2 [10].

$$k' = Kv(C_{sf} - \mathrm{CMC}) \quad (2)$$

where K is the distribution coefficient of the analyte, and v and C_{sf} are the partial specific volume of the micelle and the surfactant concentration, respectively.

The plots of calculated k' *vs.* concentration of DBTD are shown in Figs. 3 and 4 for the naphthalene and benzene derivatives, respectively. Table I gives the results of the regression analyses. For each analyte, a good linear relationship (correlation coefficient range 0.993–

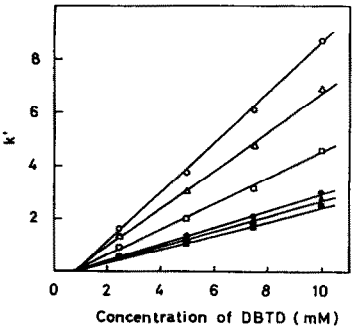

Fig. 3. Dependence of capacity factor (k') of the naphthalene derivatives on concentration of DBTD (C_{sf}). Analytes: ■ = 1-naphthalenemethanol; ▲ = 1,6-dihydroxynaphthalene; ● = 1-naphthylamine; □ = 1-naphthaleneethanol; △ = 2-naphthol; ○ = 1-naphthol.

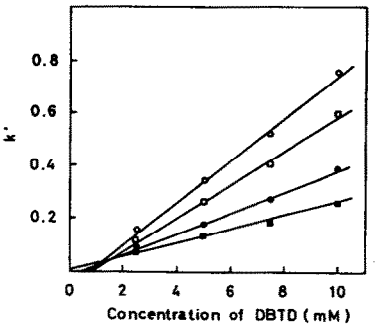

Fig. 4. Dependence of capacity factor (k') of the benzene derivatives on concentration of DBTD (C_{sf}). Analytes: ■ = 1,3-dihydroxybenzene; ● = phenol; □ = nitrobenzene; ○ = *p*-nitroaniline.

0.998) was obtained, which suggests that the distribution coefficients remain constant at least at the DBTD concentrations measured. Similar linear plots were also obtained for SDS.

From eqn. 2, the intercepts of plots in Figs. 3 and 4 extrapolated to $k' = 0$ can be interpreted to be the CMC of the surfactant under the conditions used. The averaged CMC value estimated is 0.76 m*M*, except for 1,3-dihydroxybenzene and phenol exhibiting considerably low k' values. This CMC value is too high in comparison with the measured value of 0.014 m*M* in water. The reason for this discrepancy is not clear at present.

Comparison of selectivity

It is quite interesting and of great value to compare the migration behaviour in DBTD and SDS solutions. As mentioned above, the six naphthalene derivatives were baseline separated at 7.5 m*M* DBTD. Similar separation of the five analytes except for 2-naphthol was attained at an SDS concentration of 50 m*M*. The capacity factors of the analytes at these concentrations are listed in Table II, together with those of the other analytes used here. In MEKC with DBTD, the analytes were eluted in the order given in the first column in the table (from 1,3-dihydroxybenzene to 1-naphthol). Apparently, this elution order is different from that in MEKC with SDS, especially for the naphthalene derivatives. The elution order in the DBTD solution is 1-naphthalenemethanol < 1,6-dihydroxynaphthalene < 1-naphthylamine < 1-naphthaleneethanol < 2-

TABLE I

CORRELATION BETWEEN CONCENTRATION OF DBTD (C_{st}) AND CAPACITY FACTOR (k')

y = Capacity factor of the analyte; x = concentration of DBTD (2.5–10.0 mM).

Analyte	Regression equation	Correlation coefficient
1,3-Dihydroxybenzene	$y = 0.0248x + 0.0095$	0.993
Phenol	$y = 0.0396x - 0.0171$	0.995
Nitrobenzene	$y = 0.0629x - 0.0475$	0.993
p-Nitroaniline	$y = 0.0793x - 0.0520$	0.995
1-Naphthalenemethanol	$y = 0.260x - 0.176$	0.995
1,6-Dihydroxynaphthalene	$y = 0.296x - 0.207$	0.995
1-Naphthylamine	$y = 0.323x - 0.259$	0.996
1-Naphthaleneethanol	$y = 0.496x - 0.417$	0.994
2-Naphthol	$y = 0.738x - 0.575$	0.995
1-Naphthol	$y = 0.940x - 0.825$	0.998

naphthol < 1-naphthol and 1,6-dihydroxynaphthalene < 1-naphthylamine < 1-naphthalenemethanol < 1-naphthol < 2-naphthol < 1-naphthaleneethanol in the SDS solution. 1-Naphthaleneethanol eluted last and the elution order of 1- and 2-naphthol was reversed in the SDS system. In the SDS system, the isomers could not be baseline separated, though this was readily done in the DBTD system, as described above.

A wide migration time window between t_0 and t_{mc} is favourable for high resolution, although a long analysis time may be required. The t_{mc}/t_0 value is directly related to the width of the migration time window. The larger the value of t_{mc}/t_0, the wider the migration time window. The t_{mc}/t_0 value is about 5.0 for 50 mM SDS and about 6.3 for 7.5 mM DBTD. The value decreases from 5.0 for 50 mM SDS to 4.3 for 7.5 mM SDS.

TABLE II

CAPACITY FACTORS OF THE ANALYTES IN 7.5 mM DBTD AND 50 mM SDS

Analyte	Capacity factor	
	DBTD	SDS
1,3-Dihydroxybenzene	0.182	0.220
Phenol	0.261	0.524
Nitrobenzene	0.392	1.36
p-Nitroaniline	0.507	1.08
1-Naphthalenemethanol	1.66	6.70
1,6-Dihydroxynaphthalene	1.89	2.08
1-Naphthylamine	2.04	5.32
1-Naphthaleneethanol	3.07	13.34
2-Naphthol	4.66	7.18
1-Naphthol	6.06	6.99

Thus DBTD shows remarkably different selectivity and a wider migration time window compared with SDS. In general, introduction of new surfactants exhibiting essentially different selectivity and wider migration time windows is desirable for enhancing MEKC performance. Several surface-active properties of DBTD were measured, but its micellar size and shape have not yet been characterized. Therefore, further work is needed to discuss and convincingly explain the above-mentioned selectivity change of DBTD in detail. Attempts to do this are now in progress.

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