Effect of alkyl chain length of sodium N-acyl sarcosinates on migration behavior in micellar electrokinetic chromatography

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Abstract

Application of sodium N-acyl sarcosinates for micellar electrokinetic chromatography was investigated. Three homologous sodium N-acyl sarcosinates were able to be used as pseudo-stationary phase. The migration behavior of some aromatic test samples was investigated with these surfactants under the same micellar concentrations. The migration time window became wider as the alkyl chain length of surfactant decreased. The capacity factors increased with an increase in the alkyl chain length of surfactant. Three sarcosinates gave almost the same separation selectivity for hydrophilic solutes. The separation selectivity for hydrophobic solutes was slightly different from one another.

Keywords: Chain length; Pseudo-stationary phases; Buffer composition; Sodium acyl sarcosinates; Surfactants; Anilines; Phthalates; Sarcosinates

1. Introduction

One of the most powerful analytical methods for the separation of neutral and charged solutes is micellar electrokinetic chromatography (MEKC) [1,2]. The major advantage of MEKC over other analytical methods is high separation efficiency that can be achieved with minimal usage of reagent and solvent. The migration behavior of solutes is influenced by the nature of the surfactant. The most widely used surfactants are anionic species, such as sodium dodecyl sulfate (SDS) [3–6], sodium dodecane sulfonate [5], bile salts [7,8], sodium N-lauroyl-N-methyltaurate [9,10] and sodium tetradecene sulfonate [10]. However, it is clear that as yet the optimum surfactant for use as the pseudo-stationary phase still remains to be discovered.

In order to aid design of new anionic surfactants for MEKC, we have undertaken a study of the effect of chain length on migration behavior. Possible surfactants were selected by a literature search of homologous surfactants for use as the pseudo-stationary phase in MEKC. The criteria of the search were: (1) for ease of use, their Krafft point should be lower than room temperature and (2) their critical micelle concentrations (CMC) values should be low in order to avoid degradation of separation efficiency. SDS was first employed and is the most commonly used surfactant, however, homologues of SDS seem to be less suitable for surfactants in MEKC [3,6].

From this search we discovered that a series of three sodium N-acyl sarcosinates; sodium N-lauroyl sarcosinate (SLN), sodium N-myristoyl sarcosinate (SMN) and sodium N-parmitoyl sarcosinate (SPN) satisfied the criteria with their reported values of

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Krafft point and CMC [11]. In general, sodium salts of N-acyl sarcosinates have good solubility, good detergency and lime resistance [12]. However, their employment in MEKC has not previously been reported. In this paper we have investigated the above three sodium N-acyl sarcosinates for use as pseudo-stationary phase in MEKC. The effect of changes in alkyl chain length on the migration time window, capacity factor and separation selectivity of some aromatic test compounds are presented.

2. Experimental

2.1. Apparatus

MEKC was performed with a CAPI-3000 automated CE System (Otsuka Electronics, Osaka, Japan). A 50 μm I.D. fused-silica capillary (GL Sciences, Tokyo, Japan) of 62 cm total length (50 cm to the detector) was used. The instrument control and data collections were performed with a PC-9801 personal computer (NEC Corporation, Tokyo, Japan).

Measurement of CMC was performed by a conductometry with a CDM3 conductivity meter (Radiometer, Copenhagen, Denmark).

2.2. Reagents

Aromatic test samples, aniline derivatives were obtained from Nacalai Tesque (Kyoto, Japan). Phthalate esters were obtained from Wako (Osaka, Japan). All samples were of analytical grade and were used without further purification. Sodium N-acyl sarcosinates were donated by Nikko (Tokyo, Japan). Their purity was checked by elemental analysis. SLN and SMN were used as received. SPN was recrystallized from ethanol.
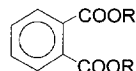
2.3. Procedure

Running solutions were prepared by dissolving sodium N-acyl sarcosinates with their micellar concentrations at 50 mM in a mixture of 0.02 M sodium dihydrogenphosphate solution and 0.02 M sodium tetraborate solution adjusted to pH 7.0. These solu-
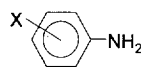
Fig. 1. Aromatic test samples for MEKC analysis with sodium N-acyl sarcosinates.

tions were filtered through a 0.45-μm membrane filter prior to use.

Chemical structures of aromatic test samples are shown in Fig. 1. Aniline derivatives are hydrophilic compounds and phthalate esters are relatively hydrophobic compounds. All solutes are electrically neutral at pH 7.0. Stock solutions of these samples were prepared in water or methanol. Sample solutions for injection were made by tenfold diluting the stock solution with running buffer. Each concentration of solute was about 100 mg/l.

When the running solution was changed, the capillary was rinsed with 0.1 M NaOH for 5 min using a vacuum at the detector reservoir, followed by subsequent rinses of distilled water for 3 min and running buffer for 4 min. After each run, the capillary was rinsed with running buffer for 4 min.

Sample injections were made hydrostatically, 20 mm × 30 s. The injection volume was about 1.7 nl. The set-up voltage and temperature were 20 kV and 30°C, respectively, throughout all experiments. Migrated samples were detected by on-column measurement of UV absorption at 270 nm.

3. Results and discussion

3.1. Physicochemical properties of three sodium N-acyl sarcosinates in buffer

Structures, literature values of Krafft points and CMC of sodium N-acyl sarcosinates and sodium alkyl sulfates are shown in Table 1. These values were obtained in distilled water [11,13]. The Krafft points of sodium N-acyl sarcosinates are lower than

Table 1
Structures and physicochemical properties of sodium N-acyl sarcosinates and sodium alkyl sulfates

Surfactant	Krafft point/°C (in distilled water [11,13])	CMC/mM (in distilled water [11,13])	CMC/mM (in buffer)
Sodium N-acyl sarcosinate: $C_nH_{2n+1}CON(CH_3)CH_2COONa$			
Sodium N-lauroyl sarcosinate (SLN, $n=11$)	<0	13.0 (25°C)	6.0 (30°C)
Sodium N-myristoyl sarcosinate (SMN, $n=13$)	<0	3.4 (25°C)	0.9 (30°C)
Sodium N-palmitoyl sarcosinate (SPN, $n=15$)	ca. 8	0.8 (25°C)	0.2 (30°C)
Sodium alkyl sulfate: $C_nH_{2n+1}OSO_3Na$			
Sodium decyl sulfate ($n=10$)	<0	33 (40°C)	
Sodium dodecyl sulfate (SDS, $n=12$)	16	8.6 (40°C)	4.6 (30°C)
Sodium tetradecyl sulfate ($n=14$)	30	2.2 (40°C)	

those of sodium alkyl sulfates. Thus good solubility is due to the additional amido linkage [12].

In buffer solutions the Krafft point is increased and the CMC is reduced. We investigated these values in buffer, employing 0.02 M borate–phosphate (pH 7.0). At ambient temperature (ca. 20°C), three sarcosinates were easily dissolved in the buffer. Hence their Krafft points are below ca. 20°C. In order to confirm micellar formation and to determine CMC, the dependence of conductivity on the concentration of these surfactants was measured at 30°C.

The specific conductance of solutions initially showed a linear increase with increasing concentration up to a point and after this again a linear increase in specific conductance was observed but at a slower rate. The point of intersection of these two straight lines can be taken as the CMC of the surfactants [12]. The determined CMC values in the buffer are also shown in Table 1. These values are considered to be sufficiently low for usage in MEKC.

3.2. Effect of the alkyl chain length on the migration time window

The chromatograms of aniline derivatives using three sodium N-acyl sarcosinates are shown in Fig. 2. All solutes were migrated between t_0, the migration time of electroosmotic flow (methanol as a tracer) and t_{mc}, the migration time of micelle (Sudan IV as a tracer). The migration time window became wider with a decrease of alkyl chain length in surfactants. For phthalate esters, similar results were

obtained. Relative standard deviations of migration time were within 1.1% ($n=9$) for all surfactants.

The value of t_{mc}/t_0 can be regarded as a parameter to indicate the width of the total range of elution for electrically neutral solutes [3]. The comparison of t_{mc}/t_0 with three sarcosinates is shown in Fig. 3. The electroosmotic velocity (v_{eo}) and the electrophoretic velocity of micelle (v_{ep}) is also shown in the same figure. The dependence of v_{eo} on alkyl chain length is more remarkable than that of v_{ep}. Therefore the extension of migration-time window is mainly due to the decrease of electroosmotic velocity. The increase of electroosmotic velocity is probably caused by the increase of hydrophobic interaction of monomeric surfactants on capillary wall [14].
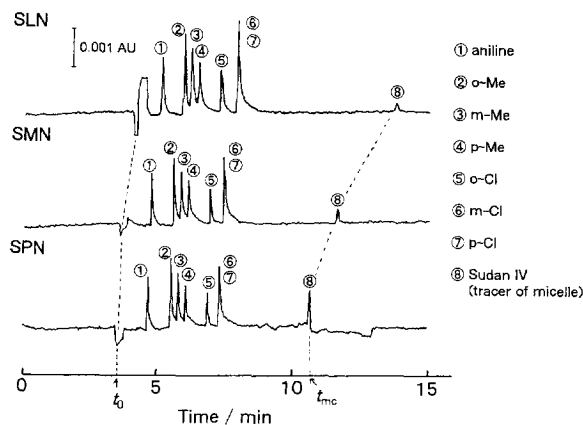

Fig. 2. Chromatograms of aniline derivatives using three sodium N-acyl sarcosinates. Conditions: capillary, 62 cm (50 cm to the detector) × 50 μm I.D.; micellar concentration, 50 mM; running buffer, 0.02 M borate–phosphate (pH 7.0); applied voltage, 20 kV; detection wavelength, 270 nm; temperature, 30°C.
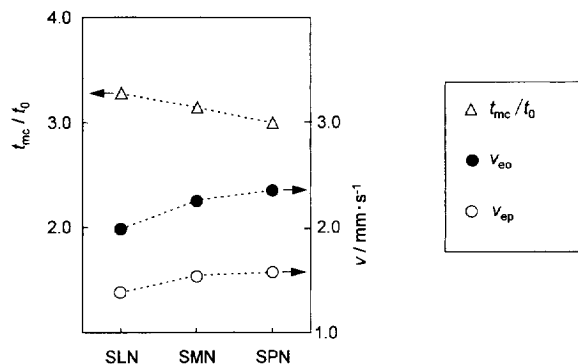
Fig. 3. The dependence of t_{mc}/t_0, electroosmotic velocity (v_{eo}) and electrophoretic velocity of micelle (v_{ep}) on three sarcosinates.

3.3. Effect of the alkyl chain length on the capacity factor

In MEKC [3], the capacity factor of an electrically neutral solute is given by:

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

where t_R is the migration time for the solute. The capacity factor can be related to the partition coefficient K_{mw} of a solute between micelle and aqueous phase through:

$$k' = K_{mw} \cdot \frac{V_{mc}}{V_{aq}} \approx K_{mw}\bar{v}(c_{sf} - \text{CMC}) \tag{2}$$

where V_{mc}/V_{aq} is the volume ratio of the micelle and aqueous phase, $\bar{v}$ is the partial molar volume of the micelle, c_{sf} is the total concentration of surfactant.

The capacity factor (k') of all test samples became larger as the alkyl chain length of surfactants became longer. The dependence of capacity factor of aniline derivatives on the alkyl chain length of sarcosinates is shown in Fig. 4. The capacity factor using SDS is also shown in the same figure. The separation selectivity is apparently different between three sarcosinates and SDS.

Phthalate esters migrated in the order of hydrophobicity with all surfactants used including SDS. They are neutral and are more hydrophobic than aniline derivatives. Accordingly, their migration behavior depends on their hydrophobicity.
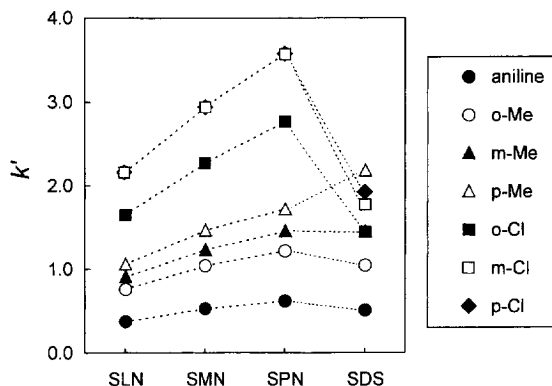
In MEKC, the time interval between peaks of

Fig. 4. Dependence of the capacity factor k' of aniline derivatives on surfactants.

hydrophobic solutes increases as k' decreases, resulting in peak separation of hydrophobic solutes. Therefore, the use of SLN gave the best separation of phthalate esters among all surfactants used. The separations of phthalate esters using SLN and SDS are shown in Fig. 5. From Eq. 2, the product of the partition coefficient and the partial molar volume of the micelle becomes larger as the alkyl chain length of surfactants increases.

3.4. Effect of the alkyl chain length on the separation selectivity

To compare the selectivity of each surfactant, we calculated separation factor (α) between aniline and each derivative. Similar calculations were made for
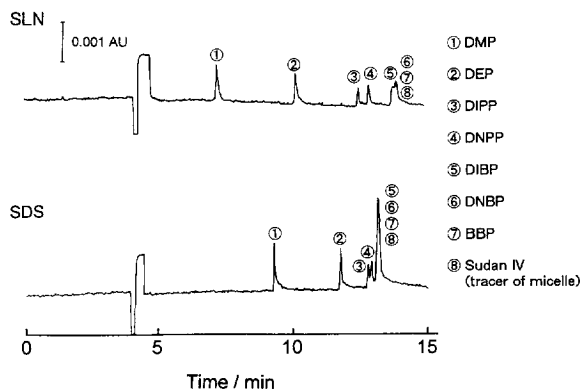

Fig. 5. Chromatograms of phthalate esters with SLN and SDS. Other conditions are the same as in Fig. 2.
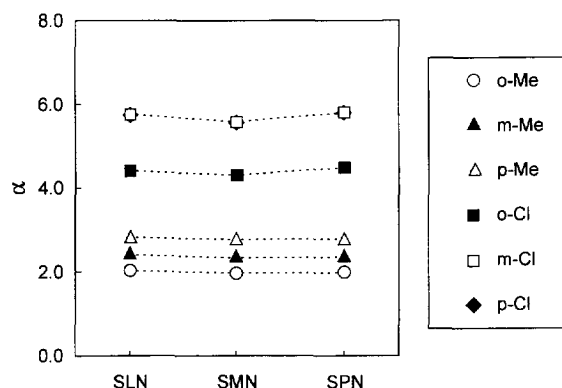
Fig. 6. Dependence of separation factor of aniline derivatives ($\alpha = k'_{derivative}/k'_{aniline}$) on three sarcosinates.

phthalate esters. The results of these calculations for aniline derivatives are shown in Fig. 6.

The separation factors for aniline derivatives were almost independent among the three surfactants, that is, these surfactants showed almost the same selectivity. For phthalate esters, however, the selectivity varied with surfactants as shown in Fig. 7. This indicates that the separation mechanisms among these surfactants are slightly different between hydrophilic and hydrophobic solutes. This can be easily explained if one assumes that hydrophilic solutes are solubilized by adsorption on the micellar surface whereas hydrophobic solutes incorporate into the interior of the micelle [3].
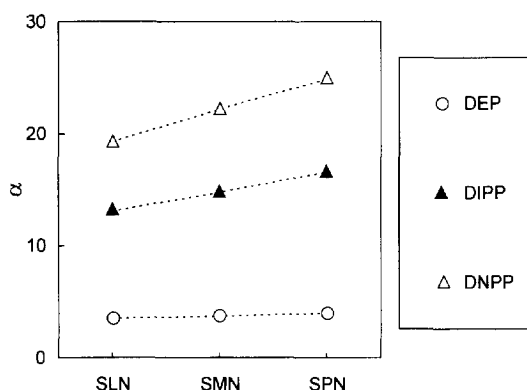

Fig. 7. Dependence of separation factor of phthalate esters ($\alpha = k'_{phthalate}/k'_{DMP}$) on three sarcosinates.

4. Conclusions

Three homologous sodium N-acyl sarcosinates were used for MEKC as pseudo-stationary phase. The effect of alkyl chain length of these surfactants on migration-time window, capacity factor and separation selectivity was investigated. Under the same micellar concentrations, SLN with the shortest alkyl chain gave the best separation for hydrophobic solutes such as phthalate esters among the three surfactants studied. The effect of alkyl chain length on capacity factor mainly contributed to this result. For hydrophilic solutes, the three sarcosinates gave almost the same separation selectivity. The structure of polar moiety of surfactant is significant to change separation selectivity for hydrophilic solutes.

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