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Capillary Electrochromatography Using a Strong Cation-Exchange Column with a Dynamically Modified Cationic Surfactant

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A novel mode of capillary electrochromatography (CEC), called dynamically modified strong cation-exchange CEC (DMSCX-CEC), is described in this paper. A column packed with a strong cation-exchange (SCX) packing material was dynamically modified with a long-chain quaternary ammonium salt, cetyltrimethylammonium bromide (CTAB), which was added to the mobile phase. CTAB ions were adsorbed onto the surface of the SCX packing material, and the resulting hydrophobic layer on this packing was used as the stationary phase. Using the dynamically modified SCX column, neutral solutes were separated with the CEC mode. The highest number of theoretical plates obtained was about 190 000/m, and the relative standard deviations (RSD's) for migration times and capacity factors of alkylbenzenes were less than 1.0% and 2.0% for five consecutive runs, respectively. The effects of CTAB and methanol concentrations and the pH value of the mobile phase on the electroosmotic flow and the separation mechanism were investigated. Excellent simultaneous separation of the basic and neutral solutes in DMSCX-CEC with a high-pH mobile phase was obtained. A mixture containing the acidic, basic, and neutral compounds was well separated in this mode with a low-pH mobile phase; however, peak tailing for basic compounds was observed in this mobile phase.

Capillary electrochromatograpy (CEC) has developed exponentially since its potential was first demonstrated by Pretorius et al. in 1974.[1] CEC combines the high efficiency of capillary zone electrophoresis (CZE) and the high selectivity of high-performance liquid chromatography (HPLC) and has received increasingly more attention. Up to now, two types of the stationary phases have been used in CEC. The first is the commercially available standard HPLC stationary phases, including silica-based ODS,[2−6] ion-exchange stationary phases,[7−10] chiral packings,[11−13] etc., which are not "end capped" and are most commonly used nowadays. The unreacted silanol groups left on the surface are capable of generating electroosmotic flow (EOF). Recently, specially developed packings for CEC[14,15] were reported, in which silica particles were coated with a mixture of sulfonic acid groups or amino groups and alkyl chain moieties. The alkyl chains act as stationary phases to retain solutes while the charged groups result in high EOF at low pH. The second type of CEC columns was prepared by polymerization of either silica or polymers inside a column to produce a monolithic bead, which was then derivatized with the stationary phases.[16−18] This technique removes the problem of the inlet and outlet frits that are required for producing particle columns.

Chromatography on dynamically modified silica in HPLC was first reported by Ghaemi et al.[19] In this procedure, an HPLC column was packed with bare silica and a certain amount of a long-chain quaternary ammonium salt, such as cetyltrimethylammonium bromide (CTAB), was added to the mobile phase.[20] The CTAB ions adsorbed on the silica surface formed an apolar layer. The separation mechanism for neutral compounds was based on reversed-phase partition according to the literature.[21,22] Garner and Yeung[23] have reported that a capillary coated with a hydrophobic stationary phase can be dynamically modified by CTAB and that the adsorbed CTAB can act as a dynamic ion exchanger, thereby increasing the selectivity for open tubular CEC. Recently, a

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dynamically modified silica column was used in CEC and a novel mode of CEC was established, called dynamically modified silica-CEC (DMS-CEC).[22] It was also found that the peak tailing of basic compounds can be effectively eliminated in this mode because the surfactant has more affinity for silanol groups than for common basic compounds. But DMS-CEC involves some problems, including the following: (1) it is difficult to be performed with a low-pH mobile phase because the ionization of silanol groups is suppressed and the amount of dynamically adsorbed CTAB decreases seriously; (2) the column efficiency of DMS-CEC is much lower than that of reversed-phase CEC due to relatively slow adsorption−desorption involving CTAB and the silanol groups. In this work, a strong cation-exchange packing (SCX) based on silica was adopted as the stationary phase and it was dynamically modified by CTAB for the performance of CEC, called dynamically modified SCX-CEC (DMSCX-CEC). The separation of neutral, acidic, and basic compounds and the mechanism of their retention in DMSCX-CEC were investigated.

EXPERIMENTAL SECTION

Instrumentation and Materials. All CEC experiments were performed on a P/ACE MDQ system (Beckman, Fullerton, CA); a Spectra-Physics pump (Spectra-Physics Inc., San Jose, CA) was used to pack the capillary columns. Fused-silica capillaries (75 μm i.d., 365 μm o.d.) were obtained from the Yongnian Optic Fiber Plant (Hebei, China), and 5 μm Spherisorb-silica gel and 5 μm Spherisorb-SCX were purchased from the Waters Phase Separation Co. (Milford, MA).

Samples and Solutions. Methanol was of chromatographic grade, ethylbenzene was of chemical grade, and the other reagents used were of analytical reagent grade. Ultrapure water used for preparing solutions was produced by a Milli-Q water system (Millipore Corp., Bedford, MA). Sample solutions were first prepared in methanol and then diluted to the appropriate concentrations by the mobile phases before injection. The stock solution of phosphate buffer (100 mM) was prepared by dissolving 3.90 g of NaH_2PO_4 in 200 mL of ultrapure water and then adjusted to the appropriate pH by NaOH or H_3PO_4 solution, after which it was transferred to a 250 mL flask. The stock solution of CTAB (25 mM) was prepared by dissolving 2.278 g of CTAB in 250 mL of ultrapure water. Mobile phases were prepared by mixing appropriate volumes of phosphate buffer, CTAB stock solution, methanol, and ultrapure water. Before running, the mobile phase was degassed in an ultrasonic bath for 30 min.

Column Preparation. CEC columns with Spherisorb-SCX and Spherisorb-silica gel were packed in house by a slurry-packing technique as reported in literature.[3] Each capillary column was flushed first with 1 M NaOH for 1 h and then with water for 10 min. The initial frit was prepared by sintering 5 μm bare silica, which was tapped into the capillary from one end. Acetone/water (70/30, v/v) and water were selected as the packing solvents for SCX and bare silica, respectively. The slurry was prepared by mixing the packings (about 0.01 g/mL) and the packing solvent and then sonicated for 10 min. Afterward, the slurry was packed into the capillary by the HPLC pump, the injection- and detection-end frits were made by sintering the stationary phase under 340 bar, and the residue particles behind the detection-end frit were flushed out by the water. Each column was 31 cm long with an effective length of 10 cm. Before an experiment, each column was flushed with the mobile phase for 30 min by using a syringe, whose needle is connected to the outlet of the CEC column with a PTFE tube. Then the column was conditioned on the instrument with the mobile phase for another 30 min. The applied voltage was first ramped from 0 to 15 kV for 10 min and then held at 15 kV for 20 min. To avoid bubble formation, 6.9 bar of pressure was also applied to both ends of the capillary.

Separation Conditions. The P/ACE MDQ system can apply 6.9 bar of pressure to both ends of the capillary. But it requires about 0.3 min to reach 6.9 bar while it requires only 0.17 min for the voltage to reach a set value. It is possible to form a bubble when the voltage has reached the separation voltage while the pressure is still low. Therefore, it is necessary to set a ramp time for the voltage longer than that for the pressure to prevent bubble formation. In this study, the ramp time for the voltage was 0.5 min and the separation voltage was 15 kV for all separations unless otherwise stated. The injections were made by applying a voltage of 5 kV for 10 s (no pressure was applied during injection). The temperature was kept at 25 °C, and the detection wavelength was set at 214 nm. Fused-silica capillaries with dimensions of 31 cm (10 cm to detector) × 75 μm i.d. were used for the CEC experiments.

RESULTS AND DISCUSSION

Column Performance. The separation mechanism of DMSCX-CEC is similar to that of DMS-CEC.[22] The adsorbed CTAB molecules form a hydrophobic layer on the SCX packing surface, which acts as the stationary phase as C_{18} chains on the ODS packings. Neutral compounds can be separated in this system based on the partitioning between the mobile phase and the hydrophobic layer. The efficiency of the columns and the reproducibility of the retention times and capacity factors in DMSCX-CEC were evaluated with five homologous benzene compounds, including benzene, toluene, ethylbenzene, *n*-propylbenzene, and *n*-butylbenzene, as the tested solutes. The capacity factors (k') of solutes were calculated as shown in eq 1, where t_r

$$k' = (t_r - t_0)/t_0 \quad (1)$$

is the migration time of a solute and t_0 is the void time of this system. Void time was measured by the disturbance of the baseline due to an injection. The highest number of theoretical plates obtained was about 190 000/m, which was comparable with that for a CEC column packed with 5 μm particles and much higher than that in DMS-CEC.[22] This result suggests that the dynamic adsorption of CTAB on the SCX packing makes much less contribution to the resistance to mass transfer of solute from the mobile to stationary phases than that on the silica packing. The relative standard deviations (RSD's) for retention times and capacity factors were less than 1.5% and 2.0%, respectively, which means that the reproducibility of retention values in DMSCX-CEC was as good as that in reversed-phase CEC with silica-based ODS as the stationary phase[24] but was better than that in DMS-CEC.[22]

Linear regression analysis for the relationship of log k' versus the number of carbons (Nc) for homologous benzene solutes was carried out, giving the following results obtained at the mobile

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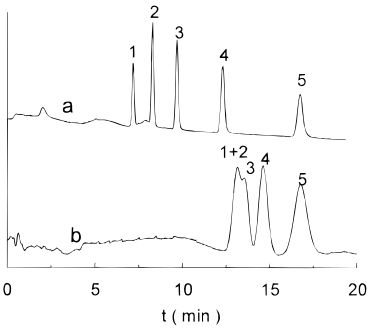

Figure 1. Chromatograms for the separation of five neutral solutes in (a) DMSCX-CEC and (b) DMS-CEC at low pH of the eluent. Experimental conditions: column, 75 μm i.d. × 375 μm o.d. packed with (a) 5 μm Spherisorb-SCX and (b) 5 μm Spherisorb-silica gel, packed/total length = 10/31 cm; applied voltage, 15 kV; electrokinetic injection 10 s × 5 kV; UV detection wavelength, 214 nm; mobile phase, methanol/25 mM CTAB/100 mM NaH_2PO_4 buffer (pH 2.21)/water = 60/8/10/26. Solutes: (1) benzene, (2) toluene, (3) ethylbenzene, (4) *n*-propylbenzene, (5) *n*-butylbenzene.

phases containing 4 and 2 mM CTAB, respectively:

CTAB: 4 mM $\quad \log k' = 0.186\text{Nc} - 1.38, \quad r = 0.9989, \quad n = 5$

2 mM CTAB: $\quad \log k' = 0.182\text{Nc} - 1.48, \quad r = 0.9988, \quad n = 5$

An excellent linear relationship ($r > 0.998$) between the logarithm of the capacity factor (log k') and the number of carbons (Nc) was observed with the two different mobile phases investigated. The positive value of the slope means that the retention value in DMSCX-CEC increases with the hydrophobicity of the solutes, which strongly supports the reversed-phase partioning mechanism in DMSCX-CEC.

Effect of pH Value. Both the bare silica and the SCX packings can be dynamically modified by CTAB, but only very few silanol groups on the bare silica surface were ionized at the low pH values of the mobile phases; therefore, the amount of CTAB adsorbed was very small and the EOF was quite low. Obviously, it is difficult to perform DMS-CEC experiments with low-pH mobile phases. However, the strong sulfonic acid groups on the SCX packing were ionized at a wide range of the pH values; thus, DMSCX-CEC can be conducted with low-pH mobile phases. The separation of five neutral solutes in DMS-CEC and DMSCX-CEC using the same mobile phase with a pH value of 2.21 is shown in Figure 1. It can be seen from Figure 1 that the five solutes were completely separated in DMSCX-CEC but only partially separated in DMS-CEC. Furthermore, μ_{eo} for DMSCX-CEC is greater than that for DMS-CEC due to the limited amount of silanol groups on the bare silica surface that were ionized at low pH. Strong acids which cannot be eluted at high pH values of mobile phases because of their high electrophoretic mobilities, however, can be separated in DMSCX-CEC at low pH values with the ion-suppressed mode. Figure 2 shows the separation of five organic acids in DMSCX-CEC at pH 2.21.

The effects of eluent pH on the electroosmotic flow and the retention of neutral solutes were investigated in this work. Five mobile phases were prepared by mixing 30 mL of methanol, 4 mL of CTAB solution (25 mM), and 5 mL of 100 mM phosphate buffers with pH values of 2.2, 3.0, 5.23, 6.3, and 7.86, respectively, and then adjusted to 50 mL by the addition of water. The apparent pH values of the prepared mobile phases were measured by a pH meter as 3.48, 4.33, 6.63, 7.73, and 9.16, respectively. The mobility of EOF (μ_{eo}) was calculated by eq 2, where L_t is the total

$$\mu_{eo} = (L_d L_t)/(V t_0) \qquad (2)$$

length of the capillary, L_d is the length of the capillary from the inlet to the detector window, V is the applied voltage, and t_0 is the void time which was measured by baseline disturbance. The μ_{eo} values at various apparent pH values of mobile phases were measured as shown in Figure 3. It can be seen that μ_{eo} value increases very quickly with pH values increasing from 3.48 to 6.63 but slightly decreases with pH values increasing from 6.63 to 9.16. The negative charge density on the SCX packing surface was provided by the sulfonic and silanol groups simultaneously. Both groups are completely ionized at the high pH values of the mobile phases; however, the ionization of the silanol group is suppressed at low pH values. With increasing pH, the amount of ionized silanol groups increases, thereby increasing the charge density also, which results in the increase of the μ_{eo} value with the pH increasing values from 3.48 to 6.63. Because both groups are

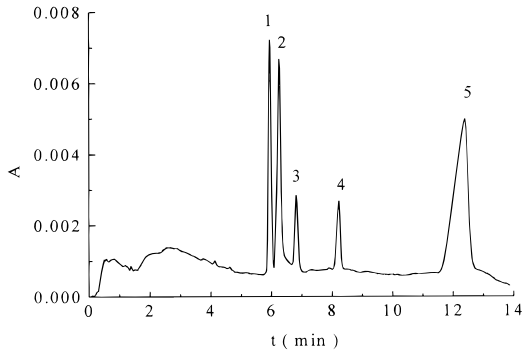

Figure 2. Chromatogram for the separation of acidic compounds in DMSCX-CEC at low pH of the eluent. Experimental conditions are the same as those for Figure 1. Solutes: (1) phenylacetic acid (pK_a 4.28), (2) benzoic acid (pK_a 4.20), (3) *o*-toluic acid (pK_a 3.91), (4) *p*-nitrobenzoic acid (pK_a 3.43), (5) phthalic acid (pK_a 2.95).

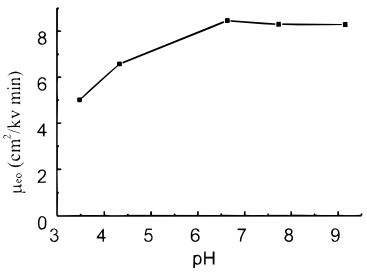

Figure 3. Effect of apparent pH values of the mobile phase on electroosmotic mobility (μ_{eo}). Experimental conditions: column, 75 μm i.d. × 375 μm o.d. packed with 5 μm Spherisorb-SCX, packed/total length = 10/31 cm; applied voltage, 15 kV; electrokinetic injection, 10 s × 5 kV; UV detection wavelength, 214 nm; mobile phases, methanol/25 mM CTAB/100 mM NaH_2PO_4 buffers (pH 2.20, 3.00, 5.23, 6.30, 7.86)/water = 60/8/10/22 with apparent pH values at 3.48, 4.33, 6.63, 7.73, and 9.16, respectively.

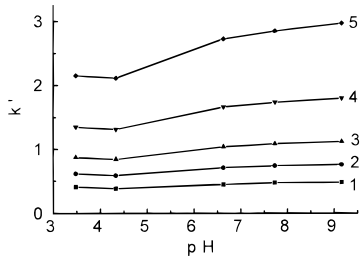

Figure 4. Effect of apparent pH of the mobile phase on capacity factors (k') of neutral solutes. Experimental conditions are the same as those for Figure 3. Solutes: (1) benzene, (2) toluene, (3) ethylbenzene, (4) *n*-propylbenzene, (5) *n*-butylbenzene.

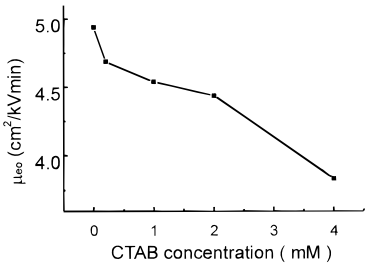

Figure 5. Effect of CTAB concentration on electroosmotic mobility (μ_{eo}). Experimental conditions: column, 75 μm i.d. × 375 μm o.d. packed with 5 μm Spherisorb-SXC, packed/total length = 10/31 cm; applied voltage, 15 kV; electrokinetic injection, 10 s × 5 kV; UV detection wavelength, 214 nm; mobile phases, methanol and 100 mM NaH_2PO_4 buffer (pH 2.21) kept at 60% and 10%, but changing the ratio of 25 mM CTAB solution to water.

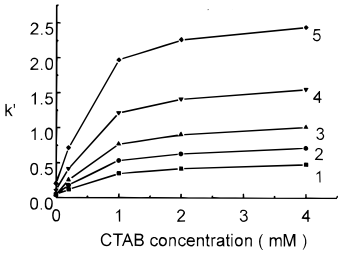

Figure 6. Effect of CTAB concentration on capacity factors (k') of neutral solutes. Experimental conditions are the same as those for Figure 5. Solutes: (1) benzene, (2) toluene, (3) ethylbenzene, (4) *n*-propylbenzene, (5) *n*-butylbenzene.

completely ionized at high pH values, the charge density does not increase with a further increase in pH; therefore, the influence of pH on the μ_{eo} value is moderate when the apparent pH is greater than 6.63. The pH of the stock buffer solutions of high pH was adjusted by NaOH solution; therefore, the higher the pH of the buffer, the higher the ionic strength, which may explain the slight decrease in μ_{eo} from pH 6.63 to 9.16. The effect of the pH values of the mobile phases on the retention values of neutral solutes was determined as shown in Figure 4. The k' values slightly increase with increasing pH. The reason for this may be that ionized silanol groups of the packing surfaces increase with increasing pH, and therefore the amount of the adsorbed CTAB increases, which results in stronger retention of neutral solutes on the packing surfaces.

Effect of CTAB Concentration. It is well-known that the magnitude and direction of EOF can be adjusted by various factors such as surfactant additive, organic modifier, and pH value of buffer in CZE. Similar phenomena were also found in CEC. Pfeffer et al.[25] have reported that the velocity of EOF in open tubular CEC (OTCEC) can be enhanced by a low concentration of CTAB in the mobile phase. Govindaraju et al.[26] and Ye et al.[22] found that EOF in CEC columns packed with bare silica decreased with the additions of spermine and CTAB, respectively. EOF in the ODS-packed columns was also found to decreased with an increase in the competing base concentration.[2,27] The EOF modifiers in CZE are always amine bases, but those in CEC can be anionic surfactants. Seifar et al.[4] found that the velocity of EOF in a column packed with ODS increased with increasing SDS concentration. EOF in packed-column CEC (PCCEC) is determined by the ζ potential at the packing surface, which is mainly related to the charge density of the packing surface. The effect of the CTAB concentration on EOF in the column packed with SCX material was studied. Figure 5 shows the plot of the electroosmotic mobility (μ_{eo}) as a function of the CTAB concentration. It can be seen that EOF decreases with increasing CTAB concentration in the mobile phase. For example, EOF with the addition of 4 mM CTAB in the mobile phase is reduced by about 25% of its value without the presence of CTAB. The reason for the decrease of μ_{eo} with increasing CTAB concentration is that part of the negative charge on the SCX packing was neutralized by the adsorbed CTAB. The EOF direction was reversed when the CTAB concentration exceeded about 0.5 mM in CE because a bilayer of CTAB molecules was formed on the capillary wall,[22] but the reversal of EOF was not found in both DMS-CEC[22] and DMSCX-CEC. The reason for this may be that the surface areas of the packings in CEC are much larger than those of the capillary walls and the charge densities are also much greater; therefore, more CTAB is needed to neutralize the charged groups in CEC and it is difficult to form bilayers on packing surfaces.

The effect of the CTAB concentration on the k' values of five neutral solutes is shown in Figure 6. It can be seen from Figure 6 that the effect of CTAB on capacity factors is very similar to the isotherm of Langmuir adsorption. In the range of low CTAB concentrations, k' values increase with increasing CTAB concentration very quickly, while in the range of high CTAB concentrations, k' values increase relatively moderately. For example, an increase in the CTAB concentration from 0 to 1 mM resulted in an 8.7-fold increase in k'(*n*-butylbenzene), but an increase in the CTAB concentration from 1 to 4 mM resulted in only a 0.24-fold increase of k'. This result means that an increase in CTAB concentration will increase the hydrophobicity of the stationary phase and, thereby, the retention of solutes.

Effect of Methanol Concentration. The influence of the methanol fraction on EOF in DMSCX-CEC was investigated. It was found that an increase in the methanol fraction from 50% to 70% (v/v) resulted in an increase in μ_{eo} by about 60% in this system. According to our previous result, it was observed that an increase in the methanol fraction from 60% to 90% resulted in an increase in μ_{eo} of only 30% in reversed-phase CEC with 3 μm Spherisorb-ODS as the stationary phase.[28] Apparently, the adsorp-

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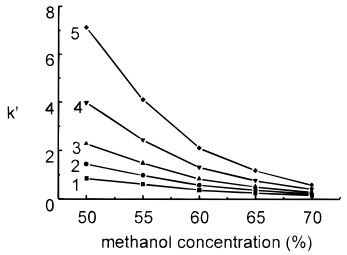

Figure 7. Effect of methanol fraction on capacity factors (k') of neutral solutes. Experimental conditions: column, 75 μm i.d. × 375 μm o.d. packed with 5 μm Spherisorb-SCX, packed/total length = 10/31 cm; applied voltage, 15 kV; electrokinetic injection, 10 s × 5 kV; UV detection wavelength, 214 nm; mobile phases, 25 mM CTAB solution and 100 mM NaH_2PO_4 buffer (pH 2.21) kept at 8% and 10%, but changing the ratio of methanol to water. Solutes: (1) benzene, (2) toluene, (3) ethylbenzene, (4) *n*-propylbenzene, (5) *n*-butylbenzene.

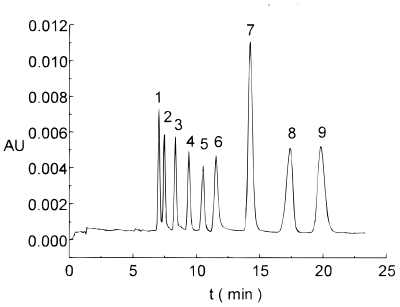

Figure 8. Simultaneous separation of acidic and neutral solutes at low pH. Experimental conditions: column, 75 μm i.d. × 375 μm o.d. packed with 5 μm Spherisorb-SCX, packed/total length = 10/31 cm; applied voltage, 15 kV; electrokinetic injection, 10 s × 5 kV; UV detection wavelength, 214 nm; mobile phase, methanol/25 mM CTAB/100 mM NaH_2PO_4 buffer (pH 2.21)/water = 55/8/10/27. Solutes: (1) phenylacetic acid (pK_a 4.28), (2) benzoic acid (pK_a 4.20), (3) *o*-toluic acid (pK_a 3.91), (4) toluene, (5) *p*-nitrobenzoic acid (pK_a 3.43), (6) ethylbenzene, (7) naphthalene, (8) phthalic acid (pK_a 2.95), (9) biphenyl.

tion of CTAB on the surface of SCX packings is strongly dependent on the methanol fraction. The amount of adsorbed CTAB decreases with increasing methanol fraction, and thereby the negative charge density increases, which very quickly causes an increase in μ_{eo}.

The effect of the methanol fraction on the k' values of five neutral solutes is shown in Figure 7. The separation of neutral solutes in this system was mainly based on the reversed-phase partitioning mechanism. There are two reasons for the decrease in k' with increasing methanol fraction as shown in Figure 7. First, an increase in the methanol fraction will increase the elution strength of the mobile phase as in reversed-phase CEC, which results in a decrease in the k' values of the neutral solutes. Second, the amount of CTAB adsorbed on the SCX packing surface decreases with increasing methanol fraction, and thereby the hydrophobicity of the packing decreases, which also results in a decrease in the k' values for the neutral solutes.

Simultaneous Separation of Acidic, Basic, and Neutral Solutes. Although both the ionic and neutral compounds can be separated by CEC in theory, most of the reported applications of CEC have been focused on the analysis of neutral compounds. Separation of acidic and basic compounds meets some difficulty in CEC. It is relatively difficult to apply CEC to the separation of

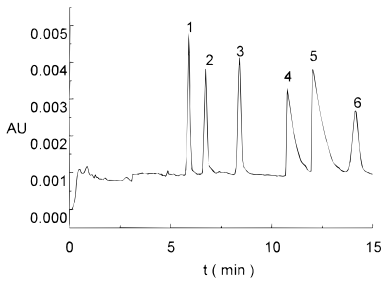

Figure 9. Simultaneous separation of acidic, basic, and neutral solutes at low pH. Experimental conditions: column, 75 μm i.d. × 375 μm o.d. packed with 5 μm Spherisorb-SCX, packed/total length = 10/31 cm; applied voltage, 15 kV; electrokinetic injection, 10 s × 5 kV; UV detection wavelength, 214 nm; mobile phase, methanol/25 mM CTAB/100 mM NaH_2PO_4 buffer (pH 2.21)/water = 60/8/10/22. Solutes: (1) phenylacetic acid (pK_a 4.28), (2) *o*-toluic acid (pK_a 3.91), (3) ethylbenzene, (4) quinoline (pK_b 9.15), (5) 2-methylquinoline (base), (6) *n*-butylbenzene.

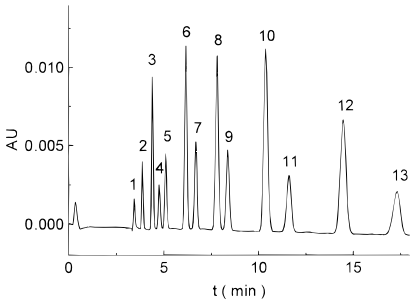

Figure 10. Simultaneous separation of neutral and basic solutes at high pH. Experimental conditions: column, 75 μm i.d. × 375 μm o.d. packed with 5 μm Spherisorb-SCX, packed/total length = 10/31 cm; electrokinetic injection, 10 s × 5 kV; UV detection wavelength, 214 nm; mobile phase, methanol/25 mM CTAB/100 mM NaH_2PO_4 buffer (pH 7.65)/water = 60/8/10/22. Solutes: (1) formamide, (2) pyridine (pK_b 8.83), (3) aniline, (4) quinoline (pK_b 9.15), (5) 2-methylquinoline (base), (6) phenetole, (7) toluene, (8) bromobenzene, (9) ethylbenzene, (10), naphthalene, (11) *n*-propylbenzene, (12) biphenyl, (13) *n*-butylbenzene.

acidic solutes in their ionized forms because they tend to migrate against the EOF, causing the migration times to become relatively long or even too long for elution to occur. To separate acidic solutes successfully, reversed-phase CEC with the ion-suppressed mode using low-pH mobile phases was recommended.[29] The major difficulty of reversed-phase CEC in separating basic compounds is peak tailing.[30] Smith and Evans[7] reported discouragingly poor peak shapes for strongly basic compounds on the silica-based ODS stationary phase. Gillott et al.[2] reported that good peak symmetry in the separation of pharmaceutical bases can be achieved by the addition of a competing base to the mobile phase. Lurie et al.[27] achieved the simultaneous separation of acidic, basic, and neutral compounds using reversed-phase CEC with Hypersil-C_8 as the stationary phase and a low-pH buffer containing hexylamine as the mobile phase.

Figure 8 shows the simultaneous separation of acidic and neutral compounds using DMSCX-CEC with a low-pH mobile phase. Excellent separation of all nine compounds was ac-

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(30) Majors, R. E. *LC-GC* **1998**, *16*, 96.

complished. Under these conditions, chromatography plays a major role in the separation process of weakly acidic and neutral compounds, but chromatography and electrophoresis processes may contribute to the separation of strong acids simultaneously. Figure 9 shows the simultaneous separation of neutral, acidic, and basic compounds using DMSCX-CEC with a low-pH mobile phase. Although all six of the compounds could be well separated, peak tailing for the basic compounds was observed, probably caused by the strongly electrostatic interactions between the positive charges on the basic compounds and the negative charges on the SCX packing. However, it was found that peak tailing of the basic solutes could be eliminated by also performing CEC in the ion-suppressed mode with a high-pH eluent. Simultaneous separation of basic and neutral compounds as shown in Figure 10 was accomplished by DMSCX-CEC using a high-pH mobile phase. All neutral and basic compounds were well separated mainly on the basis of the reversed-phase partition mechanism, and very good peak symmetry for the basic compounds was observed.

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