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Development of a chiral non-aqueous capillary electrophoretic system using the partial filling technique with UV and mass spectrometric detection

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Abstract

A chiral non-aqueous CE system with UV and mass spectrometric detection has been developed. The enantioseparation was promoted by diastereomeric complex (ion-pair) formation between the amines (e.g. salbutamol, atenolol) and the chiral selector, (−)-2,3:4,6-di-*O*-isopropylidene-2-keto-L-gulonic acid [(−)-DIKGA]. Different solvent mixtures were studied, as well as different concentrations of (−)-DIKGA and ammonium acetate in the background electrolyte. A partial filling technique was developed with a selector plug composed of (−)-DIKGA and ammonium acetate in a solvent mixture of methanol and 2-propanol. The separated enantiomers of pronethalol were detected by a Q-TOF MS system equipped with a sheath-flow electrospray ionization interface.

Keywords: Non-aqueous capillary electrophoresis; Enantiomer separation; Partial filling technique; Amines; (−)-DIKGA; Pronethalol

1. Introduction

Non-aqueous capillary electrophoresis (NACE) was introduced by Wahlbroehl et al. [1] in 1984. Different selectivity than that in aqueous media can be obtained through differences in solvation as well as dielectric constants, yielding changes in coloumbic interactions [2,3]. Furthermore, NACE enables separation of substances with poor water solubility [4]. The lower conductivity of non-aqueous media makes it possible to use high voltages without generation of high currents [5,6]. In some cases, however, disadvantages with non-aqueous media may be rapid evaporation from the vials and high UV absorption [7]. The problem with UV absorption could be overcome using NACE–MS [8].

The use of chiral selectors in NACE for analysis of enantiomers has previously been demonstrated [2,9, and references cited therein]. The low dielectric constants of many organic solvents [10] promote a high degree of diastereomeric ion-pair formation when using counter-ions as chiral selectors. Different counter-ions, e.g. quinine [11], (+)-(*S*)-camphorsul-

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phonic acid [12], (+)-18-crown-6-tetracarboxylic acid [13], benzoxycarbonylglycyl-L-proline (L-ZGP) [14] as well as (−)-2,3:4,6-di-*O*-isopropylidene-2-keto-L-gulonic acid [(−)-DIKGA] [15] have been used for chiral separations. However, all these applications were based on UV detection. The reduced sensitivity caused by inherent UV absorption of the chiral counter-ion in NACE, has in a few previous studies been improved by utilization of the partial filling technique (PFT) [16–19]. In the PFT, only a part of the capillary is filled with selector solution (Fig. 1). The solutes are separated in the selector plug (zone 1). Following separation, the enantiomers migrate out of the selector plug into zone 2 and are detected without interference from the chiral selector.

An attractive possibility in order to improve the analytical performance is by using CE with mass spectrometric (MS) detection [20,21]. This technique provides a high detection sensitivity irrespective of the UV absorbing properties of the sample. This is advantageous, e.g. for determination of enantiomeric purity of pharmaceuticals. Furthermore, the use of collision-induced dissociation (MS–MS) promotes a highly selective detection technique. However, since enantiomers have indistinguishable m/z ratios, an electrophoretic chiral separation is still necessary prior to the MS. Chiral separations based on the partial filling technique in aqueous CE–MS have previously been published [22,23].

(−)-DIKGA has been introduced by our group as a chiral selector in LC [24] and CE [15]. The present study is a continuation of this ongoing project, where we have aimed at developing a chiral NACE system with on-line MS detection for enantiomeric amines (e.g. pronethalol, atenolol). The strategy was first to investigate the influence of different background electrolyte (BGE) compositions (e.g. solvent mixtures, chiral selector and buffer concentrations) on the chiral resolution, using a capillary completely filled with the selector. The study was then continued by optimizing a partial filling system. Special care was taken to delay the arrival of the co-migrating selector zone at the detector. Finally, coupling of the separation system to a mass spectrometer was demonstrated.

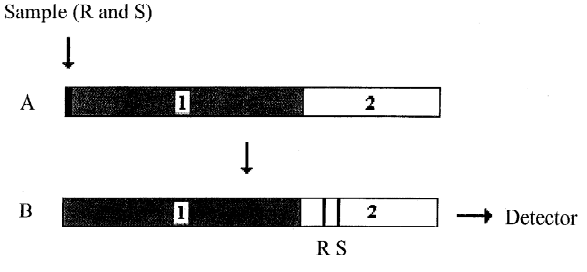

Fig. 1. Schematic illustration of the partial filling technique (PFT). (A) Sample introduction in a capillary partially filled with the chiral selector (zone 1). (B) After separation, the resolved enantiomers migrate through the zone without selector (zone 2) towards the detector.

2. Materials and methods

2.1. Instrumentation

Development of the chiral NACE system was carried out using a Beckman P/ACE 2050 (Beckman, Fullerton, CA, USA) equipped with a UV detector and System Gold software, version 7.11 and an HP ^{3D}CE (Hewlett-Packard, Waldbronn, Germany) with HP ^{3D}CE Chemstation (Rev. A. 06. 03) software. The CE–MS experiments were carried out using the HP ^{3D}CE instrument. An orthogonal Z-spray sheath liquid electrospray ionization interface (Micromass, Manchester, UK) was used for the CE–MS coupling. A Micromass Q-TOF model I, upgraded with a 3.6 GHz time-to-digital converter card (Micromass, Manchester, UK) with Masslynx version 3.3 software was used for the MS detection. The flow of sheath liquid was obtained by a Harvard Apparatus 22 syringe pump (Harvard Apparatus, Holliston, MA, USA) with a 250-μl Hamilton syringe. The fused-silica capillaries (50 μm I.D., 365 μm or 186 μm O.D.) were obtained from MicroQuartz (Munich, Germany).

2.2. Chemicals

The chiral selector (−)-DIKGA was purchased from Fluka (Buchs, Switzerland). Methanol, 2-propanol and acetonitrile, all of HPLC grade, were supplied by Fischer (Loughborough, UK). Ammonium acetate (>98%), mesityl oxide, *rac*-proprano-

lol hydrochloride, *rac*-sotalol hydrochloride, *rac*-pindolol (free base) and (+)-*ψ*- and (−)-*ψ*-ephedrine (free bases) were from Sigma (St Louis, MO, USA). *rac*-Pronethalol hydrochloride was supplied by ICI (Macclesfield, UK). *rac*-Labethalol hydrochloride (*RR*/*SS* and *RS*/*SR*) and *rac*-salbutamol sulphate were from Glaxo (Greenford, UK). *rac*-Atenolol hydrochloride, *rac*-metoprolol tartrate and *rac*-alprenolol benzoate were generous gifts from Astra Hässle (Mölndal, Sweden). *rac*-Bambuterol hydrochloride and *rac*-terbutaline sulphate were kind donations of Astra Draco (Lund, Sweden). *rac*-Isoprenaline sulphate was obtained from Apoteket (Stockholm, Sweden). Sodium hydroxide, hydrochloric acid and acetic acid (all analytical reagent-grade) were obtained from Merck (Darmstadt, Germany). All chemicals were used as received without further treatment. The water used in the experiments was purified in a Milli-Q Academic water system (Millipore, Bedford, MA, USA). The structures of the solutes and the counter-ion are given in Figs. 2 and 3.

2.3. Electrophoretic procedure

The solutes were dissolved in methanol at a concentration of 0.1 m*M*. Prior to use, all solutions were degassed by sonication. New capillaries were conditioned with: 1 *M* NaOH, water, 1 *M* HCl for 5 min each, followed by water for 20 min all at 20 p.s.i. (1.4 bar) in the Beckman P/ACE 2050. The same procedure was carried out with new capillaries on the HP ^{3D}CE system, but at a pressure of 1.0 bar. The capillaries were flushed with the BGE for 5 min at 20 p.s.i. (1.4 bar) on the Beckman P/ACE 2050 and at 1.0 bar on the HP ^{3D}CE between analyses. The samples were introduced hydrodynamically at the anodic end of the capillary by a pressure of 0.5 p.s.i. (35 mbar) for 5 s on both instruments. Mesityl oxide was used as a marker of the electroosmosis. The separations were carried out at 25 kV in capillaries with a total length of 67 cm unless otherwise stated. In the experiments with UV detection, the amines were detected at 214 nm through a window made 7 or 8.5 cm (in the Beckman P/ACE 2050 and HP ^{3D}CE instruments, respectively) from the cathodic end of the capillary. The separations were carried out at ambient temperature on the Beckman instrument and at a temperature set to 25 °C on the HP ^{3D}CE instrument. The resolution, R_s, was calculated as $2(t_{mig2}-t_{mig1})/(w_{t1}+w_{t2})$, where t_{mig2} and t_{mig1} are the migration times of enantiomer 2 and 1, respectively, and w_{t1}, w_{t2} are the base widths of the peaks between the tangents in time units. The apparent efficiency, N_{app}, was calculated as $16(t_{mig}/w_{base})^2$ where t_{mig} and w_{base} are the migration time and the peak base width between the tangents in time units, respectively. In the partial filling experiments, Δt was calculated as $(t_{front}-t_{mig2})$, where t_{front} represents the time to 50% of the maximum response of the selector plug. The term filling degree corresponds to the percentage of the effective capillary length filled with the selector plug.

2.4. Mass spectrometric procedure

The MS parameters were tuned using pronethalol or *ψ*-ephedrine dissolved at 0.01 m*M* in the background electrolyte without selector and with the CE voltage switched on at +20 kV. The sheath liquid used was a mixture of methanol–water (50:50, v/v) with addition of 0.25% (~44 m*M*) acetic acid. The sheath flow was set to 1 μl/min. In order to avoid suction from the outlet end of the capillary, the sheath gas (nitrogen), sheath liquid and the MS capillary voltage were switched off during CE injection. Samples were injected into the capillary for 1 s at a pressure of 35 mbar, followed by an injection of pure methanol for 2 s at 35 mbar. During the separation, a counter pressure of 15 mbar was applied. The electrospray ionization (ESI) interface parameters were set as follows: capillary 4500 V, cone 35 V and extractor 10 V. Prior to the MS–MS experiment, the hexapole collision cell was filled with argon at an inlet pressure of 15 p.s.i. (1.0 bar), and a collision energy of 30 eV was used. The quadrupole was set to transmit the parent ion m/z $[M+H]^+$, and the time-of-flight (TOF) detector was set to acquire data in a mass-to-charge (m/z) range over the daughter ions of interest. Injection of different enantiomeric ratios of *ψ*-ephedrine was used to verify the chiral separation of this compound in the CE–MS system.

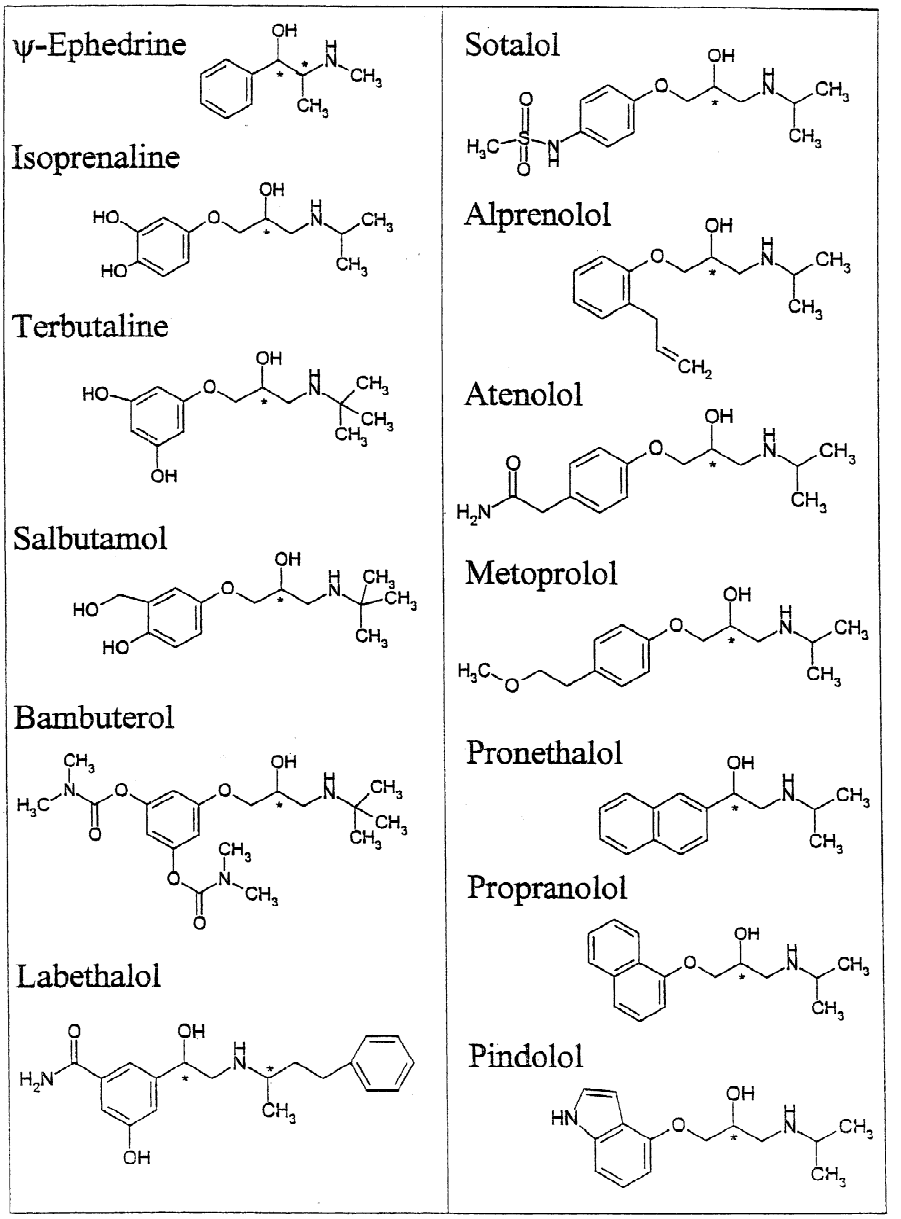

Fig. 2. Solute structures.

3. Results and discussion

The structure of the chiral selector (−)-DIKGA is given in Fig. 3. This selector (counter-ion) has a rigid structure and possesses hydrogen accepting ether functions in the vicinity of the carboxylic group. Thus, (−)-DIKGA should enable multipoint interactions with enantiomeric amines, cf. the ''three-point interaction model'' for chiral discrimination by complexation with a chiral selector [25,26]. In our previous study, it was demonstrated that (−)-DIKGA promotes enantioseparation of amines using a methanolic BGE [15]. Sodium hydroxide was added to the BGE to ionize the (−)-DIKGA. The possibility to

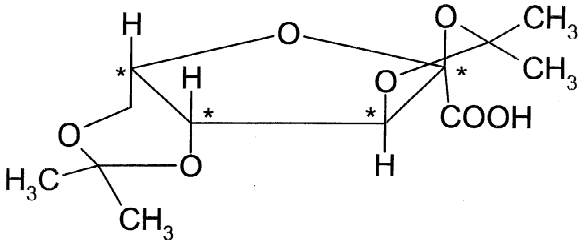

Fig. 3. The chiral selector (−)-DIKGA.

improve the enantioresolution by exchanging the BGE solvent was not investigated in that study.

3.1. Chiral resolution and solvent composition

Addition of organic modifiers to the BGE affects the dielectric constant (ε), the viscosity (η) as well as the solvation of the ion-pair and its components. Thus, the effect of organic additives on the chiral resolution is difficult to predict. The initial experiments were carried out to evaluate the influence of different solvent mixtures on the enantioseparation of pronethalol and atenolol (Table 1). The study was made in a capillary completely filled with the selector solution. The composition of the BGEs was 100 m*M* (−)-DIKGA and 40 m*M* sodium hydroxide dissolved in methanol or in methanol with addition of 2-propanol, acetonitrile or water. Due to poor solubility of (−)-DIKGA and sodium hydroxide, some initially intended solvent mixtures could not be evaluated. Addition of acetonitrile to the methanolic BGE improved $\Delta\mu$ but a decrease in chiral resolution (R_s) was observed (Table 1). This reduced enantioresolution was probably a result of the increased electroosmotic flow (EOF). The ratio between dielectric constant and viscosity (ε/η) has proven to be an important factor for determination of EOF [27]. The increased EOF at higher contents of acetonitrile may be a result of the increase in ε [28] and/or the decrease in η [29] in the acetonitrile–methanolic solvent mixtures.

Addition of water to the methanolic BGE reduced the EOF as well as $\Delta\mu$ and the chiral resolution. The reduction in EOF was probably due to the increasing viscosity at higher concentrations of water [30]. Addition of 25 or 50% (v/v) of the more viscous 2-propanol to the methanolic BGE decreased the EOF and improved the enantioresolution for pronethalol although the $\Delta\mu$ was lowered. BGEs with 25% of 2-propanol were preferable since a 50% addition significantly increased the migration times, while only slightly improving the resolution.

3.2. Different electrolytes

In the earlier study [15], sodium hydroxide was added to methanol in order to obtain the anionic

Table 1
Enantioseparations in different solvent mixtures

Co-solvent	% (v/v)	μ_{eo} ($cm^2\ V^{-1}\ s^{-1}$), $\times 10^5$	Pronethalol		Atenolol	
			$\Delta\mu$ ($cm^2\ V^{-1}\ s^{-1}$), $\times 10^6$	R_s	$\Delta\mu$ ($cm^2\ V^{-1}\ s^{-1}$), $\times 10^6$	R_s
–		5.4	6.2	2.7	2.8	1.4
Acetonitrile	25	17	6.8	1.9	[a]	[a]
	50	25	6.7	1.7	[a]	[a]
	75	32	6.5	1.3	[a]	[a]
Water	5	4.6	4.3	2.3	1.4	0.9
	25	4.5	1.3	0.8	0.4	0.4
2-Propanol	5	3.9	6.2	3.5	2.4	1.4
	25	3.6	4.9	3.9	1.8	1.5
	50	<2.2	3.1	4.2	0.9	1.5

Conditions: completely filled capillary. BGE: 100 m*M* (−)-DIKGA and 40 m*M* NaOH dissolved in co-solvent and MeOH.
[a] Not separated.

(−)-DIKGA, i.e. the counter-ion. Only the selector was used as buffer, in order to avoid ion-pair formation with achiral buffer anions. However, to facilitate the MS coupling, sodium hydroxide was exchanged for the more volatile ammonium acetate buffer in the BGE. An additional buffer, e.g. ammonium acetate is also required in PFT to obtain sufficient conductivity in the zone without the selector (zone 2, Fig. 1).

The exchange of sodium hydroxide for ammonium acetate did not impair the enantioresolution (Table 2). Thus, ammonium acetate was chosen for the remaining experiments.

Evaluation of different concentration ratios between the chiral selector and buffer were carried out (Fig. 4). The ammonium acetate concentration was varied between 10 and 150 m*M* at a constant concentration of 100 m*M* (−)-DIKGA. There seemed to be a maximum enantioresolution for pronethalol between 25 and 75 m*M* ammonium acetate. Because of decreased $\Delta\mu$, the enantioresolution decreased at higher concentrations of ammonium acetate. The reduced mobility difference may be caused by increased competition between the ammonium ions and the solute for binding the negatively charged (−)-DIKGA or increased interaction between the solutes and the acetate ions present in the BGE. As a result of the noisy baseline at higher concentrations

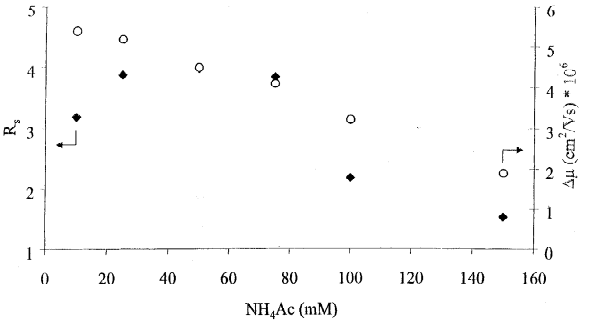

Fig. 4. Ammonium acetate concentration and chiral separation. Conditions: completely filled capillary. BGE: 100 m*M* (−)-DIKGA with varied concentrations of NH_4Ac in MeOH with 25% of 2-PrOH. Solute: pronethalol. ♦, R_s; ○, $\Delta\mu$.

of ammonium acetate, the concentrations of 100 m*M* (−)-DIKGA and 25 m*M* ammonium acetate were considered to be the most useful.

Separations of the enantiomers of pronethalol were subsequently carried out with different total concentrations of the chiral selector and ammonium acetate (Fig. 5), but at a constant concentration ratio of 4:1. At higher concentrations than 50 m*M* (−)-DIKGA, a decreased $\Delta\mu$ was observed. However, the resolution was improved between 20 and 70 m*M* (−)-DIKGA after which it leveled off, probably depending on the EOF which decreased from 1.4×10^{-4} to 5.6×10^{-5} cm^2 V^{-1} s^{-1} in the interval

Table 2
Comparison between NaOH and NH_4Ac in the BGE

Solute	X			
	NaOH[a]		NH_4Ac[b]	
	$\Delta\mu$ (cm^2 V^{-1} s^{-1}), $\times10^6$	R_s	$\Delta\mu$ (cm^2 V^{-1} s^{-1}), $\times10^6$	R_s
Pronethalol	4.4	4.1	4.4	4.5
Sotalol	3.4	2.8	3.5	3.0
Ephedrine	2.4	2.0	2.3	2.1
Bambuterol	2.3	2.3	2.3	2.5
Terbutaline	2.2	2.3	2.3	2.4
Metoprolol	1.6	1.6	1.6	1.7
Pindolol	1.3	1.3	1.4	1.4
Atenolol	1.3	1.4	1.3	1.5
Alprenolol	0.9	0.9	1.0	1.0
Propranolol	0.9	0.9	1.0	1.0

Conditions: Completely filled capillary; 100 m*M* (−)-DIKGA and 40 m*M* X in MeOH with 25% of 2-PrOH.
[a] $\mu_{eo}=5.9\times10^{-5}$ cm^2 V^{-1} s^{-1}.
[b] $\mu_{eo}=5.0\times10^{-5}$ cm^2 V^{-1} s^{-1}.

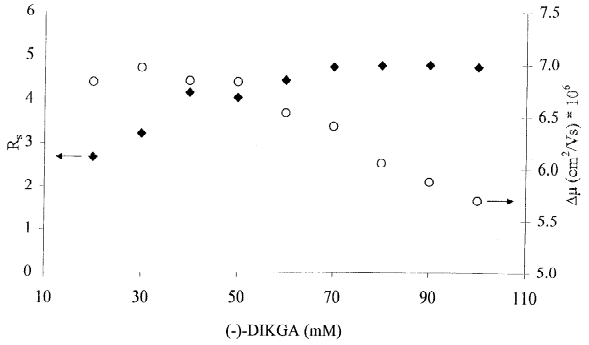

Fig. 5. (−)-DIKGA concentration and chiral separation. Maintained ratio of 4:1 between (−)-DIKGA and NH_4Ac. Other conditions as in Fig. 4. ♦, R_s; ○, $\Delta\mu$.

studied. In order to avoid high currents, (−)-DIKGA concentrations above 100 m*M* were not evaluated.

The resolution power of the system was demonstrated by a simultaneous separation of the enantiomers of *ψ*-ephedrine, isoprenaline, sotalol and (*RR*/*SS*)-labethalol in a BGE of 80 m*M* (−)-DIKGA and 40 m*M* ammonium acetate in methanol with 25% of 2-propanol (Fig. 6).

3.3. Partial filling

Introduction of a high concentration of a non-volatile compound, like (−)-DIKGA, into the ESI-MS system, can lead to a decreased detection sensitivity. In order to avoid this problem, a partial filling technique (PFT) [31] was used (Fig. 1). Initial

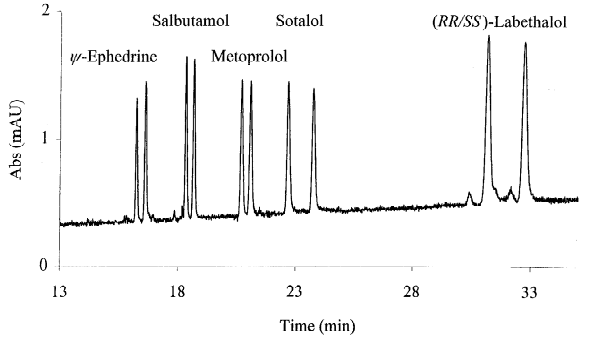

Fig. 6. Enantioseparation of *ψ*-ephedrine, salbutamol, metoprolol, sotalol and (*RR*/*SS*)-labethalol. Conditions: completely filled capillary (total length, 68.5 cm). BGE: 80 m*M* (−)-DIKGA and 40 m*M* NH_4Ac in MeOH with 25% of 2-PrOH.

experiments were carried out using the results obtained with a completely filled capillary and UV detection. However, these conditions were not directly transferable to the partial filling experiments, as a few other complications are introduced using the latter technique (e.g. band broadening in the zone interface).

Although (−)-DIKGA is a carboxylic acid, its net migration was towards the cathode (i.e. the same direction as the solutes). The following main objective was thus to enable undistorted detection of the solutes ahead of the selector zone (zone 1, Fig. 1). This could be done by delaying the arrival of zone 1 at the detector, by a decrease in EOF. Acetic acid was added to the zone without selector (zone 2, Fig. 1) for this purpose. It was found to successfully retard the selector, possibly by neutralizing the wall charges in the zone without selector, i.e. reducing the EOF (Fig. 7). Hence, addition of acetic acid enables more slowly migrating solutes to reach the detector ahead of the selector and thus the use of a longer selector plug. The influence of different concentrations of 2-propanol in zone 2 on the resolution of pronethalol was also evaluated (Table 3). Higher concentrations of 2-propanol than 25% (i.e. the same concentration as in the selector plug) increased the migration time, without further improving the enantioresolution or Δt.

The migration velocity of a solute through the

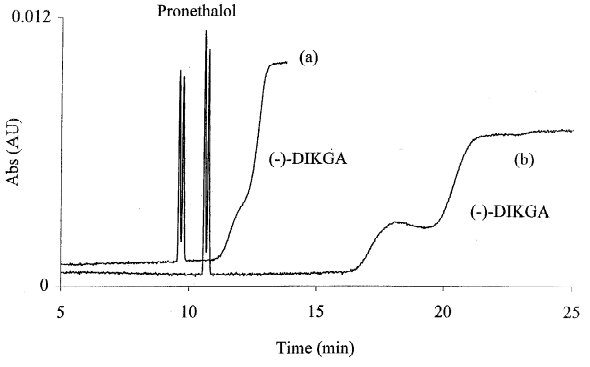

Fig. 7. Acetic acid and front migration. Conditions: partially filled capillary (total length, 57 cm), plug length 28 cm. Solute: pronethalol. BGE with selector (zone 1): 80 m*M* (−)-DIKGA and 40 m*M* NH_4Ac in MeOH. BGE without selector (zone 2): (a) 40 m*M* NH_4Ac or (b) 40 m*M* NH_4Ac and 30 m*M* HAc in MeOH. Separation voltage: +20 kV.

Table 3
Different concentrations of 2-propanol in zone 2

2-Propanol (%, v/v)	R_s	Δt (min)[a]
15	2.0	2.5
25	2.3	3.7
50	1.3	1.8

Conditions: Partially filled capillary. Plug length: 36 cm. Solute: pronethalol. BGE with selector (zone 1): 80 m*M* (−)-DIKGA and 20 m*M* NH_4Ac in MeOH with 25% of 2-PrOH. BGE without selector (zone 2): 20 m*M* NH_4Ac in MeOH with varied concentration of 2-PrOH.

[a] $\Delta t = (t_{front} - t_{mig2})$.

detector window is difficult to determine in a partial filling experiment, as the local electrical field strength is dependent on the length and composition of the selector plug. Thus, the separation efficiency expressed as the number of theoretical plates cannot be easily determined. Therefore, an apparent number of theoretical plates (N_{app}) was used to evaluate the peak efficiency at different selector plug lengths, using (*S*)-propranolol as the model compound (Fig. 8). Initially, a decreased apparent efficiency was observed at increasing selector plug length. However, N_{app} was dramatically increased at plug lengths where the solute peak appeared on top of the selector plug (Fig. 8, at plug lengths exceeding 21 cm), indicating that a major contribution to the zone broadening occurs when the solute departs from the plug containing the selector.

In order to improve the resolution, enantioseparations of pronethalol were carried out at seven plug lengths ranging from 33 to 45 cm. The highest enantioresolution of pronethalol, still allowing the enantiomers to reach the detector ahead of the selector, was obtained using a 39-cm selector plug corresponding to a filling degree of 65% of the effective capillary length (Fig. 9). Although the plug length varied, the differences in migration times of the pronethalol enantiomers were small in the three electropherograms in Fig. 9. This might be due to the fact that the total migration times of solutes in a partial filling system is dependent on several, possibly counteracting factors, that vary with the selector plug length, such as total EOF, local electrical field strengths in the different zones and local electrophoretic mobilities.

3.4. CE–MS

The developed partial filling system was coupled to a Q-TOF mass spectrometer using a sheath-liquid electrospray ionization interface. Even though a counter pressure was applied in these experiments, the migration of the selector was faster than in UV mode, probably due to a hydrodynamic pressure incurred by the ESI. Thus, a shorter selector plug than maximally allowed in UV detection, had to be used. The extracted ion electropherogram of a daughter ion (*m*/*z* 153.4) of the resolved parent

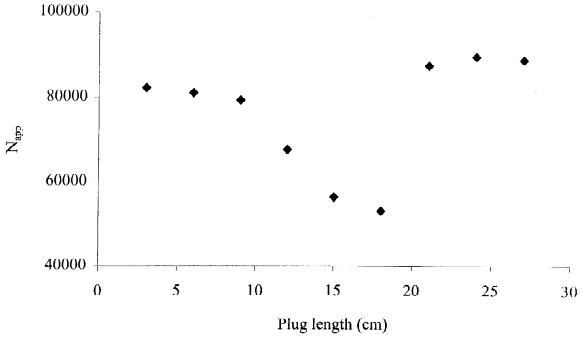

Fig. 8. Plug length and apparent efficiency (N_{app}). Conditions: partially filled capillary (total length, 37 cm). BGE with selector (zone 1): 80 m*M* (−)-DIKGA and 20 m*M* NH_4Ac in MeOH with 25% 2-PrOH. BGE without selector (zone 2): 20 m*M* NH_4Ac and 20 m*M* HAc in MeOH with 25% of 2-PrOH. Solute: (*S*)-propranolol. Separation voltage: +30 kV.

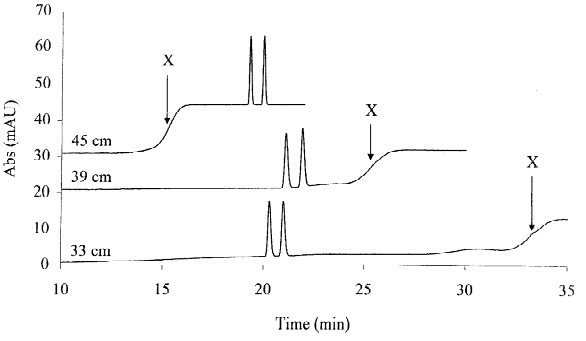

Fig. 9. Selector plug length and chiral separation. Conditions: Partially filled capillary (total length, 68.5 cm). BGE with selector (zone 1): 80 m*M* (−)-DIKGA and 20 m*M* NH_4Ac in MeOH with 25% of 2-PrOH. BGE without selector (zone 2): 20 m*M* NH_4Ac and 20 m*M* HAc in MeOH with 25% of 2-PrOH. Solute: pronethalol. X = Selector front.

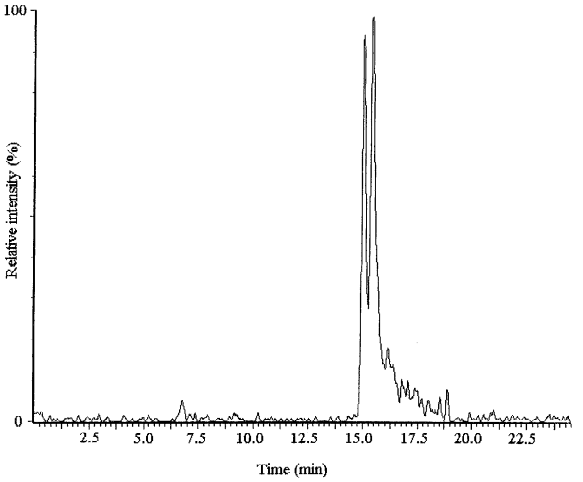

Fig. 10. MS–MS electropherogram of pronethalol. Conditions: partially filled capillary (total length, 58 cm). Plug length: 32 cm. Separation voltage: +30 kV. Parent ion $m/z=230$, daughter ion $m/z=153.4$. BGE with selector (zone 1): 80 m*M* (−)-DIKGA and 20 m*M* NH_4Ac in MeOH with 25% of 2-PrOH. BGE without selector (zone 2): 20 m*M* NH_4Ac and 20 m*M* HAc in MeOH with 25% of 2-PrOH.

enantiomers of *rac*-pronethalol (m/z 230 $[M+H]^+$) is presented as an example in Fig. 10. The enantiomers of ψ-ephedrine were also separated in the same system (results not shown). These results demonstrate that this chiral partial filling NACE system can be successfully hyphenated to a mass spectrometric detector. However, the enantioresolution was generally lower in MS than in UV mode. An increased band broadening occurred even though a counter pressure was applied. This was probably caused by suction from the outlet side of the capillary, giving rise to a parabolic flow profile. Another reason might be dilution of the sample by the sheath liquid.

4. Conclusions

A partial filling method for NACE enantioseparations has been developed using (−)-DIKGA as the chiral selector. The influence of different BGE compositions was first investigated using a capillary completely filled with the selector. Then, the special complications of the partial filling technique were investigated. A selector plug composed of (−)-DIKGA and ammonium acetate in a mixture of 25% of 2-propanol in methanol gave the highest enantioresolution. Addition of acetic acid to the zone without selector facilitated detection of more slowly migrating solutes and the use of a longer selector plug. The use of a 39-cm selector plug (corresponding to a filling degree of 65% of the effective capillary length) enabled the resolved enantiomers to reach the UV detector ahead of the selector plug. Furthermore, the aminoalcohol pronethalol served as an example to show that this chiral partial filling NACE system could be hyphenated to mass spectrometric detection.

5. Nomenclature

(−)-DIKGA	(−)-2,3:4,6-di-*O*-isopropylidene-2-keto-L-gulonic acid
2-PrOH	2-propanol
BGE	background electrolyte
MeOH	methanol
NACE	non-aqueous capillary electrophoresis
PFT	partial filling technique
Q-TOF™	quadrupole time-of-flight
ESI	electrospray ionization

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