Received: 20 April 2017 | Revised: 27 May 2017 | Accepted: 2 June 2017

DOI: 10.1002/chir.22731

REGULAR ARTICLE

WILEY

Comparison of three S-β-CDs with different degrees of substitution for the chiral separation of 12 drugs in capillary electrophoresis

Zhaokun Wang[1] | Qiongwen Zhang[1] | Linda Luo[1] | Tiemin Sun[2] | Xingjie Guo[1]

[1] Department of Pharmaceutical Analysis, School of Pharmacy, Shenyang Pharmaceutical University, Shenyang, Liaoning Province, P.R. China

[2] Department of Pharmaceutical Chemistry, School of Pharmaceutical Engineering, Shenyang Pharmaceutical University, Shenyang, Liaoning Province, P.R. China

Correspondence
Xingjie Guo, Department of Pharmaceutical Analysis, School of Pharmacy, Shenyang Pharmaceutical University, No. 103, Wenhua Road Shenhe District 110016, Shenyang, Liaoning Province, P. R. China.
Email: gxjhyz@aliyun.com

Tiemin Sun, Department of Pharmaceutical Chemistry, School of Pharmaceutical Engineering, Shenyang Pharmaceutical University, No. 103, Wenhua Road Shenhe District 110016, Shenyang, Liaoning Province, P.R. China.
Email: suntiemin8150@126.com

Funding information
National Natural Science Foundation of China, Grant/Award Number: 81273476

Abstract

Three kinds of sulfated β-cyclodextrin (S-β-CD), including a single isomer, heptakis-6-sulfato-β-cyclodextrin (HS-β-CD), degree of substitution (DS) of 7, which was synthesized in our laboratory and another two commercialized randomly substituted mixtures, a sulfated β-cyclodextrin with DS of 7 to 11, as well as a highly sulfated-β-cyclodextrin with DS of 12 to 15, were used for the enantioresolution of 12 drugs (the β-blockers, phenethylamines, and anticholinergic agents) in capillary electrophoresis. The enantioseparation under varying concentrations of S-β-CD and background electrolyte pH were systematically investigated and compared. Based on the experimental results, the effect of the nature of S-β-CD and analyte structure on the enantioseparation is discussed.

KEYWORDS

analyte structure, chiral selector, degree of substitution, enantioseparation, sulfated β-cyclodextrin

1 | INTRODUCTION

In recent years, capillary electrophoresis (CE) has been shown to be a powerful and versatile technique for chiral separation due to its advantages of high efficiency and resolution, short analysis time, as well as low cost and small amount of waste solution.[1,2] The enantioseparation in CE requires adding chiral selectors into the background electrolyte (BGE). Among the great number of chiral selectors used in CE, cyclodextrins (CDs) are the most employed because of their excellent enantiorecognition abilities, good water solubility, and UV transparency.[3]

Native β-CD has many applications in chiral separation. However, the low solubility of β-CD has limited its usefulness.[4] Chemical modification of native CDs leads to a significant improvement in their physicochemical properties and chiral recognition abilities. The first class of derivatized CDs are neutral CDs, such as 2,3,6-tri-O-methyl-β-CD (TMCD), 2,6-O-methyl-β-CD (DMCD), and hydroxypropyl-β-CD (HPCD).[5] One limitation is that neutral analytes are not enantioresolvable, simply in the presence of neutral CDs.[6] More effective separation strategies in CE has been achieved with charged CDs as chiral selectors, especially with the anionic CDs. Sulfated β-cyclodextrin (S-β-CD) is one of the most widely used anionic CDs, in that the sulfonic groups provide negative charges over the entire pH range and can thereby lead to ionic interaction in addition to the hydrophobic inclusion.[4] As reported, S-β-CD had the

ability to separate neutral, zwitterionic, basic, and even anionic analytes.[7]

S-β-CD can be obtained as the single isomer of a randomly sulfated mixture. For the single isomer, the degree of substitution (DS) and distribution of the substituent groups are clear. For example, heptakis-6-sulfato-β-cyclodextrin (HS-β-CD), with all the sulfate esters at the C-6 primary hydroxyl of β-CD, has been used for the separation of the enantiomers of numerous analytes.[8,9] However, most of the commercially available S-β-CD is a mixture of randomly sulfated species. For randomly substituted CDs in general, only the average value or the range of DS is known with certainty. This randomly sulfated β-CD has been successfully used for the separation of enantiomers of various kinds of drugs.[6,10,11]

In this study, we synthesized a single isomer β-CD derivative, heptakis-6-sulfato-β-cyclodextrin(HS-β-CD). HS-β-CD, together with two randomly substituted S-β-CDs (DS 7–11 and 12–15),were used for the enantioseparation of 12 drugs (the β-blockers, phenethylamines, and anticholinergic agents) in CE. The separation with different S-β-CDs as chiral selectors were compared under varying concentrations of chiral selectors and BGE pH. Further, the effect of DS of S-β-CD and substituent groups of analytes on the enantioseparation is discussed.

2 | MATERIALS AND METHODS

2.1 | Chemicals and reagents

Sotalol (SOT), bevantolol (BEV), metoprolol (MET), propranolol (PRO), terbutaline (TER), salbutamol (SAL), clenbuterol (CLE), clorprenaline (CLO), atropine (ATR), atropine methyl bromide (AMB), homatropine (HOM), and homatropine methyl bromide (HMB) were purchased from Chinese Food and Drug Inspection Institute (Beijing, China) as racemic mixtures. Their structures are provided in Figure 1 . Randomly substituted S-β-CDs (DS 7–11) and S-β-CD (DS 12–15) were from Sigma-Aldrich (St. Louis, MO). Sodium hydroxide, sodium dihydrogen phosphate and phosphoric acid were of analytical grade and obtained from Tianjin Bodi Chemical Plant (Tianjin, China). Methanol of high-performance liquid chromatography (HPLC) grade was from Tianjin Concord Technology (Tianjin, China). Double-distilled water was used throughout the study.

2.2 | Apparatus

The enantioseparation experiments were performed with a Beckman P/ACE MDQ Capillary Electrophoresis System (Beckman, Fullerton, CA), equipped with a diode-array detection. 32 Karat 8.0 Software (Beckman) was used for instrument control, data collection, and data analysis. ^{13}C–NMR spectra were recorded on Bruker (Billerica, MA) 400 MHz spectrometer with the samples diluted in deuterated water (D_2O). An uncoated, fused silica capillary (Ruifeng, Yongnian, Hebei, China), 50 cm × 50 μm (effective length 40 cm) was used for separation.

2.3 | Methods

The back ground buffer solutions were prepared as follows. First, sodium phosphate was dissolved in water and chiral

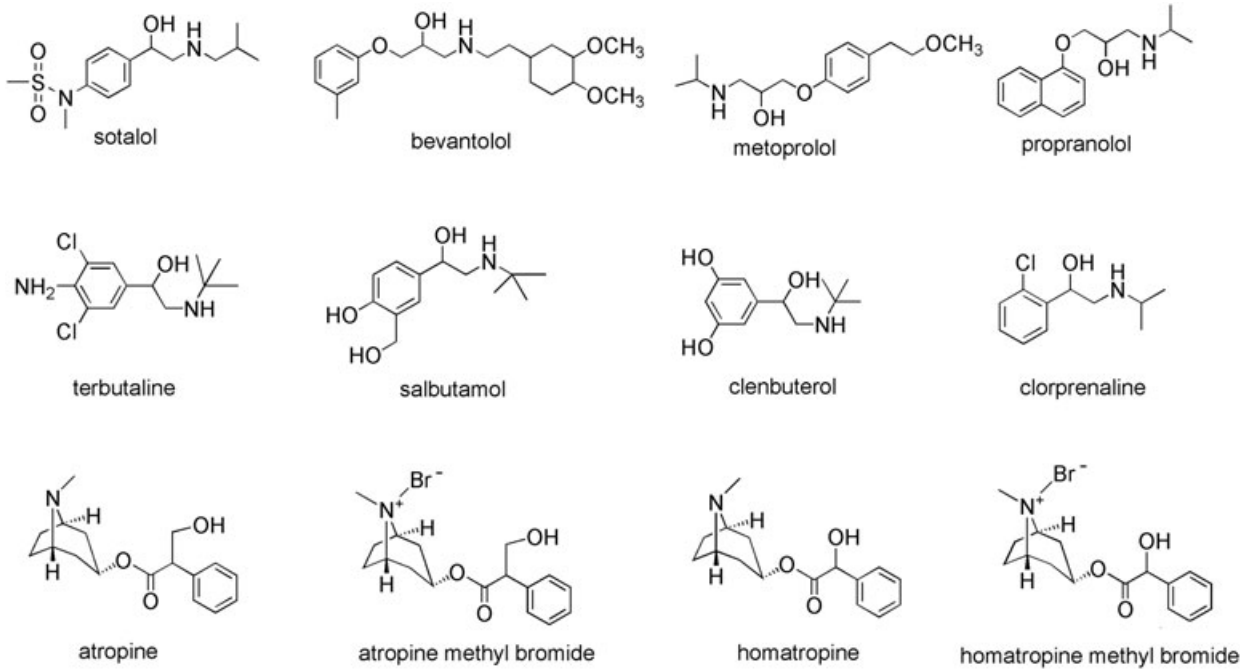

FIGURE 1 Chemical structures of 12 analytes

selectors were added, then adjusted to the desired pH with 1.0 M phosphoric acid or 1.0 M sodium hydroxide solution to make the BGE. Each sample was dissolved in water or water–methanol (90:10,*v*/v) at a concentration of 0.1 mg/ml. All solutions were passed through the 0.45 μm syringe filter before injection. When a new capillary was installed, it was rinsed with 1.0 M sodium hydroxide solution for 30 min and water for 10 min at 20 psi. Each day the capillary was rinsed with 0.1 M sodium hydroxide solution for 10 min, then water for 10 min and conditioned with BGE for 10 min. Between consecutive runs, the capillary was rinsed with BGE for 2 min. Injections were made hydrodynamically at 0.5 psi for 5 sec. Separations were performed at voltage of ±20 kV and all samples were detected at 210 nm. The capillary was thermostated at 25 °C during runs.

2.4 | Calculations

The resolution (Rs) was calculated using the following equation:

$$Rs = 2(t_2 - t_1)/(W_1 + W_2)$$

where t_1 and t_2 are the migration times and W_1 and W_2 are the baseline peak widths of the two enantiomers, respectively.

2.5 | Synthesis of HS-β-CD

The synthetic route of HS-β-CD was designed as follows, which was first reported in the previous literature.[8] In the first step, heptakis-[6-(tert-butyldimethyl)silyl] was synthesized to protect C-6 hydroxy, then C-2, C-3 hydroxy was acetylated by reaction with acetic anhydride. To remove the protecting group at C-6, the second intermediate was reacted with boron trifluoeride diethyl ether. The third intermediate was purified,[12] then sulfated with SO_3·pyridine. To deacetylate, the last intermediate was dissolved in water and the solution pH was adjusted to 12.0 with sodium hydroxide solution. After precipitation in ethanol, the solid was collected by filtration, then washed with ethanol and dried, yielding the end product. The characterization of the final product is shown in Figure 2. ^{13}C–NMR spectra were compared between β-CD and HS-β-CD. It was observed that the chemical shift of C-6 in β-CD was 60.2 ppm; however, in HS-β-CD the chemical shift was 69.8 ppm due to the hydroxyl groups being converted to sulfate ethers.[13]

3 | RESULTS AND DISCUSSION

3.1 | Effect of buffer type and buffer concentration on the enantioseparation

It is known that the chemical composition and concentration of the buffer could affect the separation selectivity, baseline stability, and peak shape. Therefore, the influence of buffer composition and concentration on the enantioseparation was first investigated. Three different buffer solutions, namely, phosphate, Tris–HCl, and acetate solutions, were tested as BGE for the separation of the tested analytes, using 20 g/L S-β-CD (DS 7–11) as chiral selector at pH 3.0. Among the tested buffer compositions, phosphate buffer was found to give the highest resolution values in most cases, and thus it was selected as BGE. In order to determine the optimum concentration, the effect of buffer concentration upon migration times and resolution was sequentially investigated in the range of 10–50 mM. It was found that with increasing the buffer concentration in the range of 10–30 mM, the enantiomeric resolutions were improved. At higher concentration only a small influence on the separation could be observed. Nevertheless, when the buffer concentration increased, the migration times slightly increased due to a decrease in the EOF. Hence, a 30 mM phosphate buffer was selected for the following optimization.

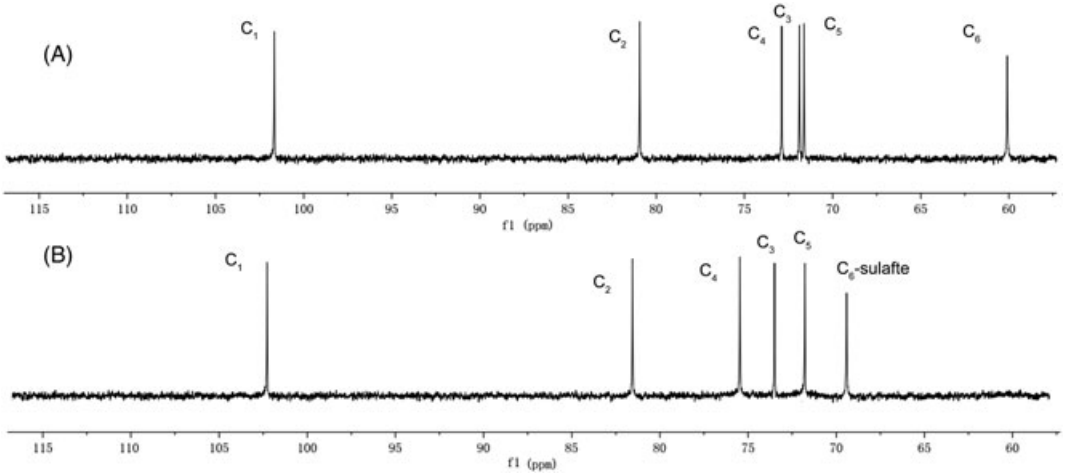

FIGURE 2 ^{13}C–NMR spectra of (**a**) β-CD and (**b**) HS-β-CD (400 MHZ, Solvent: D_2O)

3.2 | Effect of the concentration of S-β-CD on the enantioseparation

CD concentration was an important parameter to be optimized during the separation, and the effect of S-β-CD concentration on enantioresolution was investigated in the range of 5–30 g/L. As two randomly substituted S-β-CDs were mixtures, the concentration of three S-β-CDs was given by mass concentration (g/L) instead of molar concentration (mol/L). The detection of all analytes was at the anodic side, with other conditions of 30 mM phosphate buffer, at pH 3.0.[14]

It was found that both the migration times and resolutions of 12 drug enantiomers tended to decrease with an increase of three S-β-CD concentrations. In general, a high S-β-CD concentration may enhance the interaction between analytes and CDs selectors, which resulted in a prolonged migration time and higher resolution. On the other hand, with an increase of S-β-CD concentration, the elevated viscosity and ionic strength of buffer might cause lower electroosmotic mobility (μ_{eof}) and higher apparent mobility (μ_{app}), leading to decreased resolution and reduced migration times. And obviously, the results indicated that the latter reason might play a more dominant role in the enantioseparation. Figure 3 shows the variation in resolutions of 12 chiral drugs, along with the varied S-β-CD concentrations. For most of the drugs, resolution values decreased when CD concentration changed from 5 to 30 mg/L. However, there were some notable exceptions. Some drugs might not obtain accurate resolutions at lower CD concentrations due to long migration times of their enantiomers. For example, no resolution values were given at the variation curves of MET, CLO, and HMB at HS-β-CD concentration of 5 g/L (Figure 3a). Using two different S-β-CDs with various degrees of substitution (DS 7–11 and 12–15) as chiral selectors, the missing resolution values for some analytes (Figure 3b,c) could also be ascribed to their prolonged migration times. Thus, 20 g/L of HS-β-CD, S-β-CD (DS 7–11) and S-β-CD (DS 12–15) was applied to the separation as a compromise between resolution and migration time.

3.3 | Effect of the BGE pH on the enantioseparation

The pH of the buffer is another important parameter affecting chiral resolution.[15,16] In this study, the enantioseparation of 12 chiral analytes was investigated over the pH range of 2.5–8.0 at 20 g/L of chiral selector concentration. The separation results with HS-β-CD, S-β-CD (DS 7–11), and

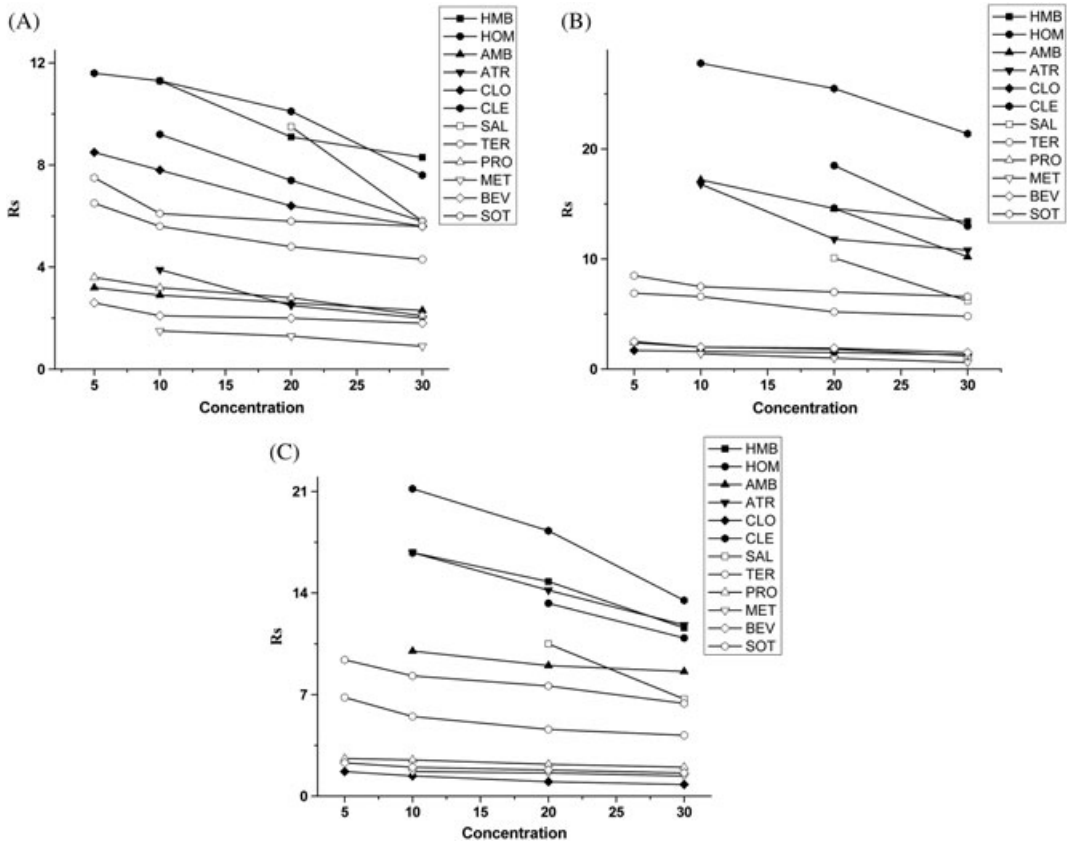

FIGURE 3 Resolutions of 12 analyte enantiomers, containing (**a**) HS-β-CD, (**b**) S-β-CD (DS 7–11), and (**c**) S-β-CD (DS 12–15) from 5 to 30 g/L. Buffer condition: 30 mM phosphate, pH 3.0; Voltage: −20 kV

S-β-CD (DS 12–15) as chiral selector at different BGE pH are shown in Table 1.

When BGE pH varied from 2.5–4.0, the analytes could only be detected at the anode because electrophoretic mobility (μ_{ep}, negative value) was larger than electroosmotic mobility (μ_{eof}, positive value). With a pH increase, both the migration times and resolution of the enantiomers tended to increase, mainly due to increasing EOF. However, SAL was a special case of 12 drugs. Its two enantiomers had no elution within 40 min using the two randomly substituted S-β-CDs at pH 4.0. Interestingly, similar phenomena were not scarce at pH 5.0. For example, no peak was observed for SOT, MET, PRO, SAL, and CLE with HS-β-CD as a chiral selector. In the case of two randomly sulfated CDs, most of the drugs (except TER and CLO) couldn't be eluted within 40 min, indicating that stronger electrostatic interaction might be formed between analytes and two randomly substituted S-β-CDs. At higher pH, the increased EOF might cause a change of apparent mobility (μ_{app}) toward the cathode (positive value); therefore, over the pH range of 6.0–8.0, separation was carried out at normal polarity mode. It was shown that migration times of analytes were shortened with increasing buffer pH. Also, a decreasing tendency of resolution was observed, which might be due to the decreased opportunity for the interaction of analytes with chiral selectors. Based on the above results, we concluded that an increase in pH has a favorable effect on enantiomeric resolution of the analytes, but only if the inclusion complex directed toward anode at low pH range (range of 2.5–4.0). Thus, the best results with respect to resolution values were all obtained at reversed polarity mode.

Simultaneously, we investigated the effect of buffer pH on mobility parameters of analytes to provide a better understanding of the host–guest interaction. Take CLO, for example: Figure 4 shows the variation of μ_{eof}, μ_{ep}, and μ_{app} with 20 g/L S-β-CD (DS 7–11), increasing pH from 2.5–8.0. CLO, as its pKa ~13.6, it could be completely positively charged over the pH range of 2.5–8.0, and changes in BGE pH caused a minor effect on its μ_{ep}. In cases of three negatively charged S-β-CDs, as salts of strong acid, their charge-state also showed pH independence. Thus, the electrostatic interaction between the chiral selector and analyte would remain constant along with the increase of pH. This was also applicable for the other 11 analytes, according to their chemical properties of quaternary ammonium salts (AMB and HMB) or basic analytes (pKa ≥9.3 for the other drugs).

TABLE 1 Separation results of 12 analytes with three S-β-CD at pH 2.5–8.0

		β-blockers				Phenethylamines				Anticholinergic agents			
		SOT	BEV	MET	PRO	TER	SAL	CLE	CLO	ATP	AMB	HOM	HMB
Analyte		$t_{1/min}$ Rs	$t_{1/min}$ Rs	$t_{1/min}$ Rs	$t_{1/min}$ Rs	$t_{1/min}$ Rs	$t_{1/min}$ Rs	$t_{1/min}$ Rs	$t_{1/min}$ Rs	$t_{1/min}$ Rs	$t_{1/min}$ Rs	$t_{1/min}$ Rs	$t_{1/min}$ Rs
HS-β-CD	2.5[a]	11.9 5.1	8.6 1.7	14.8 1.3	12.5 1.9	4.5 4.0	15.3 6.7	8.5 9.6	7.5 5.6	8.3 2.0	6.5 2.3	9.8 6.2	8.7 7.9
	3.0[a]	13.3 6.6	10.0 2.0	17.2 1.4	14.3 2.3	4.8 5.3	27.4 8.5	9.6 10.1	8.7 6.4	10.5 2.5	7.8 2.6	11.3 7.4	9.7 9.1
	4.0[a]	14.3 7.7	15.6 2.3	21.6 1.6	19.6 2.5	6.5 6.3	36.8 10.5	15.8 12.0	12.3 8.6	14.6 3.7	9.9 2.7	16.4 8.5	14.3 11.2
	5.0[a]	nd	18.7 2.5	nd	nd	9.3 6.6	nd	nd	20.6 9.8	24.8 4.2	15.2 3.0	22.3 9.7	18.9 11.8
	6.0	11.3 5.9	9.7 1.9	16.7 1.4	5.2 1.9	15.2 2.8	8.4 3.1	12.4 8.6	15.3 8.7	10.1 3.4	16.5 2.8	15.5 7.9	12.5 9.2
	7.0	8.5 4.6	7.8 1.4	12.1 1.3	12.4 1.8	10.9 2.3	6.2 2.5	9.8 6.5	10.6 6.5	7.8 3.0	12.1 2.2	11.9 5.4	8.9 7.8
	8.0	5.6 3.9	6.8 1.2	10.2 1.1	9.8 1.5	7.5 1.8	4.8 2.3	7.6 5.3	7.4 4.3	5.6 2.7	9.8 1.8	9.8 3.3	7.1 5.7
S-β-CD **pH**	2.5[a]	13.2 6.1	13.5 1.8	14.5 1.3	11.3 2.0	5.8 4.0	14.4 8.1	15.6 19.8	7.0 1.4	10.4 10.2	12.2 10.8	19.8 14.2	14.7 12.8
(DS 7–11)	3.0[a]	16.1 7.0	15.3 2.1	18.6 1.5	12.9 2.2	7.0 5.2	20.8 10.1	9.6 21.4	8.8 1.5	15.8 11.8	14.8 14.6	18.9 16.1	18.4 14.6
	4.0[a]	25.9 7.5	18.6 2.4	25.4 1.6	14.6 2.4	8.3 6.2	nd	35.8 28.6	10.4 1.6	28.6 12.5	20.8 18.9	35.6 18.5	26.7 18.2
	5.0[a]	nd	nd	nd	nd	13.1 5.6	nd	nd	20.6 1.7	nd	nd	nd	nd
	6.0	14.9 5.2	16.5 2.3	14.5 1.4	13.5 2.3	10.8 3.4	10.5 3.4	9.8 15.3	12.4 1.5	15.3 10.6	15.5 14.0	22.7 12.3	21.7 13.5
	7.0	10.8 4.3	12.4 1.8	12.4 1.3	10.9 2.2	8.8 2.4	8.3 3.0	7.3 10.2	10.5 1.4	10.4 6.3	10.5 9.4	16.6 9.8	16.5 9.8
	8.0	7.9 4.0	9.8 1.7	10.1 0.9	8.8 1.9	6.7 2.0	6.3 2.4	6.5 8.2	8.4 1.3	7.3 4.1	7.6 6.3	12.4 8.6	10.5 7.9
S-β-CD	2.5[a]	14.5 6.6	13.0 1.8	16.7 1.4	12.8 2.0	5.1 4.3	25.4 9.0	14.5 16.2	6.4 1.3	13.6 12.7	8.7 8.0	18.5 10.1	14.2 12.3
(DS 12–15)	3.0[a]	18.4 7.6	16.7 2.2	21.7 1.6	14.9 2.3	6.5 4.6	32.1 10.5	17.9 18.3	8.6 1.5	15.6 14.1	10.9 9.3	22.9 13.3	16.4 15.8
	4.0[a]	27.3 7.9	17.8 2.3	25.6 1.6	18.5 2.4	7.2 6.2	nd	28.7 20.5	10.8 1.6	30.6 15.8	15.6 10.2	36.8 15.9	20.5 18.7
	5.0[a]	nd	nd	nd	nd	10.7 5.8	nd	nd	16.4 1.7	nd	nd	nd	nd
	6.0	15.4 6.0	20.1 2.0	18.9 1.4	16.7 2.1	12.6 3.5	8.7 2.9	11.4 13.6	10.2 1.5	15.8 9.3	16.3 9.8	16.4 9.8	15.6 13.3
	7.0	12.7 5.1	14.6 1.5	15.2 1.3	13.3 2.0	9.7 3.0	6.4 2.3	9.2 9.7	8.6 1.4	12.1 7.2	12.3 6.4	12.2 7.4	12.4 10.8
	8.0	8.5 3.6	10.8 1.3	12.0 1.2	10.2 1.8	7.9 2.3	5.9 2.2	7.8 7.0	6.8 1.3	6.9 4.8	9.9 4.5	10.0 5.5	9.9 8.2

BGE condition: 20 g/L chiral selectors, 30 mM phosphate; voltage: ± 20 kV.

[a]Detected at reverse polarity.

Nd, not detected in 40 min.

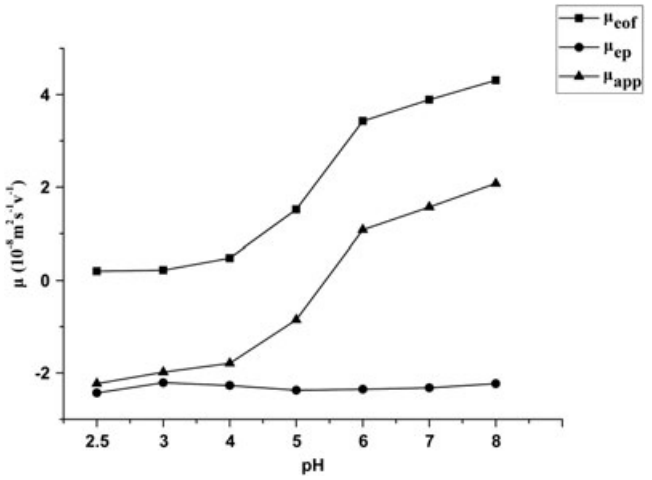

FIGURE 4 Variation of μ_{eof}, μ_{ep}, and μ_{app} of CLO with increasing pH from 2.5 to 8.0. BGE conditions: 20 g/L S-β-CD (DS 7–11), 30 mM phosphate; voltage: ±20 kV (pH 2.5–5.0 detected at reverse polarity; pH 6.0–8.0 detected at normal polarity)

Figure 5 shows the typical electropherograms of the 12 analytes at BGE pH 3.0, with 20 g/L S-β-CD (DS 7–11) as chiral selector.

3.4 | Effect of different S-β-CDs on the enantioseparation

Conclusively, three S-β-CDs with various degrees of substitution showed enantioselectivity toward the tested analytes. However, for the enantioseparation of specific analytes, some difference in the separation efficiency was also observed with S-β-CD of different DS. Take CLO, for example: a satisfactory resolution value (Rs = 6.4) was obtained with 20 g/L HS-β-CD as chiral selector. In contrast, two randomly substituted S-β-CDs seemed to be the least effective chiral selectors under the same condition, since only baseline separation was observed for CLO enantiomers (Figure 6a). However, this was not true for the enantioseparation of anticholinergic agents, which got higher resolution values with two randomly substituted S-β-CDs (Figure 6b). These results seemed to confirm the fact that the enantioselectivity was affected by the DS of S-β-CD, and the optimum selector depended on the structures of the tested analytes were observed.[17] Figure 6 gives the electropherograms of (a) CLO and (b) ATP with three kinds of S-β-CDs at 20 g/L as chiral selectors.

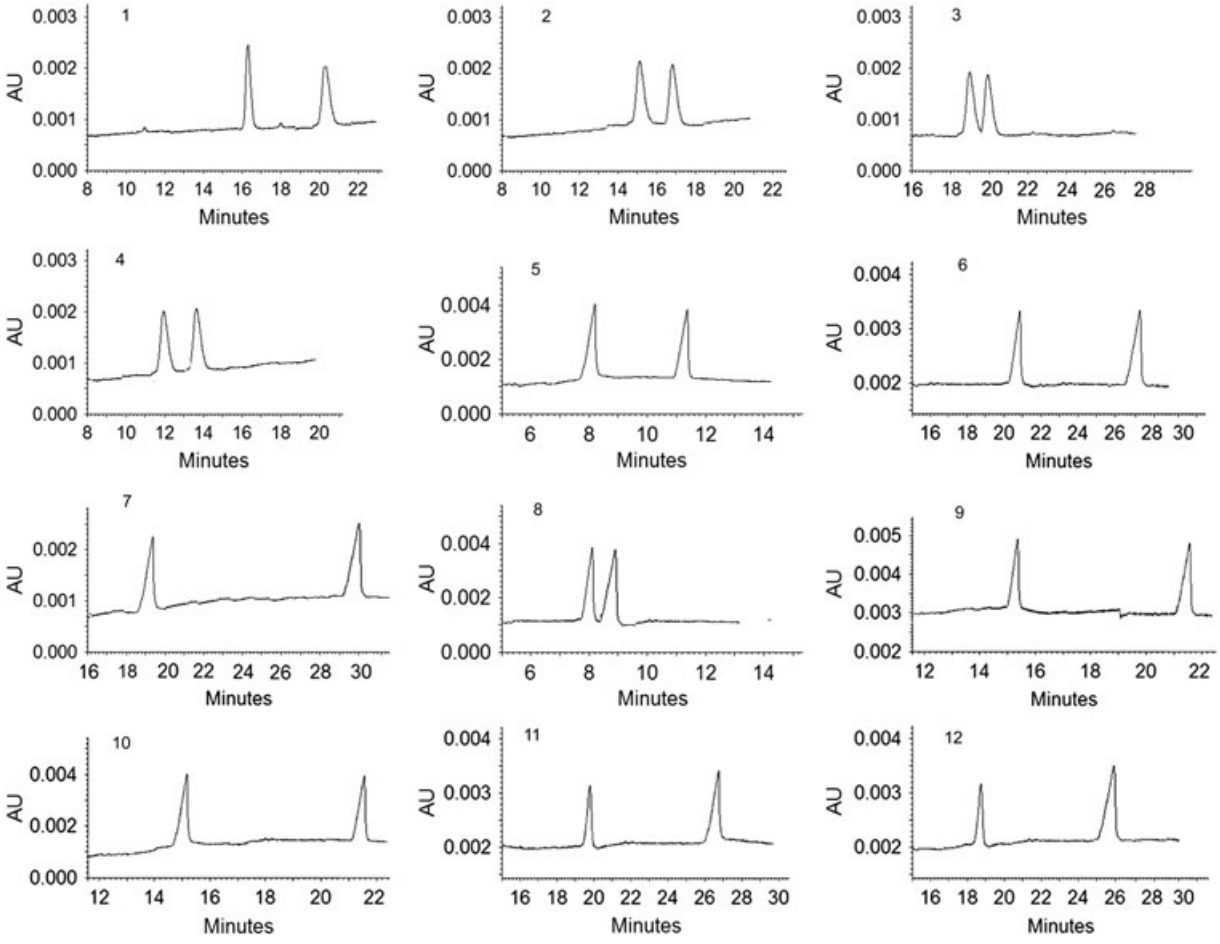

FIGURE 5 Typical electropherograms of the 12 analytes. BGE conditions: 20 g/L S-β-CD (DS 7–11), 30 mM phosphate, pH 3.0; voltage: −20 kV. SOT (2) BEV (3) MET (4) PRO (5) TER (6) SAL (7) CLE (8) CLO (9) ATP (10) AMB (11) HOM (12) HMB

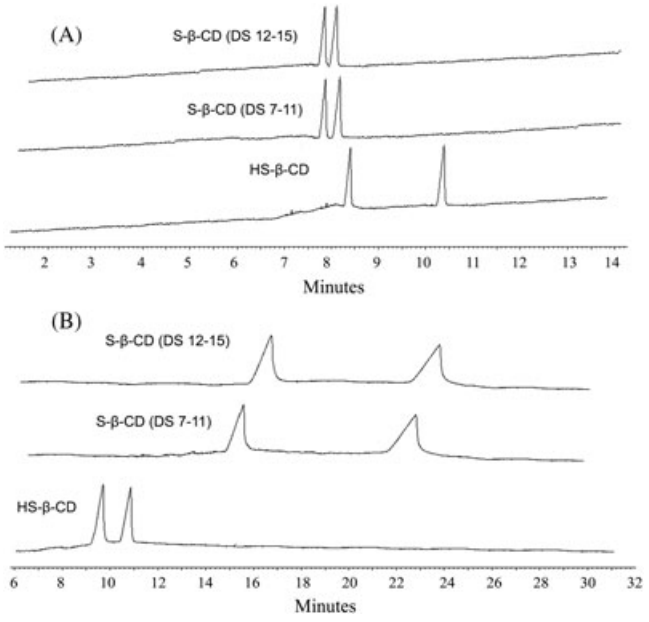

FIGURE 6 Enantioseparation electrophoretograms of (**a**) CLO and (**b**) ATP with three S-β-CDs at 20 g/L as chiral selectors. BGE conditions: 30 mM phosphate, pH 3.0; Voltage: −20 kV

3.5 | Structural influence of analytes on the enantioseparation

Under the optimized separation conditions, the 12 analytes could be completely enantioresolved, and resolution values higher than 5.0 were obtained for most analytes. These separation results were closely related to the structures of analytes. The driving force of S-β-CD responsible for host–guest interactions were reported to be electrostatic interaction, hydrophobic inclusion, and hydrogen bonding.[18] Besides amine or ammonium, which can provide an ionic interaction, analytes in our research also had the aromatic ring that should fit into the hydrophobic cavity of S-β-CD. Therefore, it was reasonable to expect that hydrophobic inclusion may play a role in the chiral recognition. In addition, as all the tested analytes had some functional groups around chiral carbon such as hydroxyl, amide, and carbonyl, we could deduce that hydrogen bonding between these substituents of the analytes and the hydroxyl groups on the CD-rim was also responsible for the chiral recognition.

It should be noted that only three analytes, BEV, MET, and PRO, obtained worse resolution values as compared to the other nine analytes. These three analytes presented similar structural characteristics that might be unfavorable for chiral discrimination, i.e., greater distance between aromatic rings and chiral carbon. Conversely, the elegant separations for the nine analytes might be related to the presence of an aromatic ring in the vicinity of the chiral center.[19]

4 | CONCLUSION

In this work, a single isomer HS-β-CD and two randomly substituted S-β-CDs (DS 7–11 and 12–15) were used as chiral selectors to separate the enantiomers of 12 drugs in capillary electrophoresis. Two important parameters affected the enantioseparation: the concentration of chiral selectors and BGE pH were systematically studied. Comparative results indicated that the single isomer HS-β-CD and two randomly substituted S-β-CDs differed in the separation of CLO and four anticholinergic agents. This can be explained by the different degree of substitution of S-β-CD. In addition, the structural influence of chiral drugs on the enantioseparation was also speculated. Especially, an aromatic ring as a substituent group directly connected to chiral carbon was considered to be an important factor in enantiomeric recognition.

ACKNOWLEDGEMENT

This work was supported by the National Natural Science Foundation of China (no. 81273476).

REFERENCES

1. Maier NM, Franco P, Lindner W. Separation of enantiomers: needs, challenges, perspectives. *J Chromatogr A*. 2001;12(906):3-33.
2. Wang Y, Zhang S, Breitbach ZS, Petersen H, Ellegaard P, Armstrong DW. Enantioseparation of citalopram analogues with sulfated β-cyclodextrin by capillary electrophoresis. *Electrophoresis*. 2016;37:841-848.
3. Asensi-Bernardi L, Escuder-Gilabert L, Martín-Biosca Y, Medina-Hernández MJ, Sagrado S. Modeling the chiral resolution ability of highly sulfated β-cyclodextrin for basic compounds in electrokinetic chromatography. *J Chromatogr A*. 2013;1308:152-160.
4. Le Potiera I, Tamisier-Karolaka SL, Morinb P, Megelc F, Tavernaa M. Comparison of native, alkylated and charged cyclodextrins for the chiral separation of labetalol stereoisomers by capillary electrophoresis. *J Chromatogr A*. 1998;829:341-349.
5. Matthijs N, Hemelryck SV, Maftouh M, Massart DL, Heyden YV. Electrophoretic separation strategy for chiral pharmaceuticals using highly-sulfated and neutral cyclodextrins based dual selector systems. *Anal Chim Acta*. 2004;525:247-263.
6. Stalcup AM, Gahm KH. Application of sulfated cyclodextrins to chiral separations by capillary zone electrophoresis. *Anal Chem*. 1996;68:1360-1368.
7. Rd E, Vigh G. Comparison of charge state distribution in commercially available sulfated cyclodextrins used as chiral resolving agents in capillary electrophoresis. *J Chromatogr A*. 2012;1226:24-30.
8. Vincent JB, Kirby DM, Nguyen TV, Vigh G. A family of single-isomer chiral resolving agents for capillary electrophoresis. 2.Hepta-6-sulfato-β-cyclodextrin. *Anal Chem*. 1997;69:4419-4428.
9. Süß F, Griend SVD, Scriba GKE. Migration order of dipeptide and tripeptide enantiomers in the presence of single isomer and

randomly sulfated cyclodextrins as a function of pH. *Electrophoresis*. 2003;24:1069-1076.

10. Guo L, Lin SJ, Yang YF, Qi L, Wang MX, Chen Y. Fast enantioseparation of arylglycine amides by capillary electrophoresis with highly sulfated-β-cyclodextrin as a chiral selector. *J Chromatogr A*. 2003;998:221-228.
11. Wang J, Yuan Q, Evans DG, Yang L, Zheng G, Sun W. Enantioselective separation of phenylglycidates by capillary electrophoresis employing sulfated β-cyclodextrin as chiral selector. *J Chromatogr B*. 2007;850:560-563.
12. Takeo K, Mitoh H, Uemura K. Selective chemical modification of cyclomalto-oligosaccharides via tert -butyldimethylsilylation. *Carbohydr Res*. 1989;187:203-221.
13. Chen FT, Shen G, Evangelista RA. Characterization of highly sulfated cyclodextrins. *J Chromatogr A*. 2001;924:523-532.
14. Rudaz S, Calleri E, Geiser L, Cherkaoui S, Prat J, Veuthey JL. Infinite enantiomeric resolution of basic compounds using highly sulfated cyclodextrin as chiral selector in capillary electrophoresis. *Electrophoresis*. 2003;24:2633-2641.
15. Yu J, Jiang Z, Sun T, et al. Enantiomeric separation of meptazinol and its three intermediate enantiomers by capillary electrophoresis: quantitative analysis of meptazinol in pharmaceutical formulations. *Biomed Chromatogr*. 2014;28:135-141.
16. Zuo L, Zhao Y, Ji F, et al. Determination of the enantiomeric and diastereomeric impurities of RS-glycopyrrolate by capillary electrophoresis using sulfated-β-cyclodextrin as chiral selectors. *Electrophoresis*. 2014;35:3339-3344.
17. Tamisier-Karolak SL, Stenger M, Bommart A. Enantioseparation of β-blockers with two chiral centers by capillary electrophoresis using sulfated β-cyclodextrins. *Electrophoresis*. 1999;20:2656-2663.
18. Liu Y, Deng M, Yu J, Jiang Z, Guo X. Capillary electrophoretic enantioseparation of basic drugs using a new single-isomer cyclodextrin derivative and theoretical study of the chiral recognition mechanism. *J Sep Sci*. 2016;39:1766-1775.
19. Hellriegel C, Händel H, Wedig M, et al. Study on the chiral recognition of the enantiomers of ephedrine derivatives with neutral and sulfated heptakis(2,3-O-diacetyl)-beta-cyclodextrins using capillary electrophoresis, UV, nuclear magnetic resonance spectroscopy and mass spectrometry. *J Chromatogr A*. 2001;914:315-324.

How to cite this article: Wang Z, Zhang Q, Luo L, Sun T, Guo X. Comparison of three S-β-CDs with different degrees of substitution for the chiral separation of 12 drugs in capillary electrophoresis. *Chirality*. 2017;1–8. https://doi.org/10.1002/chir.22731