

ChiralTek Chiral Column Manual and Applications in HPLC and UPLC

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Two Series of New Chiral Columns from ChiralTek for UPLC and HPLC

ChiralTek provides two new series of chemically-bonded silica particles-packed high-efficiency chiral columns for both traditional HPLC and modern UPLC.

The first series is ChiralCE columns. The ChiralCE columns are a new type of chemically-modified cellulose-bonded silica particles-packed chiral columns. A series of chemically-modified celluloses were chemically-bonded in the ChiralCE columns, both standard and non-standard mobile phases (e.g., chloroform and dichloromethane etc.) can be used to achieve optimized separation.

High column efficiency can be easily achieved on the ChiralCE columns since only one-step reaction was used to anchor the chemically-modified cellulose onto the silica support. At present, most of commercial cellulose-based columns are cellulose-coated columns and there are only few cellulose-bonded columns on the market. Compared to the commercial cellulose-coated columns, the bonded ChiralCE columns can provide better reducibility and stability. Compared to other commercial cellulose-bonded columns, ChiralCE columns have higher content of anchored-celluloses and could provide higher sample capacity and better separation selectivities.

ChiralTek provides two types ChiralCE columns ChiralCE-1 and ChiralCE-2. The I.D. of the column is 2mm only. ChiralCE columns have five different lengths (50mm, 100mm, 150mm, 200mm, and 250mm). The photo of the ChiralCE-1 columns is shown below.



Figure (A). Typical photo of the ChiralCE-1 columns with different length

The second series is ChiralCD columns. They are new types of chemically-substituted

cyclodextrin-bonded silica particles-packed chiral columns. The ChiralCD particles were synthesized by bonding series of functional groups-substituted α -, β -, or γ -cyclodextrins onto surface of high-quality porous silica (2 μ m or 3 μ m) gel by linking the spacer arms at the wider torus rims of the cyclodextrins.

The ChiralCD columns are the first commercial available chiral columns by anchoring cyclodextrins through the less reactive secondary hydroxyl at the wider torus rim of the cyclodextrins, the spacer arms can provide special unique steric interactions with solutes when entering the cyclodextrin cavities. Therefore, excellent separation selectivity can be achieved on the ChiralCD columns for separation a wide range of both chiral and non-chiral compounds under multiple modes chromatographic conditions.

ChiralTek provides four types of ChiralCD columns: ChiralCD-1, ChiralCD-2, ChiralCD-3 and ChiralCD-4. The I.D. of the column is 2mm only. There are five different column lengths (50mm, 100mm, 150mm, 200mm, and 250mm). The following figure shows the photo of ChiralCD-1 columns.



Figure (B). Typical photo of the ChiralCD-1 columns with different length

At the moment, ChiralCD and ChiralCE columns are analytical scales. ChiralTek can also provide the semi-preparative and preparative columns upon receiving confirmed request from customer.

ChiralTek also provide free screening tests for chiral separation if customer can provide the test compounds with clear chemical structure.

Order Information

<i>Part number</i>	<i>Type</i>	<i>Column Dimension</i>	<i>Remarks</i>
812-CE1-01	ChiralCE-1	2μm, 50 x 2mm	Type 1 chemically-modified cellulose-bonded column
812-CE1-02	ChiralCE-1	2μm, 100 x 2mm	Type 1 chemically-modified cellulose-bonded column
812-CE1-03	ChiralCE-1	2μm, 150 x 2mm	Type 1 chemically-modified cellulose-bonded column
812-CE1-04	ChiralCE-1	2μm, 200 x 2mm	Type 1 chemically-modified cellulose-bonded column
812-CE1-05	ChiralCE-1	2μm, 250 x 2mm	Type 1 chemically-modified cellulose-bonded column
813-CE1-01	ChiralCE-1	3μm, 50 x 2mm	Type 1 chemically-modified cellulose-bonded column
813-CE1-02	ChiralCE-1	3μm, 100 x 2mm	Type 1 chemically-modified cellulose-bonded column
813-CE1-03	ChiralCE-1	3μm, 150 x 2mm	Type 1 chemically-modified cellulose-bonded column
813-CE1-04	ChiralCE-1	3μm, 200 x 2mm	Type 1 chemically-modified cellulose-bonded column
813-CE1-05	ChiralCE-1	3μm, 250 x 2mm	Type 1 chemically-modified cellulose-bonded column
812-CE2-01	ChiralCE-2	2μm, 50 x 2mm	Type 2 chemically-modified cellulose-bonded column
812-CE2-02	ChiralCE-2	2μm, 100 x 2mm	Type 2 chemically-modified cellulose-bonded column
812-CE2-03	ChiralCE-2	2μm, 150 x 2mm	Type 2 chemically-modified cellulose-bonded column
812-CE2-04	ChiralCE-2	2μm, 200 x 2mm	Type 2 chemically-modified cellulose-bonded column
812-CE2-05	ChiralCE-2	2μm, 250 x 2mm	Type 2 chemically-modified cellulose-bonded column
813-CE2-01	ChiralCE-2	3μm, 50 x 2mm	Type 2 chemically-modified cellulose-bonded column
813-CE2-02	ChiralCE-2	3μm, 100 x 2mm	Type 2 chemically-modified cellulose-bonded column
813-CE2-03	ChiralCE-2	3μm, 150 x 2mm	Type 2 chemically-modified cellulose-bonded column
813-CE2-04	ChiralCE-2	3μm, 200 x 2mm	Type 2 chemically-modified cellulose-bonded column
813-CE2-05	ChiralCE-2	3μm, 250 x 2mm	Type 2 chemically-modified cellulose-bonded column
822-CD1-01	ChiralCD-1	2μm, 50 x 2mm	Type 1 substituted β-cyclodextrin-bonded column
822-CD1-02	ChiralCD-1	2μm, 100 x 2mm	Type 1 substituted β-cyclodextrin-bonded column
822-CD1-03	ChiralCD-1	2μm, 150 x 2mm	Type 1 substituted β-cyclodextrin-bonded column
822-CD1-04	ChiralCD-1	2μm, 200 x 2mm	Type 1 substituted β-cyclodextrin-bonded column
822-CD1-05	ChiralCD-1	2μm, 250 x 2mm	Type 1 substituted β-cyclodextrin-bonded column
823-CD1-01	ChiralCD-1	3μm, 50 x 2mm	Type 1 substituted β-cyclodextrin-bonded column
823-CD1-02	ChiralCD-1	3μm, 100 x 2mm	Type 1 substituted β-cyclodextrin-bonded column
823-CD1-03	ChiralCD-1	3μm, 150 x 2mm	Type 1 substituted β-cyclodextrin-bonded column
823-CD1-04	ChiralCD-1	3μm, 200 x 2mm	Type 1 substituted β-cyclodextrin-bonded column
823-CD1-05	ChiralCD-1	3μm, 250 x 2mm	Type 1 substituted β-cyclodextrin-bonded column

Order Information

<i>Part number</i>	<i>Type</i>	<i>Column Dimension</i>	<i>Remarks</i>
823-CD2-01	ChiralCD-2	3μm, 50 x 2mm	Type 2 substituted β-cyclodextrin-bonded column
823-CD2-02	ChiralCD-2	3μm, 100 x 2mm	Type 2 substituted β-cyclodextrin-bonded column
823-CD2-03	ChiralCD-2	3μm, 150 x 2mm	Type 2 substituted β-cyclodextrin-bonded column
823-CD2-04	ChiralCD-2	3μm, 200 x 2mm	Type 2 substituted β-cyclodextrin-bonded column
823-CD2-05	ChiralCD-2	3μm, 250 x 2mm	Type 2 substituted β-cyclodextrin-bonded column
823-CD3-01	ChiralCD-3	3μm, 50 x 2mm	Type 3 substituted γ-cyclodextrin-bonded column
823-CD3-02	ChiralCD-3	3μm, 100 x 2mm	Type 3 substituted γ-cyclodextrin-bonded column
823-CD3-03	ChiralCD-3	3μm, 150 x 2mm	Type 3 substituted γ-cyclodextrin-bonded column
823-CD3-04	ChiralCD-3	3μm, 200 x 2mm	Type 3 substituted γ-cyclodextrin-bonded column
823-CD3-05	ChiralCD-3	3μm, 250 x 2mm	Type 3 substituted γ-cyclodextrin-bonded column
823-CD4-01	ChiralCD-4	3μm, 50 x 2mm	Type 4 substituted α-cyclodextrin-bonded column
823-CD4-02	ChiralCD-4	3μm, 100 x 2mm	Type 4 substituted α-cyclodextrin-bonded column
823-CD4-03	ChiralCD-4	3μm, 150 x 2mm	Type 4 substituted α-cyclodextrin-bonded column
823-CD4-04	ChiralCD-4	3μm, 200 x 2mm	Type 4 substituted α-cyclodextrin-bonded column
823-CD4-05	ChiralCD-4	3μm, 250 x 2mm	Type 4 substituted α-cyclodextrin-bonded column
833-SK1-03	Screening Kit-1	3μm, 150 x 2mm	Chiral Separation Screening Kit-1 containing 813-CE1-03 (ChiralCE-1), 823-CD1-03 (ChiralCD-1), and 823-CD4-03 (ChiralCD-4) three columns.
833-SK2-03	Screening Kit-2	3μm, 150 x 2mm	Chiral Separation Screening Kit-2 containing 813-CE1-03 (ChiralCE-1), 813-CE2-03 (ChiralCE-2), 823-CD1-03 (ChiralCD-1), 823-CD2-03 (ChiralCD-2), 823-CD3-03 (ChiralCD-3), and 823-CD4-03 (ChiralCD-4) six chiral columns.

ChiralCE Column Notes and Applications in HPLC and UPLC

Introduction

ChiralCE columns are a new type of chemically-modified cellulose-bonded silica particles-packed chiral columns. The ChiralCE particles were prepared through a specially-designed procedure by bonding the different functional groups-substituted celluloses onto surface of high-quality porous silica (2 μm or 3 μm) gel particles. Due to the multiple functional groups, e.g., halogen groups, hydroxyl groups, aromatic rings, and cellulose moiety etc., available in the bonded chiral stationary phases (CSP), the ChiralCE columns can be used under both normal phase and reversed-phase conditions. Since the chemically-modified cellulose were chemically-bonded in the ChiralCE columns, both standard and non-standard mobile phases (e.g., chloroform and dichloromethane etc.) can be used to achieve optimized separations.

Unique characteristics

High column efficiency can be easily achieved on the ChiralCE columns since only one-step reaction was used to anchor the chemically-modified cellulose onto the silica support. The chemically-modified celluloses in the Chiral CE columns were anchored onto the surface of porous silica support by chemical bonds as shown in Figure (C) below.

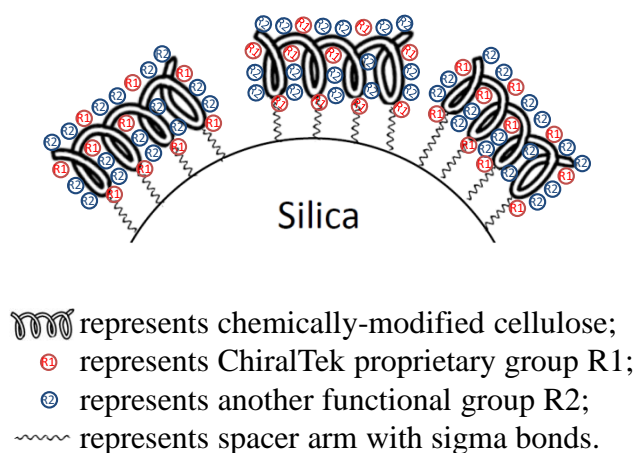


Figure (C). Schematic diagram of the bonded ChiralCE phase from ChiralTek.

Unlike other cellulose-coated columns (as shown in Figure (D)) in which all the hydroxyl groups of cellulose were substituted by a single type of functional group, the hydroxyl groups of the cellulose in ChiralCE columns were substituted by two different functional groups R1 and R2. R1 is a ChiralTek proprietary chiral group which can provide extra unique chiral recognition function. R2 is another common functional group.

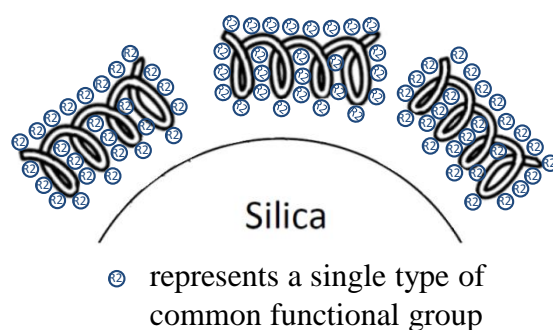


Figure (D). Schematic diagram of other supplier's cellulose-coated phase

Due to the cooperative functioning of the multiple functional groups available, the ChiralCE columns can be used for separation a wide range of both chiral and non-chiral compounds under multiple modes conditions. Compared to other supplier's cellulose-bonded columns (as shown in Figure (E)), ChiralCE columns have higher content of the celluloses.

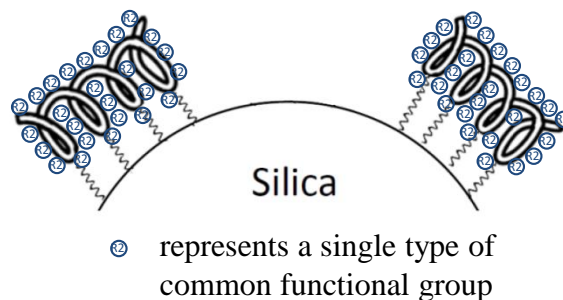


Figure (E). Schematic diagram of other supplier's cellulose-bonded phase.

ChiralCE Column Notes and Applications in HPLC and UPLC

Since the bonded ChiralCE phases from ChiralTek have higher density of the bonded celluloses and contain more types of functional groups, the ChiralCE columns can provide different and sometimes better chiral separation abilities than other supplier's cellulose-bonded columns. The chemical structure of the chiral selectors of ChiralCE phases are also different from other supplier's cellulose-based phases. The typical structure of the bonded cellulose selector unit in ChiralCE columns is showed in the following figure (F).

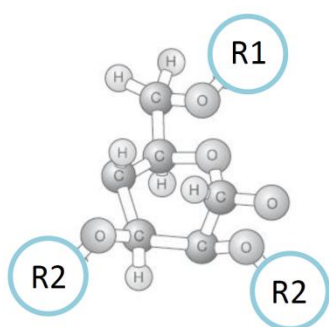


Figure (F). Typical structure of general chiral selector unit of the ChiralCE phases. R1 is a ChiralTek proprietary chiral group and R2 is common functional group.

In the ChiralCE-1 phase (Figure (G)), R2 is a Phenylcarbamate group. The ChiralCE-1 column can provide high column efficiency and excellent reducibility due to the contribution of the Phenylcarbamate group introduced into the bonded phase.

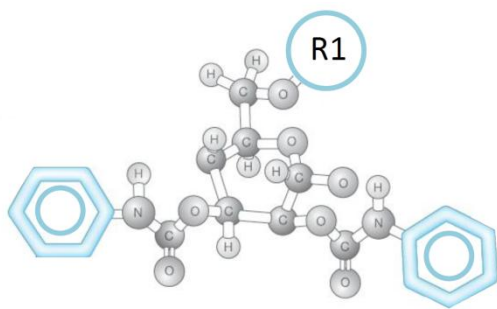


Figure (G). Typical structure of the chiral selector unit of the ChiralCE-1 column.

In the ChiralCE-2 phase (Figure (H)), R2 is a 3,5-Dimethylphenylcarbamate group which was widely used in Phenomenex Lux Cellulose-1 column and Chiralcel OD, OD-H, OD-3, OD-RH, and OD-3R columns. Therefore, the ChiralCE-2 column is an ideal alternative column to the Phenomenex Lux Cellulose-1 column and Chiralcel OD, OD-H, OD-3, OD-RH, and OD-3R columns. Due to the extra chiral recognition function of R1 group, ChiralCE-2 can also provide some unique chiral separations for some solutes.

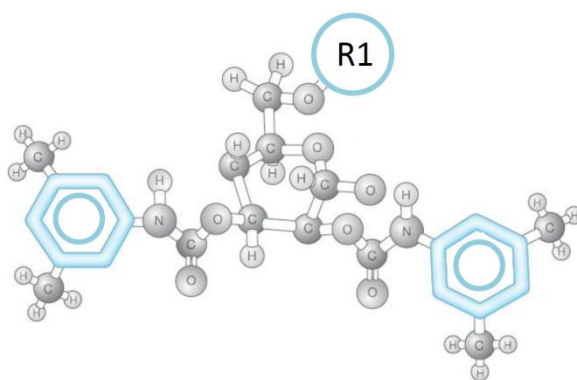


Figure (H). Typical structure of the chiral selector unit of the ChiralCE-2 column.

Most of commercial cellulose-based columns are cellulose-coated columns which can only be used under standard mobile phases. There are only a few cellulose-bonded columns on the market. We provide two new types of cellulose-bonded columns: ChiralCE-1 and ChiralCE-2. Generally, ChiralCE-2 can separate a wider range of chiral compounds than ChiralCE-1 since the former contains more functional groups than the latter.

Pre-processing and Conditioning

The ChiralCE columns were shipped with a mixture of methanol/isopropanol. For use under reversed-phase conditions, the columns need to be first flushed with methanol followed by mobile phase until reaching a constant column pressure. Similarly, for use

ChiralCE Column Notes and Applications in HPLC and UPLC

under normal phase conditions, the columns need to be firstly flushed with isopropanol following by mobile phase until achieving a stable baseline signal. A standard C18 guard column can be used for reversed-phase conditions and a Diol guard column can be used for reversed-phase conditions.

The ChiralCE columns can be used for both traditional HPLC and modern UPLC. However, since packing particles (2 μm or 3 μm) and the inner diameter (2 mm) of the ChiralCE columns are quite small, a low flow rate (e.g., 0.1 to 0.3 mL/min) should be applied when used in traditional HPLC with high viscous mobile phases in order to avoid extreme high back pressure. However, there is no special flow rate limitation for use in UPLC.

Notes and Applications

The positional isomers of disubstituted

benzenes are typical non-chiral compounds. Mixture of *o*-Nitrophenol (2-Nitrophenol, *o*-NP), *m*-Nitrophenol (3-Nitrophenol, *m*-NP), *p*-Nitrophenol (4-Nitrophenol, *p*-NP), and/or mixture of *o*-Nitroaniline (2-Nitroaniline, *o*-NA), *m*-Nitroaniline (3-Nitroaniline, *m*-NA), *p*-Nitroaniline (4-Nitroaniline, *p*-NA) were used to evaluate of the performance of the ChiralCE columns for separation of non-chiral compounds.

A wide range of chiral drug compounds were used to evaluate the enantiomeric separation performance of the ChiralCE columns. Excellent enantiomeric separations were achieved for those testing chiral compounds.

The chromatographic separations were performed on Agilent 1100 HPLC-UV or Agilent 1290 UPLC-MS system under both normal and reversed phase conditions.

ChiralCE Column Notes and Applications in HPLC and UPLC

Disubstituted benzenes on ChiralCE columns

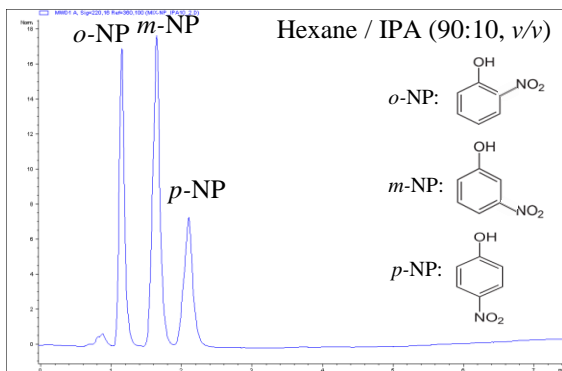


Figure 1-1. Positional isomers *o*-, *m*-, *p*-Nitrophenols (NP) on ChiralCE-1 column.

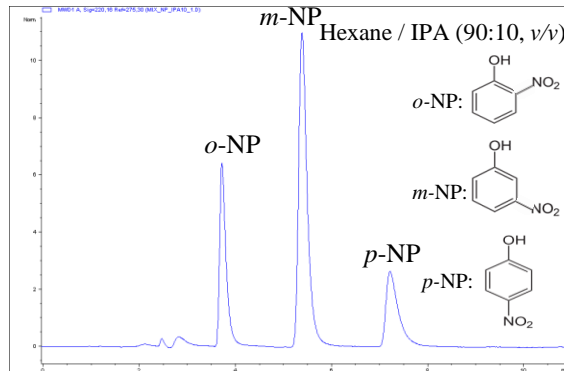


Figure 1-4. Positional isomers *o*-, *m*-, *p*-Nitrophenols on ChiralCE-2 column.

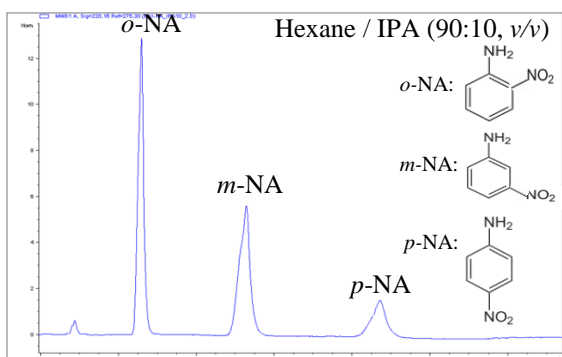


Figure 1-2. Positional isomers *o*-, *m*-, *p*-Nitroanilines(NA) on ChiralCE-1 column.

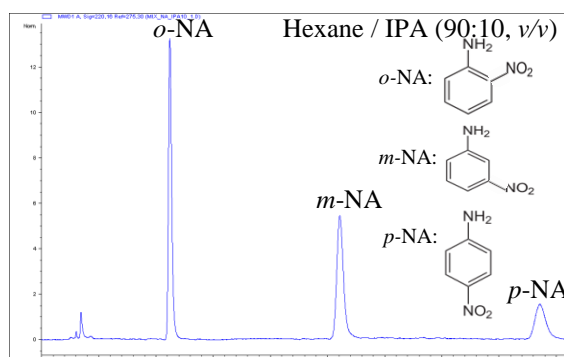


Figure 1-5. Positional isomers *o*-, *m*-, *p*-Nitroanilines on ChiralCE-2 column.

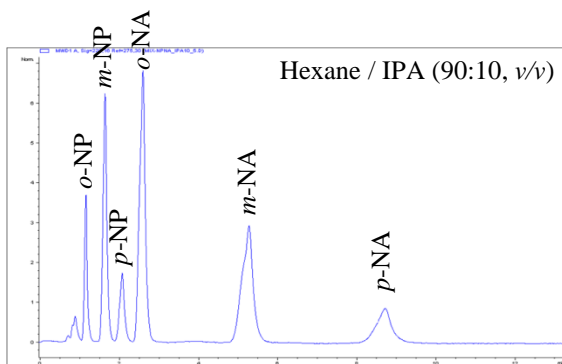


Figure 1-3. Mixture of positional isomers *o*-, *m*-, *p*-Nitrophenols and *o*-, *m*-, *p*-Nitroanilines on ChiralCE-1 column.

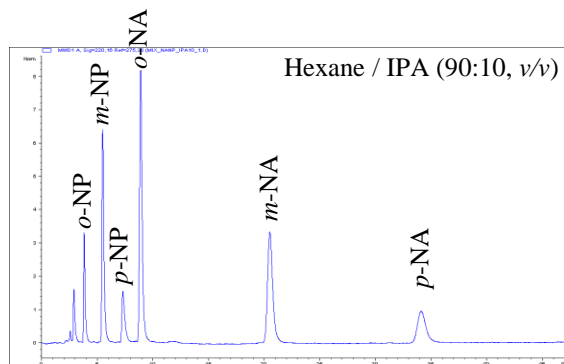


Figure 1-6. Mixture of positional isomers *o*-, *m*-, *p*-Nitrophenols and *o*-, *m*-, *p*-Nitroanilines on ChiralCE-2 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@220nm
Temperature: Ambient

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@220nm
Temperature: Ambient

ChiralCE Column Notes and Applications in HPLC and UPLC

Disubstituted benzenes on ChiralCE columns

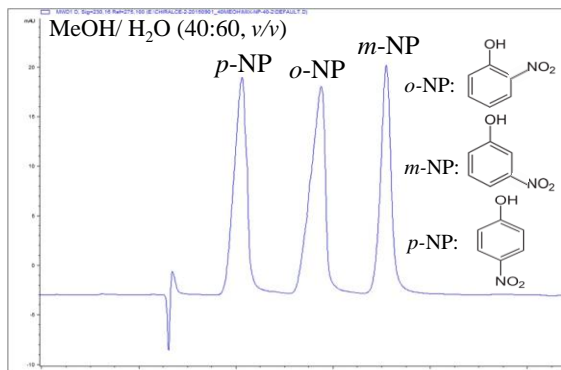


Figure 1-7. Positional isomers *o*-, *m*-, *p*-Nitrophenols on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.2 mL/min
Detection: UV@230nm
Temperature: Ambient
Note: MeOH: Methanol

The elution order for the positional isomers of *o*-, *m*-, *p*-Nitrophenols under normal phase (*o* > *m* > *p*, as shown in Figure 1-4) is different from that under reversed phase (*p* > *o* > *m*) as shown in Figure 1-7 above. This demonstrates that retention mechanisms under normal and reversed phase conditions are different for ChiralCE-2 column. This suggests the ChiralCE columns can provide different and complementary separation abilities under the normal and reversed phase conditions.

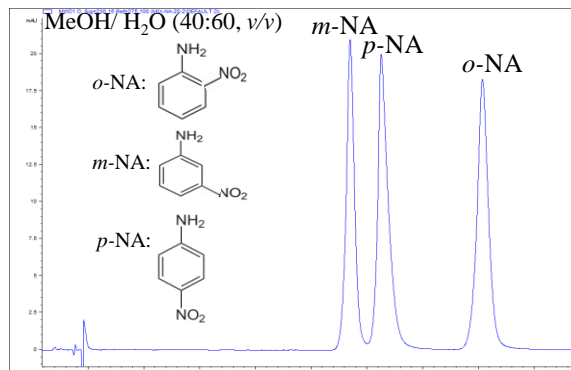


Figure 1-8. Positional isomers *o*-, *m*-, *p*-Nitroanilines on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.2 mL/min
Detection: UV@230nm
Temperature: Ambient
Note: MeOH: Methanol

The elution order for the positional isomers of *o*-, *m*-, *p*-Nitroanilines under reversed phase (*m* > *p* > *o*) as shown in Figure 1-8 is also different from that under normal phase (*o* > *m* > *p*, as shown in Figure 1-5). This confirms that retention mechanisms under normal and reversed phase conditions are different for ChiralCE-2 column. This confirms that the ChiralCE columns can provide different and complementary separation abilities under the normal and reversed phase conditions.

ChiralCE Column Notes and Applications in HPLC and UPLC

Praziquantel on ChiralCE columns

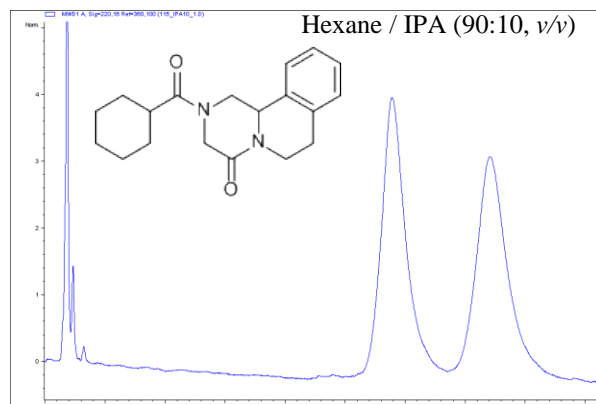


Figure 1-9. Chiral separation of Praziquantel on ChiralCE-1 column.

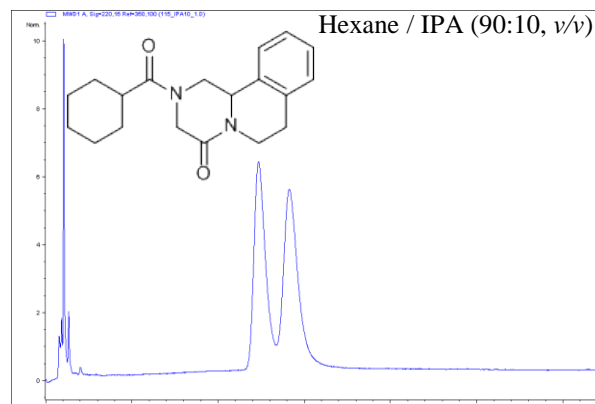


Figure 1-10. Chiral separation of Praziquantel on ChiralCE-2 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@220nm
Temperature: Ambient
Note: IPA: Isopropanol

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@220nm
Temperature: Ambient
Note: IPA: Isopropanol

ChiralCE Column Notes and Applications in HPLC and UPLC

Flavonoids Naringenin and 4'-Hydroxyflavanone on ChiralCE columns

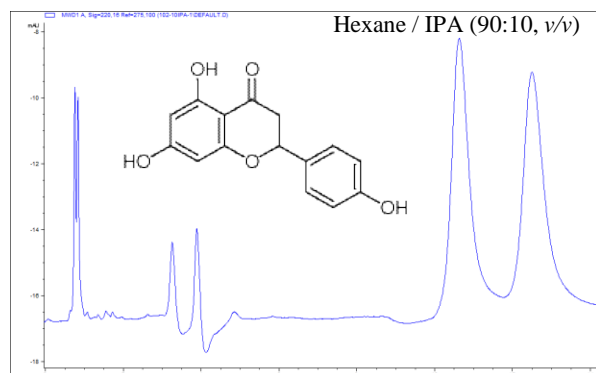


Figure 1-11. Chiral separation of Naringenin on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@220nm
Temperature: Ambient

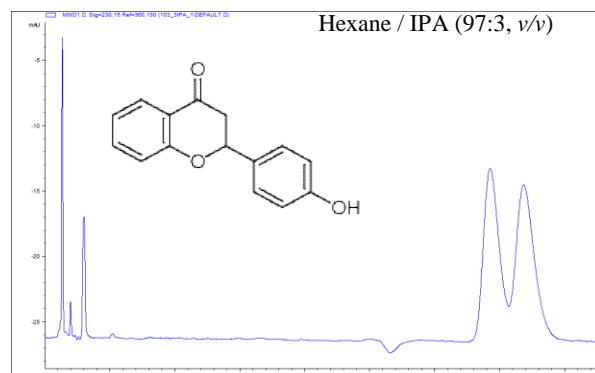


Figure 1-12. Chiral separation of 4'-Hydroxyflavanone on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.6 mL/min
Detection: UV@230nm
Temperature: Ambient

ChiralCE Column Notes and Applications in HPLC and UPLC

Flavonoids Flavanone on ChiralCE columns

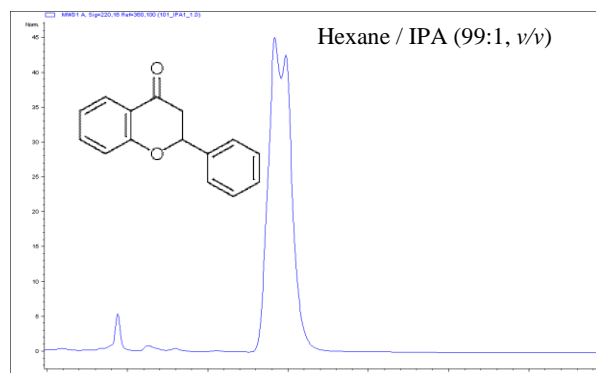


Figure 1-13. Chiral separation of Flavanone on ChiralCE-1 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.8 mL/min
Detection: UV@220nm
Temperature: Ambient

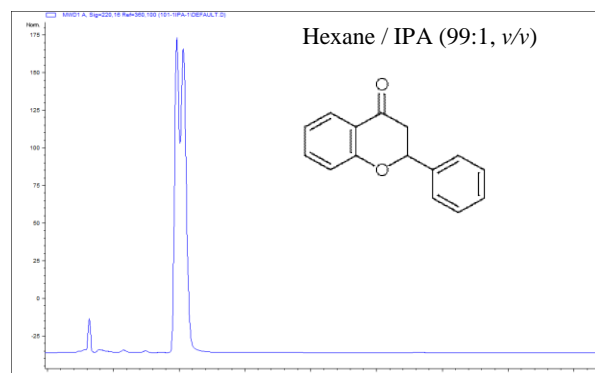


Figure 1-14. Chiral separation of Flavanone on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.8 mL/min
Detection: UV@220nm
Temperature: Ambient

ChiralCE Column Notes and Applications in HPLC and UPLC

Flavonoids Methoxyflavanone on ChiralCE columns

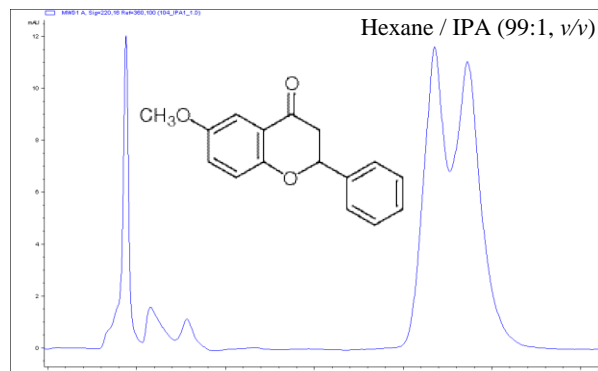


Figure 1-15. Chiral separation of Methoxyflavanone on ChiralCE-1 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.8 mL/min
Detection: UV@220nm
Temperature: Ambient

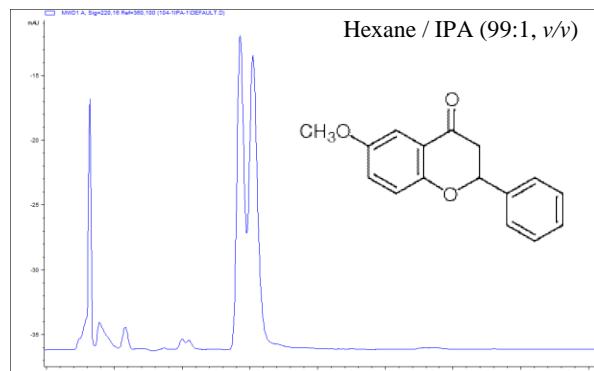


Figure 1-16. Chiral separation of Methoxyflavanone on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.8 mL/min
Detection: UV@220nm
Temperature: Ambient

ChiralCE Column Notes and Applications in HPLC and UPLC

Chromanol on ChiralCE columns

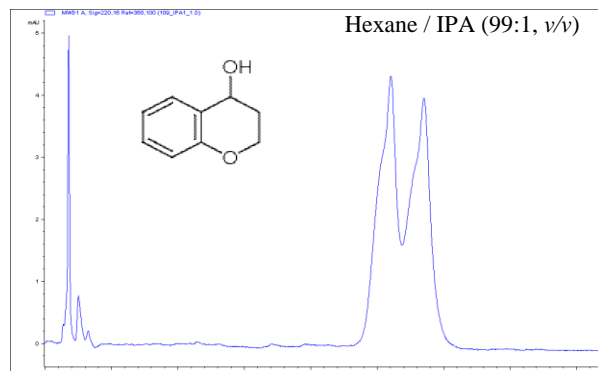


Figure 1-17. Chiral separation of Chromanol on ChiralCE-1 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.8 mL/min
Detection: UV@220nm
Temperature: Ambient

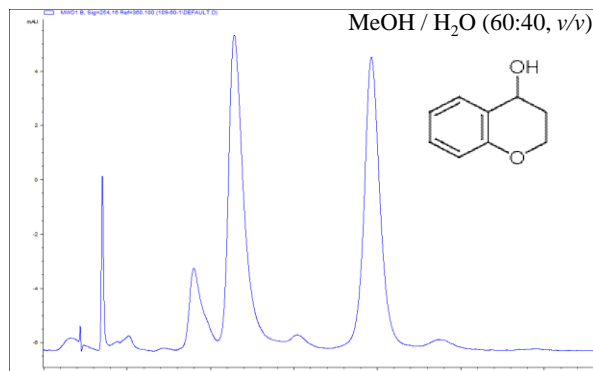


Figure 1-18. Chiral separation of Chromanol on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.2 mL/min
Detection: UV@254nm
Temperature: Ambient

ChiralCE Column Notes and Applications in HPLC and UPLC

Trichloromethiazide and Bicalutamide on ChiralCE columns

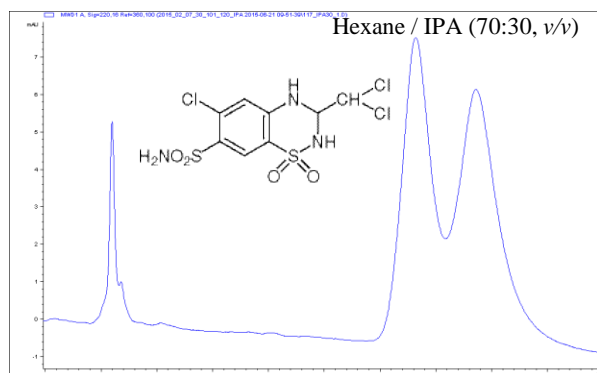


Figure 1-19. Chiral separation of Trichloromethiazide on ChiralCE-1 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.4 mL/min
Detection: UV@220nm
Temperature: Ambient

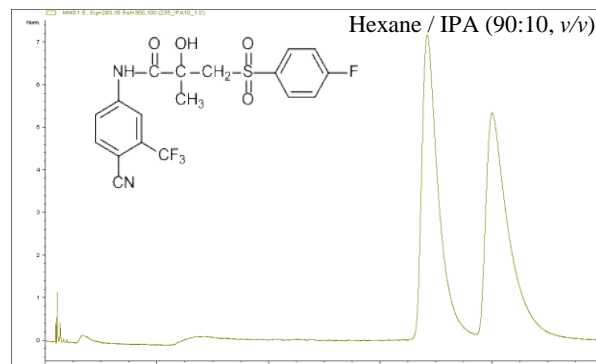


Figure 1-20. Chiral separation of Bicalutamide on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@280nm
Temperature: Ambient

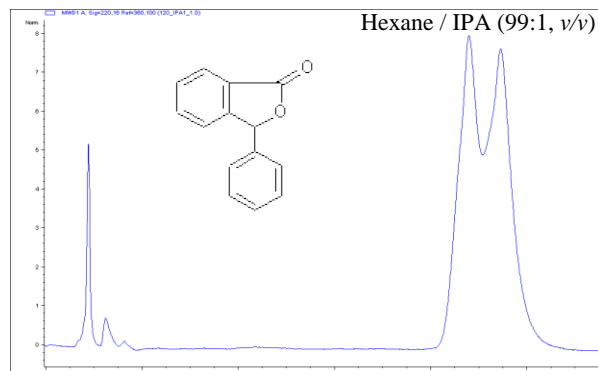
ChiralCE Column Notes and Applications in HPLC and UPLC**3-Phenylphthalide on ChiralCE columns**

Figure 1-21. Chiral separation of 3-Phenylphthalide on ChiralCE-1 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.8 mL/min
Detection: UV@220nm
Temperature: Ambient

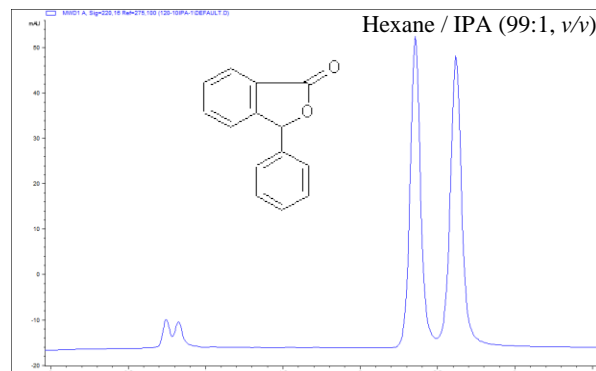


Figure 1-22. Chiral separation of 3-Phenylphthalide on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.8 mL/min
Detection: UV@220nm
Temperature: Ambient

ChiralCE Column Notes and Applications in HPLC and UPLC

3-Phenylphthalide on ChiralCE columns

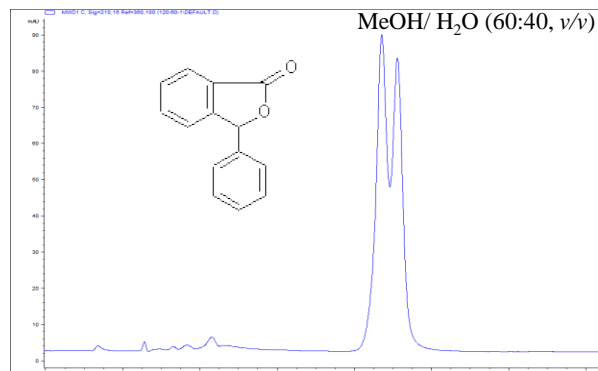


Figure 1-23. Chiral separation of 3-Phenylphthalide on ChiralCE-1 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.2 mL/min
Detection: UV@210nm
Temperature: Ambient

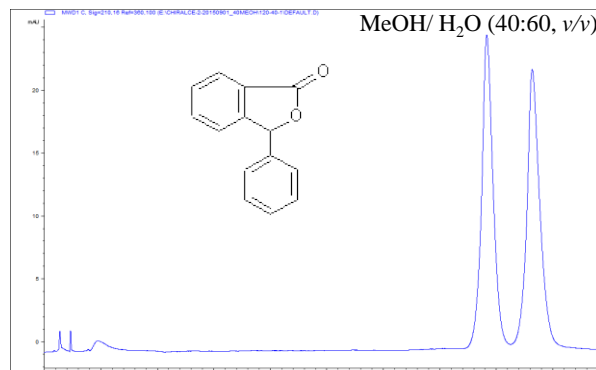


Figure 1-24. Chiral separation of 3-Phenylphthalide on ChiralCE-2 column.

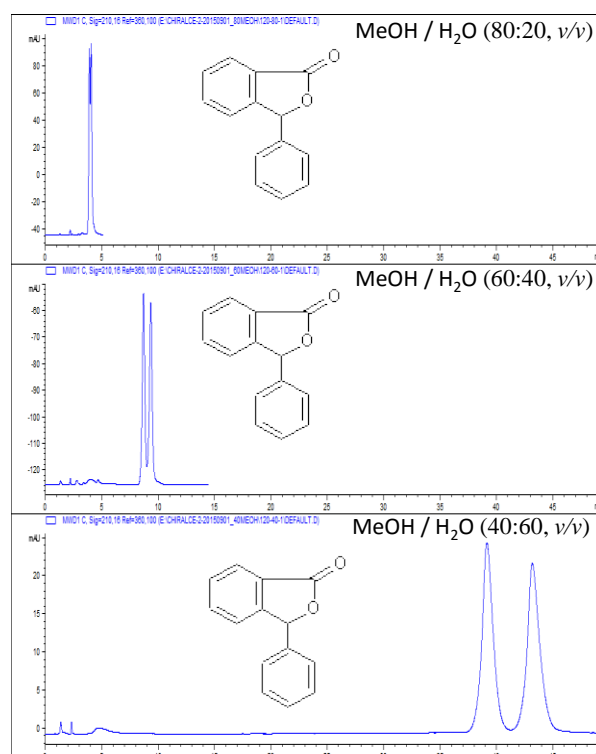


Figure 1-25. Progressive chiral separation of 3-Phenylphthalide on ChiralCE-2 column as composition of mobile phase is varied.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.2 mL/min
Detection: UV@210nm
Temperature: Ambient

ChiralCE Column Notes and Applications in HPLC and UPLC

Promethazine on ChiralCE columns

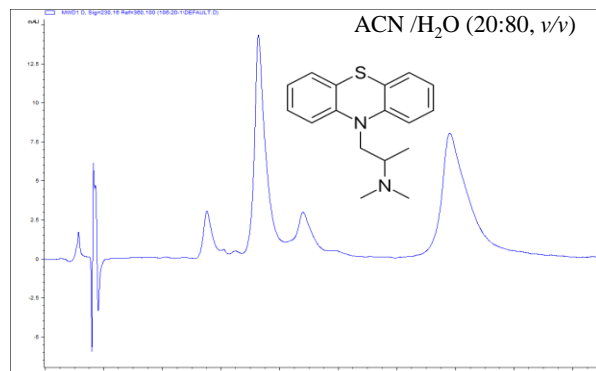


Figure 1-26. Chiral separation of Promethazine on ChiralCE-1 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.3 mL/min
Detection: UV@230nm
Temperature: Ambient

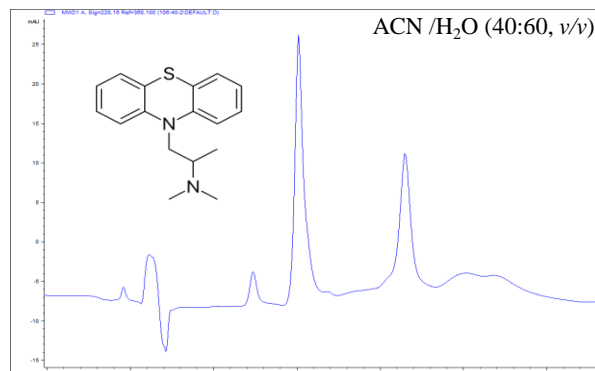


Figure 1-27. Chiral separation of Promethazine on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.35 mL/min
Detection: UV@220nm
Temperature: Ambient

ChiralCE Column Notes and Applications in HPLC and UPLC

Benzoin and Anisoin on ChiralCE columns

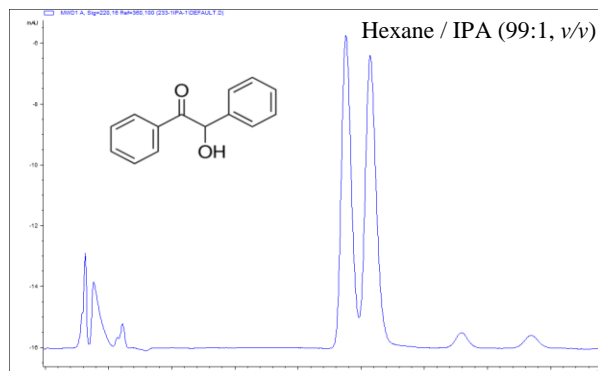


Figure 1-28. Chiral separation of Benzoin on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.8 mL/min
Detection: UV@220nm
Temperature: Ambient

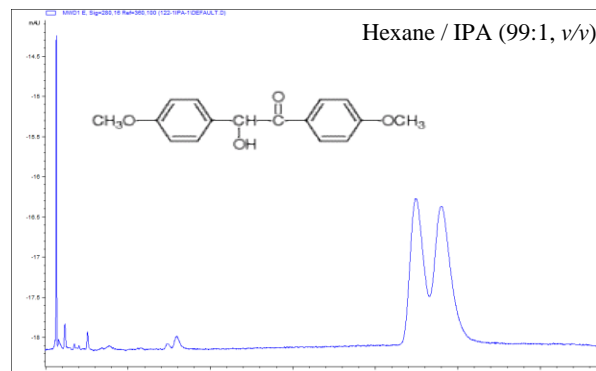


Figure 1-29. Chiral separation of Anisoin on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.8 mL/min
Detection: UV@280nm
Temperature: Ambient

ChiralCE Column Notes and Applications in HPLC and UPLC

Stilbene Oxide on ChiralCE columns

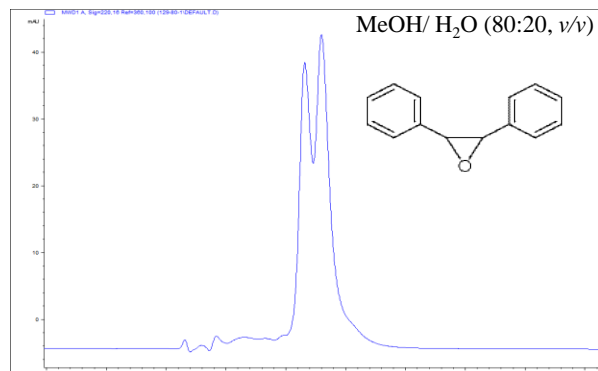


Figure 1-30. Chiral separation of Stilbene Oxide on ChiralCE-1 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.2 mL/min
Detection: UV@220nm
Temperature: Ambient

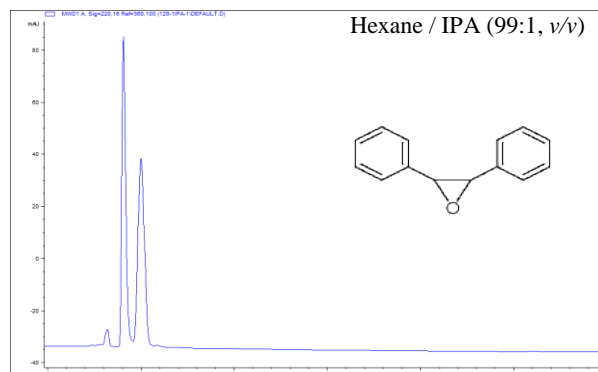


Figure 1-31. Chiral separation of Stilbene Oxide on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.8 mL/min
Detection: UV@220nm
Temperature: Ambient

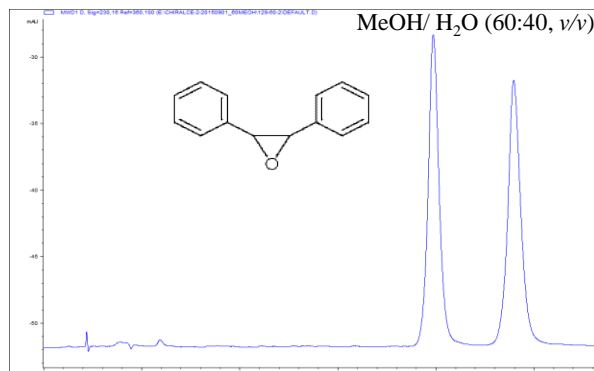


Figure 1-32. Chiral separation of Stilbene Oxide on ChiralCE-2 column.

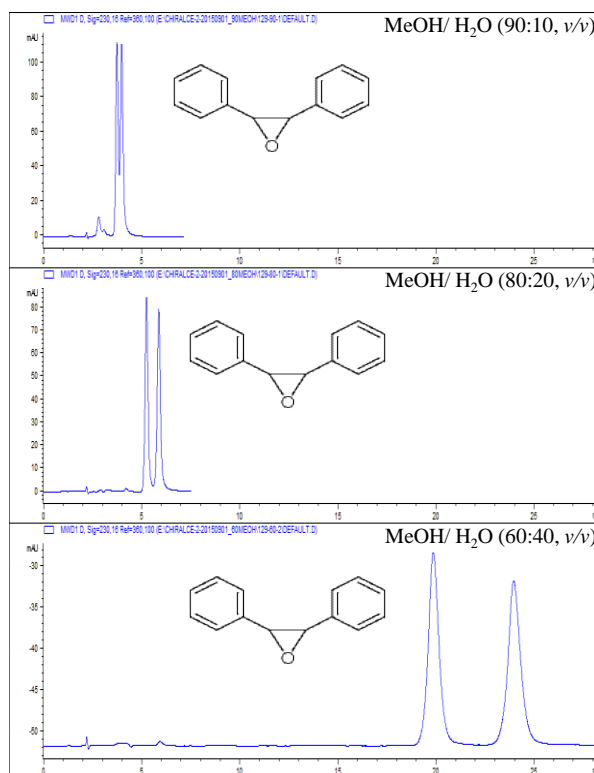


Figure 1-33. Progressive chiral separation of Stilbene Oxide on ChiralCE-2 column as composition of mobile phase is varied.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.2 mL/min
Detection: UV@230nm
Temperature: Ambient

ChiralCE Column Notes and Applications in HPLC and UPLC

Fenoprofen and Chlormezanone on ChiralCE columns

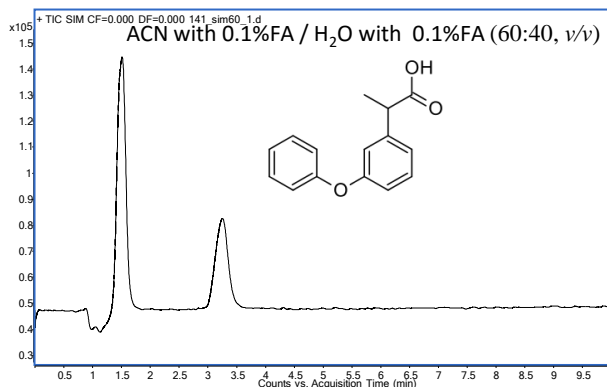


Figure 1-34. Chiral separation of Fenoprofen on ChiralCE-1 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE1-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.4 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient
Note: ACN: Acetonitrile
 FA: Formic Acid

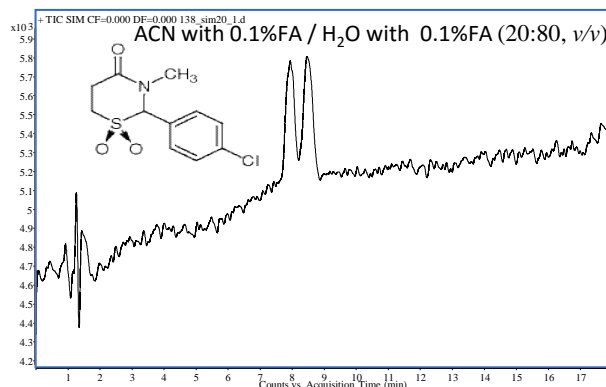


Figure 1-35. Chiral separation Chlormezanone on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.4 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient
Note: ACN: Acetonitrile
 FA: Formic Acid

ChiralCE Column Notes and Applications in HPLC and UPLC

Trichloromethiazide on ChiralCE columns

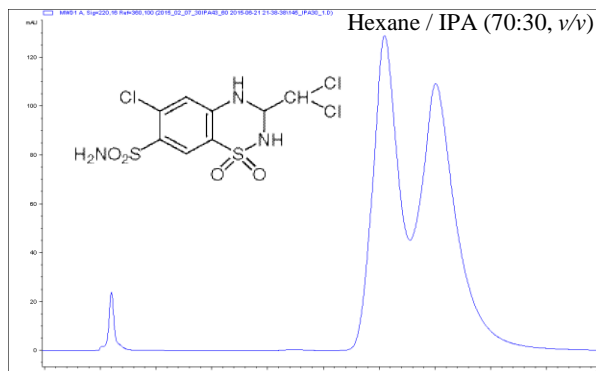


Figure 1-36. Chiral separation of Trichloromethiazide on ChiralCE-1 column.

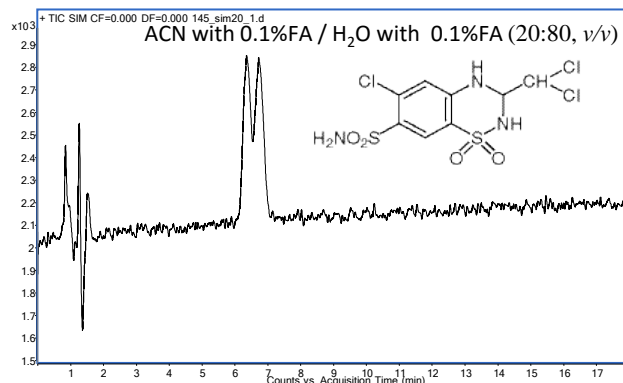


Figure 1-37. Chiral separation Trichloromethiazide on ChiralCE-2 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.4 mL/min
Detection: UV@220nm
Temperature: Ambient

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.4 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

ChiralCE Column Notes and Applications in HPLC and UPLC

Salbutamol and Bupivacaine on ChiralCE columns

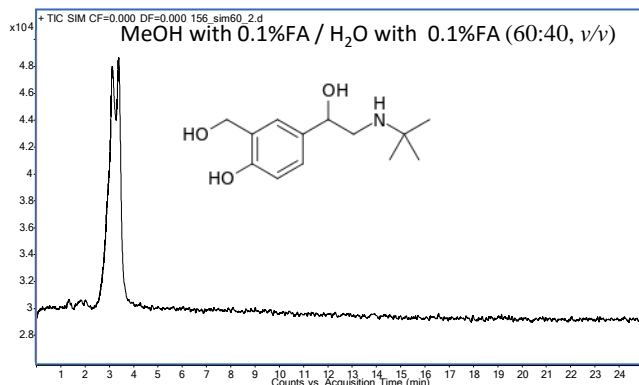


Figure 1-38. Chiral separation of Salbutamol on ChiralCE-1 column.

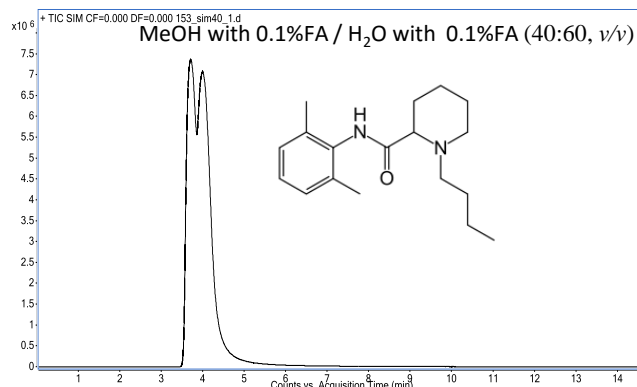


Figure 1-39. Chiral separation Bupivacaine on ChiralCE-2 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE1-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.25 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient
Note: MeOH: Methanol
 FA: Formic Acid

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.25 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient
Note: MeOH: Methanol
 FA: Formic Acid

ChiralCE Column Notes and Applications in HPLC and UPLC

Amlodipine and Fluoxetine on ChiralCE columns

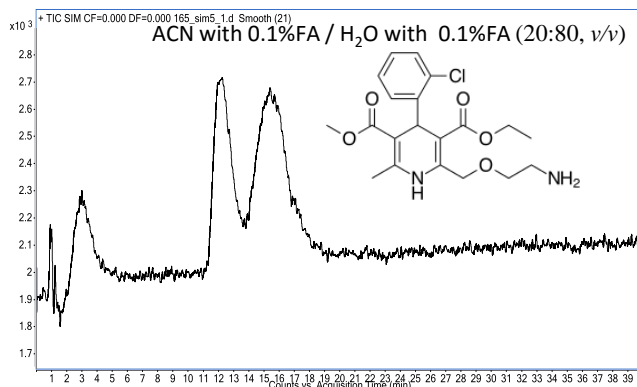


Figure 1-40. Chiral separation of Amlodipine on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.4 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

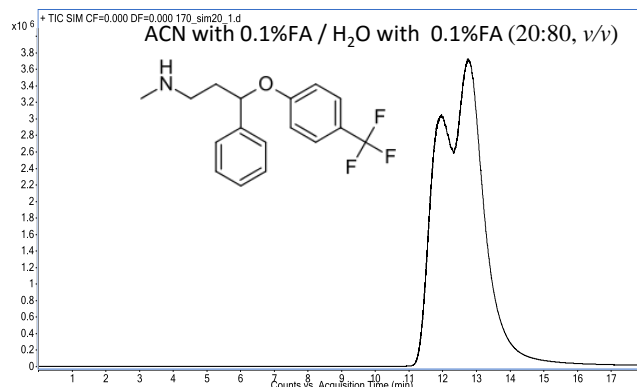


Figure 1-41. Chiral separation Fluoxetine on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.4 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

ChiralCE Column Notes and Applications in HPLC and UPLC

Bifonazole on ChiralCE columns

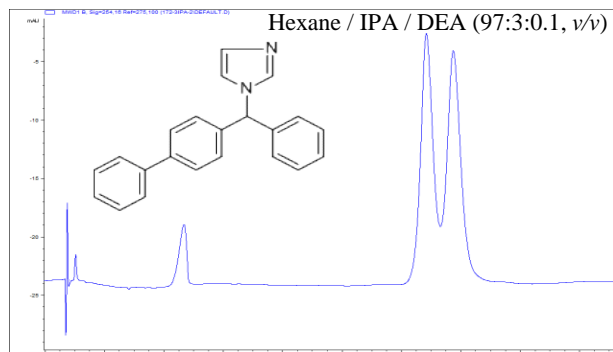


Figure 1-42. Chiral separation of Bifonazole on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.7 mL/min
Detection: UV@254nm
Temperature: Ambient

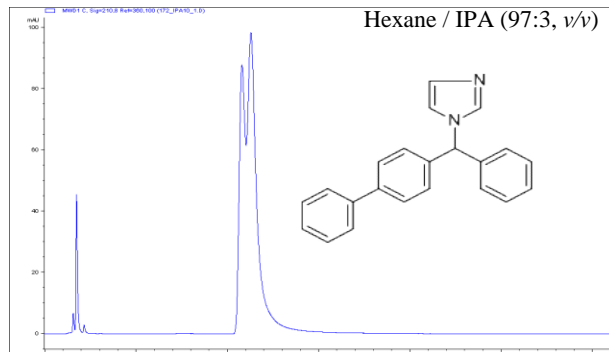


Figure 1-43. Chiral separation of Bifonazole on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@210nm
Temperature: Ambient

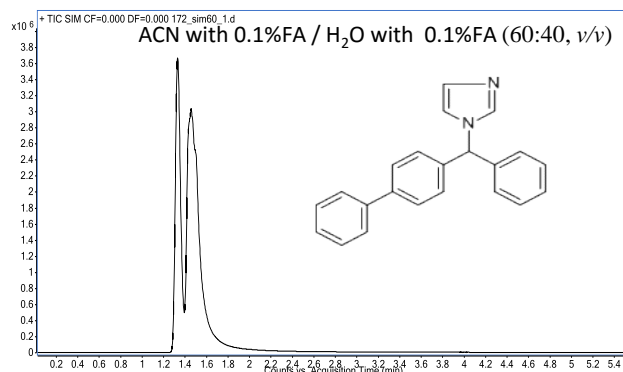


Figure 1-44. Chiral separation of Bifonazole on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.4 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

ChiralCE Column Notes and Applications in HPLC and UPLC

Econazole and Indoprofen on ChiralCE columns

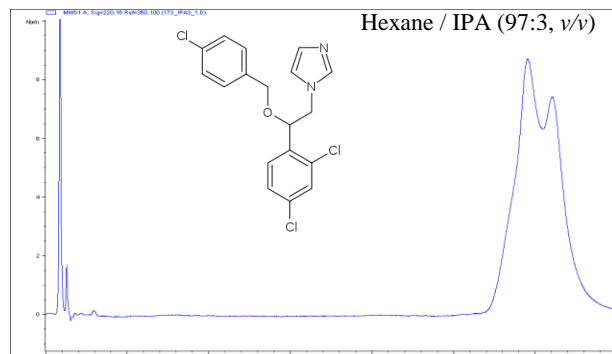


Figure 1-45. Chiral separation of Econazole on ChiralCE-1 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.6 mL/min
Detection: UV@220nm
Temperature: Ambient

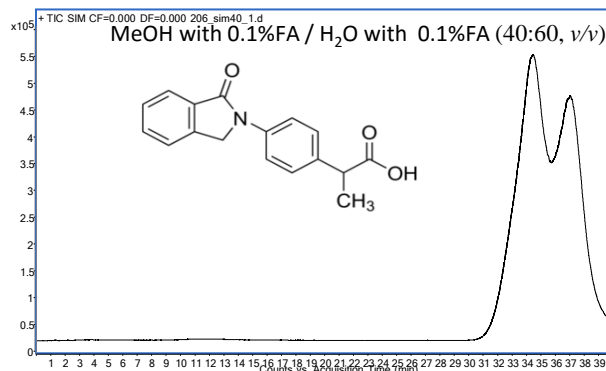


Figure 1-46. Chiral separation Indoprofen on ChiralCE-1 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE1-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.25 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

ChiralCE Column Notes and Applications in HPLC and UPLC

Indapamide on ChiralCE columns

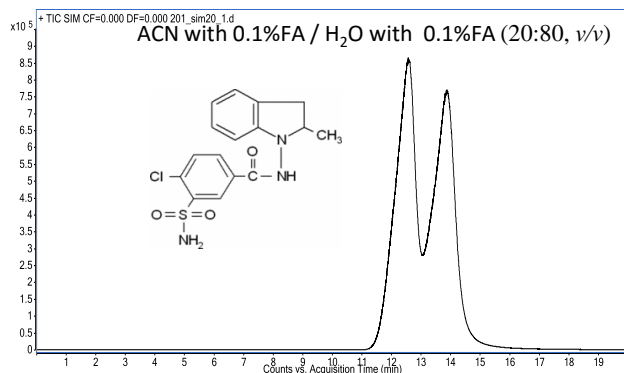


Figure 1-47. Chiral separation of Indapamide on ChiralCE-1 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE1-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.4 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

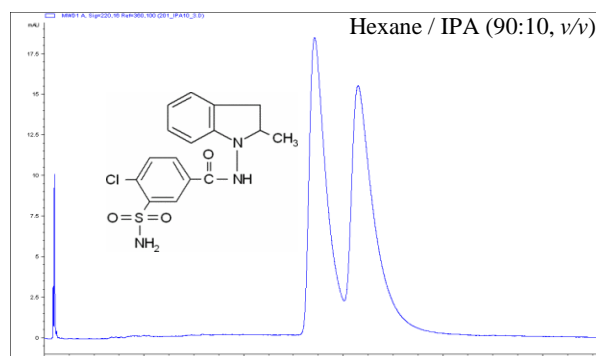


Figure 1-48. Chiral separation of Indapamide on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@220nm
Temperature: Ambient

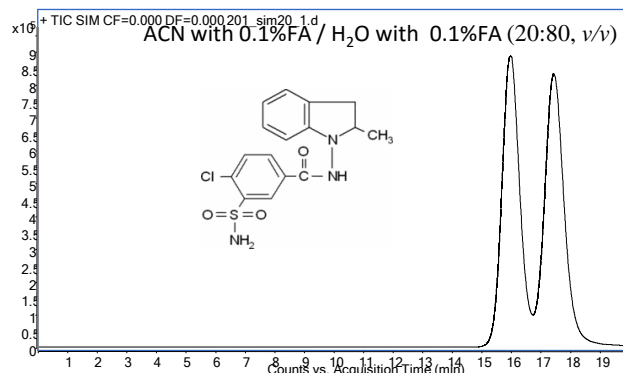


Figure 1-49. Chiral separation Indapamide on ChiralCE-2 column.

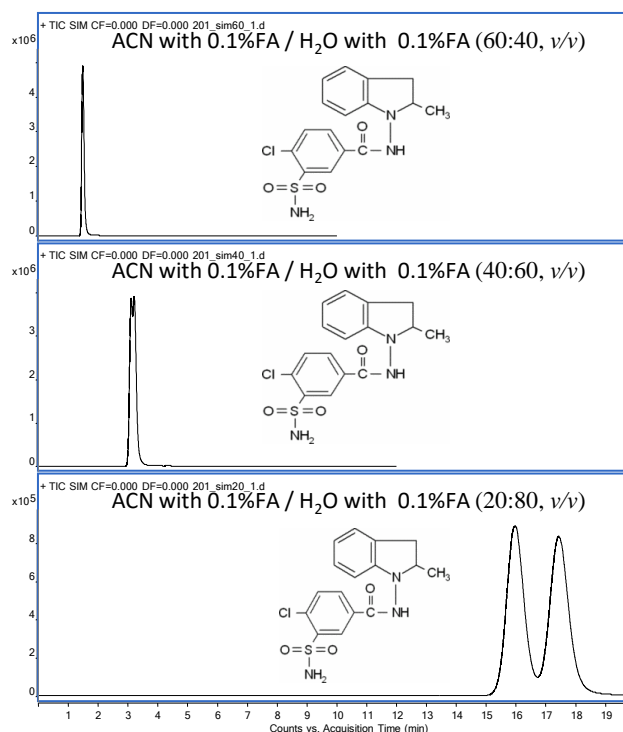


Figure 1-50. Progressive chiral separation of Indapamide on ChiralCE-2 column as composition of mobile phase is varied.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.4 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

ChiralCE Column Notes and Applications in HPLC and UPLC

Proglumide on ChiralCE columns

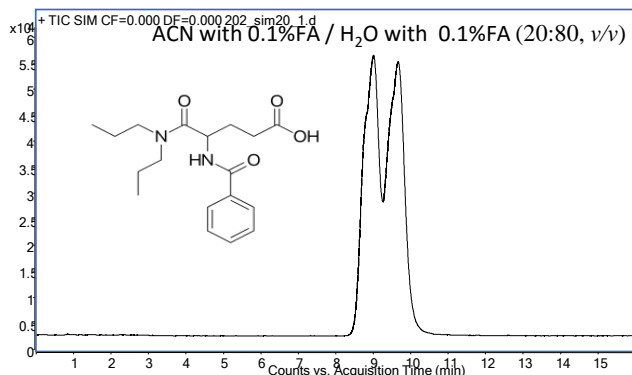


Figure 1-51. Chiral separation of Proglumide on ChiralCE-2 column.

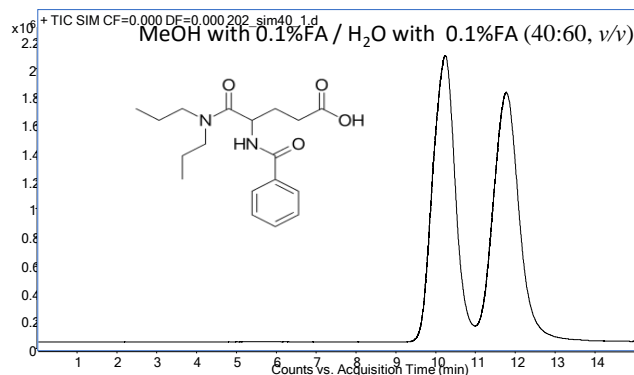


Figure 1-52. Chiral separation of Proglumide on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.4 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

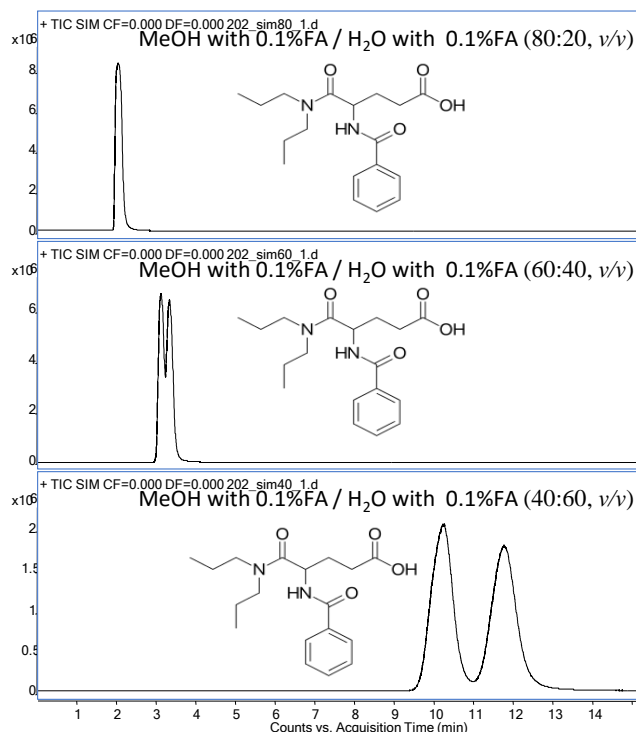


Figure 1-53. Progressive chiral separation of Proglumide on ChiralCE-2 column as composition of mobile phase is varied.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.25 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

ChiralCE Column Notes and Applications in HPLC and UPLC

Warfarin on ChiralCE columns

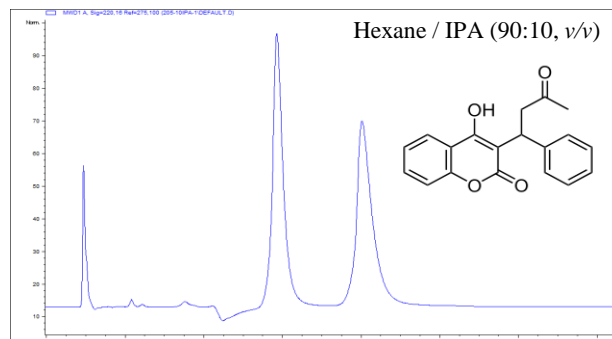


Figure 1-54. Chiral separation of Warfarin on ChiralCE-1 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@220nm
Temperature: Ambient

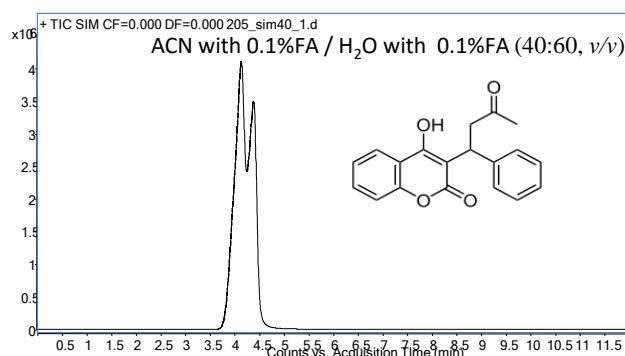


Figure 1-55 Chiral separation of Warfarin on ChiralCE-1 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE1-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.4 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

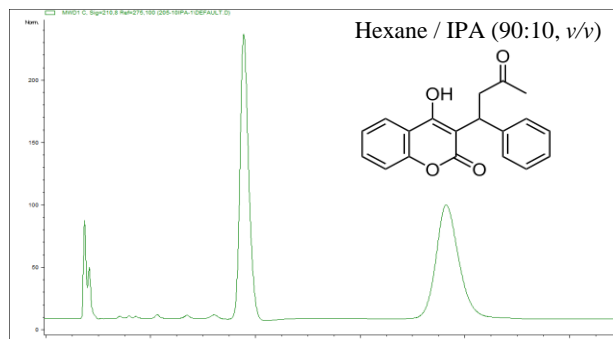


Figure 1-56. Chiral separation of Warfarin on ChiralCE-2 column.

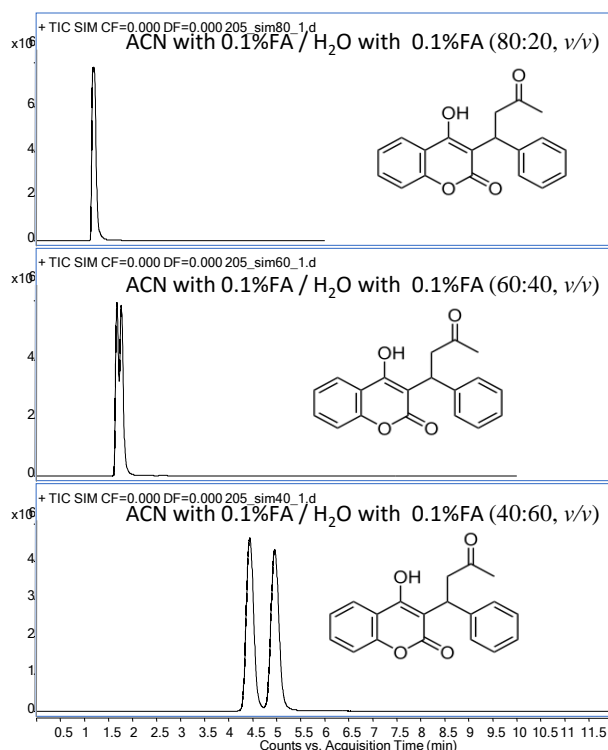


Figure 1-57. Progressive chiral separation of Warfarin on ChiralCE-2 column as composition of mobile phase is varied.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.4 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

ChiralCE Column Notes and Applications in HPLC and UPLC

Warfarin on ChiralCE columns

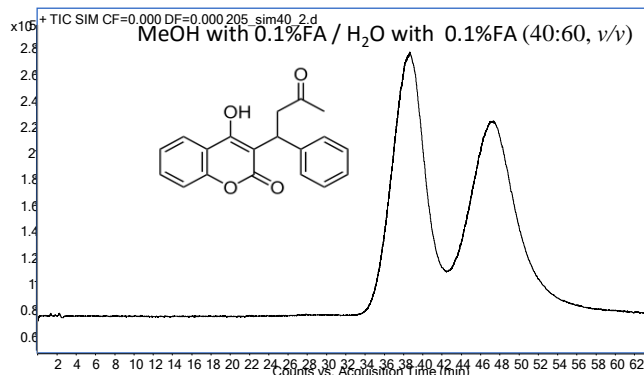


Figure 1-58. Chiral separation of Warfarin on ChiralCE-1 column.

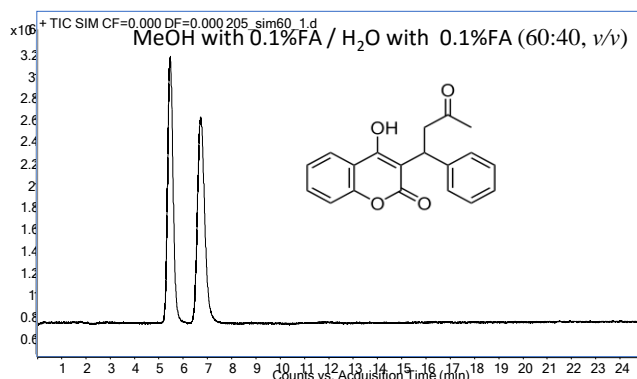


Figure 1-60. Chiral separation of Warfarin on ChiralCE-2 column.

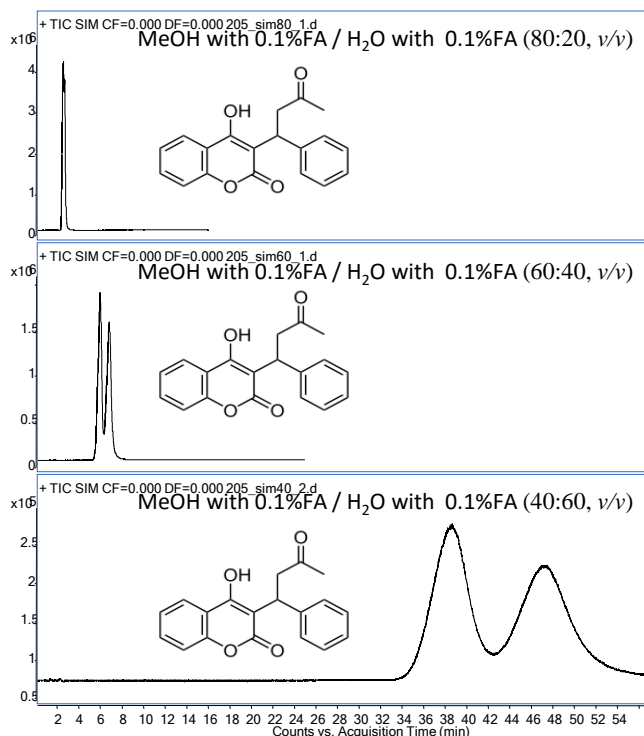


Figure 1-59. Progressive chiral separation of Warfarin on ChiralCE-1 column as composition of mobile phase is varied.

Column: ChiralCE-1
 Dimension: 3 μ m, 150 \times 2 mm I.D.
 Part No. 813-CE1-03
 LC Mode: UPLC (Agilent1290)
 Flow rate: 0.25 mL/min
 Detection: MSD@SIM *m/z*
 Temperature: Ambient

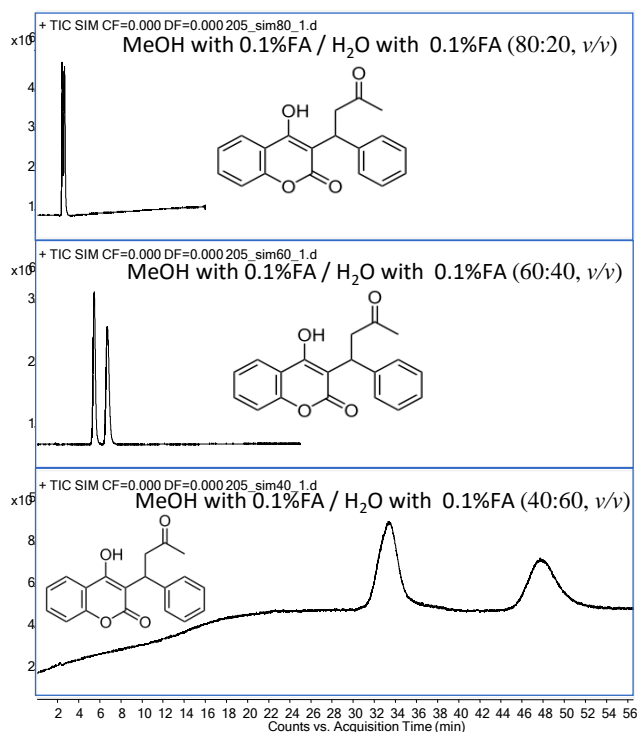


Figure 1-61. Progressive chiral separation of Warfarin on ChiralCE-2 column as composition of mobile phase is varied.

Column: ChiralCE-2
 Dimension: 3 μ m, 150 \times 2 mm I.D.
 Part No. 813-CE2-03
 LC Mode: UPLC (Agilent1290)
 Flow rate: 0.25 mL/min
 Detection: MSD@SIM *m/z*
 Temperature: Ambient

ChiralCE Column Notes and Applications in HPLC and UPLC

1-(1-Naphthyl)ethanol on ChiralCE columns

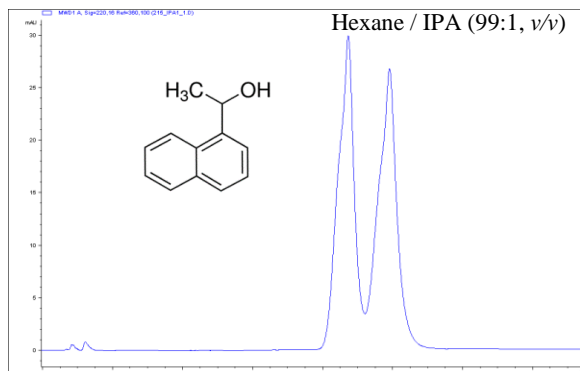


Figure 1-62. Chiral separation of 1-(1-Naphthyl)ethanol on ChiralCE-1 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 801-CE1-01
LC Mode: HPLC (Agilent1100)
Flow rate: 0.6 mL/min
Detection: UV@220nm
Temperature: Ambient

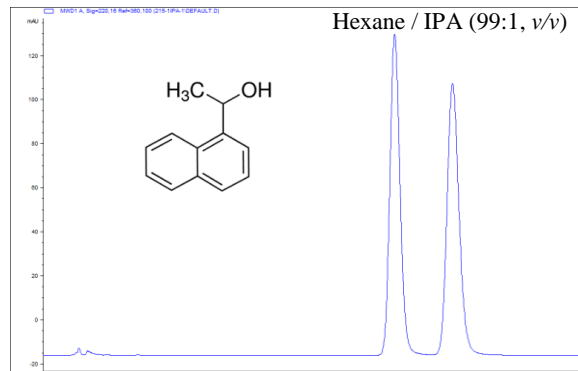


Figure 1-64. Chiral separation of 1-(1-Naphthyl)ethanol on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 801-CE2-01
LC Mode: HPLC (Agilent1100)
Flow rate: 0.6 mL/min
Detection: UV@220nm
Temperature: Ambient

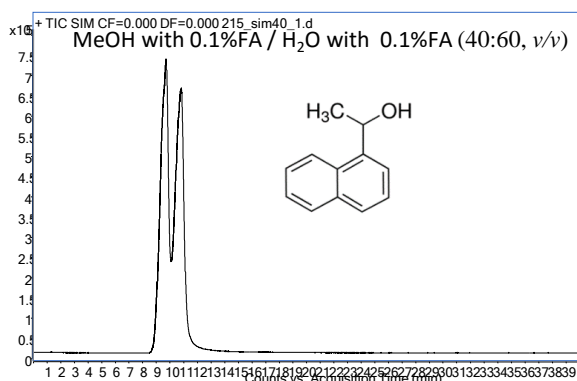


Figure 1-63. Chiral separation of 1-(1-Naphthyl)ethanol on ChiralCE-1 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE1-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.25 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

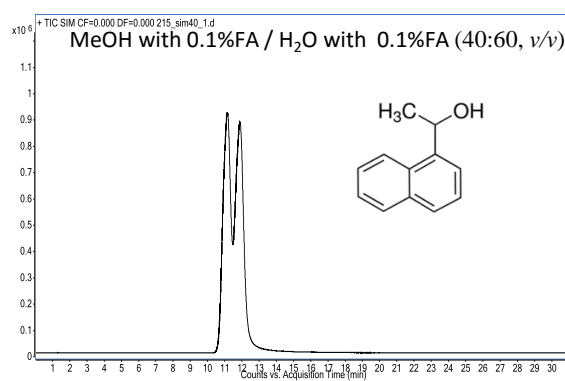


Figure 1-65. Chiral separation 1-(1-Naphthyl)ethanol on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.25 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

ChiralCE Column Notes and Applications in HPLC and UPLC

Furoin on ChiralCE columns

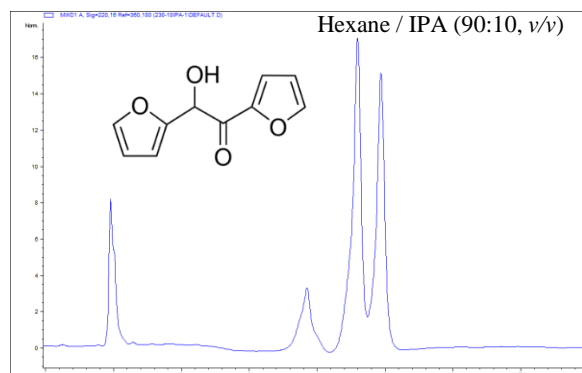


Figure 1-66. Chiral separation of Furoin on ChiralCE-1 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@220nm
Temperature: Ambient

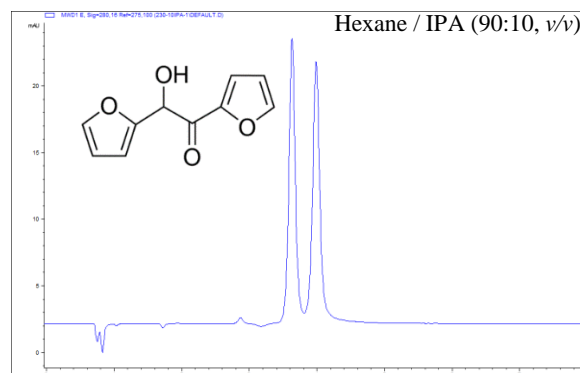


Figure 1-67. Chiral separation of Furoin on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@280nm
Temperature: Ambient

ChiralCE Column Notes and Applications in HPLC and UPLC

Zearalanol on ChiralCE columns

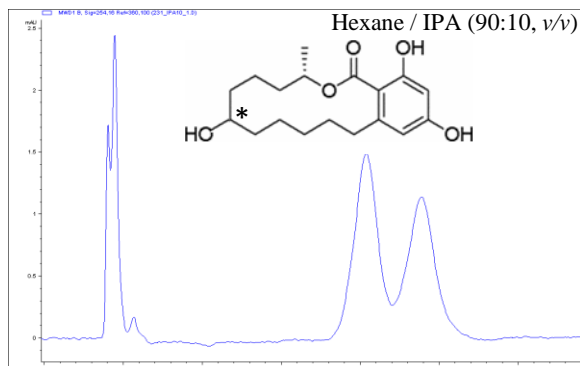


Figure 1-68. Chiral separation of (α and β)-Zearalanol on ChiralCE-1 column.

Column: ChiralCE-1
Dimension: 3 μm, 150 × 2 mm I.D.
Part No. 813-CE1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@220nm
Temperature: Ambient

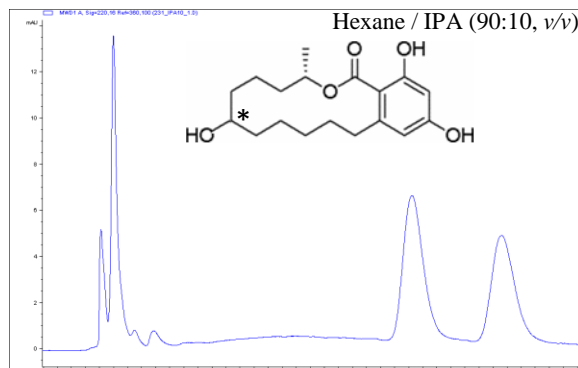


Figure 1-70. Chiral separation of (α and β)-Zearalanol on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μm, 150 × 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@280nm
Temperature: Ambient

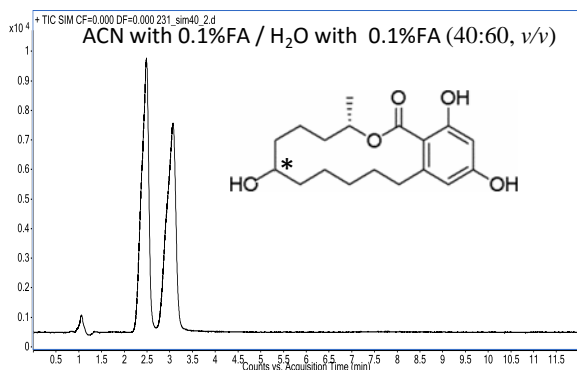


Figure 1-69. Chiral separation of (α and β)-Zearalanol on ChiralCE-1 column.

Column: ChiralCE-1
Dimension: 3 μm, 150 × 2 mm I.D.
Part No. 813-CE1-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.4 mL/min
Detection: MSD@SIM *m/z*
Temperature: Ambient

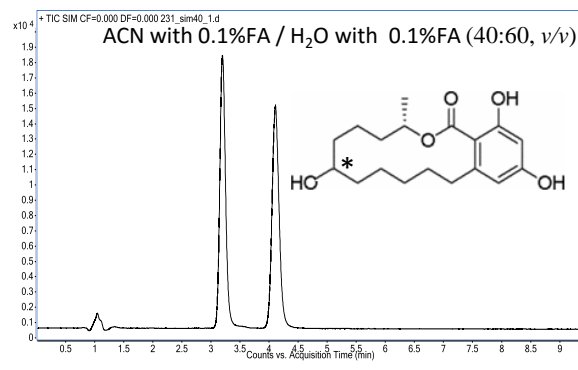


Figure 1-71. Chiral separation (α and β)-Zearalanol on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μm, 150 × 2 mm I.D.
Part No. 813-CE2-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.4 mL/min
Detection: MSD@SIM *m/z*
Temperature: Ambient

ChiralCE Column Notes and Applications in HPLC and UPLC

Zearalanol on ChiralCE columns

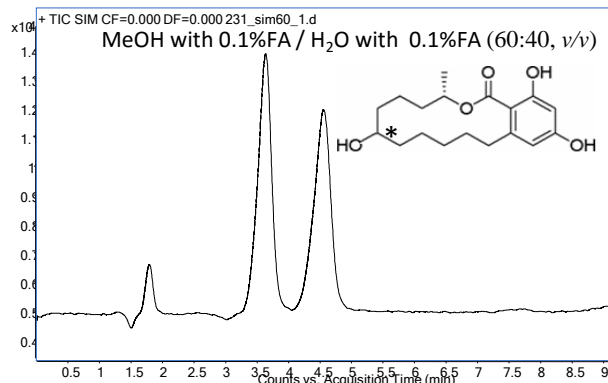


Figure 1-72. Chiral separation of (α and β)-Zearalanol on ChiralCE-1 column.

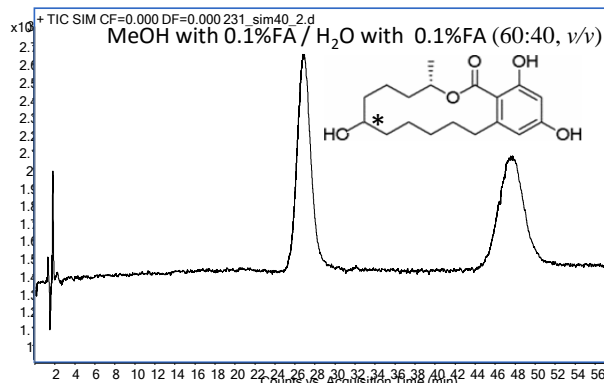


Figure 1-74. Chiral separation of (α and β)-Zearalanol on ChiralCE-2 column.

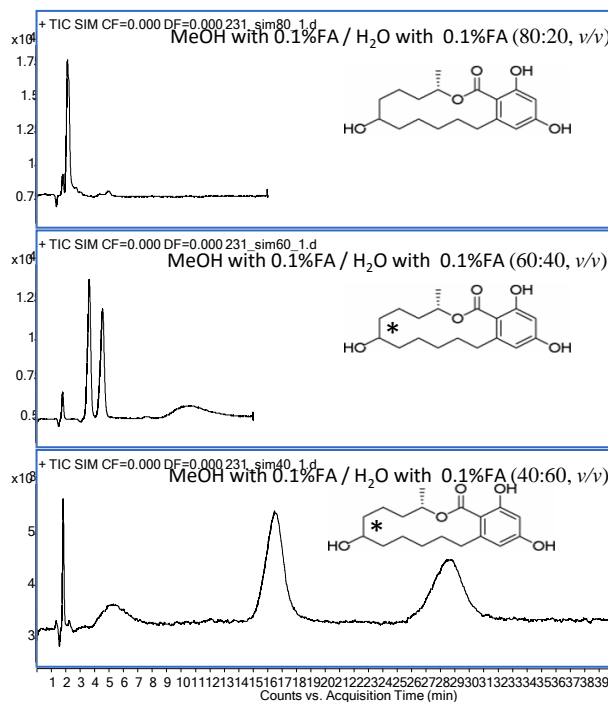


Figure 1-73. Progressive chiral separation of (α and β)-Zearalanol on ChiralCE-1 column as composition of mobile phase is varied.

Column: ChiralCE-1
Dimension: 3 μm, 150 × 2 mm I.D.
Part No. 813-CE1-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.25 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

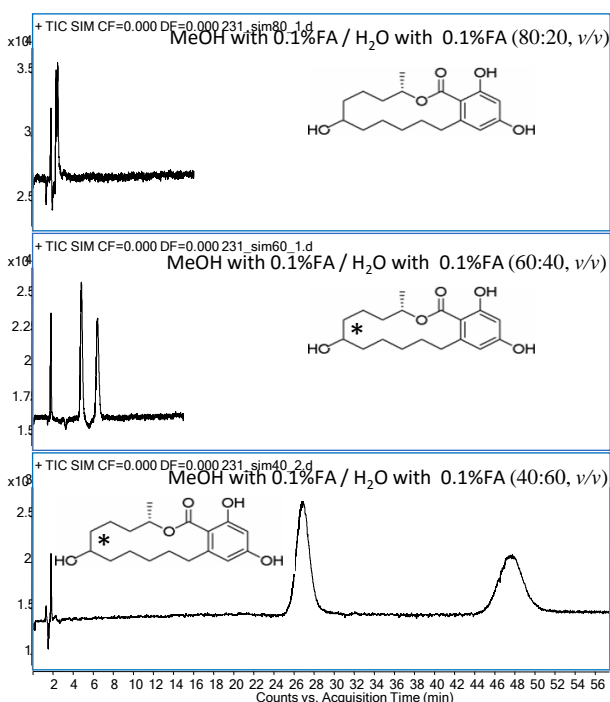


Figure 1-75. Progressive chiral separation of (α and β)-Zearalanol on ChiralCE-2 column as composition of mobile phase is varied.

Column: ChiralCE-2
Dimension: 3 μm, 150 × 2 mm I.D.
Part No. 813-CE2-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.25 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

ChiralCE Column Notes and Applications in HPLC and UPLC

Vinclozolin and Prilocaine on ChiralCE columns

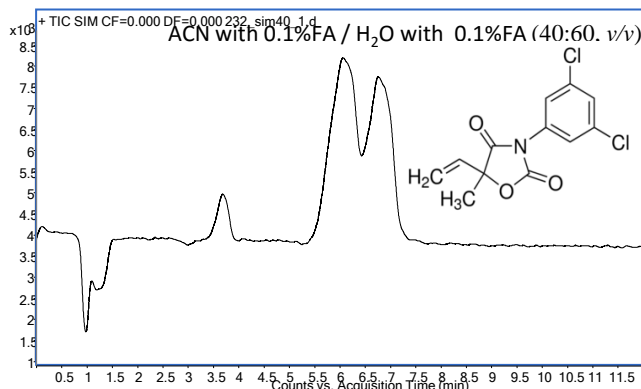


Figure 1-76. Chiral separation of Vinclozolin on ChiralCE-1 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE1-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.4 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

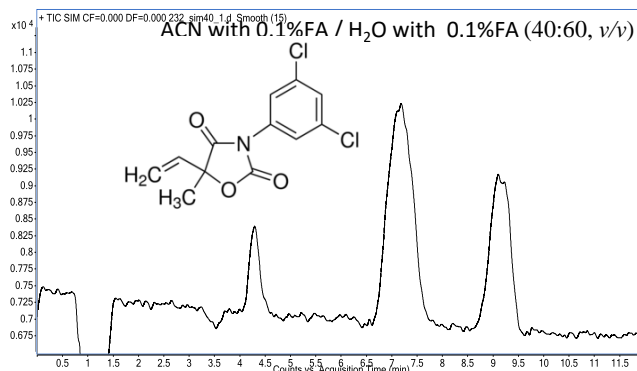


Figure 1-77. Chiral separation Vinclozolin on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.4 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

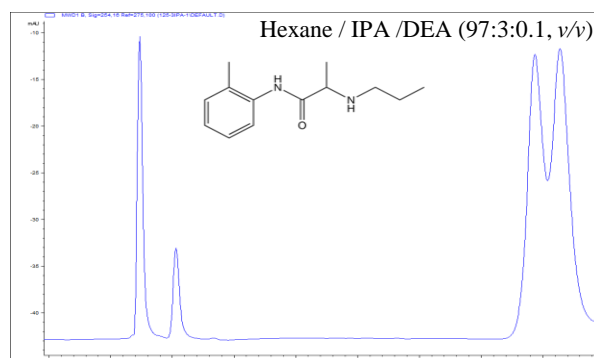


Figure 1-78. Chiral separation of Prilocaine on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.7 mL/min
Detection: UV@254nm
Temperature: Ambient
Note: DEA: Diethylamine

ChiralCE Column Notes and Applications in HPLC and UPLC

Thalidomide and Prilocaine on ChiralCE columns

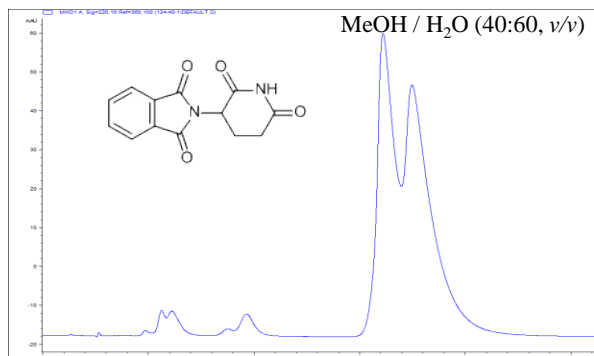


Figure 1-79. Chiral separation of Thalidomide on ChiralCE-1 column.

Column: ChiralCE-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.18 mL/min
Detection: UV@220nm
Temperature: Ambient

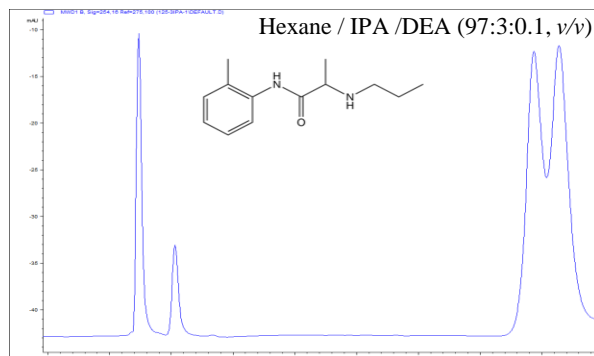


Figure 1-80. Chiral separation of Prilocaine on ChiralCE-2 column.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.7 mL/min
Detection: UV@254nm
Temperature: Ambient
Note: DEA: Diethylamine

ChiralCE Column Notes and Applications in HPLC and UPLC

Thalidomide on ChiralCE columns

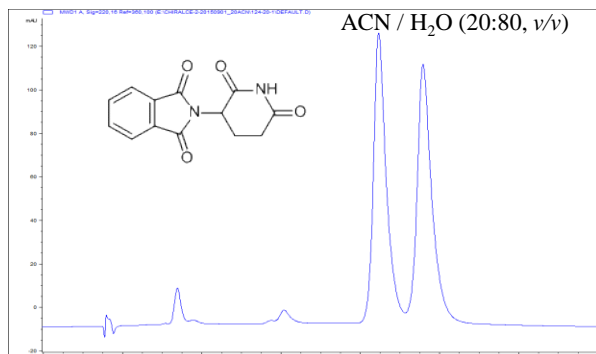


Figure 1-81. Chiral separation of Thalidomide on ChiralCE-2 column.

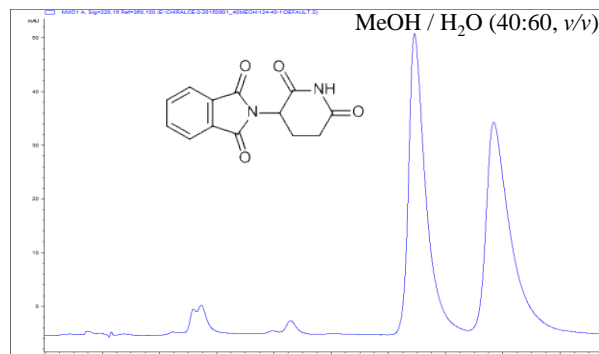


Figure 1-83. Chiral separation of Thalidomide on ChiralCE-2 column.

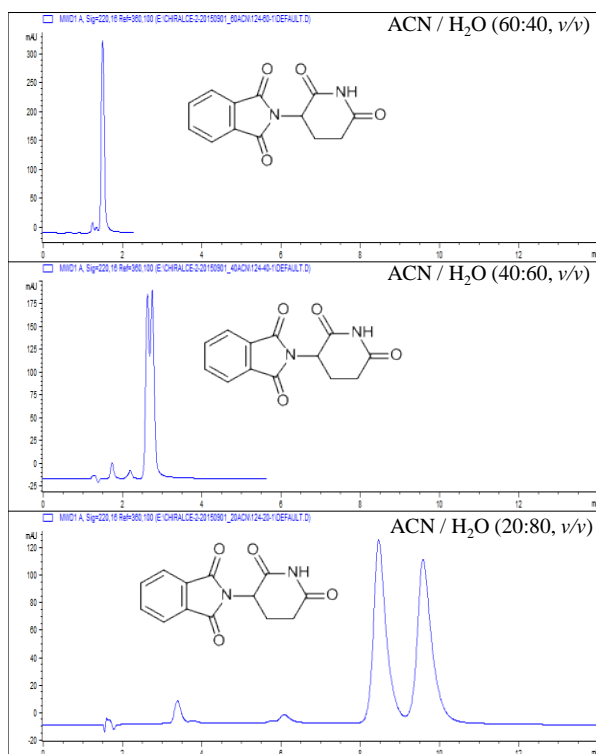


Figure 1-82. Progressive chiral separation of Thalidomide on ChiralCE-2 column as composition of mobile phase is varied.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.3 mL/min
Detection: UV@220nm
Temperature: Ambient

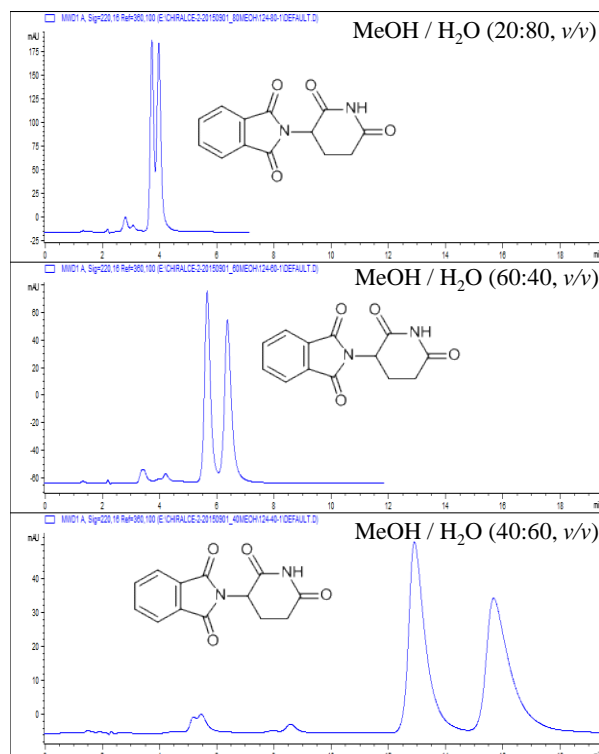


Figure 1-84. Progressive chiral separation of Thalidomide on ChiralCE-2 column as composition of mobile phase is varied.

Column: ChiralCE-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 813-CE2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.2 mL/min
Detection: UV@210nm
Temperature: Ambient

ChiralCD Column Notes and Applications in HPLC and UPLC

Introduction

ChiralCD columns are a series of new types of chemically-substituted cyclodextrin-bonded silica particles-packed chiral columns. The ChiralCD particles were synthesized by bonding a series of different functional groups-substituted α -, β -, or γ -cyclodextrins onto surface of high-quality porous silica (2 μm or 3 μm) gel particles by linking the spacer arms at the wider torus rim of the cyclodextrins as shown in the following Figure (I). Due to the multiple functional groups, e.g., halogen groups, hydroxyl groups, aromatic rings, and cyclodextrin moiety etc., available in the bonded stationary phases, the ChiralCD columns can be used under both normal phase and reversed-phase conditions.

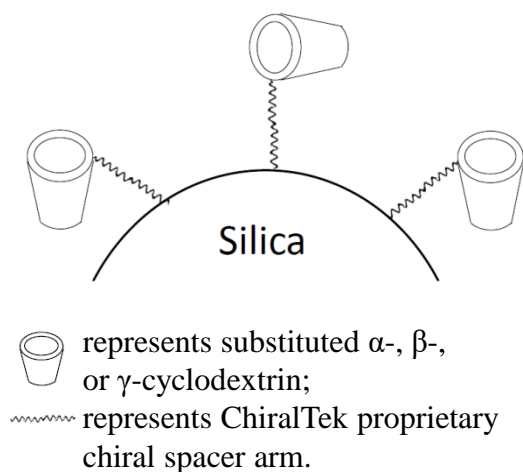


Figure (I). Schematic diagram of the bonded ChiralCD phase.

Currently, the other cyclodextrin-based chiral columns in the market are prepared by anchoring cyclodextrin through the more reactive primary hydroxyl groups at the narrow torus rim of cyclodextrins as shown in Figure (J). Since the ChiralCD columns are the first commercial available chiral columns by anchoring cyclodextrins through the less reactive secondary hydroxyl at the wider torus rim of the cyclodextrins, the ChiralTek proprietary chiral spacer arms can provide

extra unique steric interactions with solutes when entering the cyclodextrin cavities through the wider torus rim. Therefore, enhanced chiral selectivity can be easily achieved on the ChiralCD columns. Excellent separation selectivity can be obtained on the ChiralCD columns for separation a wide range of both chiral and non-chiral compounds under multiple modes of mobile phase conditions.

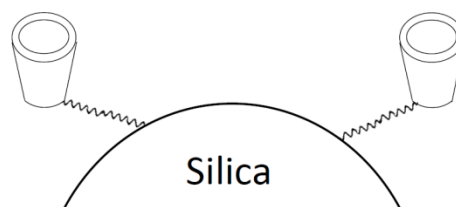


Figure (J). Schematic diagram of other supplier's cyclodextrin-bonded phase.

Since the ChiralCD phases from ChiralTek contain higher content of bonded cyclodextrins than other supplier's cyclodextrin-bonded phases, the ChiralCD columns can exhibit much higher sample capacity. Due to the different chemical structure of the substituted cyclodextrins in ChiralCD phases, the new ChiralCD columns can provide different and generally better separation abilities than other supplier's cyclodextrin-based columns.

Pre-processing and Conditioning

The ChiralCD columns were shipped with mixture of methanol/isopropanol. For use under reversed-phase conditions, the columns need to be firstly flushed with methanol following by mobile phase until reaching a constant column pressure. Similarly, for use under normal phase conditions, the columns need to be firstly flushed with isopropanol following by mobile phase until achieving a stable

ChiralCD Column Notes and Applications in HPLC and UPLC

baseline signal. A standard C18 guard column can be used for reversed-phase conditions and a Diol guard column can be used for reversed-phase conditions to protect ChiralCD column.

The ChiralCD columns can be used for both traditional HPLC and modern UPLC. However, since packing particles (2 μm or 3 μm) and the inner diameter (2 mm) of the ChiralCD columns are quite small, a low flow rate (e.g., 0.1 to 0.3 mL/min) should be applied when used in traditional HPLC with high viscous mobile phases in order to avoid extreme high back pressure. However, there is no flow rate limitation for use in UPLC.

Notes and Applications

The positional isomers of disubstituted benzenes are typical non-chiral compounds.

Mixture of *o*-Nitrophenol (2-Nitrophenol, *o*-NP), *m*-Nitrophenol (3-Nitrophenol, *m*-NP), *p*-Nitrophenol (4-Nitrophenol, *p*-NP), and/or mixture of *o*-Nitroaniline (2-Nitroaniline, *o*-NA), *m*-Nitroaniline (3-Nitroaniline, *m*-NA), *p*-Nitroaniline (4-Nitroaniline, *p*-NA) were used to evaluate of the performance of the ChiralCD columns for separation of non-chiral compounds.

A wide range of chiral drug compounds were used to evaluate the enantiomeric separation performance of the ChiralCD columns. Excellent enantiomeric separations were achieved for those testing chiral compounds.

The chromatographic separations were performed on Agilent 1100 HPLC-UV or Agilent 1290 UPLC-MS system under both normal and reversed phase conditions.

ChiralCD Column Notes and Applications in HPLC and UPLC

Disubstituted benzenes on ChiralCD columns

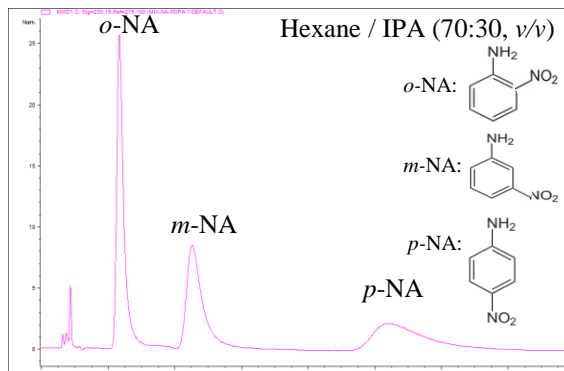


Figure 2-1. Positional isomers *o*-, *m*-, *p*-Nitroanilines on ChiralCD-1 column.

Column: ChiralCD-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.4 mL/min
Detection: UV@230nm
Temperature: Ambient

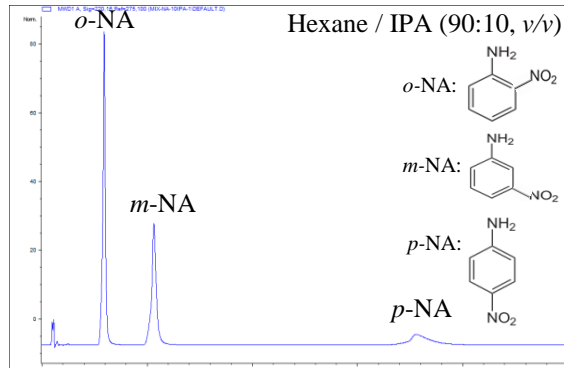


Figure 2-3. Positional isomers *o*-, *m*-, *p*-Nitroanilines on ChiralCD-2 column.

Column: ChiralCD-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@220nm
Temperature: Ambient

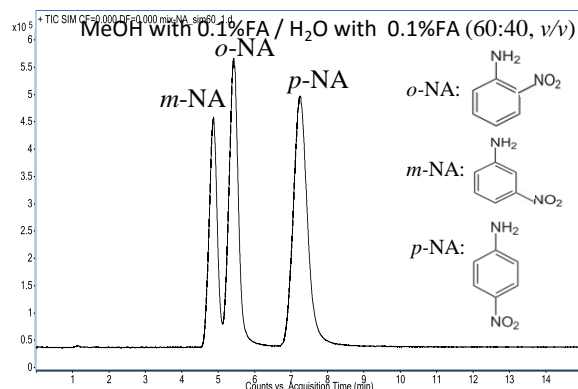


Figure 2-2. Positional isomers *o*-, *m*-, *p*-Nitroanilines on ChiralCD-1 column.

Column: ChiralCD-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD1-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.25 mL/min
Detection: MSD@SIM *m/z*
Temperature: Ambient

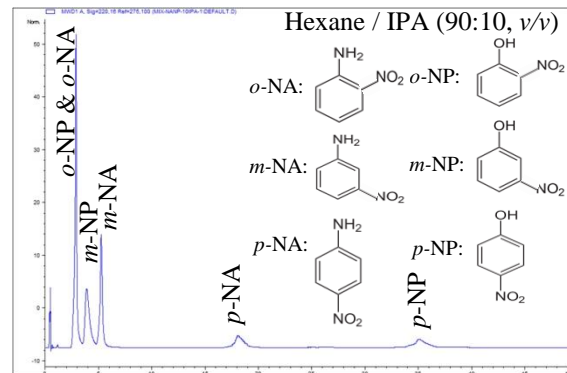


Figure 2-4. Mixture of positional isomers *o*-, *m*-, *p*-Nitrophenols and *o*-, *m*-, *p*-Nitroanilines on ChiralCD-2 column.

Column: ChiralCD-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@220nm
Temperature: Ambient

ChiralCD Column Notes and Applications in HPLC and UPLC

Disubstituted benzenes on ChiralCD columns

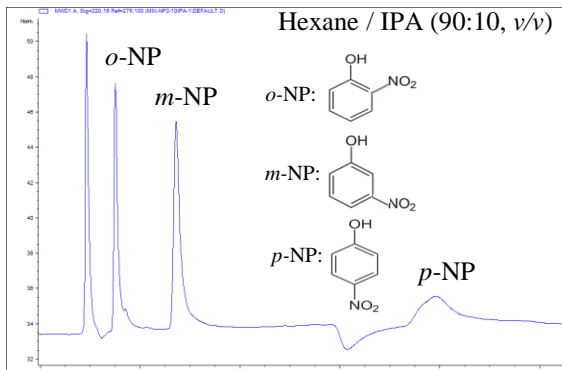


Figure 2-5. Positional isomers *o*-, *m*-, *p*-Nitrophenols on ChiralCD-3 column.

Column: ChiralCD-3
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD3-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@220nm
Temperature: Ambient

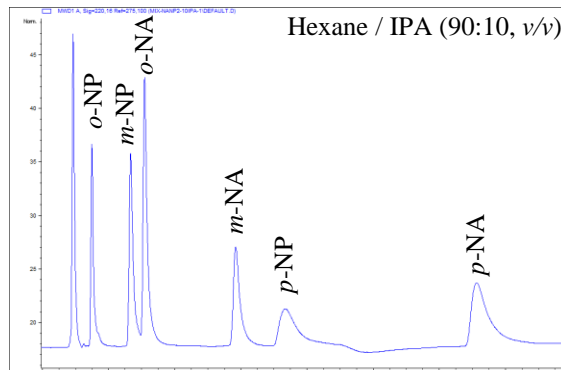


Figure 2-7. Mixture of positional isomers *o*-, *m*-, *p*-Nitrophenols and *o*-, *m*-, *p*-Nitroanilines on ChiralCD-3 column.

Column: ChiralCD-3
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD3-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@220nm
Temperature: Ambient

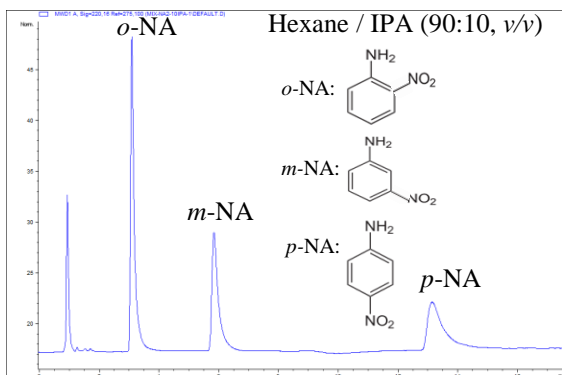


Figure 2-6. Positional isomers *o*-, *m*-, *p*-Nitroanilines on ChiralCD-3 column.

Column: ChiralCD-3
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD3-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@220nm
Temperature: Ambient

The elution order for the positional isomers of *o*-, *m*-, *p*-Nitroanilines under normal phase ($o > m > p$, as shown in Figure 2-1) is different from that under the reversed phase ($m > o > p$) as shown in Figure 2-2. This demonstrates that retention mechanisms under normal and reversed phase conditions are different for ChiralCD-1 column. This suggests the ChiralCD columns can provide different and complementary separation abilities under the normal and reversed phase conditions.

ChiralCD Column Notes and Applications in HPLC and UPLC

Flavonoids Flavanone on ChiralCD columns

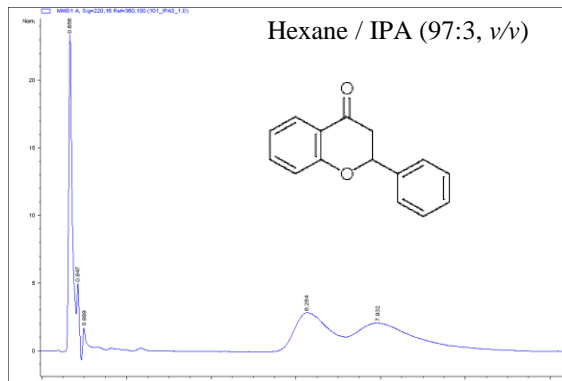


Figure 2-8. Chiral separation of Flavanone on ChiralCD-1 column.

Column: ChiralCD-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.6 mL/min
Detection: UV@220nm
Temperature: Ambient

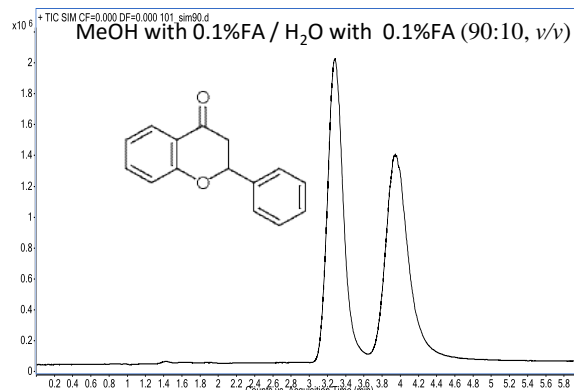


Figure 2-9. Chiral separation of Flavanone on ChiralCD-1 column.

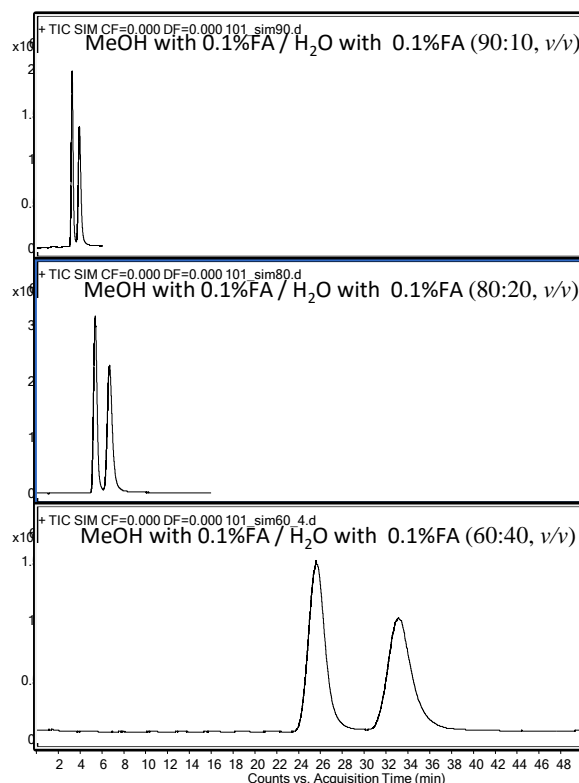


Figure 2-10. Progressive chiral separation of Flavanone on ChiralCD-1 column as composition of mobile phase is varied.

Column: ChiralCD-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD1-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.25 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

ChiralCD Column Notes and Applications in HPLC and UPLC

Flavonoids Flavanone on ChiralCD columns

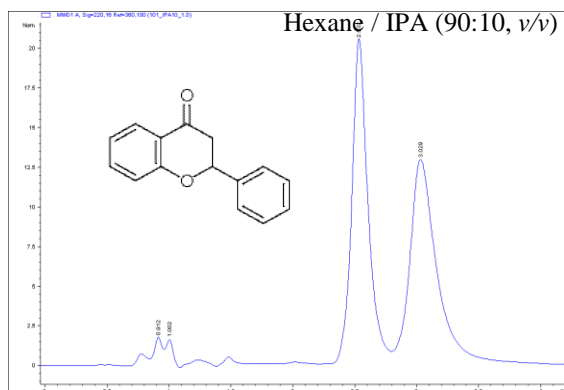


Figure 2-11. Chiral separation of Flavanone on ChiralCD-2 column.

Column: ChiralCD-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@220nm
Temperature: Ambient

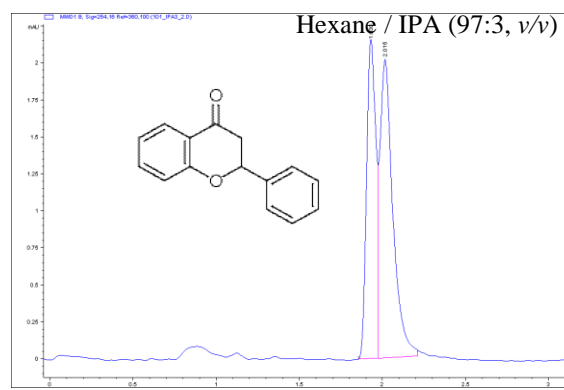


Figure 2-12. Chiral separation of Flavanone on ChiralCD-3 column.

Column: ChiralCD-3
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD3-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@254nm
Temperature: Ambient

ChiralCD Column Notes and Applications in HPLC and UPLC

Flavonoids Naringenin on ChiralCD columns

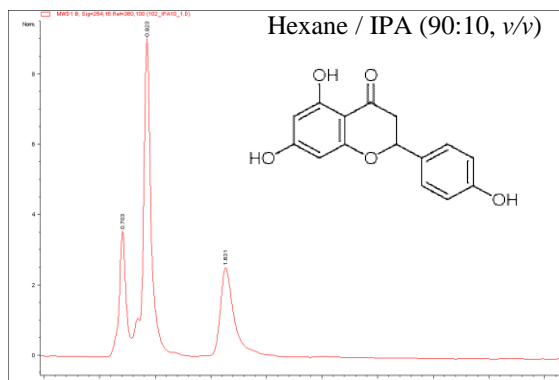


Figure 2-13. Chiral separation of Naringenin on ChiralCD-1 column.

Column: ChiralCD-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@254nm
Temperature: Ambient

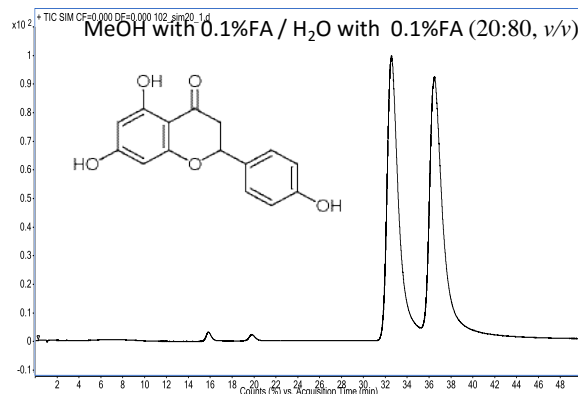


Figure 2-14. Chiral separation of Naringenin on ChiralCD-4 column.

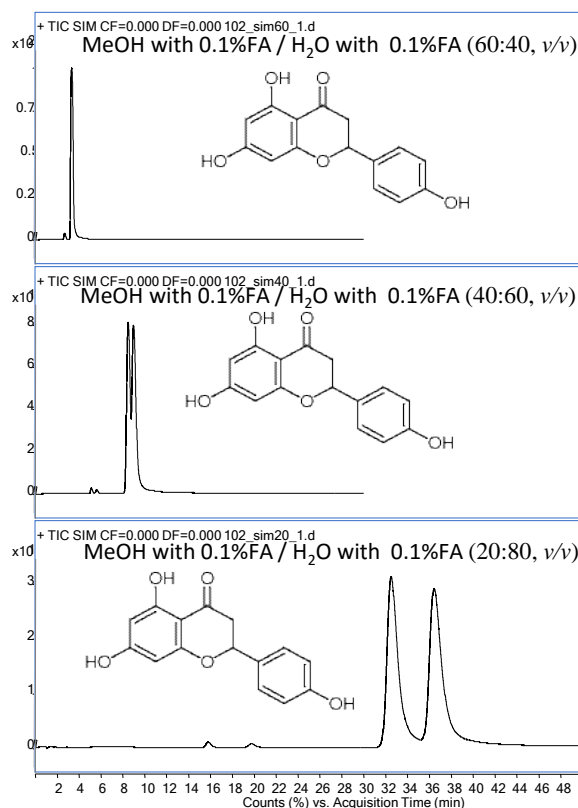


Figure 2-15. Progressive chiral separation of Naringenin on ChiralCD-4 column as composition of mobile phase is varied.

Column: ChiralCD-4
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD4-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.25 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

ChiralCD Column Notes and Applications in HPLC and UPLC

Flavonoids 4'-Hydroxyflavanone on ChiralCD columns

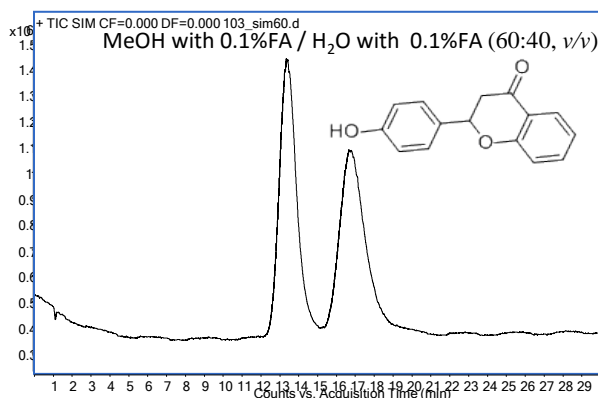


Figure 2-16. Chiral separation of 4'-Hydroxyflavanone on ChiralCD-1 column.

Column: ChiralCD-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD1-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.25 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

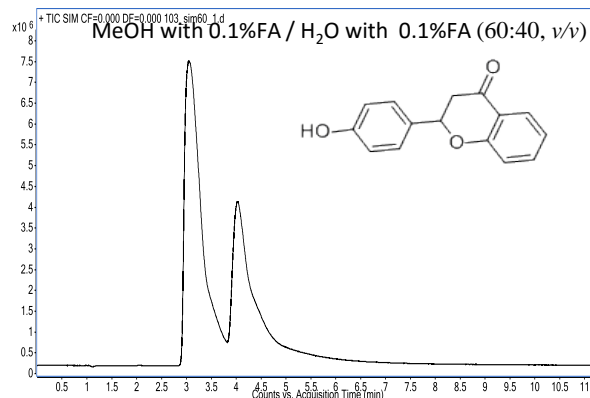


Figure 2-18. Chiral separation of 4'-Hydroxyflavanone on ChiralCD-3 column.

Column: ChiralCD-3
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD3-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.25 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

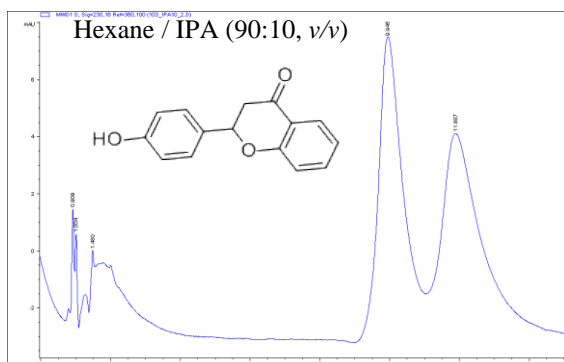


Figure 2-17. Chiral separation of 4'-Hydroxyflavanone on ChiralCD-2 column.

Column: ChiralCD-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@230nm
Temperature: Ambient

ChiralCD Column Notes and Applications in HPLC and UPLC

Flavonoids 4'-Hydroxyflavanone on ChiralCD columns

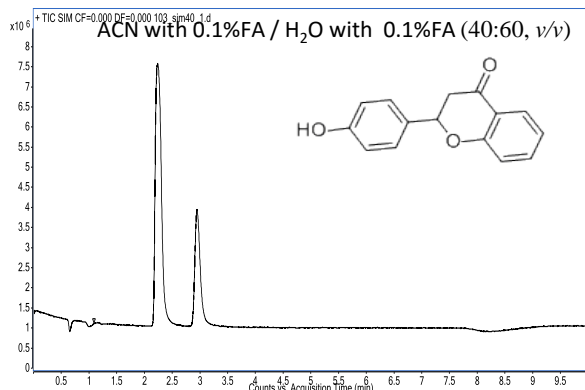


Figure 2-19. Chiral separation of 4'-Hydroxyflavanone on ChiralCD-4 column.

Column: ChiralCD-4
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD4-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.40 mL/min
Detection: MSD@SIM *m/z*
Temperature: Ambient

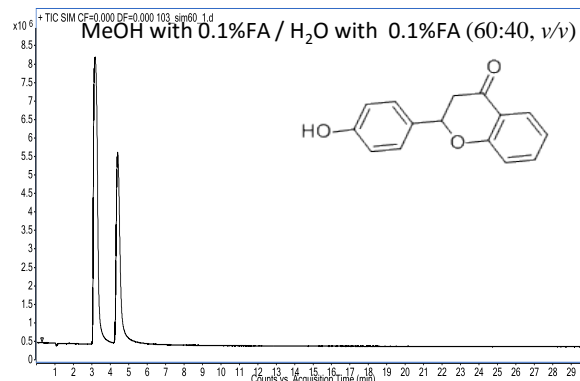


Figure 2-20. Chiral separation of 4'-Hydroxyflavanone on ChiralCD-4 column.

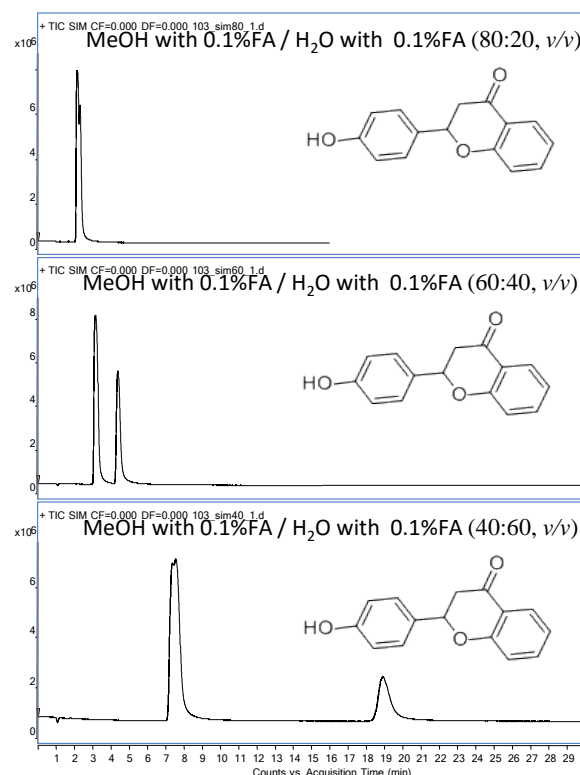


Figure 2-21. Progressive chiral separation of 4'-Hydroxyflavanone on ChiralCD-4 column as composition of mobile phase is varied.

Column: ChiralCD-4
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD4-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.25 mL/min
Detection: MSD@SIM *m/z*
Temperature: Ambient

ChiralCD Column Notes and Applications in HPLC and UPLC

Flavonoids Methoxyflavanone on ChiralCD columns

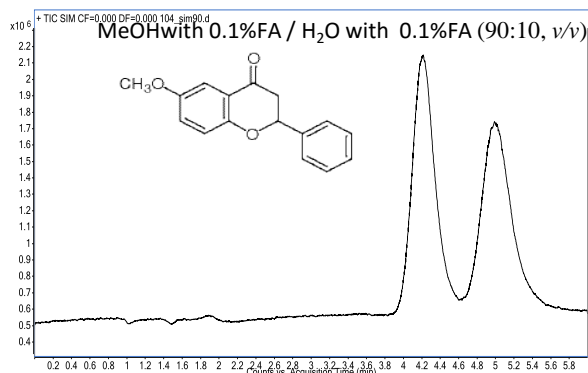


Figure 2-22. Chiral separation of Methoxyflavanone on ChiralCD-1 column.

Column: ChiralCD-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD1-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.25 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

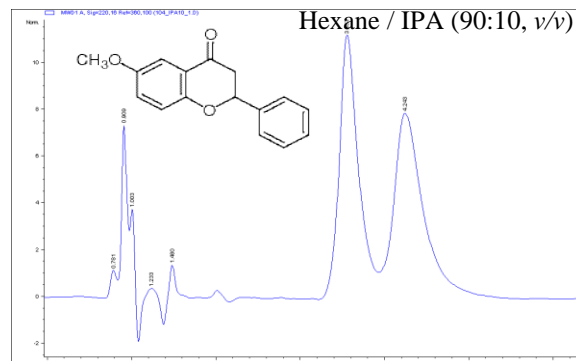


Figure 2-23. Chiral separation of Methoxyflavanone on ChiralCD-2 column.

Column: ChiralCD-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@220nm
Temperature: Ambient

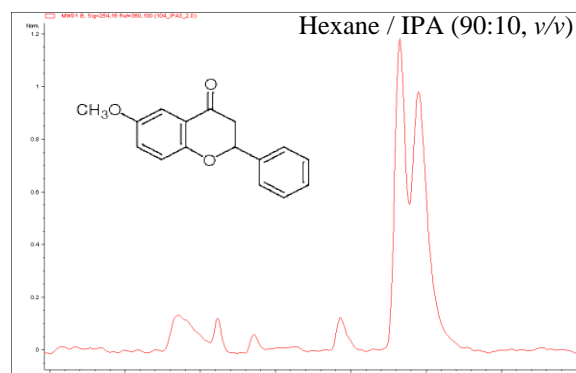


Figure 2-24. Chiral separation of Methoxyflavanone on ChiralCD-3 column.

Column: ChiralCD-3
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD3-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@254nm
Temperature: Ambient

ChiralCD Column Notes and Applications in HPLC and UPLC

4-Chromanol on ChiralCD columns

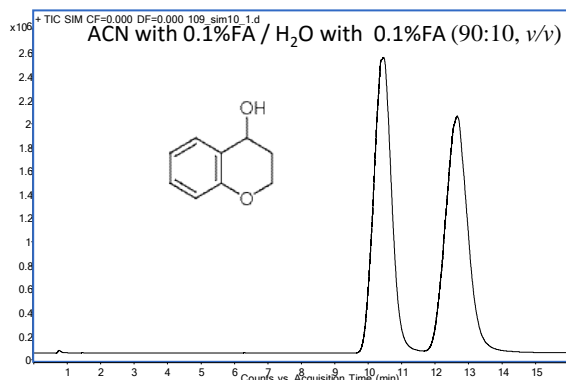


Figure 2-25. Chiral separation of 4-Chromanol on ChiralCD-1 column.

Column: ChiralCD-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD1-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.4 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

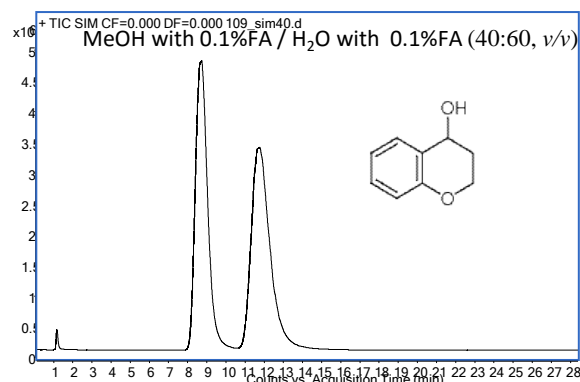


Figure 2-26. Chiral separation of 4-Chromanol on ChiralCD-1 column.

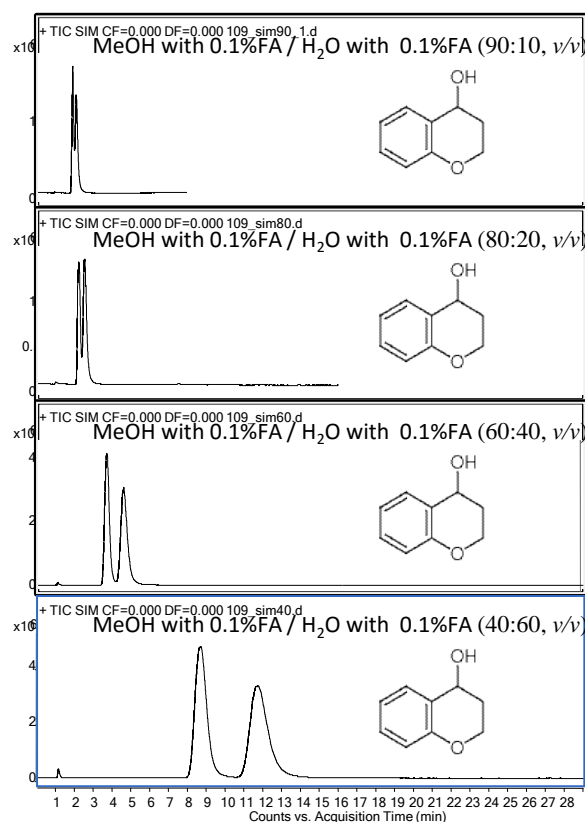


Figure 2-27. Progressive chiral separation of 4-Chromanol on ChiralCD-1 column as composition of mobile phase is varied.

Column: ChiralCD-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD1-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.25 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

ChiralCD Column Notes and Applications in HPLC and UPLC

4-Bromo- α -methylbenzyl Alcohol on ChiralCD columns

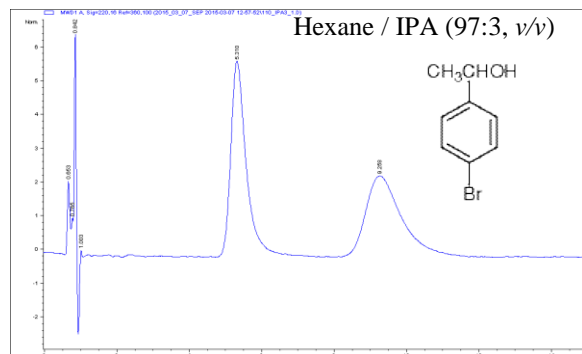


Figure 2-28. Chiral separation of 4-Bromo- α -methylbenzyl Alcohol on ChiralCD-1 column.

Column: ChiralCD-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.6 mL/min
Detection: UV@220nm
Temperature: Ambient

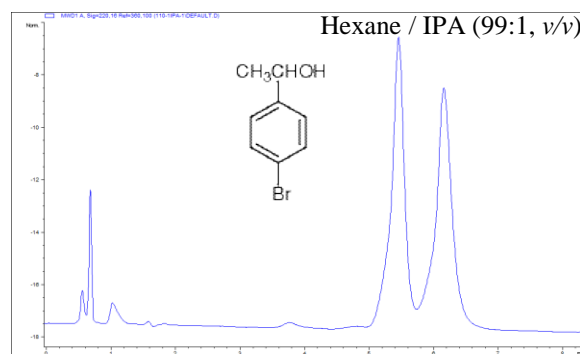


Figure 2-29. Chiral separation of 4-Bromo- α -methylbenzyl Alcohol on ChiralCD-2 column.

Column: ChiralCD-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.7 mL/min
Detection: UV@220nm
Temperature: Ambient

ChiralCD Column Notes and Applications in HPLC and UPLC

4-Chloro- α -methylbenzyl Alcohol on ChiralCD columns

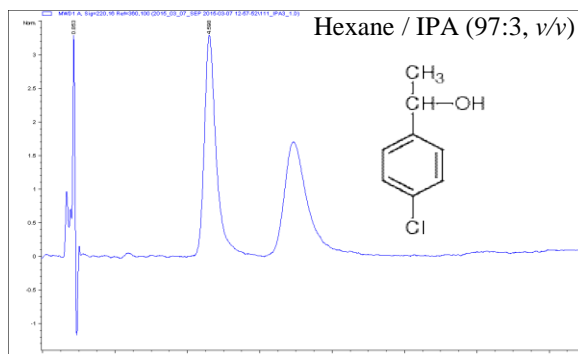


Figure 2-30. Chiral separation of 4-Chloro- α -methylbenzyl Alcohol on ChiralCD-1 column.

Column: ChiralCD-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.6 mL/min
Detection: UV@220nm
Temperature: Ambient

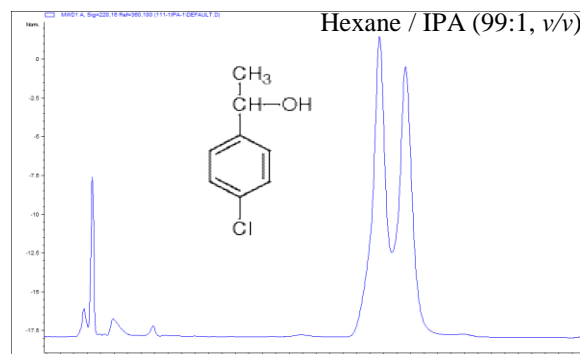


Figure 2-31. Chiral separation of 4-Chloro- α -methylbenzyl Alcohol on ChiralCD-2 column.

Column: ChiralCD-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.7 mL/min
Detection: UV@220nm
Temperature: Ambient

ChiralCD Column Notes and Applications in HPLC and UPLC

4-Chloro- α -methylbenzyl Alcohol on ChiralCD columns

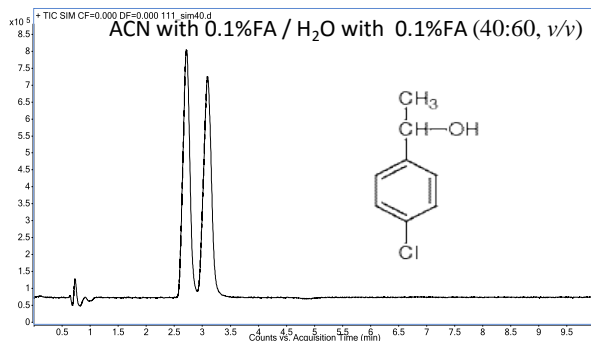


Figure 2-32. Chiral separation of 4-Chloro- α -methylbenzyl Alcohol on ChiralCD-1 column.

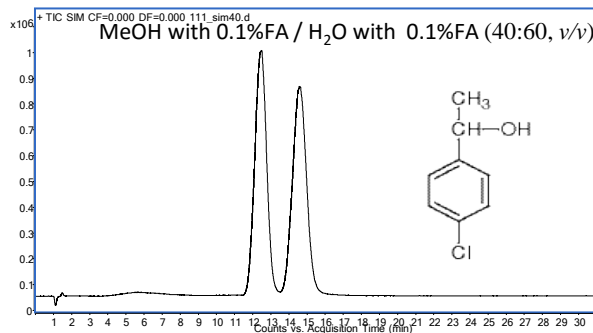


Figure 2-34. Chiral separation of 4-Chloro- α -methylbenzyl Alcohol on ChiralCD-1 column.

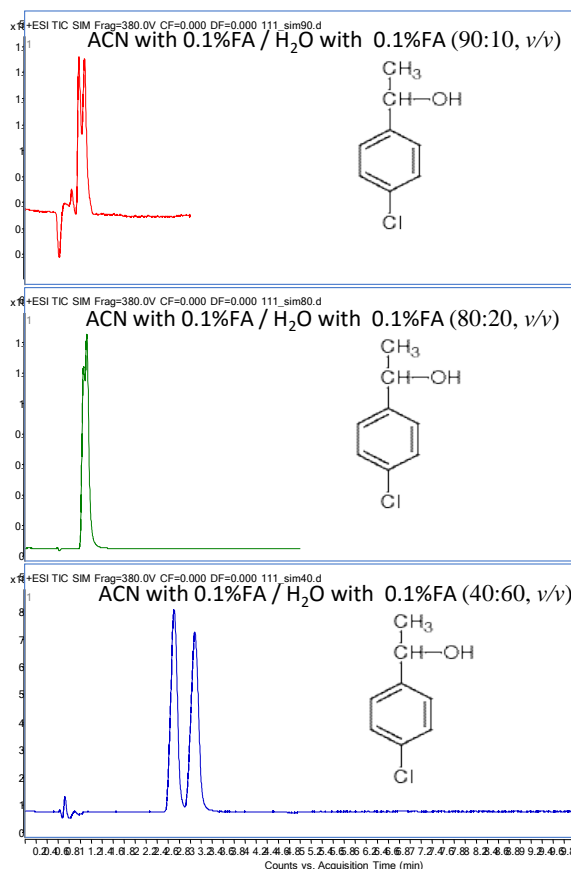


Figure 2-33. Progressive chiral separation of 4-Chloro- α -methylbenzyl Alcohol on ChiralCD-1 column as composition of mobile phase is varied.

Column: ChiralCD-1
 Dimension: 3 μ m, 150 \times 2 mm I.D.
 Part No. 823-CD1-03
 LC Mode: UPLC (Agilent1290)
 Flow rate: 0.4 mL/min
 Detection: MSD@SIM m/z
 Temperature: Ambient

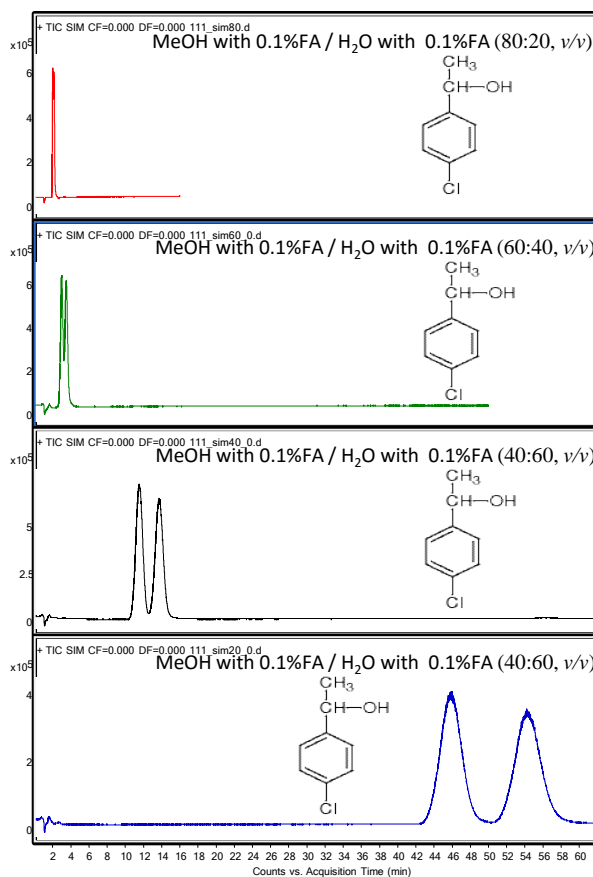


Figure 2-35. Progressive chiral separation of 4-Chloro- α -methylbenzyl Alcohol on ChiralCD-1 column as composition of mobile phase is varied.

Column: ChiralCD-1
 Dimension: 3 μ m, 150 \times 2 mm I.D.
 Part No. 823-CD1-03
 LC Mode: UPLC (Agilent1290)
 Flow rate: 0.25 mL/min
 Detection: MSD@SIM m/z
 Temperature: Ambient

ChiralCD Column Notes and Applications in HPLC and UPLC

Praziquantel on ChiralCD columns

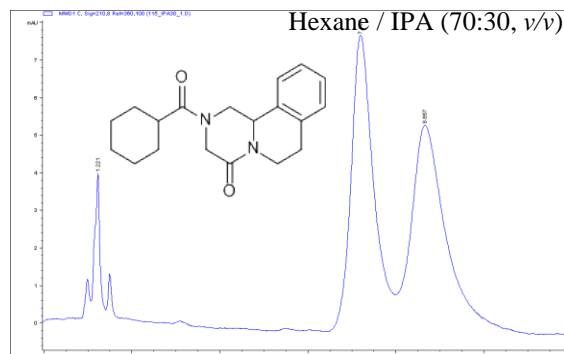


Figure 2-36. Chiral separation of Praziquantel on ChiralCD-2 column.

Column: ChiralCD-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.4 mL/min
Detection: UV@210nm
Temperature: Ambient

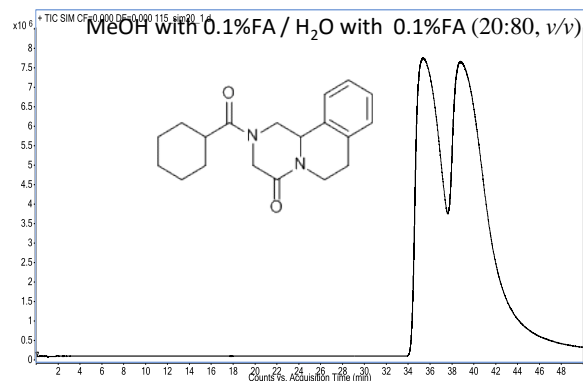


Figure 2-37. Chiral separation of Praziquantel on ChiralCD-4 column.

Column: ChiralCD-4
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD4-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.25 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

ChiralCD Column Notes and Applications in HPLC and UPLC

4-Phenyl-2-butanol on ChiralCD columns

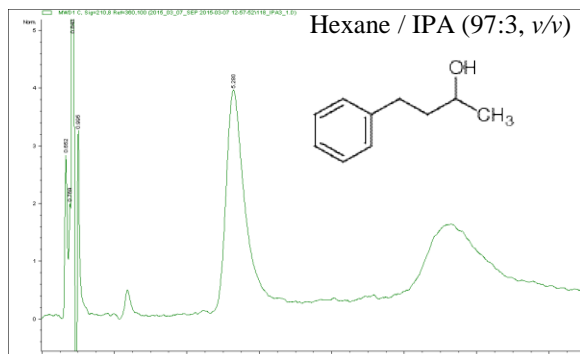


Figure 2-38. Chiral separation of 4-Phenyl-2-butanol on ChiralCD-1 column.

Column: ChiralCD-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.6 mL/min
Detection: UV@210nm
Temperature: Ambient

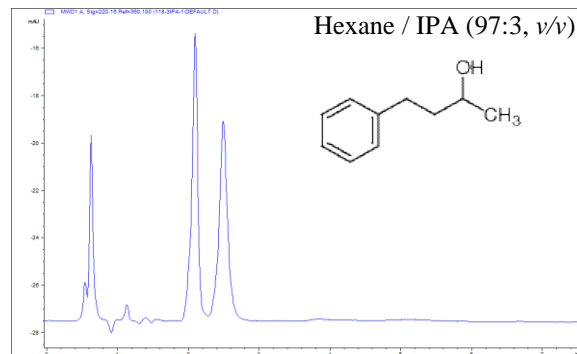


Figure 2-39. Chiral separation of 4-Phenyl-2-butanol on ChiralCD-2 column.

Column: ChiralCD-2
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD2-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.6 mL/min
Detection: UV@220nm
Temperature: Ambient

ChiralCD Column Notes and Applications in HPLC and UPLC

Thalidomide on ChiralCD columns

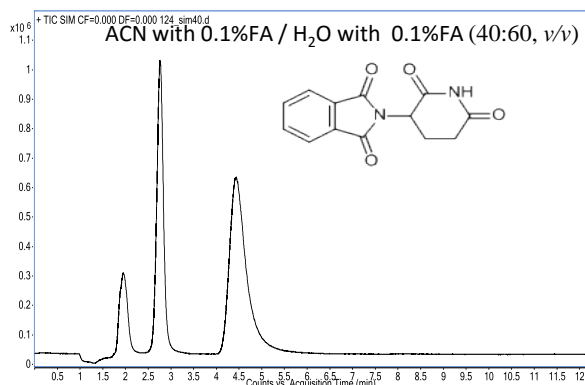


Figure 2-40. Chiral separation of Thalidomide on ChiralCD-1 column.

Column: ChiralCD-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD1-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.4 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

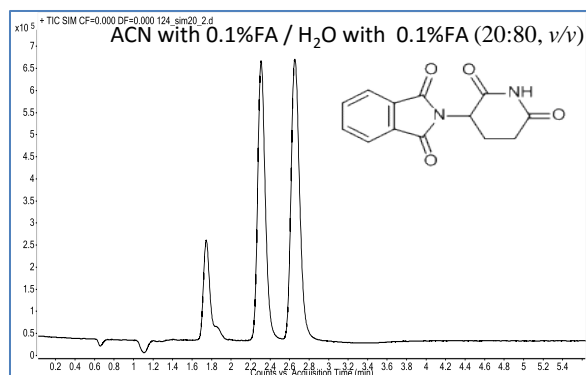


Figure 2-41. Chiral separation of Thalidomide on ChiralCD-4 column.

Column: ChiralCD-4
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD4-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.4 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

ChiralCD Column Notes and Applications in HPLC and UPLC

Thalidomide on ChiralCD columns

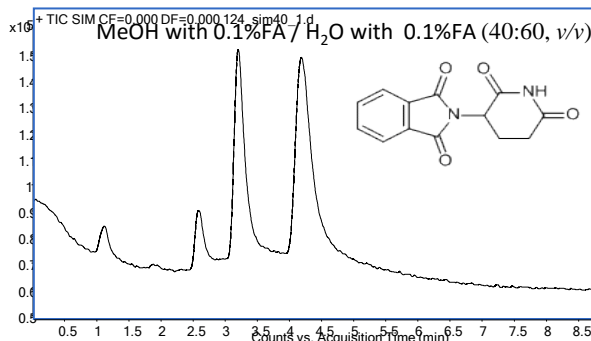


Figure 2-42. Chiral separation of Thalidomide on ChiralCD-3 column.

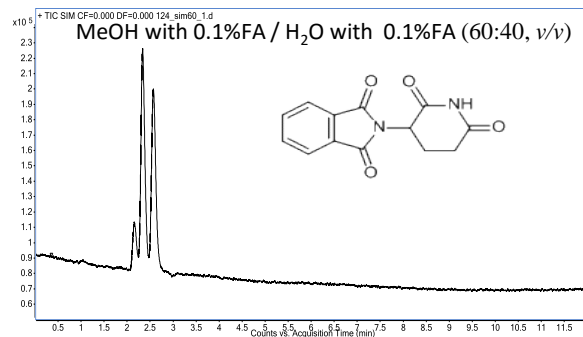


Figure 2-44. Chiral separation of Thalidomide on ChiralCD-4 column.

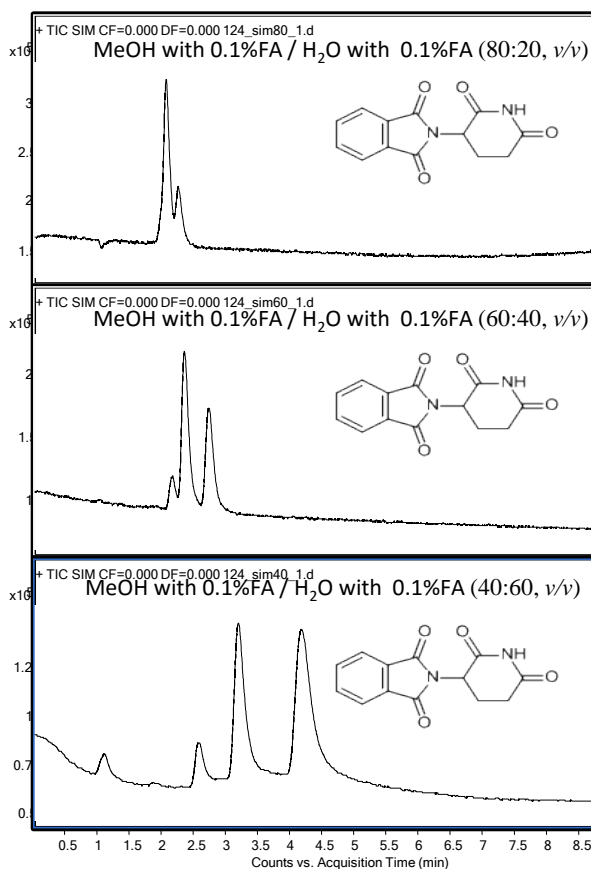


Figure 2-43. Progressive chiral separation of Thalidomide on ChiralCD-3 column as composition of mobile phase is varied.

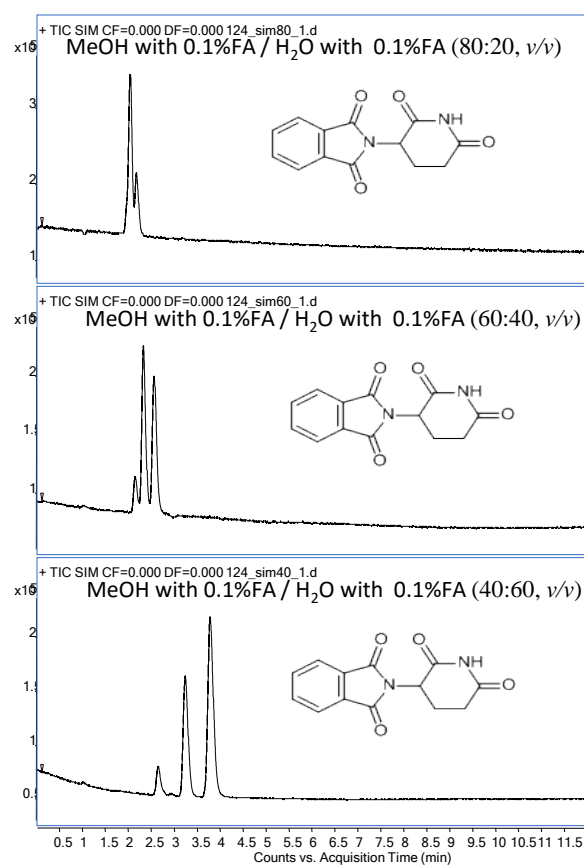


Figure 2-45. Progressive chiral separation of Thalidomide on ChiralCD-4 column as composition of mobile phase is varied.

Column: ChiralCD-3
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD3-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.25 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

Column: ChiralCD-4
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD4-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.25 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

ChiralCD Column Notes and Applications in HPLC and UPLC

Trans-Stilbene Oxide and Lansoprazole on ChiralCD columns

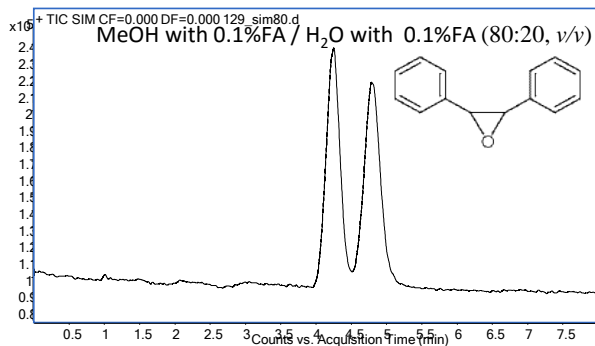


Figure 2-46. Chiral separation of trans-Stilbene Oxide on ChiralCD-1 column.

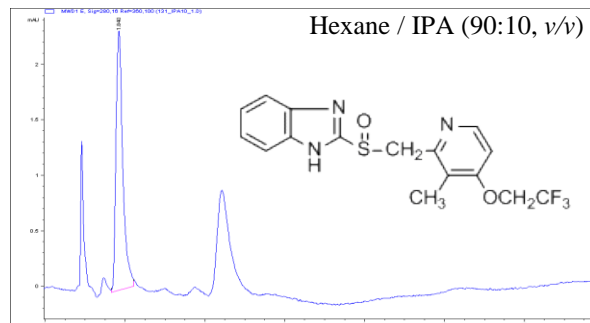


Figure 2-48. Chiral separation of Lansoprazole on ChiralCD-4 column.

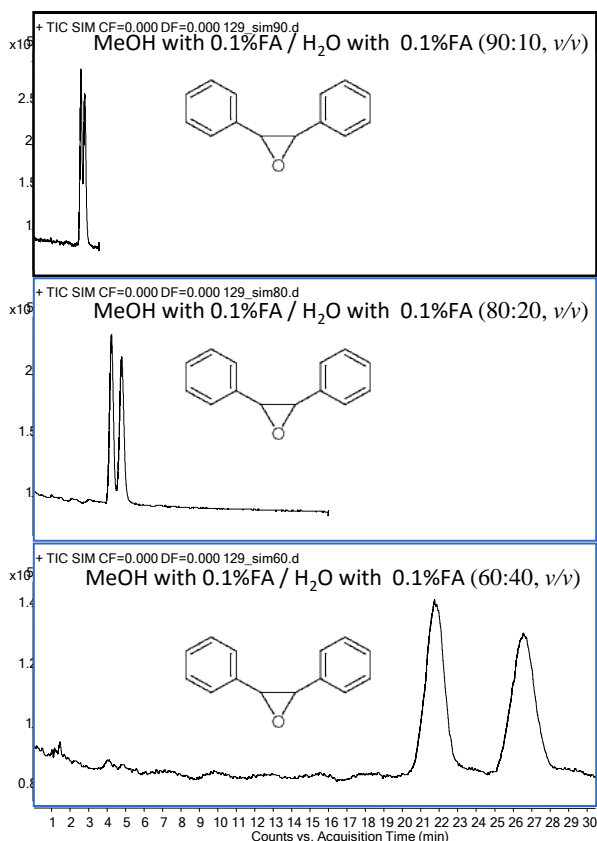


Figure 2-47. Progressive chiral separation of trans-Stilbene Oxide on ChiralCD-1 column as composition of mobile phase is varied.

Column: ChiralCD-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD1-03
LC Mode: UPLC (Agilent1290)
Flow rate: 0.25 mL/min
Detection: MSD@SIM m/z
Temperature: Ambient

Column: ChiralCD-4
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD4-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@280nm
Temperature: Ambient

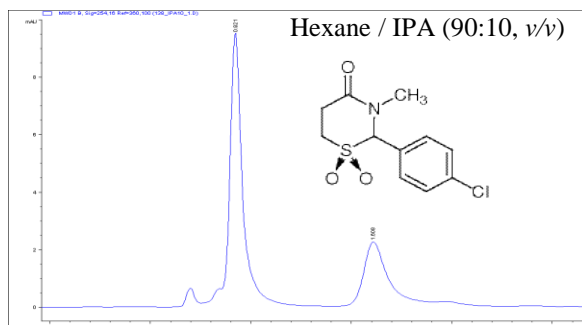
ChiralCD Column Notes and Applications in HPLC and UPLC**Chlormezanone and Flurbiprofen on ChiralCD columns**

Figure 2-49. Chiral separation of Chlormezanone on ChiralCD-1 column.

Column: ChiralCD-1
Dimension: 3 μ m, 150 \times 2 mm I.D.
Part No. 823-CD1-03
LC Mode: HPLC (Agilent1100)
Flow rate: 0.5 mL/min
Detection: UV@254nm
Temperature: Ambient