MERCK ChiraDex®

Specially for the separation of enantiomers

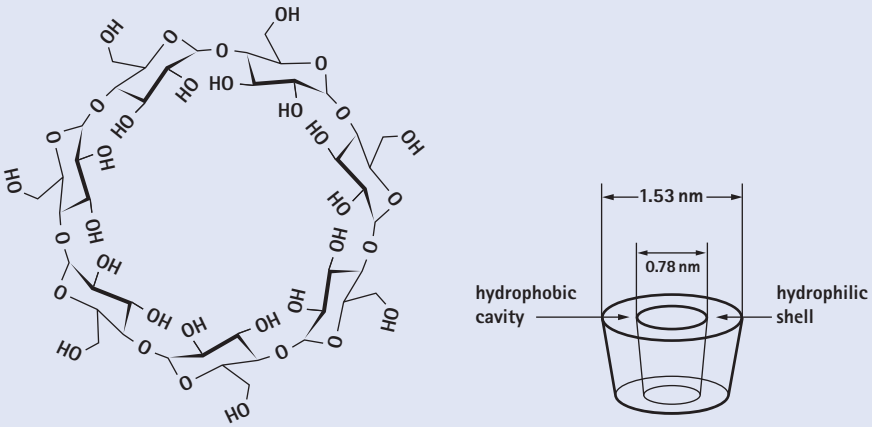
ChiraDex® is a versatile HPLC column characterized by broad enantioselectivity and can be used for the separation of enantiomers of numerous different classes of substances. ChiraDex® is based on beta-cyclodextrin covalently linked to spherical particles of silica and is well suited for the chiral separation of hydrocarbons, steroids, phenol esters and derivatives, aromatic amines, heterocycles with 5-membered ring to 7-membered ring. Simply composed RP-eluents can be used in most separations.

Characterization of ChiraDex®

ChiraDex® is characterized by broad enantioselectivity and can be used for the separation of enantiomers of numerous different classes of substances. Cyclodextrins are cyclic oligosaccharins consisting of α-1,4-glycosidically linked D-glucose units. β-cyclodextrin consist of 7 glucose units, respectively. Geometrically seen, cyclodextrins may be described as truncated cones, where all the secondary hydroxy groups are directed towards the larger opening, whereas the smaller opening at the other end is formed by primary hydroxy groups.

Thus, a hydrophobic inner cavity results, contrasting with the two hydrophilic openings. Since cyclodextrins are made up of chiral D-glucose units, its structure may be regarded as a chiral selector. The enantiomers of a racemic substance mixture, due to their opposite configurations, can now be associated – to different degrees – with the cyclodextrin molecule. Thus, diastereomeric "inclusion complexes" are formed, based on hydrophobic interaction (between cavity and guest molecule) and stereo selective hydrogen bonds (between the C2 and C3 hydrogen groups of glucose molecules and the guest molecule).

β-cyclodextrin



Specifications of ChiraDex®

Sorbent characteristics	Spherical silica particles with covalently bonded beta-cyclodextrin particles
Particle shape	spherical
Particle size	5 µm
Efficiency	>25 000 N/m
HighResolution	>37 000 N/m
Pore size	10 nm (100 Å)
Spec. surface area	300 – 360 m^2/g
Chiral selector	Beta-cyclodextrin
pH range	pH 3 – 7.5
Shipping eluent	Methanol/Water

Accessories for particulate HPLC columns:

- **manu-CART® cartridge holder** for LiChroCART® cartridges
  page 370
- **LiChroCART® cartridge** Different lengths, different internal diameter
  page 373

Ordering information – ChiraDex®, stainless steel columns Hibar®

Product	Ordering No.	Particle size	Dimension length	Dimension i.d.	Contents of one package
ChiraDex®	1.50013.7004	5 µm	100 mm	4.6 mm	1 piece

The Hibar® columns are complete with endfittings. When using a guard column with a Hibar® column, we recommend part number 1.51487.0001 guard column cartridge holder for 4–4 mm guard column cartridges LiChroCART®. Additional dimensions available as customized packings see page 366.

Ordering information – ChiraDex®, stainless steel cartridges LiChroCART®

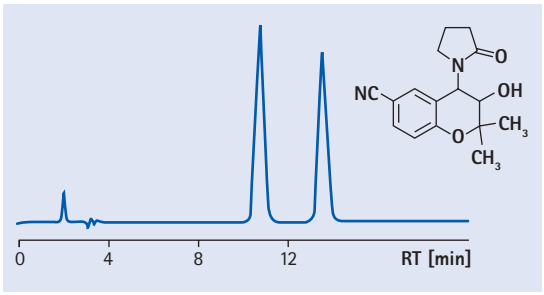
Product	Ordering No.	Particle size	Dimension length	Dimension i.d.	Contents of one package
ChiraDex®	1.50117.0001	5 µm	4 mm	4 mm	10 pieces
ChiraDex®	1.51333.0001	5 µm	250 mm	4 mm	1 piece
ChiraDex® HighResolution	1.51000.0001	5 µm	250 mm	4 mm	1 piece

The LiChroCART® columns in the list above require part number 1.51486.0001 manu-CART® cartridge column holder, which can be used to hold one cartridge column with or without a 4–4 mm guard column.

Separation examples of chiral pharmaceutical active ingredients on ChiraDex®

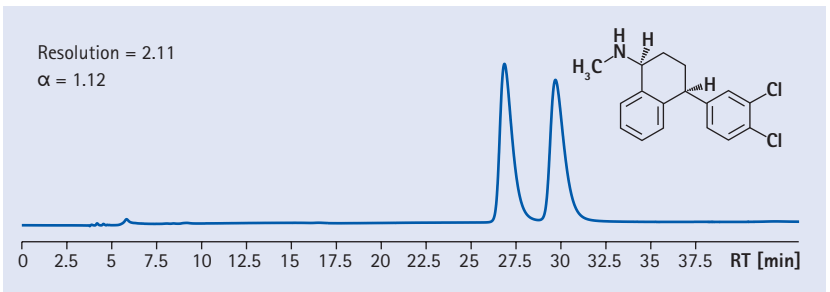
Cromakalim

Column	LiChroCART® 250-4 ChiraDex®
Mobile phase	Water/Methanol 80/20 (v/v)
Flow rate	0.8 mL/min
Detection	UV 254 nm



Selevtivity with Sertralin

Column	LiChroCART® 250-4 ChiraDex® HighResolution
Mobile phase	Acetonitril/10 mM Phosphat buffer adjusted with Triethylamin to pH=7.0 30/70 v/v
Flow rate	0.6 mL/min
Detection	UV 220 nm
Injection volume	5 µL
Sample	1 mg/mL Sertralin



Supelco®

1.51000.0001
1.51333.0001

LiChroCART®
LiChroCART® 250-4 ChiraDex® (5 µm)

ChiraDex® is a stationary phase based on spherical particles of silica gel with β-cyclodextrin covalently bonded. Cyclodextrins are naturally occuring oligosaccharides with hydrophobic cavities, which enable them to form inclusion complexes with organic substances in aqueous solutions. These properties make β-cylodextrin an ideal stationary phase in chromatography. As β-cyclodextrin is built from chiral glucose units, it is possible to use β-cyclodextrin as a chiral selector in the separation of enantiomers.

Specifications

Column: LiChroCART® 250-4 ChiraDex®
Particle size: 5 µm
Pore diameter: 100 Å
Specific surface area: 300–360 m2/g
Optimal flow rate: 0.8 –1.0 mL/min
pH-stability: pH 3.0–7.5

Column equilibration

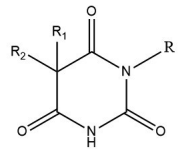
LiChroCART® 250-4 ChiraDex® is supplied in methanol/water (40/60) (v/v) and should be stored in the same solvent mixture. Before use, methanol/water (50/50) (v/v) should be pumped through the column for 30–60 minutes with a flow rate of 0.8 mL/min.

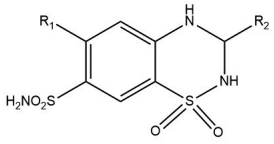
Conditions for chromatographic separations

For separations of enantiomers with ChiraDex®, mixtures of methanol and water or buffer (methanol/water or buffer: 5/95–60/40) (v/v) have been proven as well suited. The organic solvent plays an important role on enantioselectivity. The higher the polarity of the organic solvent (methanol > ethanol > acetonitrile), the better is the separation of enantiomers. The separation of optical isomers can be improved by using buffers (especially Na_2HPO_4/NaH_2PO_4) or 0.1%TEAA (triethyl amine acetate). The separation of enantiomers is depen-dent on the concentration of the buffer and on pH. The enantioselec-tivity of ChiraDex® can be altered by varying the temperature.

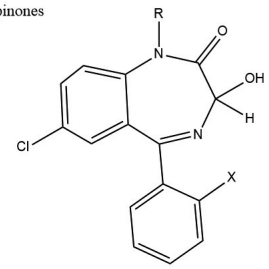
Racemates, that had been separated with ChiraDex®:

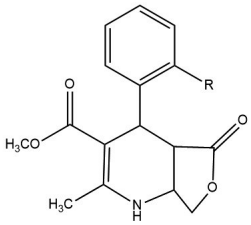
Barbiturates



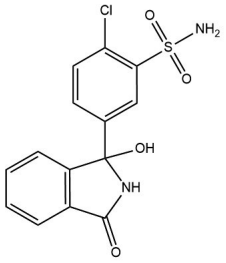


Benzodiazepinones

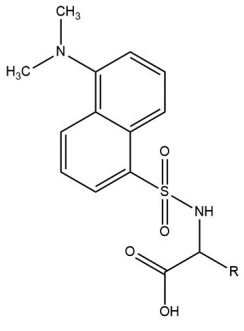




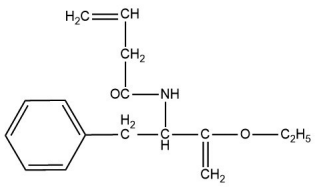
Chlorthalidone



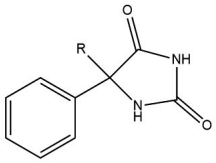
Dansylamino Acids



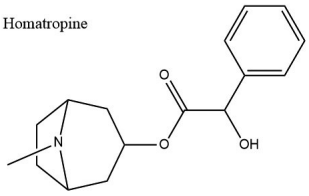
N-Acryloylphenylalanineethyl ester



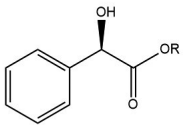
Hydantoin



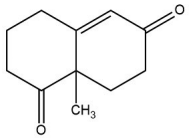
Homatropine



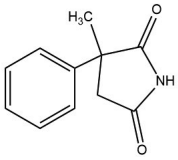
Mandelic Acid Esters



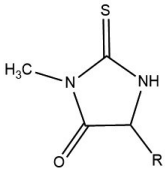
8-alpha-Methyl-1,2,3,4,7,8-alpha-octahydronaphthalene-1,6-dion



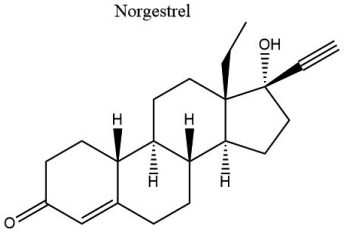
alpha-Methyl-alpha-phenylsuccinimide



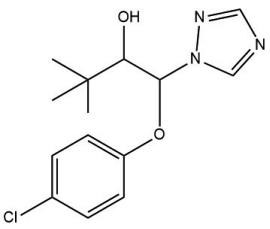
Methylthiohydantoinamino acids (MTH-amino acids)



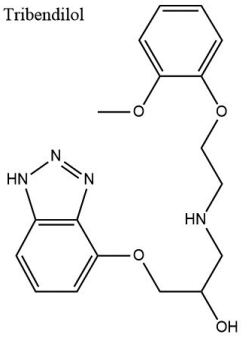
Norgestrel



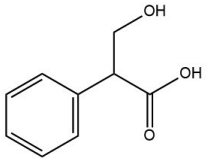
Triadimenol



Tribendilol



Tropic Acid



Handling of LiChroCART® HPLC cartridges

LiChroCART® cartridges are used with re-usable endfittings (manuCART®) which fit different cartridge lengths (75 mm–250 mm) and inner diameters(4.6 mm, 4 mm, 3 mm or 2 mm).
The endfittings (manuCART®) are designed to allow the cartridges to be hand-sealed at normal working pressures of 150 to 200 bar without the need for any tools. Only at higher pressures may further tightening with a wrench become necessary.

CARTRIDGE EQUILIBRATION

LiChroCART® cartridges were flushed with acetonitrile/water.
As they can dry out during stockings and shipping they have to be activated by purging 10–20 column volumes of pure organic component (e.g. acetonitrile, methanol) before equilibrating them with the mobile phase. Please increase the flow rate gradually stepwise from 0 mL/min to the desired value. If the eluent contains a buffer salt, you have to flush the cartridges with water after the activation step and before the equilibration step.

Mounting the manuCART® to a LiChroCART® cartridge without precolumn

The manuCART® (Cat. No. 1.51486.0001) consists of the following pieces. Check that you have all of these pieces before you begin to assemble your column.

No.	Description	Quantity
1	Cap nut	2
2	Spin collet	4
3	End fitting	2

The following procedure describes how to fit the pieces to just one end of your cartridge. Repeat steps 1–4 for the other end.

1. Insert the cartridge through the end fitting, so that the outer thread is at the outermost end.
2. Place 2 split collets around the neck of the cartridge as shown in Figure 1.
3. Push the end fitting up over the 2 split collets.
4. Screw the end cap over the assembled endfitting. Tighten finger tight.
5. When you install your column into the flow path of your LC, tighten the end fittings leak tight using wrenches.

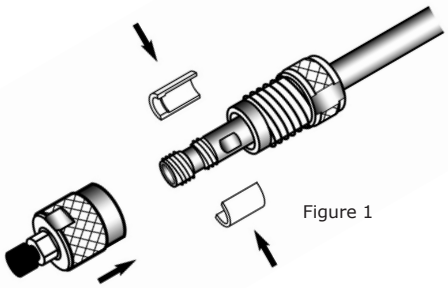


Figure 1

Mounting the manuCART® to a LiChroCART® cartridge precolumn

1. Insert the cartridge through the end fitting, so that the outer thread is at the outermost end.
2. Place 2 split collets around the neck of the cartridge as shown in Figure 2.
3. Place the guard column cartridge inside the space at the end of the collets.
4. Push the end fitting up over the assembled guard column cartridge.
5. Screw the end cap over the end fitting. Tighten finger tight.
6. When you install your combined column and guard column into the flow path of your LC, tighten the end fittings leak-tight using wrenches.

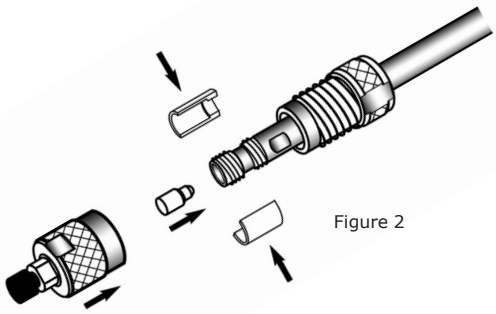


Figure 2

Exchanging the sieve and glass fiber filter

Content of the tool for replacement of frits (Cat. No. 1.15576.0001):
1 pin, 1 plastic tool, 1 centering sleeve

1. Using a wrench, loosen the cap nut from the end fitting.
2. Unscrew the cap nut, and remove the semicylindrical collets from the neck of the column cartridge. Put them aside for re-use later.
3. Using the pin, gently remove the sieve from the end of the column cartridge (Figure 3).
4. Using a small spatula, remove the remains of the glass fiber filter and any soiled packing material.
5. Fill the void with freshly prepared packing material and smooth-off the surface.
6. Place a new filter on the open end of the cartridge. Do not attempt to push it into place with your fingers – it may wrinkle.
7. Using the plastic tool, push the filter inside the cartridge (Figure 4).
8. Place the cylindrical sleeve over the open end of the cartridge.
9. Place a sieve inside the funnel.
10. Using the plastic tool, push the sieve firmly into place (Figure 5).

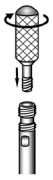


Figure 3

Figure 4

Figure 5

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Made in Germany

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