



HICHROM

Chromatography Columns and Supplies

LC COLUMNS Chiral Technologies

Catalogue 9

Hichrom Limited

1 The Markham Centre, Station Road
Theale, Reading, Berks, RG7 4PE, UK

Tel: +44 (0)118 930 3660 Fax: +44 (0)118 932 3484

Email: sales@hichrom.co.uk www.hichrom.co.uk

- Excellent resolution of racemates
- Well established immobilized and coated polysaccharide phases
- HPLC and SFC columns
- Wide range of applications

Daicel chiral HPLC columns are the most widely referenced chiral columns. Chiral Technologies, a subsidiary of Daicel Corporation, offers the complete range of Daicel chiral columns and has the largest portfolio of chiral stationary phases for the separation of racemic mixtures. This includes the CHIRALPAK®, CHIRALCEL® and CROWNPAK® trademarks. Columns from microbore to preparative dimensions can be supplied, allowing smooth transition from laboratory to development, to pilot plant and production. In addition to the well established traditional CHIRALPAK and CHIRALCEL polysaccharide coated silica columns, Daicel manufacture the immobilized polysaccharide CHIRALPAK phases. Also see page 5 for new CHIRALPAK ZWIX zwitterionic chiral phases.

CHIRALPAK® IA, IB, IC, ID, IE and IF Immobilized Phases

- Immobilized chiral selector
- Broad application range
- Universal solvent compatibility – phases suitable for both NP and RP applications
- Higher screening success rate
- Robustness and extended durability

CHIRALPAK IA, IB, IC, ID, IE and IF HPLC and SFC columns are a newer generation of chiral stationary phases (CSPs), in which the polysaccharide chiral selector has been immobilized on to a wide pore silica matrix. This confers universal solvent compatibility on these highly selective chiral stationary phases, without compromising phase stability. The broader range of solvents used as eluents, introduces new selectivity profiles, improved productivity and additional robustness compared with traditional coated polysaccharide phases.

Chiral Selectors

CHIRALPAK IA - based on amylose *tris*(3,5-dimethylphenyl)carbamate (as in CHIRALPAK AD)

CHIRALPAK IB - based on cellulose *tris*(3,5-dimethylphenyl)carbamate (as in CHIRALCEL OD)

CHIRALPAK IC - based on cellulose *tris*(3,5-dichlorophenyl)carbamate

CHIRALPAK ID - based on amylose *tris*(3-chlorophenyl)carbamate

CHIRALPAK IE – based on amylose *tris*(3,5-dichlorophenyl)carbamate

CHIRALPAK IF – based on amylose *tris*(3-chloro, 4-methylphenyl)carbamate

Figures 1, 2 and 3 show examples of separations achieved on CHIRALPAK IA, IB and IC respectively. Figure 4 shows the baseline resolution of enantiomers and conformers of diastereomeric tofisopam on CHIRALPAK ID.

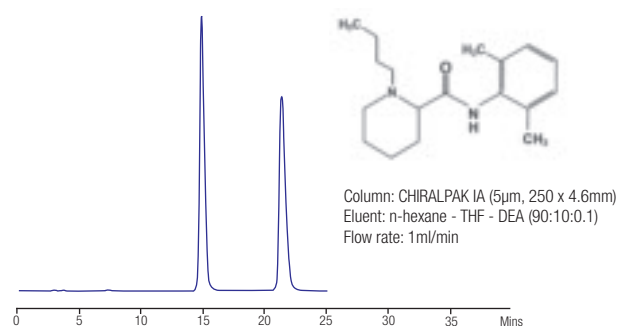


Figure 1. Analysis of bupivacaine on CHIRALPAK IA

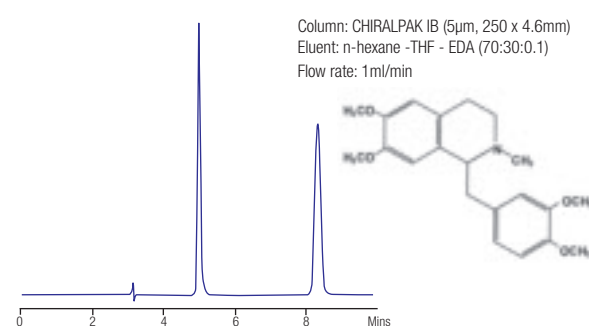


Figure 2. Analysis of laudanosine on CHIRALPAK IB

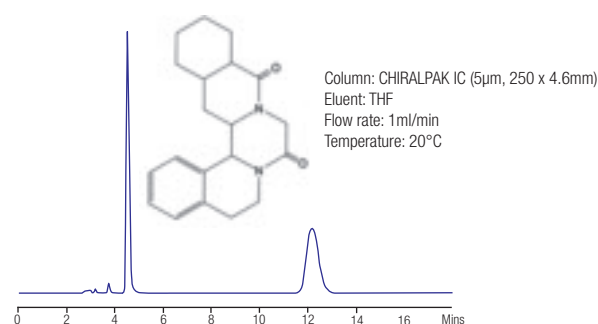


Figure 3. Analysis of praziquantel on CHIRALPAK IC

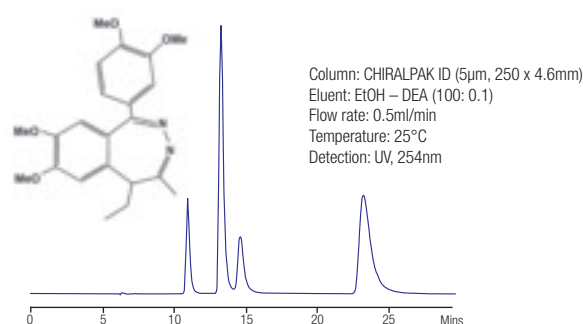


Figure 4. Analysis of tofisopam on CHIRALPAK ID

CHIRALPAK® Immobilized Phases (continued)

3µm Fast Analysis Immobilized Phase Columns

The immobilized product line has been extended by the introduction of 3µm versions of the CHIRALPAK IA, IB, IC, ID, IE and IF phases (CHIRALPAK IA-3, IB-3, IC-3, ID-3, IE-3 and IF-3). These columns are intended for high speed, high efficiency separations of enantiomers and offer the same selectivity and stability characteristics as their 5µm immobilized counterparts. Methods can be transferred directly from 5µm to 3µm particle size columns. Figure 5 shows the fast separation of ornidazole on CHIRALPAK IA-3 in less than one minute.

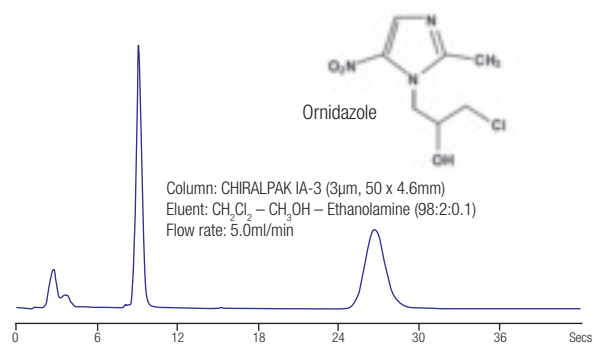


Figure 5. Fast separation on CHIRALPAK IA-3

Features of Immobilized Phases

1) Wide Solvent Compatibility – The immobilization of the selector confers a wide solvent compatibility to these highly selective chiral stationary phases, without compromising phase stability. This is in contrast to traditional, coated polysaccharide phases which have restricted solvent compatibility due to solubility of the polymer coating in certain solvents, including chloroform, methylene chloride, ethyl acetate, acetone, THF and DMF. Immobilized phases can be used in all chromatographic modes: normal- and reversed-phase HPLC, SFC, analytical and preparative, and the same column can be used with all eluent combinations. Figure 6 shows the use of CHIRALPAK IC in different chromatographic modes.

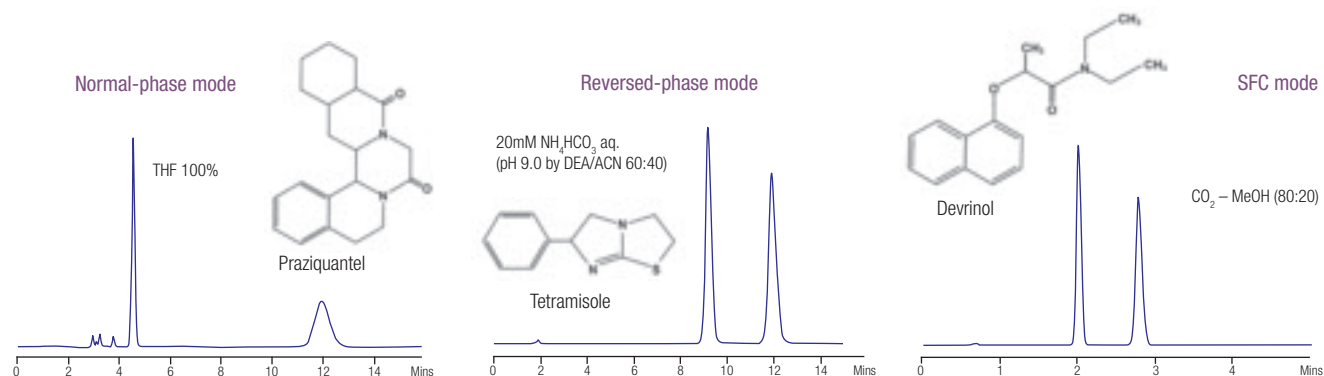


Figure 6. Different chromatographic modes with CHIRALPAK IC

2) Column Regeneration – CHIRALPAK immobilized columns are more robust than their coated analogues. If columns have been used with additives or with multiple solvent changes, a regeneration procedure may be implemented to eliminate any change in chiral recognition. Please contact Hichrom for further details.

3) Screening Strategies – Column screening is simpler, faster and more successful using the four main Daicel immobilized phases (IA, IB, IC and ID). The recommended eluent combinations and typical starting conditions for screening these phases are shown in Table 1.

Table 1. Immobilized primary screening solvents

Primary screening mixtures	Alkane-2-PrOH	Alkane/EtOH	Alkane/MtBE/EtOH	Alkane/DCM/EtOH
Typical starting conditions	80:20	80:20	0:98:2	50:50:2
Advised optimization range	99:1 to 50:50	99:1 to 50:50	80:20:0 to 0:40:60	85:15:0 to 0:100:0

When used in alkane/alcohol solvents, the immobilized columns can separate a significant number of small molecules, combined with the advantage of speed and ease of injecting in any suitable solvent. Figure 7 shows the increased success rate due to the introduction of the newer CHIRALPAK ID phase into a column screen of 123 compounds using a single eluent combination of hexane – 2-PrOH. When these immobilized phase columns are used with the set of four primary screening solvents, the separation success rate can approach 99%. If the desired chiral separation is not achieved, then an extended secondary screen can be applied.

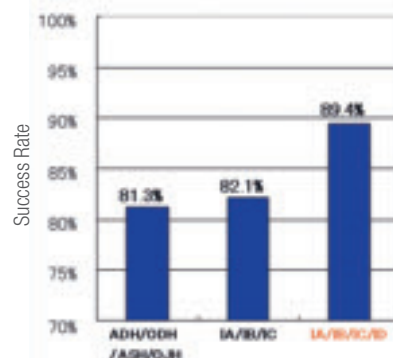


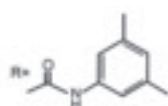
Figure 7. Screening success rates with single eluent

Please see page 86 for ordering information for CHIRALPAK immobilized phases.

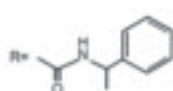
Daicel Polysaccharide Coated Phase Columns

- Original coated polysaccharide phases
- Fast easy method development
- Traditional 10 μ m, high resolution 5 μ m and ultra fast 3 μ m phases

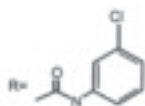
AMYLOSE-O-R



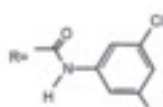
CHIRALPAK® 3A
CHIRALPAK® 3A-3
CHIRALPAK® AD-H
CHIRALPAK® AD
CHIRALPAK® AD-3
CHIRALPAK® AD-3R



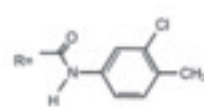
CHIRALPAK® AS-H
CHIRALPAK® AS
CHIRALPAK® AS-3
CHIRALPAK® AS-3R



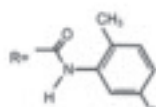
CHIRALPAK® ID
CHIRALPAK® ID-3



CHIRALPAK® IE
CHIRALPAK® IE-3

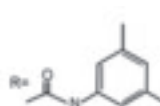


CHIRALPAK® AZ-H
CHIRALPAK® AZ
CHIRALPAK® AZ-3
CHIRALPAK® AZ-3R

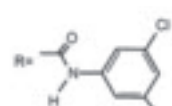


CHIRALPAK® AY-H
CHIRALPAK® AY
CHIRALPAK® AY-3
CHIRALPAK® AY-3R

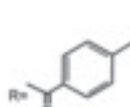
CELLULOSE-O-R



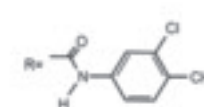
CHIRALPAK® IB
CHIRALPAK® IB-3
CHIRALCEL® OD-H
CHIRALCEL® OD
CHIRALCEL® OD-3
CHIRALCEL® OD-3R
CHIRALCEL® OD-I



CHIRALPAK® IC
CHIRALPAK® IC-3



CHIRALCEL® OJ-H
CHIRALCEL® OJ



CHIRALCEL® OZ-H
CHIRALCEL® OZ
CHIRALCEL® OZ-3
CHIRALCEL® OZ-3R

Figure 8. Structures of polysaccharide CSPs

Traditional Normal-Phase Columns (10 μ m)

The predecessors to the newer generation of immobilized columns, traditional CHIRALPAK® and CHIRALCEL® coated polysaccharide chiral stationary phases are based on a spherical high quality silica support on to which the polymeric chiral selector is physically coated. Due to the coated nature of these chiral supports, solvents should be carefully selected. CHIRALPAK AD and AS (amylose derivative coated on silica) and CHIRALCEL OD and OJ (cellulose derivative coated on silica) are the most widely used of the traditional 10 μ m phases.

High Resolution Reversed-Phase Columns (5 μ m)

Reversed-phase versions of the most popular normal-phase columns have been designed for high efficiency reversed-phase applications. These include CHIRALPAK AD-RH and AS-RH together with CHIRALCEL OD-RH and OJ-RH. These phases have the same coated chiral selector as the normal-phase columns but are coated to a hydrophobic high quality silica support. These reversed-phase columns were developed specifically for aqueous-organic eluents. They are suited for applications where the sample is in aqueous media (eg. biological samples) or for samples that require flexibility in terms of pH range. Figure 9 shows the separation of flurbiprofen enantiomers on CHIRALPAK AD-RH.

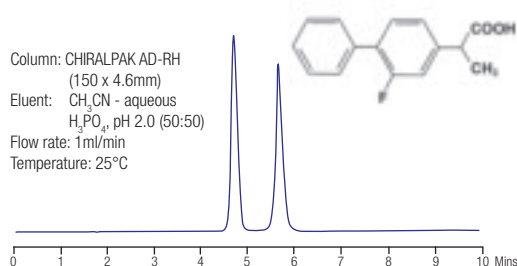


Figure 9. Analysis of flurbiprofen on CHIRALPAK AD-RH.

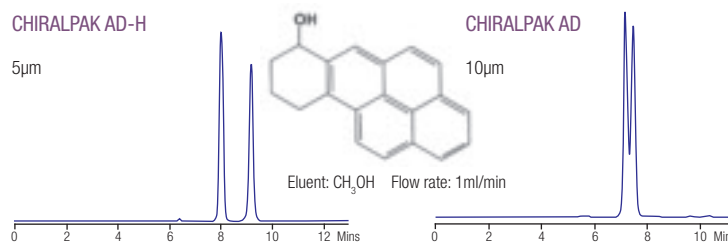


Figure 10. Analysis of 7, 8, 9, 10-tetrahydrobenzo[a]pyren-7-ol

High Resolution Normal-Phase Columns (5 μ m)

CHIRALPAK AD-H and AS-H together with CHIRALCEL OD-H and OJ-H are higher efficiency 5 μ m particle size normal-phase columns, used to resolve compounds requiring greater resolution than the traditional 10 μ m phases. Figure 10 shows the improved resolution obtained using a CHIRALPAK AD-H (5 μ m) compared to a CHIRALPAK AD (10 μ m) column.

Daicel Polysaccharide Coated Phase Columns (continued)

Ultra Fast NP and RP Columns (3 μ m)

More recently, 3 μ m particle size NP and RP phases have been introduced for higher resolution, fast analyses (CHIRALPAK AD-3, AD-3R, AS-3, AS-3R and CHIRALCEL OD-3, OD-3R, OJ-3, OJ-3R). These phases enable enhanced chromatographic separations to be achieved using conventional HPLC systems, without experiencing excessive pressure problems. Figure 11 demonstrates that enantiomer separation can be achieved in less than 30 seconds.

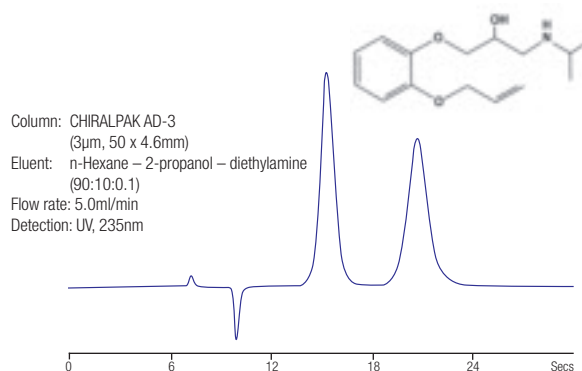


Figure 11. Fast analysis of oxprenolol on CHIRALPAK AD-3

Complementary Cellulose Chiral Phases

In addition to the four main phase types (AD, AS, OD and OJ), a complementary range of derivatized cellulose speciality phases is available. These phases were designed to resolve compounds that are not separated on these more versatile four key phases and include CHIRALCEL OA, OB, OC, OF, OG and OK.

New Polysaccharide Coated Phases

CHIRALPAK AY-H, AZ-H and CHIRALCEL OZ-H, OX-H 5 μ m and CHIRALPAK AY-3, AZ-3 and CHIRALCEL OZ-3, OX-3 3 μ m HPLC and SFC columns contain significantly different chiral selectors to the traditional phases discussed above. They show new recognition profiles, allowing effective method development for compounds not fully resolved on other Daicel columns. They are an excellent complement to the primary screen systems of CHIRALPAK IA, IB, IC and ID or the alternative CHIRALPAK AD, AS and CHIRALCEL OD, OJ.

Figure 12 shows the enhanced separation of the enantiomers of methyl-1-benzyl-5-oxo-3-pyrrolidinecarboxylate using CHIRALCEL OZ-H over that using CHIRALCEL OD-H.

The herbicide metolachlor has two chiral elements due to steric hindrance, which gives rise to four diastereomers. Figure 13 shows a comparison of the separation of these diastereomers using CHIRALPAK AY-H and CHIRALCEL OD-3.

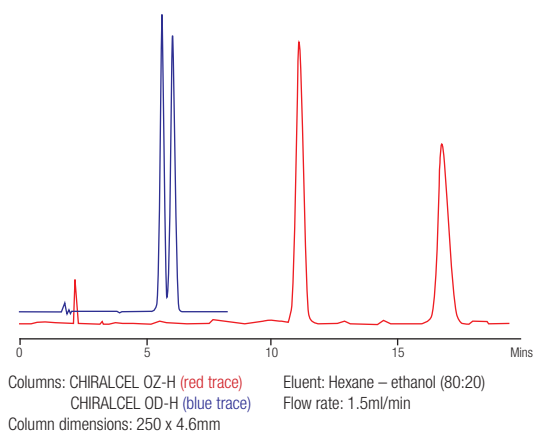


Figure 12. Separation of methyl-1-benzyl-5-oxo-3-pyrrolidinecarboxylate enantiomers

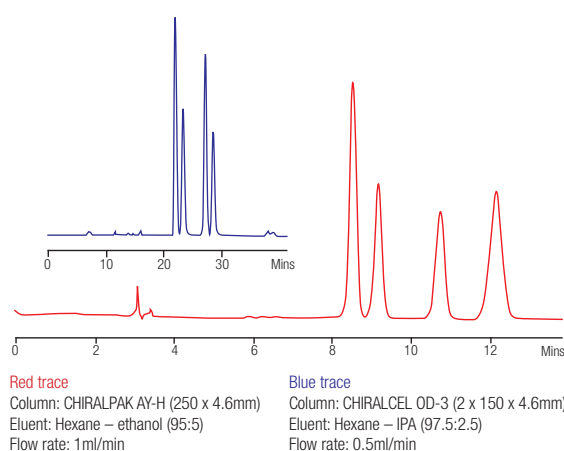


Figure 13. Separation of metolachlor diastereomers

Please see page 86 and 87 for ordering information for CHIRALPAK and CHIRALCEL coated phase columns.

Chiral Screening Service

Chiral Technologies Europe offers a chiral screening service in both HPLC and SFC, for customer supplied samples. A range of their chiral columns are screened under a variety of eluent compositions, in order to identify the most appropriate column. For further details of this service and to obtain a sample submission form, please contact Hichrom. Confidentiality agreements can be set up if required.

Application Guide

Chiral Technologies Europe have produced a fully searchable Application Guide on CD. This contains over 1,100 chiral applications, including new applications on the immobilized CSPs. Please contact Hichrom to obtain a free copy.



Daicel Columns for SFC

Daicel polysaccharide columns are well established for use in SFC separations. In addition to the benefits of speed of separation, speed of method development and improved column efficiency, green SFC technology reduces the use of organic solvents. Although the standard Daicel analytical HPLC columns (see pages 81- 84) can be used in SFC, semi-preparative CHIRALPAK and CHIRALCEL SFC columns are packed specifically to withstand the higher pressures of SFC.

Screening is recommended on the four main immobilized phases (IA, IB, IC and ID) and the four main coated columns (AD, OD, AS and OJ) with varying concentrations of organic modifier. Figures 14 and 15 illustrate the fast and efficient analyses possible using Daicel SFC columns.

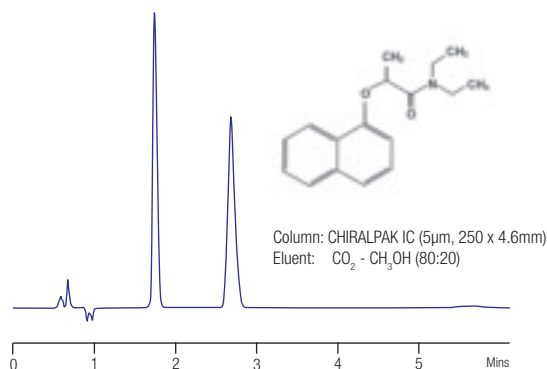


Figure 14. SFC separation of devrinol

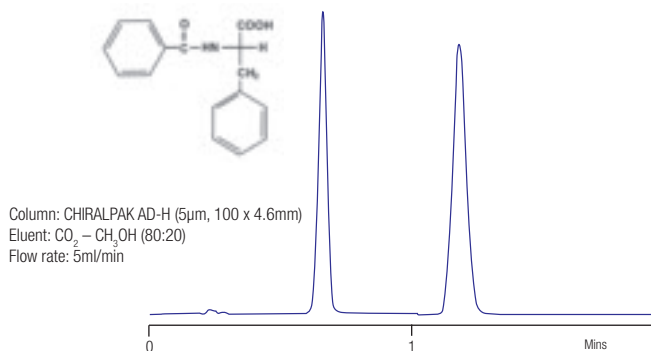


Figure 15. SFC separation of 1-naphthylethanol

Please see page 87 for ordering information for CHIRALPAK and CHIRALCEL SFC columns.

CHIRALPAK® QN-AX and QD-AX (Lindner Phase Columns)

- Reversed-phase or polar organic modes
- Wide solvent compatibility
- pH range 2 – 8

CHIRALPAK QN-AX and QD-AX are enantioselective weak anion-exchange (AX) columns. They are based on complementary stereoisomeric quinine (QN) and quinidine (QD) derivatives. Due to their pseudo enantiomeric character they usually reveal reversed elution order for opposite enantiomers.

These phases are designed specifically for enantioselective HPLC of chiral acids and show exceptional separation capabilities for acidic chiral compounds containing carboxylic, phosphonic, phosphinic, phosphoric or sulphonic acid groups. In some cases, weakly acidic compounds such as phenols can also be separated.

These two columns can be used in reversed-phase or polar organic modes. The separation of chiral basic and neutral compounds may also be possible, but usually under normal-phase conditions, when the phases behave like standard Pirkle type phases.

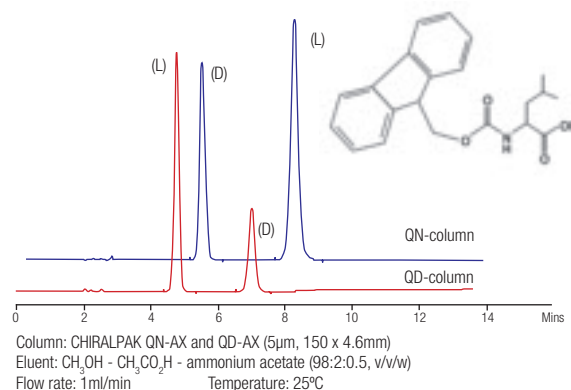
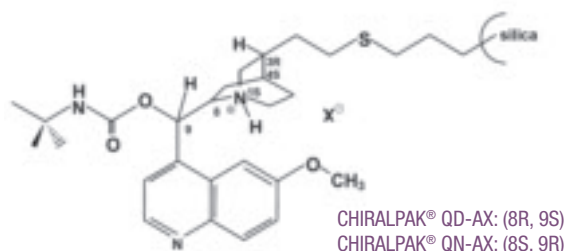


Figure 16. Analysis of Fmoc-Leu on CHIRALPAK QN-AX and QD-AX

Ordering Information

CHIRALPAK 5µm Phase	Column Dimensions (mm)			Analytical Guard Cartridges ¹ (3/pk) (For 4.6mm i.d. Columns)
	150 x 2.1	150 x 4.6	150 x 20	
QD-AX	31394	31324	31344	31311
QD-AX SFC	-	-	31444	-
QN-AX	32394	32324	32344	32311
QN-AX SFC	-	-	32444	-

¹ Use with holder 00011

and column coupler 000D1

Ordering Information - Immobilized Phases

CHIRALPAK 5µm Phase	Column Dimensions (mm)					Analytical Guard Cartridge ¹ (For 4.6mm i.d. Columns)	Semi-prep Guard Column ² (For 10 and 20mm i.d. Columns)
	150 x 2.1	150 x 4.6	250 x 4.6	250 x 10	250 x 20		
IA	80394	80324	80325	80335	80345	80311	80337
IB	81394	81324	81325	81335	81345	81311	81337
IC	83394	83324	83325	83335	83345	83311	83337
ID	84394	84324	84325	84335	84345	84311	84337
IE	85394	85324	85325	85335	85345	85311	85337
IF	86394	86324	86325	86335	86345	86311	86337

CHIRALPAK 3µm Phase	Column Dimensions (mm)					Analytical Guard Cartridge ¹ 3/pk (For 4.6mm i.d. Columns)
	150 x 2.1	50 x 4.6	100 x 4.6	150 x 4.6	250 x 4.6	
IA-3	80594	80522	80523	80524	80525	80511
IB-3	81594	81522	81523	81524	81525	81511
IC-3	83594	83522	83523	83524	83525	83511
ID-3	84594	84522	84523	84524	84525	84511
IE-3	85594	85522	85523	85524	85525	85511
IF-3	86594	86522	86523	86524	86525	86511

¹ Use with holder 00011

and column coupler 000D1

² No holder required

Ordering Information - Traditional Phases

10µm Phase ¹	Column Dimensions (mm)				Analytical Guard Cartridge ² (For 4.6mm i.d. Columns)	Semi-prep Guard Column ³ (For 10mm i.d. Columns)
	150 x 2.1	250 x 4.6	250 x 10	250 x 20		
Normal-phase						
CHIRALPAK AD	19094	19025	19035	19045	19311 ⁴	19032
CHIRALPAK AS	20094	20025	20035	20045	20311 ⁴	20032
CHIRALCEL OD	-	14025	14035	14045	14311 ⁴	14032
CHIRALCEL OJ	-	17025	17035	17045	17311 ⁴	17032
Reversed-phase						
CHIRALCEL OD-R	-	14625	-	-	14611	-

5µm Phase	Column Dimensions (mm)					Analytical Guard Cartridge ² 3/pk (For 4.6mm i.d. Columns)
	150 x 2.1	150 x 4.6	250 x 4.6	250 x 10 ⁵	250 x 20	
Normal-phase						
CHIRALPAK AD-H	19394	19324	19325	19335	19345	19311
CHIRALPAK AS-H	20394	20324	20325	20335	20345	20311
CHIRALCEL OD-H	14394	14324	14325	14335	14345	14311
CHIRALCEL OJ-H	17394	17324	17325	17335	17345	17311
CHIRALPAK AZ-H	61394	61324	61325	61335	61345	61311
CHIRALPAK AY-H	47394	47324	47325	47335	47345	47311
CHIRALCEL OZ-H	42394	42324	42325	42335	42345	42311
CHIRALCEL OX-H	63394	63324	63325	63335	63345	63311
Reversed-phase						
CHIRALPAK AD-RH	19794	19724	-	-	-	19711
CHIRALPAK AS-RH	20794	20724	-	-	-	20711
CHIRALCEL OD-RH	14794	14724	-	-	-	14711
CHIRALCEL OJ-RH	17794	17724	-	-	-	17711
CHIRALPAK AZ-RH	61794	61724	-	-	-	61711
CHIRALPAK AY-RH	47794	47724	-	-	-	47711
CHIRALCEL OZ-RH	42794	42724	-	-	-	42711
CHIRALCEL OX-RH	63794	63724	-	-	-	63711

¹ Other phases and column dimensions available

² Use with holder 00011 and column coupler 000D1

³ No holder required

⁴ 5µm material

⁵ Semi-prep guard column available – please enquire

Ordering Information - Traditional Phases (continued)

3µm Phase	Column Dimensions (mm)					Analytical Guard Cartridge ¹ 3/µk (For 4.6mm i.d. Columns)
	150 x 2.1	50 x 4.6	100 x 4.6	150 x 4.6	250 x 4.6	
Normal-phase						
CHIRALPAK AD-3	19594	19522	19523	19524	19525	19511
CHIRALPAK AS-3	-	20522	20523	20524	20525	20511
CHIRALCEL OD-3	14594	14522	14523	14524	14525	14511
CHIRALCEL OJ-3	-	17522	17523	17524	17525	17511
CHIRALPAK AZ-3	-	61522	61523	61524	61525	61511
CHIRALPAK AY-3	-	47522	47523	47524	47525	47511
CHIRALCEL OZ-3	-	42522	42523	42524	42525	42511
CHIRALCEL OX-3	63594	63522	63523	63524	63525	63511
Reversed-phase						
CHIRALPAK AD-3R	19894	19822	19823	19824	-	19811
CHIRALPAK AS-3R	-	20822	20823	20824	-	20811
CHIRALCEL OD-3R	14894	14822	14823	14824	-	14811
CHIRALCEL OJ-3R	-	17822	17823	17824	-	17811
CHIRALPAK AY-3R	-	47822	47823	47824	-	47811
CHIRALPAK AZ-3R	-	61822	61823	61824	-	61811
CHIRALCEL OZ-3R	-	42822	42823	42824	-	42811
CHIRALCEL OX-3R	63894	63822	63823	63824	-	63811

¹ Use with holder 00011 and column coupler 000D1

Ordering Information - SFC Phases

5µm Phase	Column Dimensions ¹ (mm)		
	100 x 4.6	250 x 10	250 x 20
CHIRALPAK IA SFC	80423	80435	80445
CHIRALPAK IB SFC	81423	81435	81445
CHIRALPAK IC SFC	83423	83435	83445
CHIRALPAK ID SFC	84423	84435	84445
CHIRALPAK IE SFC	85423	85435	85445
CHIRALPAK IF SFC	86423	86435	86445
CHIRALPAK AD-H SFC	19423	19435	19445
CHIRALPAK AS-H SFC	20423	20435	20445
CHIRALCEL OD-H SFC	14423	14435	14445
CHIRALCEL OJ-H SFC	17423	17435	17445
CHIRALPAK AZ-H SFC	61423	61435	61445
CHIRALPAK AY-H SFC	47423	47435	47445
CHIRALCEL OZ-H SFC	42423	42435	42445
CHIRALCEL OX-H SFC	63423	63435	63445

¹ Please enquire regarding larger i.d. prep columns


CHIRALPAK® Protein Phase Columns

- Protein-bonded silica
- Reversed-phase applications
- Unique separation characteristics
- Hundreds of applications available

Protein stationary phases were originally developed and manufactured by ChromTech Ltd, but are now manufactured by Chiral Technologies Europe. The range consists of three protein-based columns – CHIRALPAK AGP, CHIRALPAK CBH and CHIRALPAK HSA (previously called CHIRAL-AGP, CHIRAL-CBH and CHIRAL-HSA), where the protein is immobilized on 5µm porous spherical silica particles.

Phase	Chiral Selector	Particle Size (µm)	Applications
CHIRALPAK AGP	α_1 -acid glycoprotein	5	Most compound types – amines, acids, alcohols, amides, esters, sulphoxides
CHIRALPAK CBH	Cellobiohydrolase	5	Nitrogen-containing compounds also containing alcohol, phenol, carbonyl, amide, ether or ester group(s)
CHIRALPAK HSA	Human serum albumin	5	Weak and strong acids, zwitterionic and non-protolytic compounds

CHIRALPAK AGP has the broadest applicability of the three chiral phases, separating a wide range of compound types and is often the column of choice for method development. CHIRALPAK CBH has a narrower applicability, preferentially separating compounds containing one or more nitrogen atoms together with one or more hydrogen accepting or donating groups, and is particularly suited for the analysis of very hydrophilic amines. CHIRALPAK HSA is also more suitable for specific applications, particularly very hydrophilic acids.

	Amines	Non-protolytes	Acids
CHIRALPAK AGP	[Red bar spanning all three categories]		
CHIRALPAK CBH	[Red bar]		
CHIRALPAK HSA			[Red bar]

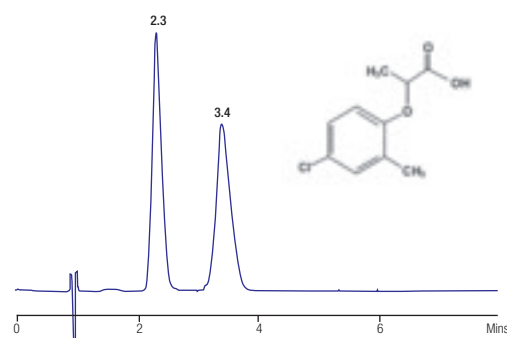
These columns function in the reversed-phase mode, using buffers with low organic content and at moderate pH.

Method Development

Analytes are retained on these protein-bonded phases by a combination of ionic binding (charged solutes), hydrophobic interaction and hydrogen bonding. Consequently, separations are affected by pH and the nature and concentration of aqueous buffer and organic modifier.

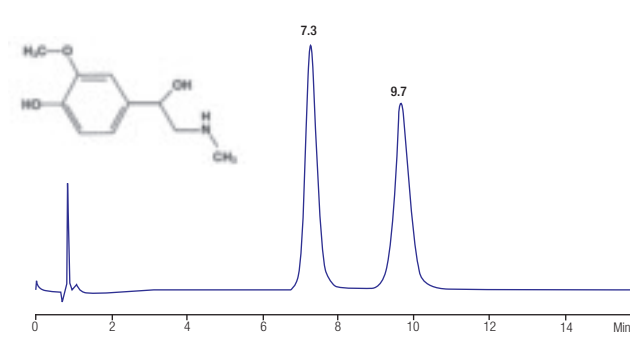
Parameter	Typical Conditions	Effect of Varying Parameter
pH	4 - 7	Variable
Buffer	Phosphate or acetate (0.01 – 0.1M)	Increase in buffer concentration can increase retention and enantioselectivity
Organic Solvent	Propan-2-ol, acetonitrile, methanol (0 – 15%)	Selection of solvent strongly affects enantioselectivity. Higher organic solvent ratios reduce retention time for each phase. For both CHIRALPAK AGP and HSA columns, enantioselectivity is simultaneously reduced, whilst for CHIRALPAK CBH columns it is often increased.

Figures 17 to 19 show typical applications on CHIRALPAK protein phase chiral columns.



Column: CHIRALPAK AGP (100 x 4.0mm)
Eluent: 5% 2-Propanol in 10mM ammonium acetate pH 5.8
Flow rate: 0.9ml/min

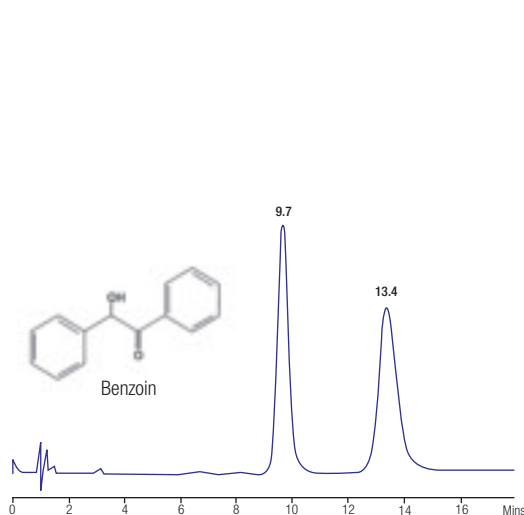
Figure 17. Mecoprop



Column: CHIRALPAK CBH (100 x 4.0mm)
Eluent: 5% 2-Propanol in 10mM Na phosphate, pH 7 + 50µM Na₂ EDTA
Flow rate: 0.9ml/min

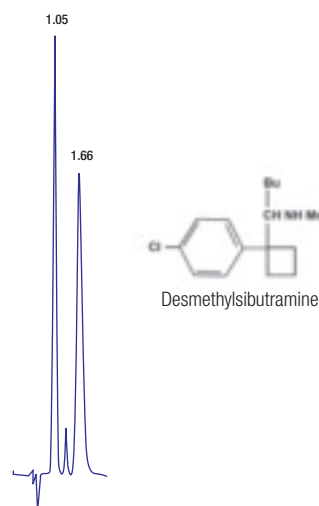
Figure 18. Metanephrine

CHIRALPAK Protein Phase Columns (continued)



Column: CHIRALPAK HSA (100 x 4.0mm)
 Eluent: 2% CH₃CN in 100mM Na phosphate pH 7.0
 Flow rate: 0.9ml/min

Figure 19. Benzoin



Column: CHIRALPAK AGP (50 x 4.0mm)
 Eluent: 5% CH₃CN in 10mM ammonium acetate buffer, pH 4.1
 Flow rate: 0.9ml/min

Figure 20. Fast chiral separation suitable for MS detection

Columns for LC-MS

Shorter CHIRALPAK protein phase columns are available for rapid analysis and LC-MS applications. In order to convert from a UV to LC-MS method, in addition to decreasing column dimensions, phosphate buffers are replaced with ammonium acetate and the concentration of buffer and organic modifier reduced. Figure 20 shows the rapid separation of desmethyisbutramine using a short CHIRALPAK AGP column.

Drug – Plasma Protein Binding Studies

Another application of these protein-based columns is their use in drug-plasma protein binding studies. As the degree of drug-protein binding directly affects pharmacokinetic and pharmacodynamic characteristics of a pharmaceutical compound, a drug's potency may be dependent on the degree to which it binds to the plasma proteins and other blood constituents. HPLC analysis using CHIRALPAK AGP and CHIRALPAK HSA columns has been shown to be useful in drug binding studies.

Ordering Information

CHIRALPAK Phase	Column Dimensions (mm)						Guard Cartridges ¹ (2/pk)	
	50 x 2.0	100 x 2.0	150 x 2.0	50 x 3.0	100 x 3.0	150 x 3.0	10 x 2.0 ²	10 x 3.0 ³
AGP	30792	30793	30794	30782	30783	30784	30791	30781
CBH	33792	33793	33794	33782	33783	33784	33791	33781
HSA	34792	34793	34794	34782	34783	34784	34791	34781

CHIRALPAK Phase	Column Dimensions (mm)					Guard Cartridges ¹ (2/pk)
	50 x 4.0	100 x 4.0	150 x 4.0	100 x 10.0	150 x 10.0	10 x 4.0 ⁴
AGP	30712	30713	30714	30733	30734	30711
CBH	33712	33713	33714	33733	33734	33711
HSA	34712	34713	34714	34733	34734	34711

¹ Use with free-standing holder 00081

² For use with 2.0mm i.d. columns and column coupler 000D2

³ For use with 3.0mm i.d. columns and column coupler 000D2

⁴ For use with 4.0mm i.d. columns and column coupler 000D1

Crown Ether Chiral Columns

The CROWNPAK® CR(+) and CR(-) phases contain a chiral crown ether as chiral selector, which is coated on to a 5µm silica support. They can be used for the enantiomeric separation of amino acids and other small molecules containing a primary amino group near the chiral centre. The CR(-) column gives the reversed elution order compared to the CR(+) column. Acidic eluents such as perchloric acid at pH 1 to 2 are used as standard with these columns. Figure 21 shows the separation of arginine on CROWNPAK CR.

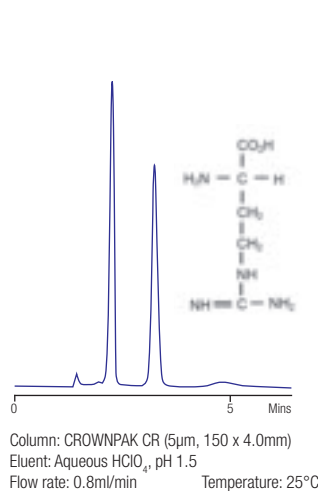


Figure 21. Analysis of arginine on CROWNPAK CR

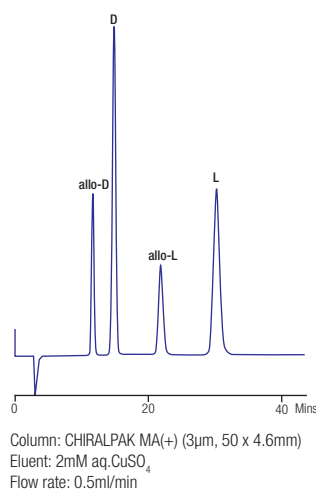




Figure 22. Analysis of isoleucine on CHIRALPAK MA(+)

Ligand Exchange Chiral Columns

The ligand exchange phases, CHIRALPAK MA(+) and CHIRALPAK WH (see Table 2) consist of amino acids and their derivatives coated on to silica supports. They are used in conjunction with an aqueous CuSO₄ eluent (0.1 to 2mM) and are useful for the separation of amino acids and their derivatives (see Figure 22).

Table 2. Crown Ether and Ligand Exchange Chiral Phases

Phase	Particle Size (µm)	Adsorbent	Applications
CROWNPAK CR(+)	5		Amino acids and compounds with a primary amino group near an asymmetric centre
CROWNPAK CR(-)	5		
CROWNPAK MA(+)	3	N,N-Dioctyl-L-alanine ligand	α-Hydroxycarboxylic acids, α-amino acids
CROWNPAK WH	10		Amino acids and their derivatives

Ordering Information

Crown Ether phases

CROWNPAK Phase	Column Dimensions (mm)		CROWNPAK CR Guard Column
	150 x 4.0		10 x 4.0
CROWNPAK CR(+)	27714		27711
CROWNPAK CR(-)	28714		27711

Ligand Exchange Phases

CHIRALPAK Phase	Column Dimensions (mm)		
	250 x 4.6	50 x 4.6 (guard)	50 x 4.6
CHIRALPAK WH	25625	25622	-
CHIRALPAK MA(+)	-	-	21822