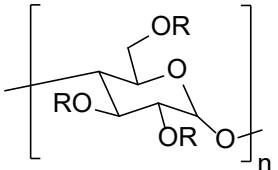
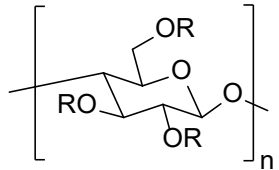
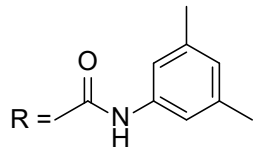
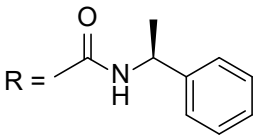
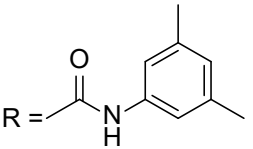
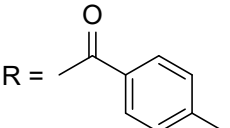
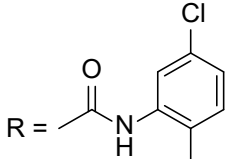
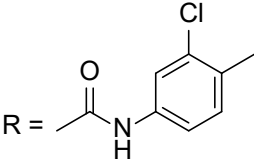
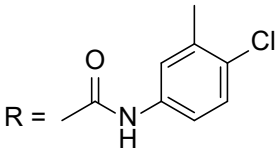
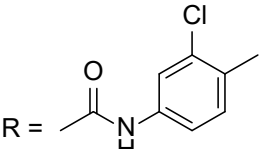


**INSTRUCTION MANUAL FOR Analytical Columns of
CHIRALPAK® AD-3, AS-3, AY-3, AZ-3
CHIRALCEL® OD-3, OJ-3, OX-3, and OZ-3**

<Supercritical Fluid Chromatography (SFC)>

Please read this instruction sheet completely before using these columns

Column Description

AMYLOSE-BASED		CELLULOSE-BASED	
 <p>Coated on 3 µm silica gel</p>		 <p>Coated on 3 µm silica gel</p>	
CHIRALPAK® AD-3	CHIRALPAK® AS-3	CHIRALCEL® OD-3	CHIRALCEL® OJ-3
Amylose tris(3,5-dimethylphenylcarbamate)	Amylose tris[(S)-α-methylbenzylcarbamate]	Cellulose tris(3,5-dimethylphenylcarbamate)	Cellulose tris(4-methylbenzoate)
			
CHIRALPAK® AY-3	CHIRALPAK® AZ-3	CHIRALCEL® OX-3	CHIRALCEL® OZ-3
Amylose tris(5-chloro-2-methylphenylcarbamate)	Amylose tris(3-chloro-4-methylphenylcarbamate)	Cellulose tris(4-chloro-3-methylphenylcarbamate)	Cellulose tris(3-chloro-4-methylphenylcarbamate)
			
Shipping Solvent: Hexane/IPA = 90:10 (v/v)			
All columns have been pre-tested before packaging. Test parameters and results, as well as the Column Lot Number, were included with the column when purchased.			

THIS INSTRUCTION MANUAL IS NOT APPLICABLE TO ANY OTHER DAICEL COLUMNS

Because different columns are shipped in different solvents, we recommend flushing them with 100% Ethanol or Isopropanol before their first use in SFC to avoid any damage (see column transfer conditions between LC and SFC on page 4).

CAUTION

The entire SFC system, including the injector and the injection loop, must be flushed with a solvent compatible with the column and its storage solvent prior to connecting the column. Many of the solvents commonly used as SFC modifiers including acetone, chloroform, DMF, dimethylsulfoxide, ethyl acetate, methylene chloride, and THF, may DESTROY the chiral stationary phase if they are present, even in residual quantities, within the system.

If an auto-sampler is used, then the solvent employed to flush this unit between injections should also be changed to something compatible and the relevant solvent lines flushed.

Operating Instructions

	50 x 2.1 mm i.d. 100 x 2.1 mm i.d. 150 x 2.1 mm i.d. 250 x 2.1 mm i.d. Analytical Column	50 x 4.6 mm i.d. 100 x 4.6 mm i.d. 150 x 4.6 mm i.d. 250 x 4.6 mm i.d. Analytical Columns
Flow Rate Direction	As indicated on the column label	
Typical Flow Rate	0.1-0.5 ml/min	0.5-2.5 ml/min
Pressure Limitation ^①	Please contact Technical Support for details	
Temperature	0 to 40°C	
Column Fitting	Please contact Technical Support for details	

① The relevant pressure value is the one generated by the column itself (pressure drop). The pressure drop is the difference between the inlet pressure (P_{inlet}) and the outlet pressure (P_{outlet}) in the system. The pressure drop generated by the system alone (without any column) has to be subtracted from the total value (system + column).

The column can be operated up to 300 Bar (pressure drop). However, it is necessary to check if the SFC system has been designed to withstand these conditions. The flow rate has to be adapted considering the pressure drop in the column (this pressure being dependent upon flow rate as well as the amount and type of modifier in the mobile phase).

 **Please contact Chiral Technologies for further assistance before trying any solvents not mentioned below.**

A - Mobile Phases

CAUTION

Basic conditions SHOULD BE AVOIDED, both in the sample solution and the mobile phase, for CHIRALPAK® AZ-3.

Primary Solvent Mixtures	CO ₂ / MeOH	CO ₂ / EtOH	CO ₂ / 2-PrOH	CO ₂ / ACN ^❶
Typical Starting Conditions	80:20	80:20	80:20	70:30 ^❶
Advised Optimization Range	99:1 to 40:60	99:1 to 40:60	99:1 to 40:60	99:1 to 40:60 ^❶

❶ For strongly retained compounds, an alcohol can be added into CH₃CN to enhance the eluting strength.

Note: The retention is generally shorter with Ethanol than with 2-Propanol, and the retention is generally shorter with higher alcohol contents. The use of other alcohols such as 1-Propanol, 1-BuOH, 2-BuOH, etc. is possible, but effectiveness is not predictable.

B – General Comments

The typical starting conditions consist in mobile phases of upper middle eluting strength. Under such conditions, most of the analytes can be eluted within a reasonable time range with a good probability of full resolution of the enantiomers.

C – Additives

For basic samples, it is necessary to incorporate an additive into the mobile phase in order to optimize the chiral separation.

Acidic samples **do not always** require the presence of an additive. In fact, the acidic properties of the carbon dioxide (CO₂) are sometimes enough to elute the product properly.

- ❶ In practice, 1% of the additive is incorporated with the modifier. The total amount of additive into the mobile phase will be dependent upon the percentage of modifier. For example, if the mobile phase is CO₂ / EtOH = 90:10, with EtOH containing 1% of additive, then the mobile phase composition will be CO₂ / EtOH / additive = 90:10:0.1).

Basic Samples require Basic additives ^❶	Acidic Samples require Acidic additives ^❶
Isopropylamine (IPAm) Diethylamine (DEA) Triethylamine (TEA)	Trifluoroacetic acid (TFA) Acetic acid Formic acid

⇒ **STRONGLY BASIC solvent additives or sample solutions MUST BE AVOIDED, because they are likely to damage the silica gel used in this column**

Column Care / Maintenance

- ❑ Samples should preferably be dissolved in the modifier.
- ❑ Sample solutions should be filtered through a membrane filter of approximately 0.5 µm porosity to ensure that there is no precipitate before use.

☞ Column transfer between modes:

From LC to SFC

- Flush with 100% EtOH at 0.25 ml/min^(*) for 45 min
- Flush with 100% CO₂ or CO₂+modifier at 0.25 ml/min^(*) for 45 min

From SFC to LC

- Flush with 100% EtOH at 0.25 ml/min^(*) for 45 min
- Flush with the mobile phase at 0.25 ml/min^(*) for 45 min

() This is the recommended flow rate for a 4.6 mm i.d. analytical columns. The flow rate of all other inner diameter columns should be adjusted proportional according to the cross-sectional area of the column.*

Column Storage

- ❑ For column storage, remove the acidic or basic additives by flushing the column with several column volumes of 100% EtOH or 100% methanol, without additives.
- ❑ Columns can be stored with ends capped in the additive-free mobile phase, or the shipping solvent, at room temperature.

Important Notice

⇒ STRONGLY BASIC solvent additives or sample solutions MUST BE AVOIDED, because they are likely to damage the silica gel used in these columns.

Operating these columns in accordance with the guidelines outlined here will result in a long column life.

⇒ If you have any questions about the use of these columns, or encounter a problem, contact:

In the USA: questions@cti.daicel.com or call 800-6-CHIRAL

In the EU: cte@cte.daicel.com or call +33-388-795-200

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