# Column Care and Use Instructions

# Alcyon SFC CSP Amylose-SA, Cellulose-SB, Cellulose-SC

Supercritical Fluid Chromatography / Optical Isomer Separation Column (Immobilized type)

#### 1. Introduction

Thank you for purchasing an Alcyon SFC Column for Supercritical Fluid Chromatography (SFC).

Alcyon SFC CSP is designed for separating optical isomers. The immobilized chiral selectors, polysaccharide derivatives, provide high compatibility with wide range of organic solvents, and superior separation and selectivity.

Alcyon SFC Columns, which are manufactured under highly controlled conditions, must pass a series of strict tests before being accepted for shipment (Please refer to the column inspection report). To ensure optimal performance and durability of the column, please read these instructions carefully before using this column.

## 2. Specifications

ltem	Alcyon SFC CSP Amylose-SA	Alcyon SFC CSP Cellulose-SB	Alcyon SFC CSP Cellulose-SC	
Particle size	3, 5 µm			
Chiral selector	Amylose tris(3,5- dimethylphenylcarbamate)	Cellulose tris(3,5- dimethylphenylcarbamate)	Cellulose tris(3,5- dichlorolphenylcarbamate)	
Туре	Immobilized type			
Shipping solvent <sup>1</sup>	2-propanol			
Usable temperature range	0-40°C			
Pressure limit <sup>2</sup>		,,	30 MPa 20 MPa	
Recommended flow rate <sup>3</sup>	3.0 mml.D. : 0.4 ~ 1	0.6 mL/min 10 mr 1.2 mL/min 20 mr 3.0 mL/min	n I.D. : 5 ~ 15 mL/min n I.D. : 20 ~ 60 mL/min	

<sup>1:</sup> If you intend to store the column for a long time, replace the mobile phase in the column with shipping solvent.

#### 3. Precautions for use

- · The column endfitting is Waters style connection.
- Tubing must have flat ends and must bottom out in the column endfitting. Tubing must be connected to the column correctly to
  avoid creating a void between the column frit and tubing, which can cause a leak and result in poor column performance (e.g.
  peak tailing, loss of theoretical plate number).
- · The correct direction of the solvent flow is indicated by an arrow on the column identification label.
- · Do not disconnect a column from the SFC system before the pressure drops to zero.

<sup>2:</sup> Avoid using a column repeatedly near the pressure limit or abrupt change in the pressure in order to prevent from shortening the column lifetime.

<sup>3:</sup> Adjust flow rate according to the recommendation in the table above to obtain the optimum results under the application.

<sup>2,3:</sup> Pressure changes depending on column length, temperature, types of organic solvent etc. If pressure exceeds the upper limit, reduce flow rate to below the lower rate of recommended range.

### 4. Mobile phase and sample solvent

- When a target compound is ionic, addition of additives listed below can improve peak shape and/or separation reproducibility. High
  concentrations of additives can result in reducing column lifetime. Add/reduce the additives according to the notes in the table.
   Additive concentration below is concentration for the entire mobile phase.
- When possible, the sample should be dissolved in the same modifier as the mobile phase. Using a stronger solvent than mobile phase for sample dissolution might result in distorted peak symmetry and degraded resolution.
- In order to avoid blockage which can cause pressure increase, the sample solution should be filtered through a membrane filter (0.2 µm or smaller porosity).

#### [Recommended solvents]

	Acidic compounds	Basic compounds	Neutral compounds	
Modifiers	alcohols(methanol, ethanol, 2-propanol), acetonitrile, ethyl acetate, tetrahydrofuran (THF), dichloromethane, chloroform, methyl <i>tert</i> -butyl ether (MTBE), etc			
Additives	0.1% (Upper limit 0.5%) trifluoroacetic acid (TFA), acetic acid, formic acid, etc	0.1% (Upper limit 0.5%) diethylamine (DEA), butylamine, ethanolamine, etc	None	
Composition ratio	CO <sub>2</sub> /modifiers (99/1 – 40/60)			

# 5. Column cleaning (general method)

- Flush the column with solution containing a higher ratio of modifiers (for example, for CO<sub>2</sub>/methanol mobile phase, concentration of methanol should be increased) for washing out the compounds that have a great capacity for retention in the column. When further cleaning is required, flush with 100% ethanol is effective.
- If 100% ethanol does not improve the column performance, flush10 (ten) column volumes (30 (thirty) column volumes: when
  mobile phase contains additives) of N, N-dimethylformamide (DMF) on Amylose-SA column and of ethyl acetate on Cellulose-SB
  column. There is a possibility that the column performance is restored after storing the column with the corresponding solvent for
  several days.
- When a mobile phase containing acid or amine is used, replace with CO<sub>2</sub>/modifiers containing neither of them (at the same ratio
  as the mobile phase), then wash as above procedure. Storing a column with a mobile phase containing additive is not
  recommended even for a short period of time.
- The column needs to be replaced when these cleaning methods do not regenerate the column performance. To extend the column lifetime, especially for samples containing large amount of impurities, we recommend a sample pretreatment conducted carefully prior to introducing the sample to the column.