# Care and Use Instructions CHIRAL ART Packing Material

# Immobilized type

## 1. Introduction

Thank you for purchasing CHIRAL ART packing material. CHIRAL ART (immobilized type) 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. With its advantages, CHIRAL ART (immobilized type) is suitable for separating a variety of chiral compounds.

CHIRAL ART packing materials, which are manufactured under highly controlled conditions, must pass a series of strict tests before being accepted for shipment (Please refer to the inspection report). To ensure optimal performance and stability of the packing material, please follow these instructions before use.

## 2. Specifications

	CHIRAL ART Amylose-SA	CHIRAL ART Cellulose-SB	CHIRAL ART Cellulose-SC	CHIRAL ART Cellulose-SJ
Particle size (µm)	10, 20			
Chiral selector	Amylose tris(3,5- dimethylphenylcarbamate)	Cellulose tris(3,5- dimethylphenylcarbamate)	Cellulose tris(3,5- dichlorophenylcarbamate)	Cellulose tris(4-methylbenzoate)
Туре	Immobilized type			
Usable pH range	2.0 – 9.0			
Bulk density (g/cm <sup>3</sup> )	ca. 0.53			

# 3. Packing instructions [for dynamic axial compression (DAC) columns]

### 3-1 Amount of packing material required

Calculate the amount of packing material by using the column volume and the bulk density (see section 2).

### 3-2 Preparation of packing slurry and packing

2-propanol is recommended as the slurry and packing solvent. Add the solvent to obtain a slurry at a concentration of 30 - 40%, and transfer the slurry to a DAC column. Packing pressure of 6 - 8 MPa is recommended for packing. Take care not to exceed the maximum usable pressure of DAC column.

\*slurry concentration (%, w/v) = amount of packing material (kg) / total volume of slurry (L) x 100

## 3-3 Testing the packed column (Evaluation of column performance)

Once packing is completed, check the theoretical plate number (N) and peak shape. In the case where appropriate theoretical plate number (N) or asymmetry factor (As) is not obtained, please optimize the packing condition.

# Example conditions of column performance evaluation

Eluent: n-hexane / 2-propanol (90/10, v/v)

Linear velocity: 180 cm/hr
Detection: UV at 230 nm

Sample: 1,3,5-Tri-tert-butylbenzene (0.5 mg/mL) [void volume marker]

trans-Stilbene oxide (0.1 mg/mL) [peak 1, peak 2]

Sample solvent: Eluent

Injection: 30 x cross-sectional areas of columns (cm²) µL

Evaluation: Theoretical plate number (N) of trans-Stilbene oxide (peak 2)

# Expected theoretical plate number (N/m) \*

10 µm	20 μm
16,000	6,000

<sup>\*</sup> Values might be influenced by column or LC system

#### 4. Precautions for use

- CHIRAL ART (immobilized type) is suitable for any mobile phase from aqueous to non-aqueous solvents. The packing material
  can be used for both normal phase and reversed phase (aqueous mobile phase). We recommend that the packing material is
  dedicated to either phase. Please refer to the recommendation of the solvents in the table below.
- Make sure of miscibility between the organic solvents. When switching the mobile phase from alkane/alcohol mobile phase to polar organic solvents (methanol, acetonitrile, etc.), flush the column with a compatible solvent such as ethanol or 2-propanol beforehand. After that, sufficiently equilibrate the column with the mobile phase.
- · Be careful of the solvent resistance of LC system and PEEK tubing when using them on normal phase mode.
- To protect a column/packing material, a sample containing a lot of impurities should be filtered out before injection. Also it is recommended that a guard column should be used.
- · Operating pressure should not exceed the packing pressure.

#### [Recommended solvents for Normal phase]

	Acidic compounds	Basic compounds	Non-ionic compounds	
Organic solvents	alkane ( <i>n</i> -hexane or <i>n</i> -heptane), alcohols (methanol, ethanol, 2-propanol), acetonitrile, ethyl acetate, tetrahydrofuran (THF), dichloromethane, chloroform, methyl <i>tert</i> -butyl ether (MTBE)			
Modifiers	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	Any ratio (these must be miscible)			

#### (Recommended solvents for Reversed phase)

	Acidic compounds	Basic compounds	Non-ionic compounds		
Organic solvents	acetonitrile, methanol, ethanol, 2-propanol, THF, etc.				
Aqueous phase (Modifiers)	0.1% phosphoric acid, 0.1% formic acid, 50 – 100 mM phosphate buffer (pH 2.0 – 3.5), etc.	20 mM NH₄HCO₃-DEA buffer (pH 9.0), etc.	Water		
Composition ratio	Organic solvent/ aqueous solution (10/90 – 100/0)				

## 5. Column cleaning and storage

Clean and store the used column according to the following procedure. During storage, keep away from heat and moisture, and put airtight stoppers on the both column ends to prevent the packed bed from drying out. Avoid storing the column with a mobile phase containing buffer salts/modifiers even if it is only for a short period.

## [Normal phase]

- Flush the column with a solution containing a higher ratio of the stronger solvent (for example, for alkane/alcohol mobile phase, concentration of alcohol should be increased) for washing out the compounds that have a great capacity for retention in the column. When further cleaning is required, flushing with 100% ethanol is effective.
- When a mobile phase containing acid or amine is used, replace the mobile phase with solvent containing neither acid nor amine (at the same ratio as the mobile phase). Then flush the column in accordance with the method described above.
- Store the column in *n*-hexane/2-propanol (90/10) or an organic solvent such as 2-propanol or ethanol.

#### [Reversed phase]

- Flush the column with a solution containing a higher ratio of organic solvent for washing out the compounds that have a great capacity for retention in the column after using mobile phases not containing buffer salts/additives. Usable concentration of organic solvent is up to 100%.
- When a mobile phase containing buffer salts/additives is used, replace the mobile phase with a water/organic solution
  containing no buffer salts/modifiers (A ratio of water to organic solvent should be set at the same proportions as a mobile
  phase). Then flush the column in accordance with the method described above.
- Store the column in an organic solvent such as 2-propanol or ethanol.

## 6. Packing material storage

**Unused packing material:** Store the packing material in the original container, and keep away from heat and moisture. **Used packing material:** At first, clean the packing material in accordance with the method described in section 5.

[Storage in a dry form]

Flush the column with organic solvents such as 2-propanol or ethanol, and then remove the packing material from the column. After drying the unpacked material at 60 °C or below, transfer it to an appropriate container. Keep away from heat and moisture.

[Storage in organic solvent]

Flush the column with organic solvents such as 2-propanol or ethanol, and then remove the packing material from the column. Transfer the unpacked material to an appropriate container and store it in the same solvent. Please ensure that the container is tightly sealed.

NOTE - We do not guarantee the used packing material, and cannot accept any return of it.