

ULTRON AF-HILIC-CD Series

Instruction Manual

1. Introduction

Thank you for purchasing a ULTRON AF-HILIC-CD series column for High-performance liquid chromatography. The ULTRON AF-HILIC-CD series is a Hydrophilic Interaction Chromatography (HILIC) column based on silica gel (2 μm/5μm) bonded withβ-cyclodextrin via a spacer. The ULTRON AF-HILIC-CD series column is able to retain and separate highly polar compounds that are not retained in reversed phase chromatography, with a mobile phase containing high concentration of organic solvent.

ULTRON AF-HILIC-CD series columns, which are manufactured under highly controlled conditions, must pass a series of strict tests before being accepted for shipment. To ensure optimal performance and durability of the column, please read these instructions carefully before using this column.

2. specification

Descriptions	Column name	Particle size (μm)	Details
Fitting	ULTRON AF-HILIC-CD	2, 5	Waters compatible
	ULTRON AF-HILIC-CD (PLSS)	2	
pH range	ULTRON AF-HILIC-CD	2, 5	Recommended range: 2.0~7.0
	ULTRON AF-HILIC-CD (PLSS)	2	
Organic solvent concentration range	ULTRON AF-HILIC-CD	2, 5	Recommended range: 80~95% (Allowable range: 50%~95%)
	ULTRON AF-HILIC-CD (PLSS)	2	
Analytical temperature range	ULTRON AF-HILIC-CD	2, 5	Recommended range: 25~40°C (Maximum: 60°C)
	ULTRON AF-HILIC-CD (PLSS)	2	
Analytical pressure range	ULTRON AF-HILIC-CD	5	Recommended range Up to 20 MPa (Maximum: 30 MPa)
	ULTRON AF-HILIC-CD (PLSS)	2	Recommended range Up to 40 MPa (Maximum: 60 MPa)

※ The degradation of column performance is likely to occur when used at higher temperatures and lower concentrations of organic solvents.

※ Avoid using a column repeatedly near the pressure limit or abrupt change in pressure to prevent shortening of the column life.

3. Shipping Solvent

- ULTRON AF-HILIC-CD series columns are shipped containing 100% acetonitrile. When using a mobile phase containing buffer salts, care must be taken to avoid salt precipitation.

4. mobile phase and sample

- The most suitable mobile phase in HILIC mode is acetonitrile/water or buffer.

[The relative solvent strength for HILIC mode]

Acetone < Acetonitrile < 2-propanol < Ethanol < Methanol < Water

- ULTRON AF-HILIC-CD (PLSS) uses PEEK resin as the column material, so we do not recommend the use of tetrahydrofuran.
- The lower polarity of mobile phase and the higher concentration of organic solvents increase the retention. It is recommended to contain at least 3% water in a mobile phase to enhance the separation reproducibility by forming a stable hydrated layer on the surface of packing material.
- Suitable buffer for HILIC mode is ammonium acetate or ammonium formate. A buffer concentration in the range 10 – 20 mM is recommended. (Depending on separation or solubility, adjustment in the range of 5 – 200 mM can be made.)
- In case of gradient elution, each mobile phase composition should be adjusted to maintain a constant salt concentration during analysis.

- Avoid phosphate salt and other low solubility buffers to organic solvents.
- Ensure to filter samples and mobile phases using a membrane filter with a mesh size of 0.45 μm or smaller before using. Failure to filter mobile phases, etc. can lead to blockages of column filters and increases in analytical pressures.
- Ensure to thoroughly degas mobile phases prior to use. Insufficient degassing of mobile phases can lead to the formation of bubbles inside analytical instruments and columns resulting in problems with analyses.
- It is recommended to dissolve the sample in a solvent that is of the same composition as the initial mobile phase. Dissolving the samples in a stronger solvent than the initial mobile phase will result in distorted peak symmetry and degraded resolution.
- The pH of the sample solution must be set in acceptable pH range for packing material.

5. Precautions for Column Installation

- Before installation the column, replace the liquid in the system with the mobile phase to be used.
(Note: Ensure the compatibility of solvents/buffers when replacing to avoid precipitation of salts. The details about the shipment solvent is described in the column performance report enclosed with the column.)
Tubing must have flat ends and must bottom out in the column end fitting. 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.
- Install the column according to the direction of the arrow.
- Do not remove the column from LC system before the pressure drops zero.

(Precautions for use of ULTRON AF-HILIC-CD (PLSS))

- Do not overtighten the male nuts to the column during installation. This may damage the fittings.
- Please make sure not to overtighten the column end plug when sealing the column.
- Hold the end fitting when installing and removing the column. If the end fitting is loosened, leakage may be caused.

6. Column cleaning and column storage

- Replace the buffer solution of the mobile phase with purified water, and then clean the column using a solution with an organic solvent concentration the same as the mobile phase. Finally, replace the column with 100% acetonitrile. Close the column with end stop plugs tightly and store at room temperature.
- Wash with 50 to 70% acetonitrile for removing polar contaminants.
- If does not improve, purge with acetonitrile / water with 95% -5% gradient or 5% acetonitrile.
- The degradation of column performance is likely to occur when used at lower concentrations of organic solvents. Therefore, please adjust the organic solvent ratio and purge amount.
- Performance of the column should be carried out in accordance with the enclosed "COLUMN PERFORMANCE REPORT" .

ULTRON series packed columns are shipped under highly controlled conditions. However, if you should find any defect, please contact your dealer or Shinwa. Note that Shinwa does not warrant the product against column life or deterioration caused by the failure to follow the above instructions.

