

# YMC-UltraHT<sup>TM</sup> Series

## Instructions for use

### 1. Introduction

Thank you for purchasing a YMC-UltraHT series column.

YMC-UltraHT series is an ODS column based on ultra-high purity silica. The column is designed for ultra-fast analysis.

YMC-UltraHT series can reduce analysis time with excellent separation capacity. Two (2) micron YMC-UltraHT series has high separation capacity across broad flow and pressure ranges compared to 3  $\mu\text{m}$  and 5  $\mu\text{m}$  materials.

YMC manufactures YMC-UltraHT series to strict quality standards and delivers only those columns that pass such stringent tests prior to shipment (please refer to the Test Report and the Column Inspection Report). In order to ensure optimal performance and durability of the column, please follow these instructions.

### 2. Recommendations for column connection, detector settings and data processing considerations

- When the tubing connection area results in unnecessary extra-column volume, the user may experience a decrease in the number of theoretical plates and/or increase in peak shape distortion. Please make sure of the integrity of fluidic connections to the column.
- Fluidic connections between the column, injector, and detector should be as short as is practical and extra-column volumes minimized by using small inner diameter tubing (less than 0.15 mm).
- Please use a detector flow cell designed for use with narrow-bore columns (for example a semi-micro or for micro-bore flow cell). If the volume of the detector flow cell is too large, the sample band expands and peak performance degrades considerably. Optimize the response of detector (time constant) and the data-sampling speed of data-processing equipment in order to accept peaks having short band widths.

### 3. Mobile phase and sample solvent

- The solvent enclosed at shipment is acetonitrile/water (60/40). If you intend to store the column for a long time replace the mobile phase in the column with shipping solvent.
- Solvent should flow in the direction of the arrow as indicated on the column label.
- Aqueous to nonaqueous solvents are applicable. However continuous replacement of solvents, which have wide differences in polarity may degrade the column performance.
- When preparing mobile phase, the pH should be kept within the range of 2.0 to 8.0. When using mobile phase pH near the upper and lower limits, mobile phase containing 10% concentration of organic solvent should be used. The column durability will shorten under certain conditions by temperature and mobile phase composition.
- When replacing the mobile phase, solvents need to be miscible. Depositoin of salts should be carefully monitored and salts should be washed from the column before storage.
- When possible, the sample should be dissolved in a solvent in the same composition as the starting mobile phase composition. Using solvent compositions that are stronger than the mobile phase for sample dissolution may result in distortion of peak symmetry and/or degradation of peak resolution and hence an decrease in analytical reproducibility.
- In order to prevent exposure of the column to excessive pressures, the mobile phase should be filtered through a 0.2  $\mu\text{m}$  membrane filter and sample solution should be filtered through an YMC Duo-Filter (0.2  $\mu\text{m}$ ) before injection.

### 4. Column cleaning ( general method )

- When a mobile phase containing neither buffer nor salts is used and you are trying to remove substances that show great retention on the column, wash the column with a solvent composition of higher organic solvent concentration to remove substances that otherwise would remain in the column. When a lipophilic component is stubbornly retained on the column, the addition of THF to the column wash solvent may be effective in removing strongly retained substances.
- When a mobile phase containing buffer or salts is used, wash as above after replacement of the mobile phase with a mixture of water and organic solvent (at the same ratio as the mobile phase but containing neither buffer nor salts). When a mobile phase containing buffer or salts is used at about 100 mM, the mobile phase can be replaced directly with about 60% acetonitrile aqueous solution.
- After using a column near the usable pH limit, washing the column with water alone may cause column deterioration. Use the mixture of water and organic solvent described above or use about 60% acetonitrile aqueous solution to replace the mobile phase.
- When macromolecules including proteins and sugars adsorb to the column, it is usually difficult to wash out the macromolecules with organic solvents. When a sample containing macromolecules is analyzed, it is preferable to conduct sample pretreatment prior to introducing the sample to the column.

### 5. Other environments

- The upper limit of column pressure is about 50 MPa, and 40 MPa for the length of 30 mm.
- The upper limit of column temperature is 50 °C. However, we recommend using the column between 20 °C and 40 °C because some conditions of usage, such as operating at the extremes of pH of the mobile phase, may shorten the column lifetime especially at the upper temperature limit of the column.