Column Care and Use Instructions
YMC-Pack Diol-NP
YMC-Pack PVA-Sil

1. Introduction

Thank you for purchasing a YMC high-performance liquid chromatography (HPLC) column. YMC HPLC columns, which are manufactured under highly controlled conditions, must pass a series of stringent 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. Column connections

The "WT" or "QT" at the end of the product code indicates the style of column endfittings.
WT = Waters style / QT = Parker style

3. Shipping solvent

Indicated in the COLUMN INSPECTION REPORT. Replace with this solvent for storage. When replacing mobile phases, make sure of the miscibility among the solvents. When the displacement from non-aqueous to aqueous solvent or vice versa, flush the column with more than 10 column volume of compatible solvent like 2-propanol and THF.

4. Mobile phase

In general, the column is used as normal phase. The column can be used as Hydrophilic Interaction Chromatography (HILIC). We recommend using the same column exclusively for separation mode for column lifetime.

【Normal phase】

- In general, alkane (n-hexane or n-heptane), alcohols (methanol, ethanol, 2-propanol), ethyl acetate, dichloromethane, chloroform, are recommended for regular use.
- Addition of alcohol to alkane is basic. Acetonitrile, tetrahydrofuran (THF), dioxane etc. are also usable. When preparing mobile phases, make sure of the miscibility among the solvents.
- When a target compounds is ionic, addition of modifier at trifluoroacetic acid (TFA), acetic acid etc. can improve peak shape and separation reproducibility. High concentrations of modifiers can result in reducing column lifetime. In this case, the same column is not recommended for developing a new method, because exposure to acid or alkali may change the retention characteristics of a column.

【HILIC】

- The most suitable mobile phase is acetonitrile/water or buffer (approx. 90/10 – 60/40). In addition, general water-miscible organic solvents.
- Ammonium acetate or ammonium formate is most recommended as a buffer salt. Usually, 10 – 20 mM final salt concentration in a mobile phase is sufficient. Depending on separation or solubility, adjustment in the range of 5 – 200 mM can be made.

【For all products】

- The correct direction of the solvent flow is indicated by an arrow on the column identification label.
- Non-aqueous or aqueous solvent can be used as a mobile phase. Repetitive replacement among solvents with large difference in polarities might degrade the column performance.
- When the displacement from non-aqueous to aqueous solvent or vice versa, flush the column with more than 10 column volume of compatible solvent like 2-propanol and THF.
- Recommended pH ranges of the column are between 2.0 – 7.5. For PVA-Sil, pH ranges of 2.0 – 9.5 are available. Column lifetime is strongly dependent on pH, temperature, and mobile phase composition. In general, high temperature and concentration of buffer or additive, low organic solvent concentration will shorten column lifetime

5. Column cleaning (general method)

【Normal phase】

- Flush the column with 2-propanol etc.
- Replace with the mixture of *n*-hexane and alcohol etc. for storage.

【HILIC】

- Use the mixture of organic solvent and water with a higher solvent strength than the mobile phase, such as acetonitrile/water (50/50), to remove strongly retained substances. Usually, a solvent consisting of 50% water is sufficient to remove polar contaminants. If further cleaning is required, flush the column with acetonitrile/water (5/95).
- Once macromolecules such as proteins or polysaccharides are adsorbed onto the gel, they are hardly removed, even if solvents with high eluting capability are used. To avoid contamination of the column by them, conduct sample pretreatment carefully before introduction into the column. Alternatively use a guard column.
- Replace with the mixture of acetonitrile etc. for storage.

6. Other environments

- The operating pressure should be kept under 20 MPa (2900 psi) for 150 mm or less than 150 mm length column, under 25 MPa (3625 psi) for 250 mm length column, under 10 MPa (1450 psi) for 10 mm I.D. or more than 10 mm I.D. column.
- To prevent exposure of the column to excessive pressure, the sample solution should be filtered through a 0.2 μm membrane or smaller to remove particulates. We recommend using a pre-column filter to prevent the column frit from being clogged with samples.
- Avoid using a column repeatedly near the pressure limit or abrupt change in pressure to prevent shortening of the column life.
- Adjust the flow rate appropriately because the pressure changes depending on the column length, temperature, types of organic solvent etc.
- The upper limit of column temperature is 50 °C. However, we recommend using the column at 20 – 40 °C, because column lifetime varies depending on conditions such as pH.

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