Care & use of Avantor® ACE® UltraCore BIO columns

Thank you for purchasing this Avantor[®] ACE[®] UltraCore BIO column. Every Avantor[®] ACE[®] UltraCore BIO column is individually manufactured and validated to exceed stringent specifications. The following measures will enhance its performance and lifetime.

PRODUCT DESCRIPTION

Avantor® ACE® Ultracore BIO columns contain high purity modified silica with a wide-pore (300 Å and 500 Å) semi porous particle design. The superficial porous particle provides a thin porous shell of high-purity silica surrounding a solid silica core. This particle design exhibits very high column efficiency due to the shallow diffusion paths in the thin porous shell and the small overall particle size of 2.5 or 3.5 microns. The densely bonded, end-capped stationary phases provide a stable, reversed-phase packing that can be used for separating high molecular weight compounds such as proteins, mAbs and ADCs. The certificate shows a sample Quality Assurance test chromatogram for a 2.1 mm x 150 mm column. Also on the certificate is the actual Column QC chromatogram and performance results for the individual column.

Performance testing

It is recommended that the performance of columns is tested on arrival and periodically during use.

COLUMN INSTALLATION

System compatibility

Avantor® ACE® UltraCore® BIO columns are compatible with UHPLC instrumentation offered by all leading manufacturers. Avantor® ACE® UltraCore® BIO columns are compatible for use on HPLC systems, provided that system pressure limits are not exceeded when using smaller particles and/or elevated flow rates. For improved compatibility with a Waters Acquity UHPLC system (containing a 1/16" Waters Acquity fitting and ferrule on the inlet tubing), the use of a pre-column filter incorporating the unique Waters Acquity column port profile (p/n EXL-PCF10/ACQ, 10 pack) is additionally recommended.

System dead volume

Always ensure that the injection valve and flow cell introduce the smallest possible dead volume. Connections between injector, column and detector should be kept as short as possible, preferably using tubing with an internal diameter of 0.010" (0.25mm) or less.

Column connection

For UHPLC applications (up to 15,000psi / 1000bar), the use of Avantor[®] ACE[®] Excel UHPLC Column Connectors (p/n EXL-CC10, 10 pack) are recommended.

At the outlet end of the UHPLC column (where pressure demands are lower, but a correct connection remains important), or for HPLC applications Avantor® ACE® Fingertight HPLC Column Connectors (p/n ACE-CC10, 10 pack) may alternatively be used. The direction of flow is marked on the column.

Mechanical damage

Protect the column from mechanical shock. A flow rate ramp is recommended. Dropping a column can impair its performance.

Equilibration

Flush onto the desired mobile phase (ensure that precipitation does not occur). The column is supplied in 100% acetonitrile. Ensure that the column is fully equilibrated prior to starting an analysis.

Precolumn filters

The use of a precolumn filter is strongly recommended to prevent inlet frit blockage. Avantor ACE® Excel UHPLC precolumn filters (p/n# EXL-PCF05 or EXL-PCF05/ACQ for Waters Acquity systems) can operate at pressures up to 15,000psi (1000bar) without affecting column performance or retention, due to their ultra low dispersion design.

Guard Cartridges

The use of guard cartridges is recommended to prevent both inlet frit blockage and irreversible sample adsorption onto the top of the column. Guard cartridges are available for Avantor® ACE® UltraCore® BIO columns - for assistance identifying the correct guard cartridge for this column, please contact our Technical Support Department at chromsupport@avantorsciences.com

OPERATIONAL GUIDELINES

Solvents

Use only UHPLC/HPLC grade solvents and freshly prepared aqueous buffer solutions to minimise bacterial growth. A slip-on pump filter will remove extraneous particles.

Mobile phase pH and temperature

Avantor[®] ACE[®] Ultracore BIO columns have been application tested at operational temperatures up to 90 °C and using low pH mobile phases (pH 2 – 2.5). They have been verified to exhibit long column life at 60 °C at intermediate pH (up to pH 6). For the longest lifetime we would recommend operation at pH 6 - 9 using 40 °C. The column should not be used above pH 9.

Pressure

Avantor® ACE® Ultracore BIO columns in 2.5 µm particle size are stable to operating pressures up to 1000bar for 2.1mm ID, 1000bar for 3.0mm ID and 600 bar (9000 psi) for 4.6 mm ID. Avantor® ACE® Ultracore BIO columns in 3.5 µm particle size are stable to operating pressures up to 600bar (9000 psi). Should the operating pressure of the column suddenly increase beyond normal levels, reversing the flow direction of the column may be attempted to remove debris on the inlet frit. To remove strongly retained materials from the column, flush the column in the reverse direction with very strong solvents such as 100% of the organic component of the mobile phase in use. A mixture (95/5 v/v) of dichloromethane and methanol is often effective at this task. Extreme cases may require the use of very strong solvents such as dimethylformamide (DMF) or dimethylsulfoxide (DMSO).

Column warranty

All columns are warranted to meet the specifications stated on the Test Chromatogram. Removal of the column end fittings will automatically invalidate the column warranty.

Storage

Wash out any buffer (ensure that precipitation does not occur) and flush onto the storage solvent defined overleaf. Replace the end-stops to prevent the packing bed drying out and store in a cool area. Store in 100% acetonitrile with end stops securely in place.

Safety and disposal

This column contains amorphous silica which may be hazardous to health if the column is unpacked and the silica allowed to dry. The silica presents no hazard whilst contained within the column. When the column has reached the end of its useful life, dispose of it in a similar manner to the samples that have been injected onto it. Alternatively, please contact our Technical Support Department at **chromsupport@avantorsciences.com** for details of our column disposal program.