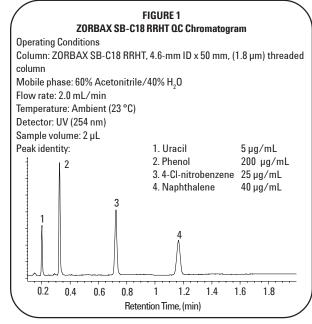


General Description

ZORBAX SB-C18 RRHT threaded columns are specially designed for higher pressure operation (up to 600 bar) and are packed with a high performance microparticulate C18 packing for high-speed reversed phase HPLC. The StableBond packing is made by chemically bonding a sterically-protected C18 stationary phase to a specially prepared, high purity ZORBAX porous silica microsphere. The special ZORBAX silica support is designed to reduce or eliminate strong adsorption of basic compounds. The densely covered, sterically protected, diisobutyl n-octadecylsilane stationary phase is chemically stable and gives longer column life. As a result, ZORBAX SB-C18 is a stable, reversed-phase packing that can be used for basic, neutral, or acidic samples. It is particularly well suited for use with aggressive mobile phases (for example, pH <2, high ionic strength, ion-pair additives, etc.) since the steric protection of the bonded phase resists degradation caused by such mobile phases. This material has also been shown to be stable when operated at temperatures up to 90 °C. These characteristics are particularly important for use in methods that need longterm stability and reproducibility. ZORBAX SB-C18 is especially suited in applications that use highsensitivity detectors that require low backgrounds (for example, mass spectrometers).



Agilent ZORBAX SB-C18 RRHT Threaded Column

Datasheet

The uniform, spherical, ZORBAX SB-C18 particles have a controlled pore size of 80Å. Columns are loaded to a uniform bed density using a proprietary, high-pressure, slurry-loading technique to give optimum column efficiency.

Column Characteristics

A typical Quality Control test chromatogram for a 1.8 μm ZORBAX SB-C18 RRHT 4.6 mm ID \times 50 mm threaded column is shown in Figure 1. The actual QC test and performance of your column is described on the Column Performance Report enclosed with your column.

Safety Considerations

- All points of connection in liquid chromatographic systems are potential sources of leaks. Users of liquid chromatographic equipment should be aware of the toxicity or flammability of their mobile phases.
- These RRHT assembled columns are mechanically stable and have been tested to very high pressures to assure safe lab operation on a variety of LC instruments. The 2.1- and 3.0-mm ID columns will support 20,000 psi (1300 bar) operation and 4.6-mm ID columns will support 16,000 psi (1000 bar) operation. Opening columns may compromise these pressure limits. Chromatographic performance has not been tested above 600 bar.
- Because of its small particle size, dry ZORBAX packings are respirable. Columns should only be opened in a well-ventilated area.

Operational Guidelines

- The direction of flow is marked on the column.
- While generally not harmful to the column, reversing flow should be avoided except to attempt removal of inlet pluggage (see "Column Care" section).
- These columns are packed and assembled for high pressure (up to 600 bar) use. Disassembling the column will degrade column performance.
- ZORBAX SB-C18 is compatible with water and all common organic solvents.
- \bullet Avoid use of columns below pH 0.8 or above pH 8.0.
- Maximum operating pressure is 600 bar (9000 psi).
- Maximum operating temperature is 90 °C.
 NOTE: StableBond columns are designed for high stability at low pH (for example, pH <5). However, all silica-based packings have some solubility in pH >6 aqueous mobile phases. Therefore, when using

silica-based columns under conditions of pH >6, maximum column lifetime is obtained by operation at low temperatures (<40 °C) using low buffer concentrations in the range of 0.01 to 0.02M. Column stability at pH >6 is also enhanced by avoiding phosphate and carbonate buffers [ref.: H.A. Claessens, M.A. van Straten, and J.J. Kirkland, J. Chromatogr. (A), 728 (1996) 259].

Mobile Phase Selection

The bonded stationary phase is nonpolar in nature and is best used with mobile phases such as methanol/water or acetonitrile/water mixtures. Increasing the amount of organic component usually reduces the retention time of the sample. Due to the relatively high viscosity of recommended mobile phases, increased efficiency can be achieved with the use of column temperatures in the range of 40-65 °C. Gradient elution techniques for this packing often use 5% methanol or acetonitrile in water as the initial solvent, and 100% methanol or acetonitrile as the final solvent. Additional information on solvent selection may be found in Chapters Six and Seven, Introduction to Modern Liquid Chromatography, Second Edition, L. R. Snyder and J. J. Kirkland, (John Wiley & Sons, 1979), and Chapters Six, Seven and Eight, Practical HPLC Method Development, Second Edition, L.R. Snyder, J.J. Kirkland, and J.L. Glajch, (John Wiley & Sons, 1997).

Applications

ZORBAX SB-C18 can be used with basic, neutral or acidic analytes. For many basic compounds, it will normally not be necessary to use basic modifiers, such as triethylamine, to achieve efficient, symmetrical peaks. However, very basic compounds may require the addition of basic modifiers such as 10-20 mM dimethyl-octylamine or 20-30 mM triethylamine. Such samples are often best chromatographed with mobile phases of pH \leq 3. One highly recommended mobile phase for very basic compounds is 0.1% trifluoroacetic acid adjusted to pH=3 with triethylamine and an appropriate concentration of methanol or acetonitrile.

ZORBAX SB-C18 can be used at 90 °C at low pH and is therefore a good choice for higher temperature separations. High temperature can reduce mobile phase viscosity, lower operating pressure and change band spacing (selectivity).

Column Care

Samples that contain particulate matter may plug the column inlet frit and should be filtered before injection into the column. If solvent flow appears to be restricted (unusually high column-back-pressure), check first to see that solvent flow is unobstructed up to the column inlet. If the column has the restriction, there may be particulate matter on the inlet frit. An attempt should be made to remove any inlet debris by back-flushing 25-30 mL of mobile

phase through the column. If this fails to return the column to near its original back-pressure, the column should be replaced.

To remove strongly retained materials from the column, flush the column with stronger (less polar) solvents. Solvents such as methanol, acetonitrile, or a 95/5 mixture of dichloromethane and methanol should remove most highly retained compounds. In extreme cases, dimethyl sulfoxide or dimethylformamide at low flow rates may also be used for this purpose. When switching between solvents with vastly different polarities, it may be necessary to first purge the column with a mutually miscible solvent such as isopropanol.

Since columns have 3/8-inch end nuts, a short 3/8-inch wrench should be used to attach the columns to the instrument to avoid any additional tightening of the end fittings. Over-tightening the end fittings will cause damage and require column replacement.

Storage Recommendations

Long term storage of silica-based, bonded phase columns should be in a pure organic solvent, preferably an aprotic liquid such as 100% acetonitrile. If the column was previously used with a buffered mobile phase, the buffer should first be removed by purging the column with 20-30 column volumes of a 50/50 mixture of methanol or acetonitrile and water, followed by 20-30 column volumes of the pure solvent. Before storing the column, the end-fittings should be tightly capped with end-plugs to prevent the packing from drying out.

Columns may be safely stored for short periods in most mobile phases. However, to protect equipment, it is desirable to remove salts from the instrument and column by purging the column with the same mobile phase without the buffer (for example, using $60/40~\rm{ACN/H_2O}$ to remove a $60/40~\rm{ACN/0.02}$ M phosphate buffered mobile phase). Re-equilibration is rapid with the original mobile phase when using this approach, and any danger of corrosion from the salts is eliminated.

Agilent Ordering Information

For more information on our products, visit our Agilent Technologies home page on the World Wide Web at: http://www.agilent.com/chem/supplies For Technical Support in the US and Canada, call 1-800-227-9770 or call your local Agilent sales office.

