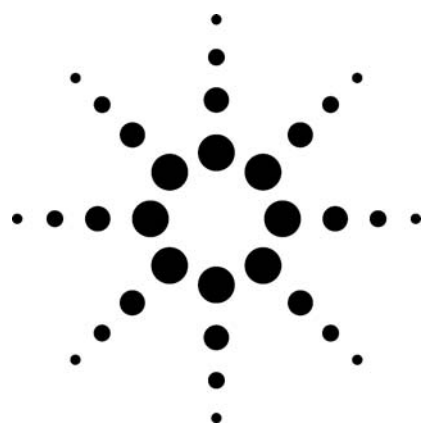


Agilent ZORBAX Rx-Sil RRHT Threaded Column

Data Sheet



General Description

ZORBAX Rx-Sil RRHT threaded columns are specially designed for higher pressure operation (up to 600 bar) and are packed with a high-performance microparticulate silica packing for high-speed liquid-solid adsorption chromatography. ZORBAX Rx-Sil particles are produced by the agglutination of colloidal silica to form spherical particles of uniform diameter and pore size. However, ZORBAX Rx-Sil is specially treated to create a highly homogeneous surface throughout the silica support. This homogeneity is achieved by utilizing ultra-high-purity silica (less than 100 ppm total of impurities such as sodium, aluminum, iron, etc.) and by applying patented technology for fully hydroxylating the silica surface. As a result, ZORBAX Rx-Sil can be used for basic, neutral, or acidic samples. It is particularly well suited for use with basic samples, since these solutes can be chromatographed without peak tailing or irreversible adsorption.

The uniform, spherical, ZORBAX Rx-Sil 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.

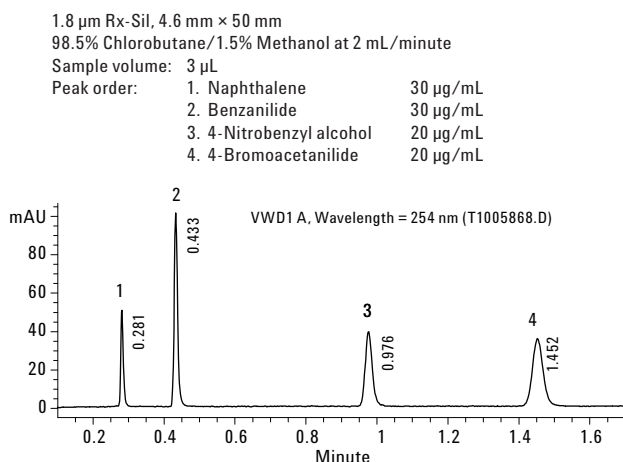


Figure 1. ZORBAX Rx-Sil RRHT QC chromatogram.

Column Characteristics

A typical quality control test chromatogram for a 1.8-mm ZORBAX Rx-Sil RRHT 4.6 mm id × 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 ensure safe lab operation on a variety of LC instruments. The 2.1- and 3.0-mm id columns will support 20,000 psi (1,300 bar) operation and 4.6-mm id columns will support 16,000 psi (1,000 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 blockage (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 Rx-Sil columns are shipped containing hexane. Care should be taken not to pass any mobile phase through the column that might cause a precipitate.
- ZORBAX Rx-Sil is compatible with water and all common organic solvents.



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- Avoid use of this column below pH 0.8 or above pH 8.
- Maximum operating pressure is 600 bar (9,000 psi).
- Maximum operating temperature of unbonded silica columns is typically limited only by the temperature limits of the mobile phase with the exception of the following note.

NOTE: ZORBAX 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 0.02 M. 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

ZORBAX Rx-Sil is compatible with all common organic solvents. When switching between solvents with vastly different polarities, it is advisable to first rinse the column with a mutually miscible solvent such as isopropanol. The eluotropic solvent series described by Snyder is helpful in selecting mobile phases that will elute compounds in the proper retention range (normally, k' between 1–10) and with the highest selectivity. To maintain a reproducible activity of the silica surface, it is often desirable to employ water-modified mobile phases. Snyder and Kirkland describe a method for adjusting the water content for non-polar solvents in a convenient and reproducible manner. Alternatively, it is often possible to help maintain reproducibility by adding 0.1–1% alcohol (such as methanol) or 1–3% acetonitrile to the primary solvent. In the case of methylene chloride, a methanol concentration of about 0.15% (v/v) is equivalent to 50% water saturation in deactivation of the packing. When gradient elution is employed, both primary and secondary solvents should be modified with alcohol or acetonitrile and at least 30 column volumes of solvent should be allowed to flow through the column after the completion of each run to allow the column to re-equilibrate to its original activity level. 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 Rx-Sil 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 ~ 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 Rx-Sil can be used at 80 °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). Rx-Sil columns may also provide unique selectivity when separating aromatic compounds.

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 (more polar) solvents. Even water may be used without damaging the column. The column should be flushed with mobile phase after cleaning (40 column volumes) to equilibrate the column. Continue to pump mobile phase through the column until reproducible k' values are obtained for a test sample. 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

To avoid potential metal corrosion, long-term storage of any HPLC column in halogenated solvents (for example, butyl chloride, methylene chloride, etc.) should be avoided. If the column has been used with a buffered mobile phase, the column should be purged with 20–30 column volumes of acetonitrile and water followed by 20–30 column volumes of the pure organic solvent. Storage of unbonded silica columns in most other liquids is typically acceptable.

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