Pursuit UPS columns are designed for ultimate performance on any ultra-high pressure LC system. Pursuit UPS columns are available in multiple chemistries and on both 1.9 and 2.4 micron silica for tailored application in the high throughput, ultra-high pressure HPLC environment.

Pursuit UPS $^{1.9}$ columns deliver sub 2 μm efficiencies for those applications where sensitivity, resolution, and throughput are critical.

Pursuit UPS ^{2.4} columns couple highly efficient, fast separations with superior ease of use. They are recommended for viscous solvent applications or particulate laden samples where lifetime and backpressure are important concerns.

Pursuit UPS columns are designed for enhanced performance with ultra-high pressure, low dead volume chromatographic systems. To achieve similar results on conventional systems, please use our Pursuit Ultra products. See www.varianinc.com for details.

I. COLUMN USE

Column Equilibration

The columns are shipped in acetonitrile based solvents. Equilibrate the column with a minimum of ten column volumes of the mobile phase until constant baseline and backpressure are achieved.

pH Range

The recommended pH range is between 1.5 and 10. The column lifetime depends on the temperature and composition of the mobile phase, especially type and concentration of buffer used. The combination of extreme pH values, high temperature and high backpressure will reduce the column lifetime.

Solvent Filtration

To maintain low working backpressures and extend column lifetime, filter all LC solvents through at least a 0.2 μm particulate filter. Filtration of solvents with a 0.45 μm filter is insufficient for maximum column lifetime.

Pressure

Pursuit UPS columns are suited for use with ultra-high pressure chromatographic systems.

Temperature

The working temperature of Pursuit UPS is between 20-60 °C. Selectivity and efficiency may improve when working at higher temperatures due to changes in the solvent viscosity and the rate of mass transfer. However, column use at high temperatures and extreme pH may decrease column lifetimes.

Cleaning and Regeneration

If the column is contaminated by particles or compounds from the matrix, changes in peak shape (fronting, tailing), peak splitting, shoulders, changes in retention times and increasing backpressure might occur. If this happens, purge the column 40-60 minutes at 40% of the optimum flow rate with subsequent solvents:

5% MeOH in water MeOH ACN THF MeOH mobile phase.

Column temperature can be increased during this process to further increase the cleaning efficiency of the neat solvents.

Storage

For periods longer than 3 days, store the columns in 100% acetonitrile. Do not store in buffered eluents. If the mobile phase contains a buffer, then flush the column with 10 column volumes of ultra-HPLC grade organic modified water and replace with 100% acetonitrile for storage. Completely seal the column to prevent solvent evaporation and drying out the column bed.

II. SAMPLE PREPARATION

Sample Solvent

To achieve the best possible peak shapes, the sample should be dissolved in the mobile phase or in a solvent that is weaker than the mobile phase.

Particulate Removal

Filtration of the sample with a 0.2 μ m particle filter will avoid backpressure build-up and is recommended. Varian's Captiva filtration products are available in different membrane materials and pore sizes (www. varianinc.com).



Pursuit[™] UPS ^{1.9} and Pursuit UPS ^{2.4} HPLC Columns

Matrix Removal

Sample matrix components, especially those from bioanalytical samples, often contribute to a degradation in column performance. Insufficient removal of these components can lead to high backpressures, system contamination, and reduced column lifetimes. Care should be taken to ensure removal of common matrix interferences. For most samples, solid-phase extraction (SPE) is the method of choice. For SPE method assistance, please contact helpdesk@varianinc.com.

For bioanalytical samples, phospholipids can be removed by filtration using Captiva[™] ND^{Lipids}. Please contact your Varian sales representative for more information.

III. GENERAL CONSIDERATIONS

Most ultra-high pressure systems have been optimized for the following:

Injector

The maximum injection volume should be no more than 1% of the column's void volume. The injection system should have a low volume design. For isocratic separations, injection volumes should be kept as low as possible (< 2 μ L). For gradient runs, sample volumes are less critical, especially when the sample is diluted in a weak solvent.

Tubing

Extra-column volume should be minimized wherever possible. Tubing lengths and diameters should also be minimized and care should be taken in making all tubing and column connections.

Detector

A detector cell with the correct cell volume is necessary. The best choice would be a cell volume that is smaller or equal to $1.2 \ \mu$ L. The detector response time should be set to the fastest setting (100Hz) to ensure sufficient data across fast eluting peaks and to minimize artificial peak broadening due to a slow sampling rate.

Other System Parts

When speed is critical, system volume prior to the injector should be optimized. Pay attention to the mixer volume and tubing volume prior to the injector to avoid gradient delays.

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