

## General Guideline for the Use of Silica Based Reversed Phase HPLC-Columns

Because of their vast applicability and their easy manageability silica based reversed phase columns are the most commonly used columns in liquid chromatography. In general these columns are rugged as long as some basic rules are being observed.

### Product Specifications:

Refer to the enclosed HPLC Column Performance Report for specific information and test results for your column.

Pressure Ratings: 0 – 3.500 psi (many columns up to 5.000 psi)

pH-Stability: 2 – 7,5

Temperature Limits: 5 – 60 °C

Shipping Solvent: Acetonitrile/water (80:20)

### Using Silica Based RP Columns

#### Column Life:

The column life depends on the cleanliness of sample and mobile phase. Column life can be extended by pre-filtering the mobile phase and using degassed solvents.

Use guard columns to prevent particulates and highly adsorptive compounds from reaching the column.

Impure samples can lead to a decrease in column performance. A partially clogged inlet frit may result in unusually high operating pressure.

Clear a clogged frit by reversing the column flow for 10 – 20 mL or changing the inlet frit system (details see the backside of your column performance report).

#### Changing Mobile Phases:

When changing from one mobile phase to another, determine if the mobile phases are miscible, and that precipitation will not occur. Re-equilibrate the column by pumping 10 or more column volumes of mobile phase through the column. Equilibration is complete when the base line is stable and peak location is reproducible. Commonly used mobile phases are acetonitrile, methanol, tetrahydrofuran (THF) or isopropanol in combination with water or aqueous buffers. For the use of mobile phases with less than 5 % organic modifier AQ-columns are recommended.

#### Column Storage:

For short term storage flush your column with your mobile phase without buffer or other additives. (i.e. mobile phase = 50% MeOH, 50% buffer; flush with 50% MeOH, 50% water.

For overnight storage keep the mobile phase flowing at 0.1-0.2 mL/min.

Return the column to acetonitrile for long term storage (avoid precipitating buffer salts).