

RECOMMENDED COLUMN CLEANING AND REGENERATING PROCEDURES

Use the cleaning routine that matches the properties of the column and what you believe is contaminating it. Flush columns with 20 column volumes (80 mL total for 4.6 x 250 mm column) of HPLC-grade solvents. Run columns in reverse flow direction, with the outlet disconnected from the detector. Cleaning efficiency is increased by increasing mobile phase temperature to 35-55 °C. If the column performance is poor after regenerating and cleaning, call us.

Silica-based particles

Non-polar-bonded **phases** (Carotenoid, C18, Octyl, YMCbasic™, J'sphere™, Phenyl, Butyl, TMS):

Polar Samples

1. Water
2. Methanol
3. THF
4. Methanol
5. Water
6. Mobile phase

Non-polar Samples

1. Isopropanol
2. THF
3. Dichlormethane
4. Hexane
5. Isopropanol
6. Mobile phase

Proteinaceous Samples

- Option 1: Inject repeated aliquots of DMSO
 Option 2: Gradient of 10 to 90% B where:
 A = 0.1% TFA in water
 B = 0.1% TFA in CH₂CN
 Option 3: Flush column with 7M guanidine
 HCl, or 7M urea

Polar-bonded **phases** (Cyano, Diol, Amino, PVA-sil™, Silica):

Polar Samples

1. Water
2. Methanol
3. THF
4. Methanol
5. Water
6. Mobile phase

Non-polar Samples

1. Chloroform
2. Methanol
3. Dichlormethane
4. Heptane or Isocyanate
5. Isopropanol
6. Mobile phase

Polymer-based particles: Polymer C18™

1. Flush column with mobile phase but omit buffers or salts (i.e. just organic and water, acetonitrile is preferable)
2. Run a gradient to 100 % organic
3. Flush with twenty column volumes of THF
4. Flush with twenty column volumes of acetonitrile
5. Run a gradient back to starting mobile phase conditions, omitting buffers and salts
6. Re-equilibrate in mobile phase