Column Cleaning Procedures

Changes in peak shape, elution time, or elevated column pressure may be resolved by cleaning the column. This section describes general indications of column deterioration and column cleaning procedures.
For details of column cleaning procedures, refer to each column's specific operation manual.

Typical indicators of column deterioration possibility

1. Elevated column pressure
2. Abnormal peak shapes (broadening, leading, or tailing) and split peaks
3. Change in retention time
4. Unstable baseline

Selection guide to the cleaning solvent

Solvents capable of dissolving the adsorbed substances

Solvents with high eluting power (variable depending on separation mode)

*Use the solvent specified in the operation manual.

Standard cleaning procedures

For an efficient cleaning, reverse the direction and reduce the flow rate to 1/3 of the regular flow.

Columns for reversed phase chromatography	Clean the columns with solvent containing higher concentration of organic solvent such as methanol, acetonitrile, or THF. (In case of using buffer as a mobile phase, miscibility of the buffer solution and the organic solvents need to be checked)
Columns for sugar analysis chromatography	**[Ligand exchange columns (SUGAR series)]** • **In case of counter ion detachment** Flush or inject solvent containing the salt corresponding to the modified counter-ligand. **[Polymer-base amino columns (NH2P series)]** • **In cases where an acidic substance has been bound to the amino functional group** Flush with solvents in the following sequence: water, 0.1M perchloric acid (aq.), water, 0.1M NaOH (aq.), water, and mobile phase.
Columns for aqueous SEC (GFC) chromatography	• **In cases where an ionic substance has been adsorbed** Use a solvent with higher salt concentration or solvent with different pH from the mobile phase. • **In cases where a hydrophobic substance has been adsorbed** Use a solvent containing organic solvent. (In case of using buffer as a mobile phase, miscibility of the buffer solution and the organic solvents need to be checked)
Columns for ion exchange chromatography	• **In cases where an ionic substance has been adsorbed** Use a solvent with higher salt concentration or solvent with different pH from the mobile phase. • **In cases where a hydrophobic substance has been adsorbed** Use a solvent containing organic solvent. (In case of using buffer as a mobile phase, miscibility of the buffer solution and the organic solvents need to be checked)
	• **In cases where protein have been adsorbed** Inject 1-2 mL of 0.1 M NaOH (aq.) or 30% (v/v) acetic acid (aq.) several times.
Columns for hydrophobic interaction chromatography	• **In cases where protein have been adsorbed** Inject 1-2 mL of 0.1 M NaOH (aq.) or 30% (v/v) acetic acid (aq.) several times.

*The required volume of the cleaning solvent is 5-10 times the column volume.
*Avoid pressure elevation during the cleaning.
*The cleaning is limited and does not guarantee the full regeneration of the column to its original condition.

For your information

One typical cause of the column pressure elevation is the clogging of solid substances at the inlet filter of the column. In this case, reverse the direction and reduce the flow to 1/3 of the regular flow rate. This may remove the solid substance causing the elevated pressure.

*Use the solvent specified in the operation manual.