

SurePac Protein RP MDi and SurePac Oligo RP MDi columns

Getting started

Prior to using the Thermo Scientific™ SurePac™ Protein Reversed-Phase Monodisperse and Inert (MDi™) Column and Thermo Scientific™ SurePac™ Oligo Reversed-Phase Monodisperse and Inert (MDi™) Column, review all the information in this section on column operation. Following these specifications for your column will help to ensure the column performs as it is intended and maximize the lifetime of your column. For more detailed information on column use please reference the column manuals online.

Column use and physical specifications

To ensure that you do not damage the column hardware or packed bed, take care to operate within the limits of the column. Table 1 indicates the operational limits for each column format in terms of flow rate, maximum column pressure drop from inlet to outlet, temperature, and mobile phase pH.

Table 1. Column use and physical specifications

Column dimension (mm)	Recommended flow rate ¹ (mL/min)	Max. column pressure drop ² psi (bar)	Temp. (°C)	pH
0.3 × 50	0.001–0.012	8000 (552)	5–90	2–10
2.1 × 20	0.2–1.0	6500 (448)	5–90	2–12
2.1 × 50	0.2–0.6	6500 (448)	5–90	2–12
2.1 × 100	0.2–0.6	6500 (448)	5–90	2–12

¹The maximum usable flow rate is dependent on backpressure. Ensure the column pressure does not exceed the specified pressure limit.

²The column pressure drop for a given flow rate is calculated as the pressure of the system with column minus the pressure of system with union in place of column.

Table 2. Column specifications

Parameter	SurePac Protein RP MDi column	SurePac Oligo RP MDi column
Common mobile phases*	For LC-UV experiment - Mobile phase A: H₂O/TFA (99.9:0.1 v/v) - Mobile phase B: Acetonitrile/H₂O/TFA (90:9.9:0.1 v/v/v) For LC-MS experiment - Mobile phase A: H₂O/FA (99.9:0.1 v/v) - Mobile phase B: Acetonitrile/H₂O/FA (90:9.9:0.1 v/v/v)	For LC-UV experiment - Mobile phase A: 0.1 M TEAA or 0.1 M HAA - Mobile phase B: Acetonitrile For LC-MS experiment - Mobile phase A: 15 mM DBA, 25 mM HFIP, pH 9.0 - Mobile phase B: Methanol
Organic solvent compatibility**	Compatible with 100% acetonitrile, isopropanol, and methanol	
Cleaning agents	- Column washing procedure: carry-over may occur if the system is not clean or previous sample may not have completely eluted from the column. Wash the system and the column with acetonitrile/0.01 M NaOH (90:10 v/v). - Please reference column manual for further guidance on column cleaning.	
Storage solution	- Short term (< 24hr): Mobile phase - Long term (≥ 24hr): Acetonitrile/H₂O (50:50 v/v)	

*TFA: Trifluoroacetic acid, FA: Formic acid, TEAA: Triethylammonium acetate, HAA: Hexylammonium acetate, DBA: Dibutylamine, HFIP: 1,1,1,3,3,3-Hexafluoro-2-propanol.

**Acetonitrile and methanol have viscosity maxima when mixed with water at certain ratios. This may cause unexpectedly high pressure. Always use low flow rates until the pressure behavior is understood when using these chemicals. Mixtures of ACN and MeOH should be introduced and removed gradually from the column using a gradient over 20 minutes to ensure a sharp viscosity front does not result in a rapid pressure difference in-column that may damage the packed bed.

Additional requirements for safe column operation

- Always set up the mobile phase flow direction as indicated on the column tag.
- Avoid exposing the column bed to sharp pressure fluctuations that may disrupt the column bed.
- When starting, stopping, or changing the flow rate, a flow ramp rate (mL/min²) of ~1/3 of the maximum flow rate for the specific column format is recommended.
- Avoid leaving the column in 100% aqueous mobile phase for an extended period of time.
- For increased 0.3 × 50 mm format longevity and result consistency, it is recommended to use fresh Thermo Scientific™ nanoViper™ Fingertight Fittings in combination with a torque wrench (PN 6250.2110) for easy and consistent column attachment. Regularly check for nanoViper fingertight fitting thread deterioration. If thread screw-in resistance increases, a change of nanoViper fingertight fitting is recommended. 20 µm and 50 µm internal diameter viper connections are recommended.

Recommended buffers and cleaning agents

Please consult the table below for recommended buffer conditions to achieve optimal separations and maintain good column performance throughout its lifetime.

Column conditioning

Your column has been designed to minimize secondary interactions and for low carryover. Depending on the nature of your sample, column conditioning may be required prior to achieving optimal performance. To quickly condition your column, we recommend performing 2-3 sample overload injections of 10× your standard sample loading and standard gradient method.

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