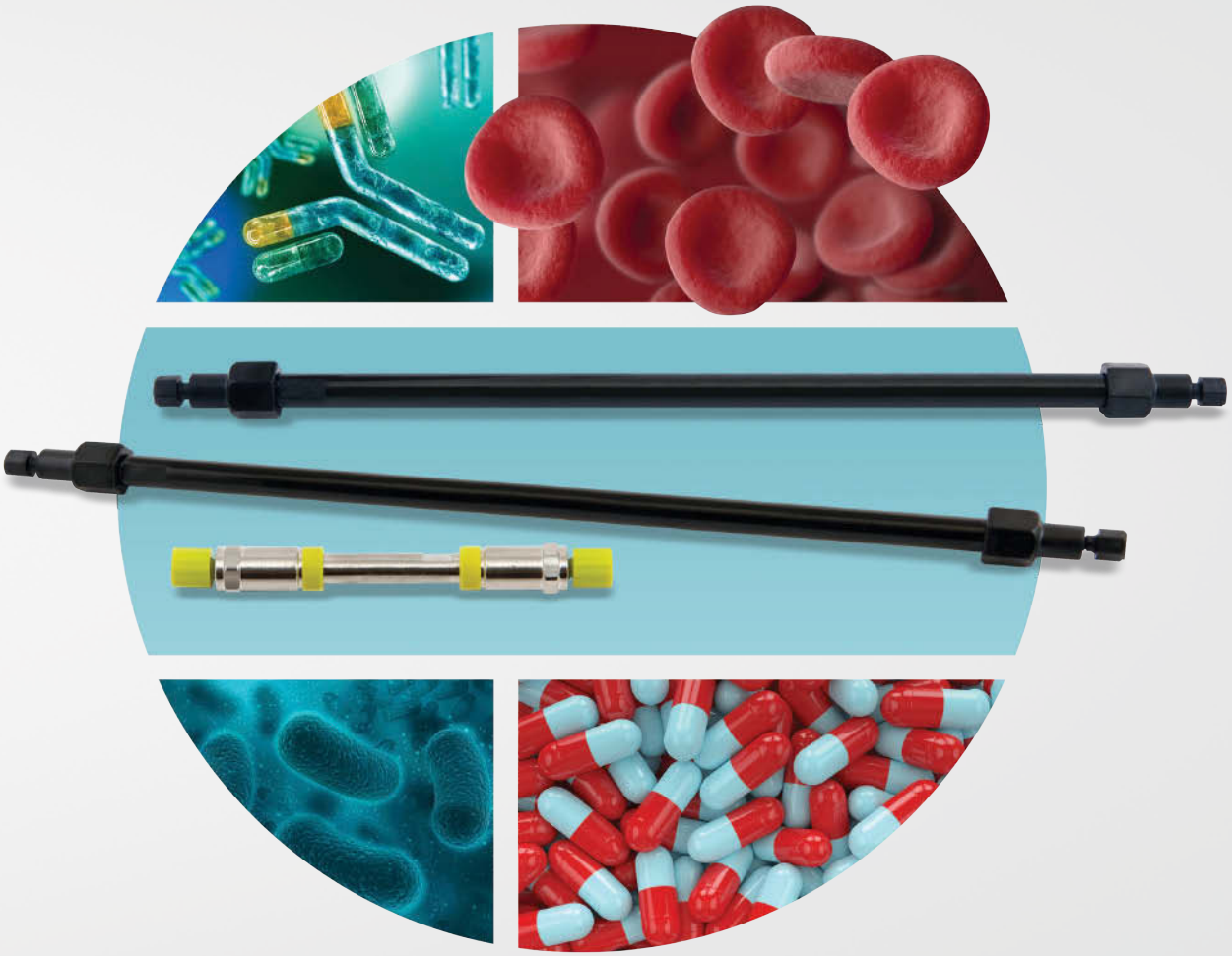
Thermo Fisher SCIENTIFIC



BioLC columns

Connected chromatography solutions

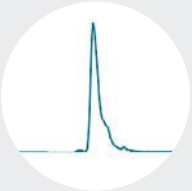
BioLC columns and accessories

thermo scientific

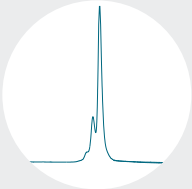
Introduction

Your complete tool kit

Thermo Fisher Scientific has innovative Thermo Scientific™ BioLC columns for each step of your therapeutic protein characterization, no matter how challenging your separation. Here is just one example, a fully characterized model sample of pertuzumab. Discover our full range in this catalogue.

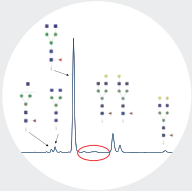


Intact or subunit analysis
Thermo Scientific™ MAbPac™ RP Column is ideal for intact and subunit analysis by MS or UV detection. The polymeric packing material offers column longevity, high resolution and the wide pores to allow for low carryover profiling of your sample.

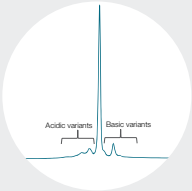


Oxidation monitoring
Deduce protein folding errors or charge-neutral amino acid modifications with the Thermo Scientific™ MAbPac™ HIC-20 Column. Our range of innovative Hydrophobic Interaction Chromatography (HIC) chemistries deliver native separations not seen on other columns.

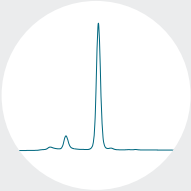
Peptide mapping
Experience reproducible peptide mapping and quantitation. The combination of rapid digestion from the Thermo Scientific™ SMART Digest™ Kit and separation with the high resolution Thermo Scientific™ Hypersil™ GOLD Peptide Column delivers outstanding, reproducible and efficient peptide mapping separations.



Released glycan analysis
Fully characterize your released N-glycans with the Thermo Scientific™ Accucore™ 150 Amide-HILIC Column. This solid core column offers high resolution, durability, and the ability to run separations at lower temperatures to reveal the complete glycan profile.



Charge variant analysis
For charge variant analysis by LC-UV or LC-MS/MS Thermo Scientific™ ProPac™ 3R SCX and SAX Columns deliver outstanding resolution on a highly robust, reproducible and high-resolution platform. Combine ProPac 3R SCX columns with our proprietary Thermo Scientific™ CX-1 Gradient Buffer formulations to enable fast, robust and reproducible pH gradients that are simple to optimize and easily automated — without the need for time-consuming mobile phase adjustments.



Aggregate analysis
Thermo Scientific™ SurePac™ Bio 550 SEC MDi™ Column and Thermo Scientific™ MAbPac™ SEC-1 Column offer excellent size exclusion separation even under challenging conditions for aggregate analysis. Compatible with mass spectrometry for native LC-MS/MS workflows.

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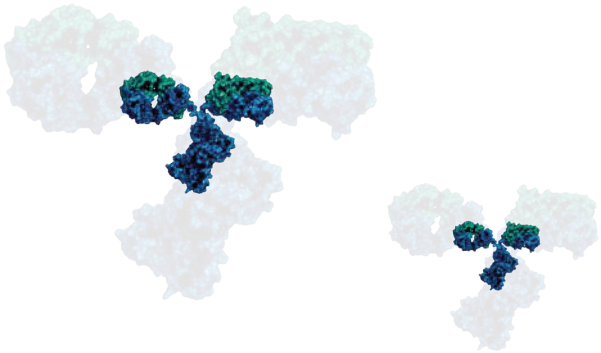


Affinity columns

MAbPac Protein A column

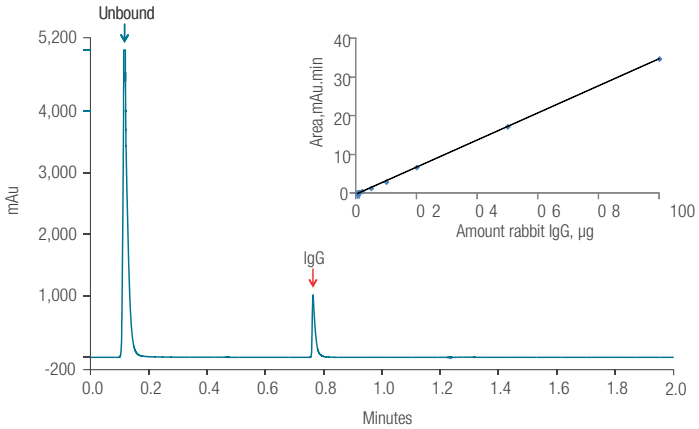
Providing fast, accurate titer analysis of monoclonal antibodies in harvest cell cultures, the nonporous, polymeric Thermo Scientific™ MAbPac™ Protein A Column delivers reproducible, highly efficient separations.

Additional reading

Links	Type	Description
	Application note	MAbPac Protein A: A novel affinity Protein A column

Harvest cell culture titer analysis

MAbPac Protein A column, 12 µm, 35 x 4.0 mm	
Flow rate	2 mL/min
Mobile phase A	50 mM sodium phosphate, 150 mM NaCl, 5% acetonitrile, pH 7.5
Mobile phase B	50 mM sodium phosphate, 150 mM NaCl, 5% acetonitrile, pH 2.5
Gradient	0% B for 0.2 mins, 100% B for 0.60 mins, 0% B for 1.20 mins
Temperature	30 °C
Injection volume	10 µL
Detection	UV at 280 nm
Sample	mAb B, 5 mg/mL harvest cell culture



MAbPac Protein A column ordering information

Particle size (µm)	Format	Length (mm)	4.0 mm ID
12	HPLC column	35	082539

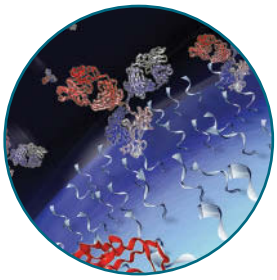
Intact analysis by HIC

MAbPac HIC-10, HIC-20, HIC-Butyl columns

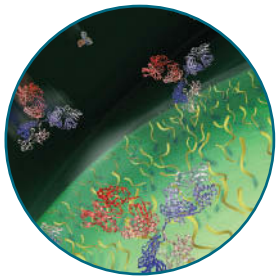
Orthogonal to IEX, SEC, and HIC offers selectivity to resolve charge neutral protein oxidations and protein misfolds. Our proprietary 1000 Å silica Thermo Scientific™ MAbPac™ HIC-10 and MAbPac HIC-20 Columns provide unique separation profiles offering high resolution for protein samples. For more hydrophobic samples, select the Thermo Scientific™ MAbPac™ HIC-Butyl column.

Additional reading

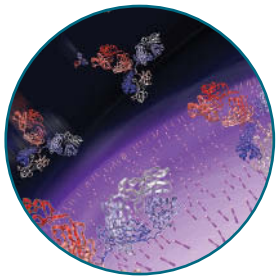
Links	Type	Description
	Application note	High resolution separation of a fusion protein on MAbPac HIC-10 column
	Application note	HIC as a complementary, confirmatory tool to SEC for the analysis of mAb aggregates
	Application note	High resolution separation of mAb fragments on MAbPac HIC-20 column
	Application note	High resolution separation of monoclonal antibody (mAb) oxidation variants
	Application note	High resolution separation of cysteine-conjugated antibody drug mimics



MAbPac HIC-10



MAbPac HIC-20

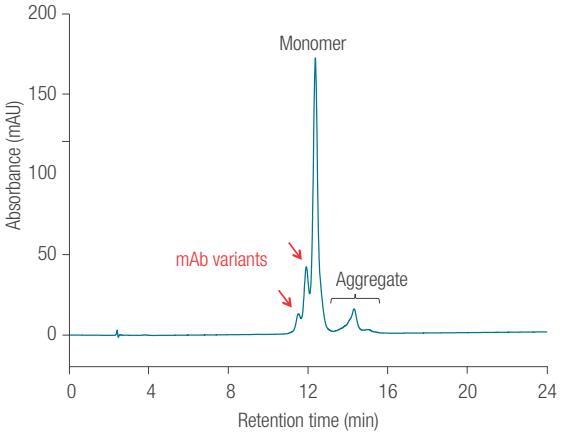


MAbPac HIC-Butyl

Intact analysis by HIC

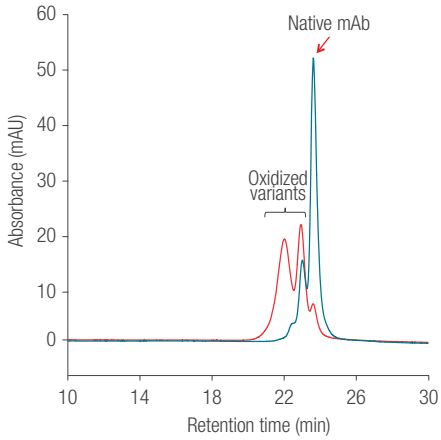
Separation of mAb aggregates

MAbPac HIC-10 column, 5 µm, 100 x 4.6 mm			
Flow rate	0.5 mL/min		
Mobile phase A	2 mM ammonium sulfate, 100 mM sodium phosphate, pH 7.0		
Mobile phase B	100 mM sodium phosphate, pH 7.0		
Temperature	20 °C		
Injection volume	15 µL		
Detection	UV at 280 nm		
Sample	Monoclonal antibody (4 mg/mL)		
Gradient	Time (min)	%A	%B
	-5.0	60	40
	0.0	60	40
	1.0	60	40
	29.0	0	0
	34.0	0	0



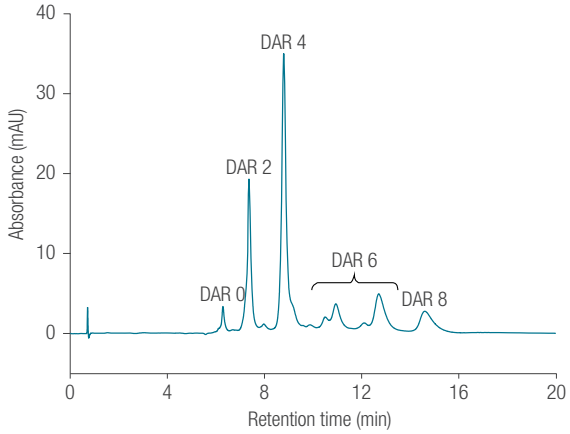
Separation of mAb fragments

MAbPac HIC-20 column, 5 µm, 250 x 4.6 mm			
Flow rate	0.5 mL/min		
Mobile phase A	2 mM ammonium sulfate, 100 mM sodium phosphate, pH 7.0		
Mobile phase B	100 mM sodium phosphate, pH 7.0		
Temperature	30 °C		
Injection volume	Untreated mAb: 20 µL (1.25 mg/mL) Oxidized mAb: 20 µL (1.25 mg/mL)		
Detection	UV at 280 nm		
Sample	Untreated mAb H_2O_2 oxidized mAb		
Gradient	Time (min)	A%	%B
	-6.0	50	50
	0.0	50	50
	2.0	50	50
	30.0	0	100
	35.0	0	100



Separation of antibody drug conjugates (ADCs)

MAbPac HIC-Butyl column, 5 µm, 100 x 4.6 mm			
Flow rate	1.0 mL/min		
Mobile phase A	1.5 mM ammonium sulfate, 50 mM sodium phosphate, pH 7.0/ isopropanol (95:5 v/v)		
Mobile phase B	50 mM sodium phosphate, pH 7.0/isopropanol (80:20 v/v)		
Temperature	25 °C		
Injection volume	5 µL		
Detection	UV at 280 nm		
Sample	Cys-conjugated ADC mimic (5 mg/mL)		
Gradient	Time (min)	%A	%B
	-5.0	100	0
	0.0	100	0
	1.0	100	0
	15.0	0	100
	20.0	0	100



Intact analysis by HIC

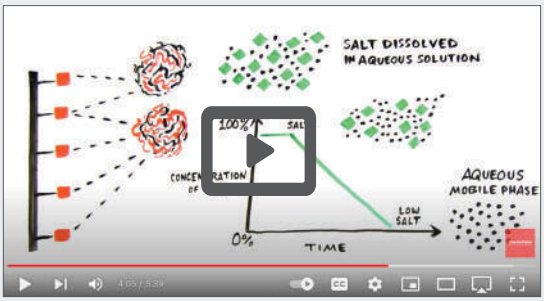
MAbPac HIC selection guide

Column	MAbPac HIC-10	MAbPac HIC-20	MAbPac HIC-Butyl
Intact mAbs/proteins	++++	+++	++
mAb aggregates	++++	+++	++
mAb fragments (F_{ab} and F_c)	+++	++++	+++
Oxidized mAbs	+++	++++	+++
Antibody Drug Conjugates (ADCs)	+++	+++	++++
Bispecific mAbs	+++	++++	++

Greater number of ++++ denotes greater suitability

MAbPac HIC family columns ordering information

Particle size (µm)	Description	Format	Length (mm)	4.6 mm ID
5	MAbPac HIC-10 column	Guard cartridges	10	088482
		HPLC column	100	088480
			250	088481
5	MAbPac HIC-20 column	Guard cartridges (2/pack)	10	088555
		HPLC column	100	088553
			250	088554
5	MAbPac HIC-Butyl column	Guard cartridges	10	088559
		HPLC column	100	088558
—	Guard cartridge holder	—	—	069580



Video:
Introduction to hydrophobic interaction chromatography

Released glycan analysis

Accucore 150-Amide-HILIC column

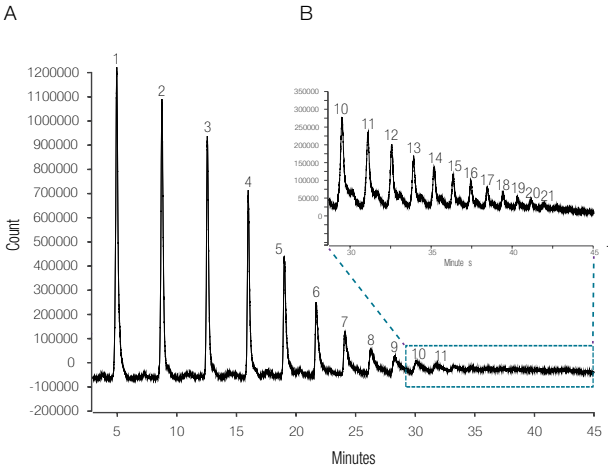
For monoclonal antibodies, or protein samples with mainly neutral glycans, the Thermo Scientific™ Accucore™ 150-Amide HILIC Column offers outstanding separation on a solid core particle. The low backpressure of this particle allows users to experiment with optimum temperature of their separation, to maximize the elucidation of their released glycan profile. For proteins with charged glycans, we offer two mixed mode column chemistries combining anion exchange with HILIC or RP separations. Thermo Scientific™ GlycanPac™ AXH-1 Columns separates the glycan profile by charge, size, and hydrophilicity. Thermo Scientific™ GlycanPac™ AXR-1 Columns separates the profile by charge, size, and branch isomers.

Additional reading

Links	Type	Description
	Application note	Analysis of human IgG glycans on a solid core amide HILIC stationary phase

2-AB labeled dextran ladder

Accucore 150-Amide-HILIC column, 2.6 µm, 100 x 2.1 mm	
Flow rate	500 µL/min
Mobile phase A	Acetonitrile
Mobile phase B	50 mM ammonium formate, pH 4.5
Temperature	60 °C
Injection volume	2 µL to 5 µL
Backpressure at starting conditions	110 bar
Injection wash solvent	80:20 (v/v) acetonitrile:water
Detector	Fluorescence, 330 nm excitation wavelength; 420 nm emission wavelength; acquisition start after 3 min from gradient start
Run time	50 min
Gradient	20–50% B in 40.0 minutes; 50% B for 5.0 minutes 50–20% B in 0.5 minutes; 50% B for 4.5 minutes



(A) 2 µL injection of sample, where 11 glycans were separated

(B) 5 µL injection of sample, zoomed-in to the later part of the gradient rise. A further 10 glycans were detected

Accucore 150-Amide-HILIC columns ordering information

Particle size (µm)	Format	Length (mm)	2.1 mm ID	3.0 mm ID	4.6 mm ID
2.6	Defender guard (4/pk)	10	16726-012105	—	—
	HPLC column	50	16726-052130	16726-053030	—
		100	16726-102130	16726-103030	16726-104630
		150	16726-152130	16726-153030	16726-154630
		250	16726-252130	—	—
—	Guard cartridge holder	—	852-00	852-00	850-00

Released glycan analysis

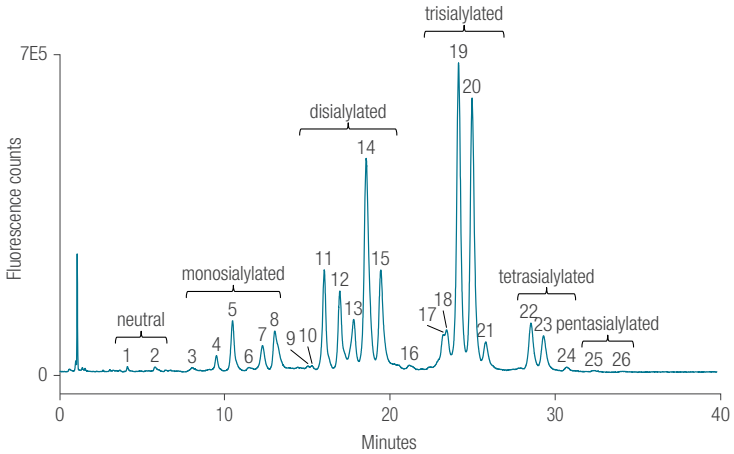
GlycanPac AXH-1 column

Additional reading

Links	Type	Description
	Application note	Separation of 2AB-labeled N-linked glycans from bovine fetuin
	Application note	Separation of 2AA-labeled N-linked glycans from human IgG
	Application note	Separation of 2AA-labeled N-linked glycans from glycoproteins

Separation of 2AB labeled N-glycans from bovine fetuin by charge, size and polarity

GlycanPac AXH-1 column, 1.9 µm, 150 x 2.1 mm				
Flow rate	0.4 mL/min			
Mobile phase A	Acetonitrile (100%)			
Mobile phase B	Water			
Mobile phase C	Ammonium formate (100 mM, pH = 4.4)			
Temperature	30 °C			
Injection volume	5 µL			
Detection	Fluorescence, 320/420 nm			
Sample	2AB labeled N-glycan from bovine fetuin			
Curve	5			
Gradient	Time (min)	%A	%B	%C
	-10.0	78	20	2
	0.0	78	20	2
	30.0	70	20	10
	35.0	60	20	20
	40.0	50	20	30



GlycanPac AXH-1 columns ordering information

Particle size (µm)	Format	Length (mm)	2.1 mm ID	3.0 mm ID	4.6 mm ID
1.9	UHPLC column	100	082473	—	—
		150	082472	—	—
		250	082521	—	—
3	Guard cartridges (2/pk)	10	082476	082475	082474
	HPLC column	150	082470	082469	082468
—	Guard cartridge holder	—	069580	069580	069580

Released glycan analysis

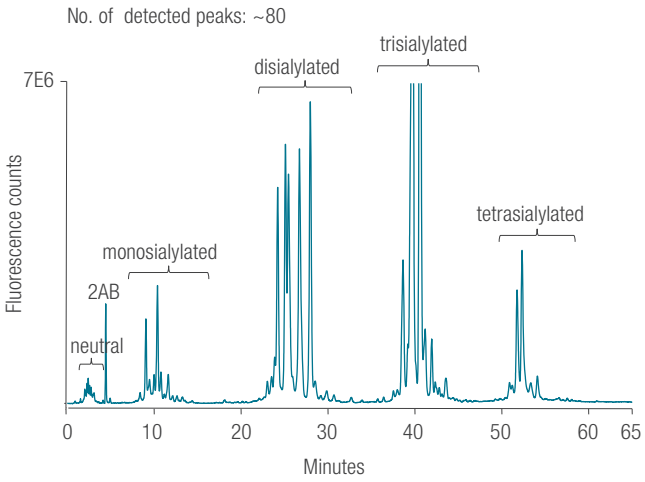
GlycanPac AXR-1 column

Additional reading

Links	Type	Description
	Application note	Separation of 2AB labeled *N*-glycans from bovine fetuin
	Application note	Structural analysis of native *N*-glycans released from proteins

Separation of 2AB labeled N-glycans from bovine fetuin

GlycanPac AXR-1 column, 1.9 µm, 150 x 2.1 mm				
Flow rate	0.4 mL/min			
Mobile phase A	Acetonitrile			
Mobile phase B	Water			
Mobile phase C	Ammonium formate (100 mM, pH = 4.4)			
Temperature	40 °C			
Sample load	100 pmoles			
Detection	Fluorescence, 320/420 nm			
Sample	2AB labeled N-glycan from bovine fetuin			
Curve	5			
Gradient	Time (min)	%A	%B	%C
	-10.0	0	95	5
	0.0	0	95	5
	1.0	0	95	15
	30.0	1	74	25
	65.0	20	50	30



GlycanPac AXR-1 columns ordering information

Particle size (µm)	Format	Length (mm)	2.1 mm ID	3.0 mm ID	4.6 mm ID
1.9	UHPLC column	150	088136	—	—
		250	088135	—	—

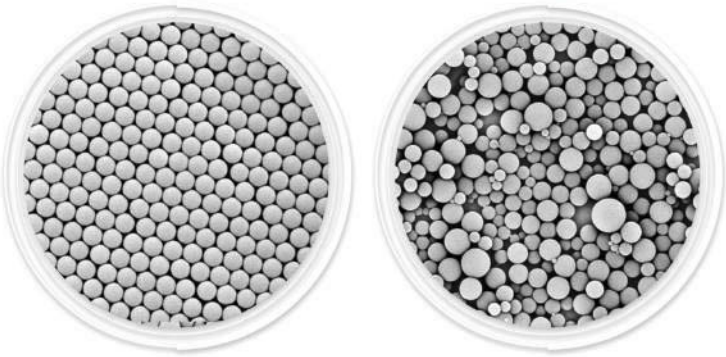
MDi particle innovation for bioseparations

Thermo Fisher Scientific's MDi particle innovation for bioseparations is a cutting-edge solution designed to transform the process of separating and purifying biomolecules. This innovative technology leverages advanced materials and particle engineering to enhance the efficiency and scalability of bioseparation applications, offering high-performance solutions for the biopharmaceutical, biotechnology, and life sciences industries.

The advantages of monodisperse particles

In liquid chromatography column technology, the particle size distribution is crucial in achieving optimal separation reproducibility. Offering consistent size and uniformity, our MDi monodisperse particles ensure consistent particle size distribution for column-to-column and batch-to-batch reproducibility, offering reproducible results and robust, easy-to-validate methods.

- **Reproducibility:** Offering consistent size and uniformity, our proprietary MDi monodisperse particles ensure column-to-column and batch-to-batch reproducibility, offering consistent results and robust, easy-to-validate methods.



Inert column hardware

Inert hardware refers to the use of materials that eliminate interactions with the sample, ensuring sample integrity and recovery. This type of hardware is particularly advantageous in biopharmaceutical analysis, where the accuracy and reliability of results are crucial. By reducing unwanted interactions, inert hardware helps to maintain sample integrity, prevent degradation or adsorption, and promote injection-to-injection reproducibility.

- **Maintains the integrity** of the target molecule by minimizing interactions between the column hardware and the sample.

  Promotes reproducibility and consistency in analysis by eliminating or reducing interactions between the hardware and the sample.

  Offers excellent chemical compatibility, allowing for a wide range of solvents and mobile phases to be used without compromising the integrity of the column.

  Improves reproducibility by maintaining consistent separation performance and sample recovery across multiple runs from the first injection.

Size exclusion columns

SurePac Bio 550 SEC MDi column

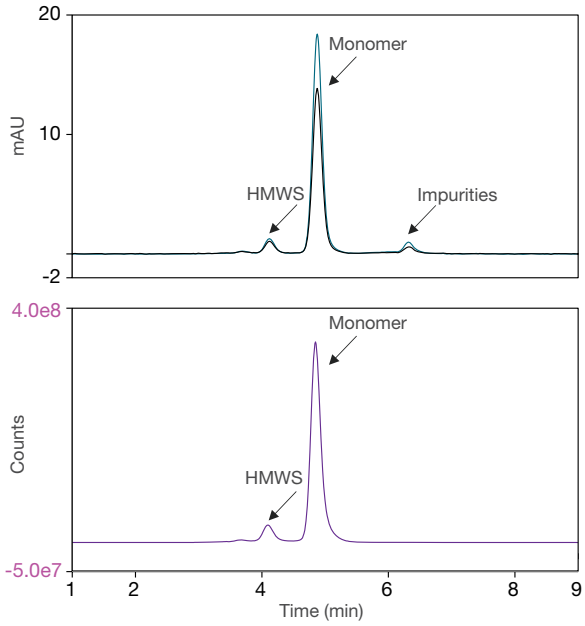
The SurePac Bio 550 SEC MDi columns are designed to provide fast, high efficiency, high resolution separations of Adeno-Associated Virus (AAV) monomeric capsids and aggregates based on their size difference. SEC operates on the principle of excluding molecules from pores in a stationary phase matrix, with larger molecules eluting faster than smaller ones. This unique mechanism makes SEC particularly well-suited for applications where the size and structure of molecules are of paramount importance.

Additional reading

Links	Type	Description
	Application note	High-resolution separation of monomeric AAV and aggregates by size-exclusion LC

Separation of AAV3 sample. UV (top) and FLD (bottom)

SurePac Bio 550 SEC MDi column, 3 µm, 4.6 × 150 mm	
Flow rate	0.3 mL/min
Mobile phase	50 mM phosphate buffer and 300 mM MaCl, pH 6.5
Gradient	0% B for 0.2 mins, 100% B for 0.60 mins, 0% B for 1.20 mins
Temperature	30 °C
Injection volume	UV: 1 µL FLD: 400 nL
Detection	UV, 280 NM (BLACK) AND 260 NM (BLUE) FLD, EX 280 NM AND EM 330 NM (PINK)
Sample	AAV3-CMV-GFP (2 × 10^{13} vg/mL)



SurePac Bio 550 SEC MDi columns ordering information

Particle size (µm)	Format	Length (mm)	2.1 mm ID	4.6 mm ID	7.8 mm ID
3	Guard column	30	43903-032131	43903-034631	43903-037831
—	HPLC column	150	43903-152131	43903-154631	43903-157831

Size exclusion columns

MAbPac SEC-1 column

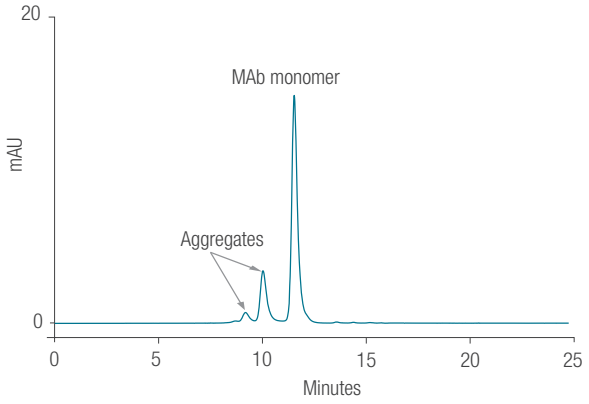
For mAb samples, our 300 Å silica MAbPac SEC-1 column provides separation of protein aggregates and fragments samples to characterize your analyte by LC-UV or LC-MS.

Additional reading

Links	Type	Description
	Application note	Lifetime stability of size-exclusion chromatography columns for protein aggregate analysis
	Application note	Analysis of monoclonal antibodies and their fragments

Monoclonal antibody aggregate separation

MAbPac SEC-1 column, 5 µm, 300 x 4.0 mm (PEEK)	
Flow rate	0.20 mL/min
Mobile phase	0.3 mM NaCl in 50 mM phosphate buffer pH 6.8
Gradient	0% B for 0.2 mins, 100% B for 0.60 mins, 0% B for 1.20 mins
Temperature	30 °C
Injection volume	2 µL
Detection	280 nM
Sample	mAb (10 mg/mL)



MAbPac SEC-1 columns ordering information

Particle size (µm)	Format	Length (mm)	2.1 mm ID	4.0 mm ID	7.8 mm ID
5	Guard column	50	—	074697	—
—	HPLC column	150	088790	075592	—
		300	088789	074696	088460

Intact and subunit analysis (RP)

MAbPac RP column

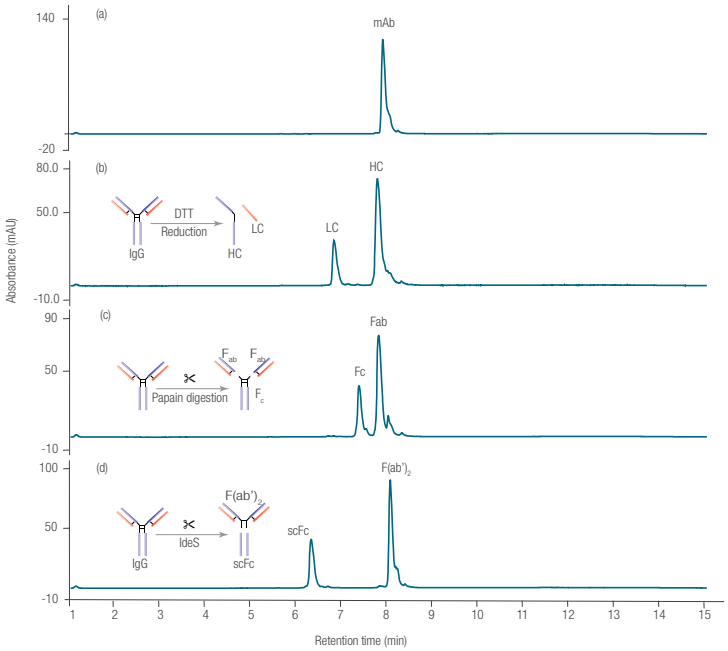
The wide pore (1500 Å) polymeric MAbPac RP columns offers high resolution separation and minimal carryover for monoclonal antibody samples. Excellent lifetime and ability to separate intact and protein subunits, compatible with LC-UV and LC-MS/MS applications.

Additional reading

Links	Type	Description
➜	**Application note**	Confident monoclonal antibody sequence verification by complementary LC-MS techniques
➜	**Application note**	Fast analysis of therapeutic monoclonal antibody fragments

mAb and mAb fragments analysis

MAbPac RP column, 4 µm, 50 x 3.0 mm			
Flow rate	0.5 mL/min		
Mobile phase A	H_2O/FA/TFA (99.88 : 0.1 : 0.02 v/v/v)		
Mobile phase B	ACN/H2O/FA/TFA 90 : 9.88 : 0.1 : 0.02 v/v/v/v)		
Temperature	80 °C		
Injection volume	5 µL		
Detection	UV at 280 nm		
Sample	(a) trastuzumab (5 mg/mL) (b) trastuzumab + DTT (4 mg/mL) (c) trastuzumab + Papain (2 mg/mL) (d) trastuzumab + IdeS (2 mg/mL)		
Gradient	Time (min)	%A	%B
	0.0	80	20
	1.0	80	20
	11.0	55	45
	12.0	55	45
	14.0	80	20
	16.0	80	20



Intact and subunit analysis (RP)

MAbPac RP columns ordering information

Particle size (µm)	Format	Length (mm)	2.1 mm ID	3.0 mm ID
4	Guard cartridges (2/pk)	10	088649	088646
	HPLC column	50	088648	088645
		100	088647	088644
		150	303270	303269
—	Guard cartridge holder	—	069580	069580

MAbPac RP 1 mm columns ordering information

Particle size (µm)	Length (mm)	1 mm ID
4	50	303182
	100	303183
	150	303184

Webinars

Analytical and life science webinars live and on-demand

NIBRT collaboration information

A collaboration built for Biopharma between the National Institute for Bioprocessing Research and Training (NIBRT) and Thermo Fisher Scientific

thermofisher.com/nibrt

Intact and subunit analysis (RP)

ProSwift RP column

The monolithic Thermo Scientific™ ProSwift™ RP Columns offer unique selectivity, high throughput separations for a wide range of protein sizes. These columns provide high loadability and operate under very low backpressure.

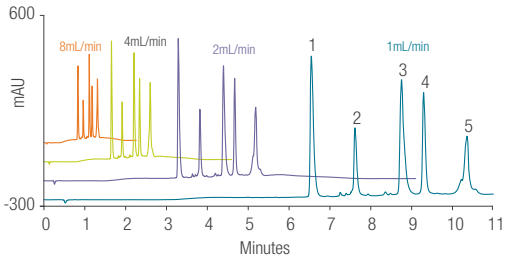
Additional reading

Links	Type	Description
	Application note	Simple and sensitive quantitation of large therapeutic proteins in plasma in 90 minutes

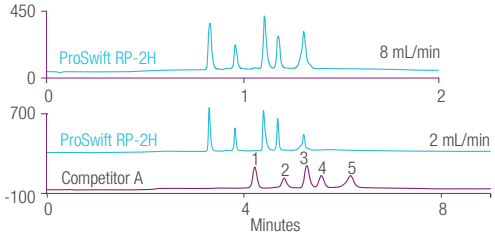
ProSwift column

ProSwift RP-2H column, 50 x 4.6 mm	
Flow rate	1, 2, 4, or 8 mL/min
Mobile phase A	H_2O/ACN (95:5; V/V) + 0.1% TFA
Mobile phase B	H_2O/ACN (5:95; V/V) + 0.1% TFA
Injection volume	2 µL
Detection	UV at 214 nm
Sample	Mixture of five proteins
Gradient	1 mL/min: 1-75% B in 12 min 2 mL/min: 1-75% B in 6 min 4 mL/min: 1-75% B in 3 min 8 mL/min: 1-75% B in 1.5 min
Analytes	1. Ribonuclease A 1.5 mg/mL 2. Cytochrome C 0.5 mg/mL 3. BSA 1.5 mg/mL 4. Carbonic anhydrase 0.9 mg/mL 5. Ovalbumin 1.5 mg/mL

Proteins



Competitive comparison



ProSwift RP columns ordering information

Functional group	Length (mm)	1.0 mm ID	4.6 mm ID
RP-1S	50	—	064297
RP-2H	50	—	064296
RP-3U	50	—	064298
RP-4H	50	069477	—
	250	066640	—

Charge variant analysis

For charge variant analysis by LC-UV or LC-MS ProPac 3R SAX and ProPac 3R SCX columns deliver outstanding resolution on a highly roust, reproducible and high-resolution platform. Combine ProPac 3R SCX columns with our proprietary CX-1 buffers formulations to enable fast, robust and reproducible pH gradients that are simple to optimize and easily automated - without the need for time-consuming mobile phase adjustments.

Protein isoelectric point (pI)				
<7	>7			
ProPac 3R SAX column	**ProPac 3R SCX column**	**MAbPac SCX-10 column**	**ProPac WCX-10 column**	**ProPac Elite WCX column**
• Works well with salt and pH gradient buffers • Best choice for proteins with acidic pI • Analyze full/empty AAV capsid ratios	• Highest resolution with excellent reproducibility • Works well with CX-1 buffers	• Alternative selectivity to WCX, scalable from short methods over comprehensive analysis to semi-prep formats • Works well with CX-1 buffers	• Industry gold-standard widely used and published	• Improved resolution, speed and reproducibility over ProPac WCX-10 column • Works well with CX-1 buffers

Video:
Tips to improve your charge variant analysis by ion exchange

Charge variant analysis

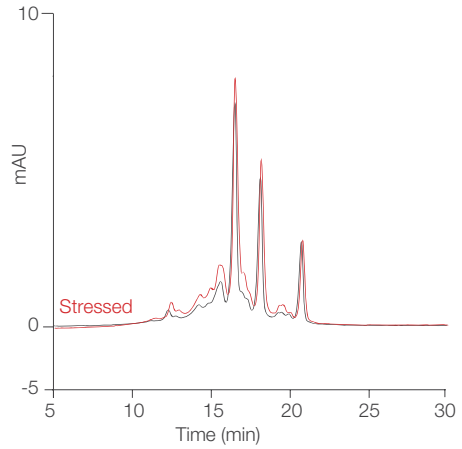
ProPac 3R SCX column

Additional reading

Links	Type	Description
	Application note	Salt gradient analysis of monoclonal antibodies using a 3 µm monodisperse SCX column
	Application note	Method development for pH gradient analysis of monoclonal antibodies using SCX column

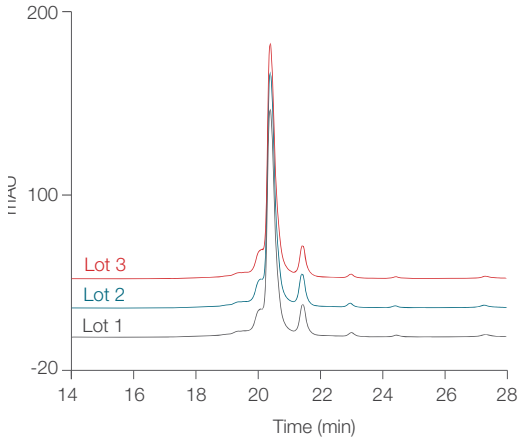
Salt gradient analysis of infliximab

ProPac 3R SCX column, 3 µm			
Format		4 × 100 mm	
Mobile phase		A: 20 mM MES, pH 6.5 B: 20 mM MES, pH 6.5 + 0.5 M NaCl	
Flow rate		0.3 mL/min	
Injection		2 µL	
Temp		30 °C	
Detection		UV, 280 nm	
Sample		Infliximab – 5 mg/mL	
Gradient		%A	%B
Time (min)	0.0	93	7
	30.0	78	22
	30.1	20	80
	33.0	20	80
	33.1	93	7
	40.0	93	7



Lot-to-lot reproducibility of NISTmAb salt gradient separation

ProPac 3R SCX column, 3 µm			
Format		4 × 100 mm	
Mobile phase		A: 20 mM MES, pH 6.5 B: 20 mM MES, pH 6.5 + 0.5 M NaCl	
Flow rate		0.3 mL/min	
Injection		2 µL	
Temp		30 °C	
Detection		UV, 280 nm	
Sample		NISTmAb – 10 mg/mL	
Gradient		%A	%B
Time (min)	0.0	95	10
	30.0	75	30
	30.1	20	80
	33.0	20	80
	33.1	95	10
	40.0	95	10



ProPac 3R SCX 3 µm columns ordering information

Particle size (µm)	Length (mm)	2.0 mm ID	4.0 mm ID
3	50	43103-052068	43103-054068
	100	43103-102068	43103-104068

Charge variant analysis

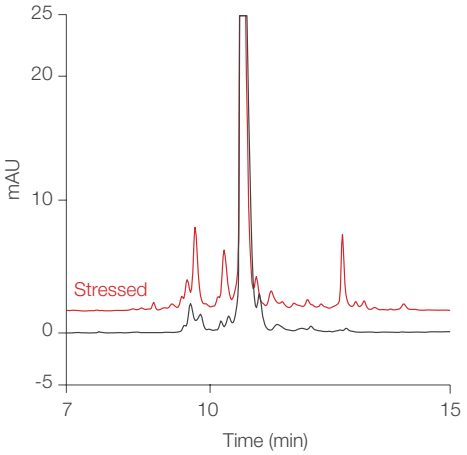
ProPac 3R SAX column

Additional reading

Links	Type	Description
	Application note	Salt gradient analysis of Protein G using a 3 µm monodisperse SAX column
	Application note	Salt gradient separation and analysis of adeno-associated virus samples using SAX column

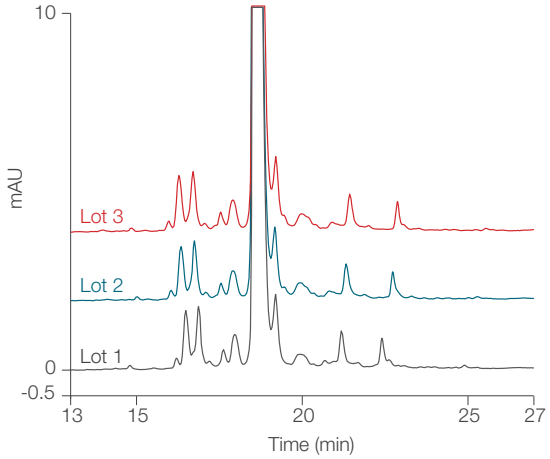
Salt gradient analysis of protein G

ProPac 3R SAX column, 3 µm			
Format		4 × 100 mm	
Mobile phase		A: 20 mM Tris, pH 8.0 B: 20 mM Tris + 500 mM NaCl, pH 8.0	
Flow rate		0.5 mL/min	
Injection		1 µL	
Temp		30 °C	
Detection		UV, 280 nm	
Sample		Protein G – 5 mg/mL	
Gradient		%A	%B
Time (min)	0.0	88	12
	1.0	88	12
	16.0	58	42
	16.1	0	100
	18.0	0	100
	18.1	88	12
	30.0	88	12



Lot-to-lot reproducibility of protein G salt gradient separation

ProPac 3R SAX column, 3 µm			
Format		4 × 100 mm	
Mobile phase		A: 20 mM Tris, pH 8.0 B: 20 mM Tris + 500 mM NaCl, pH 8.0	
Flow rate		0.5 mL/min	
Injection		1 µL	
Temp		30 °C	
Detection		UV, 280 nm	
Sample		Protein G – 5 mg/mL	
Gradient		%A	%B
Time (min)	0.0	88	12
	1.0	88	12
	31.0	58	42
	31.1	0	100
	33.0	0	100
	33.1	88	12
	45.0	88	12



ProPac 3R SAX 3 µm columns ordering information

Particle size (µm)	Length (mm)	2.0 mm ID	4.0 mm ID
3	50	43203-052068	43203-054068
	100	43203-102068	43203-104068

Charge variant analysis

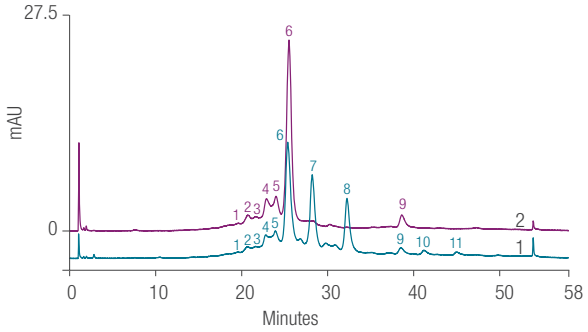
MAbPac SCX-10 column

Additional reading

Links	Type	Description
	Application note	A global pH-gradient based charge variant analysis
	Application note	High throughput, high resolution monoclonal antibody analysis

Baseline resolution of C-terminal lysine variants of a monoclonal antibody

MAbPac SCX-10 column, 5 µm, 250 x 4.0 mm	
Flow rate	1 mL/min
Mobile phase A	20 mM MES (pH 5.6) + 60 mM NaCl
Mobile phase B	20 mM MES (pH 5.6) + 300 mM NaCl
Gradient	15–36% B in 50 min
Temperature	30 °C
Injection volume	5 µL
Detection	UV at 280 nm
Sample	1. mAb B, 900 µg in 100 µL (no carboxypeptidase) 2. mAb B, 900 µg in 100 µL + carboxypeptidase, 50 µg, incubation at 37 °C for 3 h
Both chromatograms	Peaks 1–5: acidic variants
Sample 1	Peaks 6-8: C-Terminal lysine truncation variants of main peak. Peaks 9-11: C-Terminal lysine truncation variants of minor variant peak
Sample 2	Peak 6 results from peaks 6, 7, and 8 after CBP treatment. Peak 9 results from peaks 9, 10, and 11 after CBP treatment



MAbPac SCX-10 columns ordering information

Particle size (µm)	Format	Length (mm)	2.0 mm ID	4.0 mm ID	9.0 mm ID
5	HPLC column	50	—	078656	—
		150	—	085198	—
		250	—	078655	—
10	Guard column	50	075749	074631	—
	HPLC column	50	—	075603	—
		150	—	075602	—
		250	075604	074625	088784

Charge variant analysis

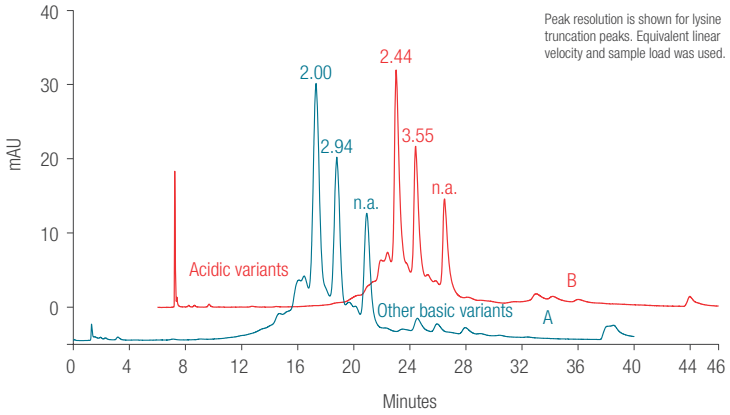
MAbPac SCX-10 RS column

Additional reading

Links	Type	Description
	Technical note	Development of ultra-fast pH gradient ion exchange chromatography
	Application note	A novel pH gradient separation platform for MAb

Lysine variants

MAbPac SCX column, 5 µm, 250 x 4.6 mm	
Flow rate	1.5 mL/min
Mobile phase A	20 mM MES pH 5.6 + 60 mM
Mobile phase B	20 mM MES pH 5.6 + 3 mM NaCl
Injection volume	15 µL
Detection	UV at 280 nm
Sample	mAb 5 mg/mL
Both chromatograms	Peaks 1–5: acidic variants
Chromatogram A	Gradient: 33-53% B in 30 min
Chromatogram B	Gradient: 33-53% B in 20 min



MAbPac SCX-10 RS columns ordering information

Particle size (µm)	Format	Length (mm)	2.1 mm ID	4.6 mm ID
5	UHPLC column	50	082675	082674
		150	088242	085209
		250	082515	082673

Charge variant analysis

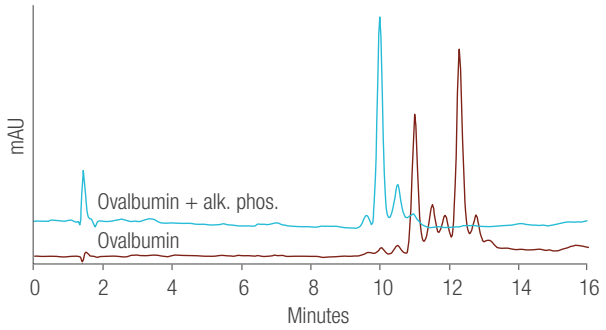
ProPac SAX-10 column

Additional reading

Links	Type	Description
	Application note	Simple and sensitive quantitation of large therapeutic proteins in plasma in 90 minutes

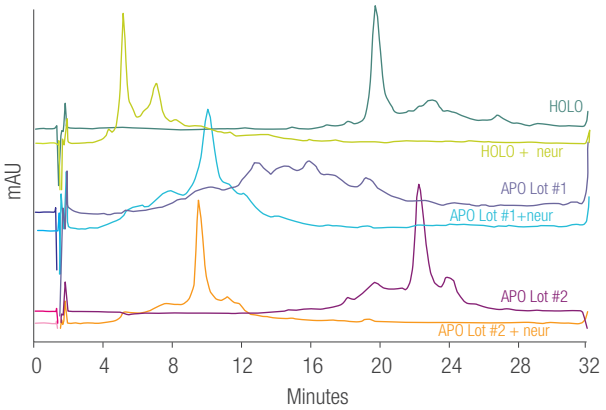
Resolution of phosphorylation variants of ovalbumin

ProPac SAX-10 column, 10 µm, 250 x 4.0 mm				
Flow rate	1.0 mL/min			
Mobile phase A	Water			
Mobile phase B	2.0 mM NaCl			
Mobile phase C	0.1 mM Tris/HCl (pH 8.5)			
Injection volume	1.0 µL			
Detection	UV at 214 nm			
Sample	Ovalbumin before and after alkaline phosphatase treatment			
Gradient	Time (min)	%A	%B	%C
	0.0	80	0	20
	15.0	67.5	12.5	20



Effect of sialylation on transferrin chromatography

ProPac SAX-10 column, 10 µm, 250 x 4.0 mm				
Flow rate	1.0 mL/min			
Mobile phase A	Water			
Mobile phase B	2.0 mM NaCl			
Mobile phase C	0.2 mM Tris/HCl (pH 9)			
Injection volume	50.0 µL			
Detection	UV at 214 nm			
Sample	HOLO (iron rich) and APO (iron poor) human transferrin samples before and after neuraminidase treatment. Digestions were carried out overnight at 37 °C in sodium acetate buffer at pH 5.			
Gradient	Time (min)	%A	%B	%C
	0.0	87	3	10
	30.0	83	7	10



ProPac SAX-10 columns ordering information

Particle size (µm)	Format	Length (mm)	2.0 mm ID	4.0 mm ID	9.0 mm ID	22.0 mm ID	4 x 50 mm ID
10	Guard column	50	063454	054998	–	–	–
	HPLC column	250	063448	054997	063703	088770	078990

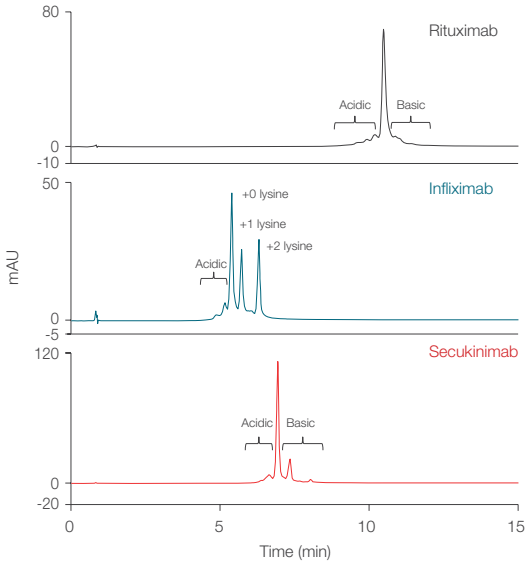
Charge variant analysis

ProPac Elite WCX column

Additional reading

Links	Type	Description
	Application note	Confident monoclonal antibody sequence verification by complementary LC-MS techniques
	Application note	Fast analysis of therapeutic monoclonal antibody fragments

ProPac Elite WCX column, 5 µm, 150 x 4.0 mm			
Flow rate	1.0 mL/min		
Mobile phase A	1x CX-1 pH gradient buffer A		
Mobile phase B	1x CX-1 pH gradient buffer B		
Temperature	30 °C		
Injection volume	2 µL		
Detection	UV at 280 nm		
Sample	Top: Rituximab, 5 mg/mL Middle: Infliximab, 5 mg/mL Bottom: Secukinimab, 5 mg/mL		
Gradient	Time (min)	%A	%B
	0.0	80	20
	15.0	20	80
	15.1	0	100
	17.0	0	100
	17.1	80	20
	25.0	80	20



ProPac Elite WCX columns ordering information

Particle size (µm)	Format	Length (mm)	2.0 mm ID	4.0 mm ID
5	HPLC column	50	303028	302973
		100	303027	302972
		250	303026	303025

ProPac Elite WCX kits ordering information

Particle size (µm)	Set contents	Length (mm)	4.0 mm ID
5	3 columns from 1 lot	150	302976
	3 columns from 3 lots	150	302977
	3 columns from 1 lot	250	303061
	3 columns from 3 lots	250	303062

Charge variant analysis

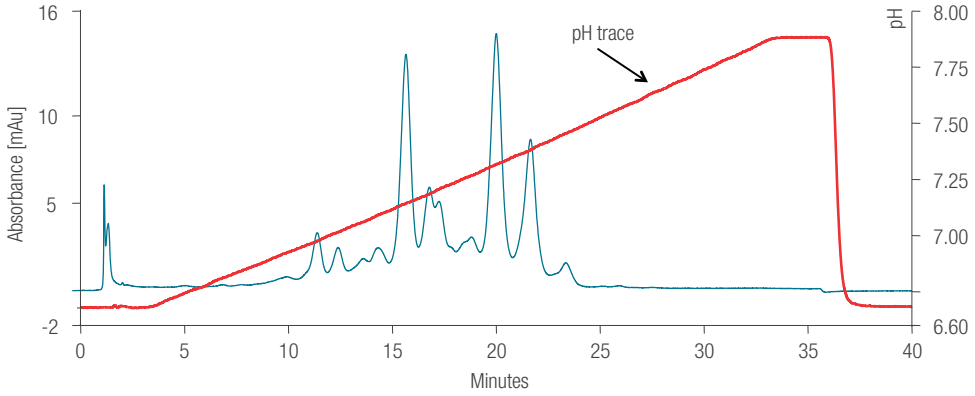
pH gradient buffers

Ready-to-use buffers for simple method development during charge variant characterization

Thermo Scientific pH gradient platform accelerates method development and facilitates method transfer to QA/QC for a wide range of protein and mAb charge variants through a generic LC-based approach to charge variant characterization.

- Patented buffer formulations enable fast, robust and reproducible pH gradients that are simple to optimize and easily automated
- Ready to use with existing LC columns and systems, without the need for time consuming mobile phase adjustments
- Applicable to the majority of mAbs

Optimization of mAb charge variant separation using a linear pH gradient: 25% B (pH 6.75) to 50% B (pH 7.9)



pH gradient buffers ordering information

Description	Buffer bottle size			
Buffer (10×)	125 mL	250 mL	500 mL	1000 mL
CX-1 pH gradient buffer A (pH 5.6)	083273	085346	302779	303274
CX-1 pH gradient buffer B (pH 10.2)	083275	085348	302780	303275

Peptide mapping and MAM

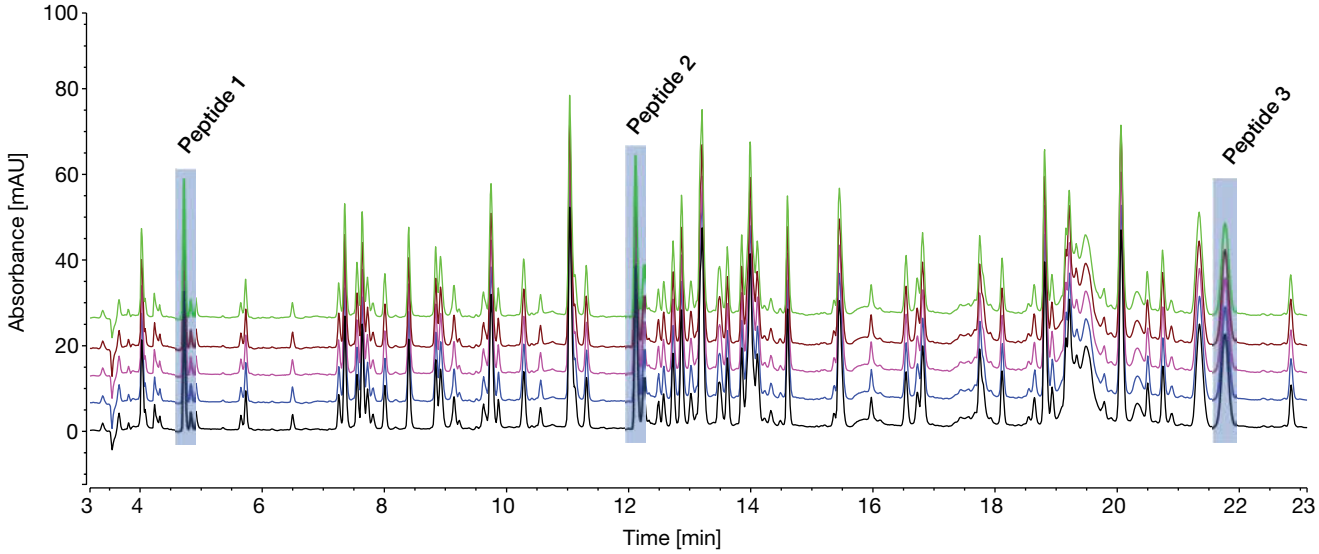
Hypersil GOLD Peptide column

Thermo Scientific™ Hypersil GOLD™ Peptide C18 UHPLC Columns are an excellent column choice for a broad range of peptides, offering high resolution for all critical quality attributes, without extremely long retention for more hydrophobic peptides.

Additional reading

Links	Type	Description
	Flyer	Resolve all your peptide challenges
	Application note	A comparison of reversed-phase C18 columns for peptide mapping of monoclonal antibodies
	Application note	Reliable monitoring of glycopeptide variants in monoclonal antibodies by LC-UV-MS analysis

UV chromatograms of five consecutive injections of adalimumab tryptic digest sample with detection wavelength of 214 nm



Hypersil GOLD Peptide columns ordering information

Particle size (µm)	Length (mm)	2.1 mm ID
1.9	50	26002-052130
	100	26002-102130
	150	26002-152130

Nucleic acids/oligonucleotides

DNAPac RP column

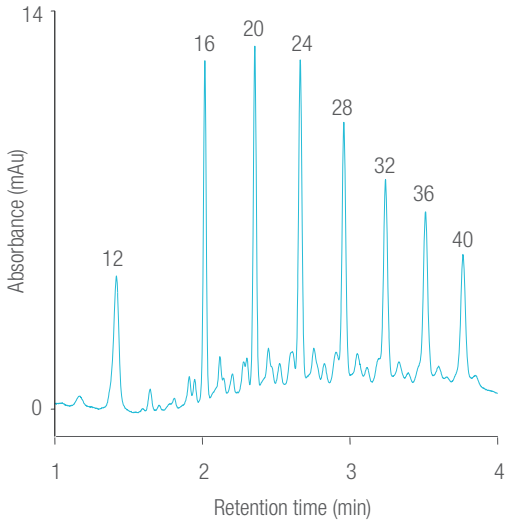
Thermo Scientific™ DNAPac™ RP Columns offers ion-pair reversed phase separations of nucleic acid mixtures. Samples from siRNA to mRNA easily resolve on this polymer chemistry. Compatible with LC-UV and LC-MS/MS methodologies this column delivers outstanding separations. The semi-preparative 21.2 mm ID column is now available with inert hardware.

Additional reading

Links	Type	Description
	Brochure	DNAPac columns: Superior oligonucleotide analysis

Fast analysis of mixed base DNA

DNAPac RP column, 4 µm, 50 x 2.1 mm			
Flow rate	0.8 mL/min		
Mobile phase A	25 mM HAA, pH 8.5		
Mobile phase B	25 mM HAA, pH 8.5/acetonitrile (50:50 v/v)		
Temperature	65 °C		
Injection volume	4 µL		
Detection	UV at 260 nm		
Sample	8-Combo DNA		
Gradient curve	3		
Peak label	Length of DNA		
Gradient	Time (min)	%A	%B
	-0.1	67	33
	0.0	67	33
	3.0	41	59
	3.1	5	95
	4.9	5	95
	5.0	67	33
	8.0	67	33



DNAPac RP columns ordering information

Particle size (µm)	Format	Length (mm)	2.1 mm ID	3.0 mm ID	10.0 mm ID	21.2 mm ID
4	Guard cartridges	10	088925	088921	–	080922-0121232
	HPLC column	50	088924	088920	–	–
		100	088923	088919	–	–
		150	–	–	SP6998	080922-1521232
		250	303324	–	–	–
—	Guard cartridge holder	—	069580	069580	–	950-00

Nucleic acids/oligonucleotides

DNAPac PA200 column

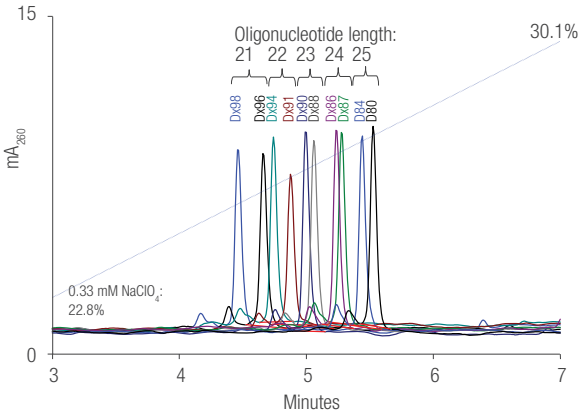
Thermo Scientific™ DNAPac™ PA200 Columns are strong anion exchange columns for n-1 separation of oligo samples. Compatible with LC-UV, these columns offer orthogonal separation to reversed phase columns, separating the oligonucleotide sample by size and charge.

Additional reading

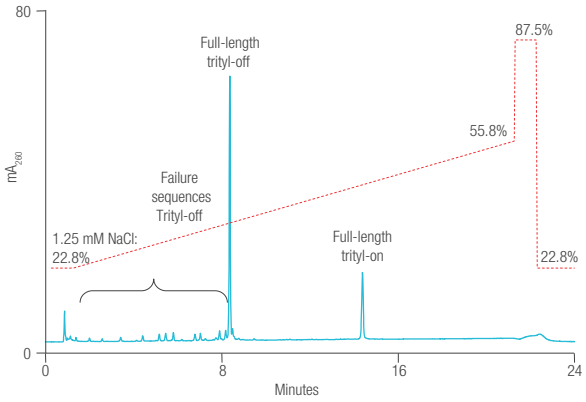
Links	Type	Description
	Brochure	DNAPac columns: Superior oligonucleotide analysis

DNAPac PA200, 8 µm, 250 x 4.0 mm	
Flow rate	1.2 mL/min
Mobile phase	$NaClO_4$, pH 6.5 with 20% ACN
Detection	UV at 260 nm
Flow rate	1.2 mL/min

Separation of oligonucleotides by length



Target, failure and trityl-on oligonucleotides



DNAPac PA200 columns ordering information

Particle size (µm)	Format	Length (mm)	2.0 mm ID	4.0 mm ID	9.0 mm ID	22.0 mm ID
8	Guard column	50	063423	062998	063419	088780
	HPLC column	250	063425	063000	063421	088781

Nucleic acids/oligonucleotides

DNAPac PA200 RS column

Thermo Scientific™ DNAPac™ PA200RS Columns are strong anion exchange columns for n-1 separation of oligo samples. Compatible with LC-UV, these columns offer orthogonal separation to reversed phase columns, separating the oligonucleotide sample by size and charge.

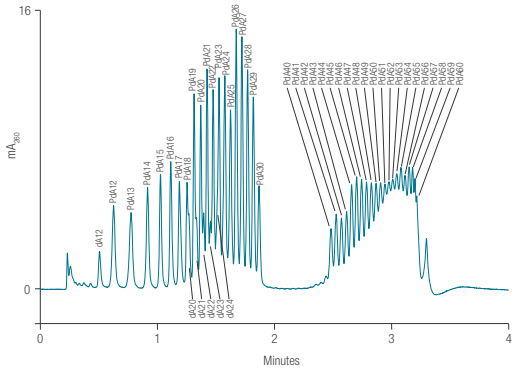
Additional reading

Links	Type	Description
	Brochure	DNAPac columns: Superior oligonucleotide analysis
	Application note	High-resolution separation of oligonucleotides
	Application note	Ultra-high-resolution separation of oligonucleotides by UHPLC
	Application note	Separation of mixed-base oligonucleotides

Partial resolution of 46 oligonucleotides

DNAPac PA200 RS, 4 µm, 50 x 4.6 mm	
Flow rate	1.30 mL/min
Mobile phase A	20 mM Tris pH 8
Mobile phase B	A + 1.25 mM NaCl
Temperature	30 °C
Injection volume	2.5 µL
Gradient	28–43% B in 4 CV* (2.56 min) curve 3**
Sample	PdA12–30, 40–60

*CV = column volumes
**Curve 3 indicates continuously changing gradient, asymptotically approaching a maximum salt concentration. Programed in Thermo Scientific™ Chromeleon™ Chromatography Data System (CDS).



DNAPac PA200 RS columns ordering information

Particle size (µm)	Format	Length (mm)	4.6 mm ID
4	BioRS column	50	082508
		150	082509
		250	082510

DNASwift SAX-1S column

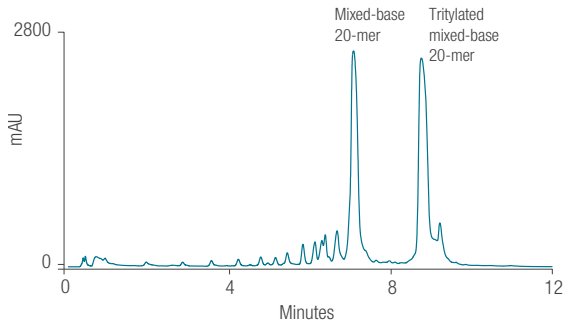
Thermo Scientific™ DNASwift™ Column is a monolithic column designed for users who would like to do SAX purification of oligonucleotide samples using their analytical HPLC. These monolithic columns offer high loadability, with slightly less resolution than our analytical columns.

Additional reading

Links	Type	Description
	Brochure	DNAPac columns: Superior oligonucleotide analysis

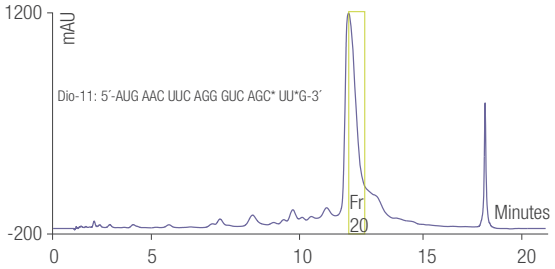
Tritylated oligonucleotide

DNASwift SAX-1S, 150 x 5.0 mm	
Flow rate	1.5 mL/min
Mobile phase A	15 mM Tris, pH 8
Mobile phase B	15 mM Tris, pH 8, 1.25 M NaCl
Temperature	30 °C
Injection volume	20 µL
Detection	UV at 260 nm
Gradient	8–64% B in 10 min



Purification of a 21-base RNA sample with aberrant 2'-5' linkages at the 1 and 3 positions from the 3' end

DNASwift SAX-1S, 150 x 5.0 mm	
Flow rate	1.5 mL/min
Mobile phase A	40 mM Tris, pH 7
Mobile phase B	40 mM Tris, pH 7 + 1.25 M NaCl
Temperature	30 °C
Injection volume	125 µg
Detection	UV at 260 nm
Gradient	26–42% B in 10 column volumes



DNASwift SAX-1S column ordering information

Length (mm)	5.0 mm ID
150	066766

BioLC column quick selection guide

Target applications	Column type	Mode of analysis	Recommended column	Particle size (µm)	Pore size (Å)	pH range	Maximum backpressure (psi)	Solvent compatibility
Affinity	Affinity columns	Affinity	**MAbPac Protein A**	12	Non-porous	2.5-7.5	1,000	—
Intact analysis by HIC	Silica-based hydrophobic interaction chromatography columns	Hydrophobic interaction	**MAbPac HIC-10**	5	1,000	2-8	4.6 × 100 mm = 6,000 4.6 × 250 mm = 8,000	Compatible with organic solvents and aqueous mobile phases
			MAbPac HIC-20	5	1,000	2-9		
			MAbPac HIC Butyl	5	Non-porous	2-12	4,000	Compatible with up to 50% organic solvents
Released glycan analysis	Silica based, mixed-mode columns	Mixed-mode	**GlycanPac AXH-1**	1.9	175	2-8	10,000	0 – 90% aqueous buffer; 10 – 100% acetonitrile or alcohols
				3	120	2-8	6,000	
			GlycanPac AXR-1	1.9	175	2-8	10,000	Compatible with 0 – 100% aqueous and common HPLC solvents (except acetone)
	Silica-based HILIC columns	HILIC	**Accucore 150 Amide HILIC**	2.6	150	2-8	14,500	—
aggregate and fragment analysis	Silica-based size exclusion chromatography phases	Size exclusion	**SurePac Bio 550 SEC MDi**	3	550	2-8	3,500	100% organic solvent
			MAbPac SEC-1	5	300	2.5-7.5	1,000 for 300 mm 600 for 150 mm	100% organic solvents
Intact and subunit analysis	Polymeric reversed-phase columns	Reversed-phase	**MAbPac RP**	4	1,500	2.1, 3.0 mm (0-14) 1 mm (1-7)	4,000	Up to 100% ACN, IPA, MeOH
	Polymeric reversed-phase columns	Reversed-phase	**ProSwift RP-1S**	Monolith	Monolith	1-14	2,800	—
			ProSwift RP-2H				2,800	Most common organic solvents
			ProSwift RP-3U				2,800	
			ProSwift RP-4H				1 × 50 mm = 2,000 2 × 250 mm = 3,000	
Charge variant analysis	Monolithic ion-exchange columns	Ion-exchange	**ProPac 3R SCX**	3	Non-porous	2-12*	4,500	—
			ProPac 3R SAX	3			4,500	—
			MAbPac SCX-10RS	5			7,000	—
			MAbPac SCX-10	5, 10			3,000 for 10 µm 5,000 for 5 µm	*Please consult column manual
			ProPac SAX-10	10			3,000	*Please consult column manual
			ProPac Elite WCX	5			4,500	*Please consult column manual
Peptide mapping	Silica based, reversed-phase columns	Reversed-phase	**Hypersil GOLD Peptide**	1.9	175	1-11	18,130	—
Nucleic acids and oligonucleotides	Polymeric ion-exchange columns	Ion-exchange	**DNAPac PA200**	8	Non-porous	2.5-12.5*	4,000	*Please consult column manual
			DNAPac PA200RS	4	Non-porous	2.5-12.5*	10,000	—
			DNASwift SAX-1S	Monolith	Monolith	3-14*	1,500	*Please consult column manual
	Polymeric reversed-phase	Reversed-phase	**DNAPac RP**	4	Proprietary wide pore	0-14	4,000	—

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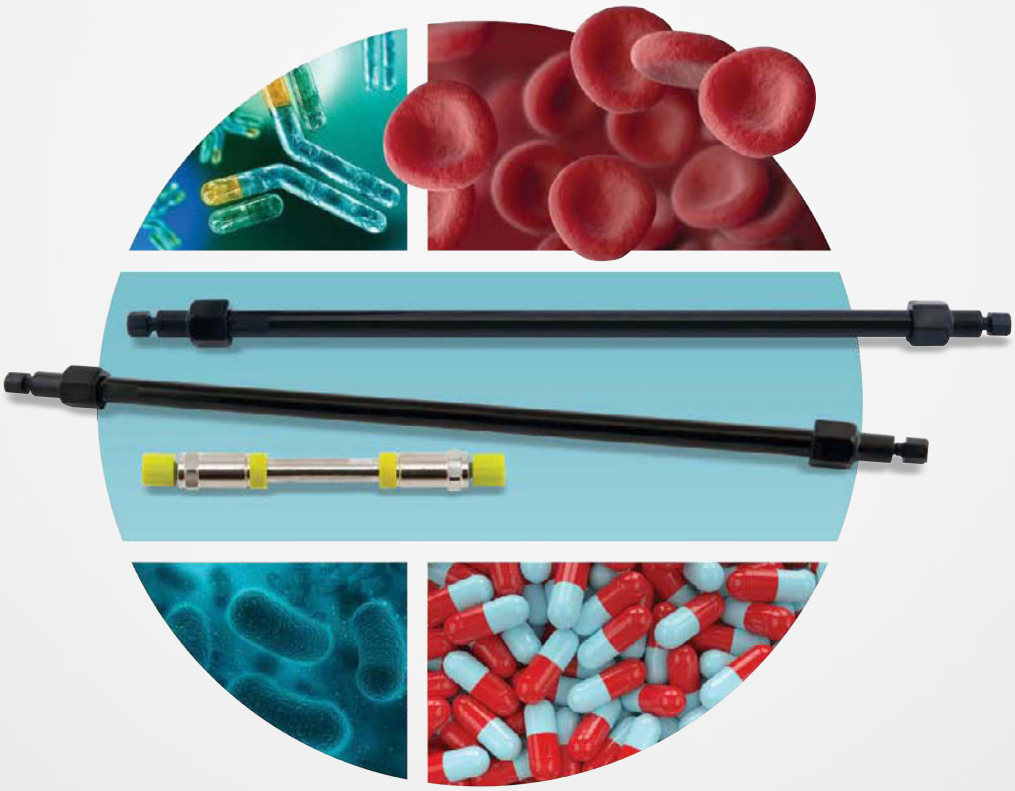

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