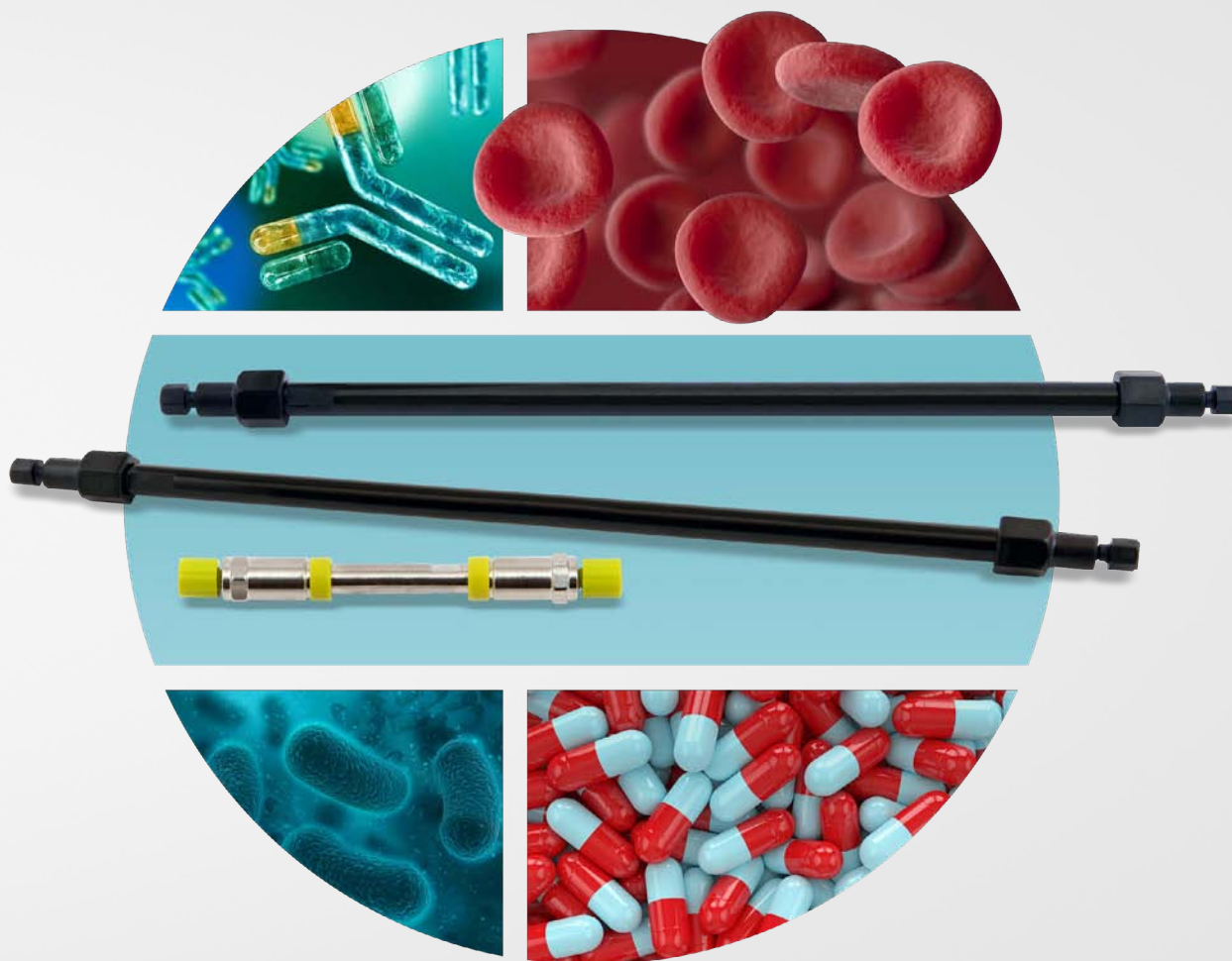




BioLC columns

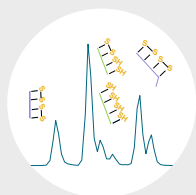
Connected chromatography solutions

BioLC columns and accessories

Introduction

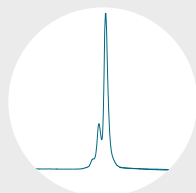
Your complete tool kit

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.



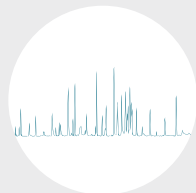
Intact or subunit analysis

Thermo Scientific™ MAbPac™ RP columns are 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.



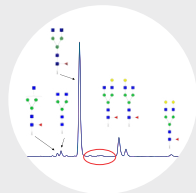
Oxidation monitoring

Deduce protein folding errors or charge-neutral amino acid modifications with the Thermo Scientific™ MAbPac™ HIC-20 hydrophobic interaction column. Our range of innovative HIC chemistries deliver native separations not seen on other columns.



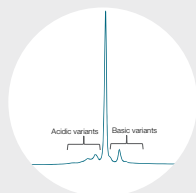
Peptide mapping

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 column delivers outstanding, reproducible and efficient peptide mapping separations.



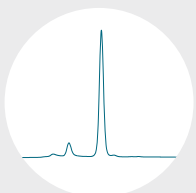
Released glycan analysis

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 Thermo Scientific™ ProPac™ 3R 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 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.

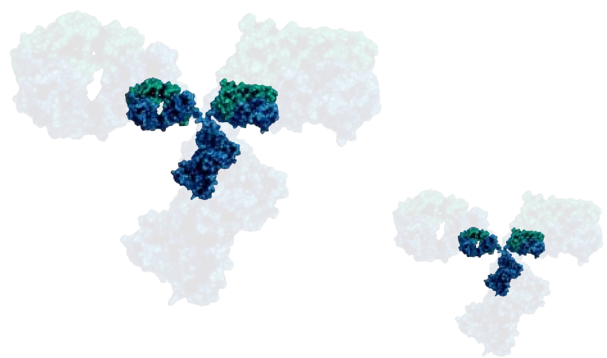


Aggregate analysis

Thermo Scientific™ MAbPac™ SEC-1 offers excellent size exclusion separation even under challenging conditions for aggregate analysis. Compatible with mass spectrometry for native LC-MS/MS workflows.

Contents

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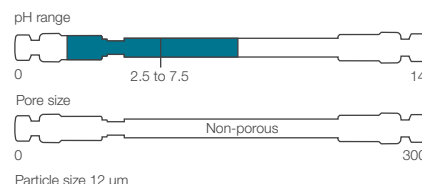
BioLC column selection quick guide

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)
			Accucore 150 Amide HILIC	2.6	150	2-8	14,500	—
Aggregate fragment analysis	Silica-based size exclusion chromatography phases	Size exclusion	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	Most common organic solvents
			ProSwift RP-2H				2,800	
			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 C18	1.9	175	1-11	18,130	—
				3	175		5,800	—
			Acclaim 120 C18	2.2	120	2-8	Various	—
				3	120			—
				5	120			—
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	—

Affinity columns

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

MAbPac Protein A column

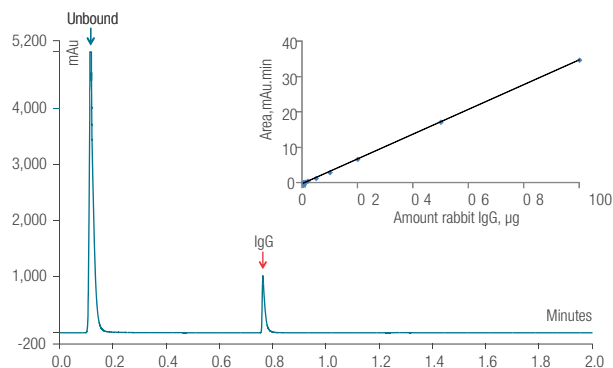


Additional reading

Links	Type	Description
	Application note	MAbPac Protein A: A novel affinity Protein A column
	Learn more	thermofisher.com/biolc

Harvest cell culture titer analysis

MAbPac Protein A, 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



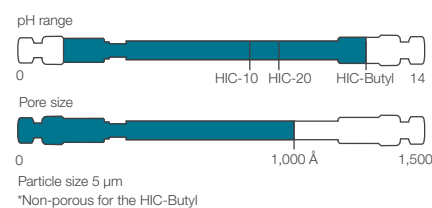
MAbPac Protein A column

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

Intact analysis by HIC

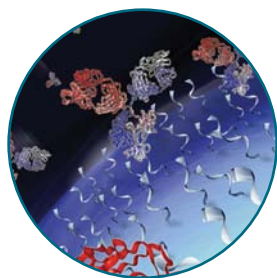
Orthogonal to IEX and SEC, Hydrophobic Interaction Chromatography (HIC) offers selectivity to resolve charge neutral protein oxidations and protein misfolds. Our proprietary 1000 Å silica Thermo Scientific™ MAbPac™ HIC-10 and Thermo Scientific™ MAbPac™ HIC-20 provide unique separation profiles offering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

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

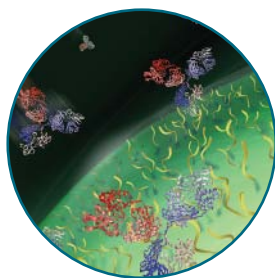


Additional reading

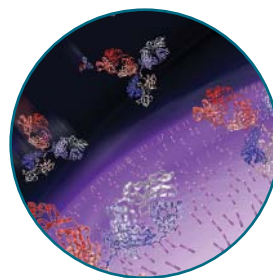
Links	Type	Description
	Application note	MAbPac HIC-10 High resolution separation of a fusion protein on MAbPac HIC-10 column
	Application note	HIC as a complementary, confirmatory tool to SEC for the analysis of mAb aggregates
	Application note	MAbPac HIC-20 High resolution separation of mAb fragments 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
	Learn more thermofisher.com/biolc	



MAbPac HIC-10



MAbPac HIC-20

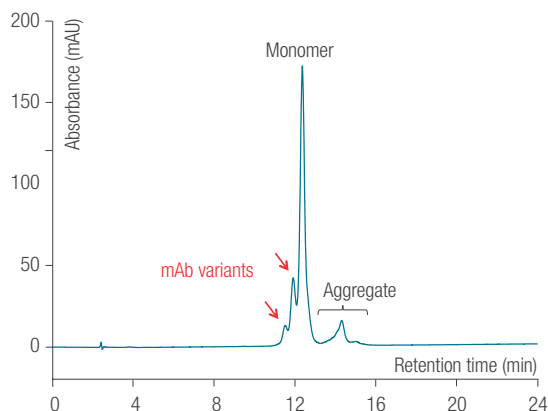


MAbPac HIC-Butyl

Intact analysis by HIC continued

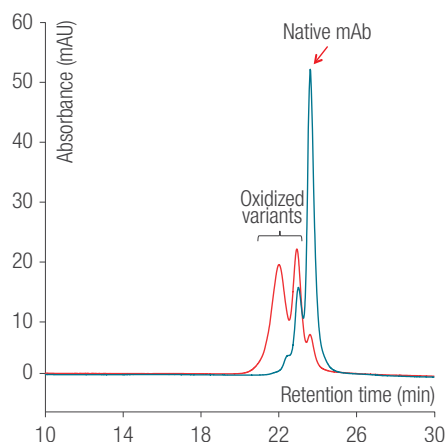
Separation of mAb aggregates

MAbPac HIC-10, 5 μ m, 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 μ L		
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



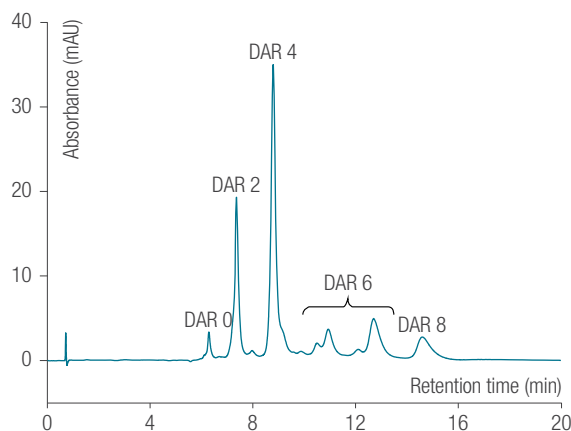
Separation of mAb fragments

MAbPac HIC-20, 5 μ m, 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 μ L (1.25 mg/mL) Oxidized mAb: 20 μ L (1.25 mg/mL)		
Detection	UV at 280 nm		
Sample	Untreated mAb H ₂ O 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, 5 μ m, 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 μ L		
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 continued

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

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



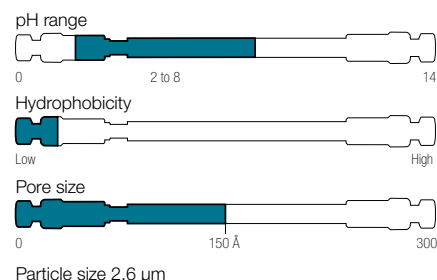
Video:

Introduction to hydrophobic interaction chromatography

Released glycan analysis

For monoclonal antibodies, or protein samples with a lot of neutral glycans, the Thermo Scientific™ Accucore™ 150-Amide HILIC 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 separates the glycan profile by charge, size, and hydrophilicity. Thermo Scientific™ GlycanPac™ AXR-1 separates the profile by charge, size, and branch isomers.

Accucore 150-Amide-HILIC column

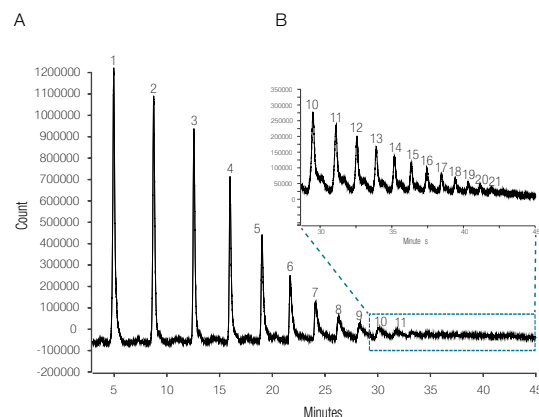


Additional reading

Links	Type	Description
	Application note	Analysis of human IgG glycans on a solid core amide HILIC stationary phase
	Learn more	thermofisher.com/biolc

2-AB labeled dextran ladder

Accucore 150-Amide-HILIC, 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

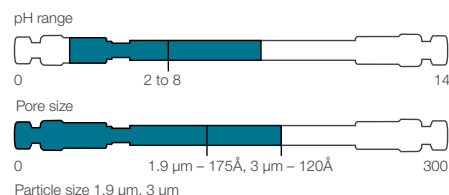


Accucore 150-Amide-HILIC columns

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 continued

GlycanPac AXH-1 column

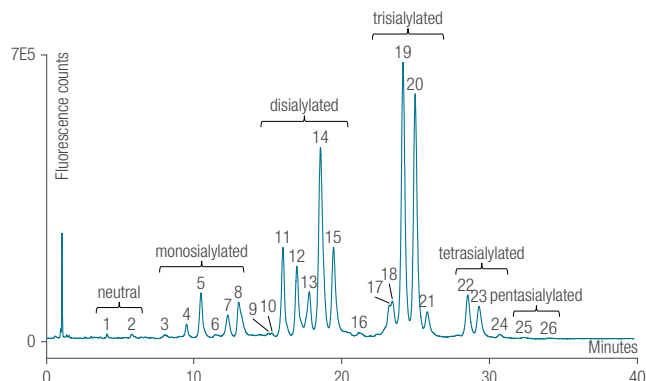


Additional reading

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
	Learn more thermofisher.com/biolc	

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

GlycanPac AXH-1, 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

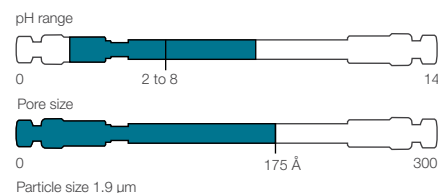


GlycanPac AXH-1 columns

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 continued

GlycanPac AXR-1 column

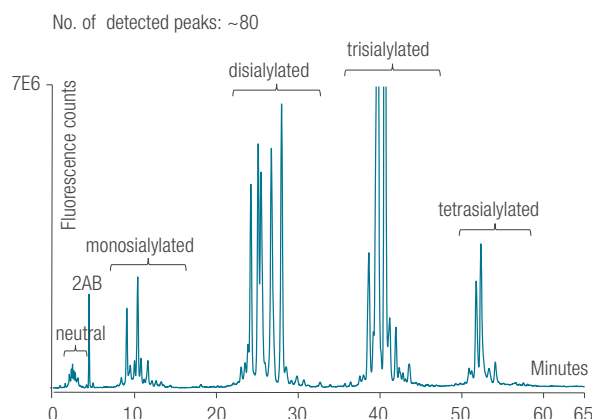


Additional reading

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
	Learn more thermofisher.com/biolc	

Separation of 2AB labeled N-glycans from bovine fetuin

GlycanPac AXR-1, 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



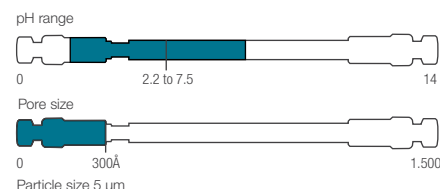
GlycanPac AXR-1 columns

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	—	—
—	Guard cartridge holder	—	069580	069580	069580

Aggregate fragment analysis

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

MAbPac SEC-1 column

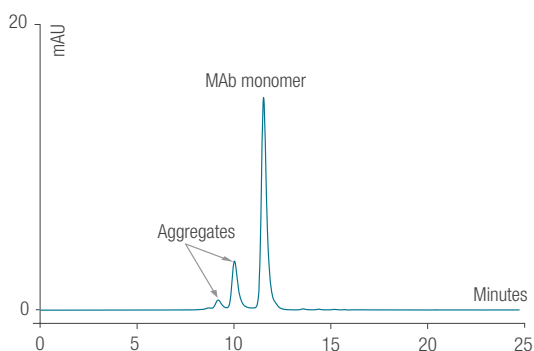


Additional reading

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
	Learn more thermofisher.com/biolc	

Monoclonal antibody aggregate separation

MAbPac SEC-1, 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)



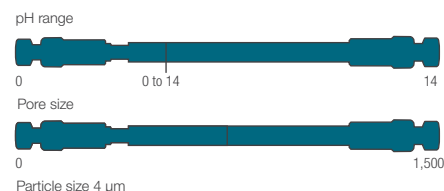
MAbPac SEC-1 columns

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)

The wide pore (1500 Å) polymeric Thermo Scientific™ 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. 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.

MAbPac RP column

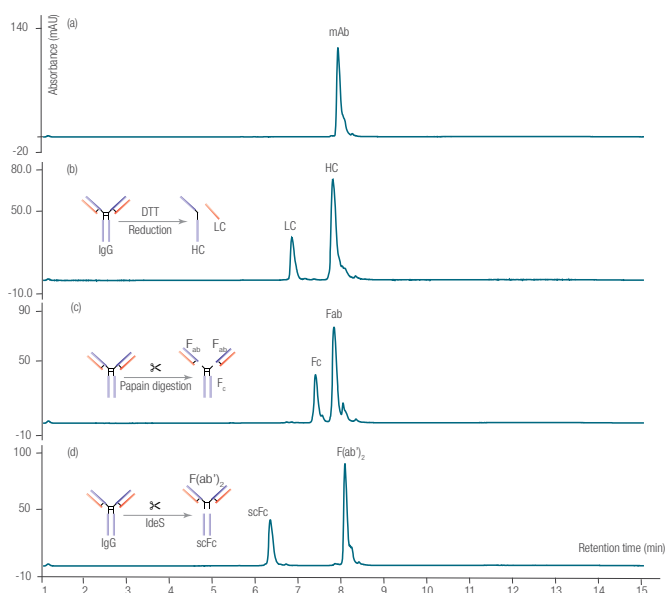


Additional reading

Links	Type	Description
	Application note	Confident monoclonal antibody sequence verification by complementary LC-MS techniques
	Application note	Fast analysis of therapeutic monoclonal antibody fragments
	Learn more thermofisher.com/biolc	

mAb and mAb fragments analysis

MAbPac RP, 4 µm, 50 x 3.0 mm			
Flow rate	0.5 mL/min		
Mobile phase A	H ₂ O/FA/TFA (99.88 : 0.1 : 0.02 v/v/v)		
Mobile phase B	ACN/H ₂ O/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) continued



MABPac RP columns

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

MABPac RP 1 mm columns

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

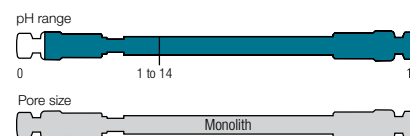


NIBRT collaboration information

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) continued

ProSwift RP column



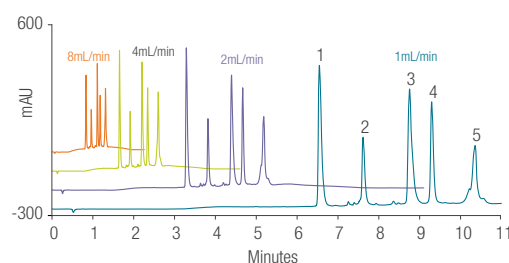
Additional reading

Links	Description
	Learn more thermofisher.com/biolc

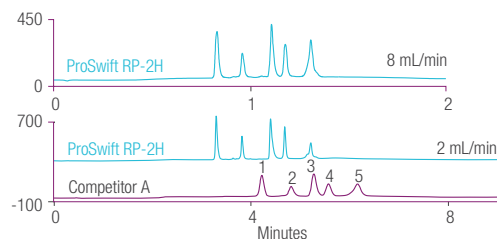
ProSwift column

ProSwift RP-2H, 50 x 4.6 mm	
Flow rate	1, 2, 4, or 8 mL/min
Mobile phase A	H ₂ O/ACN (95:5; V/V) + 0.1% TFA
Mobile phase B	H ₂ O/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

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/MS Thermo Scientific™ ProPac™ 3R SAX and Thermo Scientific™ ProPac™ 3R SCX columns deliver outstanding resolution on a highly robust, 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

- Works well with salt and pH gradient buffers
- Best choice for proteins with acidic pI
- Analyze full/empty AAV capsid ratios

ProPac 3R SCX column

- Highest resolution with excellent reproducibility
- Works well with CX-1 buffers

MABPac SCX-10 column

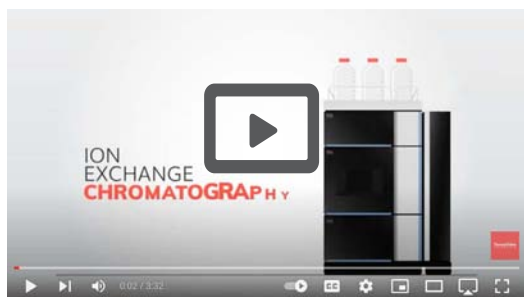
- Alternative selectivity to WCX, scalable from short methods over comprehensive analysis to semi-prep formats
- Works well with CX-1 buffers

ProPac WCX-10 column

- Industry GOLD standard – widely used and published

ProPac Elite WCX column

- Improved resolution, speed and reproducibility over ProPac WCX-10 column
- Works well with CX-1 buffers

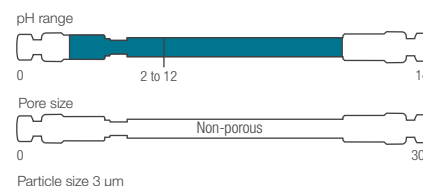


Video:

Tips to improve your charge variant analysis by ion exchange

Charge variant analysis continued

ProPac 3R SCX column

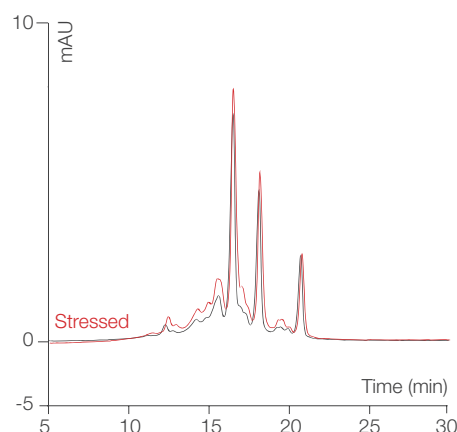


Additional reading

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
	Learn more thermofisher.com/propac	

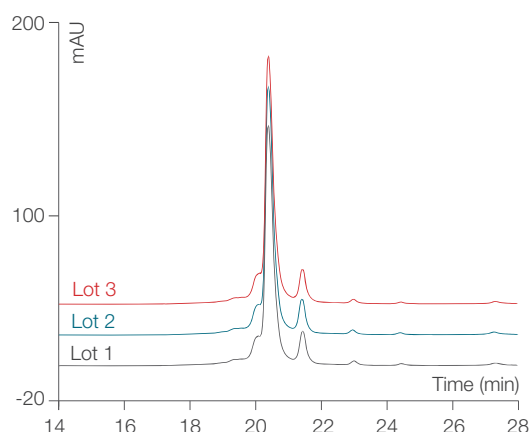
Salt gradient analysis of infliximab

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

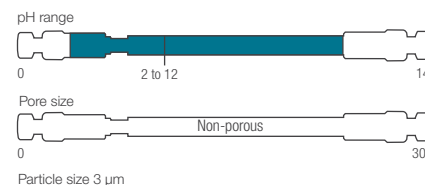


ProPac 3R SCX 3 µm columns

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 continued

ProPac 3R SAX column

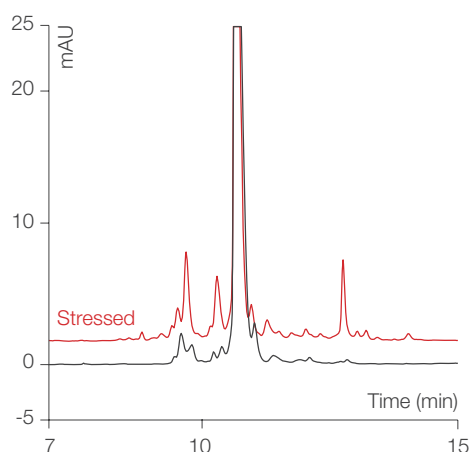


Additional reading

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
	Learn more thermofisher.com/propac	

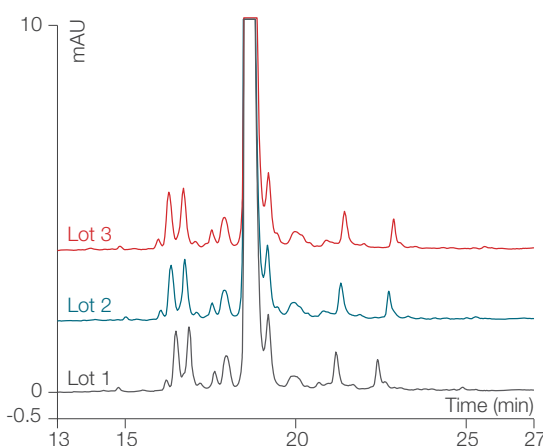
Salt gradient analysis of protein G

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

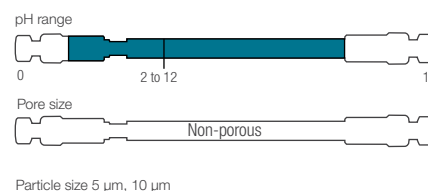


ProPac 3R SAX 3 µm columns

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 continued

MABPac SCX-10 column

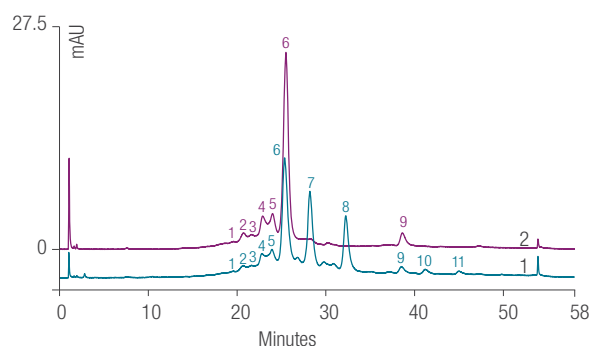


Additional reading

Links	Type	Description
→	Application note	A global pH-gradient based charge variant analysis
→	Application note	High throughput, high resolution monoclonal antibody analysis
🏠	Learn more thermofisher.com/biolc	

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

MABPac SCX-10, 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

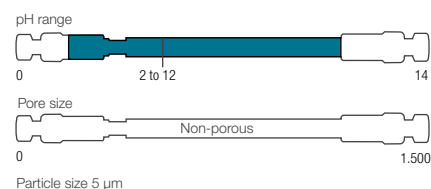


MABPac SCX-10 columns

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
		250	—	—	—

Charge variant analysis continued

MAbPac SCX-10 RS column

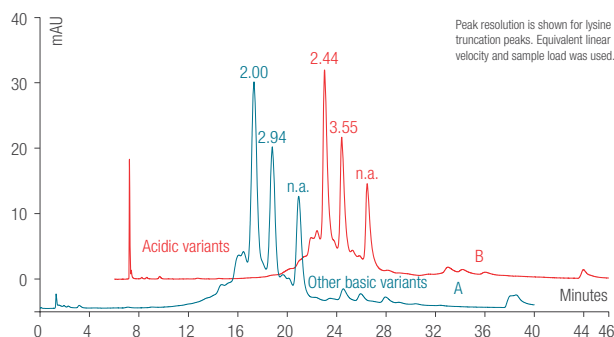


Additional reading

Links	Description
 Learn more thermofisher.com/biolc	

Lysine variants

MAbPac SCX, 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

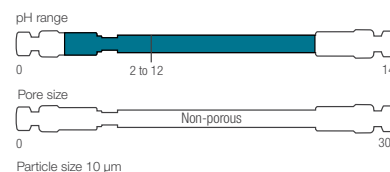


MAbPac SCX-10 RS columns

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 continued

ProPac SAX-10 column

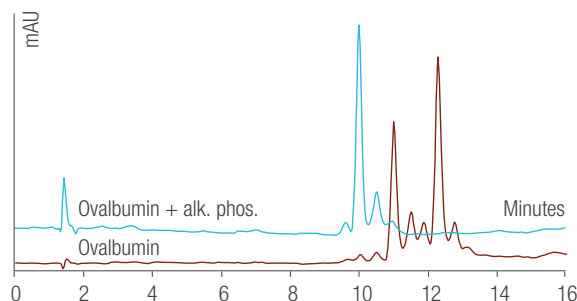


Additional reading

Links	Description
Learn more thermofisher.com/biolc	

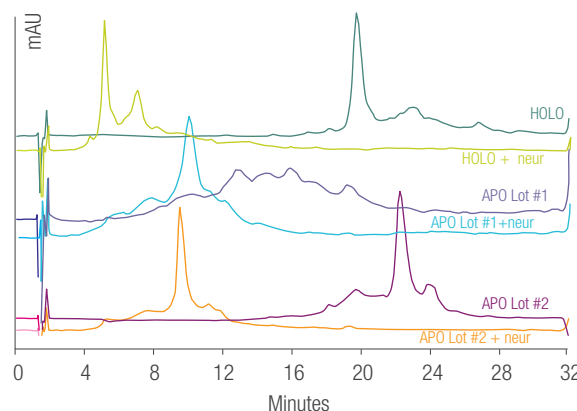
Resolution of phosphorylation variants of ovalbumin

ProPac SAX-10, 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, 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

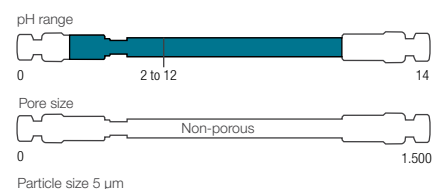


ProPac SAX-10 columns

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 continued

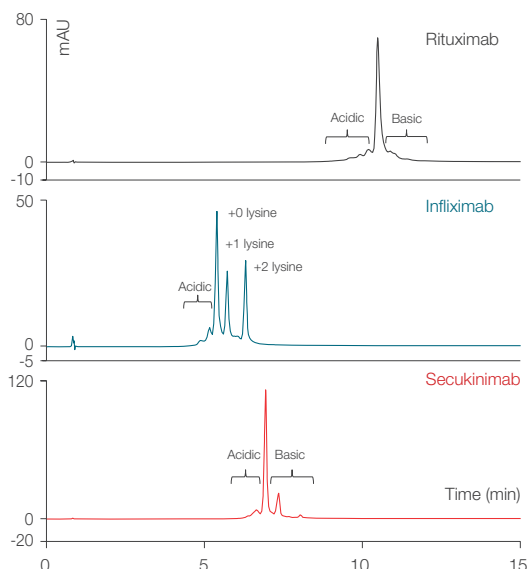
ProPac Elite WCX column



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
	Learn more thermofisher.com/biolc	

ProPac Elite WCX, 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

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

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 continued

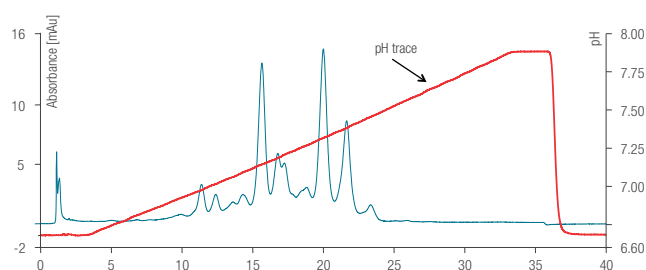
pH gradient buffers

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

Description	Buffer bottle size			
Buffer	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



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

Peptide mapping and MAM

Thermo Scientific™ Hypersil GOLD™ VANQUISH™ 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. For faster separation of peptide samples select the Thermo Scientific™ Accucore™ C18 VANQUISH™ column. The column offers sub-2 µm particles providing ultra-short diffusion paths that result in extremely efficient separations.

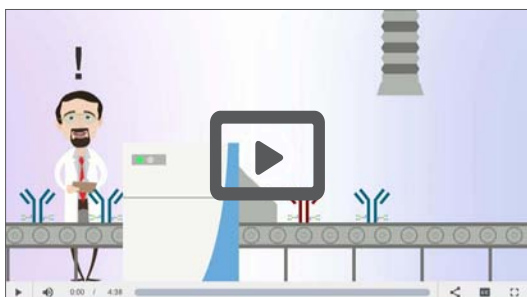
Additional reading

Links	Type	Description
	Landing page	Multi-Attribute Method (MAM): Straight through to breakthrough
	Learning centre	Biopharmaceutical Multi-Attribute Method (MAM)
	Learn more thermofisher.com/biolc	



Video:

End-to-end MAM solution to move biopharma forward



Video:

Learn how innovation and monitoring strategies can reduce the number of tests and enhance the methodology of validating impurity

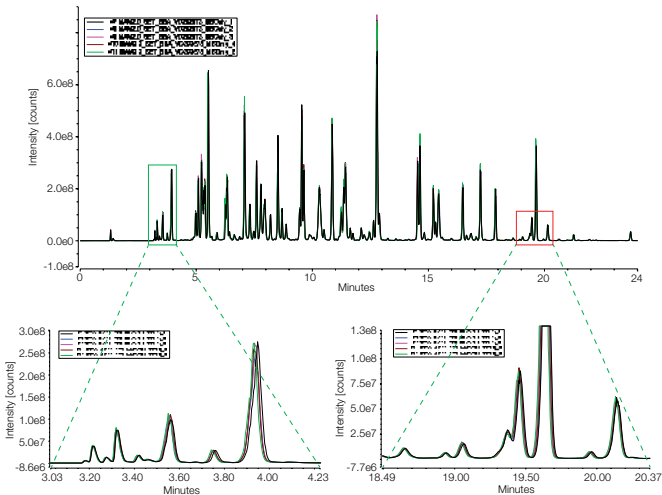
Hypersil GOLD VANQUISH column

Additional reading

Links	Type	Description
	Flyer	VANQUISH UHPLC columns. Delivering powerful separations
	Application note	An integrated LC-MS system performance evaluation test
	Learn more thermofisher.com/biolc	

Overlay of 5 TIC traces from the SET injection sequence

Hypersil GOLD VANQUISH C18 UHPLC column, 150 x 2.1 mm, 1.9 µm	
Flow rate	0.25 mL/min
Mobile phase A	H ₂ O + 0.1% FA
Mobile phase B	ACN + 0.1% FA
Injection volume	5 µL
Detection	Mass spectrometer – Full scan
Sample	Pierce BSA protein digest standard, MS grade, UD294474 (P/N 88341)
Chromatogram B	Gradient: 33-53% B in 20 min



Hypersil GOLD Vanquish columns

Particle size (µm)	Length (mm)	2.1 mm ID
1.9	50	25002-052130-V
	100	25002-102130-V
	150	25002-152130-V

Peptide mapping and MAM continued

Accucore VANQUISH C18+ column

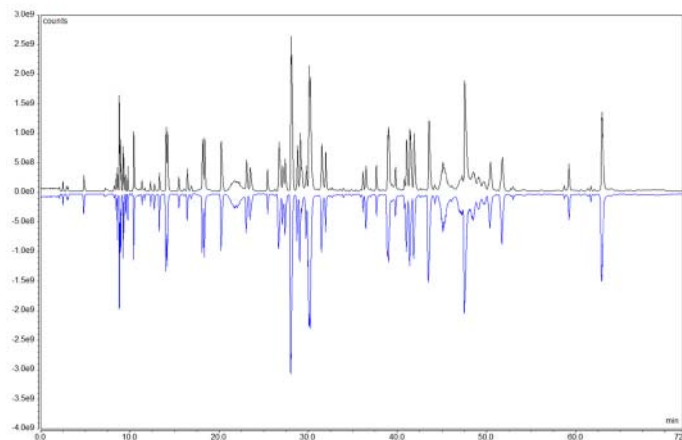
Additional reading

Links	Type	Description
	Application note	Comparative analysis of innovator and biosimilar monoclonal antibodies using MAM method
	Technical guide	Powerful separations are our core performance
	Poster	Application of a MS in QC method for characterization and attribute monitoring
	Learn more thermofisher.com/biolc	

Mirrored base peak chromatograms of rituximab innovator (black) and its biosimilar product (blue)

Accucore Vanquish C18+ UHPLC column, 1.5 μ m, 2.1 $\times$ 150 mm

Flow rate	0.25 mL/min
Mobile phase A	H ₂ O + 0.1% FA
Mobile phase B	ACN + 0.1% FA
Injection volume	8 μ L
Detection	Mass spectrometer
Sample	Rituximab innovator
Temperature	50 $^{\circ}$ C



Accucore Vanquish C18+ columns

Particle size (μ m)	Length (mm)	2.1 mm ID
1.5 μ m	50 mm	27101-052130
	100 mm	27101-102130
	150 mm	27101-152130

Nucleic acids/oligonucleotides

Thermo Scientific™ DNAPac™ RP column 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.

Thermo Scientific™ DNAPac™ PA200 and 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.

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.



Webinars

Analytical and life science webinars live and on-demand



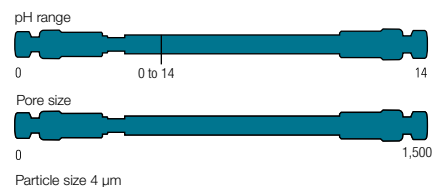
NIBRT collaboration information

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

thermofisher.com/nibrt

Nucleic acids/oligonucleotides continued

DNAPac RP column

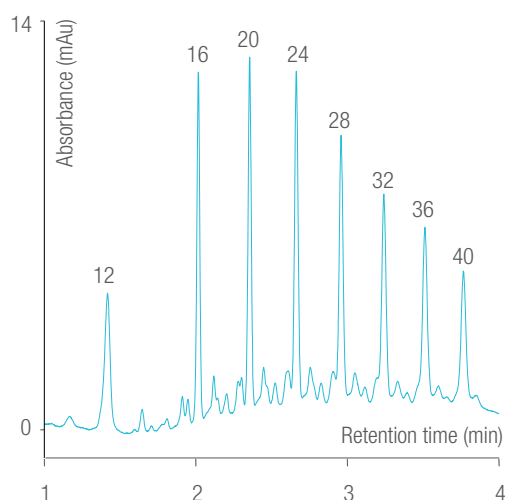


Additional reading

Links	Description
 Learn more thermofisher.com/biolc	

Fast analysis of mixed base DNA

DNAPac RP, 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		
	Time (min)	%A	%B
	-0.1	67	33
	0.0	67	33
	3.0	41	59
Gradient	3.1	5	95
	4.9	5	95
	5.0	67	33
	8.0	67	33

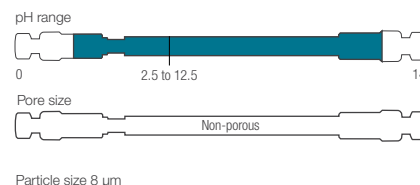


DNAPac RP columns

Particle size (µm)	Format	Length (mm)	2.1 mm ID	3.0 mm ID
4	Guard cartridges (2/pk)	10	088925	088921
	HPLC column	50	088924	088920
		100	088923	088919
		250	303324	—
—	Guard cartridge holder	—	069580	069580

Nucleic acids/oligonucleotides continued

DNAPac PA200 column



Additional reading

Links

Description



Learn more thermofisher.com/biolc

DNAPac PA200, 8 µm, 250 x 4.0 mm

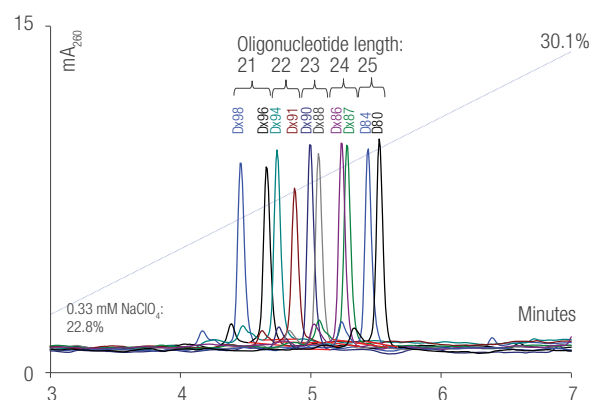
Flow rate: 1.2 mL/min

Mobile phase: NaClO₄, pH 6.5 with 20% ACN

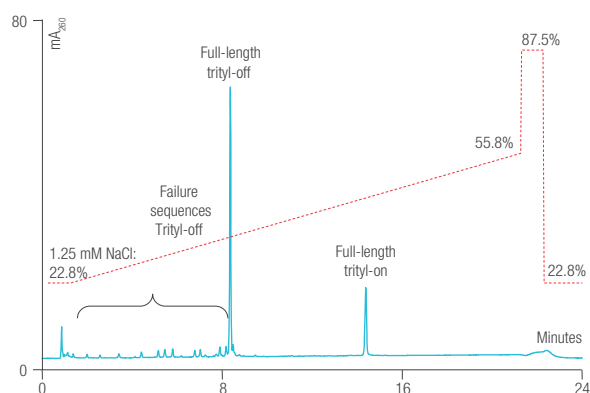
Detection: UV at 260 nm

Flow rate: 1.2 mL/min

Separation of oligonucleotides by length



Target, failure and trityl-on oligonucleotides

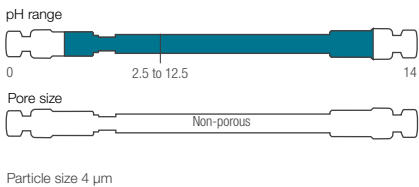


DNAPac PA200 columns

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 continued

DNAPac PA200 RS column



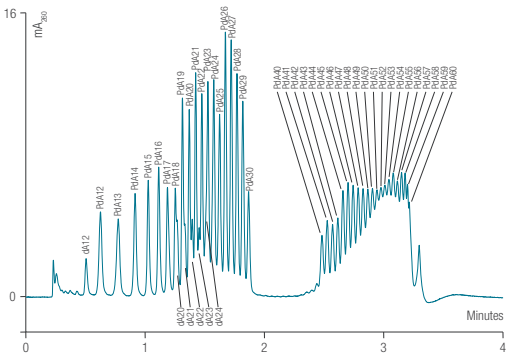
Additional reading

Links	Type	Description
	Brochure	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
	Learn more thermofisher.com/biolc	

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

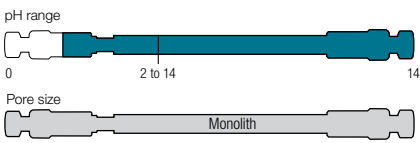
*CV = column volumes
**Curve 3 indicates continuously changing gradient, asymptotically approaching a maximum salt concentration. Programmed in Thermo Scientific™ Chromeleon™ 6.8.



DNAPac PA200 RS columns

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

DNASwift SAX-1S column

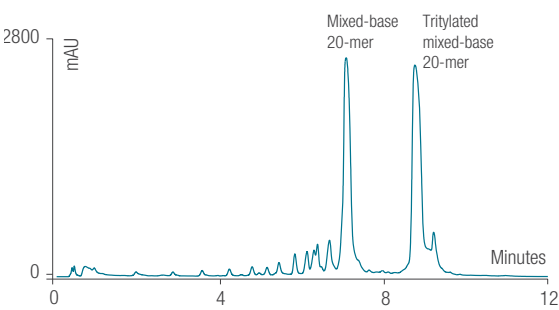


Additional reading

Links	Description
 Learn more thermofisher.com/biolc	

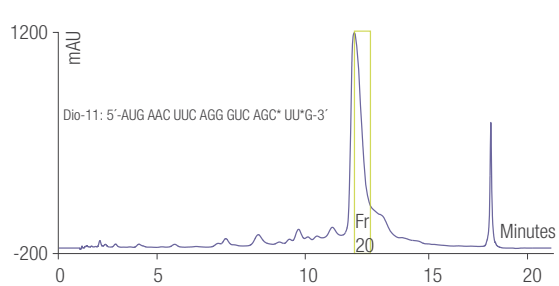
Tritylated oligonucleotide

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



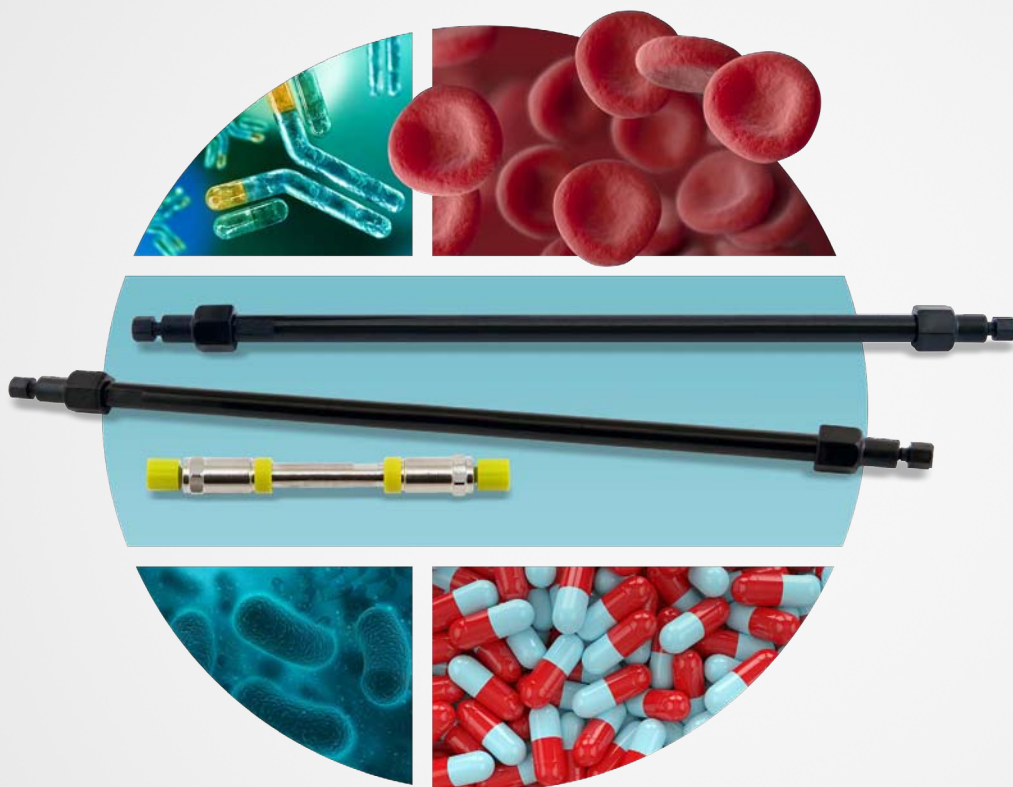
Purification of a 21-base RNA sample with aberrant 2'-5' linkages at the 1 and 3 positions from the 3' end

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

Length (mm)	5.0 mm ID
150	066766



Expect reproducible results with sample prep, columns and vials



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