

# Connected chromatography Solutions



# **BioLC Columns** and Accessories

The analysis of proteins, peptides, oligonucleotides and other biomolecules demands a range of sample separation modes, column chemistries, column configurations and detection techniques. The range of Thermo Scientific<sup>™</sup> polymeric and silica columns in analytical and nano-scale formats are designed to handle these challenging separations.

# Featured Products



#### **MAbPac SEC-1**

A size exclusion chromatography (SEC) column specifically designed for the high-resolution separation and characterization of monoclonal antibodies (mAbs) and their aggregates.

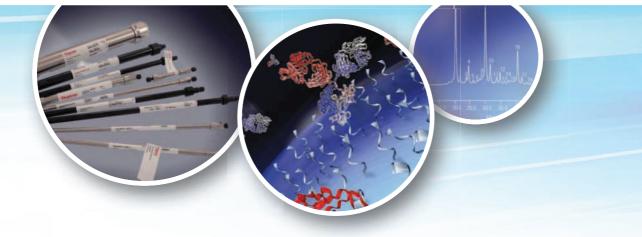
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#### **MAbPac SCX**

Strong cation exchange columns designed specifically for the highresolution, high efficiency charged variant analysis of monoclonal antibodies and associated variants.

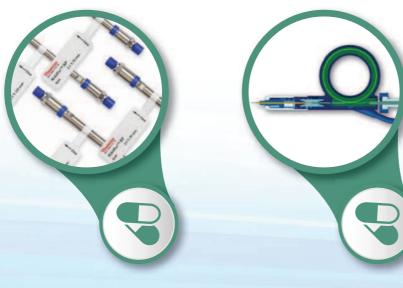
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#### **MAbPac RP**

Columns designed for high-resolution accurate mass analysis of monoclonal antibodies (mAbs), antibody drug conjugates (ADCs) and other proteins using reversed-phase HPLC and LC-MS for high-resolution separations.

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### **EASY-Spray**

EASY-Spray columns offer outstanding peak capacity for comprehensive proteome characterization and temperature control for maximum reliability and performance.

**PAGE 3-040** 



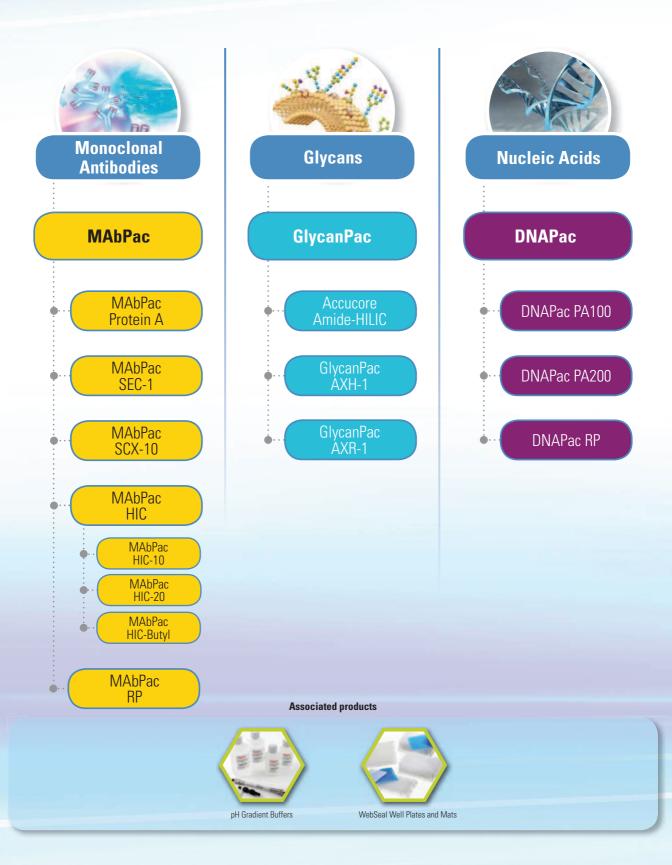


# visit **www.thermoscientific.com/chromexpert** to access the following information:

- BioLC Columns Selection Guide
- HPLC Phases for Biomolecules
- Columns for Protein Separations
- Columns for Monoclonal Antibody Separations
- Columns for Carbohydrate Separations
- Columns for Oligonucleotide Separations



# Columns for Biomolecules BioLC Column Lines



3-004



# **MAbPac HPLC and UHPLC Columns**

## MAbPac Protein A

Fast mAb titer analysis

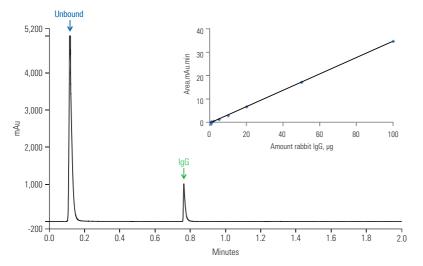
- High efficiency column
- Rugged, long column lifetime
- Excellent sample recovery
- Designed for ease of use and automation



# monoclonal antibody (mAb) titer analysis of samples such as harvest cell cultures (HCC). This HPLC column offers high throughput and accurate analysis through a combination of low back pressure and high efficiency. The MAbPac Protein A column format allows rapid automation of loading, binding, elution and collection using Thermo Scientific biocompatible systems. The column is based on a novel non-porous polymeric resin consisting of a divinylbenzene core and a hydrophilic surface, optimized for affinity separation.

Thermo Scientific<sup>™</sup> MAbPac<sup>™</sup> Protein A is an affinity column designed to provide fast

#### Harvest cell culture titer analysis



#### MAbPac Protein A, 12µm, 35 x 4.0mm Flow Rate: 2 mL/min

now nate.	2 1112/11111
Mobile Phase A:	50mM Sodium Phosphate, 150mM
	NaCl, 5% acetonitrile, pH 7.5
Mobile Phase B:	50mM Sodium Phosphate, 150mM
	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:	10µL
Detection:	280nm
Sample:	MAb B, 5mg/mL Harvest Cell Culture

#### **MAbPac Protein A**

Particle Size (µm)	Format	Length (mm)	4.0mm ID
12	HPLC Column	35	082539



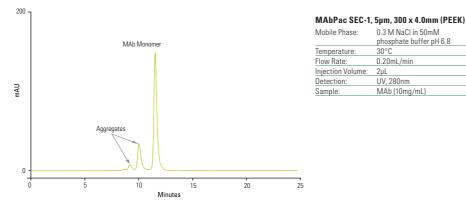
Thermo Scientific Chromatography Columns and Consumables 2016-2017

# MAbPac SEC-1

A size exclusion chromatography (SEC) column specifically designed for the high-resolution separation and characterization of monoclonal antibodies (mAbs) and their aggregates

- Analysis of monoclonal antibodies (mAbs) and their aggregates
- Analysis of mAb Fab and Fc fragments, even using high and low salt concentrations
- Hydrophilic bonded layer for minimal nondesired interactions between the biomolecules and the stationary phase
- Stable surface bonding leads to low column bleed and compatibility with MS, ELSD and Corona Charged Aerosol Detection (CAD)
- Separation range for globular proteins 10,000–1,000,000; exclusion limit for globular proteins >1,000,000

#### Monoclonal antibody aggregate separation



#### **MAbPac SEC-1**

Particle Size (µm)	Format	Length (mm)	2.1mm ID	4.0mm ID	7.8mm ID
5	Guard Column	50	-	074697	-
	HPLC Column	150	088790	075592	-
		300	088789	074696	088460



0.3 M NaCl in 50mM phosphate buffer pH 6.8

30°C

0.20mL/mir

2µL UV, 280nm

MAb (10mg/mL)

# MAbPac SCX-10

Strong cation exchange column designed specifically for the high-resolution, high efficiency charged variant analysis of monoclonal antibodies and associated variants

 pH Range

 0
 2 to 12

 0
 2 to 12

 14

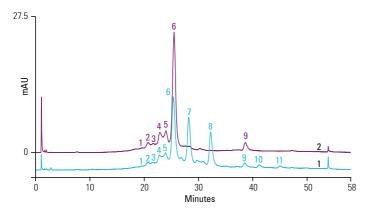
 Pore Size

 0
 Non-porous

Particle Size 3µm, 5µm, 10µm

- Exceptionally high-resolution for monoclonal antibody charged variants separation
- Ideal for characterization and quality control assessment of monoclonal antibodies
- Unmatched column-to-column and lot-to-lot reproducibility
- Hydrophobic interactions greatly minimized
- Ideal for stability studies
- Meets the regulatory requirements for biopharmaceutical characterization

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



MAbPac SCX-1	), 5µm, 250 x 4.0mm
Mobile Phase A:	20mM MES (pH 5.6) + 60mM NaCl
Mobile Phase B:	20mM MES (pH 5.6) + 300mM NaCl
Gradient:	15-36% B in 50 min
Temperature:	30°C
Flow Rate:	1mL/min
Injection Volume:	5µL
Detection:	UV at 280nm
Samples:	1. MAb B, 900µg in 100µL
	(no carboxypeptidase)
	2. MAb B, 900µg in 100µL +
	carboxypeptidase, 50µg,
	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

Particle Size (µm)	Format	Length (mm)	2.0mm ID	4.0mm ID	9.0mm ID
3	HPLC Column	50	-	077907	-
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



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14

1,500

USP N/A

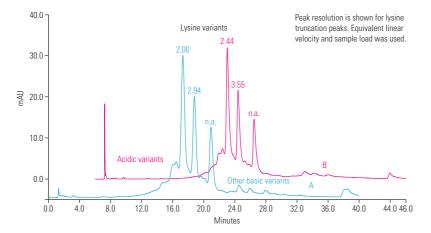
# MAbPac SCX-10 RS

BioRS (Rapid Separation), strong cation exchange column designed for monoclonal antibodies and associated charged variants

- UHPLC, high throughput analysis
- Specially developed bio-inert PEEK lined stainless steel column hardware
- High pressure compatibility
- Suitable for operation up to 7,000 psi

Higher resolution and throughput of mAb charge variant UHPLC separations can be achieved using the MAbPac SCX-10 RS strong cation-exchange phase with specially developed bio-inert PEEK lined stainless steel column hardware. These columns are designed to be used at higher UHPLC conditions to maximize the resolution of mAb variant separation. Higher pressure compatibility of the column hardware allows use of high flow rates for faster separation.

#### Improved mAb resolution



#### MAbPac SCX, 5µm, 250 x 4.6mm

pH Range

Pore Size

0

2 to 12

Particle Size 3µm, 5µm, 10µm

	<b>-</b> , <b>-</b>
Mobile Phase A:	20 mM MES pH 5.6 + 60 mM
Mobile Phase B:	20 mM MES pH 5.6 + 300 mM NaCl
Flow Rate:	1.5 mL/min
Injection Volume:	15µL
Sample:	MAb 5mg/mL
Chromatogram A:	Gradient: 33-53% B in 30 min
Chromatogram B:	Gradient: 33-53% in 20 min

#### MAbPac SCX-10 RS

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



# pH Gradient Buffers

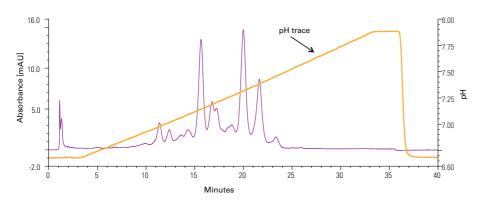
Ready-to-use buffers for simple method development during charge variant characterization



The 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

Thermo Scientific pH buffer concentrates can be purchased individually or as a pair, in quantities of 125mL or 250mL. For added convenience, the 125mL buffers can also be bundled with columns in a number of specifically preconfigured kits.



#### 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 Bot	Buffer Bottle size		
Buffer	125mL	250mL		
CX-1 pH Gradient Buffer A (pH 5.6)	083273	085346		
CX-1 pH Gradient Buffer B (pH 10.2)	083275	085348		

Kits		Buffer Bottle size		
Buffer	MAbPac SCX-10 Column format	125mL	250mL	
Gradient Buffer Kit: Includes both Buffer A & Buffer B (available in either 125mL or 250mL size – one bottle each/ kit)	-	083274	085349	
Gradient Starter Kit: Includes both Buffer A & Buffer B + MAbPac SCX-10	10µm, 4 × 250mm column	083381	-	
Gradient High Throughput Kit: Includes both Buffer A & Buffer B + MAbPac SCX-10	5µm, 4 × 50mm column	083378	-	
Gradient High Resolution Kit: Includes both Buffer A & Buffer B + MAbPac SCX-10	5µm, 4 × 250mm column	083272	-	

# Monoclonal Antibody Characterization and Analysis Kits

MAb Charge Variant Analysis IEX Column Kit

MAb Charge Variant Analysis IEX Column Kit includes two ion-exchange (IEX) specialty columns for mAb charge variants analysis. This kit is a convenient starter kit for researchers at the beginning of a mAb analysis projects, and facilitates the screening of two columns for determination of the best column for their specific monoclonal antibody sample.

Included in the Kit:

- ProPac WCX-10 Analytical column, 4 × 250mm (P/N 054993)
- MAbPac SCX-10 Analytical column, 4 × 250mm (P/N 074625)

#### **MAb Charge Variants Kit**

Description	Cat. No.
MAb Charge Variant Analysis IEX Column Kit	076196

# MAb Analysis IEX and SEC Column Kit

The MAb Analysis IEX and SEC Column Kit includes two columns: an ion-exchange (IEX) column and a size-exclusion (SEC) column. This kit is a convenient starter and column replacement kit for mAb analysis projects. It is useful for researchers at the beginning of mAb analysis projects, and facilitates the screening of aggregates and variants in two columns.

Included in the kit:

- MAbPac SCX-10 Analytical column, 4 × 250mm (P/N 074625)
- MAbPac SEC-1 Analytical column, 4 × 300mm (P/N 074696)

#### **MAb Analysis Kit**

Description	Cat. No.
MAb Analysis IEX and SEC Column Kit	076197



# MAbPac HIC Family

- Advanced column chemistry designed for separating mAbs and related biologics
- Broad selectivity coverage for most challenging separations of mAbs
- Excellent bio-compatibility
- High column efficiency
- Rugged column packing

#### **Applications**

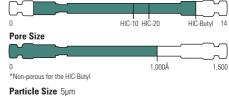
The MAbPac HIC-10 is the column of choice for intact mAbs/proteins and mAb aggregates while the MAbPac HIC-20 is suited to resolve mAb fragments, oxidized mAbs and bispecific mAbs. When it comes to ADCs, MAbPac HIC-Butyl is ideal for cystein-conjugated ADC while MAbPac HIC-10 and MAbPac HIC-20 have proven useful for several cysteine proprietary ADC molecules, as shown below.

#### **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	+++	++++	++

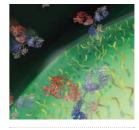
Greater number of ++++ denotes greater suitability

#### pH Range





MAbPac HIC-10



MAbPac HIC-20

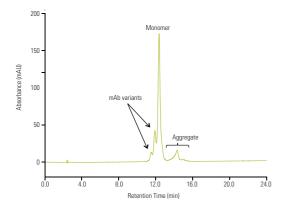


MAbPac HIC-Butyl

#### **MAbPac HIC Family**

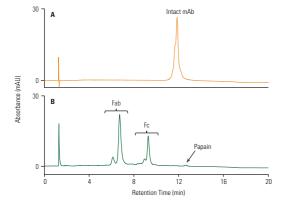
Description	Particle Size (µm)	Format	Length (mm)	4.6mm ID
MAbPac HIC-10	5	Guard Cartridges (2/pk)	10	088482
		HPLC Column	100	088480
			250	088481
MAbPac HIC-20	5	Guard Cartridges (2/pk)	10	088555
		HPLC Column	100	088553
			250	088554
MAbPac HIC-Butyl	5	Guard Cartridges (2/pk)	10	088559
		HPLC Column	100	088558
Guard Cartridge Holder				069580

#### Separation of mAb Aggregates



MAbPac HIC-10, 5µm, 100 x 4.6mm				
Mobile Phase A:	2 M ammonium sulfate, 100mM			
	sodium phosp	ohate, pH 7.0		
Mobile Phase B:	100mM sodiu	ım phosphate	e, pH 7.0	
Gradient:	Time (min)	%A	%B	
	-5.0	60	40	
	0.0	60	40	
	1.0	60	40	
	29.0	0	100	
	34.0	0	100	
Temperature:	20°C			
Flow Rate:	0.5mL/min			
Injection Volume:	10µL			
Detection:	UV, 280nm			
Sample:	Monoclonal antibody (4mg/mL)			

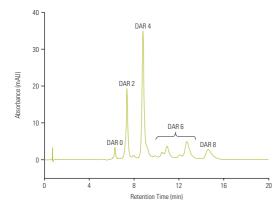
#### Separation of mAb Fragments



#### MAbPac HIC-20, 5μm, 100 x 4.6mm Mobile Phase Δ: 2 M ammonium sulfat

Mobile Phase A:	2 M ammonium sulfate, 100mM sodium phosphate, pH 7.0			
Mobile Phase B:			hosphate, pH 7.0	
Gradient:	Time (min)	%A	%B	
	-5.0	60	40	
	0.0 60 40			
	1.0	60	40	
	15.0	0	100	
	20.0	0	100	
Temperature:	30°C			
Flow Rate:	1.0mL/min			
Injection Volume:	Intact mAb: 5µL			
	Papain digest: 12µL			
Detection:	UV, 280nm			
Sample:	a. Intact mAb (2.5mg/mL)			
	b. Papain digest (1mg/mL)			

#### Separation of Antibody Drug Conjugates (ADCs)



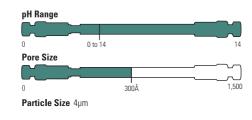
#### MAbPac HIC-Butyl, 5µm, 100 x 4.6mm

Mobile Phase A:	1.5 M ammonium sulfate, 50mM sodium phosphate, pH 7.0/ isopropanol (95:5 v/v)			
Mobile Phase B:	50mM sodium phosphate, pH 7.0/ isopropanol (80:20 v/v)			
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	
Temperature:	25°C			
Flow Rate:	1.0mL/min			
Injection Volume:	5µL			
Detection:	UV, 280nm			
Sample:	Cys-conjugated ADC mimic (5mg/mL)			

# MAbPac RP

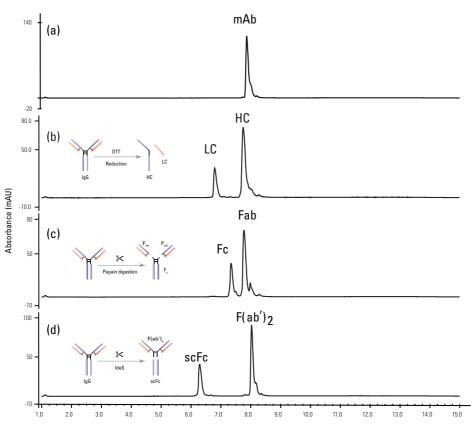
High resolution accurate mass determination of monoclonal antibody variants, antibody drug conjugates (ADC) and proteins

- Superior resolution power for monoclonal antibodies and related substances
- High efficiency with low carry-over
- Excellent MS compatibility
- Wide operating pH range: 0 14
- High temperature stability: up to 110°C
- High throughput



The Thermo Scientific<sup>™</sup> MAbPac<sup>™</sup> RP is a reverse phase (RP) liquid chromatography column designed for mAb characterization. Highly efficient separations can be achieved for mAbs and their variants, light chain (LC) and heavy chain (HC), Fc and Fab fragments, scFc and F(ab')2 fragments. The unique column chemistry provides stability under wide pH range, high temperature, and organic mobile phases and is fully compatible with both UV and MS detection.

#### mAb and mAb Fragments Analysis



#### MAbPac RP, 4µm, 50 x 3.0mm

Mobile Phase A: Mobile Phase B:	H <sub>2</sub> O/FA/TFA (99.88 : 0.1:0.02 v/v/v) MeCN/ H <sub>2</sub> O/FA/TFA (90: 9.88 :0.1:0.02 v/v/v/v)		
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
	12.0	80	20
Temperature:	80°C		
Flow Rate:	0.5mL/min		
Injection Volume:	5µL		
Detection:	UV, 280nm		
Sample:	(a) trastuzumab (5mg/mL) (b) trastuzumab + DTT (4mg/mL) (c) trastuzumab + Papain (2mg/mL) (d) trastuzumab + IdeS (2mg/mL)		

#### MAbPac RP

Particle Size (µm)	Format	Length (mm)	2.1mm ID	3.0mm ID
4	Guard Cartridges (2/pk)	10	088649	088646
	HPLC Column	50	088648	088645
		100	088647	088644
	Guard Cartridge Holder		069580	069580