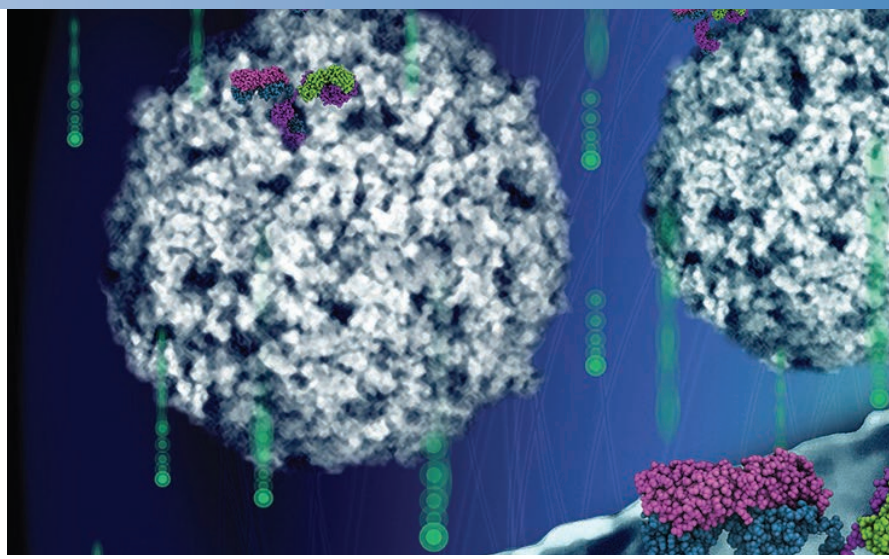
MSPac DS-10 Desalting Cartridge

Superior desalting performance

The Thermo Scientific™ MSPac™ DS-10 desalting cartridge reduces the background contamination and noise frequently seen in LC-MS analysis of samples containing non-volatile salts. Removal of these salts prior to analysis enhances analyte response, increases signal to noise ratio and improves sensitivity, providing a more reliable identification.

- Low peak carryover for sample to sample confidence
- High capacity for antibody and protein desalting in identification, biomarker discovery and systems biology.
- High pH stability for superior lifetime with high pH samples
- Use with salt based fractions from IEX, pH-gradient, SEC etc.



Improved Sensitivity in LC-MS Analysis

The MSPac DS-10 desalting cartridge reduces background contamination and noise seen in LC-MS analysis of samples containing non-volatile salts. Removal of these salts prior to analysis enhances analyte response and improves sensitivity aiding a more reliable identification in MS. The cartridge can be used alone or in combination with an analytical column for high resolution separations.

Run to Run Reliability

The MSPac DS-10 desalting cartridge was developed to be robust and able to handle complex sample matrices without significant carryover.

High Loading Capacity

The high-capacity desalting cartridge is used for the on-line desalting of biological samples for analysis by reversed phase LC-MS. The high loading capacity of the desalting cartridge makes it suitable for use in identification and quantitation in clinical research, biomarker discovery and systems biology applications.

Technology

The MSPac DS-10 desalting media is based on a 5 µm divinylbenzene co-polymer resin. The reversed-phase media retains proteins under aqueous conditions while salts and hydrophilic matrix components are eluted from the cartridge; the large pore structure of the supermacroporous resin allows the use of high flow rates at low temperatures with a low column backpressure; the lower hydrophobicity of the phenyl functionalized co-polymer results in reduced carryover between analyses.

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Applications

Rapid LC-MS Analysis of a mAb

Monoclonal Antibodies (mAbs) as biopharmaceutical therapeutics have grown in use rapidly over the last decade. To make the MS characterization process economical, high throughput methods must be developed that enable rapid and inexpensive characterization of mAbs so that promising cell lines can be selected for further development.

The ability to load and desalt an intact mAb (Rituximab) using the fast 4 minute analysis, as shown in Figure 1 is key for this high-throughput application area. Glycoforms of the intact mAb ≥ 10% and ≥ 30% were characterized by MS up to 100 and 300 runs, respectively, as shown in Figure 2.

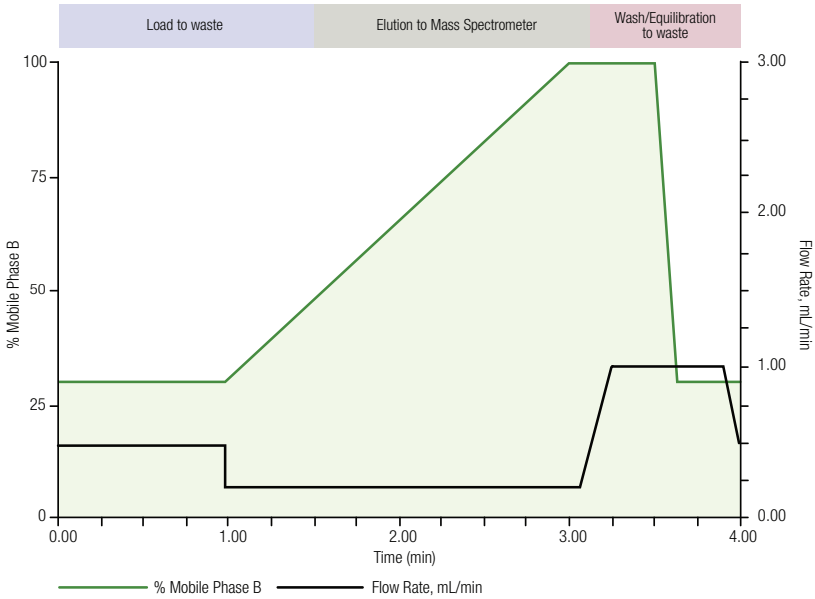


Figure 1. Chromatographic method schematic. Mobile phase A: water + 0.1% formic acid. Mobile phase B: 80/20 (v/v) acetonitrile/water + 0.1% formic acid.

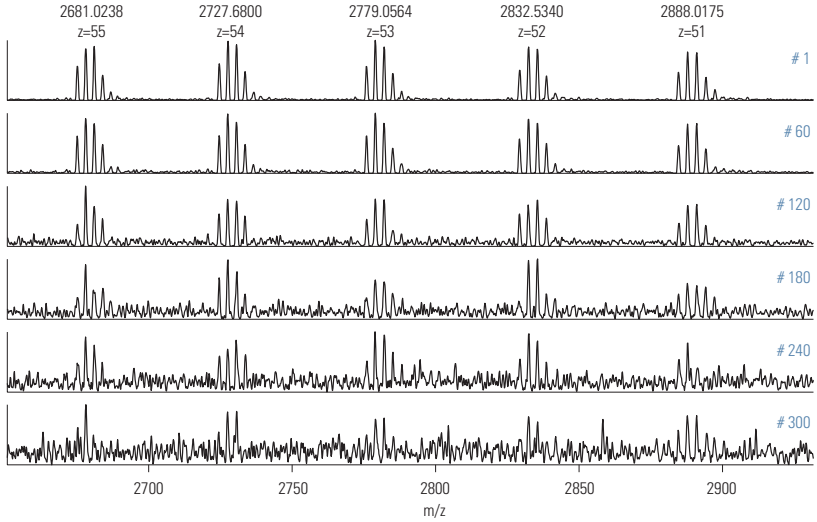
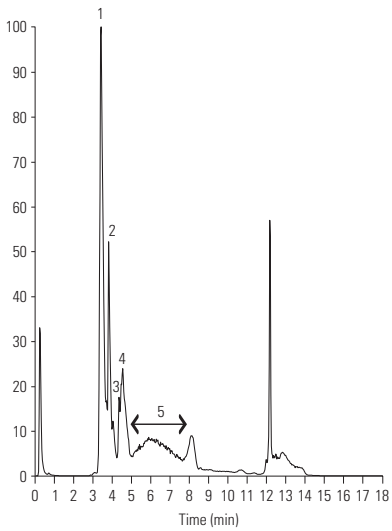


Figure 2. Ruggedness as measured by spectrum quality >300 injections before significant deterioration of MS signal is observed.

LC-MS Analysis of mAb Fragments

As seen in Figure 3, MS is an essential tool in the characterization of mAbs, providing molecular weight determinations and structural information of intact as well as digested mAbs. Direct infusion of the sample into the MS is the simplest approach to characterize the sample. Desalting, separation and MS characterization of digested monoclonal antibody fragments of the pharmaceutical mAb Rituximab have been analyzed. The Lc, Fc/2, and Fd' antibody fragments were separated and characterized with HRAMS as seen in Figure 3.



Cartridge:	**MSPac DS-10 Desalting Cartridge**
Format:	2.1 × 10 mm
Mobile Phase A:	Water + 0.1% formic acid
Mobile Phase B:	20/80 (v/v) water/acetonitrile + 0.1% formic acid

Gradient:	Time (min)	A	B
	0.0	75	25
	1.0	75	25
	11.0	30	70
	11.1	0	100
	13.0	0	100
	14.0	75	25
	18.0	75	25

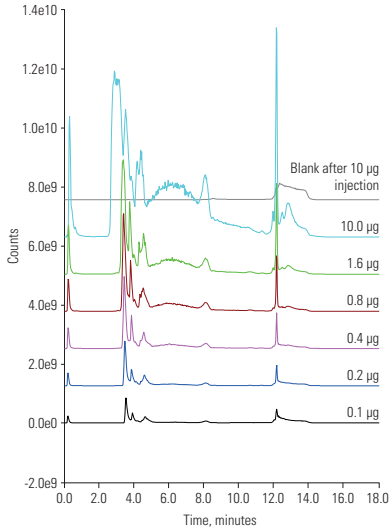
Flow Rate:	0.2 mL/min
Inj. Volume:	0.8 µL
Temp.:	50 °C
Detection:	MS
Sample:	Rituximab digested with IdeS and reduced with TCEP; 1 mg/mL

Peaks:
1. Fc/2 – C-terminal fragment of heavy chain with glycosylated N301
2. Lc – Light chain
3. Fd' – N-terminal fragment of heavy chain
4. Intact mAb
5. Undigested and partially reduced species

Figure 3. Separation of fragments and intact mAb from digested and reduced rituximab.

Protein Loading and Carryover

It is important to load a sufficient amount of protein onto the cartridge to obtain sufficient signal strength for effective MS characterization and to obtain clean MS spectra. The spectra for increasing mass loading of a digested and reduced mAb ranging from 0.1–10 µg of total protein and the blank run after the 10 µg injection indicates effective desalting even at high mass loading amounts. It can even be seen that while it is common in reversed phase chromatography to observe hydrophobic sample artifacts eluting as several peaks during the step wash at 100% mobile phase B, no detectable carryover was observed following the 10 µg protein injection using the MSPac DS-10 desalting cartridge.



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	11.0	30	70
	11.1	0	100
	13.0	0	100
	14.0	75	25
	18.0	75	25
Flow Rate:	0.2 mL/min		
Inj. Volume:	0.1–10 µL		
Temp.:	50 °C		
Detection:	MS		
Sample:	Rituximab digested with IdeS and reduced with TCEP; 1 mg/mL		

Figure 4. Loading analysis of rituximab fragments, including blank injection after 10.0 µg.

Operational Specifications

Parameter	Recommendation
Flow Rate Range:	Loading: 0.1–0.5 mL/min Analysis: 0.1–0.3 mL/min Re-equilibration: 0.5–1.0 mL/min Maximum flow rate: 1.0 mL/min
Cartridge Storage	Long term: Acetonitrile Short term: High organic content mobile phase
Common Mobile Phase	Mobile phase A: Water + 0.1% formic acid Mobile phase B: Acetonitrile/water (80/20 v/v) + 0.1% formic acid
Solvent Compatibility	Common reverse phase solvents including acetonitrile, acetone, methanol, ethanol, n-propanol, and isopropyl alcohol
Maximum Temperature:	90 °C
Pressure Limit	6,000 psi
pH Range	0–14
Protein Loading	0.01–10 µg
Antibody Frontal Loading Capacity	150 µg Human IgG Antibody
Suggested Tubing	Thermo Scientific™ Viper™ Capillary Kit for Biocompatible RSLC Systems

Ordering Information

Description	Particle Size	Part Number
MSPac DS-10, 2.1 × 10 mm (2/pk)	5 µm	089170
Cartridge Holder		069580

www.thermoscientific.com/desalter

Thermo Fisher Scientific, Sunnyvale, CA USA is ISO 9001 Certified.

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